TEXT PROBLEM WITHIN THE BOOK ONLY

UNIVERSAL
LIBRARY

OU_162568

UNIVERSAL
LIBRARY

ORGANIC MEDICAMENTS
AND THEIR
PREPARATION

A TEXT-BOOK OF PHARMACOLOGY AND THERAPEUTICS.

By ARTHUR R CUSHNY, M D , F R S 8th Edition 73 Illustrations **24s.**

THE PLANT ALKALOIDS.

By T. A. HENRY, D Sc 2nd Edition. 8 Plates. **28s.**

A TEXT-BOOK OF MATERIA MEDICA.

By H. G GREENISH, D ÉsSc , F I C. 4th Edition 270 Illustrations. **25s.**

LEE AND GATENBY'S MICROTOMISTS' VADE-MECUM.

8th Edition. **28s.**

CRITICAL MICROSCOPY, How to get the best out of the Microscope. 7s. 6d.

PREGL'S QUANTITATIVE ORGANIC MICRO-ANALYSIS.

Translated by E. FYLEMAN, Ph D. 42 Illustrations **12s. 6d.**

A GERMAN-ENGLISH DICTIONARY OF TERMS USED IN MEDICINE AND THE ALLIED SCIENCES.

By H LANG, B A , and MILTON K. MEYERS, M D 3rd Edition **28s.**

RECENT ADVANCES IN MEDICINE, Clinical-Laboratory-Therapeutic.

By G E BEAUMONT, D M , and E C DODDS, M B , B S 37 Illustrations. **10s. 6d.**

J. & A. CHURCHILL

ORGANIC MEDICAMENTS
AND THEIR
PREPARATION

BY

ERNEST FOURNEAU

Head of the Laboratory for Therapeutical Chemistry in the Pasteur Institute; Member of the Academy of Medicine; formerly Director of the Poulenc Laboratories

Authorised Translation

BY

W. A. SILVESTER

M.Sc.(Sheffield)

Chemist with the British Dyestuffs Corporation, Ltd.

With Prefaces,

To the French Edition by

ÉMILE ROUX

Member of the Institute; Director of the Pasteur Institute,

And to the English Edition by

GEORGE BARGER

M.A., D Sc., F.R.S., Professor of Medical Chemistry, University of Edinburgh

22 ILLUSTRATIONS

LONDON

J. & A. CHURCHILL

7 GREAT MARLBOROUGH STREET

1925

Printed in Great Britain

PREFACE

TO THE ENGLISH EDITION

BY

GEORGE BARGER, M.A., D.Sc., F.R.S.

Professor of Medical Chemistry in the University of Edinburgh

MR. SILVESTER has asked me to write a preface to his translation of M. Fourneau's *Préparation des médicaments organiques*. I do so with considerable hesitation, for the writing of a preface to the work of others seems to imply scientific eminence to which I cannot lay claim. If it merely implied appreciation and admiration, I should write with less misgiving.

M. Fourneau has long been known as a master of drug synthesis, a field of organic chemistry which has been little cultivated outside Germany. Not only has he done much work of an academic kind, but as a former director of the Poulenc Laboratories he has also an intimate acquaintance with industrial practice. With Stovaine he has permanently enriched our store of medicaments; the subtle allusion in the naming of this drug should appeal to English-speaking students.

The present book shows M. Fourneau in yet another capacity, that of the teacher, for it originated in a brief course of instruction in the preparation of synthetic drugs, given by him at Madrid, in 1917, on the invitation of the Junta para ampliacone de Estudios. As the title implies, this book should make a direct practical appeal to manufacturers and technologists, but it does much more than that. Since the various preparations start from common materials, and all the successive operations are described in detail, the book is a veritable manual of organic preparative chemistry, of a novel and interesting kind. Most of these books have been influenced by the dyestuff industry, but quite as much chemistry may be learned from that other branch of organic chemical manufacture which produces synthetic drugs. A student who works through M. Fourneau's book will, moreover, gain an appreciation of other aspects in which the ordinary text-book is lacking. Thus, in the

first chapter he is introduced to technical considerations of yields and prices, of patents and secret processes. He will learn how guaiacol came to be introduced into medicine, and why its clinical use is not based on a secure theoretical foundation. As he passes on to antipyretics, hypnotics, antiseptics and organic arsenic compounds the chemist will learn much about pharmacology, disinfection and chemotherapy. The chapter on adrenaline provides an opportunity for discussing the relationship between physiological action and chemical constitution; those on phosphatides and on nucleic acid illustrate the application of organic chemistry to biological problems. On reading through these theoretical chapters a student, whether he be primarily interested in organic chemistry, or in pharmacology, or in manufacture, will have his outlook widened, his interest in cognate subjects stimulated. The practical section of the book is no less attractive; even veterans in organic research will be interested in M. Fourneau's advice to beginners, and might profit by close attention to his injunctions.

Why is this book so attractive? Because it is not an ordinary text-book. Its author was not constrained by the need of being complete; he has written only of what interested him most, of what he had most experience, and the writing of it was evidently a labour of love. It is strange that the language in which it was written should have restricted the circulation of such a book in English and American laboratories, but we may hope that this restriction has been removed by the enthusiasm of the translator. The original or the translation should find a place in every laboratory where organic synthesis is practised, and should be in the possession of all who take a scientific interest in drugs, whether they be manufacturers, or pharmacists, or pharmacologists.

GEORGE BARGER.

DEPARTMENT OF MEDICAL CHEMISTRY,
UNIVERSITY OF EDINBURGH.

PREFACE

TO THE FRENCH EDITION

BY

DR. ROUX, Director of the Pasteur Institute

PROPERLY to appreciate the work that I have the honour to present needs some acquaintance with the circumstances in which it was composed. In 1917, by request of the Junta para ampliacone de Estudios, M. Fourneau went to Madrid to arrange and direct a course of theoretical and practical instruction in the synthesis of the principal organic medicaments. The present volume contains his lectures and the practical training that he organised, with the valuable help of Professor Madinaveitia, in Professor Carracido's laboratory.

M. Fourneau had but a short time at his disposal (three months), so an exhaustive summary of our knowledge of the properties and applications of pharmaceutical products or of methods for their preparation must not be looked for here. Several important groups of medicinal chemicals—anthelmintics, purgatives, etc.—are not even mentioned. Moreover, the book is not planned to fit any official syllabus, and shows no signs of being written to satisfy an existing demand. All the same, the scheme of instruction contained in it makes it a most valuable book.

Some chapters are very complete summaries of our knowledge along certain lines—for example, those on arsenicals, phosphatides and adrenaline.

Above all, it is a programme. Research in chemotherapy is of little renown in our country. Most new medicaments that we use are of foreign origin. This comes about partly because we have few chemists who have specialised in drug synthesis, and further, because no instruction in the subject is given in either technical schools or universities. M. Fourneau is a recognised missionary convinced of the need for making France no longer dependent on the foreigner for important pharmaceutical products. Indeed, he thinks that chemotherapeutical research is just the kind which should best develop here. Such a book as this will do much to encourage our

young chemists to enter the field in which the inventor of stovaine has worked for so many years.

In conclusion, it should be pointed out that preparative experiments in manuals of practical organic chemistry usually bear no relation to one another; for that reason students often lose interest. The second part of M. Fourneau's book is really very interesting, because there the preparation of any particular compound is a step towards a definite end, namely, the synthesis of a drug. There are included, nevertheless, examples illustrating most of the fundamental reactions of organic chemistry. M. Fourneau's experience in Madrid of the great interest shown by students in practical work thus organised leads us to hope that in France, in university chemistry departments, and particularly in schools of pharmacy, courses of lectures and practical work will be organised in the same fashion even if this means engaging the services of outside teachers.

DR. ROUX.

TRANSLATOR'S NOTE

M. FOURNEAU cordially agreed to the preparation of this translation. With his sanction and help, for which I am much indebted, a number of corrections and minor alterations have been made, some notes on and references to recent developments have been added, and a little bibliography has been appended. The latter is intended only to suggest where further information and references to the original literature may readily be found. Trade names, except when frequently used, are in italics.

I am very grateful to Professor Barger for encouraging me, and for contributing the preface to the English edition. Mr. T. R. Parsons has helped me with such sections as deal with physiological questions, and he and Mr. E. J. Amies have read the proofs. To these gentlemen also my best thanks are due.

W. A. SILVESTER.

CRUMPSALL.

CONTENTS

PART I—DESCRIPTIVE

CHAPTER		PAGE
I.	GUAIACOL AND PHENACETINE	1
II.	GUAIACOL AND PHENACETINE (*continued*)	8
III.	ANTIPYRETICS	15
IV.	PREPARATION OF ANTIPYRINE	33
V.	HYPNOTICS	41
VI.	LOCAL ANÆSTHETICS	55
VII.	ANTISEPTICS	70
VIII.	ORGANIC COMPOUNDS OF ARSENIC	82
IX.	ORGANIC COMPOUNDS OF MERCURY	108
X.	ADRENALINE	117
XI.	PHOSPHATIDES	136
XII.	NUCLEIC ACIDS	153
XIII.	ALKALOIDS	161
XIV.	GENERAL REMARKS ON PHARMACEUTICAL PRODUCTS	172

PART II—PRACTICAL

Setting up Apparatus—Recommendations to Beginners		179
PREPARATION of Guaiacol (from *o*-nitrophenol) [1]		192
,,	Phenacetine (from phenol or *p*-nitrophenol) by three methods)	197
,,	Acetanilide (from nitrobenzene)	204
,,	Antipyrine (from phenylhydrazine)	206
,,	Acetylsalicylic acid (from salicylic acid)	208
,,	Stovaine (from chloracetone)	210
,,	Ethyl aminobenzoate (from *p*-nitrotoluene)	219
,,	Quietol (from chloracetone)	221
,,	Acetophenone (Friedel and Crafts's Method)	223
,,	Bromovalerylurea (from valeric acid)	224
,,	Diethylbarbituric acid and of Bromodiethylacetylurea (from ethyl malonate)	226

[1] The parent compound indicated is in each case the subject of the first preparation of the series.

PAGE

ADRENALINE :

Preparation of Adrenaline from the suprarenal glands of the horse 230

Synthesis (from catechol), in part 230

MERCURIALS :

PREPARATION of "Mercury benzoate" 232

,, Mercury diaminodihydroxydiphenyl (from *p*-nitrophenol) 232

ARSENICALS :

Preparation of Salvarsan (from arsanilic acid). 235

PREPARATION of Tyramine (from benzyl cyanide) 237

,, Calcium glycerophosphate 240

,, Lecithin from egg-yolk 241

NUCLEIC ACIDS :

Preparation from yeast and from calf-thymus 243

ALKALOIDS AND GLUCOSIDES :

Determination of the Nicotine-content of tobacco . . . 244

Preparation of Atropine from belladonna root 244

Oxidation of Quinine to quitenine 245

Preparation of Diacetylmorphine hydrochloride . . . 246

Preparation of Digitaline from digitalis 246

PREPARATION of Betaine hydrochloride (from chloracetic acid) . 248

,, Sodium cinnamate (from benzaldehyde) . . 251

,, Allylthiourea (from allyl iodide) 254

,, *p*-Iodoanisole 255

BIBLIOGRAPHY 256

INDEX 259

PART I—DESCRIPTIVE

CHAPTER I

GUAIACOL AND PHENACETINE

We will begin with a study of *Guaiacol* and *Phenacetine*. These two substances are related because their manufacture starts with the two nitro-compounds produced when phenol is nitrated, namely, *ortho-* and *para*-nitrophenol.

From o-*nitrophenol* guaiacol may be prepared thus:

(i) Treat it with a methylating agent and so produce *nitroanisole*,

(ii) Reduce the nitroanisole to o-*anisidine*,

(iii) Diazotise the anisidine by treatment in sulphuric acid solution with sodium nitrite,

(iv) Decompose the diazonium compound by heating it with dilute sulphuric acid or copper sulphate solution and so produce *guaiacol*.

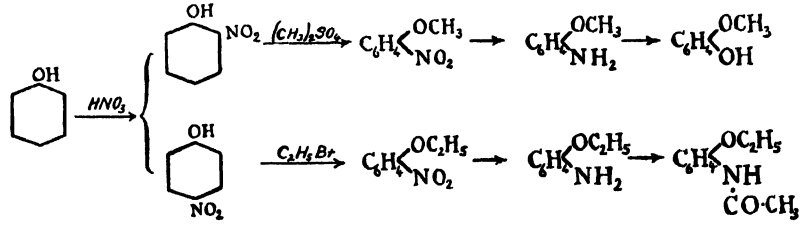
From p-*nitrophenol* phenacetine may be prepared thus:

(i) Treat with an ethylating agent to give p-*nitrophenetole*,

(ii) Reduce the nitrophenetole to *phenetidine*,

(iii) Acetylate the phenetidine and obtain *phenacetine*.

These transformations are represented thus:



This series of reactions is actually used in industrial practice. Nevertheless, the pre-war price of guaiacol was 9*s*. to 10*s*. and that of phenacetine 6*s*. to 7*s*. per kilo., and so it is easily seen that the yields at each stage must have been excellent if a loss instead of a profit were not to result from the manufacture.

Now, right at the start the nitration of phenol to the mononitro stage cannot be made quantitative, no matter how the operation be conducted. In the laboratory, for instance, 1,000 gm. phenol will give 500 gm. *ortho-* and 500 gm. *para*-nitrophenol, instead of 1,470 gm.

in all. Although since methyl sulphate became an industrial product the methylation of *o*-nitrophenol may be conducted in the cold with an excellent yield, yet later on certain difficulties are met with in decomposing the diazotised anisidine. Except for descriptions in the patent literature of two methods, neither of which gives, actually, even when implicitly followed, more than a mediocre yield, the last stage in the manufacture of guaiacol has been kept secret.

So, to prepare the various intermediate products needed for manufacturing guaiacol and phenacetine other methods than those just outlined had to be devised; indeed, alternatives were all the more necessary because the original processes were covered by patents.

The case of guaiacol will first be considered.

GUAIACOL

It has already been shown that the stages in the manufacture of this compound are *o-nitrophenol, nitroanisole, anisidine.* Let us first see how these substances may be obtained in other ways, and afterwards we can discuss the possibility of preparing guaiacol by methylating catechol.

o-Nitrophenol may be prepared by the following methods as alternatives to the nitration of phenol.

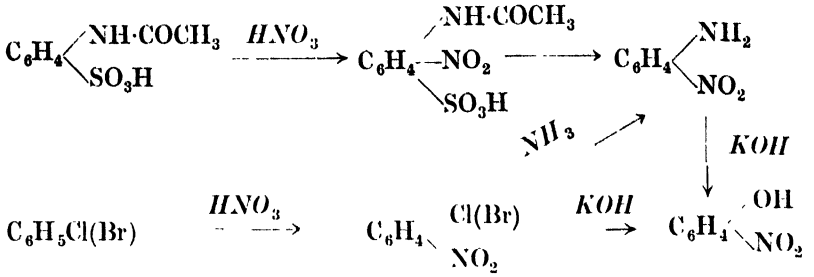
Preparation of *o*-Nitrophenol.—(i) When *o*-nitraniline is treated with caustic potash, *o*-nitrophenol is produced. Now *o*-nitraniline is the least easily obtained of the three isomerides, yet there are two methods by which it may be prepared. In the first of these, acetyl sulphanilic acid is nitrated, whereupon the nitro-group enters in the *ortho-* position to the acetylamino radical, and when the product is heated with a mineral acid the sulphonic group is split off and *o*-nitraniline formed.

In practice there is no need to isolate the acetyl-sulphanilic acid. Acetanilide (50 gm.) is heated with fuming sulphuric acid (20% SO_3—150 gm.) for half an hour at 100°. Sulphuric acid (92%—about 100 gm.) is added and the cooled mixture treated with nitric acid (63%—37 gm.). It is then poured into water (140 c.c.) and boiled for half an hour to split off the acetyl and sulphonic groups. By adding water in small quantities the nitraniline is precipitated, and at a certain stage separates in a nearly pure condition.

(ii) A second method is based on the fact that *o*-chloronitrobenzene yields *o*-nitraniline when heated with ammonia under pressure.

But *o*-nitraniline is not alone in giving *o*-nitrophenol when treated with caustic potash. *o*-Dinitrobenzene and both chloro- and bromonitrobenzenes lose one nitro-group or a halogen atom respectively when similarly treated, likewise producing *o*-nitrophenol.

These reactions are summarised thus :



Rules of Nitration.—A short digression may profitably be made here as the above reactions present several points of general interest. In the first place the rules of nitration may be reviewed.

The radicals ·CH_3, ·CH_2R, ·CH_2Cl, ·Cl, ·Br, ·I, ·OH, ·OR, ·NHCOR, and others, direct the entering nitro-group into the *ortho-* and *para-* positions, whilst the groups ·CHO, ·CO_2H(R), ·COR, ·CH_2SO_3H, ·NR_2, ·CCl_3 and ·NO_2 have a *meta*-directive influence.

Thus, when chloro- or bromobenzene is nitrated, the *o-* and *p-* derivatives are produced, there being formed about 1 part of *o-* to 2 of *p-* at low temperatures, whilst at higher temperatures the proportion of *ortho-* derivative increases.

Chlorobenzene is manufactured in large quantities, and its net cost is barely double that of benzene, as chlorine costs only a penny or so the kilo. on the works.[1] As the nitration is simple and furnishes two products both in industrial use, these, the *o-* and *p-* chloronitrobenzenes, are likewise cheap and can be most advantageously used.

To attain the same end one cannot chlorinate nitrobenzene, although the latter is so easily made on the large scale, for in this case, as we have already seen, the principal product is *meta*-chloronitrobenzene. But when nitrobenzene is nitrated further, besides *m*-dinitrobenzene, which is the chief product, small quantities of the *ortho-* and *para-* isomerides are formed; these may be separated from the mother liquors in a large scale preparation and used for making *o-* and *p-* nitrophenols.

This, a typical example, shows how difficult it was formerly to compete with the great dyestuff manufacturing firms, who often prepared from valueless by-products pharmaceutical chemicals which could not be made independently except at a prohibitive cost.

One need only refer to a pre-war catalogue of "fine chemicals" and compare the price of *m*-dinitrobenzene with that of its isomerides, to be impressed with the difficulty of making the latter. When the *meta-* cost 6*s.* per lb., the *o-* and *p*-dinitrobenzenes cost about £12 per lb.

To return to the reactions by which nitrophenols are produced; these also are of general interest :

[1] Pre-war. In 1924 about 2*d.* per lb.

Reactivity of Halogens and Nitro-groups when present together as Substituents in the Benzene Nucleus. -In monosubstituted derivatives of benzene, such as nitro- and chlorobenzene, the halogen and the nitroxyl radical are very firmly attached, so that they resist the most energetic reagents unless these are aided by such catalysts as copper or magnesium. But if a second nitro-group be introduced, the conditions are completely changed and the halogen, or one of the nitro-groups, may become mobile. Thus, *o*- or *p*-chloronitrobenzene and *o*- or *p*-dinitrobenzene react with ammonia, sodium methoxide, or caustic soda more or less readily, and the chlorine in the former or one of the nitro-groups in the latter is replaced by $\cdot NH_2$, $\cdot OCH_3$ or $\cdot OH$ respectively (just as in the aliphatic series). Further introduction of nitro-groups renders the halogen still more reactive so that chlorotrinitrobenzene (picryl chloride) behaves exactly like a true acid chloride.

The two noteworthy observations to be made here are: (1) The mobility of the halogen (or nitro-group) is limited to the *ortho-* and *para-* derivatives; *m*-chloronitrobenzene and *m*-dinitrobenzene are quite inert towards alkalis and ammonia.[1] (2) In these reactions chlorine is the most mobile of the three halogens, bromine next and iodine least. This is the exact opposite of what takes place in the aliphatic series, particularly in the Grignard reaction.

Other Procedures for making Nitrophenol. There remain to be noted two curious ways of preparing nitrophenol, starting from nitrobenzene. These processes have been patented, but it is doubtful whether the first has ever been much used, because it can result in serious explosions. It is as follows: a mixture of nitrobenzene and caustic potash, dried and finely powdered, is heated at 70°. *Ortho*-nitrophenol is formed. Thus, from 20 gm. nitrobenzene 6 gm. of nitrophenol may be obtained and 10 gm. nitrobenzene recovered unchanged.

According to the second specification, benzene is nitrated in presence of mercury. For example, 400 gm. benzene with 50 gm. mercuric nitrate and 625 gm. nitric acid (sp. gr. 1·39) yields 200 gm. *o*-nitrophenol.

Most of the processes by which *o*-nitrophenol may be obtained have now been reviewed.

Nitroanisole.—The preparation of *o*-nitroanisole from other compounds than *o*-nitrophenol may next be considered.

When *o*-dinitrobenzene, *o*-chloro- or *o*-bromonitrobenzene are heated with sodium methoxide in methyl alcoholic solution, nitroanisole is produced. There is no need to discuss these reactions at

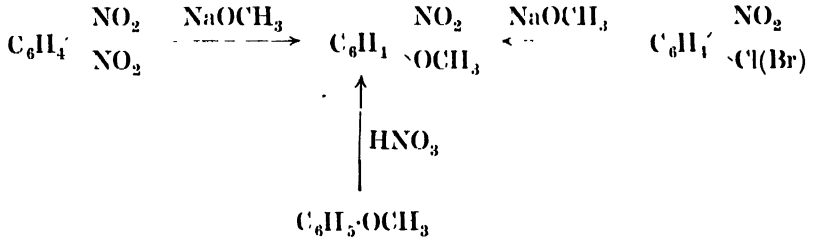
[1] This again is one of the numerous cases in which the close relationship between the *ortho-* and *para-* positions and the special place occupied by the *meta-* is shown. It is difficult to find an explanation of these relationships if benzene has the structure usually attributed to it, viz., with three double bonds. (The original Kekulé formula is more frequently used in France than in England.– *Tr.*)

length; they run parallel to those above, sodium methoxide being used here instead of the hydroxide.

Finally, there remains the preparation of nitroanisole by nitrating anisole.

The nitration of anisole yields a mixture or *para*- and *ortho*-nitroanisole, in agreement with the rule quoted above. Now, although *o*-nitroanisole is used in large quantities for making dianisidine as well as guaiacol, there is practically no outlet for the *para*-isomeride. We must therefore look for a process which will give *o*-nitroanisole almost exclusively. There is, in fact, one, widely used in laboratory work but not known to be applied in industry. The anisole is treated at a low temperature with acetyl nitrate (obtained by distilling *in vacuo* a mixture of 100% nitric acid and acetic anhydride). 90% of the theoretical yield of *o*-nitroanisole has been obtained in this way.

The reactions just discussed may be displayed thus:



Nitroanisole must now be reduced to anisidine and the anisidine must be diazotised and converted into guaiacol.

The **reduction** needs no particular comment; it may be carried out by any method applicable to nitrobenzene to produce aniline. But of all the stages in the production of guaiacol, the **diazotisation** and subsequent treatment of the anisidine need the most care and attention. Usually, simple heating with water will transform a diazonium salt into the corresponding phenol, but occasionally a higher temperature is necessary. This is the case with the diazotised anisidine.

So as to be able to heat to the required degree and at the same time protect the diazonium compound against side-reactions, the solution in which it is treated consists of 60 per cent. sulphuric acid saturated with sodium sulphate.

A still better method is to use a concentrated solution of copper sulphate, since copper has a catalytic action in all the reactions of diazonium compounds. The details of this latter process will be found in Part II. (the practical section of the book).

Methylation of Catechol.—There is an alternative to the method just described in which anisidine is the intermediate, which is theoretically simpler and, indeed, would be the first to come to mind. This consists in *methylating catechol*, guaiacol being simply catechol monomethyl ether.

To methylate catechol is easy—too easy, in fact, because the reaction goes too far and gives the dimethyl ether, *veratrole.*

If the calculated proportions of catechol and methylating agent for monomethylation are caused to react, the product actually has the composition :

Guaiacol	20 parts
Veratrole	45 ,,
Catechol recovered	40 ,,

In consequence, attempts have been made to demethylate veratrole, and several processes for doing so have been patented. In some, the demethylation is effected by caustic soda or by hydrochloric or hydrobromic acid at a high temperature in an autoclave ; in others, the veratrole is treated with aluminium chloride.

But here again an equilibrium mixture is formed and part of the veratrole is completely demethylated to give catechol. This is the case, at any rate, if the patent specifications are to be believed ; but as these processes are actually used by certain manufacturers, one must suppose that they have special skill and can make them practicable.

As catechol could be made for little over 3*s.* 6*d.* per kilo., its transformation into guaiacol was always an interesting problem.

This discussion of guaiacol may be brought to an end by a cursory survey of the methods for producing **catechol,** which are as follows :

(i) The fusion of ***o*-benzenedisulphonic** acid with caustic soda. This procedure is hardly feasible because the *ortho*-disulphonic acid is not easily obtained, and, moreover, when fused with caustic soda yields a large proportion of resorcinol.

(ii) Caustic soda fusion of **phenolmono-, di-** or **trisulphonic acid.** In the two latter cases there are formed catechol mono- or disulphonic acids from which the sulphonic groups are removed by treatment with steam.

The following is the way in which the cost of manufacturing catechol by one of these methods may be reckoned :

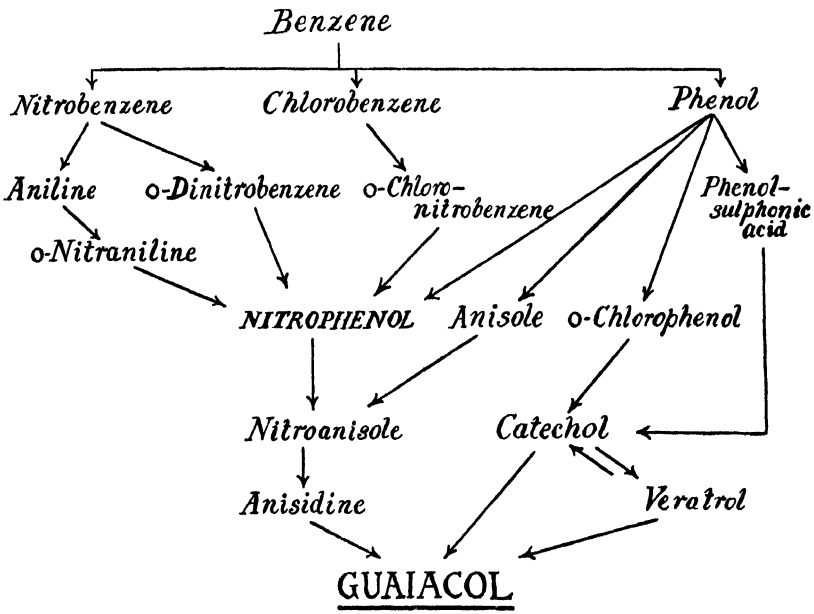
		Fr.
100 kg.	carbolic acid	100
270 ,,	"oleum" at 60% SO_3 . . .	32
120 ,,	sodium carbonate (soda ash) . .	15
600 ,,	caustic soda	175
1,300 ,,	sulphuric acid (B.O.V.) . . .	100
		422

The yield is 60 kg. catechol, so the cost is 7·03 fr. per kilo,[1] and if the manufacturer himself makes his raw materials, this can be considerably reduced.

[1] All prices are pre-war. (In England, costs are usually reckoned on a pence per *pound* basis.—*Tr.*)

(iii) *o*-**Chlorophenol,** which is easily obtained by chlorinating phenol, is heated with caustic soda. This process is probably the most advantageous. According to DRP 269554, if 26 gm. *o*-chlorophenol, 130 c.c. 5N caustic soda, and a trace of copper sulphate be heated at 190°, the yield is 83% of that calculated by theory.

The most important methods of preparing guaiacol have now been described. All can be put to industrial use, but for our laboratory work we must choose the most characteristic, viz., those in which phenol and bromobenzene are the primary materials. A full description of the first and of certain stages in the second of these methods will be found in Part II.



CHAPTER II

GUAIACOL AND PHENACETINE—*continued*

Use and Derivatives of Guaiacol.—Guaiacol is an important industrial product. It is used for making vanillin and is applied in medicine to the treatment of pulmonary consumption.

Guaiacol occurs in considerable amount in creosote (hardwood tar) mixed with other phenols, notably the methyl homologue, creosol.

The employment of guaiacol in medicine was preceded by that of creosote, which seems to have been most widely used in France during the period 1885–89. The creosote was administered either in cod-liver oil, or in wine, or more often in pills containing iodoform and tolu-balsam.

In 1887 the creosote treatment was spreading into Germany under Sommerbrodt's influence when MM. Béhal and Choay succeeded in isolating guaiacol in a crystalline form for the first time. Since then guaiacol has largely replaced creosote, mainly because of its absolute purity and much less disagreeable smell, but, apparently, for no definite therapeutical reason.

The manner in which creosote and guaiacol act in cases of consumption is not known. It is not even known if they have any action at all. Certainly *guaiacol is an antiseptic*, but it cannot be introduced into the organism in quantities sufficient to raise the concentration in the blood and tissues to a degree fatal to tuberculosis bacilli. And, like all phenols, it is mostly eliminated as a sulphuric ester, that is to say, in a form possessing no antiseptic action.

Moreover, many renowned specialists on diseases of the respiratory tract deny, rightly or wrongly, that guaiacol has any effect. In their opinion, the treatment of consumption should be almost entirely hygienic : pure air, appropriate diet, physical and mental tranquillity, some phosphates and chalk, and a little cryogenine to combat fever at night, such should be the consumptive patient's regimen.

All the same, the antiseptic action of guaiacol ought not to be quite negligible, upon digestive processes in the stomach and intestine at any rate. Possibly it aids secretion and expectoration to some extent. In actual fact, the guaiacol treatment, if wisely controlled, is of proved value in the early stages of consumption ; the patients feel generally livelier and have a healthier appetite.

Derivatives of Guaiacol.—The irritating action of phenol on mucous membranes is naturally also possessed by guaiacol, and although its taste and odour are not very offensive, they cannot but annoy patients

in time. Attempts have therefore been made to mask the one or the other in various ways, all of which really come to the same thing, namely, esterification of the hydroxyl group. Another reason for esterifying is that the guaiacol in that form will act only after it has reached the intestine. Endeavours have also been made to prepare soluble derivatives so as to facilitate application in pharmacy. Some of these derivatives of guaiacol, notably guaiacol carbonate and potassium guaiacolsulphonate (*Thiocol*), are now commercially very important. These two compounds, particularly, are typical of the two classes just mentioned, but there is an abundance of others, and it is interesting to glance over as many examples as possible and so appreciate the ingenuity of the pharmaceutical manufacturers.

Of **insoluble derivatives,** guaiacol carbonate or *Duotal* has the formula $(CH_3O\cdot C_6H_4\cdot O)_2CO$ and is prepared by treating a solution of guaiacol in caustic soda with phosgene. *Monotal* is guaiacol methylglycollate: $CH_3O\cdot C_6H_4\cdot O\cdot CO\cdot CH_2OCH_3$. *Benzosol* is guaiacol benzoate: $CH_3O\cdot C_6H_4\cdot O\cdot CO\cdot C_6H_5$. Their formulæ show how these latter are prepared. Other derivatives may be mentioned, such as *Geosote* or the valeric ester, the oleic ester and the phosphite, *Guaiacophosphal.*

The most important **soluble derivatives** are *Guaiasanol* and *Thiocol. Guaiasanol* is diethylaminoacetylguaiacol hydrochloride:

$$C_6H_4(OCH_3)\cdot O\cdot CO\cdot CH_2N(C_2H_5)_2\cdot HCl.$$

Einhorn was the first to prepare this compound, using a novel method of rendering organic medicaments soluble, by which many amino-derivatives may be obtained.

The process is as follows. Guaiacol is treated with chloracetyl chloride, or better with chloracetic acid in presence of pyridine and phosphorus trichloride:

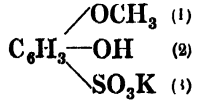
$$CH_3O\cdot C_6H_4\cdot OH + CH_2Cl\cdot COCl \rightarrow CH_3O\cdot C_6H_4\cdot O\cdot CO\cdot CH_2Cl + HCl.$$

The chloroacetyl derivative of guaiacol is thus formed and is then treated, in the cold, with diethylamine:

$$CH_3O\cdot C_6H_4\cdot O\cdot CO\cdot CH_2Cl + NH(C_2H_5)_2 \rightarrow CH_3O\cdot C_6H_4\cdot O\cdot CO\cdot CH_2N(C_2H_5)_2\cdot HCl.$$

Guaiasanol, as hydrochloride soluble in water, is precipitated as base from its aqueous solution by alkali carbonates. It is not very poisonous, but is seldom used, probably because of its high price.

After the carbonate, *Thiocol* is the most widely employed derivative of guaiacol. This is the potassium salt of guaiacol-*o*-sulphonic acid:



It is prepared by treating guaiacol with its own weight of concentrated sulphuric acid at temperatures below 80°. If this temperature limit be passed, the *para*-isomeride is produced. When the reaction is complete the mixture is diluted with water and neutralised with barium carbonate. After being filtered the solution containing barium guaiacolsulphonate is treated with potassium carbonate until no more precipitate is formed. It is again filtered and concentrated by evaporation to get out the Thiocol.

Thiocol is freely soluble in water. It is not decomposed in the organism and shows no appreciable antiseptic activity. Theoretically it should not have the least efficacy, and yet there are few medicaments with so considerable a market.

PHENACETINE

The steps taken in preparing *Phenacetine* by one method have already been outlined. In brief, they are as follows: *p*-nitrophenetole is made from *p*-nitrophenol and reduced to phenetidine, which, acetylated, forms phenacetine.

As in the case of guaiacol, we will review the various industrial methods of producing these various intermediates, but since derivatives of *p*-nitrophenol may be obtained by reactions resembling those which lead to the formation of the *ortho*-isomerides, our survey here need not be so detailed.

Nitrophenol.—*p*-Nitraniline is heated with caustic soda solution; the NH_2 group is replaced by OH and so *p*-nitrophenol is formed. *p* Nitraniline is an industrial product, easily made by nitrating acetanilide and subjecting the acetylnitraniline so formed to hydrolysis.

Similarly, *p*-chloro- and *p*-bromonitrobenzene are converted into nitrophenol when heated with caustic soda.

Although when chlorobenzene is nitrated a mixture of the two isomerides, *ortho*- and *para*-, is formed, for the above purpose it is not necessary to effect a separation at this stage. Each compound gives the corresponding nitrophenol when treated with caustic soda. The mixture can therefore be brought into reaction as such and the two components isolated from the product very easily by steam-distillation, as already described.

p-Nitrophenol may also be prepared from *p*-nitraniline by another method, namely, *viâ* the diazonium compound. A solution of the nitraniline in dilute sulphuric acid is treated with sodium nitrite and then boiled, nitrogen is evolved and the nitrophenol thrown down.

Nitrophenetole.—The general methods employed for preparing nitroanisole serve also for nitrophenetole, but in this case their application is not so practicable. Thus, *p*-dinitrobenzene and

p-chloronitrobenzene react with sodium ethoxide to give nitrophenetole :

$$C_6H_4\begin{matrix}NO_2\\NO_2(Cl)\end{matrix} + NaOC_2H_5 \dashrightarrow C_6H_4\begin{matrix}NO_2\\OC_2H_5\end{matrix} + NaNO_2(Cl).$$

But a great excess of alcohol must be used or, *e.g.*, chloronitrobenzene will give a considerable amount of dichloroazoxybenzene. In this side-reaction sodium ethoxide plays the part of a reducing agent and is itself oxidised to acetate :

$$2C_6H_4Cl\cdot NO_2 \dashrightarrow Cl\cdot C_6H_4\cdot N\!\!<\!\!\begin{matrix}\\O\end{matrix}\!\!>\!\!N\cdot C_6H_4Cl.$$

Yet another interesting method of preparing nitrophenetole has been patented. Phenyl toluene-sulphonate, produced by treating phenol in alkaline solution with toluene-sulphonic chloride, is nitrated ; the nitro-group enters the *para*- position in the phenyl radical and *p*-nitrophenyl toluene-sulphonate is obtained. This interacts with sodium ethoxide to give nitrophenetole and sodium toluene-sulphonate :

$$C_6H_5\cdot OH + CH_3\cdot C_6H_4\cdot SO_2Cl \dashrightarrow C_6H_5\cdot OSO_2\cdot C_6H_4\cdot CH_3$$

$$\xrightarrow{HNO_3} NO_2\cdot C_6H_4\cdot OSO_2\cdot C_6H_4\cdot CH_3$$

$$\xrightarrow{NaOC_2H_5} NO_2\cdot C_6H_4\cdot OC_2H_5 + CH_3\cdot C_6H_4\cdot SO_3Na.$$

Phenetidine.—Phenetidine may be obtained in other ways than by reducing nitrophenetole. The alternatives are of two kinds, viz. :

(i) Ethylation of *p*-aminophenol, or better, of its acetyl or benzylidene derivative ;

(ii) Ethylation, followed by reduction, of *p*-hydroxyazobenzene (or *p*-hydroxy-*p'*-ethoxyazobenzene) obtained by "coupling" diazotised aniline (or phenetidine) with phenol.

For the first of these, *p*-aminophenol must be prepared.

***p*-Aminophenol.**—This aminophenol is largely used in industry, and so the principal ways of making it will be outlined. The simplest method is to reduce *p*-nitrophenol, using sodium sulphide ; or tin or iron and hydrochloric acid ; zinc dust and caustic soda ; hydrosulphite[1] ; or ferrous sulphate and ammonia. There is also Wislicenus's method of reduction, now coming into considerable use, in which the active agent is aluminium amalgam.

To prepare **aluminium amalgam,** aluminium borings are treated with caustic soda until hydrogen is briskly evolved ; they are then quickly washed, immersed in a 5 per cent. solution of mercuric chloride (corrosive sublimate) for a few minutes, again washed with water, then with alcohol and finally with ether.

[1] Throughout the book the incorrect but well established name *sodium hydrosulphite* is used instead of the orthodox *sodium hyposulphite* for the compound $Na_2S_2O_4$. The latter name is rarely found in technical literature.—*Tr.*

Nitrophenol is reduced by this method as follows. Half its weight of aluminium amalgam is added to a cold solution of the compound in 50 per cent. alcohol. The mixture is stirred for an hour, filtered, and the filtrate evaporated to dryness in a current of carbon dioxide.

This procedure may be recommended for any unstable compound to be obtained by reduction in neutral solution.[1]

Aminophenol may also be produced directly from nitrobenzene by two methods, both in industrial use and both of considerable theoretical interest. These are, namely, (1) electrolytic reduction of nitrobenzene in strong sulphuric acid solution with platinum electrodes, and (2) reduction by zinc and strong sulphuric acid. Here two unexpected reactions take place: the first product, it appears, is phenylhydroxylamine,

$$C_6H_5{\cdot}NO_2 \longrightarrow C_6H_5{\cdot}NH{\cdot}OH$$

which then undergoes a rearrangement into *p*-aminophenol,

$$C_6H_4\begin{cases}NH_2\\OH.\end{cases}$$

Aminophenol may also be obtained by oxidising sulphanilic acid with manganese dioxide and sulphuric acid.

$$C_6H_4\begin{cases}NH_2\\SO_3H\end{cases} + O + H_2O \longrightarrow C_6H_4\begin{cases}NH_2\\OH\end{cases} + H_2SO_4.$$

Diazobenzene (sulphate) will couple with phenol in alkaline solution giving quantitatively *hydroxyazobenzene* (benzene-azo-phenol)

$$C_6H_5{\cdot}N_2{\cdot}SO_4H \mid C_6H_5{\cdot}OH \longrightarrow C_6H_5{\cdot}N:N{\cdot}C_6H_4{\cdot}OH + H_2SO_4$$

and when the product is reduced (by, *e.g.*, sodium hydrosulphite) aminophenol is produced and aniline regenerated.

$$C_6H_5{\cdot}N:N{\cdot}C_6H_4{\cdot}OH + 2H_2 \longrightarrow C_6H_5{\cdot}NH_2 + C_6H_4\begin{cases}NH_2\\OH.\end{cases}$$

Finally, acetyl-*p*-phenylenediamine may be diazotised and the diazo compound boiled with water to give acetyl-*p*-aminophenol:

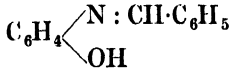
$$C_6H_4\begin{cases}NH{\cdot}CO{\cdot}CH_3\\NH_2\end{cases} \longrightarrow C_6H_4\begin{cases}NH{\cdot}CO{\cdot}CH_3\\N:N{\cdot}SO_4H\end{cases} \longrightarrow C_6H_4\begin{cases}NH{\cdot}COCH_3\\OH.\end{cases}$$

There are thus a number of ways by which *p*-aminophenol may be prepared.

p-Aminophenol is a widely used product. Besides serving as an intermediate in the manufacture of phenacetine, it is employed in fur-dyeing to give brown shades and, under the trade name, ***Rodinal***, as a photographic developer. Two derivatives, namely, *p*-hydroxyphenylglycine, $HO{\cdot}C_6H_4{\cdot}NH{\cdot}CH_2{\cdot}COOH$, known as *Glycine*, and the N-monomethyl compound, $CH_3{\cdot}NH{\cdot}C_6H_4{\cdot}OH$, known as *Metol*, are also used as photographic developers.

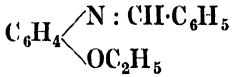
[1] Ferrous hydroxide is also an excellent reducing agent for nitrophenol.

Phenetidine.—When the hydroxyl group in *p*-aminophenol is ethylated, phenetidine results; but ethylation cannot be directly applied to the free aminophenol because the amino-group also is attacked. This group, therefore, must first be protected. This may be done either by acetylation, or by condensation with benzaldehyde in weakly alkaline solution; the latter reaction produces quantitatively *benzylidene aminophenol:*

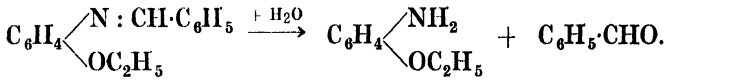


(a "Schiff's base").

This compound is unaffected by weak alkali and may be ethylated by treatment with ethyl bromide in alkaline alcoholic solution. The product, *benzylidene aminophenetole,*

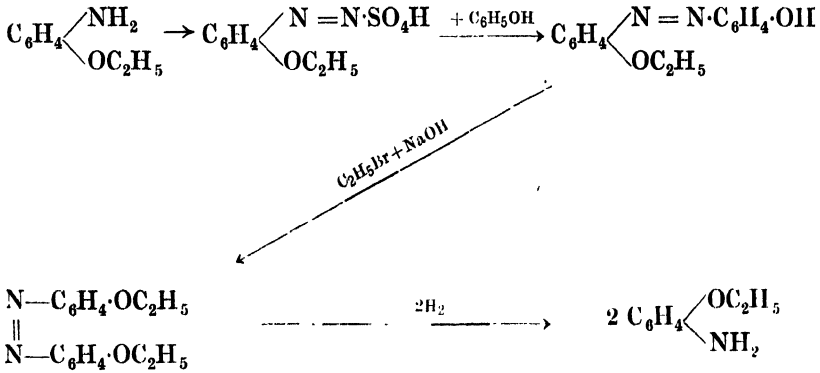


by being simply warmed with dilute hydrochloric acid, is split up again, giving phenetidine and regenerating benzaldehyde:



The last method on our list is the most elegant of all, because, except for a certain amount of phenetidine which is kept in use, the only organic raw materials needed are phenol and ethyl bromide.

The phenetidine, dissolved in dilute sulphuric acid, is diazotised with sodium nitrite and added to a solution of phenol containing an excess of sodium carbonate. *p*-Ethoxy-*p'*-hydroxyazobenzene (*p*-phenetole-azo-phenol) is formed and is then ethylated further by treatment with ethyl bromide, giving *diethoxyazobenzene* (*p*-azophenetole), which, when reduced, yields two molecular proportions of phenetidine. One of these is acetylated to make phenacetine; the other serves again in the cycle of operations:

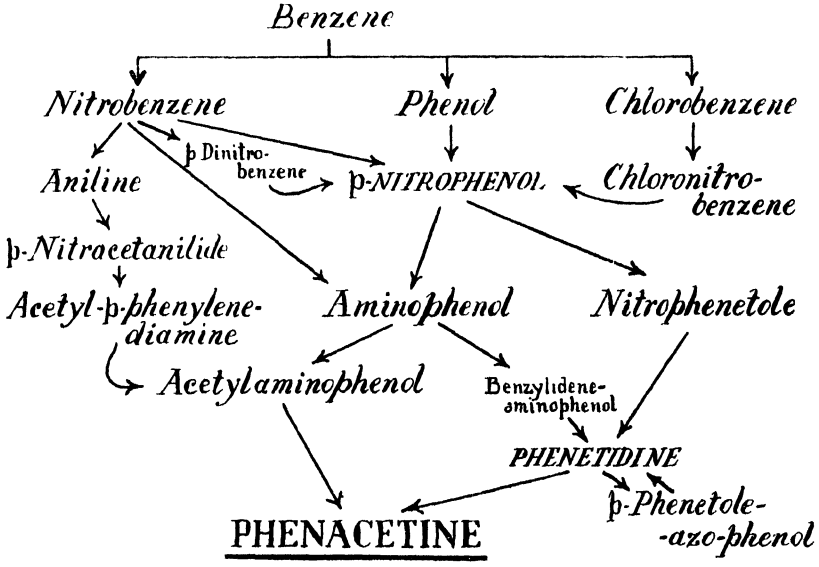


This "**coupling**" of diazo-compounds with phenols is of outstanding importance in the dyestuff industry. It is also of considerable theoretical interest. Two noteworthy rules hold :

(1) Coupling only takes place when the *ortho-* or *para-* position to the hydroxyl group is free ;

(2) Coupling always takes place in the *para-* position unless it be occupied ; otherwise, in the *ortho-*.

Little need be said about the last stage, namely, **acetylation.** The reaction takes place quantitatively when an aqueous suspension of phenetidine is stirred with acetic anhydride, or when phenetidine and acetic acid are boiled together in a vessel so fitted that the water produced in the reaction may distil off.



CHAPTER III

ANTIPYRETICS

PHARMACOLOGISTS explain the effect of antipyretics on the temperature of those suffering from fever by saying that the heat-regulating centres are acted upon. It cannot, however, be stated precisely where these centres are nor how the action takes place.

When one speaks of the centres regulating animal heat, the underlying idea is hardly of a localisation of the regulating function in an anatomical sense, but of a grouping of systems having the same effect. That such systems do exist is evident from the fact that warm-blooded animals (homoiothermic) keep at their own temperature whatever be that of the surroundings, whilst cold-blooded animals (poikilothermic) change in temperature with their environment. Further, the temperature of warm-blooded animals may be altered artificially, as will be shown later, by operating on the particular area of the brain that controls the physiological mechanism of heat-regulation.

The physiology of the conservation of animal heat is complex and at present that dealing with the blood vessels is the most satisfactorily explored field. The skin, with its network of capillary canals, is an excellent conductor of heat, and so is very sensitive to cold air outside; but a general lowering of temperature is speedily limited by a constriction of the surface blood vessels that takes place all over the body and not merely in the parts subjected to cooling. The result is that less blood circulates in the surface areas of the body; that which returns to the heart is not so cold; and, as the total amount in circulation remains the same, a larger proportion passes through the greater organs and the digestive tract where much heat is being generated. Thus with the blood alone as agent, a fair explanation may be made of the phenomenon of temperature-equilibrium. But there are other factors. External cooling provokes more active oxidation and more material is burnt up in the body—as can be experimentally shown by measuring the carbon dioxide eliminated. And indeed everyone knows that cold weather engenders hunger—and hunger for food with the highest calorific value.

The influence of a hot atmosphere is counteracted by a contrary kind of mechanism: sweat evaporates and cools the blood that rapidly circulates at the surface of the body.

In a normal individual temperature regulation is controlled by the nervous centres; it is, to all appearances, localised in the mid-brain.

In pathological cases, infectious fevers, and so forth, it is supposed that the regulating centres are upset by toxic albuminoid substances generated by the protoplasm and the actual infectious agents.

Experimentally, elevation of body temperature in animals may be brought about by intravenous injection of albumin or bacteria, by thermal or electrical stimulation of certain parts of the brain, or, particularly in the dog and the rabbit, by puncture in the neighbourhood of the *corpus striatum*.[1]

Wood was the first to demonstrate that there are cerebral areas specially devoted to thermo regulation. His conclusions have been confirmed by Richet, Aronsohn and others. These investigators have shown that an injury to the part of the forebrain adjacent to the *corpus striatum* causes a sudden drop in temperature followed almost immediately by a pronounced rise of over 2°. This lesion may be carried out experimentally by a fine trocar, under the conditions described below, or by a special mode of cauterisation (Thermopuncture, Wärmestich). It is outside the scope of this treatise to recount the controversies to which experiments on the thermo-regulating centres have given rise. Isenschmiedt's recent work demonstrates the great importance of the *tuber cinereum*, that is to say, of the region near the pituitary body (hypophysis), but the sympathetic system also plays a part, principally in thermal phenomena of a chemical nature, and it is now agreed that physical thermo-regulation is particularly under the influence of the cervical portion of the spinal cord. Communication between the two systems seems to be carried on by particular hormones.

In the pharmacological study of antipyretics, the puncture near the *corpus striatum* is a most important operation, and so a short description will be given, taken from the most interesting thesis of Sanchis Banus (*Contribución al estudio de los Antipyreticos*, Valencia, 1918, p. 32).

The rabbit is bound, its head being left free until it is anæsthetised (by ether), then also firmly fixed. Its scalp is shaved and treated with iodine solution. The maintenance of perfectly antiseptic conditions is, indeed, absolutely essential. An incision is made to the bone, starting from near the nostrils, a little in front of the eyes, and continuing over to that occipital protuberance particularly prominent in the rabbit. The edges of the incision are retracted and the surface of the skull scraped with a periosteotome until the sutures appear. The junction of the frontoparietal suture with that which crosses it at right angles at the level of the ears should now be visible. In the area thus marked out and in the upper angle anterior to the parietal craniectomy is performed. An opening 6 mm. in diameter is big enough, the thickness of the bone here being about 1·5 mm. If the

[1] The *corpus striatum* is a collection of nerve cells situated at the base of the cerebrum. —*Tr.*

trepanation is carefully performed the surface of the *dura-mater* will be uncovered.

The wound bleeds copiously, but hæmorrhage ceases when the first plug is applied; if it be prolonged the technique has been faulty. The *meninges* being now visible, a sterilised needle is plunged into the middle of the area. The *dura-mater* offers some resistance, which must be carefully overcome so as not to penetrate too far into the brain. The needle should not be held vertically but inclined a little forward so that it points backwards. It is to be inserted to a depth of 5 to 6 mm. and then its point moved forwards so as to destroy a large enough area of the heat-regulating region. When the operation is complete, the wound is stitched up. Anæsthesia is now interrupted and the animal kept at a temperature of 20°.

Thermopuncture, or puncture of the *corpus striatum*, is not the only way of bringing about artificially a state of fever. An equally satisfactory effect results from injecting sterile preparations of certain bacterial toxins.

Thanks to these methods, antipyretics may be studied experimentally, and they are particularly valuable because, curiously enough, the depression in temperature brought about in a normal individual by an injection of antipyretic is hardly comparable in intensity with that observed in an animal in a state of either pathological or artificial fever. The elevation of body temperature in this case causes, in fact, the body to be in much more unstable equilibrium than in a normal subject, and disturbances are easily produced, sometimes by quite small doses of antipyretic.

Thus, and this is the fact to remember, *antipyretics act with much more effect* when the subject is *in a state of fever* of either experimental or infectious origin, than when he or it is in a normal condition.

In brief, then, we may say that antipyretics act on the excited heat-regulating centres and regularise their activity.

One of the ways in which their complicated action is effected is the dilatation of the surface blood-vessels so that more blood is cooled as it circulates. At the same time they act on the sensory and vaso-motor centres of the brain and cause narcosis and analgesia.

Actually all known antipyretics do act to some extent as analgesics and narcotics. With some, such as *Lactophenin* and *Phenacetine*,[1] hypnotic action is very marked; with others, such as *Cryogenin*, antipyretic action alone stands out, almost masking the other effects. Yet the three physiological properties are not indissolubly connected; changes in chemical constitution can make one property predominant and weaken the others, as several of the following examples will show.

An interesting problem for pharmacology is that of passing from a pure antipyretic to a pure narcotic by progressive modifications in chemical constitution.

[1] The addition of a terminal *e* in these trade names seems to depend on whether or no an attempt has been made to anglicise the name.—*Tr.*

That which precedes aims at making more intelligible one of the objects of this chapter, which is, namely, an inquiry into the effect of alterations in chemical composition on antipyretic activity. We shall also want to know in what classes of chemical compounds antipyretics are to be found, which radicals are the active ones, and how the substances are made. These questions will now be answered.

It will be seen at once that antipyretics belong to the aromatic (cyclic) series, and so stand in sharp contrast with most hypnotics. This does not mean that compounds possessing analgesic and antipyretic properties are never to be met with in the aliphatic series; Curtius has stated, for example, that pyrazolones containing no benzenoid nucleus depress the body temperature as readily as Antipyrine, and, moreover, several aliphatic amino-alcohol derivatives are known which act in the same way (Fourneau). Nevertheless, no marketable competitor with Antipyrine, Phenacetine and Aspirin, the three great "protagonists" of antipyretic action, has yet been found in the aliphatic series, and, indeed, Filehne has shown that even among pyrazolones activity is feeble unless there be an aromatic nucleus in the molecule.

The aromatic nucleus may therefore be considered to have a specific influence, and, in a general way, it may be stated that *all oxidisable benzene derivatives*, and their easily hydrolysed compounds, are antipyretics to some extent. The maximum activity is shown by such compounds as produce p-*aminophenol* in their passage through the organism.

A second observation may be made, viz., antipyretics have a comparatively simple basis: aniline, phenylhydrazine, phenol, aminophenol. There are certainly some, for example, *Salophen*, with fairly big molecules, but such as these are rapidly hydrolysed by the organism, and on this hydrolysis their activity depends.

The two primary compounds from which all antipyretics are derived are **aniline** and **phenol.** From aniline originate phenylhydrazine, quinoline, hydroquinoline; from phenol salicylic acid; and from both aniline and phenol the aminophenols and the hydroxy-quinolines.

By simple methods of treatment these substances yield the antipyretics that are now well established and many others that have had but a brief career. Aniline gives acetanilide; phenylhydrazine, *Antipyrine*, *Pyramidon* and *Cryogenin*. From *p*-aminophenol *Phenacetine* is produced; from phenol itself, *Antodyne*, and from salicylic acid, *Aspirin*. These relationships are shown in the following table:

Benzene	Aniline	Acetanilide or	*Antifebrine*
		Phenylhydrazine	*Cryogenin*, *Antipyrine*, *Pyramidon*
	Phenol	*p*-Aminophenol	*Phenacetine*
		Salicylic Acid	*Aspirin*
		Phenyl ether of glycerine	*Antodyne*

Antipyrine, *Aspirin* and *Phenacetine* always stand supreme in practical importance. Then follow *Pyramidon*, *Lactophenin* and *Cryogenin*; the latter is a more recent production, it is the nearest approximation to a pure antipyretic and is becoming more and more important therapeutically because of the mildness and duration of its action.

It should be noted that the first member of a series of compounds to be put on the market has nearly always held the field later. Yet evidently it is not simply that the best product is that which was discovered first. The event depends on many other factors: skill acquired in manufacture, selling price and advertisement, all play a part. And so it often happens that far greater efforts must be made to displace a known product by another of the same family than to put out a new one of a hitherto unexploited series.

Such considerations as these have not discouraged chemists and pharmacologists from planning and, with the help of the great chemical manufacturing firms, making and marketing many derivatives of the better known products, but usually their efforts have been in vain. Another reason for the failure to replace the old product by the new lies in this, that the claims of those who will profit by the replacement are so often exaggerated.

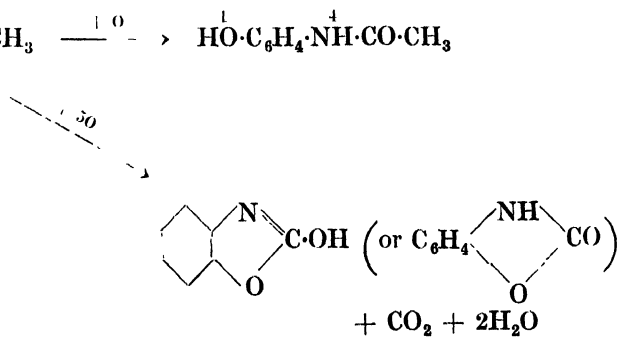
We might, therefore, content ourselves with studying simply the three chief antipyretics, but, for the reasons given above, it is more useful and interesting to review the greatest possible number of such compounds, or, at any rate, those which have actually been in therapeutic use even if only for a short time. Each product has its virtues and its failings: the virtues helped to justify its inventor's hopes; the failings will show us what should be avoided. Doubtless, having collected together items of information in this way, one may some time be able to lay down exact laws in therapeutics.

Our review of compounds proposed or actually utilised as antipyretics may well begin with derivatives of aniline.

Antipyretics derived from Aniline.—Aniline itself acts on the nervous system, causing a fall in body temperature followed by narcosis. It cannot be used therapeutically because it is very toxic and has a definite action on the red corpuscles of the blood. When it is present, hæmoglobin forms with oxygen methæmoglobin, a compound too stable to serve in tissue respiration. The fatal dose (one that will kill rapidly) of aniline is 0·005 gm. per kilo. of body weight.

Substitution in the amino-group brings about a considerable decrease in toxicity. Acetanilide—*Antifebrine*—behaves like aniline, but in a much milder fashion. Its activity partly depends on hydrolysis into aniline and acetic acid, but unless very large doses are administered enough aniline to cause serious results is not liberated. Moreover, the stability of acetanilide has another important conse-

quence in that whilst passing through the organism some of the compound suffers nuclear oxidation to *p*-acetyl-aminophenol, which is much less toxic than aniline. Besides *p*-acetyl-aminophenol, free aminophenol and oxycarbanil (benzoxazolone) are produced. The latter is formed by oxidation of the acetyl group as well as of the nucleus,



p-Aminophenol and oxycarbanil are eventually eliminated in combination with glycuronic or sulphuric acid.

Acetanilide was the first great success of synthetic chemical industry in the medical field. Attempts have been made to replace the acetyl group by other radicals, but no advantage has been gained. Formanilide, for instance, $C_6H_5 \cdot NH \cdot CHO$, being readily hydrolysed in the organism, is very toxic. On the other hand, the anilides of higher homologues of acetic acid are less soluble than acetanilide, and consequently both less toxic and less active. In fact, throughout the series, antipyretic activity and toxicity are proportional to one another.

In this case the most valuable medicament was hit upon at the first attempt.

The three isomeric **acetotoluidides** do not all behave like acetanilide in the organism. The *ortho*- derivative certainly undergoes a similar change, giving $CH_3 \cdot C_6H_3 \langle \begin{matrix} N \\ O \end{matrix} \rangle C \cdot OH$, but has no influence on the body temperature; neither has the *para*- compound, although it suffers oxidation to *p*-acetylaminophenol. The *meta*- isomeride, on the other hand, has a well-marked effect on the temperature, but, nevertheless, is oxidised in a different way, acetylaminobenzoic acid being formed. Here it is difficult to trace a connection between antipyretic action and chemical constitution.

Another class of aniline derivatives contains a free acidic group, as is present, for instance, in **phenylglycocoll,** or phenyglycine, $C_6H_5 \cdot NH \cdot CH_2 \cdot CO_2H$. This compound is prepared by treating aniline with chloracetic acid. It is an important intermediate in synthetic indigo manufacture. All derivatives of this kind are, however,

inactive. One may indeed, state, as a general rule that the introduction of an acidic group into a molecule renders the compound less toxic and completely alters its physiological properties. This is well shown by another example, namely, *Cosaprin*, sodium acetylsulphanilate, $CH_3 \cdot CO \cdot \overset{1}{N}H \cdot C_6H_4 \cdot \overset{4}{S}O_3Na$, which has no antipyretic activity, and was soon abandoned.

The **urethanes** form a special class of amine derivatives. They are substituted alkyl carbamates. The compound represented by the formula: $NH_2 \cdot CO \cdot OC_2H_5$ is ethyl urethane or, simply, urethane. The phenyl derivative of this, $C_6H_5 \cdot NH \cdot CO \cdot OC_2H_5$, is known as *Euphorine*. This product is only slightly toxic, and is as useful a febrifuge as acetanilide, but, like *Exalgine*, $C_6H_5 \cdot N(CH_3)CO \cdot CH_3$, which also was in vogue for a time, it has been displaced by *Phenacetine*, to which we will now turn. But, having referred to Exalgine, it may be noted in passing that this methylation of the amino group in acetanilide produces a more poisonous compound. The surprisingly deleterious effect of simple methylation of amino groups will be observed again later in our study of the arsenic compounds used in therapeutics.

That aniline and its derivatives are, to a great extent, oxidised to aminophenols during their passage through the organism has already been pointed out; they are in this way converted into compounds possessing similar antipyretic properties but a much weaker toxic action. Considerations such as these, however, did not lead to the discovery of **Phenacetine.** This was the result of reasoning of a much more practical kind, and its history makes curious reading.

In 1887, shortly after the therapeutic properties of acetanilide had been discovered, Duisberg, then director of research to the Farbwerke vorm. F. Bayer & Co., Elberfeld, and his assistant, Hinsberg, conceived the notion of making a pharmaceutical product from about 30 tons of *p*-nitrophenol which had accumulated in the works stores as a useless by-product of the manufacture of dianisidine. *p*-Aminophenol, although less toxic than aniline, had nevertheless a marked action on the blood. Acetylaminophenol should, by analogy with acetanilide, be a step in the right direction; this also, however, acted upon the red corpuscles and so could not be used. Eventually it was found that ethylation of the hydroxyl group gave the desired result. Methoxyacetanilide already is an improvement on the free phenol; it has hardly any effect on the blood, and is more active as an antipyretic and as a narcotic; but the therapeutic value of the ethyl ether is much greater; it is superior to the lower homologue as an antipyretic, narcotic, and analgesic, and is still less poisonous.

Thus a quite short investigation led Duisberg and Hinsberg into discovering *Phenacetine* and laying the foundations of an important branch of industry.

A number of variations on the phenacetine theme may now pass under review. Phenacetine is a simple derivative of an aminophenol; its parent contains two atomic groupings, both eminently adaptable for substitution, so it can well be imagined that a vast number of derivatives were made and investigated under the stimulus provided by the success of Phenacetine and Antipyrine. Yet of the many products of this great effort there are now only two survivors, namely, *Lactophenin* and *Salophen*; all the others are either forgotten or neglected.

Nevertheless, two of the failures, *Phenocoll* and *Dulcin*, are worthy of a brief reference: the first because it appears to have a similar specific action to that of quinine on the microorganisms responsible for infectious fevers, and its antiseptic properties may provide a starting point for further research; the second because it is remarkably sweet to the taste.

As happened with the aniline derivatives, replacement of the acetyl radical in phenacetine by that of an acid lower or higher in the series has not produced substances of any great interest. The formyl derivative is quite different from phenacetine and is only feebly antipyretic, but it has a depressive influence on the activities of the spinal cord which makes it the best " antagonist " for strychnine.

Derivatives of hydroxy-acids are, on the other hand, more valuable. Lactylphenetidine or *Lactophenin* is, after Phenacetine, the most widely used medicament in this group. Because of its free hydroxy group it is more soluble in water than phenacetine. It is a weaker antipyretic, but acts more strongly as an analgesic and hypnotic. Further, its action is more speedy as a result of its being easily hydrolysed, for a readiness to undergo hydrolysis is characteristic of all derivatives of the hydroxy acids. It is prepared by heating phenetidine lactate at 170–180°, or by digesting together phenetidine and lactic ester.

If glyceric acid, containing two hydroxyl groupings, be used instead of lactic acid, the product obtained shows no antipyretic activity.

Apolysin, the citryl derivative:

$$\begin{array}{l} CH_2\cdot CO_2H \\ | \\ C(OH)\cdot CO_2H \\ | \\ CH_2\cdot CO\cdot NH\cdot C_6H_4\cdot OC_2H_5 \end{array}$$

contains two free carboxyl groups and has hardly any action.

Salicylphenetidine:

$$C_6H_4\begin{array}{l} \diagup CO\cdot NH\cdot C_6H_4\cdot OC_2H_5 \\ \diagdown OH \end{array}$$

(not to be confused with *Salophen*) has also been prepared. It has no action, since it is not hydrolysed in passing through the organism.

After the amides come the urethanes, derivatives of ethyl carbamate. *Thermodin* is an acetylated urethane of the constitution :

$$C_2H_5O{\cdot}C_6H_4{\cdot}N(COCH_3){\cdot}CO_2C_2H_5.$$

It is still in use ; no dangerous symptoms follow its administration.

We will next review various attempts made to produce more soluble analogues of phenacetine. The usual procedure has been followed, namely, introduction of acidic or basic groupings, or the radical of glycocoll, and so forth.

Two kinds of glycocoll derivative have been prepared : phenetylglycocoll, $C_2H_5O{\cdot}C_6H_4{\cdot}NH{\cdot}CH_2{\cdot}CO_2H$, is a representative of the first class. This has no physiological action. Aminoacetylphenetidine, $C_2H_5O{\cdot}C_6H_4{\cdot}NH{\cdot}CO{\cdot}CH_2NH_2$, *Phenocoll*, is typical of the second class. It has already been mentioned, and is a useful drug. It may be administered either as free base or as a salt ; in the latter case, its action is more rapid than that of phenacetine. The salicylate of Phenocoll is *Salocoll*.

Dulcin is *p*-ethoxyphenylurea : $C_2H_5O{\cdot}C_6H_4{\cdot}NH{\cdot}CO{\cdot}NH_2$. It is 250 times sweeter than sugar, and has only been used as a substitute for Saccharin.

Many other efforts have been made to produce useful derivatives of phenetidine, particularly by treating it with aldehydes, but nothing of practical importance has ensued. The same can be said for the attempts made to replace the phenolic hydrogen in the aminophenol by radicals other than ethyl. Neither acetylanisidine, nor *p*-amyloxyacetanilide, nor *p*-propyloxyacetanilide is of any value.

Quite recently, derivatives of glycol of the type $R{\cdot}NH{\cdot}CH_2\ CH_2OH$ have been patented, but the results of clinical investigation have yet to be recorded.

To all the above cases the following rules apply :

(i) The methyl derivatives are the more toxic ; the ethyl derivatives the more narcotic.

(ii) In a homologous series activity decreases as molecular weight increases.

There have also been prepared a number of esters of *p*-aminophenol, based on acetylaminophenyl benzoate as a type. The only one of these still in pharmaceutical use is *Salophen*, the salicylic ester of acetylaminophenol : $C_6H_4(OH){\cdot}CO_2{\cdot}C_6H_4{\cdot}NH{\cdot}COCH_3$. This compound is also a salol, acetylaminosalol in fact, and, as a derivative of salicylic acid, will be studied later.

In bringing the discussion of aminophenols to an end, the following remarks may be made :

(i) The *ortho-* and *meta-* isomerides are more toxic than the *para-*.

(ii) Among aminophenolic compounds, only those which produce *p*-aminophenol or its simple derivatives in passing through the organism have any antipyretic activity.

(iii) The absorption of antipyretics of the aminophenol series is

always followed by the appearance in the urine of substances giving the **indophenol** reaction. The test is carried out in this way. To a sample of the urine are added a few drops of sodium nitrite solution followed by a few drops of hydrochloric acid; the treated urine is then mixed with a dilute alkaline solution of β-naphthol. An intense red coloration is produced which changes to blue-violet when more acid is added. If, after administering a derivative of *p*-aminophenol, the test gives a negative result, one may generally conclude that the substance under investigation is not an antipyretic.

Aminophenols may be regarded as forming a class intermediate between the amines and phenol itself. For that reason the study of the latter compound and its derivatives has been deferred.

Derivatives of Phenol.—The only derivative of phenol, leaving salicylic acid out of the reckoning, that has been used in therapeutics is the glycerol ether *Antodyne*, $C_6H_5O\cdot CH_2\cdot CH(OH)\cdot CH_2OH$. This is an antipyretic with a well-marked analgesic and hypnotic action; it is soluble in water and has hardly any poisonous properties. It is too early yet to judge, but as this is a simple compound and breaks new ground for antipyretics, it is possible that useful derivatives will be made. Already one German firm has patented the urethane of phenyl-glyceryl ether and the urethane of the corresponding glycol ether:

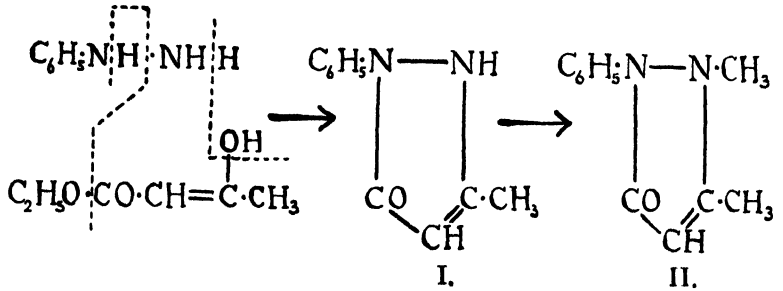
$$C_6H_5O\cdot CH_2\cdot CH(OH)\cdot CH_2O\cdot CONH_2$$
$$C_6H_5O\cdot CH_2\cdot CH_2O\cdot CONH_2,$$

and another has protected acetylamino-antodyne, and no doubt these will shortly appear on the market.

Derivatives of Pyrazolone.—When aniline dissolved in dilute mineral acid is treated with sodium nitrite, diazobenzene is produced, and when diazobenzene is reduced it is converted into phenylhydrazine (Emil Fischer, 1875).

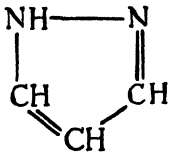
Phenylhydrazine is a violent poison acting on the red blood corpuscles even more vigorously than aniline. Every attempt made to render it less toxic has failed.

When phenylhydrazine is heated with acetoacetic ester, phenyl-methylpyrazolone (I.) is formed, and subsequent methylation produces phenyldimethylpyrazolone (II.), the trade name of which is *Antipyrine*:—



I. II.

These compounds are derivatives of **pyrazole**:

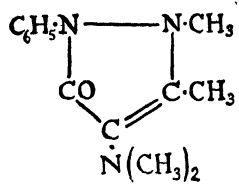


We will consider in detail the preparation of Antipyrine and its derivatives in the next chapter, and for the present confine our attention to their application in therapeutics.

At first antipyrine was thought to be a pure antipyretic, and several years elapsed before Germain Sée and Gley discovered that actually it had marvellous analgesic properties. This made it a signal success. Consequently, also, it received in France the name *Analgésine*.

Extensive pharmacodynamical investigations have been carried out on antipyrine and its derivatives and on pyrazolones in general, with results that cannot be studied here in detail for lack of space. They give no satisfactory explanation of the varying action of different members of the group. Indeed, the precise chemical formulation of antipyrine and its isomerides and derivatives is still a matter of controversy.

No other derivatives of antipyrine but *Pyramidon*, *Salipyrin* and *Melubrin* have established themselves in therapeutical use. *Pyramidon* or *Amidopyrin* is dimethylaminoantipyrine, and is prepared by methylating aminoantipyrine. It is both more active and more toxic than antipyrine itself. Its formula is:

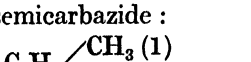


Melubrin is sodium aminoantipyrine-methane-sulphonate. It is readily soluble in water.

A few remarks may be made here on the **phenylsemicarbazides**, of which two representatives are used in therapeutics. These are *Cryogenin* and *Maretin*.

Cryogenin is *m*-benzamidosemicarbazide [1]:

Maretin is *m*-tolysemicarbazide:



[1] Cryogenin is described as benzamidosemicarbazide, but a new analysis (Fourneau, *priv. comm.*) proves that it is only phenyl-semicarbazide, $C_6H_5 \cdot NH \cdot NH \cdot CONH_2$.

Cryogenin (Lumière) is an excellent product; its antipyretic action manifests itself slowly, endures for a considerable time, and has no objectionable accessory effects.

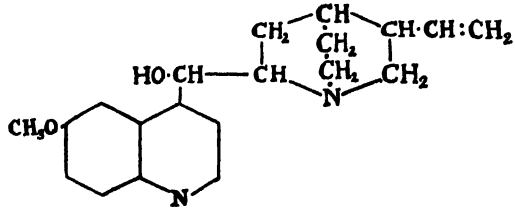
Maretin has no therapeutical value; its use has resulted in many mishaps.

All the medicaments discussed hitherto have acted on the *fever-symptoms*. Quinine and salicylic acid, on the other hand, have a further action on the actual agents responsible for certain diseases. Quinine is a specific for malaria, and salicylic acid for acute rheumatism.

QUININE

More than fifty years ago the action of quinine on the paramœcia in an infusion of hay was observed. The organisms were killed immediately by a 1 in 400 solution and ceased to move even in a dilution of 1 in 2,000.[1] But only since the celebrated discovery by Laveran in 1880 has the influence of quinine on the microorganism of malaria been systematically investigated.

The latest researches indicate that quinine has the structure represented below:



It is an aminoalcohol, containing both a quinoline radical and another of a special type which may be regarded as a fusion of two piperidine nuclei. It contains a methoxy group in the quinoline half of the molecule: the phenol of which quinine is the methyl ether is known as cupreine.

In the quinine series chemotherapeutical investigation can be, and has been, carried out in two ways. First, one may start with quinine itself and attempt to improve its properties by familiar methods, or, secondly, one may begin with quinoline and try to make more effective compounds by successive introduction of substituent groups.

Before the organism causing malaria was known, much attention was directed towards preparing quinine derivatives which should be more conveniently administered through being either less bitter or more soluble. Greater solubility was achieved by using other salts than the sulphate, for instance, the hydrochloride in presence of urethane, or better the formate, which is indeed still in considerable use in France. Tastelessness results when the quinine is so combined that the product is insoluble in water. Quinine base itself is very sparingly

[1] *Bioch. Z. f. med. Wiss.*, 1867, 310.

soluble and almost tasteless and can therefore be quite satisfactorily employed. But the most widely used product has been *Euquinine*, or quinine ethyl carbonate. This is prepared by treating quinine with a benzene solution of ethyl chloroformate; the hydrochloride of Euquinine being formed from which the base is set free by means of ammonia. Euquinine base is quite tasteless, but its salts are very bitter.

Aristoquinine, or quinine carbonate, obtained by treating quinine with phosgene, has similar properties.

As yet there is only one derivative of quinine which is at once soluble in water, neutral and tasteless. This is the sodium salt of the benzarsinic acid derivative, *Arsenobenzoylquinine* (Oechslin)

$$C_6H_4 \begin{cases} AsO_3NaH \\ CO—\text{Quinine}. \end{cases}$$

Since chemotherapeutical methods became more precise, quinine has been investigated from another point of view, the object here being to strengthen its antiseptic properties so that it shall act not only on malaria microbes but on others against which therapeutics has at present failed. Morgenroth's efforts in this direction some years ago led to the manufacture of *Optochin* (*Optoquinine*, = ethylhydrocupreine). Already Grimaux had prepared homologues of quinine (with the methyl group replaced by ethyl and propyl), and found that they were much more active than quinine itself, but unfortunately for development along this line, cupreine, the parent phenol, rarely occurs naturally and cannot be made by simply demethylating quinine, because the product of that reaction, it appears, is not cupreine, but an isomeride, apoquinine.

When quinine is reduced, hydrogen is taken up at the double bond and hydroquinine is produced. This derivative, like quinine itself, readily suffers demethylation when heated at 150° with 20% hydrochloric acid, but here isomerisation does not take place and the product is hydrocupreine. When hydrocupreine is ethylated by treatment with ethyl bromide in the usual way *Optoquinine* is obtained.

Optoquinine acts on the microbes responsible for pneumonia. It is too early yet for its use to be well established, as the tests have been interrupted by the war.[1]

Other derivatives are amylhydrocupreine or *Eucupin*, and *iso*-octylhydrocupreine or *Vuzin*. The latter is a powerful antiseptic and was largely used on the German side during the war for dressing wounds. It will act on microorganisms in presence of the body-fluids with as much vigour as in physiological saline *in vitro*.

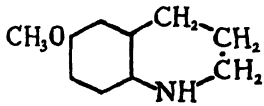
Another line of research has started from quinotoxin (eucupino-

[1] The hopes based on optoquinine appear not to have been justified; its use is followed by ocular trouble (cf. Atoxyl).

toxin), which is the ketone corresponding to quinine. The conversion of the alcoholic into the carbonyl grouping, markedly augments the antiseptic activity.

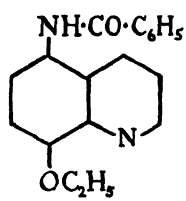
The second general method employed in chemotherapeutical research on quinine-like compounds may be called a centrifugal method, because it starts with a simple nucleus and makes more and more complicated compounds by successive introduction of substituents.

The problem of replacing quinine by simpler compounds of the quinoline series was tackled in the early days of synthetic medicaments: but although some measure of success was gained in that the substances were quite good antipyretics, yet there it stopped short, as they had no action on the microorganisms of malaria. Among the compounds of this order that have been used to some extent are *Thalline*, that is, tetrahydroquinanisole, 6-methoxytetrahydroquinoline:



Kairolin, N-ethyltetrahydroquinoline:

Kairin, hydroxy-N-ethyltetrahydroquinoline, and *Analgen*, a benzoylaminoethoxyquinoline of the formula:



some of which are, as can be seen, near relatives of phenacetine.

These products have been abandoned; but quite recently Kaufmann, one of Pictet's pupils, has taken out a series of patents on aldehyde derivatives of quinoline and alcohols produced therefrom. This seems to foreshadow new developments in this field of therapeutical research.

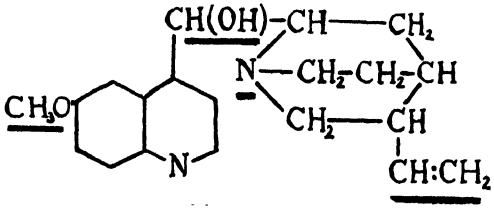
To sum up, in quinine, besides substitution in the rings, there are four groupings susceptible of variation, viz.:

The ethylene group,

The methoxy group,

The secondary alcoholic group (quinotoxin, reduction of quinotoxin),

The amino group.



During the investigations which led to the production of Optochin, modifications were made in the ethylene group and the alkyloxyl group, then later (quinotoxin) in the alcohol grouping. Numerous variations are still possible, but chemotherapeutical investigations take up much time, and one cannot expect them to be fruitful all at once.

SALICYLIC ACID

We will now turn to salicylic acid and its derivatives. *o*-Hydroxybenzoic acid, or salicylic acid, was discovered in 1838 by Piria, who obtained it by oxidising salicylaldehyde. In 1877, Kolbe devised the synthetic method employed for its industrial preparation, and this marks a stage in the development of chemical industry. Kolbe's method, more or less modified, is still in use, and consists in heating sodium phenoxide at 180–200° in a current of carbon dioxide. Only half the phenol undergoes reaction :

$$2C_6H_5ONa + CO_2 \longrightarrow C_6H_4\begin{matrix}ONa\\CO_2Na\end{matrix} + C_6H_5OH.$$

Schmitt (1905) introduced a valuable improvement, sodium phenyl-carbonate, produced by passing carbon dioxide over sodium phenoxide, being heated in an autoclave at 130°. Under these conditions, that is to say, in presence of carbon dioxide under pressure, the reaction proceeds to completion :

$$C_6H_5{\cdot}O{\cdot}CO_2Na \longrightarrow C_6H_4\begin{matrix}OH\\CO_2{\cdot}Na.\end{matrix}$$

By treating the sodium salicylate thus obtained with dilute mineral acid, salicylic acid is set free.

Industrially, the acid is purified by sublimation on a very large scale in special apparatus ; several tons a day may be thus handled. Plant of this kind is expensive ; yet the price of salicylic acid is now very low, and so evidently it becomes difficult to compete with manufacturers who have already written off the cost of their installation.

There is no need to discuss other methods for preparing salicylic

acid, such as by diazotising anthranilic acid: they are never employed in practice.

One point should be remembered, namely, that when potassium phenoxide is used instead of the sodium compound, *p*-hydroxybenzoic and not salicylic acid is formed.

It is not easy to suggest any way in which Schmitt's excellent modification of Kolbe's method may be further improved unless it be in simplifying the preparation of the alkaline phenoxide. Actually, according to a recent patent, this operation can be avoided by treating a *mixture of phenol* and *potassium carbonate* with carbon dioxide.

Action of Salicylic Acid.—At a strength of 1 in 1,000, salicylic acid inhibits the growth of bacteria, and at 1 in 10,000 it stops fermentation by yeast. It augments the antiseptic properties of sugar and salt. 0·5 part per 1,000 of milk prevents the milk from turning sour for three or four days.

Salicylic acid passes into the blood as sodium salicylate and circulates therein for a considerable time without exhibiting affinities for any particular organ, and this is perhaps its most characteristic property. It causes diabetic patients to eliminate less sugar. When added to a freshly prepared saturated solution of uric acid in sodium carbonate it prevents the precipitation of sodium urate; so also, in general, it dissolves urates already formed.

The various uses of salicylic acid are explained by this diversity of properties. But the most interesting action of all is that which it has in the treatment of acute rheumatism. What happens here is still obscure, because the actual agent responsible for this disease is still unknown; it is merely a matter of surmise that it is bacterial and not protozoal in nature.

Besides, drugs which have a specific action on bacteria alone are rare, indeed, are at present unknown. Yet, as salicylic acid is only a weak antiseptic, and its sodium salt still weaker, it must be supposed that it actually forms in the organism additive or decomposition products which do have some specific action on the agent of this disease. On the other hand, the following very plausible hypothesis has been put forward. Sodium salicylate circulates readily in the blood and is not decomposed by carbon dioxide, as the concentration of this is too low. But in the infected areas intense oxidation takes place, and the concentration of carbon dioxide is much higher and attains a value three or four times that for the blood. Possibly this is enough to set free salicylic acid itself, which, being much more energetic than its sodium salt, may thus exert its influence at the focus of the infection.

There is one fact which some day may help to account for the therapeutic action of salicylic acid, and that is that sodium salicylate can pass through fatty membranes. It does so certainly only in small amount, but quite definitely, and thus behaves to some extent like a hypnotic.

Lastly, one important characteristic of salicylic acid must be noted. It alone, of the three isomeric hydroxybenzoic acids, has this action on rheumatism; and again, of the three isomerides, it is the strongest acid. Whilst the dissociation constant of sodium salicylate is 0·102, those of the *para-* and *meta-* derivatives are 0·00286 and 0·00862, respectively. With these may be compared the figure for sodium benzoate, viz., 0·00600.

Derivatives of Salicylic Acid.—Free salicylic acid has a quite decided irritant action which after administration manifests itself by nausea, loss of appetite, and so forth. Sodium salicylate being partly decomposed already in the stomach may give rise to the same symptoms. The discovery of compounds which liberate the acid only in the bowel should, therefore, be regarded as a noteworthy advance in this branch of therapeutics. Such substances are, for example, *Salol*, *Salophen*, *Aspirin* . . . .

We will make a short survey of this class of derivatives. *Salol* is phenyl salicylate. It is not hydrolysed by the gastric juice but very readily by the bile and the pancreatic juice. One method for preparing it consists in heating together at 120° two molecular proportions of salicylic acid, two of phenol and one of phosphorus oxychloride. Industrially, phosgene (carbonyl chloride) is used instead of phosphorus oxychloride, as it is much cheaper; the procedure then is to heat a mixture of sodium salicylate and sodium phenoxide with phosgene under pressure. Finally, if salicylic acid be heated alone at 160–240°, Salol is formed, one molecule of salicylic acid losing carbon dioxide to give phenol, which then esterifies another molecule of acid.

Betol is the corresponding ester of naphthol.

A Salol derivative already mentioned is *Salophen*. This is obtained by treating acetylaminophenol with salicylic acid and phosphorus oxychloride in benzene solution.

Aspirin has been known for a long time; Gerhardt discovered it almost simultaneously with acetanilide. But only much later was its utilisation as a substitute for salicylic acid suggested. It is eminently an analgesic antipyretic.

In fever experimentally produced by puncture near the *corpus striatum*, aspirin (in small doses) behaves like a most active antipyretic and causes a considerable fall in temperature, out of all proportion with that brought about by salicylic acid. This is still a mysterious phenomenon because, *in vitro*, aspirin is speedily hydrolysed to salicylic and acetic acids.

Aspirin is prepared either by heating salicylic acid with acetic anhydride at 170°, or by treating it with acetyl chloride in presence of pyridine.

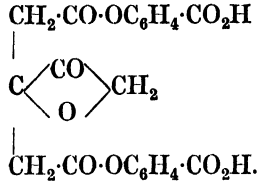
Aspirin has not been ousted from pharmacy by any derivatives except, recently, its sodium and calcium salts.

Other esters of salicylic acid may be divided into two classes : those which are solid and are used internally, and those which are liquid and are applied in lotions, liquid salicylates being able to pass into the blood through the skin.

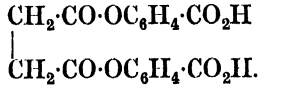
The following derivatives are solids :

Diplosal is salicylsalicylic acid : $C_6H_4(OH)\cdot CO\cdot OC_6H_4\cdot CO_2H$.

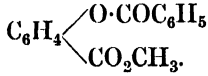
Novaspirin is methylene-anhydrocitrylsalicylic acid :



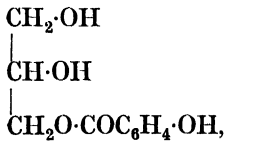
Diaspirin is succinylsalicylic acid :



Benzosalin is methyl benzoylsalicylate :



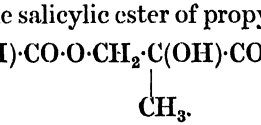
Glycosal is the monosalicylic ester of glycerol :



and *Salacetol*, salicylacetone :

$$C_6H_4(OH)CO_2\cdot CH_2CO\cdot CH_3.$$

Similarly there is quite a number of liquid derivatives. These should pass readily through the skin, be non-irritant, and have an odour neither disagreeable nor too persistent. One known for many years is methyl salicylate,[1] the smell of which is, however, very pronounced. Amyl salicylate is on the market as *Ulmaren*. Among recently employed derivatives the best seem to be *Spirosal* and *Algolan*. Spirosal is glycol monosalicylate : $CH_2(OH)\cdot CH_2O\cdot CO\cdot C_6H_4\cdot OH$, and Algolan the salicylic ester of propyl dihydroxybutyrate :



Lastly, we will mention *Mesotan*, the methoxymethyl ester : $C_6H_4(OH)\cdot CO_2\cdot CH_2\cdot OCH_3$, which has a very irritating action ; *Salen*, which is a mixture of ethyl and methyl esters of salicylglycollic acid : $C_6H_4(OH)\cdot CO\cdot OCH_2\cdot CO_2C_2H_5$(or CH_3) ; and *Salit*, *i.e.*, bornyl salicylate, etc.

[1] Oil of wintergreen.

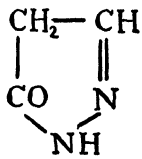
CHAPTER IV

PREPARATION OF ANTIPYRINE

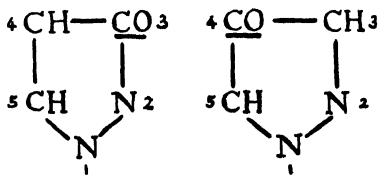
ANTIPYRINE is prepared by condensing phenylhydrazine with aceto-acetic ester and methylating the product. Such is the basis of the process; in practice it is not as simple as it sounds and cannot be carried through successfully without a sure grasp of theory. But apart from that, this is one of the most interesting branches of organic chemistry, not only because of the manifold reactivity of the compounds in question, but also because our knowledge of them results from the work of such notable investigators as Knorr and Michaelis.

From a general instructional point of view it would be difficult enough to exhibit properly all the reactions of pyrazolone compounds, they are so diverse. Here only those directly involved in the manufacture of antipyrine can be considered.

The simplest pyrazolone is a ketonic derivative of pyrazole, and may be formulated thus:

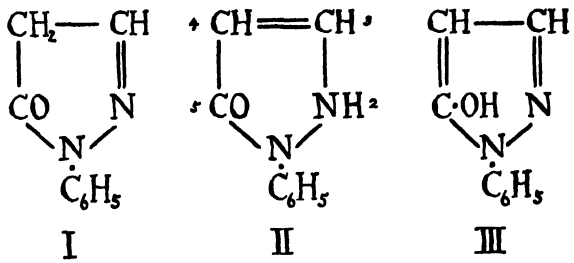


In such a five-membered heterocyclic structure as this, the ketonic group may occupy one of two positions, thus:



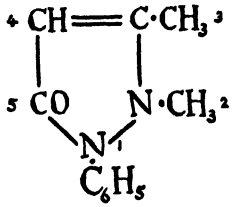
The atoms in the pyrazole ring are always numbered by beginning with one nitrogen atom and passing round by the second, as shown. That nitrogen atom which carries a substituent group is always numbered "1."

Only the 5(3)-pyrazolones, to which class antipyrine belongs, will be discussed here. Phenylpyrazolone may be represented by three formulæ:



I II III

As antipyrine is 1-phenyl- 2 : 3-dimethylpyrazolone, its structure is that of II., thus :

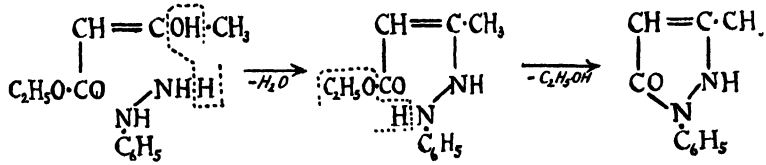


Obviously it may have a number of isomerides according to the positions of the methyl and carbonyl groups and of the double bonds, but, although these are of great interest, they must not now detain us.

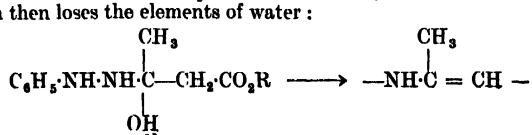
Let us follow, step by step, the preparation of antipyrine. To begin with, *phenylhydrazine is condensed with acetoacetic ester*, the reaction being carried out either without a solvent or in a neutral medium. The condensation takes place in two stages, corresponding, respectively, with the separation first of water and then of alcohol. The first of these reactions takes place in the cold, whilst the second needs a higher temperature.

Now, theoretically, acetoacetic ester may react either as the ketonic or as the enolic form and so the first stage in the reaction may be represented symbolically in two alternative ways,[1] thus :

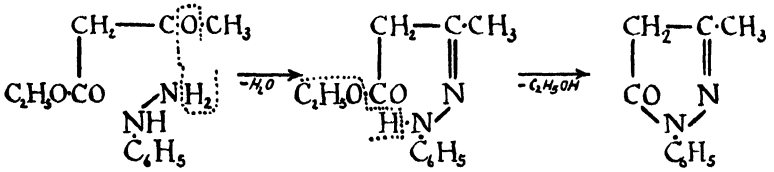
A. (From the enolic form.)



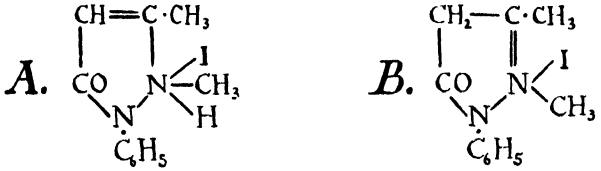
[1] Possibly the reaction follows yet another course, an addition compound being formed, which then loses the elements of water :



B. (From the ketonic form.)

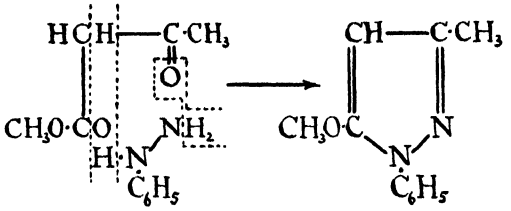


In the second stage, alcohol separates and phenylmethylpyrazolone is formed; as is also shown, this product may have one of two formulæ. And if methyl iodide acts on the compounds represented by these two formulæ, the products will be:



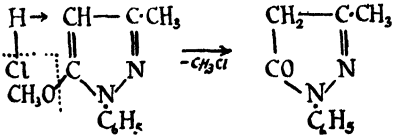
When the methiodide actually obtained is treated with caustic soda, antipyrine is produced. This result readily agrees with formula **A**, whilst formula **B** requires that the transference of a double bond take place. Thus the *formation of antipyrine is best explained by supposing that originally the ethyl acetoacetate reacted in the enolic form.*

The above is what happens when the condensation of phenylhydrazine and acetoacetic ester is brought about in either a neutral solvent or none at all. If, however, the reaction takes place in an acid medium it does not follow the same course, at any rate in the second stage. Thus, if methyl acetoacetate, for example, be used, the product is a methoxymethylpyrazole:

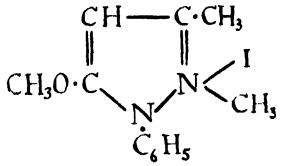


the condensation in this case having taken place by elimination of two molecules of water.

If this derivative be heated with hydrochloric acid, it loses one methyl group as CH_3Cl and gives a phenylmethylpyrazolone identical with that obtained by condensation in a neutral solvent.



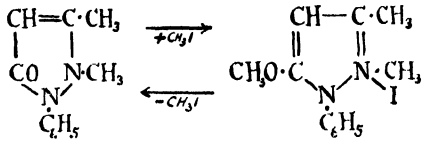
The methoxy derivative above combines with methyliodide, giving a methiodide :



Methoxymethylphenylpyrazole methiodide.

And this is the same compound as is formed when antipyrine is heated with methyl iodide.

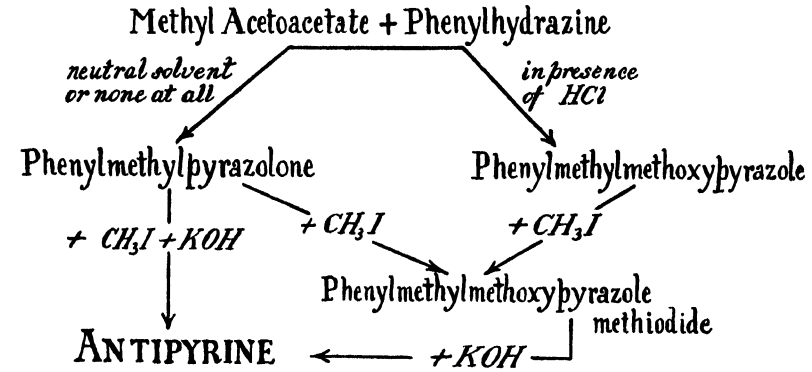
Here is a most remarkable reaction which shows that the pyrazole ring is in a state of very unstable equilibrium; by displacement of one double linkage, two more have been formed:

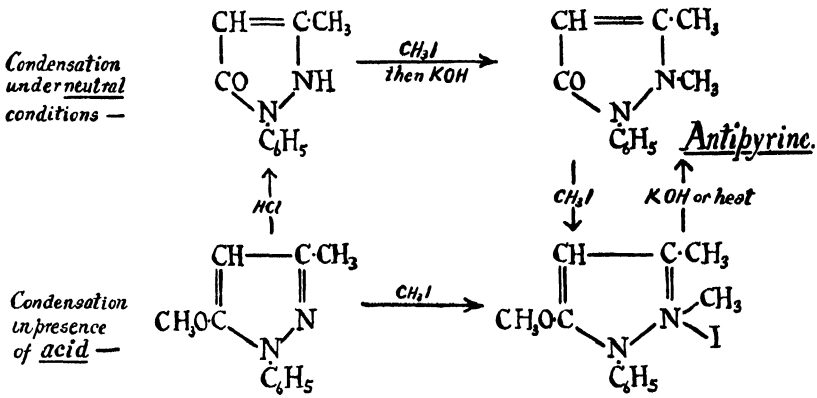


The methiodide above is a quaternary ammonium iodide. Usually such quaternary salts are very stable towards caustic alkalis : tetramethylammonium iodide $(CH_3)_4NI$, for instance, may be heated with concentrated caustic potash solution without suffering decomposition; nay, the corresponding base, tetramethylammonium hydroxide, will displace potash from its salts. To decompose quaternary ammonium iodides it is necessary to use moist silver oxide. But these quaternary methiodides derived from antipyrine behave differently : they lose methyl iodide by simply boiling with dilute alkali and thus regenerate antipyrine. More than that, it is not even necessary to boil with caustic soda; simple heating of the compound by itself will drive off the methyl iodide, the opposite reaction to that which produced the quaternary iodide taking place.

So, whether the condensation of phenylhydrazine and acetoacetic ester be carried out in acid or in neutral medium is of no great importance, as antipyrine can eventually be obtained either way.

The above relationships are set out diagrammatically below:



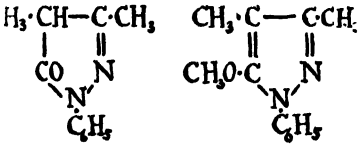


Phenylmethylpyrazolone and phenylmethylethoxypyrazole being intermediates in the manufacture of antipyrine, one ought to be familiar with their principal properties. Phenylmethylethoxypyrazole crystallises from ligroin in large colourless scales, m.p. 38·5°. It dissolves in acids but not in alkalies. Phenylmethylpyrazolone may be recrystallised from alcohol or water and then melts at 127°. It is insoluble in ether and only sparingly soluble in cold water. *It is dissolved by both acids and alkalies*, and forms characteristic salts with certain heavy metals, a noteworthy example being the diacid silver salt of the composition, $C_{10}H_9ON_2Ag \cdot C_{10}H_{10}ON_2$. Phenylmethylpyrazolone seems, then, to behave in many reactions as if it were an enol, although a better explanation of other reactions is furnished by the ketonic formula. This behaviour is quite comparable with that of acetoacetic ester. Only the ketonic formula can make intelligible, for example, the condensation of phenylmethylpyrazolone with aldehydes, but on the other hand the enolic formula offers the best explanation of the formation of antipyrine by methylation and the solubility of the compound in alkalies.

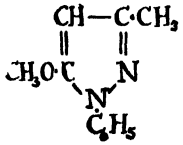
We may now study more closely the second step in the preparation of antipyrine.

Methylation.—When phenylmethylpyrazolone is treated with methyl iodide it is converted into antipyrine hydriodide, which is decomposed by caustic alkali, giving antipyrine. If, however, as is frequently done in ordinary methylations, the reaction be carried out in presence of caustic soda, it goes much too far and in other directions, and the product is a mixture of six compounds, of which two are dimethyl derivatives (one of them being antipyrine), and three trimethyl derivatives.

There is no need to give the formulæ of all these compounds here; two only will be cited to show how intricate the problem is:



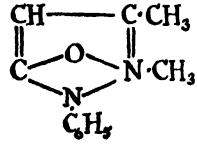
If diazomethane in methyl alcoholic solution be used as the methylating agent, the product is exclusively the methoxyl derivative:



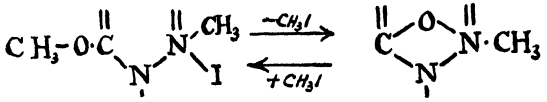
and the same product is formed when methyl sulphate is employed if the substances interact in presence of sodium methoxide, but if the reaction be carried out in *aqueous* methyl-alcoholic sodium hydroxide, antipyrine is obtained.

All this information is not useless but of value. Even now not half the observations made in this field have been mentioned. Industrial chemists should have an all-round knowledge of their subject; they should know it even better than those who work in scientific laboratories, as a mistake on their part may have more serious consequences. It is easy to see, from what has been said, that disastrous results might ensue if phenylmethylpyrazolone were methylated in presence of soda, or if, having carried out the condensation of phenylhydrazine and acetoacetic ester in presence of acid, one did not know how to convert the product into antipyrine.

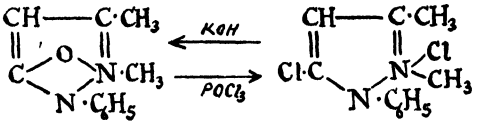
In the last chapter it was pointed out that the formula for antipyrine was still under discussion. Michaelis, in fact, urges strongly the adoption of a special formula—the phenol-betaine formula:



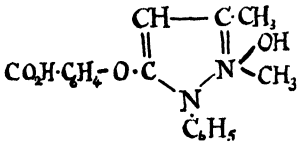
This explains better than the ketonic formula many reactions. First of all, antipyrine is exceptionally soluble compared with phenylmethylpyrazolone, and this is easily understood if there be a quaternary ammonium grouping in the molecule. Secondly, both the methylation of antipyrine and the disruption of the product by simple heating become more intelligible :



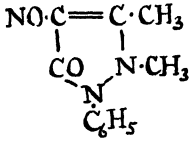
Similarly for the action of phosphorus oxychloride :



Finally, a most widely used derivative of antipyrine, namely, *Salipyrin*, is not an ordinary salt ; for reasons which cannot be discussed here, it is supposed to be an ether of salicylic acid, thus :



Antipyrine readily gives substitution products with bromine, iodine, nitrous acid, mercury, aldehydes. It contains a very mobile hydrogen atom. Nitroso-antipyrine :



is the primary intermediate in the manufacture of *Pyramidon* (dimethylamino-antipyrine) and *Melubrin* (sodium salt of phenyldimethylpyrazolone-aminomethane-sulphonic acid).

The compound produced by interaction with formaldehyde, methylene di-antipyrine, is used in estimating antipyrine ; so also is the iodo-derivative. When antipyrine combines with trichloroacetaldehyde (chloral) a substance is produced which has hypnotic properties and is sold under the name of *Hypnal*. . . .

Pyramidon.—Pyramidon is prepared by reducing nitroso-antipyrine and methylating the product. As reducing agents may be used either zinc and sodium bisulphite or, better, bisulphite alone. The latter method produces the amino-sulphonic acid which yields Pyramidon directly when treated with methyl sulphate. If the intermediate compound be aminoantipyrine itself, it is separated as the benzyli-

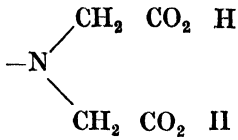
dene derivative by treatment with benzaldehyde. This derivative is insoluble in water [1] and easily obtained quite pure.

Aminoantipyrine may be methylated by using either methyl iodide, bromide, or better, methyl sulphate. Sometimes, however, these methods give a poor yield because quaternary ammonium salts are formed. Efforts have therefore been made to obtain a more nearly quantitative yield, and the following improved process has been elaborated.

Aminoantipyrine, treated with chloracetic acid, forms amino-antipyrine-diacetic acid :

$$-NH_2 + 2ClCH_2 \cdot CO_2H \longrightarrow -N(CH_2 \cdot CO_2H)_2,$$

and when this is treated with hydrochloric acid in an autoclave at 120°, carbon dioxide (2 mols.) is given off and Pyramidon produced :



[1] The compound is so sparingly soluble that on this reaction Tiffeneau has based a method for estimating benzaldehyde.

CHAPTER V

HYPNOTICS

Classification.—As matters stand it seems that the most useful thing to do is to classify hypnotics, as only by that means can one keep track with individual work in this field, or grasp some of the relationships subsisting between hypnotic properties and chemical constitution.

Usually, hypnotic compounds have been divided into three classes, namely,

(*a*) Those containing aldehyde or ketone groups,

(*b*) Halogen compounds,

(*c*) Compounds containing alkyl radicals.

This classification, put forward by Fränkel in his well-known treatise,[1] does not seem to be very satisfactory, being at once inexact and too rigid. Thus, if we adopt it, we must put chloral into both class (*a*) and class (*b*); we must separate amido compounds from their halogenated derivatives, and so forth.

As we shall see later, hypnotic activity is due mainly to particular physical properties, possessed by substances of widely varying constitution. Hypnotics may therefore be met with in any group of chemical compounds provided the following conditions are fulfilled:

(1) The compound in question may not be acidic or basic in character, or if so, the property may not be too pronounced; [2]

(2) It must be stable to a definite degree;

(3) It may not be too soluble in water, or if freely soluble, it must be still more soluble in ether or liquid fats.

Hypnotics, in contrast with antipyretics, are mostly members of the aliphatic series. But this is because the first known hypnotic compounds belonged to this series and later research has centred on them. Yet, among many compounds with hypnotic properties, such an augmentation of activity results from replacing an alkyl group by an aryl group that without doubt the aromatic series will furnish other excellent hypnotics to set beside *Luminal*, *Hypnone* and *Nirvanol*.

It would seem that the best way to classify hypnotics is to survey the chief families of organic compounds, noting down those in which substances used or suggested as hypnotics are met with; then, for the families thus endowed, to draw up a scale of narcotic potency, as far as available knowledge permits.

[1] *Die Arzneimittel-Synthese*, 5th ed., Berlin, 1921.

[2] Veronal and Luminal dissolve in alkali but are not true acids

But before setting about this classification, we must make a preliminary grouping: we must distinguish between **general anæsthetics** and **hypnotics.**

General anæsthetics are those which simultaneously alleviate pain and stop reflex-action; which bring about narcosis, but, naturally, leave unaffected the respiratory organs and the vaso-motor centres.

These again form two sub-groups, namely, volatile anæsthetics administered by inhalation, and non-volatile anæsthetics. The first of these, of which *chloroform* is a type, are characterised by the fact that their action is complete but of short duration, and abates rapidly when administration is stopped. The patient under treatment, after first passing through a state of excitement, begins to lose touch with the outside world; movement ceases, even that due to reflex-action, but almost immediately consciousness begins to fade and nearly vanishes, although a few glimmerings remain. Actually the motor centres are attacked after the sensory centres but, practically speaking, nearly all the functions of the brain are affected at once.

The second sub-group is represented by *morphine* and *scopolamine.* Morphine stands by itself, for it acts almost exclusively on the sensory centres, and is not really hypnotic except when administered in doses which approach the limit of safety. It is hypnotic inasmuch as it relieves the pain that causes sleeplessness; but if the insomnia is due to something else it is not always effective.

Scopolamine is pre-eminent in acting mainly on the motor centres; it serves thus to round off the effect produced by morphine and the hypnotics.

Hypnotics properly so called limit their action to bringing about the first stage of narcosis, *i.e.*, the loss of consciousness of outside events, and can maintain this condition for some time. They have no true action on sensibility except when used in doses nearing the lethal limit, and so are in sharp contrast with morphine.

So the effects produced by the hypnotics on the one hand and morphine on the other are essentially different in character and only come to resemble one another when either substance is administered in dangerous doses.

Having cleared up these points, we may begin the classification.

Hydrocarbons.—Nearly all aliphatic hydrocarbons of not too high molecular weight are hypnotic and belong to the group of general anæsthetics. All those may be used that have boiling points lying between 25° and 50°, so from pentane (normal) boiling at 36°, to the branched hexanes, such as $\beta\beta'$-dimethylbutane, trimethylethylmethane, boiling at 49° (Schleich). Naturally all cannot be employed, as the question of cost has always to be considered, but if ether and chloroform were unknown, these compounds would serve equally well in their place.

Ethylenic hydrocarbons have a much more pronounced action,

ethylene itself being a strong general anæsthetic.[1] The best known compound of this series is *Pental*, trimethylethylene, $(CH_3)_2C:CH{\cdot}CH_3$. This is obtained by dehydrating amyl alcohol by treatment with zinc chloride and boils at 36°, *i.e.*, much higher than the corresponding saturated hydrocarbon, which boils at 30·5°. It is, in fact, a general rule that ethylenic hydrocarbons have higher boiling points than the corresponding paraffins, although the contrary usually holds for their derivatives.

Halogen Derivatives of the Hydrocarbons.—Passing on to this group of compounds we meet with chloroform, the most widely employed general anæsthetic, and with ethyl chloride and ethyl bromide, the use of which is rapidly spreading.

All chloro- and bromo- derivatives with boiling points between 20° and 50° possess hypnotic properties. It has been stated that those with an odd number of halogen atoms are more potent than the others, thus, for instance, ethyl chloride is a more energetic hypnotic than ethylene chloride, this itself being weaker than methylchloroform,

$$CH_3{\cdot}CH_2Cl > CH_2Cl{\cdot}CH_2Cl < CH_3{\cdot}CCl_3,$$

but this generalisation does not quite agree with the facts. It has been forgotten that the location of the chlorine atoms on the carbon chain is of importance, thus ethylidene chloride, $CH_3{\cdot}CHCl_2$, with both chlorine atoms attached to the same carbon, is more active than ethylene chloride or even ethyl chloride.

Nearly all the halogen derivatives coming within the limits of volatility specified above have been tested in actual practice, thus, in England, methylene chloride CH_2Cl_2; in France, methylchloroform; in Germany, acetylene chloride, CHCl : CHCl (*Dioform*); but none has achieved permanent success.

Alcohols.—Of this series the tertiary alcohols are by far the most active, and one of them is still widely used, namely dimethylethylcarbinol, $(CH_3)_2C(OH){\cdot}C_2H_5$, or "amylene hydrate." Triethylcarbinol, a C_7 alcohol, is even more potent, but in small doses it brings about a state of excitement, and is, moreover, not easily made, while "amylene hydrate" is readily obtained by hydrating *Pental*:

$$(CH_3)_2C = CH{\cdot}CH_3 + H_2O \longrightarrow (CH_3)_2C(OH){\cdot}CH_2{\cdot}CH_3.$$

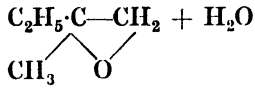
The **glycols** are not such active hypnotics as alcohols with the same number of carbon atoms, but with increase in molecular weight, and particularly if at least one hydroxyl group be tertiary, they do become powerfully hypnotic. The activity is greatest when both hydroxy groups are tertiary. It comes to the same thing to say that these effects follow increased solubility in ether.

Amylene glycol, hydroxymethylmethylethylcarbinol:

$$C_2H_5{\cdot}\underset{\displaystyle CH_3}{\overset{\displaystyle |}{C}}(OH){\cdot}CH_2OH,$$

obtained by treating the corresponding derivative of ethylene oxide with water:

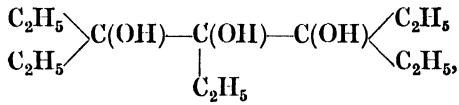
[1] Acetylene also is a valuable general anæsthetic.



is an excellent hypnotic, but has not been used because of its costliness. The chlorhydrin of this series, it may be remembered, is an intermediate in the preparation of Stovaine.

The **pinacones,** however, possess the most pronounced activity, and when methods have been discovered for producing them cheaply, which should be in the near future, doubtless some representative or derivative of this group will be employed in therapeutics. Ethylmethylpinacone, diethyldimethylethylene glycol, $C_2H_5(CH)_3) : C(OH) \cdot C(OH) : (CH_3)C_2H_5$, is quite as active as " amylene hydrate "; and the maximum activity, it appears, is shown by the C_{10} compound, diethylpinacone, tetraethylethylene glycol, $(C_2H_5)_2 : C(OH) \cdot C(OH) : (C_2H_5)_2$.

Such trihydric alcohols as have so far been tested are not hypnotic, but one may expect the property to be possessed by derivatives with secondary or tertiary hydroxyl groups or with a many-branched carbon chain. The compound :



which may be prepared by treating ethyl mesoxalate, $CO(CO_2C_2H_5)_2$, with magnesium ethyl bromide, should be interesting to study.

Halogen Derivatives of the Alcohols.—Already a generalisation may be made, namely, that if the simplest member of a series be soluble in water and not in ether, then the more this is so the higher must be the molecular weight of a derivative for it to possess hypnotic properties. However, substitution by halogens brings about the same result as mere increase in molecular size or the development of branched chains, as is shown, for example, by the fact that halogen derivatives of glycerol and its homologues are hypnotic, whilst the parent trihydric alcohols are inactive.

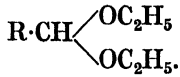
Isopral, $CCl_3 \cdot CH(OH) \cdot CH_3$, is, of the halogen alcohol derivatives, the one used to the greatest extent. It is obtained by treating chloral with magnesium methyl iodide. The preparations of this compound and that of Stovaine were the first in which Grignard's reaction was carried out on an industrial scale.

The related tertiary alcohol, $CCl_3 \cdot C(OH) : (CH_3)_2$, is *Chloretone.* This derivative is more active than Isopral. It may be prepared by causing acetone and chloroform to interact, or by treating trichloracetic ester with magnesium methyl bromide.

Ethers.—To this family belongs the compound that, with chloroform, is most widely used, namely, Ether (Diethyl ether, " sulphuric ether "), $C_2H_5 \cdot O \cdot C_2H_5$. The ether grouping is, physiologically, one of the most active, and compounds containing it differ from the

corresponding alcohols to a surprising degree. Here, again, the cost must be considered, otherwise methyl ethyl ether, or methyl propyl ether, could be utilised quite well instead of common ether.

The acetals and the orthoformates may also be included in this class. Acetals are ethers of (hypothetical) dihydric alcohols in which both hydroxy groups are attached to the same carbon atom, thus :



Nearly all these compounds show hypnotic activity. Those derived from ketones are, in fact, among the best hypnotics known. This conclusion, which contradicts earlier statements, has resulted from recent research carried out by Brissemoret on the two compounds represented by the formulæ :

Orthoformates are derivatives of the hypothetical orthoformic acid, $CH(OH)_3$; they may also be regarded as ethers of the simplest trihydric alcohol. Ethyl orthoformate, $CH(OC_2H_5)_3$ (*Ethone*), is largely used in France as a mild hypnotic and as a sedative in whooping-cough (Brissemoret, Chevalier).

Aldehydes.—Oxidation of a primary alcohol to the corresponding aldehyde brings about a considerable increase in hypnotic activity, as may be seen by comparing ethyl alcohol with acetaldehyde. But aldehydes have other, objectionable, properties : small doses induce excitation, whilst larger doses bring about asphyxia. These defects are less pronounced in the polymerides, and paraldehyde, for example, is still employed to some extent.

Of halogen derivatives of the aldehydes, those containing one halogen atom in the molecule are extraordinarily irritant and toxic, but when the proportion of halogen passes a certain point, the compounds produced have the property of forming hydrates. These substances, which may also be regarded as substituted dihydric alcohols with both hydroxy groups attached to the same carbon atom, are soluble in water, much less irritant than the corresponding unsubstituted aldehydes, and have no appreciable odour.

The best known compound of this kind is chloral hydrate, $CCl_3{\cdot}CH(OH)_2$, the first synthetic narcotic. Although discovered by Liebig, it was not employed pharmaceutically for many years. This development was due to Liebreich, who reasoned that as chloral easily breaks down to form chloroform, so it might serve actually in the organism as a source of the latter. This reasoning was unsound, but it led to a valuable discovery, that of the physiological properties of chloral.

Chloral has a most disagreeable taste and an irritant action on the

gullet. As it will combine with almost any organic compound, it is easy to imagine the vast number of derivatives that have been investigated. Compounds with sugars, alcohols, amides, antipyrine, etc., all have been tried. Of these only three are still in use, namely, *Dormiol*, which is the combination with tertiary amyl alcohol and is an excellent product; *Hypnal*, the compound with antipyrine already mentioned, and *Chloralose*. *Chloralose* is employed to only a limited extent on the human subject, but for animals it is the best anæsthetic hypnotic known. With its aid operations may be carried out without either danger or pain. It is produced by the interaction of glucose and chloral, having formed the subject of an investigation by Richet and Hanriot.

Other derivatives of chloral have had but a short career. Among such are *Viferral*, a polymeride of chloral, chloral amide, chloral-oxime, chloral urethane, $CCl_3CH(OH)\cdot NH\cdot CO_2C_2H_5$.

A homologue of chloral, viz., butylchloral, $CH_3\cdot CHCl\cdot CCl_2\cdot CH(OH)_2$, has been used in combination with pyramidon, under the names *Trigemin* or *Asciatine*. This compound of butylchloral and pyramidon is a well-characterised crystalline substance, although chloral itself will not combine with pyramidon, but only with antipyrine.

Ketones.—Ketones are produced when a secondary alcohol is oxidised. From methylethylketone onwards, all ketones are narcotic to some extent. Nowadays these compounds, even the mixed ketones, are easily prepared by Senderen's method, namely, by passing a fatty acid, or a mixture of acids, over heated thoria. It may be expected that the higher members of the series, of which at present little is known, will soon be investigated more fully.

The simplest ketone in the aromatic series, acetophenone or *Hypnone*, has long been in use and still has a certain vogue, principally in Italy.

The monohalogenated ketones, like the corresponding aldehyde derivatives, are violent irritants. But, just as with the aldehydes, when more chlorine atoms are present in the molecule, the products will form solid hydrates with no odour and a not too unpleasant taste. Thus, while monochloroacetone is one of the most objectionable compounds to handle, trichloroacetone hydrate, $CCl_3\cdot C(OH)_2\cdot CH_3$, is solid and almost odourless. Naturally it is strongly hypnotic.

Acids.—The ultimate product of oxidising a primary alcohol, short of complete breakdown, is an acid. Acids, no matter what their molecular weight or the degree of substitution by chlorine, possess no hypnotic properties at all. But their esters and amides are in a different category.

Esters.—Most esters of fatty acids up to caproic acid are hypnotics, but none has been utilised except those of valeric acid. Among the latter, the borneol ester is reputed to be the best; it is found in valerian root and is also manufactured under the name *Bornyval*. It

has mildly hypnotic and sedative properties. *Validol* is menthol valerate.

Amides.—This class of compounds is of outstanding interest, for it includes nearly all the most widely employed hypnotics of recent origin, *Veronal*, *Adaline*, and the like.

Simple amides of the aliphatic series, homologues of acetamide, possess only feeble hypnotic properties. The only one in pharmaceutical use is valeryl diethylamide. Hypnotic activity is more marked in amides of the aromatic series. Amides of halogenated acids are much more active, particularly if the parent acid contains a branched carbon chain.

The following simple classification of this group may be made :

Acetamide and its homologues . .	Feeble activity or none at all.
Benzamide and its homologues . .	Hypnotics.
Halogenated homologues of acetamide .	Hypnotics.

Hypnotic properties are possessed even by bromacetamide and in the highest degree by diethylbromacetamide (*Neuronal*).

But the substances of this class in which hypnotic properties are most highly developed are the acyl derivatives of urea (carbamide) and the urethanes. Thus *Bromural* is bromo-*iso*-valerylurea and *Adaline* is diethylbromacetylurea.

Urethanes are esters of carbamic acid ; ethyl urethane, for example, is

$$NH_2{\cdot}CO{\cdot}OC_2H_5.$$

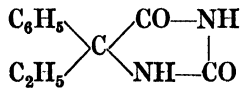
Some of the best and least objectionable hypnotics known are urethane derivatives. Of those devised from primary alcohols only, the simple ethyl urethane is in use. Urethanes of secondary alcohols are much more active : *Hedonal*, a representative of this group, is methylpropylcarbinol urethane :

$$C_3H_7{\cdot}CH(CH_3)O{\cdot}CO{\cdot}NH_2.$$

Urethanes of tertiary alcohols have also been prepared ; *Aponal*, for example, is the carbamate of tertiary amyl alcohol :

$$C_2H_5C(CH_3)_2O{\cdot}CO{\cdot}NH_2.$$

Urea, carbamide, $CO(NH_2)_2$, is not itself a hypnotic, but many of its derivatives are. The property is well developed and sometimes very pronounced, particularly if many substituent radicals are attached to the nitrogen atoms.[1] The most recent addition to this class is *Nirvanol*, phenylethylhydantoïn,



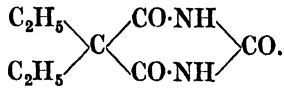
Amylurea, $CH_3{\cdot}CH_2{\cdot}C(CH_3)_2{\cdot}NH{\cdot}CO{\cdot}NH_2$, appears to be an excel

[1] Lumière and Perrin have lately found that certain homophthalimide derivatives, the dialkylhomophthalimides, are hypnotics.

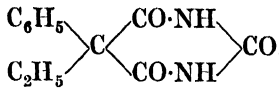
lent hypnotic, but its preparation is troublesome. The heptylurea derived from triethylcarbinol, $(C_2H_5)_3C{\cdot}NH{\cdot}CO{\cdot}NH_2$, is similarly a very potent substance.

Veronal Series.—This is the group of the ureides, among which the derivatives of the dialkyl- and arylalkyl-malonic acids have the highest reputation. *Veronal, Luminal, Proponal, Dial* and *Soneryl* are members of this series.

Veronal is diethylmalonylurea (diethylbarbituric acid):

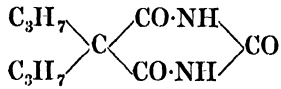


Luminal (*Gardenal, Phenylbarbital*) is phenylethylmalonylurea:



It is widely employed in treating epilepsy.

Dial is diallylmalonylurea, and *Proponal* is dipropylmalonylurea:



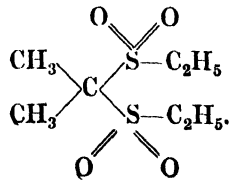
Soneryl (Tiffeneau) is ethylbutylmalonylurea.

Neither the organic bases nor the acids are possessed of hypnotic properties, but, as has just been made evident, certain combinations of bases and acids (the amides) are peculiarly well endowed in this way.

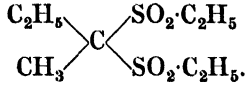
Nearly all the natural groups of aliphatic compounds—hydrocarbons, alcohols, aldehydes, ketones, acids, amides, ethers and esters, amines—have now been reviewed and we have seen that hypnotics are met with in all but two of them, namely, the acids and the amines, and that even this gap is, in a way, filled when members of these two groups are brought into combination.

Sulphonal.—There remains to consider another, very important, group of hypnotics that cannot really be classed with any of the above. These are the sulphones, one of which is *Sulphonal.* At first sight, sulphones resemble esters, but the resemblance disappears on closer inspection. Here the sulphur atom is linked directly to two carbon atoms. It is combined, further, with two atoms of oxygen, and is thus hexavalent. The only group with which the sulphones can be compared is that of the ketones.

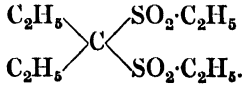
Sulphonal is diethylsulphonedimethylmethane:



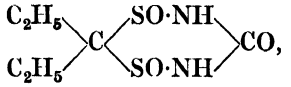
Trional is diethylsulphonemethylethylmethane :



Tetronal contains only ethyl groups :



In these compounds there is some resemblance to Veronal and Adaline, but, unfortunately, it is impossible to produce directly from a sulphone a sort of veronalsulphone :



but perhaps one may attain this end in some other way.

It need not be pointed out how much there is still to do in this field, and what vast tracks are open for research.

Relationships between Hypnotic Activity and Chemical Constitution.—We have just drawn up a chemical classification of the hypnotics. From a practical point of view this is a most important step, but a chemist engaged in tackling therapeutical problems should aim also at including in his general education some acquaintance with the theories put forward to explain the physiological action of drugs.

As has been stated, narcotic activity depends essentially on physical factors, and above all on the partition coefficient between fat and water. But precise laws, by which the physical properties of a given compound may be foretold, have not yet been formulated, except in isolated cases. Consequently theories of the action of hypnotics cannot help us very much, and our plans of research cannot depend so much on them as on conclusions drawn from particular examples already investigated.

Thus, none of the therapeutic properties of either Veronal, or Bromural, or Sulphonal, could be predicted, but now that they are known we may expect similar activity to be shown by related compounds. So before dealing with Overton's theory of the action of hypnotics, certain typical pharmacological researches along these lines will be reviewed.

The ureides of the brominated fatty acids, *e.g.*, Bromural and Adaline, will serve as illustrative examples. Adaline is a caproic acid derivative with the bromine atom in the α position and with two ethyl groups attached to the same carbon atom :

The hypnotic dose with dogs is 0·30 gm. per kilo. body-weight. The solubility in water is 0·15% at 37°; in fatty oil it is 2·39% at the same temperature. The partition coefficient is therefore 1·40. It was interesting to see if isomerides of this compound were equally hypnotic. Tiffeneau and Ardely studied the ureide of the α-bromo derivative of *normal* caproic acid. Here the solubility in water was very low, viz., 0·033%, and the partition coefficient, 0·1. The compound possessed no hypnotic properties.

Van Eckout drew the same conclusion as Tiffeneau and Ardely from his investigation of valeric acid derivatives. Thus, α-bromovalerylurea is not a hypnotic; α-bromo-*iso*-valerylurea is a hypnotic, it is Bromural; α-bromo-methyl-ethyl-acetylurea is even more active. The partition-coefficients (fatty oil/water) for these three compounds are, respectively, 0·64, 1·33 and 1·90.

Beyond the C_6 acids very few experiments have been made. However, it has been found (Tiffeneau) that the ureide of the monobromo derivative of *normal* lauric acid is almost insoluble and is physiologically inactive. Even the C_5 and C_6 series are a long way from being completely investigated.

These experiments point to one conclusion, namely, that the group $\cdot CHBr \cdot CO \cdot NH \cdot CO \cdot NH_2$ is not itself active. "It may perhaps have latent hypnotic properties, but they only display themselves when certain solubility conditions are satisfied, and these depend, above all, on the carbon chain, and in turn control the passage of the substance in question into the central nervous cells" (Tiffeneau).

Further, one may say that bromine has no specific action, as it may be replaced by chlorine or iodine. Indeed, if the presence of bromine (or halogen in general) were essential, the compounds with the highest proportion, *i.e.*, ureides of bromo acids of low molecular weight, should be the most active; yet we have just seen that among the acids below the C_5 series no activity is shown.

Yet these experimental results, interesting though they be, are far from covering the ground. Probably what would lead to more valuable conclusions would be a thorough-going study of all the possible monobromo derivatives of, for example, valeric acid. Here there are a dozen possible variations. Whoever undertook this job would indeed serve therapeutics well, and yet would have a fine piece of chemical work to his credit; because, curiously, although a number of bromovaleric acid derivatives have been investigated, in which the carbon chain has been varied, none in which the position of the bromine atom has been altered, nor even, so far as is known, any derivative of, *e.g.*, β-bromovaleric acid, has been examined.

Here is a list of the possible monobromovaleric acids, not including the optically active isomerides:

$CH_3 \cdot CH_2 \cdot CH_2 \cdot CHBr \cdot CO_2H$

$CH_3 \cdot CH_2 \cdot CHBr \cdot CH_2 \cdot CO_2H$

$CH_3 \cdot CHBr \cdot CH_2 \cdot CH_2 \cdot CO_2H$

$CH_2Br \cdot CH_2 \cdot CH_2 \cdot CH_2 \cdot CO_2H$

$(CH_3)_2 \cdot CH \cdot CHBr \cdot CO_2H$

$(CH_3)_2CBr \cdot CH_2 \cdot CO_2H$

$CH_2Br \cdot CH(CH_3) \cdot CH_2 \cdot CO_2H$

$CH_3 \cdot CH_2 \cdot CBr(CH_3) \cdot CO_2H$

$CH_3CH_2 \cdot CH(CH_2Br) \cdot CO_2H$

$CH_3CHBr \cdot CH(CH_3) \cdot CO_2H$

$CH_2Br \cdot CH_2 \cdot CH(CH_3) \cdot CO_2H$

$(CH_3)_2 \cdot C(CH_2Br) \cdot CO_2H$

In investigations of this kind it is necessary :

(i) To prepare the compounds ;

(ii) To study carefully their physical and chemical properties (solubility in water, in various other solvents, in fats ; extent to which the bromine atom is labile in presence of water or dilute alkali, etc.) ;

(iii) To determine the partition coefficient between fatty oil and water ;

(iv) To follow the elimination of the substance *viâ* the urine ;

(v) To see how the bromine is dispersed in the organism and how it is absorbed, principally in the blood and the brain ; [1]

(vi) To determine its hypnotic activity towards mammals (dog, cat), and towards fish and tadpoles.

Theory of the Action of Hypnotics.—We now turn to the various theories put forward to explain the action of hypnotics. There are here two questions to consider, viz. :

(1) The penetration of the hypnotic substance into the nerve cell ;

(2) The mechanism by which hypnotics, having once entered the nerve cells, bring about narcosis.

Only to the first of these questions has a satisfactory reply been given. The primary action of hypnotics, that is to say, their penetration into nerve cells, appears to be dependent on the possession of certain physical properties, while chemical properties, in the narrow sense, are of no account. In fact, this action is a function of the solubility of the substance in fats.

On this conclusion is based the theory put forward independently

[1] Valuable information on carrying out the necessary tests will be found in the following papers : Denigès and Chelle, *C. R. Soc. Biologie*, 1912, *145*, 101 ; Labat, *Thèse de Doctorat*, Bordeaux, 1912 ; Carnot, *C. R. Acad. des Sciences*, 1913, 197 ; 1914, *76*, 641 ; Damiens, *Bull. Soc. Pharmacie*, 1920, *27*, 609 ; 1921, *28*, 37.

of each other by Overton and Meyer, both of whom bring forward abundant experimental data to support it. The theory may be stated as follows :

(1) Any indifferent chemical compound, soluble in lipoids, may have a narcotic action on living protoplasm, provided that it can diffuse therein.

(2) The effect first becomes apparent, and then to the greatest extent, in cells containing a large proportion of lipoids, *i.e.*, where these substances are responsible for most of the cell's functions.

(3) The intensity of the narcotic action may depend, first, on the physical affinity between the narcotic substances in question and the lipoids ; and, secondly, on their affinity with other cell constituents, particularly with water. It may be expressed, therefore, in terms of the partition coefficient, which indicates how a substance disseminates itself in an emulsion of fat and water.

In brief, narcotics paralyse the protoplasm when they can pass into animal or vegetable cells in sufficient quantity; and the high proportion of lipoids in the constituents of nerve cells and the fact that lipoids will dissolve hypnotic substances make it evident that there is a close relationship between narcotic activity and solubility in fats.

Experiments have been made on the dialysis of drugs through artificial fatty membranes, the aim here being to devise a purely physical method of testing and classifying hypnotic compounds. The results obtained are of considerable interest.

To begin with, the permeability of a castor-oil collodion membrane to solutions of salts was investigated. It was found that the salt did not pass through the membrane if this contained more than 2·5% of castor oil, and only easily if there were less than 2%. The limit of permeability was between 2% and 2·5%.[1]

When, instead of solutions of salts, solutions of various drugs were placed in the dialysing vessel it was found that only hypnotic substances were able to pass through the membrane. Even such compounds as antipyrine or aspirin were held back, but veronal, sulphonal, trional and the like diffused out, some more slowly than others certainly, yet all without exception.

Some experimental details may be quoted here. The actual test is carried out by preparing a capsule or thimble of collodion containing 3% of castor oil ; introducing into it 10 c.c. of a saturated aqueous solution of the hypnotic compound ; immersing it in a small beaker containing 50 c.c. water, so that the level of liquid within and without the capsule is the same ; and after thirty-six hours titrating or otherwise determining the amount of hypnotic present in the outside solution. Working in this way it was found that :

[1] The amount of oil to add to the collodion doubtless depends on the composition of the collodion and the thickness of the membrane (Thienlin)

From a solution of—

Hedonal	containing	0·086	gm.	in 10 c.c.	there passed out	0·0524	gm.
Veronal	,,	0·081	,,	,,	,,	0·041	,,
Sulphonal	,,	0·025	,,	,,	,,	0·0074	,,
Aponal	,,	0·050	,,	,,	,,	0·022	,,
Neuronal	,,	0·10	,,	,,	,,	0·025	,,

Although no conclusion can be drawn from these results as to the action of hypnotics in the organism, yet one cannot fail to note the general concordance between them and those obtained by Overton and by Meyer in their studies of the distribution of hypnotic substances in a fat–water system.

The partition coefficient is not necessarily the simple ratio between the solubility in water and the solubility in the fat. The actual method of determining its value is as follows, and this serves also as a definition of the term for our purpose. A solution of the hypnotic compound in water, of a known titre, *e.g.*, 2%, is shaken with an equal volume of oil for several hours. The mixture is allowed to settle and the amount of the hypnotic remaining in the aqueous layer determined. Suppose that it is 0·2%. Then, for 1,000 parts : $\frac{\text{In oil}}{\text{In water}} = \frac{20 - 2}{2}$ = 9. For further details, Overton's book [1] on this subject should be consulted.

Overton's investigations are the most extensive yet made on hypnotics. His results have led him to put forward the generalisations to which reference has already been made in the section dealing with the classification of these compounds. Further discussion is unnecessary here ; [2] but the following points are worthy of notice :

When two isomeric esters are compared, for example, amyl acetate and ethyl valerate, it is observed that the latter has a much more potent hypnotic action than the former, and that this corresponds with a difference in physical properties. The penetration of hypnotics into the living cell finds, therefore, in Overton and Meyer's hypothesis and the precise experimental results they have brought forward, a satisfactory explanation. But the mechanism by which hypnotics act once they are inside the cell, with its mixture of lipoids and phosphatides, is still obscure ; although many explanations have been offered, none can be regarded as entirely satisfactory.

A number of possibly relevant facts have been observed, and one of these should be mentioned, without, however, inferring that it is significant. Certain basic dyestuffs, for example, methylene blue, form colourless *leuco*-compounds when reduced. Now Ehrlich observed that after an injection of methylene blue, the brain of a normal animal

[1] *Studien über Narkose*, Jena, 1901.

[2] According to Traube, surface tension is the most important factor determining the penetration of hypnotic substances into the cell.

was not coloured ; probably, therefore, the leuco compound had been formed. But the cortex of an animal anæsthetised with ether was found to be dyed a bright blue. Doubtless the explanation of this is that in the first case the nerve cells are systems in which active oxidation is taking place, and therefore they act as powerful reducing agents, while in the second case the cells are inactive and their reducing power diminished.

CHAPTER VI

LOCAL ANÆSTHETICS

FEW applications of chemistry to therapeutics have given results so satisfactory to investigators of all kinds as those with local anæsthetics for subject. Not only has every detail of the constitution of cocaine been revealed by analysis and its synthesis effected in confirmation, but a number of synthetic compounds, conceived by ingenious chemical brains and produced in substance from more and more simple components, have appeared as competitors with this alkaloid, and, in fact, would have displaced it altogether if its use for illicit purposes were not a chief reason for its commercial importance.

Most local anæsthetics are derivatives of amino-alcohols, or, more precisely, of compounds containing *at least one amino and one hydroxy* (alcoholic or phenolic) *group*, for the molecule may contain other groupings of not too pronounced a character, such as that of an ester (cocaine).

It seems necessary that the alcoholic hydroxyl should be esterified by an acid for anæsthetic properties to become manifest. Hitherto, the acid used has nearly always been benzoic or aminobenzoic, and one might say that all benzoic esters of amino-alcohols are anæsthetics to some extent. It will be seen later that the benzoyl radical has no specific qualities, but it does appear to be the most valuable.

Some special local anæsthetics, *i.e.*, those which act principally on the exposed dermis, such as *Orthoform*, are again both esters and amines; but here the amino group is a substituent in the acid part of the molecule and is not attached to the alkyl radical. It is obvious, in fact, that isomerism of this kind is possible, thus:

$C_6H_5{\cdot}CO_2{\cdot}CH_2{\cdot}CH_2N<$ = an aminoethyl benzoate,
$>N{\cdot}C_6H_4{\cdot}CO_2{\cdot}CH_2{\cdot}CH_3$ = an ethyl aminobenzoate.

Others are derivatives of phenolic ethers, for example, *Acoine*, *Holocaine*. Recently, in America, benzyl alcohol also has been introduced.

The following types, at least, must therefore be considered:

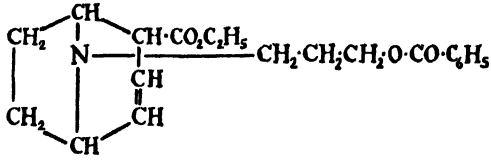
(1) $C_6H_5{\cdot}CO_2{\cdot}CH \cdot\cdot\cdot\cdot N\begin{matrix}R\\R\end{matrix}$ (Tropacocaine, *Stovaine*);

(2) $\begin{matrix}R\\R\end{matrix}\!\!>\!N{\cdot}C_6H_4{\cdot}CO_2{\cdot}CH_2 \cdot\cdot\cdot\cdot$ (*Anæsthesine*);

(3) $C_6H_5{\cdot}CO_2CH(CO_2CH_3){\cdot}N\begin{matrix}R\\R\end{matrix}$ (Cocaine);

(4) $>N\cdot C_6H_4\cdot CO_2\cdot CH \cdots N\begin{matrix}R\\R\end{matrix}$ (*Novocaine*);

(5) $R\cdot C\begin{matrix}=N-C_6H_4\cdot OR\\ NH\cdot C_6H_4\cdot OR\end{matrix}$ (*Acoine*);

(6)  (*Eccaine*—Braun);

(7) $C_6H_5\cdot CH_2\cdot OH$ (Benzyl alcohol).

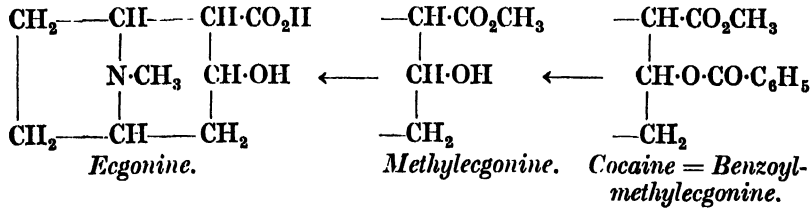
Lastly, just to show how anæsthetic properties are distributed among the different families of chemical compounds, aminopyridine may be mentioned.

Besides compounds of the above classes, whose constitution is definitely established, there are a number of anæsthetics among the naturally occurring alkaloids of as yet unknown constitution, such as ibogaine, corynanthine, yohimbine. Here, of course, it is impossible to say how far anæsthetic activity is associated with specific atomic groupings.

We will examine these various groups of compounds one after the other, paying particular attention to tropacocaine, the *Eucaines*, and cocaine itself—the typical local anæsthetic, the subject of some most important investigations of which the outlines, at any rate, should be known.

Cocaine.—Cocaine was isolated by Niemann in 1860, and has since been studied by Lossen, Einhorn, Ladenburg and Merling, to mention only important names, but it remained for Willstätter to carry through the outstanding investigations which conclusively established its constitution.

Cocaine is an ester of benzoic acid, for when it is treated with methyl-alcoholic hydrogen-chloride, it loses benzoic acid, which reappears as methyl benzoate, and yields the methyl ester of an amino-hydroxy-acid, namely *ecgonine*:



Cocaine contains a second ester-grouping, viz., CO_2CH_3, for when treated with aqueous hydrochloric acid, not only is the benzoyl radical hydrolysed off, but also the methyl group and ecgonine, the amino-hydroxy-carboxylic acid, is produced directly.

Cocaine is derived from a tertiary amine, for it takes up only one molecular proportion of methyl iodide.

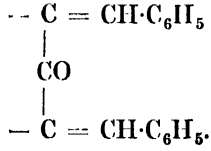
The nitrogen atom in cocaine is attached to only one methyl radical, for when the substance is oxidised by appropriate methods, it loses one methyl group to give norcaine, and further, when it is demethylated by treatment with hydriodic acid only one methyl group is split off.

The above *facts* being known, there remained to decide how the heterocyclic nucleus was constituted, and this was the aim of Willstätter's brilliant research, the essential results of which will now be outlined.

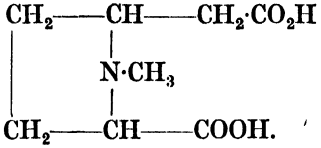
Ecgonine was oxidised with chromic acid and produced an amino ketone, namely, *tropinone*. The hydroxyl must therefore form part of a secondary alcoholic grouping. This discovery had another interesting result, in that it showed how cocaine and atropine were related, since *tropine*, the parent base of atropine, also gave *tropinone* when carefully oxidised. This relationship is demonstrated also by the fact that ecgonine, or better its dehydration product, *anhydroecgonine*, loses carbon dioxide when heated with hydrochloric acid to 280°, forming *tropidine*, that is to say, anhydrotropine, a product of the dehydration of tropine.

To establish the constitution of tropine would carry one a long way towards that of cocaine; returning therefore to tropinone, it was found that:

The ketonic group in tropinone is situated between two methylene groups, for, on treatment with nitrous acid, it gives a dinitroso-derivative, and with benzaldehyde it forms a dibenzylidene compound:



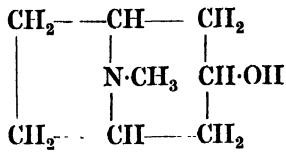
The tropinone nucleus contains a ring of seven carbon atoms, i.e., *tropinone is a derivative of cycloheptane*, for when it is oxidised it gives tropinic acid:



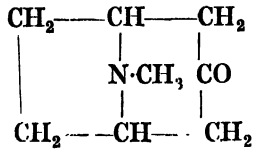
When tropinic acid is esterified, treated with methyl iodide and degraded by Hofmann's exhaustive methylation method, an unsaturated pentadiene-dicarboxylic acid is obtained which, when reduced, gives pimelic acid, identical with the synthetic product,

$$CO_2H{\cdot}CH_2{\cdot}CH_2{\cdot}CH_2{\cdot}CH_2{\cdot}CH_2{\cdot}CO_2H.$$

These transformations find their explanation only in the following formulæ :

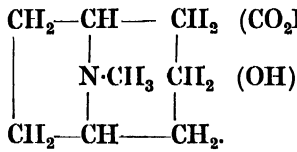


Tropine.



Tropinone.

Ecgonine, therefore, has the same nucleus as tropine, and its hydroxyl group occupies the same position as in that compound. There remains to locate the carboxyl group, and the position of this is already indicated. As ecgonine is ultimately oxidised to tropinic acid, a dibasic acid and a derivative of methylpyrrolidine, the carboxyl group cannot be in this pyrrolidine nucleus, for if it were, tropinic acid would be tribasic. But, again, it is not attached to the same carbon atom as the hydroxyl group, since Willstätter synthesised this compound and found it to be quite different from the natural product. *Ecgonine is therefore simply tropanolcarboxylic acid* (tropane being hydrotropidine), as shown here :



Willstätter has synthesised cocaine (optically inactive) starting from suberone, thus demonstrating conclusively its constitution as a derivative of *cyclo*-heptane.[1]

Tropacocaine.—In 1891 Giesel found that the leaves of coca trees grown in Java contained small quantities of another alkaloid, which he called tropacocaine. This substance was investigated by Liebermann, who obtained analytical results which showed that its empirical formula was the same as that of benzoyl-tropine, and that, in fact, it would be hydrolysed to give benzoic acid and a base isomeric with tropine, namely, *pseudo*-tropine. To Willstätter, again, is due our knowledge of the nature of the isomerism of these two compounds : they were shown by him to be the *cis* and *trans* forms of an optically active amino-alcohol.

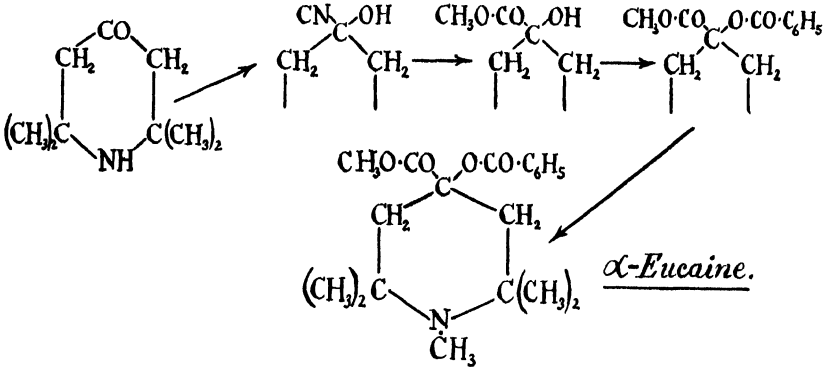
Thus, both tropine and *pseudo*-tropine were found to yield tropinone when oxidised by Beckmann's reagent (chromic acid in glacial acetic acid). Moreover, when tropinone was reduced the product was either tropine or *pseudo*-tropine, depending on the method employed, and again, when tropine was heated with sodium amyloxide in amyl alcoholic solution it was transformed into *pseudo*-tropine. *Pseudo*-tropine is therefore the stable form. Both bases could be benzoylated ; the benzoyl derivative of *pseudo*-tropine was found to be identical with

[1] See now *Annalen*, 1923, *434*, 111 (Willstatter, Wolfes, and Mader).—*Tr.*

the naturally occurring tropacocaine, a powerful anæsthetic. The benzoyl derivative of tropine seems to be both a weaker anæsthetic and less toxic, but now that local anæsthetics are becoming more intimately known, this statement needs confirmation. However, tropine derivatives do possess properties which are lacking in those of *pseudo*-tropine: they show mydriatic activity. The phenylglycollyl derivative is the most potent in this respect.

Eucaines.—The influence of stereoisomerism on physiological properties has now been demonstrated in one instance. Similar phenomena will be found to characterise the Eucaine group.

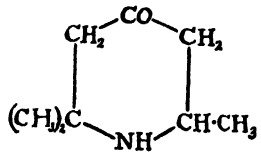
α-Eucaine is a cocaine in the sense that its molecule contains an amino-grouping, a hydroxyl group as the benzoyl ester, and a carboxyl group as the methyl ester. The parent compound of α-Eucaine is triacetonamine, the condensation product formed when acetone and ammonia react together. When triacetonamine is treated with hydrocyanic acid the cyanhydrin is produced. This is then simultaneously hydrolysed and esterified by the action of methyl-alcoholic hydrochloric acid. The hydroxyl group is benzoylated and finally the imino group is methylated, thus:



The molecule of α-Eucaine is symmetrical and it can form no stereoisomerides, optically active or otherwise. It is indeed obvious at the first glance that whether the benzoyl group be above or below the plane of the ring (the carbomethoxy group being then on the opposite side) makes no difference—it is always flanked by the four symmetrically situated methyl groups. There is therefore no α-Eucaine corresponding with *pseudo*-tropine.[1]

If one of the four methyl groups in triacetonamine be replaced by hydrogen, the symmetry of the molecule is destroyed. Vinyldiacetonamine (Heintz) is the common name of the compound of this constitution. It is obtained by condensing diacetonamine and acetaldehyde:

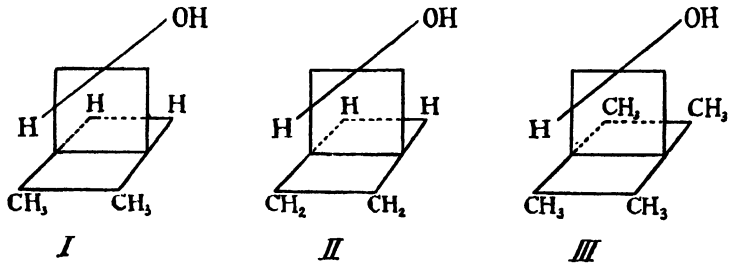
[1] It should be noted in passing that cocaine may exist in two stereoisomeric forms, and that there is no evidence that naturally occurring cocaine is not derived from *pseudo*-tropine, although its mydriatic properties would suggest its relationship with tropine. (But see now Willstätter and others, *loc. cit.*—*Tr.*)



This compound is a ketone, and the corresponding alcohol ("alkamine") may be obtained theoretically in two ways; either by reduction (Fischer) or by treating the amine, 2 : 2 : 6-trimethyl-4-amino-piperidine, with nitrous acid, $R{\cdot}NH_2 \rightarrow R{\cdot}OH$ (Harries).

Actually the products obtained by these two methods were not identical. That from the reduction of the ketone was a mixture and was separated into two isomeric compounds, melting at 138° and 168° respectively; the second of these was found to be the same as that obtained by Harries *via* the amine. Continuing the investigation, Harries showed that the isomeride melting at 168° was transformed into that of m.p. 138° when it was treated with sodium amyloxide in amyl alcohol, the conversion being analogous to that of tropine into *pseudo*-tropine, studied by Willstätter. This parallelism appears again in the physiological properties. The benzoyl derivative of the stable form is a potent anæsthetic with no mydriatic action; it is *β-Eucaine*. On the other hand, derivatives of the unstable form possess mydriatic properties, particularly the N-methyl phenyl-glycollyl derivative, which is known as *Euphtalmin*.

The diagrams below will demonstrate more clearly how it is that triacetonalkamine (III) cannot form stereoisomerides, while vinyldiacetonalkamine and tropine can (I and II).



I *II* *III*

In III transposition of the H— and the —OH does not alter the configuration.[1]

Stovaine.—The splendid research work which led to the synthesis of the eucaines had as its aim the manufacture of artificial anæsthetics with, as far as possible, similar properties to those of the natural products, cocaine and tropacocaine. The discovery of *Stovaine* by E. Fourneau resulted from considerations of quite another order.

[1] These diagrams are intended to indicate how the functional groupings are arranged in space. The student will find stereochemistry more intelligible if he makes models (cork and wire) for himself.—*Tr.*

Fourneau reasoned that as the piperidine nucleus always appears to confer toxic properties on the compounds of which it forms part, and yet seems to have no outstanding qualities as "anæsthesiophore", since a number of local anæsthetics are known which are not derived from it, so it should be possible to prepare compounds in which the groupings characteristic of cocaine and tropacocaine are attached to a simpler skeleton. His investigations, therefore, started with the simpler amino-hydroxy-acids and amino-alcohols with tertiary amino groups. The simplest member of the first class is dimethylaminolactic acid :

$$(CH_3)_2N{\cdot}CH_2CH(OH){\cdot}CO_2H.$$

It is evident that in this compound there are all the functioning atomic groupings characteristic of ecgonine, and so the benzoyl derivative of its methyl ester :

$$(CH_3)_2N{\cdot}CH_2{\cdot}CH{\cdot}CO_2CH_3$$
$$\diagup$$
$$O{\cdot}CO{\cdot}C_6H_5$$

may be regarded as cocaine reduced to its simplest terms.

This substance was not actually prepared because of certain practical difficulties, and, moreover, it was found in the course of the investigation that compounds whose molecules contained acid radicals clustered near the amino group were not well adapted to injection. Nevertheless, the next higher homologue was prepared, as being very interesting from a theoretical point of view; this compound, the N-dimethyl derivative of amino hydroxy*iso*butyric acid, is easily obtained from dimethylaminoacetone, by a series of reactions exactly similar to those by which eucaine was produced from triacetonamine. The hydrochloride of the benzoylated methyl-dimethylamino-hydroxy-*iso*-butyrate :

$$\begin{array}{l} CH_2{\cdot}N(CH_3)_2HCl \\ | \\ CH_3{\cdot}C{\cdot}O{\cdot}CO{\cdot}C_6H_5 \\ | \\ CO_2CH_3 \end{array}$$

is a potent anæsthetic, quite feebly toxic, but very irritating to the tissues. The amino-hydroxy-acid series was therefore abandoned for the time.

The simplest compound analogous to tropacocaine in the above sense is dimethylaminoethyl benzoate :

$$(CH_3)_2N{\cdot}CH_2{\cdot}CH_2O{\cdot}COC_6H_5.$$

This substance, already known, possesses very feeble anæsthetic properties, and the preparation of its homologues for systematic investigation was at one time not an easy matter. Indeed, amino-alcohols, as a class, were compounds of which very little was known; some, but not many, of the aliphatic series, and still fewer of the aromatic series, had been studied. Now, with the aid of Grignard's

reaction, an extensive series of these compounds has been prepared and several of them have been investigated by Fourneau.

Application of the Grignard reaction furnishes three types of amino-alcohol according as (1) epichlorhydrin, (2) chloracetone, or (3) β-chloropropionaldehyde is used, viz.,

(1) $R{\cdot}CH(OH){\cdot}CH_2{\cdot}N(CH_3)_2$;
(2) $CH_3{\cdot}CR(OH){\cdot}CH_2{\cdot}N(CH_3)_2$
(3) $R{\cdot}CH(OH){\cdot}CH_2{\cdot}CH_2{\cdot}N(CH_3)_2$.

The actual preparation involves treating a chlorohydrin with a secondary amine, thus:

$$CH_2\text{-}CH\ CH_2Cl \xrightarrow{R{\cdot}Mg{\cdot}Br} R{\cdot}CH_2{\cdot}CH(OH){\cdot}CH_2Cl \xrightarrow{NH(CH_3)_2} R{\cdot}CH_2CH(OH){\cdot}CH_2{\cdot}N(CH_3)_2$$

$$CH_3{\cdot}CO{\cdot}CH_2Cl \xrightarrow{R\,Mg\,Br} \begin{matrix} R \\ CH_3 \end{matrix}\!\!>\!C(OH){\cdot}CH_2Cl \xrightarrow{NH(CH_3)_2} \begin{matrix} R \\ CH_3 \end{matrix}\!\!>\!C(OH){\cdot}CH_2{\cdot}N(CH_3)_2$$

$$CHO{\cdot}CH_2{\cdot}CH_2Cl \xrightarrow{R\,Mg\,Br} R{\cdot}CH(OH){\cdot}CH_2CH_2{\cdot}Cl \xrightarrow{NH(CH_3)_2} R{\cdot}CH(OH){\cdot}CH_2{\cdot}CH_2{\cdot}N(CH_3)_2.$$

The first of these reactions cannot be carried out with aliphatic compounds alone. Of the other two, Fourneau chose the first for extended application, because at the time acrolein was not readily obtained in quantity; later, when it became possible to investigate both series, the results only served to confirm his choice.

Thus it is that **Stovaine** is a member of the series of amino derivatives of *tertiary* alcohols. It is the hydrochloride of methylethyldimethylaminomethylcarbinol benzoate:

$$\begin{matrix} & & CH{\cdot}N(CH_3)_2{\cdot}HCl \\ & & | \\ C_2H_5{\cdot}&C{\cdot}O{\cdot}COC_6H_5 \\ & & | \\ & & CH_3. \end{matrix}$$

Its preparation need not be described in detail, as it is given in the practical part of the book.

Alypine.—

$$CH_3{\cdot}CH_2{\cdot}C(CH_2{\cdot}N(CH_3)_2HCl)(CH_2{\cdot}N(CH_3)_2){\cdot}O{\cdot}COC_6H_5 \qquad \text{(Fritz Hofmann)}$$

This is dimethylamino-stovaine. It is somewhat toxic and is used to only a limited extent. It is made by taking dichloracetone through the same series of reactions as produce Stovaine from monochloracetone.

Anæsthesine and the Orthoforms, Nirvanine, Novocaine.—All these anæsthetics are members of one family: they are derivatives of amino-benzoic acid.

Anæsthesine (Bing and Kobert) is ethyl *p*-aminobenzoate. It is almost exclusively used in making pastilles for the throat. *Scuroform* is *n*-butyl *p*-aminobenzoate.

Novocaine (Stolz) is diethylaminoethyl *p*-aminobenzoate hydrochloride, $NH_2 \cdot C_6H_4 \cdot CO_2CH_2 \cdot CH_2 \cdot N(C_2H_5)_2 \cdot HCl$. Whilst the corresponding ester of benzoic acid itself is not worth calling an anæsthetic, its amino derivative, *Novocaine*, is very definitely active. Nevertheless, it is never employed by itself as it is too diffusible, but is always administered in conjunction with adrenaline, the latter causing a contraction of the blood-vessels and so retaining the anæsthetic near the place of injection.

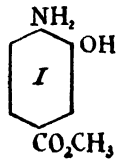
Novocaine was selected from a widely varying group of esters of *meta*- and *p*-aminobenzoic acids with numerous amino alcohols. It may be made in several ways, thus :

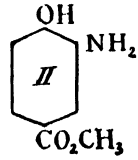
(1) The chloroethyl ester of *p*-nitrobenzoic acid is treated with diethylamine, $NO_2 \cdot C_6H_4 \cdot CO_2CH_2 \cdot CH_2Cl + NH(C_2H_5)_2$, and the nitro group then reduced. This is an unsatisfactory method, because the replacement of the chlorine by the diethylamino group is accompanied by a secondary reaction in which some of the nitrobenzoyl radical is split off and reappears as diethylnitrobenzamide ;

(2) Anæsthesine is heated with diethylaminoethyl alcohol ;

(3) Diethylaminoethyl alcohol is esterified by treatment with *p*-nitrobenzoyl chloride and the product reduced.

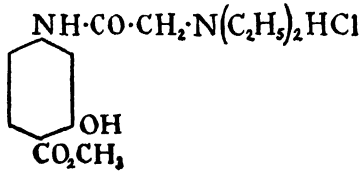
The *Orthoforms* are esters of aminohydroxybenzoic acids. The older Orthoform is methyl *p*-amino-*m*-hydroxybenzoate (I), the newer one—*Orthoform new*—is the *m*-amino-*p*-hydroxy derivative (II) :





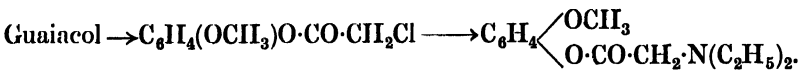
Orthoforms act only when the surface nerves are laid bare. They are used in the form of free base, not as a salt, and applied as a powder, or in ointments for dressing ulcers, sores and chapped places, etc. Orthoform salts are strongly acid to litmus ; attempts have been made to remove this defect by replacing the nuclear amino group by one forming part of an aliphatic radical (Einhorn).

Nirvanine is the hydrochloride of the diethylglycocoll derivative of methyl *p*-amino-*o*-hydroxybenzoate :



The method by which Nirvanine is obtained from an orthoform is a general one widely employed. It consists in treating the aromatic amine or phenol with chloracetyl chloride and then replacing the chlorine by an amino group:

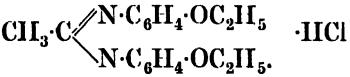
Nirvanine.



Guaiasanol.

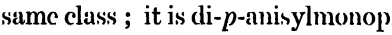
Holocaine.—*Holocaine* belongs to a series of compounds quite different from those above, which are, more or less, aminoalcohols; yet there are some points of resemblance. It is an aminophenol derivative, the hydroxyl group being, however, not benzoylated, but alkylated, *i.e.*, the substance is a phenolic ether. Thus it is really a phenetidine derivative. It is not the only aminophenolic ether to show anæsthetic activity; quinine is another example, and possibly all aminophenolic ethers have some anæsthetic properties.

Holocaine is prepared by condensing together phenetidine and phenacetine in the presence of phosphorus trichloride.



It is a powerful anæsthetic with a rapid action, but being more poisonous than cocaine, it is scarcely used.

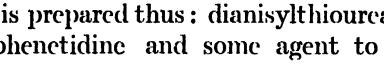
Acoine is in the same class; it is di-*p*-anisylmonophenetyl guanidine hydrochloride:



This compound is prepared thus: dianisylthiourea (-thiocarbamide) is treated with phenetidine and some agent to take up sulphur (Goldschmidt).

Acoine is not as toxic as cocaine although quite as active an anæsthetic, but it irritates the tissues and is sparingly soluble.

There should also be mentioned *Apothesine*, which is the cinnamic ester of diethylaminopropyl alcohol:



and *benzyl alcohol*, which has been put forward in America (Macht). The latter does not, however, appear capable of wide application, for it is an irritant.

Eccaine.—The most recent anæsthetic is a derivative of ecgonidine, introduced by von Braun.[1] It is stated to be only $\frac{1}{5}$ as poisonous as cocaine and a more active anæsthetic. In this new series anæsthetic

[1] *Ber.*, 1918, *51*, 238.

potency depends on the size of the aliphatic radical attached to the nitrogen atom and decreases as this increases.

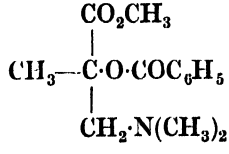
THEORETICAL CONSIDERATIONS

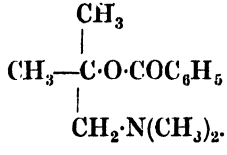
The Constitution and Anæsthetic Action of Local Anæsthetics.—We have now studied local anæsthetics from the chemical point of view in as much detail as is necessary for our purpose, and can turn our attention to the conditions regulating their essential physiological properties.

Considering cocaine first of all, it can be said that if the N-methyl group be replaced by hydrogen, no appreciable alteration in physiological properties will result. Actually β-Eucaine has no N-methyl group. All the same, when preparing these anæsthetic compounds, derived as they are from amino alcohols, it is an advantage to have the amino group tertiary, as then the hydroxyl group may be more easily benzoylated, there being no risk of a second benzoyl being taken up by the amino group.

If cocaine be subjected to hydrolysis so that the methyl group is detached and benzoyl-ecgonine formed, anæsthetic activity is lost, for it is a general rule that acidic substances have no pronounced physiological properties and are usually not very poisonous. Tyrosine, $HO{\cdot}C_6H_4{\cdot}CH_2CH(NH_2){\cdot}CO_2H$, is a typical case in point; the acid itself is not toxic, but its methyl ester is a violent poison. The replacement of methyl by other alkyl radicals appears not to bring about any noteworthy alteration in properties, but investigation along these lines has not been carried very far; aromatic radicals, for example, have not yet been studied.

Moreover, is there really any need for the carbomethoxy group to be present? Apparently not, as it is absent from many other anæsthetics. And when one considers that cocaine is more toxic than tropacocaine, it may be thought that the carbomethoxy group is actually detrimental, but then it must not be forgotten that cocaine is not a derivative of *pseudo*-tropine, but of tropine (at least if any conclusion can be drawn from its mydriatic properties), and its poisonous nature may be thus explained. On the other hand, it is possible occasionally to compare two closely related anæsthetics, differing only in that a methyl group in the one is replaced by a carbomethoxy group in the other, and in one of these rare instances it was demonstrated that the carbomethoxy derivative was much less toxic than the other (Fourneau). The two compounds in question are represented by the formulæ:





There is, therefore, not only no justification for ruling out completely the amino-hydroxy-acids as starting materials in local anæsthetic research, but, on the contrary, this observation suggests that they should be given preferential consideration.

A most important question will now occupy our attention. It is essential that the hydroxy group be esterified; neither a free amino alcohol nor an amino-hydroxy-carboxylic ester is an anæsthetic of itself. But for this purpose is benzoic acid indispensable? Not at all; aminobenzoic acid is more active, as has already been shown. In fact, the benzoyl radical may be replaced by any homologue or aromatic congener. So far, however, no advantage has been gained in this way, but it does seem that cinnamic acid intensifies the anæsthetic property. Enough experiments have not yet been made on the replacement of the aromatic acid by fatty acid radicals for sound conclusions to be drawn. Above a certain molecular weight, fatty acid derivatives certainly are anæsthetics (Fourneau). Hydrobenzoic acid, which is really an aliphatic acid, yields strongly anæsthetic esters (Madinaveitia, Cano and Ranedo).

Much depends on the nature of the alkyl radical. Derivatives of primary alcohols have hardly been investigated. They are not easily prepared, as the only general method consists in reducing the esters of dialkylamino derivatives of fatty acids (Gault). Novocaine is the only example of a primary alcohol derivative[1]; it is quite mildly anæsthetic, but as no isomerides exist, no comparisons can be made. To make a comparison derivatives of butyl alcohol should be investigated; here at least three isomerides are possible, *e.g.*:

$$CH_2(OH)\cdot CH_2\cdot CH_2\cdot CH_2\cdot N(CH_3)_2$$
$$CH_3\cdot CH_2\cdot CH(OH)\cdot CH_2N(CH_3)_2$$
$$\begin{matrix} CH_3 \\ CH_3 \end{matrix}\!\!\diagdown\!\!\diagup C(OH)\cdot CH_2\cdot N(CH_3)_2.$$

The only published instance of two isomerides being directly compared is given in a paper by Launoy and Fuyimori. The two compounds in question were the benzoyl derivatives of the following amino-alcohols:

$$CH_3\cdot CH_2\cdot CH(OH)CH_2\cdot CH_2\cdot N(CH_3)_2$$
$$CH_2\cdot CH_2\!-\!C(OH)\!-\!CH_2\cdot N(CH_3)_2 \text{ (\textit{Stovaine})}$$
$$\quad\quad\quad

another a matter of importance ? Most decidedly so—because it is one of the factors on which the physical and chemical properties depend ; the salts of benzoyl esters of β-amino-alcohols are acid to litmus, but those of corresponding esters of amino-alcohols with the groups more widely separated are quite neutral in reaction. This is a little known generalisation which may have considerable practical importance in establishing the constitutions of certain alkaloids. For example, long before cocaine and tropacocaine were thoroughly investigated one could justifiably have asserted that the hydroxyl and alkyl-amino groups were not adjacent, but that in the case of ephedrine the contrary held and the two groups were close to one another. In research work directed towards new local anæsthetics interest should therefore be concentrated chiefly on the γ-amino-alcohols. Nevertheless, the transitory irritant effect observed when the β-derivatives are dropped into the eye and the slight pain following hypodermic injection cannot be attributed to their acidic nature, as these disadvantages are possessed equally by both isomeric compounds investigated by Launoy (see above), to quote but one example. The caustic action on the tissues and the irritation to the eye certainly depend on the properties of the free base (the base is liberated immediately in either blood or tears). What effect the varying separation of the two active groups has upon anæsthetic activity is not known.

Problems relating to Local Anæsthetics still unsolved. The Need for Research. The Characteristics of a Good Anæsthetic.

Already in this chapter questions that are yet unsettled and obscure have been referred to, and now we may review them again by suggesting directions in which research may profitably be undertaken.

(1) Vary the acid used for esterification ; make a comparison, if necessary, of stereoisomerides with the same acyl group.

(2) Study amino-hydroxy-*acids* and compare them with the corresponding amino-alcohols.

(3) Investigate derivatives of aromatic alcohols.

(4) Study isomeric amino-alcohols ; for example, prepare for comparison the isomerides of the C_6-amino-alcohol series (there are nine of them, not counting optical isomerides).

(5) Compare the stereoisomeric forms of a particular amino alcohol.

This kind of investigation is of outstanding importance ; at present little has been done along such lines.

Yet it may be asked : what good will come of this sort of research ? Do we not possess excellent anæsthetics and are not better ones difficult to make ? The answer to such questions is that it *is* possible to effect great improvements and that the *ideal* anæsthetic has yet to be found.

What should be its properties ? A good anæsthetic should :

(i) Be easily soluble in water ;

(ii) Be capable of undergoing sterilisation by heat in aqueous solution ;

(iii) Be almost tasteless ;

(iv) Possess no pronounced toxic properties ;

(v) Cause no smarting sensation when introduced under the eyelids or injected under the skin ;

(vi) Have a strong anæsthetic action, intense and continuous, but not violent ;

(vii) Have no permanent effect on the nerve fibres, and should pass away completely ;

(viii) Show vasoconstrictive activity and render the tissues and surfaces with which it comes in contact bloodless ;

(ix) Be not too costly ;

(x) Not form precipitates with heavy metals (mercury).

All these conditions, except (iv) and (x), are satisfied by cocaine. The most widely used of the other anæsthetics, Stovaine and Novocaine, both fulfil condition (iv), which indeed is the most important, but neither of them complies with all the others.

Thus the ideal anæsthetic has yet to be discovered. One might now ask why it is that a given substance manifests anæsthetic activity. This question is not easily answered.

Local anæsthesia consists in diminishing or totally suppressing the action of the sensory nerves. The motor nerves and muscles should not be affected. Thus the local anæsthetic has actually an affinity for the peripheral nervous system and paralyses it without permanently altering it. What then happens when an anæsthetic is injected under the skin or administered beneath the eyelid ? First of all, the base is liberated from its salt by interaction with the alkaline body fluids. In the form of base the anæsthetic is taken up by the nerve fibres, but the exact mechanism of this process is not known. Quite likely here, again, the lipoids play an important part and the action may be (at the beginning at least) a purely physical one. One thing is certain, viz., that little by little the alkaloid is removed by the continuous irrigation taking place, and the nerve gradually recovers its normal activity. So it is possible that all substances dissolving by preference in the lipoids of the sensory nerves when injected may function as anæsthetics. Most hypnotics behave in this way and all are local anæsthetics to some extent. It has already been mentioned that benzyl alcohol is well endowed with this property. Dihydroxyphenoxypropane (glycerol monophenyl ether) is likewise a fairly potent anæsthetic, and everything suggests that many compounds, as yet untested, will come into the same class.

Note.—Of still more recent products the following merit consideration:

Butyn, $NH_2 \cdot C_6H_4 \cdot COO \cdot CH_2 \cdot CH_2 \cdot CH_2 \cdot N(C_4H_9)_2 \cdot HCl$ (or H_2SO_4).
(See *Applied Chemistry Reports*, 1923, VIII., 159.)

Tutocaine (Bayer), $CH_3 \cdot CH(O \cdot CO \cdot C_6H_4 \cdot NH_2)(1:4)$
$CH_3 \cdot CH \cdot CH_2 \cdot N(CH_3)_2$, HCl.
(See *Deutsche Medizinische Wochenschrift*, 1924, p. 539.)

Psicaine (Willstätter), acid tartrate of $d - \psi$ – cocaine, synthetic (see footnote on p. 58).

CHAPTER VII

ANTISEPTICS

It is not easy to deal with the subject of antiseptics, and to do so satisfactorily would need volumes, not a single chapter. So much preliminary knowledge is needed; one ought to know exactly the composition of the contents of the living cell, what chemical reactions and physical transformations are taking place inside it, and how the components behave towards foreign substances with which they come in contact.

A living cell may be killed, not only if its interior physical and chemical equilibrium is upset, and sometimes very little suffices to do that, but also if its environment is lacking in certain essential nutritive elements. [Zinc is needed by *Penicillium glaucum* (Raulin), and probably by other organisms (Delezenne), arsenic (Armand Gautier, Bertrand) and copper (Maquenne, Fleurent) are also required.]

While the absence of a necessary component will prevent or hinder development, the presence of certain substances in the environment, even in infinitesimal traces, will kill the cell or organism. One need only refer to the classical experiments of Nägeli on algæ, and those of Delezenne and Suzanne Ledebt on fishes. Algæ will live in pure distilled water if it has been distilled in glass apparatus, but they die if it has passed through a copper pipe or tap, although no trace of copper can be detected in the water by any known method. Delezenne and Suzanne Ledebt have shown that the same holds for fishes. Minnows immersed in ordinary distilled water died in two or three hours, but when placed in water redistilled in glass apparatus, they lived for at least twenty-five days. Curiously enough, they also lived comfortably in ordinary distilled water if a little spring water were added.

Again, Sauton states that if a piece of silver foil be put into a freshly made culture of tuberculosis bacilli, the culture does not develop, although no silver can be detected in the filtered liquor.

Even atmospheric oxygen, indispensable for life as it is, is poisonous in quite small doses for anærobic bacteria if the actual quantity is more than they can stand. They choose rather to get their oxygen in their own special way, and thus obtain only so much as their development requires.

Furthermore, all the conclusions resulting from the study of vitamins and accessory-food factors may possibly be extended to micro-organisms.

Thus considered, the meaning of the term antiseptic is widened;

it must include any substance or any medium unsuited to microbic life. Generally, however, *when antiseptics are spoken of, it is bactericides that are meant,* that is to say, substances *speedily fatal* to bacteria. So the name antiseptic should really be kept for any substance which hinders the development of, without necessarily killing, bacteria, and the term bactericide used if the microbes actually perish. Here, however, we will fall in with the common custom and use the two terms indiscriminately.

Antiseptics or bactericides are met with among inorganic substances and among organic products, both synthetic and natural. One should distinguish between internal and external antiseptics; the former fall within the province of chemotherapeutics of infectious disease. Only external antiseptics will be discussed here, and principally those not from the mineral kingdom.

Many antiseptics have an affinity for albumins or fats, with which they form more or less stable compounds. These compounds are usually less active than the parent substance. So the strength of an antiseptic in water or dilute salt solution is out of all proportion with that which it can exert in presence of the complex body fluids. This important fact has several consequences: in the first place, it is most difficult, even with antiseptics of great *in vitro* strength, to attack the actual agent of a contagious disease by intravenous or intramuscular administration (and all the more so if the mouth is the chosen channel); secondly, it is not easy to produce and maintain even imperfectly antiseptic conditions when infected wounds are undergoing treatment; and lastly, as antiseptics have at least as much affinity for the cells of the organism as for bacteria, it is at present impossible to ensure complete sterilisation without harming the living cells and impeding the processes of healing. And the solution of these problems is rendered more difficult to attain by the fact that many micro-organisms assume a more resistant form (spores) when they become subject to unfavourable conditions. Moreover, many of them are laden with waxy matter and encased in chitin, all of which protects them from attack, while the cells of the organism are less tough and lack an envelope, properly so called. It should be added, also, that bacteria can habituate themselves to antiseptics; sometimes they even adapt themselves so far as to augment their metabolic functions and their virulence.[1]

The action of antiseptics on micro-organisms thus manifests itself, unless very large relative amounts are used, in various contradictory ways, and besides those mentioned there are many others. The bacteria may not be killed, but fail to proliferate, or lose the power of forming spores. In the case of charcoal, for example, the anti-spore-forming action is accompanied also by a depression of the virulence.

[1] Even in mercuric chloride solution, in 15% caustic soda, or in 10% sulphuric acid, moulds and bacteria may thrive and develop.

Some microbes become incapable of forming their characteristic colouring matter, others lose their mobility, and others again their fermentative powers.

Knowing all this, it is not surprising that the meaning of the term antisepsis oscillated and changed about during the war—during a period which provided opportunities for experimental work on a scale undreamed of before. Diversity prevailed, above all, in opinions on the efficacy of antiseptics; some had no faith at all, others were too enthusiastic; until a mean was struck which may be expressed as follows: In using antiseptics, it should be made a rule that all mortified and badly-infected tissues be cut away, so that the antiseptic can be applied in small quantities and yet produce the desired effect; this holds both for small superficial wounds and for long-standing local sores (osteomyelitis).

Actually, the ideal antiseptic has yet to be found. Perhaps a different one will be needed for every infectious agent, because a substance acting as a general poison for micro-organisms will also be a poison for any other living cells. In every case the satisfactory antiseptic should attack the infectious agent inside the tissues without appreciably harming the healthy parts and even without being necessarily introduced *viâ* the wound.[1]

In presence of the body fluids it should act as well as, if not better than, in water. It is not impossible to fulfil these demands; certain colouring matters, *e.g.*, Malachite-Green and Trypaflavine, and Vuzin (octylhydrocupreine), are steps in the right direction.

All the same, bacteriological methods (vaccines, serums) will doubtless provide truly specific antiseptics before chemistry does. And, to sum up, one should not ask more of antiseptics than they can give; they cannot act beyond a certain distance from the wound, and when the infection is deep they are, so to speak, outflanked.

Phenol and its Derivatives.

When Lister introduced antiseptic technique into surgery phenol (carbolic acid) forthwith had a great vogue. For a long time afterwards Lister's disciples employed it almost exclusively; Lucas Championnière, in particular, owed to carbolic acid much of his success at a time when there was no general belief in antiseptic methods. Its employment in this way has now been almost entirely abandoned, probably more because of the great progress in aseptic treatment than because other antiseptics have been introduced. It is still widely used for special purposes, such as sterilising instruments, preserving bandages and general disinfection.

For uses of this kind, however, phenol is being replaced by the next

[1] It is not paradoxical to say that the best external antiseptic will be that which reaches the outer surfaces from within.

higher homologues, the cresols. These are not so toxic as phenol. Of the three isomerides the *meta* is the most active. The cresols are sparingly soluble in water, so their use in the free state is limited, not only because one cannot obtain strong enough solutions, but also because of the difficulty of removing them without copiously washing or using a volatile solvent, once they have been sprayed on the ground or on clothing and household linen. In order to overcome these disadvantages, attempts have been made to render the cresols more soluble or, at any rate, easily emulsified.

Many years ago Lebœuf, a Bayonne pharmacist, succeeded in emulsifying tar, using soap-bark (*Quillaja*) as agent, and so produced an antiseptic which was very popular for a long time (*Coaltar saponiné Lebœuf*). For the cresols, caustic soda and various soaps are usually employed, for example, resin soap (*Creoline Pearson*), sodium cresotinate (*Solveol*), sodium cresoxyacetate (*Cresine*) and turkey-red oil (sulphonated castor oil).

Lysols contain phenols and certain aromatic hydrocarbons, of which the most important is methylnaphthalene. Lysol is closely related to *Coaltar saponiné*, but the emulsifying agent used here is a linseed oil soap.

Another way of arriving at a soluble product is to introduce an acid radical into the phenol and prepare the sodium or other salt of the derivative thus obtained. Thus the use of salicylic acid instead of phenol has been tried. Phenolsulphonic acid has been given the trade name of *Aseptol*. Various salts (zinc, mercury, sodium) of diiodo-*p*-phenolsulphonic acid will be described later (*Sozoiodols*).

Other phenolic compounds that have been used are the following: thymol, guaiacol, resorcinol, pyrogallol, eugenol, allyl phenol (*Chavasol*) and the naphthols. Of homologues of phenol the most active are the xylenols, particularly the *meta*, but they have not been utilised.

α-Naphthol is the more toxic of the two isomerides. The sodium salt of β-naphthol is *Microcidin*, recommended by Lucas Championnière. But naphthol is scarcely used as an antiseptic except for preserving dressings and in lotions or ointments for the scalp. On the other hand, some of its derivatives have been utilised in various ways. *Epicarin* is the sodium salt of β-hydroxynaphthyl-*o*-hydroxy-*m*-toluic acid:

$$HO{\cdot}C_{10}H_6{\cdot}CH_2{\cdot}C_6H_2(OH){\cdot}CO_2H$$

This is a powerful antiseptic largely used in veterinary work, for treating herpes and mange in dogs.

When the naphthols are suitably reduced, tetrahydronaphthols are formed. The β-derivative is a much more active antiseptic than the parent naphthol. Its trade name is *Tetralol*.

Substitution Products of Phenol.—The most interesting of the substituted phenols are the halogen derivatives, particularly those of the

naphthols. An important investigation on the antiseptic power of these compounds was carried out by Ehrlich and Bechhold. It was found that the numerical value of the property increased with the number of halogen atoms in the molecule and that the halogenated cresols are stronger antiseptics than the corresponding phenols, while, apparently, the maximum activity is possessed by tribromonaphthol, which is actually, *in vitro*, one of the most powerful antiseptics known. Yet everything depends on the particular species of micro-organism subjected to treatment. Experiment shows that here is a quite noteworthy example of specificity, and the phenomenon has been termed by Bechhold demi-specificity. Thus, to give only one illustration, dibromonaphthol is more active than tribromonaphthol towards *B. Coli*, while the contrary is the case for *B. Staphylococcus*, the bacillus of diphtheria, etc. So, as a general rule, no compound of this series displays the same antiseptic power towards all kinds of micro-organisms.

For practical reasons, and particularly because of its feeble hæmolytic action—the mono- and dibromo-derivatives are strongly hæmolytic—tribromonaphthol was selected for manufacture and is known as *Providoform*.

With the exception of *Salol*, phenol esters (*Benzonaphthol*, *Betol*, etc.) are chiefly used as internal (intestinal) antiseptics. Guaiacol is the only phenol ether to be employed, and its preparation and utilisation have already been discussed.

No external antiseptic is known among the aminophenol series, but it might be mentioned by the way that certain complicated ureas of aminonaphthol and naphthylamine sulphonic acids (*Bayer 205*) have been put forward as specifics against trypanosomes (internal antiseptics). The exact composition of *Bayer 205* has not been disclosed.[1]

A special class of phenolic compounds are quinoline derivatives, the hydroxyquinolines. The most widely known substances of this group are *Chinosol* and *Oxychinaseptol*. *Chinosol* is prepared by heating together *o*-aminophenol, sodium sulphate, glycerine, nitrophenol and sulphuric acid. 8-Hydroxyquinoline is thus produced and its (neutral) sulphate forms the drug. It is a mild antiseptic, and when used in quite small amounts prevents the development of moulds, and so is used for preserving sera (Nicolle). *Oxychinaseptol* is a compound of hydroxyquinoline with phenol sulphonic acid.

Before passing on to a discussion of Formalin a few words may be said on the subject of the aromatic acids. Benzoic acid is a weak antiseptic principally used as a preservative in jams, etc. Its homologue, phenylacetic acid, is notably more active; activity is, in fact, the more marked the weightier the side chain, until phenyl-butyric acid is reached at any rate, but only benzoic acid itself has been

[1] See note at end of chapter.

actually utilised. Benzyl benzoate is also in use under the name *Peruscabin* and advantageously replaces balsam of Peru, being in fact quite as active, colourless, and almost odourless. It is on the market as a solution in castor oil called *Peruol*.

As salicylic acid has already been the subject of an earlier chapter, it needs no discussion here.

Formalin.—*Formalin* (formaldehyde) is chiefly employed for disinfecting dwellings and the like, and for this purpose use is made of a variety of contrivances, some of which spray the 40% solution of formaldehyde mechanically, while others simply boil it. More convenient arrangements are those in which trioxymethylene (a polymeric form of formaldehyde) is volatilised.

An easy way of disinfecting a room without using expensive apparatus is to treat a mixture of trioxymethylene and barium peroxide (*Autan*) with water. Instead of barium peroxide, bleaching-powder (*Aldogen*) or calcium permanganate may be used.

For internal use the irritant properties of formaldehyde make it necessary to prepare condensation products with, *e.g.*, starch (*Amyloform*), dextrine (*Dextroform*) or albumin, etc., but none of these products has been very successful. Of greater interest is *Formicine*, the product of interaction with acetamide :

$$CH_3{\cdot}CO{\cdot}NH{\cdot}CH_2{\cdot}OH.$$

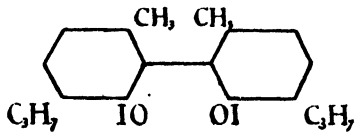
This is a liquid and has been recommended mainly for sterilising surgical instruments.

An internal antiseptic in most extensive use is hexamethylenetetramine (*Urotropin*, *Urometin*). This is a urinary antiseptic. It will not be discussed here.

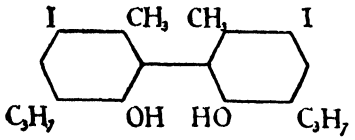
Halogen Compounds.—Iodoform and the hypochlorites are examples of this group of antiseptics. Iodoform has been employed for many years as a dry dressing for wounds and, indeed, has never been entirely superseded. But it has its disadvantages : it is costly and has an objectionable smell. Moreover, when used in quantity, it often has an intoxicating effect. For these reasons, chemical ingenuity has been applied more intensively here than in many other fields. At first efforts were made to mask the odour of iodoform, but the compounds produced were either too stable and so comparatively useless, or not stable enough, being easily decomposed by water, that is to say, they might be odourless to begin with, but the characteristic stench of iodoform soon arose.

As examples of the second class, the combinations of iodoform with hexamethylenetetramine and with tannin (*Iodoformogen*) may be noted. This line of investigation was soon abandoned and attention directed to the introduction of iodine into other compounds to form products which should be odourless and yet easily liberate the iodine again. *Aristol* is the best known substance of this kind. This is

dithymol-diiodide, to which its discoverers, Messinger and Wortman, attribute the formula :



It is not likely to have this constitution, and, according to Bougault, should be formulated as a quinone, but Moles y Marquina states that it contains two phenolic hydroxy groups and is therefore actually diiodo-dithymol :



What is almost certain, in every case, is that the iodine is in the nucleus. But anyhow, it is not absolutely necessary that the halogen be liberated, and actually, Aristol is one of the most valuable successors to Iodoform, being equally useful both internally and externally.[1]

In the same class are *Europhen*, isobutyl-*o*-cresoliodide :

$$CH_3{\cdot}C_6H_2(C_4H_9)OI{\cdot}C_6H_3O(CH_3){\cdot}C_4H_9\,;$$

Losophan, triiodocresol ; *Nosophen*, tetraiodophenolphthalein, obtained by treating the sodium salt of the phthalein with iodine chloride ; and *Iodol*, tetraiodopyrrole (Ciamician and Silber).

Isoform is *p*-iodoxyanisole, $CH_3O{\cdot}C_6H_4{\cdot}IO_2$, produced by oxidising *p*-iodoanisole with chlorine or potassium permanganate. Iodoxyanisole is explosive, so it is handled as a mixture with its own weight of calcium phosphate.

The *Sozoiodols* have already been mentioned, one of them is the potassium salt of diiodophenolsulphonic acid. Similarly constituted compounds are *Vioform*, iodochlorohydroxyquinoline ; *Iodofan*, iodo-dihydroxybenzene ; and *Chryseine* (Mouneyrat), which is an iodine derivative of *Urotropine*. . . . .

Chlorine.—For large scale and extensive disinfection chlorine is well adapted, both by reason of its cheapness and its outstanding bactericidal power. It is principally used in the form of sodium hypochlorite (*eau de Javel, liqueur de Labarraque*) or as calcium hypochlorite (bleaching-powder). Concentrated solutions of sodium hypochlorite are fairly stable, but much too poisonous, while dilute solutions are unstable. The stabilisation of hypochlorite solution has been investigated by Dakin,[2] who discovered the best conditions for

[1] The preparation is described by Moles y Marquina in *Anales de la Sociedad española de Fisica y Quimica*, 1919.

[2] See *Applied Chemistry Reports*, 1917, II, 475.

preserving and applying it. Dakin's solution contains 0·45% to 0·50%, of sodium hypochlorite neutralised with boric acid. It was used in large quantities during the war, particularly by the British and German armies, the technique of its preparation having been much improved by the famous surgeon, Carrel.

Even when boric acid is present hypochlorite solutions do not keep well for long periods. Dakin, therefore, sought for compounds which would serve as chlorine containers or carriers and would easily liberate it in contact with a wound or sore. The result of his inquiries was the preparation of *Chloramine T*, a water-soluble compound, and *Dichloramine T*, insoluble in water. *Chloramine T* is sodio-*p*-toluenesulphonchloroamide, $CH_3 \cdot C_6H_4 \cdot SO_2Na : NCl$. It is prepared by gently warming together *p*-toluenesulphonamide (1 mol.) and a 5% alkaline solution of sodium hypochlorite (1½ mol.), a saturated solution of sodium chloride (½ mol.) being then added. The product forms a crystalline precipitate containing 3 mols. water of crystallisation and is readily soluble in water. *Dichloramine T* is prepared by passing chlorine into a suspension of *p*-toluenesulphonamide (500 gm.), sodium acetate (1,000 gm.), chloroform (1,000 gm.) and water (5 l.), until no more is absorbed. The dichloramine is taken up by the chloroform, from which it is separated again by distillation. Both the isolated substance and its solutions should be kept in the dark. Dichloramine T is insoluble in water and is applied as a 5% to 10% solution in eucalyptolised oil or in chlorinated paraffin wax (*Chlorcosan*).

No other chlorine derivatives are worthy of special notice. Among bromine derivatives there are only tribromonaphthol which, as already mentioned, has been given the name *Providoform*, and was extensively used in Germany for dressing war wounds, and a compound of tribromophenol with bismuth, also used to a considerable extent, called *Xeroform*.

Sulphur Compounds.—Many sulphur compounds have been suggested as useful antiseptics, but few have kept their place in therapeutics, the chief exception being Ichthyol. Nevertheless, certain sulphur-containing essential oils, such as mustard oil, have potent antiseptic properties. Mustard oil, in fact, is a constituent of a well-known speciality, *Aniodol*. Moreover, the antiseptic qualities of oil of garlic have been known for many years : it was at one time recommended in cases of tuberculosis (Sejournet) and cholera, and quite recently has been in cases of influenza. Again, the essential oils of various crucifers (horse-radish, cress, scurvy-grass) are present in a number of antiseptic mouth-washes (dentifrices). *Thiosinamine* is a well-known derivative of mustard oil, being, in fact, allylthiourea. Di-*o*-aminophenyldisulphide has been recommended by MacDonagh as a substitute for salvarsan and has received the trade name *Intramine*.[1]

[1] *Applied Chemistry Reports*, 1916, I, 285.

However, the most widely used sulphur compound is *Ichthyol*, Certain naturally occurring bitumens of organic origin (fish remains). which are particularly plentiful in the Tyrol, contain much combined sulphur, and when distilled yield an oil. When this oil is treated with sulphuric acid a sulphonic acid is obtained, the ammonium salt of which is Ichthyol.

The essential oils of certain crucifers having been mentioned above, it may be recalled to mind in passing that other essential oils, not containing sulphur, possess marked bactericidal power and are still supplied in all kinds of ways for the household medicine store. Such are cajeput oil (Niaouli, *Gomenol*), eucalyptus oil, peppermint, cinnamon, and oil of cloves. A derivative, namely, the ethoxyacetic ester, of the principal constituent of the essential oil of mint, menthol, is used in place of menthol itself in either pastilles or ointments under the trade name *Coryfin*.

Bismuth Compounds.—Salts of bismuth are chiefly applied in internal treatment, yet certain insoluble compounds are in extensive use as antiseptic powders, and, together with Aristol, have almost entirely ousted iodoform. *Dermatol*, bismuth sub-gallate, is the best known example of this class and is an excellent preparation. *Bismal* is the trade name of a compound of dermatol and formaldehyde, a bismuth methylene-digallate. *Airol* is an iodo-dermatol, produced by bringing together bismuth oxyiodide and gallic acid, or better by treating a solution of gallic acid and potassium iodide with one of bismuth hydroxide in sodium acetate; the mixture is to be gently heated until the precipitate develops a greenish colour.

Other compounds which may be noted are *Eudoxine*, a bismuth tetraiodophenolphthalein, and *Xeroform*, already mentioned above.

Silver Compounds.—Besides colloidal silver prepared electrolytically—*Electrargol*—which does not come within our purview here, silver compounds actually used as antiseptics, either internally or externally, are nearly all colloidal preparations with albumoses or peptones as base. The best known is *Collargol*, which contains silver up to 90% by weight. It is prepared by treating an albumin with caustic soda and silver oxide alternately, continuing the process unti the silver-content is high enough. Imitations of genuine Collargol rarely contain more than 70% of silver.

The preparation of *Protargol*—silver proteinate—is based on a different principle. Here silver casein is dissolved in water with the aid of a peptone or an albumose, themselves prepared by warming an albumin with oxalic or sulphuric acid. The organic part of Protargol is thus much less completely hydrolysed than that which serves as the basis of Collargol. Protargol is not, properly speaking, colloidal silver, as it contains only 7% to 8% of silver. It has a buff-yellow colour, is taken up but slowly by water, and from the solution it is partly precipitated again on boiling.

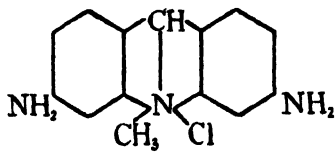
Dyestuffs.—Many dyestuffs are taken up selectively by certain tissues, and, as is well known, they are used in staining micro-organisms for microscopic examination. It was natural to suppose that the fixation of the colouring matter on the micro-organisms meant that the latter were being attacked and that this could not take place without injuring them. All the same, because micro-organisms are easily stained on the microscope slide it does not follow that they are similarly affected inside the living body. In fact, in the living organism most dyestuffs are reduced to their colourless leuco derivatives; staining results only in exceptional cases, as in that of methylene blue, which colours selectively the ends of certain nerves. And although methylene blue itself will stain malarial parasites intensely *in vitro*, it does not reach nor colour them *in vivo*; but, on the other hand, even *in vivo* a certain oxazine dyestuff can stain the centrosomes or blepharoblasts of trypanosomes.

Malachite Green is a dyestuff of the triphenylmethane group, being, in fact, the chloride of tetramethyldiaminotriphenyl carbinol. To prepare it benzaldehyde and two molecular proportions of dimethylaniline are condensed together in presence of about $1\frac{1}{3}$ equivalents of hydrochloric acid. The leuco base so obtained is oxidised with lead peroxide and the dyestuff isolated as the zinc double salt.

Malachite Green was used during the war, either alone or combined with mercuric chloride, mainly by the British (Fildes). The mercurial compound is prepared by mixing alcoholic solutions of the dyestuff and mercuric chloride. The solution obtained is employed as such and is sprayed over superficial wounds, or for treating osteomyelitis.

Brilliant Green is the tetraethyl derivative corresponding to Malachite Green. It is a powerful bactericide and has been used and recommended principally by Browning.

Trypaflavine (*Acriflavine*) is, however, the most extensively employed dyestuff, and this compound seems destined to retain its position in therapeutics. It is a potent antiseptic and possesses the valuable property of being more active in organic media (sera) than in water. *Trypaflavine* is 3 : 6-diamino-N-methylacridinium chloride.



It is prepared by heating trimethylene aniline, $(C_6H_5{\cdot}NCH_2)_3$, produced by condensing together formaldehyde and aniline, with aniline hydrochloride, which acts as a catalyst, 4 : 4′-diamino-diphenylmethane being formed. This is then nitrated with "mixed acid," and the 2 : 2′-dinitro derivative obtained, which yields the tetra-amino

compound on reduction. By heating this with a molecular proportion of hydrochloric acid to 135°–140° diaminoacridine is produced.[1] Diaminoacridine (*Proflavine*) itself possesses well marked antiseptic properties (Dakin), although it is not so active as Trypaflavine. To obtain the latter Proflavine, after protecting the amino groups, is methylated by treatment with methyl chloride. Trypaflavine is used as a 1 : 1,000 solution in normal saline. To give a rough idea of its strength, it may be stated that a *staphylococcus* culture in blood serum containing 600,000 germs per drop is sterilised completely in eight hours by an amount of trypaflavine corresponding to 0·03%, and stays sterile indefinitely. Brilliant Green does not effect complete sterilisation, and although the number of cocci is at first considerably diminished, it rises again to the original value in twenty-four hours.[2]

Besides their use in treating wounds antiseptics are also employed for disinfecting germ-carriers. During the war disinfection of this kind was carried out on a large scale; men on active service were either subjected for 10–15 minutes to an antiseptic atmosphere produced by means of a spraying device, or their nasal passages were treated independently with an antiseptic solution. This disinfection gave excellent results with *meningococcus*, but the results were less satisfactory with the diphtheria bacillus, and negative in the case of *pneumococcus*. The antiseptics used were Chloramine T, iodine, guaiacol, Argyrol, etc. Some observations showed that Eucupine had a wholesome effect on pneumococcus.

During the war it was also necessary to disinfect dwellings, etc., and to sterilise water supplies. For sterilising water the substances chiefly used in France were permanganate (*Poudre Lambert*), potassium iodate (*Poudre Vaillard*), and, last but not least, sodium hypochlorite (*Eau de Javel*). A most interesting product used for sterilising water is Dakin's *Halazone*. This is the dichloramine corresponding with sulphobenzoic acid, of the formulæ:

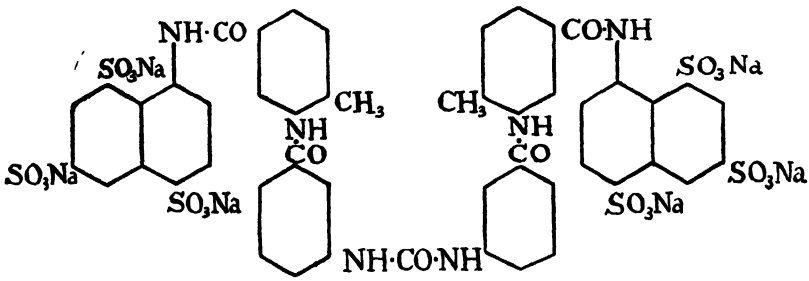
$$C_6H_4\begin{cases}CO_2H & (1)\\ SO_2NCl_2. & (4)\end{cases}$$

It is supplied for use mixed with borax in tablet form. For a litre of water 4 mgm. of the active compound suffice.

[1] Benda, *Ber.*, 1912, *45*, 1787; *Applied Chemistry Reports*, 1917, II, 477.

[2] *Rivanol*, 2-ethoxy-6: 9-diaminoacridine, is a new very potent antiseptic in this series.

Note on "*Bayer 205*": MM. Fourneau, Tréfouël, Mme. Tréfouël, and M. Vallée have recently (*Compt. rend.*, 1924, *178*, 675; *Chemical Abstracts*, 1924A, i., 382), prepared the complex carbamide:



and shown that it has the trypanocidal and other properties attributed to "*Bayer 205*." (*Applied Chemistry Reports*, 1923, VIII., 537; *cf.* also 1921, VI., 537.)

CHAPTER VIII

ORGANIC COMPOUNDS OF ARSENIC

ALIPHATIC SERIES

In spite of their great interest from the standpoint of pure chemistry, the aliphatic arsenicals will not be discussed at great length, because they have hitherto not played a great part in the therapeutics of infectious diseases; from those of the aromatic series, on the other hand, one of the most important branches of chemotherapeutics has sprung, the development of which has been brilliantly successful.

The most important arsenical medicaments of the aliphatic series are the **cacodylates** and the **methylarsinates.**

Cadet's fuming arsenical liquid seems to be the first organic arsenic compound described. The army pharmacist, Louis Cadet, discovered this substance in 1760, having obtained it by heating together arsenious acid and potassium acetate. It had a repulsive odour and took fire in contact with air. Further investigation was carried out by Thénard and notably by Bunsen, who showed that Cadet's liquid was composed of carbon, hydrogen, arsenic and oxygen, and that the oxygen was replaceable by other non-metallic elements, sulphur, iodine, chlorine, etc., just as it is in potassium oxide. The arsenic-hydrocarbon group remained intact through a series of the most diverse reactions, and so Berzelius gave to this radical, $(CH_3)_2As$, the name **Cacodyl.**

The chief constituent of Cadet's fuming liquid is cacodyl oxide, $(CH_3)_2As{\cdot}O{\cdot}As(CH_3)_2$. This substance does not fume nor is it inflammable in air, but the crude product contains also some free cacodyl, $(CH_3)_2As{\cdot}As(CH_3)_2$, which is easily oxidised and to which the inflammability is due.[1]

There are two series of cacodyl derivatives, viz., those in which the radical behaves as if it were monovalent, and those in which it is trivalent. The following are the chief members of the first series:

Cacodyl (*Dicacodyl*)	$(CH_3)_2As{\cdot}As(CH_3)_2$
Cacodyl hydride (*Dimethylarsine*)[2] . .	$(CH_3)_2AsH$
Cacodyl chloride	$(CH_3)_2As{\cdot}Cl$
Cacodyl cyanide	$(CH_3)_2As{\cdot}CN$

[1] According to a recent patent (Merck), the addition of a small amount of sulphur to Cadet's liquid makes it non-inflammable.

[2] Cacodyl hydroxide $(CH_3)_2As{\cdot}OH$, corresponding to potassium hydroxide, is not known.

And of the second series :

Cacodylic acid . . . . . $(CH_3)_2AsO{\cdot}OH$
Tetraalkylarsonium compounds, *e.g.* . $(CH_3)_4AsI$

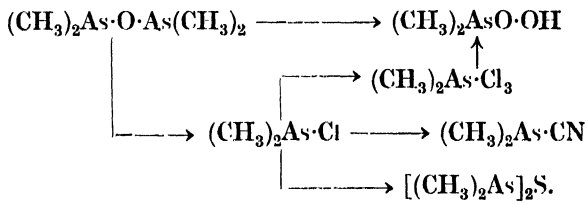
Cacodylic Acid.—Cacodylic acid, $(CH_3)_2AsO{\cdot}OH$, may be regarded as arsenic acid with two hydroxyl groups replaced by two methyl radicals. Cacodylic acid is obtained industrially solely from Cadet's fuming liquid, produced, as has been seen, when a mixture of potassium acetate [1] and arsenious acid is distilled :

$$4CH_3CO_2K + As_2O_3 = (CH_3)_2As{\cdot}O{\cdot}As(CH_3)_2 + 2K_2CO_3 + 2CO_2.$$

The product is first rectified in a current of carbon dioxide and then oxidised with mercuric oxide in aqueous suspension.

Cacodylic acid is a crystalline compound readily soluble in water and alcohol. Its great stability towards oxidising agents is its most remarkable characteristic. It is neutral to methyl orange, acid to phenol-phthalein, and is amphoteric in character, forming unstable compounds with strong acids.

Cacodyl oxide is the starting point from which not only cacodylic acid is prepared, but also all the other cacodyl derivatives. These are obtained by a series of reactions of which some are indicated in the following scheme :



Methylarsinic Acid.—Cacodylic acid is the only derivative of dimethylarsine to be utilised, but of the monomethylarsine series, in which only one methyl group is attached to the arsenic atom, a representative, namely, methylarsinic acid, is employed. (The sodium salt is *Arrhenal.*) This acid can be regarded as cacodylic acid with one CH_3 replaced by OH. In fact, as will be shown later, methylarsinic acid may be prepared *viâ* cacodylic acid. Industrially it is obtained by methylating arsenious acid. The methylation may be carried out either with methyl iodide (Meyer) or methyl sulphate (Auger), thus :

$$2CH_3I + As_2O_3 + 6NaOH \longrightarrow 2CH_3AsO_3Na_2 + 2NaI + 3H_2O.$$

Now sodium arsenite may be formulated either as

$$Na{\cdot}AsO : (ONa)_2,$$

or as

$$As(ONa)_3 ;$$

the first of these formulæ affords a better explanation of the above

[1] Sodium acetate gives poor yields.

reaction. If the second be adopted one must suppose either that the first stage is a simple addition forming the compound

or that an ether of the formula

$$As(ONa)_2OCH_3,$$

undergoes intramolecular rearrangement.

Homologues of methylarsinic acid cannot be made by the above method. For their preparation potassium arsenite must be used instead of sodium arsenite, and even then the yields are meagre (Dehn).

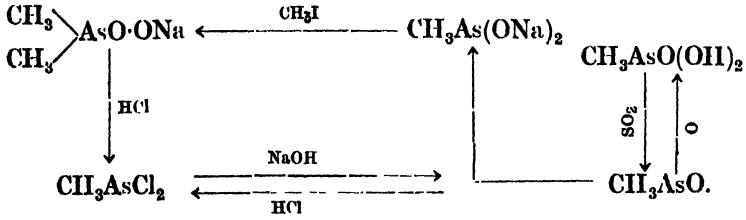
We must now turn to the various reactions by which members of the methylarsinic group may be obtained from those of the cacodyl group (or the latter from the former); and, also, direct our attention to the general reactions which, besides those applied industrially, give rise to aliphatic arsenic compounds, because many of these apply as well to the aromatic as to the aliphatic series.

(i) When sodium methylarsinate is treated in hydrochloric acid solution with sulphur dioxide in presence of a trace of potassium iodide—which acts catalytically– it yields methylarsenious chloride, CH_3AsCl_2. Methylarsenious chloride is decomposed by caustic soda, forming sodium methylarsinite, $CH_3As(ONa)_2$, which may be oxidised back again to sodium methylarsinate. *If sodium methylarsinite be treated with methyl iodide, sodium cacodylate is formed* by a reaction exactly analogous to that which results in the production of the methylarsenate from sodium arsenite (Auger):

(ii) When cacodylic acid is heated in a current of hydrogen chloride, preferably in presence of mercuric chloride, it loses a methyl group as methyl chloride, and yields methylarsenious chloride, thus:

$$(CH_3)_2AsO{\cdot}OH + 3HCl \longrightarrow CH_3AsCl_2 + CH_3Cl + 2H_2O.$$

These are the most important reactions of the kind. They are set out concisely below.



Other preparative methods are as follows:

(iii) By the action of alkyl magnesium compounds on arsenic trichloride, trialkylarsines are formed:

$$3R{\cdot}MgBr + AsCl_3 \longrightarrow AsR_3 + 3MgBrCl.$$

From the trialkyl compounds, AsR_3, first $\begin{matrix}R\\R\end{matrix}\!\!>\!AsCl$, then $\begin{matrix}R\\R\end{matrix}\!\!>\!AsCl_3$, and eventually $R{\cdot}AsCl_2$ may be obtained.

(iv) Alkyl iodides interact with potassium arsenite (Dehn) to produce homologues of Arrhenal, although, as has already been noted, sodium arsenite cannot be used.

(v) Arsenic trichloride and, *e.g.*, mercury diethyl, will react thus:

$$AsCl_3 + HgR_2 \longrightarrow R{\cdot}AsCl_2 + RHgCl.$$

(vi) Lastly, there is the Cahours-Auger method, which is based on the action of alkyl iodides on pulverised arsenic at 160° to 200° (Cahours), or on amorphous arsenic, obtained by reducing arsenic acid with hypophosphorous acid (Auger). In the latter case the reaction takes place in the cold. The products obtained are either tetraalkylarsonium iodides, or the hydriodides of methyl- and dimethyl-arsine. When Auger brought amorphous arsenic and, for example, iodoform together, he obtained a mixture of di-iodomethylarsinic and tetraiodocacodyl compounds:

$$3CHI_3 + 2As \longrightarrow CHI_2{\cdot}AsI_2 + (CHI_2)_2AsI,$$

which were then oxidised by nitric acid thus:

$$CHI_2{\cdot}AsI_2 + 4HNO_3 = CHI_2AsO{\cdot}(OH)_2 + 4NO_2 + H_2O + I_2.$$

As regards the therapeutical employment of the aliphatic arsenic compounds, it has already been said that until quite recently only the cacodylates and the methylarsinates were utilised. These, however, are used to a considerable extent for treating tuberculosis in its early stages and as general tonics. Their action here is very noteworthy (*Arrhenal, Histogenol*, cacodylate and guaiacol, etc.).[1] Bunsen, and later Kirchner, drew attention to the fact that cacodylic acid was almost harmless, and several attempts were made to use it therapeutically, but the honour of introducing organic arsenic compounds into medicine truly belongs to Armand Gautier.

AROMATIC SERIES

Methylarsinates have been recommended for treating malaria, but it is doubtful if they are effective, and they certainly have no action on spirilla and trypanosomes. The aromatic arsenic compounds, on the other hand, have all, or nearly all, a pronounced action on these agents of disease, and to them our attention will now be directed. Some of these substances have acquired extraordinary importance, and, indeed, since Ehrlich began his researches, the whole field of the arsenic derivatives has become the most interesting one in thera-

[1] There are in use, however, certain arsenic compounds of acids of the acetylene series (Solarson); for example, heptinecarboxylic acid + $AsCl_3$, and so forth, but their value as medicaments is still uncertain.

peutical chemistry. For this reason our studies will be as full as possible, and the subject will be divided up as follows :

Methods of preparation.

Methods for transforming derivatives of pentavalent arsenic (aryl-arsinic acids) *into compounds of trivalent arsenic* (arsines, arsine oxides, arseno compounds, etc.).

Methods of introducing new substituent groups into the aromatic nuclei.

Physical and chemical properties of the products.

Chemotherapy of organic arsenic compounds.

Methods of Preparation.—These may be divided into two main groups : (1) Those, three in number, for obtaining the arylarsinic acids which serve as starting materials for preparing all other compounds ; these are based on the following reactions, viz. :

I. That between arsenic acid and aromatic amines, *e.g.*, aniline, toluidine, nitraniline.

II. That between arsenic acid and phenols.

III. That between arsenic acid and diazonium compounds.

(2) Those by which derivatives of trivalent arsenic may be directly prepared. Compounds containing pentavalent arsenic may then be more or less readily produced. The methods here are based on the following reactions :

IV. That between mercury-diaryls and arsenious chloride.

V. That between arsenious chloride and salts of mono-aryl-mercuri-derivatives.

VI. That of mixtures of halogen-substituted benzene derivatives and arsenious chloride with metallic sodium. The last three reactions on our list yield arylarsenious chlorides ; the product in this case (VI.) is a mixture of arylarsenious dichloride and diarylarsenious chloride.

VII. That between aryl magnesium halides and arsenious chloride. This method furnishes exclusively triarylarsines, from which, by heating with arsenious chloride, the chlorides of the mono- and diaryl derivatives may be obtained.

VIII. That between arsenious chloride and tertiary aromatic amines, *e.g.*, dimethylaniline.

I. *Fusion of the Arsenates of Aromatic Amines.*

$$C_6H_5{\cdot}NH_2{\cdot}H_3AsO_4 \longrightarrow C_6H_4\begin{cases}NH_2\\AsO(OH)_2.\end{cases}$$

This reaction is analogous to that by which sulphanilic acid is formed, viz. :

$$C_6H_5{\cdot}NH_2{\cdot}H_2SO_4 \longrightarrow C_6H_4\begin{cases}NH_2\\SO_3H.\end{cases}$$

In this way Béchamp prepared the first aromatic arsenic derivative, arsanilic acid or *Atoxyl*. Essentially the method is as follows : basic aniline arsenate, which can be readily obtained in a crystalline form,

is heated in a flask until aniline begins to distil off (one molecular proportion will separate). The flask is then evacuated to expedite the distillation and the temperature is gradually taken up to 190°–195°. Water and a little aniline pass over and the mass becomes viscous and violet in colour. The temperature is kept at 195°–210° for two hours. The product is dissolved in dilute sodium carbonate solution, the solution filtered and the atoxyl precipitated by adding nitric acid.

This method is not generally applicable to amines containing nitro or carboxyl groups, although *p*-nitraniline yields the corresponding arsinic acid with great readiness. It should be added that the product above contains, besides arsanilic acid, a certain proportion of diaminodiphenylarsinic acid :

$$NH_2 \cdot C_6H_4 \cdot AsO(OH) \cdot C_6H_4 \cdot NH_2.$$

II. *Action of Arsenic Acid on Phenols.*—Arsenic acid and phenol, for example, react to form *p*-hydroxyphenylarsinic acid, identical with that obtained from atoxyl, together with a little of the *ortho* isomeride. When the *p*-hydroxy acid is nitrated the main product is the 3-nitro-4-hydroxyphenylarsinic acid used for making "*606*."

Arsenic acid interacts in the same way with dihydric phenols, with resorcinol in particular, yielding the corresponding acids with the greatest ease.

III. *Bart's Method.*[1] This is the application of the Sandmeyer reaction, and like all methods in which diazonium compounds are used, it is capable of wide variation. Many aromatic arsenic compounds may be made in this way. The method is simple and involves merely bringing together the appropriate diazonium compound and arsenious acid, either in an acid, or, better, in an alkaline medium. The reaction is facilitated by the presence of such catalysts as copper or copper salts.

Bart prepared, among others, the following derivatives : *p*-bromophenylarsinic acid from *p*-bromoaniline, acetylaminophenylarsinic acid from *p*-aminoacetanilide, benzarsinic acid and nitro-hydroxyphenylarsinic acid.

Arylarsinic acids, for example, *p*-nitrophenylarsinic acid, react again equally well.

The following equations illustrate the reactions involved :

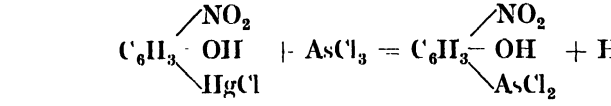
$$R \cdot N : N \cdot X + As(OK)_3 = R \cdot N : N \cdot OAs(OK)_2 + KX$$
$$R \cdot N : N \cdot OAs(OK)_2 = R \cdot AsO(OK)_2 + N_2.$$

IV. *Michaelis's Method.*—This also is available as a general method, but it has been used only to a limited extent. Mercury diphenyl compounds are the starting materials. When these substances are treated with arsenious chloride they lose their mercury either as mercuric chloride or as monoarylmercurichloride, thus :

[1] The name is spelt either Barth or Bart.

$$C_6H_5\cdot Hg\cdot C_6H_5 + AsCl_3 = C_6H_5\cdot AsCl_2 + C_6H_5HgCl$$
$$C_6H_5\cdot Hg\cdot C_6H_5 + AsCl_3 = (C_6H_5)_2As\cdot Cl + HgCl_2$$
$$C_6H_5\cdot Hg\cdot C_6H_5 + 2AsCl_3 = 2C_6H_5AsCl_2 + HgCl_2.$$[1]

V. *Roeder's Method.*—This is a development of that of Michaelis; the variation is very interesting because it involves using monoarylmercuri-salts, *e.g.*, $C_6H_5\cdot Hg\cdot Cl$. Unfortunately, in a number of cases, particularly those of the nitrophenolic derivatives, it is only applicable to compounds in which the mercury atom occupies the *para* position.

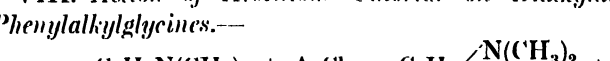


VI. A method giving excellent results in simple cases, that is, when there are no nuclear nitro or hydroxy groups, consists in treating a mixture of arsenious chloride and halogen derivatives of benzene or its homologues, or of such compounds as the phenol ethers (anisole), with *metallic sodium.*

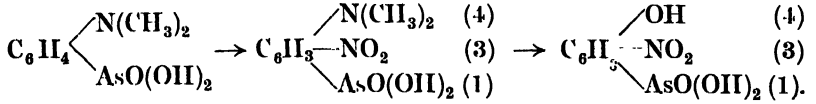
$$C_6H_5Br + AsCl_3 + 2Na = C_6H_5AsCl_2 + NaBr + NaCl.$$

VII. By applying *Grignard's reaction* only triarylarsine compounds can be obtained. The method can only be used with such halogen derivatives as will give arylmagnesium halides, and so is not of great utility.

VIII. *Action of Arsenious Chloride on Dialkylanilines and on Phenylalkylglycines.*—

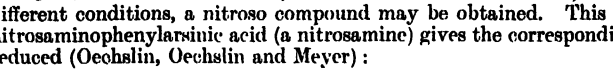


An almost quantitative yield is given by this method. It may be used to obtain, for example, *p*-dimethylaminophenylarsinic acid. This substance is readily produced and is a valuable intermediate for preparing other arsenic compounds. Thus, when it is nitrated under certain conditions, it yields 4-dimethylamino-3-nitrophenylarsinic acid, which is converted by heating with caustic potash into the corresponding *p*-hydroxy derivative:



From the latter, "*606*" can be made (Oechslin).[2]

[1] Mercury diaryls are easily obtained *via* the monoarylmercuric salts, which are formed by interaction of benzene itself or many of its derivatives, with mercuric acetate:



[2] Under different conditions, a dinitroderivative is formed, and again, under still different conditions, a nitroso compound may be obtained. This substance, methylnitrosaminophenylarsinic acid (a nitrosamine) gives the corresponding hydrazine when reduced (Oechslin, Oechslin and Meyer):

In short, therefore, methods are known for obtaining directly either arsinic acids, aryl- or diaryl-arsenious chlorides, or triarylarsines ; but the real primary materials for preparing all other arsenicals are the arsinic acids, and so the question of converting the arsines into these acids arises. If one has to deal with the monoarylarsenious chlorides (halides), simple treatment with alkaline hydrogen peroxide will effect this conversion. If the other derivatives are in question they must be first heated with arsenious chloride to convert them into monoarylarsenious chlorides, in the manner already described, a process which may or may not be readily carried out, or they may be treated with chlorine and the product, *e.g.*, triphenylarsine dichloride, heated under diminished pressure :

$$(C_6H_5)_3AsCl_2 \longrightarrow C_6H_5Cl + (C_2H_5)_2AsCl.$$

Reduction of the Arsinic Acid Group.—Arylarsinic acids may be reduced to give successively :

A. The corresponding *arsenious oxide* :
B. The corresponding *arseno-* compound ;
C. The *arsine*, $R{\cdot}AsH_2$.

A. *Conversion of Arsinic Acids into Arsenious Oxides.*

This transformation may be effected either directly or through the intermediate stage of the arylarsenious chloride, as the latter furnishes the oxide by simple treatment with dilute caustic soda or even with sodium carbonate. Some of the methods are the same as those applied in the aliphatic series.

(i) Action of sulphurous acid in presence of a little hydriodic acid. If the operation be conducted in strong hydrochloric acid solution the product is the arsenious chloride :

$$C_6H_5{\cdot}AsO_3H_2 + SO_2 + 2HCl = C_6H_5AsCl_2 + H_2SO_4 + H_2O,$$

but if, on the other hand, the solution be neutral, the arsenious oxide itself is obtained. The conversion of phenylacetamidophenylarsinic acid will serve as an example :

$$C_6H_4\begin{cases}NH{\cdot}CO{\cdot}CH_2{\cdot}C_6H_5\\ AsO_3H_2.\end{cases}$$

Phenylacetamidophenylarsinic acid (79 gm.), hydrochloric acid, sp. gr. 1·12 (80 gm.) and hydriodic acid, 48% (0·5 c.c.) are mixed together and sulphur dioxide passed into the mixture. When the reaction is complete the precipitate is filtered off, washed with hydrochloric acid and treated with ammonia to liberate the free arsenious oxide.

By this method the arsinic group may be reduced without affecting any nitro groups present, thus *p*-nitrophenylarsinic acid yields *p*-nitrophenylarsenious oxide.

(ii) The reducing agent may be phenylhydrazine :

$$C_6H_5{\cdot}AsO_3H_2 + C_6H_5{\cdot}NH{\cdot}NH_2 = C_6H_6 + N_2 + C_6H_5{\cdot}AsO + 2H_2O;$$

(iii) Hydriodic acid :

$$R{\cdot}AsO_3H_2 + 4HI = RAsI + 3H_2O + I_2;$$

(iv) Phosphorus trichloride in ethyl acetate solution :

$$C_6H_5{\cdot}AsO_3H_2 + PCl_3 = HPO_3 + C_6H_5AsCl_2 + HCl;$$

thus, dimethylaminophenylarsinic acid yields dimethylaminophenylarsenious chloride and benzarsinic acid benzarsenious chloride ;

(v) Phosphorous acid, for example, when nitrophenylarsinic acid is heated in a sealed tube at 115° with water and phosphorous acid (crystalline), the corresponding arsenious oxide separates in a crystalline form.

We have already seen that methods are available for introducing the arsenious chloride group directly into the ring. These may be briefly reviewed again here.

(i) Interaction of arsenious chloride and the mercury diaryls :

$$(C_6H_5)_2Hg + AsCl_3 = C_6H_5{\cdot}Hg{\cdot}Cl + C_6H_5AsCl_2.$$

(ii) Action of metallic sodium on a mixture of chlorobenzene and arsenious chloride. The product contains some diphenylarsenious chloride, depending on the conditions under which the reaction takes place ; this substance is converted into phenylarsenious chloride by heating it with more arsenious chloride.

(iii) Action of arsenious chloride on dialkylanilines and alkylphenylglycines (Oechslin, Michaelis).

To these should be added :

(iv) Arseno compounds (·As : As·)— among the reduction products of the arsinic acids these are the ones most easily obtained— may be converted into the corresponding arsenious chlorides, and so to the oxides, by treatment with chlorine. For example, arsenobenzene reacts with two molecular proportions of chlorine to produce phenylarsenious chloride.

B. *Conversion of Arsinic Acids and Arsenious Oxides into Arseno Compounds (— As : As—).*

Reduction of the Arsenious Oxide Group.—This reduction takes place with greater facility than that of the corresponding arsinic acid. The same reagents may generally be used and the temperature need not be elevated, that of the room will suffice. The reducing agent may be :

(i) Sodium hydrosulphite—the calculated amount. Reduction takes place in the cold or on gently heating ;

(ii) Stannous chloride and hydrochloric acid ;

(iii) Phosphorous acid (crystallised) in methyl alcohol solution ;

(iv) Sodium amalgam. For example, dimethylaminophenylarsenious oxide in alcoholic solution is treated with 3% sodium amalgam in excess at 40°–50°. The arseno compound separates out and after twelve hours may be filtered off.

Reduction of the Arsinic Acid Group.—The reagents used here are :

(i) Stannous chloride and hydrochloric acid—hot ;

(ii) Hydriodic acid with a catalyst ;

(iii) Crystallised phosphorous acid—in the warm ;

(iv) Zinc and sodium bisulphite ;

(v) Sodium hypophosphite and hydriodic acid ; thus, to prepare the compound,

$$(HO{\cdot}C_6H_4{\cdot}NH{\cdot}CH_2{\cdot}CO{\cdot}NH{\cdot}C_6H_4{\cdot}As =)_2$$

sodium hypophosphite (60 gm.), water (20 c.c.), hydrochloric acid (100 gm.), and methyl alcohol (400 gm.) are mixed together and to the filtered solution are added hydriodic acid (48% — 5 c.c.) and then *p*-hydroxyphenylaminoacetamidophenylarsinic acid (75 gm.), dissolved in a mixture of equal volumes of methyl alcohol (1,000 c.c.) and hydrochloric acid. The whole is heated to 35° and the hydrochloride of the arseno compound separates out. It is filtered off, washed with methyl alcohol, and decomposed by treatment with ammonia (Jacob, Heidelberger and others) ;

(vi) Sodium hydrosulphite ; this is the reducing agent most appropriate for the purpose and an example of its use will be given in detail later.

As with the arsenious oxides, when it is desired to reduce the arsinic acid group and leave nitro groups untouched, crystallised phosphorous acid dissolved in methyl or ethyl alcohol, or sodium amalgam, or sodium hydrosulphite in the exactly calculated amount, is used. If potassium iodide be added when the first reduction is complete, that of the nitro groups may likewise be carried out. Thus, when 4-hydroxy-3-nitrophenylarsinic acid is reduced by means of phosphorous acid, dihydroxydinitroarsenobenzene is obtained ; but if, when this stage of the reduction is finished, potassium iodide be added, the action recommences and the nitro groups are attacked, so that the final product is salvarsan.

By the above methods only symmetrical arseno compounds are produced. Unsymmetrical derivatives must be prepared by bringing together arsines and arsenious oxides, *e.g.*,

$$R{\cdot}AsO + H_2As{\cdot}R' = R{\cdot}As : As{\cdot}R' + H_2O.$$

C. *Conversion of Arsenious Oxides and Arsinic Acids into Arsines,* $—AsH_2$.

Arsines are almost exclusively obtained by reducing arsinic acids with amalgamated zinc and hydrochloric acid. Most of these substances are volatile in steam and soluble in ether (Kahn). The arsines are usually less poisonous than the corresponding arseno compounds, but are quite as active physiologically. They are oxidised on exposure to the air to arseno derivatives.

Modification of Existing or Introduction of New Substituent Groups, etc.—Turning now to the question of modifying existing or introducing new substituent groups in such a way as to leave the arsinic acid group unaffected, we find that most of the common methods used for aromatic compounds are applicable here also. Thus nitration, reduction of nitro groups, oxidation of various substituents, and so forth, may generally be carried out quite readily on arsinic acid derivatives.

I. The *reduction* of $\cdot NO_2$ to $\cdot NH_2$ is best done with ferrous sulphate and ammonia.

II. The replacement of the amino group, or the $\cdot N(CH_3)_2$ group, by hydroxyl in nitro derivatives is easily effected by treatment with sodium or potassium hydroxide when the nitro group is situated *ortho* or *para* to the amino group in question. Thus 3-nitro-4-dimethylaminophenylarsinic acid, treated with caustic potash, gives 3-nitro-4-hydroxyphenylarsinic acid, from which salvarsan may be prepared (Oechslin).

III. $\cdot OH$, $\cdot Cl$, $\cdot I$, $\cdot CN$ may be introduced in place of the amino group by *Sandmeyer's* method, *viâ* the diazonium compound.

IV. Side chains may be *oxidised*, thus, tolylarsinic acid gives benzarsinic acid; acetylamidotolylarsinic acid gives acetylamidobenzarsinic acid, from which, by removing the acetyl group by hydrolysis and replacing the amino group by hydroxyl through the diazo reaction, salicylarsinic acid may be obtained.

V. One or both of the hydrogen atoms in an amino group may be replaced by *acyl* radicals, to form compounds either of the acetanilide or of the phenylglycine type,

$$NH{\cdot}COCH_3, \quad -NH{\cdot}CH_2{\cdot}CO_2H, \quad -NH{\cdot}CH_2{\cdot}CONH_2.$$

(*a*) Type $R{\cdot}NH{\cdot}CO{\cdot}CH_3$ is formed:

(i) By the action of acid anhydrides on amino-arsinates;

(ii) By the action of the anhydrides on the amino-arsinic acids in presence of water;

(iii) By the action of acid chlorides in presence of pyridine, caustic soda or sodium carbonate, for example, atoxyl is converted in this way into *Hectine*. By heating amino-arsinic acids with certain esters of dibasic acids, derivatives are formed, thus, with ethyl oxalate oxalyl-atoxyl (oxalyl-*p*-arsanilic acid) is produced:

$$C_6H_4\begin{cases} NH{\cdot}CO{\cdot}CO_2H \\ AsO_3H_2. \end{cases}$$

(*b*) Type $R{\cdot}NH{\cdot}CH_2{\cdot}CO_2H$, or $R{\cdot}NH{\cdot}CH_2{\cdot}CONH_2$, *e.g.*, phenylglycinearsinic acid, is prepared by heating the corresponding amino acid in concentrated aqueous solution with chloracetic acid in presence of two molecular proportions of sodium hydroxide.

(*c*) Type $R{\cdot}N = CH{\cdot}R'$. Amino-arsinic acids will react with

aldehydes, for example, atoxyl combines with benzaldehyde to form the benzylidene derivative,

$$C_6H_5\cdot CH = N\cdot C_6H_4\cdot AsO_3H_2.$$

VI. Introduction of *chlorine* or *bromine* may readily be carried out. When hydroxyphenylarsinic acid is treated with sodium hypochlorite or hypobromite it yields the disubstituted derivative with the two halogen atoms *ortho* to the hydroxyl group.

VII. *Diazo* compounds from amino-arsinic acids react to form azo compounds in just the same way as those from other aromatic amines or amino-acids. A large number of such products is known. The compound,

$$AsO_3H_2\cdot C_6H_4\cdot N : N\cdot C_6H_3\begin{matrix}CO_2H\\OH\end{matrix}$$

will serve as an illustration.

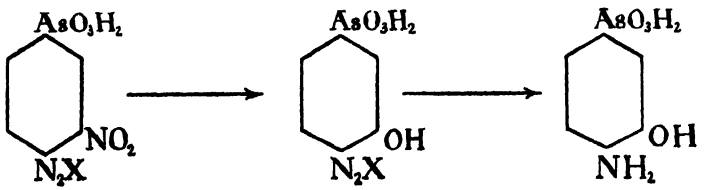
VIII. *Nitration.*—Arsanilic acid itself is not readily nitrated, its amino group must first be protected; but generally nitration is easily carried out, and many nitro-derivatives of phenylarsinic acid are known. It may be recalled that *p*-hydroxyphenylarsinic acid forms both a mononitro compound, an intermediate in preparing salvarsan, and a dinitro derivative.

IX. *Other reactions.*—Atoxyl may be oxidised by ammonium persulphate to form *phenazine* derivatives, the molecules of which contain two nitrogen atoms and two arsinic acid groups.

Mercury may be introduced into the ring by treatment with mercuric acetate. . . . .

As an interesting example of the variety of reactions which may be carried out starting from arsinic acids, the following may be quoted.

When *o*-nitroarsanilic acid is diazotised and treated with sodium acetate, a hydroxy derivative is produced, the nitro group having been replaced by hydroxyl:

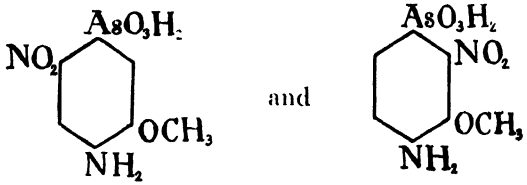


from which the aminophenol may be obtained, as indicated (by "coupling" and reduction), the arsinic acid group being unaffected. If, however, before the azo-hydroxy product is converted into the amino-hydroxy compound, it be methylated, anisidinearsinic acid,

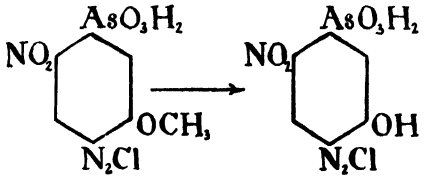
$$C_6H_3\begin{matrix}AsO_3H_2,\\OCH_3\\NH_2\end{matrix}$$

may be obtained as the final product.

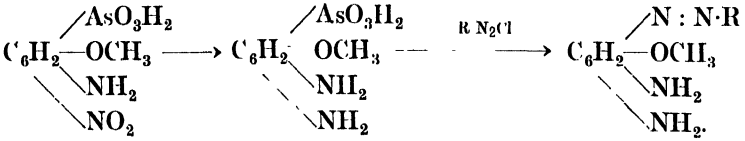
The reactions now begin to be very interesting. When anisidine-arsinic acid is nitrated it yields two products, namely,



These two isomerides are not equally soluble in water, and can therefore be separated. When the second is diazotised and the diazo-derivative warmed at 40°-50°, an unexpected replacement occurs, the methoxy group being lost and hydroxyl taking its place : [1]



If the first be reduced and then treated with a diazonium compound, for example, that from anisidine, an azo compound is formed and the arsenical group is displaced :



Our object here is to show, on the one hand, how interesting these arsenical compounds are from the theoretical point of view, and, on the other hand, how thorough must be the knowledge of organic chemical reactions possessed by those who work in this field.

General Properties of the Aromatic Arsenicals.

Acids.—Arylarsinic acids are all soluble in hot and many are tolerably soluble in cold water. Phenolic compounds are much more soluble than amino derivatives, and so also are the corresponding nitro compounds, *e.g.*, *p*-nitrophenylarsinic acid is more soluble than arsanilic acid.

Arsinic acids are not precipitated by magnesia mixture [2] in the cold, but nearly all are thrown down from a hot solution. This property allows of their separation from arsenic acid, which is precipitated by this reagent in the cold. In preparations made by Bart's method it is often necessary to purify the product in this way.

[1] The formulæ shown are incorrect : the nitro group should be in the 2–, not the 6–, position.

[2] Ammonium chloride, ammonia and magnesium chloride. (See Clowes and Coleman's *Quantitative Chemical Analysis*, 8th ed., p. 96, etc.)

Arsinic acids are acid to congo-red indicator, their amino derivatives are neutral. When arsinic acids are heated with Bougault's reagent (phosphorous acid in hydrochloric acid solution) the corresponding arseno compounds are produced *viâ* the dichlorides.

Arsenious oxides.—These substances are usually colourless, sparingly soluble in water and much more soluble in alcohol than the corresponding acids. Treated with reducing agents in the cold, they yield arseno compounds.

Arseno compounds.—These derivatives have all a yellow colour and are insoluble in water. They take up iodine and so decolorise a solution of iodine in potassium iodide. Salvarsan, diaminodihydroxyarsenobenzene dihydrochloride, does not give a precipitate with silver nitrate ; the silver chloride formed remains in solution and is probably attached to the arseno grouping.

PROPERTIES OF THE CHIEF AROMATIC ARSENICALS USED IN PHARMACY

Atoxyl.—Atoxyl is the sodium salt of arsanilic acid. As already stated, Béchamp, by whom it was discovered (in 1863), considered it to be an anilide of arsenic acid :

$$C_6H_5{\cdot}NH{\cdot}AsO_3NaH{\cdot}nH_2O.$$

Landsberger was the first to recommend the use of atoxyl in treating anæmia, skin diseases, and so forth, but at that time he was unable to establish the true constitution of the compound nor did he show that it was identical with Béchamp's product. Fourneau proved that the two substances were identical, but it remained for Ehrlich and Bertheim to settle conclusively the constitution. These investigators showed that the compound was no other than arsanilic acid, strictly comparable with sulphanilic acid, its molecule containing a free amino group and an arsenic radical attached directly to the nucleus. This discovery was of capital importance for the development of chemotherapy. If atoxyl had been an anilide, an arsenanilide, it could not have been much else than a chemically rather inert substance, but when it appeared that it really possessed a free amino group then all the transformations that aniline was capable of it also could be made to undergo. So it is that atoxyl has been a most valuable starting material for preparing other arsenical compounds.

Most of the reactions on which further treatment of atoxyl is based have already been reviewed, but in view of their importance, it will be useful to group them together here :

(i) Nitration of atoxyl produces mainly the 3 : 5-dinitro derivative, but if the oxalyl derivative, $C_6H_4(AsO_3H_2)NH{\cdot}CO{\cdot}CO_2H$, be used, the 3-mononitro compound is formed ; this is a starting point in making salvarsan.

(ii) When atoxyl is heated with hydriodic acid, *p*-iodoaniline is formed and so the constitution proved.

(iii) The diazonium compound obtained when atoxyl is diazotised will form azo compounds with phenol, salicylic acid, and the like, just like diazobenzene.

(iv) By means of the diazo reaction the amino group may be replaced by ·CN, ·AsO_3H_2, ·I, ·OH, etc. So in this way *p*-hydroxyphenylarsinic acid, the primary material in another process for making salvarsan, may be obtained.

(v) Any N-substituted derivative may be obtained just as with aniline. Some of these, acetylatoxyl, *Hectine* and others, have found employment in pharmacy.

(vi) When atoxyl is reduced with phosphorous acid and a little hydriodic acid, it yields aminophenylarsenious oxide.

(vii) If hydrosulphite be used, diaminoarsenobenzene is produced.

Atoxyl forms a white, crystalline powder, with a refreshing kind of taste, soluble in about 6 parts of water, and very sparingly so in alcohol. The crystals contain four molecular proportions of water, which are lost at 108°. The dry substance dissolves freely in methyl alcohol. A 10% solution gives a green precipitate with ferrous sulphate, and white precipitates with mercuric chloride and silver nitrate. If it be added to a solution of gold chloride and sodium bicarbonate, a very stable preparation of colloidal gold is obtained. Thomas, in England, was the first to experiment with atoxyl in trypanosome diseases, after Laveran had shown that arsenic was of value in such cases. Salmon was a pioneer in successfully using atoxyl in large doses for treating syphilis; but to Ehrlich and his pupils is due the systematic study of atoxyl and the resulting great progress in arsenical chemotherapy.

Arsacetine.—

$$C_6H_4\begin{cases}NH\cdot CO\cdot CH_3\\ AsO_3H_2.\end{cases}$$

Arsacetine is the acetyl derivative of atoxyl obtained by treating the latter with acetyl chloride or acetic anhydride. It is no longer in use.

Hectine.— $C_6H_4\begin{cases}NH\cdot SO_2\cdot C_6H_5,\\ AsO_3H_2\end{cases}$ is the benzene-sulphonic derivative, discovered by Mouneyrat.

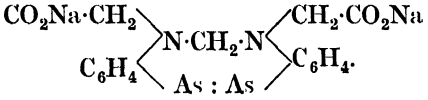
Arsenophenylglycine.—

$$CO_2Na\cdot CH_2\cdot NH\cdot C_6H_4\cdot As : As\cdot C_6H_4\cdot NH\cdot CH_2\cdot CO_2Na.$$

Phenylglycinearsinic acid is obtained by treating atoxyl with a hot solution of sodium chloracetate. The product is dissolved in boiling water and ten times its weight of sodium hydrosulphite dissolved in five parts of water added. From the hot solution arseno-

phenylglycine soon separates and is filtered off and converted into the sodium salt.

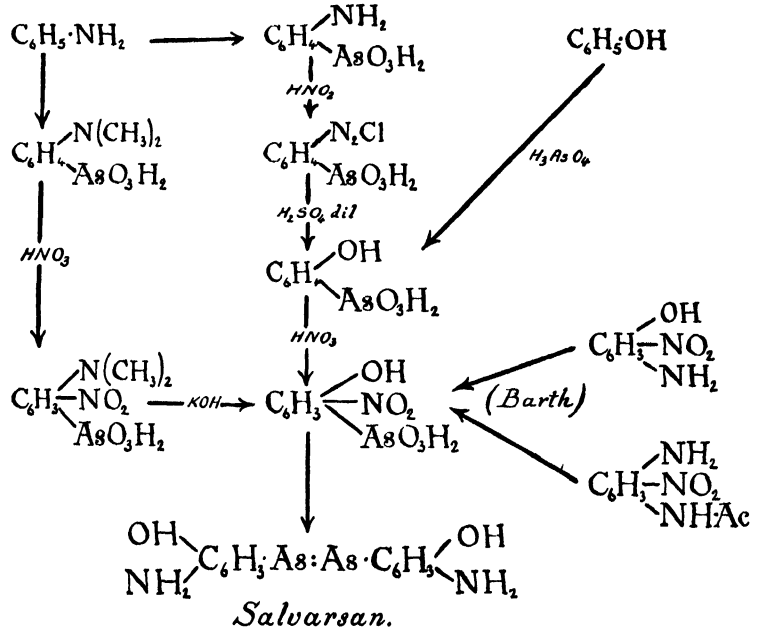
This compound has a remarkable trypanocidal action, and is, in fact, the best product available for treating trypanosomiasis. Unfortunately it is very unstable; but, according to recent patents of Poulenc and Oechslin, it may be stabilised either by treatment with acetic anhydride or with formaldehyde. A derivative, *Osarsan*, prepared in this way, has been shown by Laveran and Mesnil to be an improvement on the original arsenophenylglycine. Its structure is probably:



Salvarsan (*"606"*, *Arsenobenzol*, *Kharsivan*, *Arsphenamine*).—Salvarsan is diaminodihydroxyarsenobenzene. It is prepared as follows:

Atoxyl is diazotised and converted into *p*-hydroxyphenylarsinic acid. This is nitrated, the nitro group locating itself *ortho* to the hydroxy group. The nitrohydroxyphenylarsinic acid so obtained, reduced with sodium hydrosulphite, yields directly salvarsan.

Hydroxyphenylarsinic acid may also be obtained by treating phenol with arsenic acid; its nitro derivative by heating nitrodimethylaminophenylarsinic acid with caustic soda (Oechslin), or from nitroaminophenol or nitroaminoacetanilide, by employing Bart's



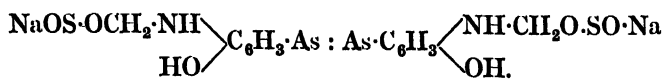
Salvarsan.

method. Lastly, the nitro acid may be prepared by nitrating oxalyl-atoxyl, and treating the product with caustic soda, which first removes the oxalyl group by hydrolysis, and then causes the replacement of the amino group by hydroxyl. The best way of converting nitrohydroxyphenylarsinic acid into salvarsan is to use sodium hydrosulphite as the reducing agent, but the reduction may be carried out in stages, using the methods described above, and a purer product is obtained in this way.

The free salvarsan base forms a yellow powder, soluble in dilute hydrochloric acid and sodium hydroxide, insoluble in acetic acid. By adding sodium sulphate to a solution of salvarsan in dilute hydrochloric acid, the material is precipitated completely as an insoluble sulphate. It is employed as the hydrochloride and must be kept in closed bottles filled with carbon dioxide. The commercial product contains certain impurities which cannot be satisfactorily traced by chemical analysis, the presence of which is shown, however, by the widely varying toxicity of different samples. Tests must therefore be made on animals; rats should endure a dose of at least 0·12 gm. per kilogramme body weight.

Neosalvarsan (*Novarsenobenzol*).—This is a compound of salvarsan with the addition product of formaldehyde and sodium hydrosulphite. It is prepared by dissolving salvarsan in water and adding first a solution of sodium formaldehyde-sulphoxylate, then, after an hour, successively, sodium carbonate (10%) and hydrochloric acid (12%). A yellow precipitate is thrown down; it is filtered off, redissolved in just as much caustic soda as is necessary, and reprecipitated by alcohol. Neosalvarsan forms a yellow powder, dissolving in water to a neutral solution; if an acid be added to this solution a precipitate is formed which is not redissolved by an excess of the acid. This property differentiates the new product from salvarsan.

Neosalvarsan usually contains certain impurities, and the percentage of arsenic is only about twenty instead of nearer thirty. The following formula probably represents its constitution:



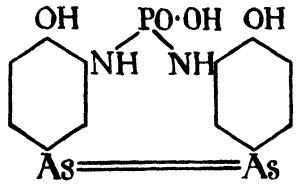
Of this product rats should stand a dose (intravenous injection) of at least 0·2 gm. per kilo.

A formaldehyde-bisulphite derivative of salvarsan,

$$-NH{\cdot}CH_2O{\cdot}SO_2Na,$$

has also been prepared.

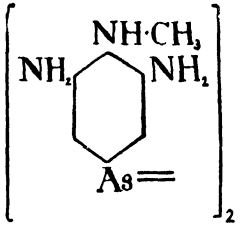
Galyl.—This derivative was discovered by Mouncyrat and is a compound of salvarsan with phosphoric acid, namely, dihydroxy-arsenobenzenephosphamic acid, probably having the structure:



Ludyl, which also was discovered by Mouneyrat, is a compound with benzene-*m*-disulphonic acid :

$$C_6H_4\begin{matrix}/SO_2\cdot NH\cdot C_6H_3(OH)\cdot As : As\cdot C_6H_3(OH)\cdot NH_2 \\ (m) \\ \backslash SO_2\cdot NH\cdot C_6H_3(OH)\cdot As : As\cdot C_6H_3(OH)\cdot NH_2.\end{matrix}$$

Arsalyte is dimethylhexaminoarsenobenzene :



Like other *meta*diamine derivatives, this base has the remarkable property of dissolving in aqueous solutions of sodium bicarbonate to form a carbamic salt.

Luargol, one of Danysz's products, is a compound of salvarsan with silver bromide and antimony oxide. The combinations of salvarsan with metallic salts were described in detail by Ehrlich at the International Medical Congress held in London in 1913. At the present time the use of silver compounds of salvarsan is being extensively developed, principally in Germany.

EXPERIMENTAL CHEMOTHERAPY OF TRYPANOSOMIASIS AND SPIRILLOSIS

The aim of chemotherapeutics is to cure infectious diseases by directly attacking their cause, *i.e.*, the microorganisms or their secretions, using only chemical substances.

Experimental chemotherapy cannot be carried out with man as the subject, and so it is necessary to infect animals with the disease it is desired to study. The microorganisms hitherto investigated are the spirilla and the trypanosomes. These organisms usually secrete but feebly toxic products, and consequently the infected animal is only slowly killed; their incubation stage is over in a short time, their development is well marked out, they circulate in the blood stream, and so are easily affected by medicaments; both kinds of organism, particularly the trypanosomes, may readily be transmitted to small animals, mice, birds, guinea-pigs, fowls, such as are used in

the laboratory; for all these reasons they can very properly be subjected to research of this kind. Further, the organisms are extremely mobile, and so it is easy to tell, simply by examining a drop of blood, whether they are present, either dead or alive, or whether they have disappeared.

In the particular case of syphilis, experimental work, already successfully begun by Metchnikoff and Roux with monkeys as subjects, became much more easily carried out when it was found that rabbits could be infected and a syphilitic condition produced experimentally.

Chemotherapeutical problems of this kind are attacked as follows:

Some substance having been found (empirically) to exhibit an action on microorganisms, every resource offered by chemistry is applied so to modify it as to produce derivatives with the maximum activity against the organisms, and a minimum activity against the organs of the body. The aim, therefore, is to augment *parasitotropic* and diminish *organotropic* action, that is, if C be the curative dose, and T the tolerated dose, the ratio $\frac{C}{T}$ should be as low as possible.

One should distinguish between action *in vitro* and action *in vivo*. Action *in vivo* is preventive or curative.

To determine the action *in vitro*, shortly after the infection, so that they do not contain many parasites, mice are taken, their throats cut and the blood mixed with physiological saline. Part of the mixture is set aside as a reference sample;[1] the rest is divided up into test-tubes containing an equal volume of physiological saline and increasing doses of the chemical product under examination. The tube contents are examined every two minutes and the time noted at which the parasites cease to move about. As a control, the mixture at this stage is injected into fresh animals, which should not take the infection.

To determine the action *in vivo*, it is first necessary to examine the toxic action of the product on the particular kind of animal serving as subject in the experiments, so as to establish the tolerated dose; this should be well above that to be given to the infected animals and should serve as a limit to be only gradually approached. This done, two ways are open: either one may inject first the dose of medicament and then the parasites, allowing varying intervals of time to elapse between the two injections, or one may wait until the infection is well developed and then administer the drug. In the first case, the degree of immunisation is measured; in the second, that of sterilisation.

After this preliminary exposition we may turn to the more interesting results of chemotherapeutical investigation.

Relations between Curative Action and Constitution among Chemical Products.—Laveran and Mesnil, Lingard, and Bruce had already found that arsenic had a beneficial effect on trypanosome diseases;

[1] "Control."

but Thomas, another Englishman, working in collaboration with Breinl, seems to have been the first to employ arsenic in the form of atoxyl. Thomas's results were so encouraging that attempts to utilise the new product were made on all sides.

Atoxyl was, indeed, a great advance on arsenious acid: not only is it effective in experimental trypanosomiasis, but it, together with arsenophenylglycine and osarsan, is the best known remedy against "sleeping sickness." A curious fact has been noticed by several observers, namely, that atoxyl has hardly any action *in vitro*. Levaditi has put forward the hypothesis that it forms some sort of combination with substances in the liver and that it is the product, "trypanotoxyl," which is the active agent. Ehrlich, however, supposes that atoxyl suffers reduction in the organism, and that the trypanocidal action is due to the reduction products. Breinl and Nierenstein, on the other hand, attribute the activation of these arsenicals to their undergoing oxidation. Uhlenhuth is of the same opinion, and has demonstrated that injection of atoxyl is followed by the appearance in the urine of a derivative of aminohydroxyphenylarsinic acid.

No matter what may come of these hypotheses, that of Ehrlich has shown itself to be fruitful from a practical point of view. Ehrlich's plan was to make a number of reduction products of arsenical compounds and compare their activity *in vitro* with that of the parent unreduced substances. He obtained surprising results. Thus, for example, *p*-hydroxyphenylarsinic acid will kill trypanosomes (*ferox*) at a dilution of 5/100, whilst the reduction product, *p*-hydroxyphenylarsenious oxide, destroys them in half an hour at a dilution of 1/1,000,000. The same relationship was found to hold between atoxyl and its reduction product.

A big step forward in experimental chemotherapy seemed to have been made. There followed a brilliant investigation carried out under Ehrlich's direction by Hata; and it is this work, dealing with a considerable number of arsenicals, which we will now review, for it is a model of its kind.

Ehrlich and Hata's results, with those of later investigators, fix certain points from which the relations between chemical constitution and action on microorganisms in this series may, to some extent, be mapped. Hata's work had as subjects spirillosis in fowls, relapsing fever and syphilis. It should be pointed out forthwith that no conclusions as to the action on trypanosomes can be drawn from that on spirilla; indeed, some products which are active enough towards the latter have no effect on the former. However, the following is a brief account of Ehrlich and Hata's results.

First of all, as regards relapsing fever, atoxyl acts very feebly, the ratio $C : T$ is not greater than 1 : 2. Acetylatoxyl is no more active, but as it is less toxic, the ratio becomes 1 : 3. Dichlorohydroxyphenylarsinic acid exhibits a much more marked action;

the poisonous dose being 1 c.c. of a 1 in 75 solution, 1 c.c. of a 1 in 100 solution is enough to bring about a definite cure. Aminohydroxyphenylarsinic acid has much the same effect.

Turning now to the arseno compounds, arsenophenylglycine is hardly more active than arsacetin (acetylatoxyl). With salvarsan, the ratio $C:T$ is no higher than for the aminohydroxyphenylarsinic acid from which it is derived. The iodoarseno derivative of aminophenol is inferior even to atoxyl.

Thus it is already evident that Ehrlich's theory is not in agreement with the facts. Arseno compounds are not more active than certain arsinic acids, at any rate, so far as relapsing fever is concerned. The real reason for choosing arseno compounds for practical use instead of arsinic acids lies in the fact that such of the latter as had been tested appeared to cause nervous disturbances, and this was not the case with the former. Arsinic acids have, for instance, a more or less serious effect on mice, the most remarkable manifestation of which is the transformation of the animals into dancing mice. Again, the use of atoxyl and its simple derivatives in treating syphilis was abandoned, mainly because it resulted, only sometimes, to be sure, in serious ocular trouble.

Returning to the main theme, we have seen that Ehrlich's theory is unsatisfactory, at any rate, as far as relapsing fever is concerned. Yet this conclusion does not appear to hold for spirillosis in fowls or syphilis. A definite statement cannot be made because the action of arsinic acids in these diseases has not been exhaustively investigated. But salvarsan has proved itself much superior to atoxyl and arsacetine as a specific for spirillosis in fowls; thus, for these compounds the ratio $C:T$ is not greater than 1 : 3, but for salvarsan the value is 1 : 58, the toxic dose being 0·10 gm. per kilo. body weight, whilst the curative dose is less than 0·003 gm.

We may review, in conclusion, the experiments on syphilis with rabbits as subjects. Rabbits may readily be made to develop syphilitic chancres; the ulcers become charged with spirochæts, and their development is slow. The usefulness of a medicinal product can be gauged by the effect it has on the cicatrization of the chancre and on the rate at which the spirochæts disappear. The following results were obtained with the three substances investigated by Hata. The ratio $C:T$ is 1 : 7 for salvarsan; the tolerated dose was 0·10 gm. per kilo. body weight, whilst the administration of only 0·014 gm. caused all the spirochæts to disappear and the syphilitic ulcers to heal in a few days. Anyone who has seen these remarkable effects of such small doses actually taking place in the animal is not surprised at Ehrlich's enthusiasm. Arsenophenylglycine was found to be much less active than salvarsan; aminohydroxyphenylarsenious oxide has the ratio $C:T = 1:5$, but it is very poisonous and difficult to deal with.

Hata's investigations were not extended to trypanosomiasis. In this field the work has been done chiefly by Laveran and Mesnil in the Pasteur Institute. So far the best products known for treating diseases of this class are, first, atoxyl, which still remains the most widely used arsenical compound, mainly for "sleeping-sickness"; secondly, arsenophenylglycine; and lastly, a derivative of arsenophenylglycine discovered by Oechslin, namely, *Osarsan*; but quite recently the Rockefeller laboratories have put out the amide of phenylglycinearsinic acid,

$$C_6H_4\begin{cases}NH\cdot CH_2\cdot CONH_2\\ AsO_3HNa.\end{cases}$$

This is only feebly toxic and seems to be superior to atoxyl.

By reviewing all the available data one may arrive at certain interesting conclusions. In the first place, although Hata investigated derivatives of both trivalent and pentavalent arsenic in their action on spirillosis in fowls and relapsing fever, he did not compare the arsinic acids with either arseno compounds or arsenious oxides as to their value in the treatment of syphilis in rabbits, and this leaves an unfortunate gap to be filled. Moreover, the experimental results with relapsing fever and spirillosis were not always in agreement with Ehrlich's theories: from experiments *in vitro*, Ehrlich concluded that trivalent arsenic derivatives should be more active than those of pentavalent arsenic, even *in vivo*; but on this very point his conclusions were not confirmed by Hata's results. Thus, taking dichlorophenylarsinic acid as one example, this acid certainly has an action on the nervous system of animals which would make its use on man dangerous; but it is a much more energetic agent against certain parasites than the arsenious oxides and, indeed, is quite as active as salvarsan. Again, to take another example, aminohydroxyphenylarsinic acid is much more active than atoxyl, of which it is a derivative, a great increase in activity having resulted from the mere introduction of a hydroxyl group; it is, in fact, quite as active at any rate towards the diseases on which it has been tried, as the corresponding arseno compound, namely, salvarsan. So in this case, conversion of an amino into an aminohydroxy derivative brings about a sharp jump in activity, but little or no alteration follows the change from the acid to the arseno derivative. Hence one may say that, in this instance, an oxidation has had just as beneficial results as a reduction.

Besides, there is no evidence that aminohydroxyphenylarsinic, or any other acid, should not be as effective as salvarsan in experimental syphilis (rabbits). If it is proved that the effect on the nervous system is always associated with the presence of the arsinic acid group, then we might hesitate to utilise the substances in question, however active they might be; but at present the examples studied

are not numerous enough for us to affirm that always a free arsinic acid group means that the nervous system will be affected.

Perhaps it will be easier to find acids causing no nervous disturbances when administered in curative doses than arseno compounds readily available for hypodermic injection. When one thinks of the advantages to be gained by using the acids, capable as they are of giving neutral salts, stable and easily injected, one wonders why research work along these lines has been so systematically neglected.

However, investigations already made still furnish results that are worthy of our attention. When, for example, a chlorine atom is introduced into the molecule sometimes the therapeutical properties are improved, sometimes they are not; no general conclusions can be made. With certain products the introduction of iodine is followed by a rise in curative activity, at least, so far as the treatment of relapsing fever and fowl spirillosis is concerned, but the derivatives obtained are more toxic and the ratio $C:T$ is not altered to any great extent. The di-iodo derivative of salvarsan is less active towards trypanosomes than the parent compound.

Arsenophenylglycine, which is so active in trypanosomiasis, and, indeed, is the best specific known for treating such diseases, has practically no action in spirillosis and experimental syphilis. Salvarsan, on the contrary, is of much less value in trypanosomiasis than arsenophenylglycine or even than atoxyl.

The introduction of a second amino group into atoxyl produces a less poisonous product. Diaminophenylarsinic acid is only $\frac{1}{25}$ as toxic as atoxyl, but, like the latter, it brings about prolonged nervous disturbances.

Methylation of the amino group has an unfavourable (dystherapeutic) influence. Every time that this expedient has been tried a more poisonous product with less curative activity has been obtained. Methylation of the hydroxyl group has the same effect.

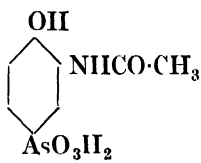
The influence of the position of the substituent groups has also been the subject of several investigations, and we will give one illustration of the kind of effect observed, viz.: *o*-aminophenylarsinic acid is much more toxic than the *para* isomeride, atoxyl.

It can be said, in summing up, that there is still plenty to do in the arsenical field. To be sure, it will be difficult to displace salvarsan and its derivatives as specifics for syphilis. The cases successfully treated with these medicaments are very numerous, and the habit of employing them has gradually developed in spite of the difficulties involved in handling them; but it cannot be said that they represent an ideal. Evidently, if one could discover some substance the administration of which could be carried out by simple hypodermic injection and which, therefore, could be carried out by any medical man, or better still, if one could find a drug that could be given by

mouth to cure syphilis as radically as salvarsan, then progress would indeed have been made.

And there are not only the ailments of mankind, but also those of the lower animals, to consider. It is reckoned that the losses due to trypanosomiasis or spirillosis amount annually to hundreds of millions. Here is fruitful ground in a vast field open to the pioneer.

Note.—M. Fourneau and his collaborators have now carried out an extensive investigation of the arsinic acid series (see *Applied Chemistry Reports*, 1923, VIII., 533). *p*-Hydroxy-*m*-acetylaminophenylarsinic acid,



has been put on the market under the name *Stovarsol*. This is the first arsenical for curing syphilis to be administered *by mouth*. If taken in time, it will also act prophylactically against syphilis.

APPENDIX—RECIPES

5-Nitro-2-methylphenylarsinic Acid.—*Preparation of o-Tolylarsinic Acid by Bart's Method.* 53 gm. *o*-Toluidine is dissolved in 500 c.c. water and 165 c.c. hydrochloric acid (sp. gr. 1·19), and the solution diazotised at 5° by 35 gm. sodium nitrite in 140 c.c. water. To the diazo solution is added 130 gm. sodium arsenite in 260 c.c. water, followed by 100 c.c. 10*N* sodium hydroxide solution. The mixture is set by for several hours, filtered, and treated successively with 300 c.c. concentrated ammonia, 75 c.c. hydrogen peroxide, and, finally, 2,000 c.c. of a $\frac{2}{3}$-*N* magnesia mixture. The precipitate of magnesium ammonium arsenate is filtered off and the solution boiled, whereupon the magnesium salt of the tolylarsinic acid is thrown down. This salt is sparingly soluble in hot water. It is collected and treated with 52 c.c. hydrochloric acid (sp. gr. 1·12), diluted with a little water, to liberate the free acid, which separates in a crystalline form, and after purification by recrystallisation from hot water, melts at 160°.

Nitration.—5 gm. Tolylarsinic acid is added to a mixture of 25 gm. concentrated sulphuric acid and 20 gm. nitric acid (sp. gr. 1·49) at 20°–25°. After standing for a quarter of an hour, the mixture is poured into six times its volume of water. Strong caustic soda is added until most of the sulphuric acid is neutralised, when the nitro acid separates in the form of silky needles. It is moderately soluble in hot water, but sparingly so in cold, and melts at about 250° after becoming discoloured at 230°.

4-Chloro-2-methylphenylarsinic Acid (*Bart's Method*).—15 gm. *p*-chloro-*o*-toluidine is suspended in a mixture of 300 c.c. water and 200 c.c. hydrochloric acid (sp. gr. 1·19), and diazotised at 5° by the addition of 150 c.c. of a 2*N* solution of sodium nitrite. To the diazo solution, 130 gm. sodium arsenite in 200 c.c. water is gradually added, followed by about 200 c.c. 10*N* caustic soda. Nitrogen is evolved. When the reaction is over, the excess arsenious acid is oxidised by adding 75 c.c. hydrogen peroxide (30%). The magnesium salt of the arsinic acid is isolated as in the example above, using two litres of magnesia mixture. The yield here is only 8 gm. The free acid is obtained by decomposing the magnesium salt with hydrochloric acid.

The arsinic acid may be nitrated as in the preceding example.

2 : 4-Dihydroxyphenylarsinic Acid (*Action of Arsenic Acid on Phenols*).

Resorcinol	110 gm.
Commercial arsenic acid, 75° Bé. (= 83%) . .	171 ,,

are mixed and heated on the water-bath. Crystals gradually separate, and after an interval of some hours the crystalline mass is crushed up with acetic acid, filtered off and washed two or three times with acetic acid. The yield of clean product is 145 gm. The compound is readily soluble in water or alcohol, sparingly so in acetic acid or acetone.

5-Nitro-2 : 4-dihydroxyphenylarsinic Acid (*Nitration of the above*).—To a solution of 46·8 gm. of the dihydroxyphenylarsinic acid in 50 c.c. concentrated sulphuric acid, cooled to 0°, a mixture of 14 c.c. nitric acid (sp. gr. 1·4), mixed with its own volume of strong sulphuric acid, is added. The mixture is to be stirred vigorously ; as nitration proceeds it thickens, so that it is an advantage not to start with all the arsinic acid but only about two-thirds, and to add the rest when about half the " mixed acid " has been introduced. The mixture is left overnight and poured on to ice. Yield : 41·7 gm.

When this product is treated with bromine in acetic acid solution the arsinic acid group is displaced and dibromonitroresorcinol formed.

5-Amino-2 : 4-dihydroxyphenylarsinic Acid (*Reduction of the Nitro Group*).—32·5 gm. of the nitro acid is dissolved in 300 c.c. water and 100 c.c. 10*N* caustic soda, and treated with 70 gm. sodium hydrosulphite. Reduction takes place with rise in temperature. To the colourless solution 62 gm. glacial acetic acid is added ; the amino acid is quantitatively precipitated. For purification it is dissolved in dilute hydrochloric acid, treated with bone-black and reprecipitated by sodium acetate.

4-Nitro-2-carboxyphenylarsinic Acid (*Bart's Method*).

5-Nitroanthranilic acid . . .	18·5 gm.
Hydrochloric acid (sp. gr. 1·12) . .	100 c.c.
Water	50 ,,

A solution of the above is made and diazotised at 5° by adding 50 c.c. of 2*N* sodium nitrite solution. To the filtered diazo solution, 26 gm. sodium arsenite, dissolved in 50 c.c. water, is added. When the nitrogen has all been evolved, the mixture is made neutral to congo-red by adding 10*N* caustic soda, and the arsinic acid thus precipitated. Yield: 22 gm.

4-Hydroxy-2-carboxyphenylarsinic Acid *viâ* the Corresponding Arseno Derivative.—14·5 gm. of the above nitro acid is dissolved in 180 c.c. water and 98 c.c. 10*N* caustic soda and mixed at 70° with a solution of 86 gm. crystallised ferrous sulphate in 200 c.c. water. The precipitated iron hydroxide is filtered off, washed with 200 c.c. boiling water, and the collected filtrates evaporated until crystallisation sets in. About 25 c.c. concentrated hydrochloric acid is added, until the solution is acid to congo-red, and it is cooled in a stream of water. The liquor is filtered off—it contains the amino acid—and diluted with 100 c.c. water, neutralised with caustic soda, 10 c.c. concentrated sulphuric acid added, cooled to 5°, and sodium nitrite solution run in until an excess is shown to be present by starch-iodide paper. The diazo solution is heated to boiling, filtered, 60 c.c. phosphorous acid (35%) and a little potassium iodide added, and the mixture heated on the water-bath until no more arseno compound separates out (about half an hour). This product is filtered off and well washed with water: it is suspended in a little water and treated with hydrogen peroxide until a colourless solution is produced. This is quickly filtered; it rapidly sets to a crystalline mass.

CHAPTER IX

ORGANIC COMPOUNDS OF MERCURY

One might have thought that the great and deserved success of salvarsan and the other arsenicals would have put an end to research among mercury derivatives, but, curiously enough at first sight, the exact contrary is what really happened, and activity in this field has never been so great as in the last few years. On reflection, however, one sees that this is not so surprising a result. Thus, in the first place, if arsenicals with more valuable therapeutic properties may be produced by suitably modifying simple derivatives, why, one might ask, should not similar successful results be obtained with mercury compounds? Secondly, as more and more cases were treated with salvarsan, it became evident that the number in which a complete cure, without relapse, was effected was relatively small and that we were, as yet, far from realising Ehrlich's "therapia sterilisans magna." Specialists on syphilis therefore tended more and more to adopt the practice of combining intensive arsenical with prolonged mercurial treatment. It should, moreover, be noted that as the technique of intravenous injection was unfamiliar to many medical men, they remained loyal to mercury.

Investigators had then two aims in view:

(i) To discover a mercurial derivative with a rapid action that should cleanse the primary lesion as quickly as arsenic, and at the same time have a more effective curative action in the secondary and tertiary stages.

(ii) Since there is decidedly a demand for mercurials, to devise improvements so as to make the injection as painless as possible.

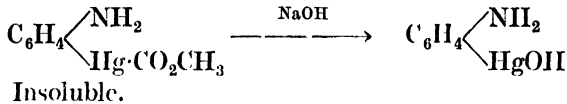
The first object has not been reached; the second has been attained but not in a completely satisfactory manner. The dominating feature of the whole history of research on mercurials is that the ratio of curative dose to toxic dose has never been modified to any considerable degree. If, occasionally, some improvement seems to have been made, as in Launoy and Levaditi's experiments on syphilitic rabbits, yet, when the tests have been continued on the human subject, it has been found either that the products in question were no better than known ones, or that, if there were any difference, it was so slight as not to encourage grappling with the difficulties of applying them.

Mercurial compounds may be divided into three classes;

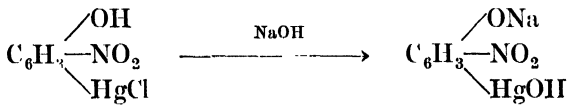
(i) True salts, *i.e.*, compounds formed by interaction of mercuric oxide with acids, such as mercuric chloride, acetate, benzoate, and so forth. It is characteristic of these compounds that all the combined mercury is set free as oxide when they are treated with a strong alkali. They are all very poisonous, no matter what acid they are derived from, and their therapeutic action is always proportional to the amount of mercury present. Experimental therapeutics cannot be carried out in this group.

We must disregard preparations containing minimal amounts of mercury, interesting though they be, as no organic mercury compounds are included among them.

(ii) Compounds in which one valency of the mercury atom is satisfied by an organic radical, *e.g.*, aminophenylmercuric acetate, methyl mercuric iodide, anhydro-*o*-hydroxy-mercurisalicylic acid, hydroxyphenylmercuric chloride. Such of these compounds as have no nuclear hydroxyl or acidic group are insoluble in caustic soda, the others are soluble. In every case, caustic soda does not throw down mercuric oxide, but the alkyl- or aryl-mercuric hydroxide :



Insoluble.



Soluble.

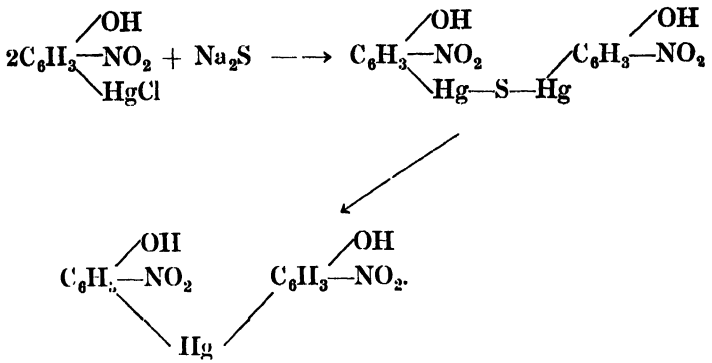
The compounds of this class are nearly all as poisonous as the salts described above.

(iii) Compounds in which the mercury atom is associated with two organic radicals, *e.g.*, mercury diethyl, mercury diphenyl.

$$C_6H_5{\cdot}Hg{\cdot}C_6H_5.$$

These substances are quite stable, they are unaffected by caustic soda.

The three classes of mercury compounds behave differently towards such other reagents as sodium sulphide and sodium hydrosulphite. Those of the first group are immediately decomposed by sodium sulphide, mercuric sulphide being precipitated. Those of the second class are also decomposed with formation of mercuric sulphide, but only half the mercury present is precipitated in this form, the other half remains attached to the organic radical and links up with the second radical set free by the precipitation, a complex sulphide being formed as an intermediate stage, thus :



Derivatives of the third class do not react with sodium sulphide; on the contrary, as we see, sodium sulphide is used in their preparation.

Sodium hydrosulphite brings about similar decompositions, but here the mercury is precipitated as metal. If any nitro groups are present they undergo reduction.

Compounds of the first two classes have all much the same action on the organism, although it does appear that the mono-aryl mercurials are less toxic than the others. But it should be pointed out that the exact degree to which a compound of this kind is toxic is difficult to determine. After the injection several weeks may elapse before death ensues, although the animal does not recover. It loses weight; it suffers from enteritis; it looks very ill; but it endures for a long time. So one must needs wait before drawing any conclusions. Furthermore, the particular resistance of each individual animal must be taken into account. May one truly say that one product is more toxic than another when the administration of the first is followed by death after fifteen days, and that of the second after nineteen days? Generally, the toxic strength is measured by the dose which causes death in two or three days, sometimes by that which takes ten days, at other times by the dose which brings about a progressive loss in weight. If, for example, a certain amount of a mercurial causes an animal to lose weight for four days and then begin to fatten again, the toxic dose may be considered to be that which, injected into an equally heavy animal, just suffices to cause progressive wasting, continuing until death supervenes.

The dialkyl and diarylmercury derivatives are not more poisonous than the corresponding uncombined organic radicals, except, perhaps, those of the lower members of the aliphatic series and certain particularly unstable compounds which will be described later (mercury diaminodihydroxydiphenyls).

Besides the substances in the above three classes, there are others which, strictly speaking, could also be placed in one or other of those groups, but which are best considered apart. Thus, there are com-

pounds in which mercury is attached to nitrogen, mercury succinimide, mercuric cyanide; others in which mercury is attached to sulphur, and so forth. Examples are:

Sodium mercury thioglycollate, $Hg\langle^{S\cdot CH_2\cdot CO_2Na}_{S\cdot CH_2\cdot CO_2Na}$.

Dithiocarbamates, $Hg\langle^{S\cdot CS\cdot N(CH_3)_2}_{S\cdot CS\cdot N(CH)_3)_2}$.

These thio derivatives are true complex sulphides stable towards sodium hydroxide, but losing their mercury when treated with sodium sulphide. The dithiocarbamates are soluble in organic solvents (Delépine), and those derived from amino acids are, as sodium salts, soluble in water (Fourneau).

$$Hg\langle^{S\cdot CS\cdot N(CH_3)\cdot CH_2\cdot CO_2Na}_{S\cdot CS\cdot N(CH_3)\cdot CH_2\cdot CO_2Na}.$$

Preparation and Properties of Some Mercurials.—In this section a description of all known organic mercury compounds will not be attempted, nor will all those actually utilised be described, but the aim will be to illuminate the more interesting features of this branch of chemistry.

If a layer of metallic mercury be covered with one of methyl iodide and exposed to sunlight, there gradually forms at the interface, together with a little mercuric iodide, a mass of crystals. In a few days all the methyl iodide—and much of the mercury—has disappeared and only these crystals remain. The substance is almost completely dissolved by ether, and when the ethereal solution is allowed to evaporate slowly by exposure it leaves behind a crop of splendid crystals of methylmercuric iodide, $CH_3\cdot Hg\cdot I$.

This curious reaction is not a general one, although it occurs also with allyl iodide. Other alkyl iodides react further, the action of the mercury here being to remove completely the iodine from two molecular proportions of the iodide. So in order to obtain the higher homologues of methylmercuric iodide, mercury dialkyls must first be made, these derivatives being readily formed by interaction of alkyl iodides and sodium amalgam in alcoholic solution.

The mercury alkyl compounds are reputed to be very poisonous, but in reality they have not been investigated to any great extent. Schaeffer's recent work on compounds prepared by Tiffeneau [1] suggests that homologues above mercury dimethyl are only feebly toxic. Mercuridipropionic acid,

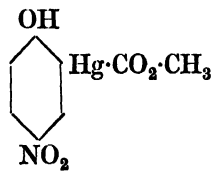
$$Hg\langle^{CH_2\cdot CH_2\cdot CO_2H}_{CH_2\cdot CH_2\cdot CO_2H},$$

may be mentioned in passing. The sodium salt of this compound is

[1] *Bulletin des Sciences Pharmacologiques*, 1924, *28*, 65–69.

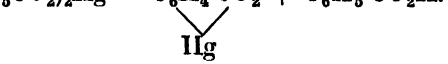
soluble in water. It is quite non-poisonous and at one time great hopes were based on it (E. Fischer). Not improbably, if this molecule were made less stable, as by the introduction of a hydroxyl or amino group, some or all of the toxic properties would be regained.

The introduction of mercury[1] takes place with outstanding facility in the aromatic series, a mercurial radical entering the ring as easily as a nitro group. Thus, if almost any aromatic compound be warmed with mercuric acetate in dilute acetic acid solution, a derivative is formed containing one, or even two, mercurial complexes, $-HgO_2CCH_3$. The presence of other substituents, provided the *ortho* and *para* positions are not blocked, facilitates the reaction. Phenol and aniline yield a mixture of *ortho*, *para* and di-substituted mercurial derivatives. *p*-Nitrophenol reacts practically instantaneously and yields the monomercurial derivative :



whilst *o*-nitrophenol gives a mixture of mono- and di-substituted derivatives.

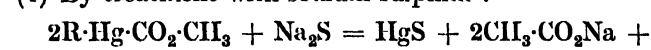
When mercury salts of aromatic acids are heated to a certain temperature (100°–160°) the mercury atom migrates into the ring ; this action takes place with acids of all kinds. Mercuric benzoate, for example, reacts thus at 120° :



The reaction is at an end when a little of the substance no longer gives an orange-coloured precipitate with sodium hydroxide.

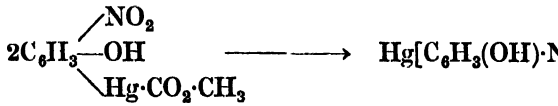
These mercury mono-aryl compounds readily yield diaryl derivatives, thus :—

(1) By treatment with sodium sulphide :



In this reaction the sulphide, R·Hg·S·Hg·R, is formed as an intermediate product.

(2) By heating with a solution of sodium hydrosulphite or stannous chloride. Any nitro groups present are simultaneously reduced. An example of this, which is mentioned again later, is the following :

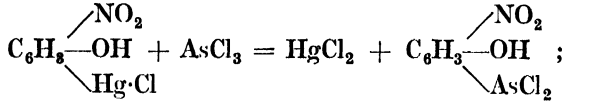


[1] "Mercuration" or "Mercurisation."

A third method of preparing diaryl derivatives is to treat aryl bromides, *e.g.*, bromobenzene, with sodium amalgam in presence of ethyl acetate :

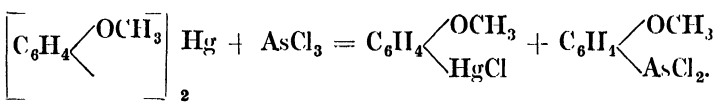
$$2C_6H_5Br + HgNa_2 = 2NaBr + Hg(C_6H_5)_2.$$

When many mono-arylmercuric salts are treated with arsenious chloride they give the corresponding arylarsenious chloride (Roeder) :



when treated with iodine, the mercury atom is displaced, an iodine atom taking its place ; sometimes another is introduced at the same time. 3-Nitro-4-hydroxyphenylmercuric acetate, for example, yields diiodonitrophenol.

Mercury diaryls interact with arsenious chloride when the two substances are heated together, the mercury being displaced by the $-As{\cdot}Cl_2$ group :



The main interest in these compounds centres round this reaction.

Sodium also will displace mercury from the diaryl derivatives : [1]

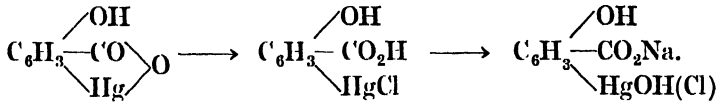
$$(C_6H_5)_2Hg + Na_2 = 2C_6H_5{\cdot}Na + Hg.$$

Mercurial Chemotherapy.—All the efforts to bring about some alteration in the ratio $C : T$ may be regarded as fruitless, as has already been observed. As far as the first class of mercury compounds is concerned, variation is impossible. The acid may be changed, with advantage, when employment is in question, but the curative value is unaffected. The toxic strength is always proportional to the amount of mercury itself present.

The most widely used mercurial medicaments are all members of the second class of compounds. It is not that these substances are more active than the salts, but that they are less troublesome to use—thus, injection is generally a comparatively painless operation. A typical example is mercury salicylate, or rather the product improperly so called. This compound is not, strictly speaking, a simple salt, as the mercury atom is directly attached to the nucleus by one of its valency bonds. It actually exists as the internal salt, anhydro-*o*-hydroxymercurisalicylic acid, in neutral solution, whilst it dissolves in alkalies by virtue of the carboxyl group. It may also be rendered more soluble by salts, *e.g.*, sodium chloride, sodium methylarsinate (the product is then called *Enesol*), or sodium hydroxyaminobutyrate (this combination is *Asurol*). But no matter what combination is

[1] Acree, *Chemical Society's Abstracts*, 1903, i., 724.

formed, the therapeutic activity is always proportional to the amount of mercury present :

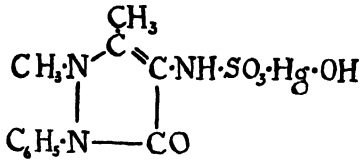


A nitrophenol derivative, namely, nitrohydroxyphenylmercuric hydroxide, has been recommended as an antiseptic for dressing wounds or chiefly for making antiseptic soap. It is soluble in caustic soda and possesses considerable bactericidal power.

Hermophenyl is a mercury-phenoldisulphonate.

Afridol, also used for preparing antiseptic soaps, is the sodium salt of the mercury derivative of toluic acid, $CH_3 \cdot C_6H_3(HgOH) \cdot CO_2Na$.

Besides the above, a considerable number of organic mercurials have been patented : mercuriphenoxyacetic acid, mercuri-di-salicylic acid, the mercuri-amino-sulphonate, $Hg : N \cdot SO_2K$; sulphamino-phenyldimethylpyrazolone mercury (Kolle and Scheitlin) :



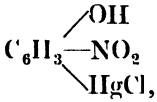
Kolle made this compound the subject of an investigation and came to the conclusion that it was superior to any other mercurial product, but nothing more has been heard of it. The same may be said of most of the others.

A characteristic property of all the compounds of the first two classes is that after injection the mercury accumulates in certain particular parts of the body, and the same effects, viz., nephritis and diarrhœa, are always produced. Moreover, and this point must be emphasised, it does not matter what agent is used to confer solubility, sodium chloride, sodium methylarsinate, and so forth, one centigram of mercury metal is, to all intents and purposes, as toxic in the one case as in the other. Specialists and manufacturing druggists now and again make play upon words ; evidently a given weight of a product containing two molecules of the methylarsinate to one of mercury salicylate will not have the same toxic effect as the same weight of another product containing the two compounds in equi-molecular proportions—but to obtain the same therapeutic results just so much more will need to be injected. There is, indeed, little to hope for in this series, at least one can hardly expect results like those obtained with arsenic compounds, where, as we have seen, a considerable alteration in toxic strength and curative activity was brought about by a small modification in chemical constitution.

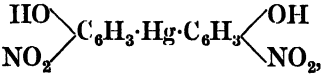
The dialkyl and diaryl mercurials are, however, not in the same

plight; here some success may be achieved. Mercury diphenyl is not poisonous: 0·5 gm. may be given to a rabbit with no more distressing result than that the animal fattens; but the compound is quite inactive in syphilis. Schrauth and Schoeller have investigated this group of compounds at length, and record certain observations, noted also by Blumenthal, that are most interesting in connection with the destination of the mercury inside the organism. A particular study was made of elimination, and for the first time it was found that mercury thus combined does not accumulate in the usual organs; this evidently means that the organic mercurial complex is not broken up and that the mercury is carried along by the rest of the molecule. That compounds which immediately lose their mercury and compounds in which it is firmly bound should be thus sharply divided, and there be no derivatives of milder character between, is obviously very doubtful. The aim of the investigator is to find these intermediates.

A recent discovery by Fourneau and Vila is of great significance. When the molecule becomes less stable, it becomes more toxic, even if the mercury atom be associated with two aryl radicals. This is the first indication of any considerable variation in toxic strength among the derivatives of a particular series. The mercurial derivatives of *p*-nitrophenol will be cited as an illustrative example. The compound,

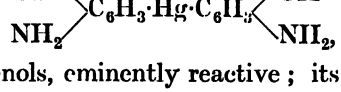


is very poisonous, nearly as poisonous as corrosive sublimate, at least if the doses represent equal amounts of mercury. When it is converted into the diaryl compound, leaving the nitro groups intact, mercury dinitrodihydroxydiphenyl is obtained. This substance,



is not toxic, except inasmuch as it is a nitro compound. It is very stable and is eliminated by the organism unchanged without any sign of mercurial poisoning appearing (Blumenthal). It has no action in syphilis experimentally induced in rabbits.

But when the nitro groups are reduced, mercury diaminodihydroxydiphenyl is formed, and this product,



is, like all aminophenols, eminently reactive; its solutions in alkalis blacken immediately, and when they are afterwards acidified, no white precipitate of mercury diaminodihydroxydiphenyl, but a black amorphous substance, is thrown down. Now this diamino derivative

is very poisonous, although its mercury is not removed either by caustic soda or hydrosulphite. It is not, however, stable towards the living cell. Probably what happens is that it is quickly "burnt up" in the organism and the mercury set free then exhibits its characteristic poisonous properties.

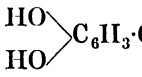
When the amino groups are acetylated the diacetyl derivative produced is more stable than the parent compound and also less toxic, but still more so than the original dinitro compound. The diacetyl derivative has a marked action on spirochæts, and, for the first time in mercurial chemotherapeutics, syphilitic chancres on rabbits were cured by administering less than the toxic dose (Launoy and Levaditi); but when attempts were made to apply the product to the human subject it was found to be useless. Thus, in the first place, it had to be injected into a vein like salvarsan or neosalvarsan, and then one or even two injections of a nearly toxic dose was insufficient to bring about as quickly as the arsenicals the disappearance of the primary symptoms of syphilis, so there was actually no advantage to be gained. Nevertheless, this is a beginning along an entirely new line.

CHAPTER X

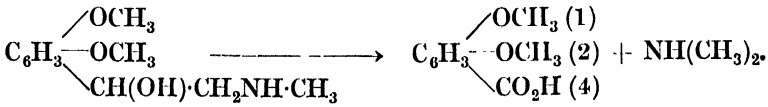
ADRENALINE

It was known in ancient times that in the bodies of animals there were certain glands that possessed toxic or curative properties. The chemical composition of the active principles responsible for the effects in question was later the subject of much investigation, but until quite recently [1] only one of them, namely, that from the suprarenal glands, had been separated in a definitely pure condition. This substance was isolated in 1901 by Takamine, and called **Adrenaline.** [2]

Constitution.—Adrenaline is derived from a homologue of catechol, in the side chain of which methylamino and secondary alcoholic groupings are present, thus :

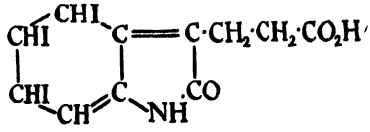


This structure was, in part, indicated by the results of oxidising the dimethyl ether. When the phenolic hydroxy groups were methylated, and so the aromatic radical protected, the product, dimethyl adrenaline, gave on oxidation veratric acid and dimethylamine (or trimethylamine if the amino group also were methylated), together with other products. As the carboxyl group represented the original side chain, the position of the latter was settled :



When adrenaline was fused with caustic potash the products were catechol, protocatechuic acid and methylamine. These results still left doubt as to the position of the alcoholic hydroxyl, but it was found that when the tri-benzenesulphonyl derivative was oxidised a ketone was produced, which was no longer optically active yet whose

[1] Kendall has now isolated the active principle of the thyroid gland, having worked up the extract from about three tons of the glands. The substance is a *cyclo*hexene derivative of the structure :



and has been named by its discoverer " Thyroxin ".

[2] With the name of Takamine should be associated those of Abel, von Fürth, Aldrich, and others.

molecule contained the same number of carbon atoms as that of the original alcohol; furthermore, when it was hydrolysed it gave adrenalone, which had already been obtained synthetically.

This constitution for adrenaline has been finally confirmed by the complete synthesis of the compound, as will be shown later.

Properties.—Adrenaline is a base and with acids forms salts which mostly do not crystallise readily (the commercial product is usually the hydrochloride). Being a dihydric phenol, it dissolves also in caustic soda.

The carbon atom to which the alcoholic hydroxyl is attached is asymmetric and so adrenaline is active towards polarised light. Natural adrenaline is lævo-rotatory; the specific rotation being $-53{\cdot}3°$ (Bertrand). The dextro-rotatory form has only $\frac{1}{12}$ to $\frac{1}{15}$ the activity of the lævo-isomeride in raising the blood-pressure, and can even render the latter less effective if injected beforehand in large enough doses. So it is that racemic adrenaline, as obtained synthetically, only half of which is the lævo variety, is somewhat less than half as active physiologically as the natural substance.

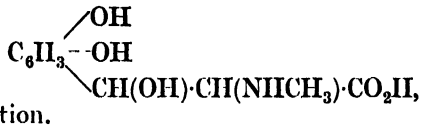
Several colour reactions are characteristic of adrenaline. These reactions are accounted for by the fact that its molecule contains two phenolic hydroxyl groups situated *ortho* to one another. Ferric chloride produces, in a dilute neutral or weakly acid solution, a green coloration changing to violet, or red on adding alkali. When a solution of adrenaline is exposed to the air it becomes brown. The oxidation of which this is the result is vastly accelerated by various oxidases, such as those found in the ink-bag of the cuttlefish and in certain toadstools; these oxidases rapidly bring about the formation of black pigments resembling the natural melanines. Maybe the oxidation of adrenaline in the animal organism takes place in a similar way.

Already in 1856 Vulpian had noted that the suprarenal glands contained a substance giving a green colour with ferric chloride and a red one with iodine. Several oxidising agents have since been used to characterise adrenaline by means of colour reactions. The best known are iodic acid and the persulphates. With iodic acid, and in presence of sulphanilic acid, a visible red coloration is obtained at a dilution of 1 : 5,000,000. With a 0·1% solution of sodium persulphate, the same coloration is obtained at the same dilution on gently warming. With a solution of sodium tungstate in phosphoric acid (Folin's reagent), adrenaline may be detected at a dilution of one in a million or even 1 : 3,000,000. Folin's reagent is prepared by dissolving 100 gm. sodium tungstate in 750 c.c. water, adding 80 c.c. phosphoric acid (85%), boiling the mixture, cooling it and diluting to 1 litre. The presence of $\frac{1}{100}$ mgm. of adrenaline is shown by this reagent.

Colorimetric methods based on these reactions have furnished a

means of estimating the amount of adrenaline in the glands of various animals. Thus, in cat's glands 0·125 gm. per kilo. has been found; in sheep's glands, 0·3 gm.; in those of the whale, 0·247 gm.

The origin of adrenaline is unknown. It is conjectured that it is derived from an amino acid, namely, dihydroxyphenylmethylserine,

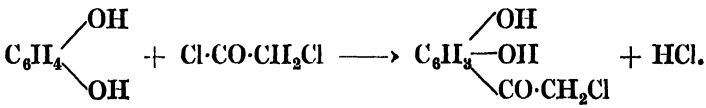


by decarboxylation.

This supposition is supported by the fact that the dihydroxyphenylalanine, $C_6H_3(OH)_2 \cdot CH_2 \cdot CH(NH_2) \cdot CO_2H$, has been isolated by Guggenheim from the pods of the broad-bean.

Synthesis of Adrenaline.—The synthesis of adrenaline has been attempted in several ways, the aim being both to establish definitely its constitution and to provide a basis for its manufacture, because the isolation of natural adrenaline is a costly process.

The classical method, which still seems to give the best results and is probably the only practicable one, was devised by Scholtz and patented by the Farbwerke vorm. Meister, Lucius and Brüning in 1904. The starting point is catechol, which is condensed with chloracetyl chloride. The latter substance is either employed as such, in which case a little zinc chloride is added to act as a catalyst, or the catechol is mixed with chloracetic acid and phosphorus oxychloride. When this mixture is heated on the water-bath, the chloracetyl chloride formed interacts with the catechol, setting free hydrogen chloride, and giving chloracetocatechol, thus:



To an alcoholic suspension of the product a cold concentrated aqueous solution of methylamine is added. At first all goes into solution because of the acidic nature of the phenolic hydroxyl groups, then soon methylaminoacetocatechol, which is sparingly soluble in water, is precipitated. It is collected and washed first with alcohol, then with ether:

$$C_6H_3(OH)_2 \cdot CO \cdot CH_2Cl + 2NH_2 \cdot CH_3 \longrightarrow C_6H_3(OH)_2 \cdot CO \cdot CH_2 \cdot NH \cdot CH_3.$$

The methylaminoacetocatechol so obtained is reduced either with aluminium amalgam or electrolytically. The ketone becomes converted into a secondary alcohol, racemic adrenaline being produced:

$$C_6H_3(OH)_2 \cdot CH(OH) \cdot CH_2NH \cdot CH_3.$$

This reduction is difficult to carry out because the amino group in an aminoketone is very readily split off.

The optical isomerides are separated by means of an optically active acid, *e.g.*, tartaric acid. The mixture of tartrates obtained from the

racemic base is treated with methyl alcohol, *d*-adrenaline *d*-tartrate dissolves, and the less soluble *l*-adrenaline *d*-tartrate remains behind. The latter is converted into the hydrochloride and then forms commercial synthetic *Adrenalin*.

By employing the above process as it stands half of the product would be lost, and synthetic adrenaline is not cheaply made. This disadvantage can be overcome, however, by heating the by-product, *d*-adrenaline, with hydrochloric acid, and so converting it into the racemic mixture. The separation may now again be effected, the dextro variety produced this time again racemised and so on until all has been converted into the lævo form. Only the latter is of therapeutic value.

Attempts have been made to synthesise adrenaline in several other ways, thus :

(1) By methylating *dihydroxyphenylethanolamine*.[1]—This base is obtained by reducing aminoacetocatechol, which itself is prepared either :

(*a*) By condensing chloracetocatechol with ammonia, or better, with hexamethylenetetramine :

$$C_6H_3(OH)_2\cdot CO\cdot CH_2\cdot NH_2;$$

or, (*b*) by hydrolysing with hydrochloric acid the product of condensing together hippuryl chloride and veratrole :

$$C_6H_4(OCH_3)_2 + Cl\cdot CO\cdot CH_2\cdot NH\cdot CO\cdot C_6H_5$$
$$\longrightarrow C_6H_3(OCH_3)_2\cdot CO\cdot CH_2\cdot NH\cdot CO\cdot C_6H_5$$
$$\longrightarrow C_6H_3(OH)_2\cdot CO\cdot CH_2\cdot NH_2.$$

(2) Barger made several attempts to produce adrenaline, starting from piperonal or methyl vanillin (veratraldehyde), but without much success. When these aldehydes are treated with magnesium methyl iodide secondary alcohols are formed, which may be dehydrated to the corresponding ethylene derivatives. These unite with bromine to form dibromides, and the dibromides are converted by the action of water into bromhydrins ; these products are then treated with methylamine :

$$\left.\begin{matrix}CH_3O\\CH_3O\end{matrix}\right\rangle C_6H_3\cdot CHO + CH_3\cdot Mg\cdot I \longrightarrow (CH_3O)_2 : C_6H_3\cdot CH(OH)\cdot CH_3$$
$$\longrightarrow (CH_3O)_2 : C_6H_3\cdot CH : CH_2 \longrightarrow (CH_3O)_2 : C_6H_3\cdot CHBr\cdot CH_2Br$$
$$\longrightarrow (CH_3O)_2 : C_6H_3\cdot CH(OH)\cdot CH_2Br$$
$$\longrightarrow (CH_3O)_2 : C_6H_3\cdot CH(OH)\cdot CH_2\cdot NH\cdot CH_3.$$

The reactions to this stage give not unsatisfactory yields. It is the final stage, the conversion of the ether into the parent phenol, that is troublesome. The elimination of the two methyl groups is always accompanied by that of the amino group. Böttcher thought that he had overcome this difficulty in the case of the piperonal derivative by

[1] This base is on the market with the trade name *Arterenol*.

removing the methylene group before treating the bromhydrin with methylamine. This he effected by treating the intermediate product first with phosphorus pentachloride, then with water, but the substance obtained, which should have been dihydroxyphenylethylene bromhydrin, gave with methylamine a product which did not crystallise, and Böttcher could not definitely prove that adrenaline was present. Mannich took up the question later and showed that it is doubtful whether the bromine atom is replaced by methylamine, as an ether is formed which may give the two isomeric amino alcohols together. Thus, for example, from dimethoxyphenylchloroethanol, $(CH_3O)_2 : C_6H_3 \cdot CH(OH) \cdot CH_2Cl$, the two isomerides,

dimethyl adrenaline $-(CH_3O)_2 : C_6H_3 \cdot CH(OH) \cdot CH_2 \cdot NH \cdot CH_3$,
and dimethyl*iso* adrenaline $-(CH_3O)_2 : C_6H_3 \cdot CH(NH \cdot CH_3) \cdot CH_2OH$,

may be obtained. These two bases differ in the way they behave towards hydriodic acid. The first, unfortunately, is not converted into adrenaline, the methylamine group being detached, but the second gives an almost quantitative yield of the demethylated product, *iso*adrenaline, which, however, has none of the physiological properties of its isomeride.

The preparation of various homologues and analogues of adrenaline has been undertaken; thus, *Arterenol* is adrenaline without the methyl group (Bayer). Dihydroxyphenyl-propanolamine, *i.e.*, homoarterenol, $C_6H_3(OH)_2 \cdot CH(OH) \cdot CH(CH_3) \cdot NH_2$, has also been made and found to resemble both adrenaline and ephedrine; it is obtained by reducing the corresponding ketone with hydrogen in presence of platinum or palladium. The ketone is prepared by a method which is noteworthy, because it enables an aminoacyl group to be introduced into a molecule. Thus, in the case in point, phthalimidopropionyl chloride is condensed with veratrol:

$$C_6H_4(OCH_3)_2 + Cl \cdot CO \cdot CH(CH_3) \cdot N\begin{matrix}CO\\CO\end{matrix}C_6H_4$$
$$\longrightarrow C_6H_3(OCH_3)_2 \cdot CO \cdot CH(CH_3) \cdot N\begin{matrix}CO\\CO\end{matrix}C_6H_4.$$

The product is hydrolysed (and demethylated), giving

$$C_6H_3(OH)_2 \cdot CO \cdot CH(NH_2) \cdot CH_3,$$

which is then reduced.

This new base has been separated into the two optically active isomerides and the lævo form again found to be more active physiologically than the dextro variety (Tiffeneau); indeed, the difference here is more marked than that between the two adrenalines, as the lævo dihydroxyphenylpropanolamine is at least thirty times more

active than the dextrorotatory form. These substances appear to be less toxic than adrenaline.

Whilst trying to prepare a similar compound, Mannich obtained, exclusively, the α-amino isomeride,

$$C_6H_3(OH)_2 \cdot CH(NH \cdot CH_3) \cdot CH(OH) \cdot CH_3,$$

which was examined by Kobert and shown to have none of the properties characteristic of adrenaline.

Various substances closely related to adrenaline are commercial products, namely, *Homorenon*, ethylaminoacetocatechol, ω-ethylamino-3 : 4-dihydroxyacetophenone, $C_6H_3(OH)_2 \cdot CO \cdot CH_2NH \cdot C_2H_5$; *Epinine*, dihydroxyphenylethylmethylamine, $C_6H_3(OH)_2 \cdot CH_2 \cdot CH_2 \cdot NH \cdot CH_3$; and, of course, *Arterenol*, $C_6H_3(OH)_2 \cdot CH(OH) \cdot CH_2 \cdot NH_2$. The first two of these are not as active physiologically as racemic adrenaline, whilst the last is almost as active as adrenaline itself. Epinine is obtained by reducing the oxime of homoveratraldehyde and demethylating the product. Homoveratraldehyde is produced by the action of ozone on eugenol methyl ether.

Commercial adrenaline is not entirely the synthetic product; some is obtained by extracting suprarenal glands. The extraction is not a simple matter, because the product is so easily oxidised. Bertrand's method is the one to be recommended. Essentially it consists in using a solution of oxalic acid in alcohol as the extracting agent. Albumins remain undissolved, whilst the base goes into solution as the oxalate. The extract is evaporated to dryness and the residue treated with petroleum ether to remove fats and lipoids. It is then dissolved in water and the oxalic acid precipitated by adding the exactly necessary amount of lead acetate. The solution is concentrated *in vacuo*, and the adrenaline finally precipitated by ammonia. In this way Bertrand obtained 1·25 gm. adrenaline from 1 kilo. of the suprarenal glands of the horse.

Physiological Action of Adrenaline.—The physiological function of adrenaline is still obscure. At one time it was considered to be very important, but nowadays, chiefly as a result of the work of Stewart in America and of Gley and his pupils in France, the tendency is to regard it more as a waste product.

When adrenaline is injected into the blood it acts on the terminations of the sympathetic nervous system; for this reason it was thought to be the chief regulator of the functions dependent on this system, and that consequently there must be a physiological adrenalinæmia without which the organs would not fulfil their functions properly.

Cannon and de la Paz based on the results of their researches a very attractive theory, which caused quite a stir at the time. According to these investigators, strong emotional changes bring about an

increased secretion of adrenaline, and the resulting hyperadrenalinæmia is responsible for all the physical manifestations accompanying the emotions (ashen appearance, hair standing on end, glycæmia, relaxed sphincters, etc.).

Certainly these phenomena result from an injection of adrenaline into the blood, but, in the first place, removal of the suprarenals or ligature of the suprarenal veins does not prevent their appearance; secondly, to produce them artificially needs much more adrenaline than is normally present in the blood, even at times when the quantity is at a maximum; lastly, adrenaline is never found in the blood of the *vena cava* above the sub-hepatic veins, nor in the blood of the heart, so how the latter can be affected by the adrenaline is not very clear. However, it is no less premature to conclude that adrenaline is a quite useless product, because as yet nothing is known of the influence glands can exert on one another, or of the indirect action of hormones, or, in general, of the action of the constituent principles (not to say the active principles) of the organs of internal secretion on the chemical systems in the organism. The fact that a poison like adrenaline disappears from the blood during its passage through the liver does not necessarily indicate that it has done its work; it may be converted into a substance of which we know, at present, neither the chemical constitution nor the physiological properties.

The footway here is somewhat insecure and we shall gain nothing by going further. Let us turn back to the pharmacological properties possessed, in fact, not only by adrenaline, but also, in varying degrees, by a series of derivatives of phenylethylamine, or, to put it still more generally, by the β-substituted ethylamines.

The characteristic effect produced by adrenaline is that which it has on the ends of the sympathetic nervous system. Its therapeutic value, and perhaps also its physiological importance, are intimately connected with the functions regulated by these nerves. Excitation of the sympathetic system causes constriction of the blood-vessels; administration of adrenaline has the same effect. Constriction takes place in every artery, except the coronary artery, which supplies the muscular walls of the heart itself, and perhaps also the arteries of the lungs. The effect is most marked on the capillaries: when adrenaline is put in contact with the skin no action takes place, but when a mucous surface is wet with the solution, it is rapidly blanched as a result of the anæmia produced by the shrinkage of the arterioles. The use of adrenaline in therapeutics depends on this vaso-constrictive action; certain kinds of hæmorrhage, as, for example, in the eye, may be arrested by direct application of the drug, and it is widely employed in association with some local anæsthetics. If the latter be injected without adrenaline they are rapidly carried away by the blood stream because they are so diffusible, but if adrenaline be injected at

the same time, the resulting vaso-constriction holds up the local anæsthetic so that it can exert its action within a definite area.

Adrenaline exerts such a vaso-constrictive action on the walls of the alimentary canal that practically nothing taken by the mouth can be absorbed in the stomach or intestine. The action is so marked that the administration of adrenaline has actually been recommended as a means of preventing a poison from taking effect.

Adrenaline causes a very marked rise in the arterial blood pressure. This is brought about by the constriction of the arteries and also, in part, by the greater vigour of the heart's contractions, for which the excitation of the sympathetic nerve endings in this muscle is responsible. Adrenaline is a powerful analeptic and is, in fact, administered to sufferers from various infectious diseases (influenza, typhoid), partly because of this action on the heart, partly also because of its antitoxic action. It is chiefly employed in serious cases of cardiac syncope.

When adrenaline is dropped into the eye of an animal, preferably an excised eye, the pupil becomes dilated. This effect is that of a stimulation of the sympathetic nerve ending associated with the muscle that controls the iris. The mydriatic action here must not be confused with that of the alkaloids of the Solanaceæ; atropine also causes dilatation of the pupil, but this is not due to excitation of the sympathetic nerves, but to paralysis of the motor nerves, which have an action on the muscle of the iris contrary to that of the sympathetic.

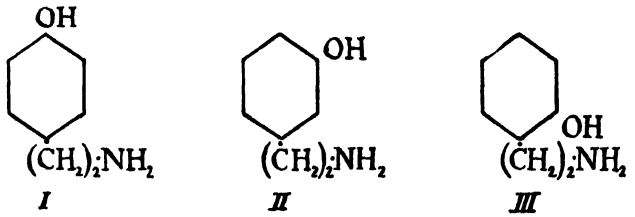
Adrenaline has an action on various internal organs; it arrests the peristaltic movements of the intestines, but, on the other hand, it stimulates the contractions of the uterus, particularly in certain animals.

If the amount of adrenaline in the blood be increased the carbohydrates stored in the liver are set free, and the concentration of glucose in the blood may rise to such a point that it passes through the kidneys and appears in the urine. This effect is known as adrenaline glycosuria.

To the action which results in all these effects at the various endings of the sympathetic nervous system Barger and Dale have given the name "sympathomimetic action." To which part of the molecular structure of adrenaline is this characteristic property due? This is the question, typical of those met with in this field, that Barger and Dale have tackled in masterly fashion.

Relation between the Chemical Constitution of Adrenaline and its Sympathomimetic Action.—First of all, adrenaline is an amine. Aliphatic amines act to some extent on the arterial blood pressure and the activity, although feeble, increases with rise in molecular weight. It is quite marked with *normal* and *iso*-amyl-amine and rises till the C_7 amine is reached. When an aromatic nucleus is attached to the

aliphatic chain, the action is augmented to a surprising degree, but, if the sympathomimetic action is to be really noteworthy, there must be *one, and only one, carbon atom between that to which the amino group is attached and the aromatic nucleus.* Thus, benzylamine, $C_6H_5 \cdot CH_2 \cdot NH_2$, and phenylpropylamine, $C_6H_5 \cdot CH_2 \cdot CH_2 \cdot CH_2 \cdot NH_2$, are both much less active than β-phenylethylamine, $C_6H_5 \cdot CH_2 \cdot CH_2 \cdot NH_2$, in which the nucleus is separated from the carbon atom carrying the amino group by only *one* other carbon atom. The introduction of a hydroxy group *para* or *meta* to the side chain in phenylethylamine brings about a remarkable increase in sympathomimetic activity, but if the substitution take place in the *ortho* position, it has a deleterious effect.



I and *II* show great activity.
III is almost inactive.

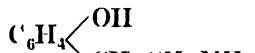
As we shall see later, *p*-hydroxyphenylethylamine is a naturally occurring base, known as tyramine, and has a therapeutic action similar, qualitatively at any rate, to that of adrenaline. The introduction of a second hydroxyl group still further increases the activity, provided that it takes up a position *ortho* to the first. Tiffeneau established this point in his researches on the lower homologues of adrenaline.

Dihydroxyphenylethylamine, or *Epinine*, already has a physiological action comparable with that of *Arterenol*, although less intense (about $\frac{1}{7}$). So the alcoholic hydroxyl group is not an essential part of the molecule[1]; its chief function is that of conferring asymmetry, so that two isomerides are possible, the activities of which may differ considerably. Aminoacetocatechol (adrenalone) and arterenol, the corresponding alcohol (racemic form), are almost equally active; the sympathomimetic activity is therefore the same for the ketonic as for the secondary alcoholic group.

Furthermore, in compounds analogous to adrenaline but possessing no phenolic groups, the alcoholic hydroxyl has no effect, the unsubstituted amines and the aminoalcohols derived from them have a like sympathomimetic action. Thus the base, $C_6H_5 \cdot CH_2 \cdot CH_2 \cdot NH_2$, has the same activity as the alcohol, $C_6H_5 \cdot CH(OH) \cdot CH_2NH_2$.

[1] Nevertheless, between racemic dihydroxyphenylhydroxyethylmethylamine (adrenaline) and dihydroxyphenylethylmethylamine, there is a much greater difference in activity.

A methylamino group is present in the adrenaline molecule: the methylamino group has the same effect as the amino group itself, but it is not so potent. If the substitution be continued, the tertiary base is obtained; in a compound of this kind the sympathomimetic action is very feeble. Thus tyramine is very active, but hordenine, the corresponding dimethylamine derivative, is a drug of which several grams per day may be taken.



Tyramine.

Hordenine.

Further interaction of a tertiary base with methyl iodide produces a quaternary ammonium iodide and a complete change in physiological properties results. Sympathomimetic activity is suppressed entirely. The iodide certainly brings about an increase in the blood pressure, but this is not due to direct excitation of the terminations of the sympathetic system, but to an action similar to that of nicotine.

Tiffeneau has studied the physiological activity of the lower homologues of hordenine and epinine, *i.e.*, of the compounds in which the aromatic nucleus and the amino group are attached to the same carbon atom. The first of these compounds is about as active as hordenine itself, that is to say, it is very feeble; the second has about a hundredth the activity of adrenaline. Thus the rule that only compounds with the aromatic radical in the β-position to the amino group are active is again substantiated.

There are various ways of measuring the physiological action of these compounds. To make sure that a product possesses sympathomimetic properties it must be shown to have an action at all the terminations of the sympathetic system. One of the simplest tests to carry out is that on the pupil of the eye. The two eyes of a frog are excised, transferred to conical test-glasses, covered with Ringer's solution, and exposed to a bright light so as to make the pupils contract as far as possible. A solution of the product under examination is run into one of the containers. The intensity of the illumination is kept constant while the diameters of the pupils are compared. In another method, the technique of which is not difficult but which also gives only qualitative results, small portions of fresh artery are used. These are fixed between clamps and treated with a solution of, *e.g.*, adrenaline, and the contraction registered graphically on a clockwork-driven drum. Cannon adopted this method in many of his investigations on the relationship between adrenaline and the emotions.

There are two chief methods of estimating quantitatively the physiological activity of the compounds in this group. In the first

of these the action on the arterial blood pressure is measured (pressor effect).

Barger and Dale usually employed cats for the purpose. Their technique is very complicated. To ensure that the effects were due solely to excitation of the sympathetic, the central nervous system, which might affect indirectly the sympathetic system by a stimulation of the suprarenal glands, had to be eliminated. This was effected by removing the brain or beheading the animal; in any case, artificial respiration was necessary to keep the heart beating. The method is unsatisfactory also, because a simple effect is not measured, since the rise in blood pressure is caused both by vaso-constrictive action and by that on the heart. Tiffeneau [1] has shown that accurate and comparative results may be obtained if the cat be simply chloralosed under the influence of atropine.

The second method is relatively simple. It also gives quantitative results and has been employed by Läwen and Trendelenburg for estimating adrenaline in very dilute solutions. An artificial circulation is set up in the lower limbs of a frog, the rate of which is measured, keeping the head of liquid constant, by the number of drops per minute leaving the system. The vaso-constrictive action is measured by the change in the rate of circulation when the adrenaline is added. The frog is decapitated and an iron wire pushed down its spine so as to destroy all the spinal cord and thus eliminate all the central nervous system. The abdominal wall is slit at each side so that it hangs down between the two hind legs; attached to this membrane is the anterior abdominal vein by which the solution is to emerge. The renal portal veins are ligatured, and then all the organs in the abdominal cavity are removed so that the dorsal aorta is exposed. Finally, both fore limbs and the whole thorax are cut away. The preparation is fixed up and a small cannula inserted into the aorta, so that Ringer's solution, contained in an aspirator bottle, may be introduced from a height of about 10 c.m.; circulation begins, and the anterior abdominal vein becomes swollen; when it is in this condition an incision is made and a small glass cannula inserted so that the liquid may drip out. Equilibrium is soon attained and the rate at which the drops fall becomes steady, the number of drops and the time, in minutes, being recorded on the revolving drum. The solution of the product to be tested is now added to the Ringer's solution by injecting it through a Pravaz syringe into the rubber tube connecting the aspirator bottle with the cannula in the aorta. The decrease in the number of drops per minute falling from the vein gives an exact measure of the vaso-constriction produced.

There is yet another method which appears to give quite good results. This consists in measuring the contractions of an isolated

[1] *Comptes rendus*, 1915, *161*, 36.

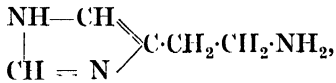
uterus. It has been chiefly employed in investigating compounds analogous to adrenaline. Other sympathomimetic substances also augment the intensity of contraction.

OTHER NATURAL PRODUCTS RESEMBLING ADRENALINE

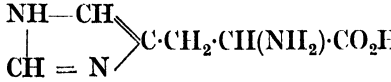
The vaso-constrictive action of an extract of ergot, which is very similar to that of adrenaline, has been known for a long time. Analogous vasomotor activity is displayed also by certain bases produced by the putrefactive decomposition of proteins. Tyramine, *p*-hydroxyphenylethylamine, $OH \cdot C_6H_4 \cdot CH_2 \cdot CH_2 \cdot NH_2$, is typical of this class; it doubtless originates from tyrosine, the corresponding amino acid. Putrefactive bacteria, and the ergot fungus, can, in fact, bring about the decarboxylation of the amino acids from which proteins are built up.

Amino acids of the aliphatic series are similarly decomposed—lysine, for example, undergoes decarboxylation, giving cadaverine.

But the bases derived from amino acids containing cyclic nuclei are the most interesting from a physiological point of view, because of the more or less marked activity towards the terminations of the sympathetic nervous system displayed by some of them. When their formulæ are examined it is seen that all consist of a nucleus with a side chain, and that there is an amino group attached to the side chain in the β-position. **β-Phenylethylamine,** $C_6H_5 \cdot CH_2 \cdot CH \cdot {}_2NH_2$, is the simplest member of the group. This compound was discovered in putrid meat by Nencki (1876). It may be prepared by reducing benzyl cyanide. **β-Iminazolyl-ethylamine**[1] (Trade names: *Ergamine*, *Histamine*),

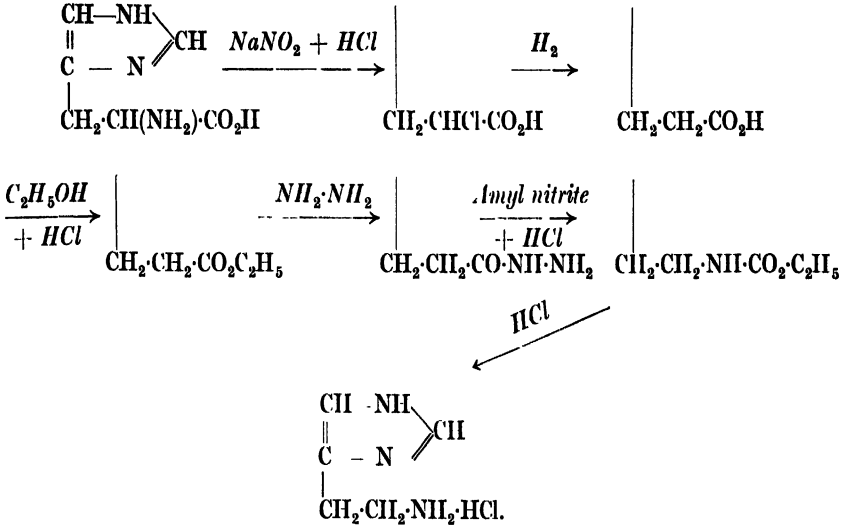


has been isolated from the ergot of rye and found to have the property, characteristic of the drug, of producing uterine contraction. For this reason it is considered to be the most important active principle in ergot. Industrially it has been obtained by bacterial putrefaction of histidine,

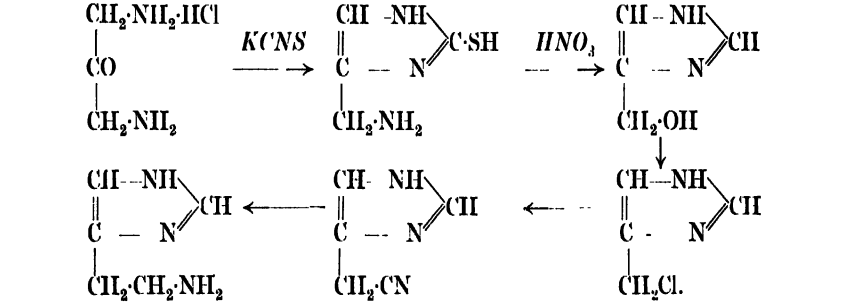


Several kinds of bacteria may bring about the decarboxylation. The most active, namely, *Bacillus aminophilus intestinalis*, has been isolated by M. Berthelot, of the Pasteur Institute. The decarboxylation is carried out in very dilute solution, viz., 1·5 parts of histidine per 1,000. Histidine may also be decarboxylated by chemical means (Windaus), the reactions involved being as follows:

[1] 4-β-Aminoethylglyoxaline. Glyoxaline is the orthodox English name.—*Tr.*



The synthesis of histamine has been effected by Pyman. Diaminoacetone hydrochloride is heated with potassium thiocyanate, and the product oxidised with nitric acid; simultaneously the nitrous acid formed in the reaction acts on the amino to give a hydroxyl group; this is then replaced first by chlorine, then the cyano compound is formed, finally this is reduced.

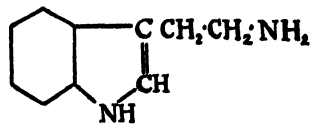


The most characteristic property of histamine is its action on the uterus in various animals. A surprisingly dilute solution will cause contraction, thus, a marked effect is obtained with a 1 in 25,000,000 solution and even a 1 in 250,000,000 solution has a perceptible action.

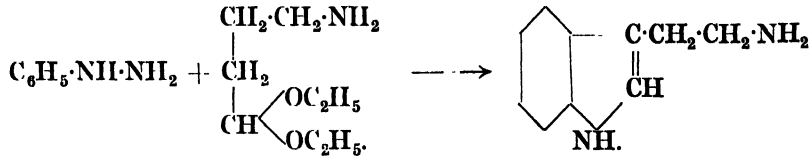
β-Iminazolylethylamine is a violent poison, the action of which causes symptoms exactly resembling those of anaphylactic shock, so much so, indeed, that it has been suggested that anaphylactic shock involves the formation of similar poisons by decomposition of histidine or related amino acids.[1]

[1] Histamine does not belong to the sympathomimetic group, as Barger has defined the term, but it has been discussed here, first of all, because it is associated with hydroxyphenylethylamine in ergot; secondly, because it is a β-substituted ethylamine; and, lastly, because it is noteworthy in being much more active than either its lower or its higher homologues.

Indolethylamine,



possesses marked sympathomimetic activity. This compound results from the putrefactive decomposition of tryptophan. It has been synthesised (Ewins and Laidlaw) by condensing γ-aminobutyrylacetal with phenylhydrazine in the presence of zinc chloride,



Of all these compounds, the one manufactured on the largest scale is **tyramine,** hydroxyphenylethylamine, $OH{\cdot}C_6H_4{\cdot}CH_2{\cdot}CH_2{\cdot}NH_2$. This substance was isolated from putrid meat by Gautier and from ergot by Barger. It is used in much the same way as adrenaline, but does not act so vigorously. Synthetically it may be obtained in the following way: Benzyl chloride is treated with potassium cyanide to give phenylacetonitrile (benzyl cyanide), which on nitration yields *p*-nitrophenylacetonitrile. The nitro compound is reduced and the amino group diazotised and replaced by hydroxyl. *p*-Hydroxyphenylacetonitrile, thus obtained, is reduced with sodium and alcohol and yields the desired amine.

$$C_6H_5{\cdot}CH_2{\cdot}CN \longrightarrow NO_2{\cdot}C_6H_4{\cdot}CH_2{\cdot}CN \longrightarrow NH_2{\cdot}C_6H_4{\cdot}CH_2CN$$

$$OH{\cdot}C_6H_4{\cdot}CH_2{\cdot}CH_2{\cdot}NH_2 \longleftarrow OH{\cdot}C_6H_4{\cdot}CH_2{\cdot}CN.$$

This process is employed on the manufacturing scale, the overall yield being quite good.

Tyramine has been synthesised by other methods, but, with the exception of that due to Rosenmund, they are of no great interest. Rosenmund condensed anisaldehyde with nitromethane by the action of sodium ethoxide in alcoholic solution, thus obtaining *p*-methoxy-ω-nitro-styrene,

$$CH_3O{\cdot}C_6H_4{\cdot}CH:CH{\cdot}NO_2,$$

which on reduction with zinc and acetic acid gave methoxyphenylacetaldoxime,

$$CH_3O\ C_6H_4\ CH_2{\cdot}CH:NOH;$$

this, reduced further with sodium amalgam, gave the methyl ether of the desired amine, from which the methyl group was removed by heating with hydriodic acid.

The dimethyl derivative of tyramine, **Hordenine,**

$$OH{\cdot}C_6H_4{\cdot}CH_2{\cdot}CH_2{\cdot}N(CH_3)_2,$$

was found by Léger to be present in malt extract. The therapeutic

value of malt extract in treating diarrhœa depends on this constituent. The compound is now employed as a drug in the form of its sulphate. Camus and, later, Barger, have shown that it has a sympathomimetic action, weak, certainly, but still strong enough for the compound to have a soothing effect on intestinal peristalsis and to act as a cardiac tonic; perhaps, also, it is quite strongly antiseptic, being a phenol derivative. It is an innocuous substance, only weakly toxic, and several grams a day may be taken. Hordenine was first synthesised by Barger, the starting point being phenylethyl alcohol, a commercial product. This, treated with thionyl chloride, gave phenylethyl chloride, which, by interaction with dimethylamine, formed dimethylaminoethylbenzene. This was nitrated, the nitro group entering the *para* position, and the nitro compound converted into the phenol by reduction and diazotisation in the usual way.

Hordenine may also be obtained from methoxyphenylethylamine by methylating the amino group with methyl chloride, and then demethylating the phenolic group, or, as another alternative, by reducing *p*-hydroxy-phenyl dimethylaminomethyl ketone,

$$HO{\cdot}C_6H_4{\cdot}CO{\cdot}CH_2{\cdot}N(CH_3)_2.$$

There remains to mention another natural base, **Ephedrine,** which is obtained from *Ephedra vulgaris*, a plant indigenous to Japan. This compound is interesting, because it also is a derivative of phenylethylamine, having, so it seems, the structure :

$$C_6H_5{\cdot}CH(OH){\cdot}\overset{\displaystyle NH{\cdot}CH_3}{\overset{|}{C}}H{\cdot}CH_3.$$

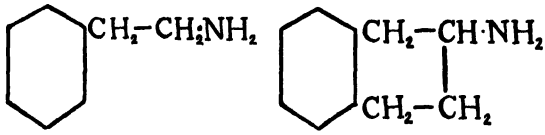
It is optically active and has a pronounced mydriatic action. Fourneau, and later Nagai, obtained the racemic form synthetically. Several structural isomerides have been similarly prepared. Quite recently Spath has succeeded in separating the optically active variety, identical with the natural product, from a synthetic preparation.

The formula above shows ephedrine to be an amine with the (methyl) amino group in the β-position relative to a phenyl radical. We might expect, then, from theoretical considerations, that it should possess sympathomimetic activity, and that this is so is shown by the effect the compound has on the pupil of the eye. Nagai compared the synthetic racemic form with the natural product and obtained results which led him to state that the two substances were equally active physiologically. This is very unlikely, and the investigation should be repeated. (See note at end of chapter.)

SYNTHETIC PRODUCTS RESEMBLING ADRENALINE

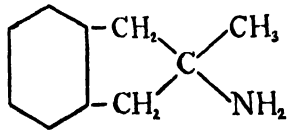
Besides the above natural bases there are a number of physiologically active synthetic compounds of great theoretical interest from the pharmacological point of view.

β-Naphthylamine resembles aniline closely in chemical properties; the resemblance also extends to the physiological properties. But when β-naphthylamine is reduced by means of sodium and alcohol the molecule undergoes a profound change; the ring to which the amino group is attached is hydrogenated and becomes aliphatic (*cyclo*-paraffinic) instead of aromatic in character. Thus a weak base, like aniline in behaviour, is converted into a strong (aliphatic) base, the salts of which are neutral in reaction. The product is no longer a naphthalene derivative, it must be regarded as a benzene derivative with an amino group attached to a side chain; and in the β-position, for if the formulæ of β-phenylethylamine and *ac*-**tetrahydronaphthylamine** be compared, one sees that they are not widely different.



A similar radical change takes place in the physiological properties; the behaviour of tetrahydronaphthylamine is that characteristic of the phenylethylamine group. It has a marked action on the sympathetic system. The symptoms described above (vaso-constriction, standing of the hair on end, etc.) appear when it is injected into an animal. This compound, moreover, is exceptional in bringing about a rise in body temperature; very few substances have this property. Dilatation of the pupil of the eye is another characteristic property of the substance.

Tetrahydronaphthylamine is, as we have just seen, a β-phenylethylamine. It is also a phenylpropylamine, as the amino group is separated from the nucleus by two carbon atoms, counting one way, and three carbon atoms, counting the other. Let us now turn to an amine which is, so to speak, doubly a β-phenylethylamine. Such is the **methylaminohydrindene** synthesised by von Braun (1917).

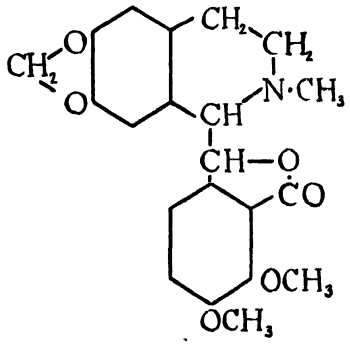


This is intensely active, as its constitutional formula would suggest, but, curiously enough, the activity is not increased by the entry of a hydroxyl group into the aromatic nucleus, although substitution of this kind augments the activity of all the simpler compounds already discussed. Von Braun supposes that the base is already so active that the introduction of a hydroxyl group cannot strengthen it (?).

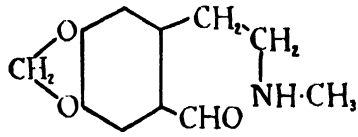
To round off our study of vaso-constrictive medicaments we must

draw attention to the properties of certain alkaloids, as these illustrate very aptly the pharmaceutical laws set forth above.

Hydrastine is an alkaloid found in *Hydrastis canadensis*, having the structure:



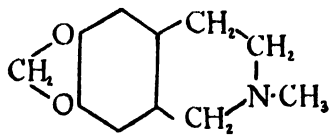
Hydrastine.



Hydrastinine.

By hydrolysis and oxidation it yields hydrastinine and opianic acid. Now the amino group in hydrastine is in the β-position to a phenyl group, as the formula shows, but it is a tertiary group, and so has practically no sympathomimetic action; this, in any case, would be checked by the opianic acid radical and the methylene group in place of the hydrogen of the phenolic hydroxyl groups. Hydrastinine, however, is different. Here the β-amino group is secondary,[1] and the compound possesses marked vaso-constrictive activity and, indeed, all the other properties which we now expect to find in this series of compounds.

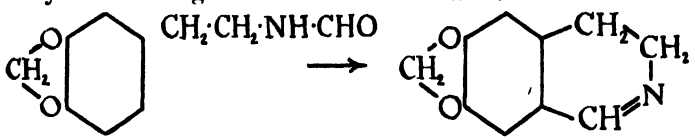
Hydrastinine treated with alkali undergoes auto-oxidation, giving (di-)hydrohydrastinine and oxyhydrastinine.



Hydrohydrastinine.

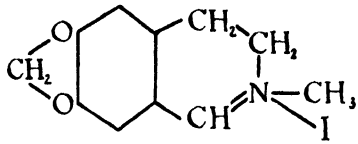
In hydrohydrastinine the amino group is again tertiary and the sympathomimetic activity is almost completely lost.

Hydrastinine has been synthesised by Decker, formyl-homopiperonylamine being caused to condense thus:



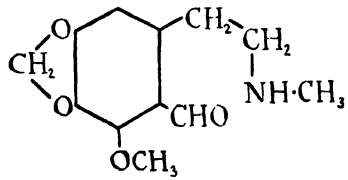
The dihydro-*iso*-quinoline obtained, treated with methyl iodide, yields hydrastinine hydriodide:

[1] For a discussion of these formulæ see, *e.g.*, Meyer and Jacobson, *Lehrbuch der organ. Chemie*, II, iii, p. 1030.—*Tr.*



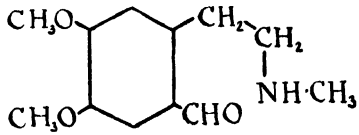
Hydrastinine hydriodide.[1]

Narcotine, one of the alkaloids in opium, is structurally very similar to hydrastine. Like the latter, it can be hydrolysed to opianic acid and hydrocotarnine, which, oxidised, gives cotarnine. Cotarnine is a styptic and is used chiefly in cases of uterine hæmorrhage, being marketed under the name *Stypticine.*



Cotarnine.

Another opium alkaloid, **Laudanosine,** breaks down like narcotine when oxidised, giving veratric aldehyde and a base very similar to hydrastinine. This compound, the trade name of which is *Lodal,* is related to hydrastinine as veratric aldehyde is to piperonal.



Lodal.

Narceine, another alkaloid closely related to cotarnine but having a tertiary amino group, has no hæmostatic properties.

The phenyl nucleus is not the only one that can serve as a foundation for compounds with sympathomimetic properties; if a β-aminoethyl group be introduced into other radicals, the products are likewise active. Madinaveitia has recently published the results of some notable work in the naphthalene series. He shows that the replacement of the phenyl by the naphthyl group augments the vaso-constrictive action in a very striking way. Basing his observations on the effect produced by 0·01 gm. of phenylmethoxyethylmethylamine,

$$C_6H_5{\cdot}CH(OCH_3){\cdot}CH_2{\cdot}NH{\cdot}CH_3,$$

he finds that the corresponding naphthyl derivative is much more active, even with a dose of 0·0005 gm.

$$C_{10}H_7{\cdot}CH(OCH_3){\cdot}CH_2{\cdot}NH{\cdot}CH_3\ (\beta)$$

This opens up a most interesting new field.

[1] See footnote on preceding page.

The introduction of a hydroxyl group into the naphthalene nucleus brings about a further increase in activity.

Our choice of the subject for this chapter from among so many possibilities was dictated by the fact that in no other class of compounds employed in pharmacy are the relations between physiological action and chemical constitution so well displayed as in the sympathomimetic group. In fact, we consider the work that has started with, or centred round, adrenaline to be among the most admirable of any done in pharmaceutical chemistry. Syntheses that have no other aim than to complete a series, or make new compounds, are of indifferent interest, but if, by their aid, we may unravel the complicated mechanism of physiological behaviour, they become really valuable.

Note on Ephedrine:—Fourneau and Kanao have recently (*Bull. Soc. Chim. de France*, 1924, **35**, 611), published a critical survey of earlier work on the synthesis of ephedrine and *pseudo*-ephedrine.

CHAPTER XI

PHOSPHATIDES

THE name **phosphatides** is given to substances containing nitrogen and phosphorus, derived from esters formed by certain polyhydric alcohols with the higher fatty acids.

The typical phosphatide is **Lecithin.**

Lecithin is an ester-like combination of choline with a glycerophosphoric acid, in which the two (glycerol) hydroxyl groups not taken up by the phosphoric acid are esterfied by fatty acids of high molecular weight (one acid is always unsaturated), mainly palmitic and oleic. To put the matter concisely, we may say that lecithin is simply a fat in which one fatty acid radical is replaced by that of choline phosphoric ester. It may then be formulated in this way:—

$CH_2 \cdot O-$ palmitic radical
|
$CH \cdot O$ -oleic radical
|
$CH_2 \cdot O$ $PO(OH) \cdot O \cdot CH_2 \cdot CH_2 \cdot N(CH_3)_3$.

Although our knowledge is by no means complete, lecithin has been the subject of more investigation than any other phosphatide, so we will discuss it in greater detail later.

Other phospho-lipoid complexes, besides lecithin, have been isolated from, *e.g.*, yolk of egg, various animal organs, or vegetable tissues. Some of these cannot satisfactorily be regarded as definite chemical individuals, and their existence as proximate constituents of the cell must only be accepted with reserve. Others, on the contrary, such as kephalin and sphyngomyelin, seem to be quite definite compounds.

Lastly, a place to itself must be kept for de-oleolecithin, *Lysocithin* (Preston Kyes's cobra-lecithide), which is a crystalline substance obtained by partially hydrolysing lecithin by means of cobra venom (Delezenne and Ledebt; Delezenne and Fourneau, 1914). Lysocithin has certainly not yet been found in the normal tissues or fluids of the organism, but it, more than any other in the phosphatide group, is a definite chemical individual, and for that reason the results of investigating it have aided in clearing up certain obscure points in the chemistry of lecithin. Moreover, its pronounced physiological properties, particularly its powerful hæmolytic action, and the part it is considered to play in certain of the processes involved in poisoning

by snake venoms, make it a most interesting substance, and so we are justified in referring to it in some detail.

Let us now review briefly the phosphatides of the first category.

Cuorin.—In 1907, Erlandsen, while investigating the phosphatides of the muscles of the heart, separated from an ethereal extract two fractions, one of which was composed of lecithin with, consequently, a nitrogen : phosphorus ratio of 1 : 1, whilst in the other the ratio was 1 : 2; it was therefore an *aminodiphosphatide* and was termed Cuorin (Ital. *cuore* = heart). When hydrolysed, cuorin is broken up into three molecules of fatty acid, instead of two as in the case of lecithin, one of glycerophosphoric acid, and one of a base which is not choline.

Cuorin may be a homogeneous substance, or it may be a mixture. In this connection it is interesting to note that several experienced investigators have isolated phosphatides containing two atoms of phosphorus to one of nitrogen, from egg-yolk, for instance (MacLean), from liver (Baskoff) and from kidneys. However, MacLean is not convinced that the products obtained are pure compounds and, indeed, the phosphatide obtained from the heart is not the same as that from the kidneys.

Levene states that cuorin is simply kephalin contaminated with stearo-glycerophosphoric acid and aminoethyl stearo-glycerophosphate, the latter compound being the first product of the hydrolysis of kephalin—and corresponding, therefore, to lysocithin.[1] Levene purified cuorin by treatment with methyl ethyl ketone, followed by precipitation of an emulsion in water with hydrochloric acid, and obtained a product having all the properties of kephalin.

Neottin.—If after treating egg-yolk with cold alcohol to take out the lecithin the residue be extracted with boiling alcohol, the second extract becomes turbid on cooling owing to the separation of a white substance. To this product Fränkel gave the name Neottin. The same substance has been obtained by other workers, and it seems to be identical with Dunham's **carnaubon,** isolated from ox kidneys. MacLean's investigations lead him to conclude that it is nothing more than a mixture of sphingomyelin and cerebrosides.

Vesalthin is the name given to a mono-amino-monophosphatide isolated by Fränkel from ox pancreas by extraction with hot acetone. Vesalthin was said to contain myristic acid and a base having four methyl groups attached to the nitrogen atom, a surprising enough conclusion, since, if that were so it must either be a tetramethyl-ammonium salt or a compound of the lower homologue of choline, viz. :

$$(CH_3)_4N{\cdot}OH \qquad \text{or} \qquad (CH_3)_3N(OH){\cdot}CH_2{\cdot}OH,$$

[1] It would seem from what we know of lysocithin, that aminoethyl stearo-glycerophosphate should render cuorin intensely hæmolytic.

and the latter should decompose immediately into formaldehyde and trimethylamine.

Vesalthin seems to belong to the group of phosphatides the composition of which depends essentially on the method of isolation.

Jecorin, isolated by Drechsel from horse-liver, is another, typical, member of this group. It is a mixture of lecithin, kephalin and glucose, containing a fair percentage of sodium. Different chemists have published different analyses, and the composition of the substance depends on the number of times it is treated with ether and alcohol. The percentage of sugar, for example, varies from 14 to 18; that of phosphorus from 1·40 to 3·50; that of sodium from 2·8 to 6; and that of nitrogen from 2·6 to 6·2. But, although there is every reason to believe that jecorin is a mixture, it should be pointed out that that the phosphatide (plant lecithin) in plants (rice, soya, oats, etc.), contains a considerable proportion of sugar (up to 18%).

The most characteristic property of jecorin is its solubility in water. A combination closely resembling jecorin has been prepared artificially (Mayer); it also is soluble in water, and by this means it is possible to administer lecithin by subcutaneous injection.

To sum up, those phosphatides of which we have just given a brief account are badly defined substances, and chemists are unfavourably disposed towards regarding them as individual compounds. The same may be said of most of those isolated from brain-substance.[1]

More than fifty years ago, an English physician, Thudichum, carried out an extensive investigation into the chemical composition of the brain. He described a large number of phosphatides, drawing very subtle distinctions between one and another. No doubt, taking everything into consideration, Thudichum really made the question unduly complicated, but it must not be forgotten that we owe to him the discovery and exact description of kephalin and sphyngomyelin.

One of the components of nerve tissue has given rise to much experimental work and busy discussion. The substance in question is **Protagon.**

Protagon forms crystals, that is to say, it appears to possess a characteristic of purity which is lacking among most phosphatides. Yet, for all that, at the present time, the general opinion is that it is a mixture of cerebrosides and sphyngomyelin. Its ability to crystallise is due to the fact that two crystallisable substances enter into its composition. Thudichum, indeed, had already stated (but his opinion was not accepted) that protagon was a mixture, but it needed further work, carried out chiefly by Thierfelder, Rosenheim and Tebb, to show that, by simply recrystallising it from alcohol or pyridine, there could be separated out phosphorus-free substances corresponding to Thudichum's cerebrosides (the cerebron of Thierfelder; phrenosin and kerasin of Thudichum, Rosenheim and Tebb).

[1] Phosphatides, the nature of which is unknown, exist also in the blood.

Protagon exhibits a curious phenomenon with which the notion that it is a homogeneous substance is not in very good accord. In 3% solution in pyridine, at 30°, it is optically active, showing $[\alpha]_D^{30} = + 6{\cdot}8°$. If the temperature be raised to 50°, the optical activity is lost. When the solution is cooled, it becomes turbid (separation of sphyngomyelin) and strongly lævo-rotatory, until $[\alpha]_D^{30} = - 116°$. Soon there is so much matter in suspension that observations cannot be made, but if one waits until the precipitate has settled, one finds that the lævo-rotation has decreased to $- 13{\cdot}3°$. When the mixture is warmed, so that the deposit is redissolved, the solution exhibits again the original dextro-rotation of $+ 6{\cdot}8°$.

We may mention in addition **leukopoliin,** a pentamino-phosphatide discovered by Fränkel, which, according to MacLean, is an impure carnithin,[1] **sahidin,** etc.

The literature on phosphatides has, we observe, been considerably enriched by Fränkel, but he has given us nothing of permanent value.

CLASSIFICATION OF THE PHOSPHATIDES

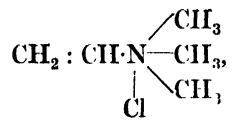
Several classifications have been put forward, but only one fits in with our present knowledge.

The phosphatides may be divided into two groups :

I. Those derived from glycerol—
   (*a*) Kephalin, almost insoluble in alcohol ;
   (*b*) Lecithin ; and (*c*) Cuorin, easily soluble in alcohol.

II. Those not derived from glycerol, but from some other alcohol, Sphingomyelin, insoluble in alcohol.

Lecithin.—Gobley, a French pharmacist, discovered lecithin in egg-yolk in 1846, and not only characterised it, but carried out what was for the time a very noteworthy investigation, in which he showed that it contained glycerophosphoric and fatty acids. He also recognised that nitrogen was present, but believed it to be combined as ammonia. Liebreich, in 1865, thought that he had identified the nitrogenous component as neurine,



but it was definitely proved to be choline by Diakonow and Strecker (1867–68).

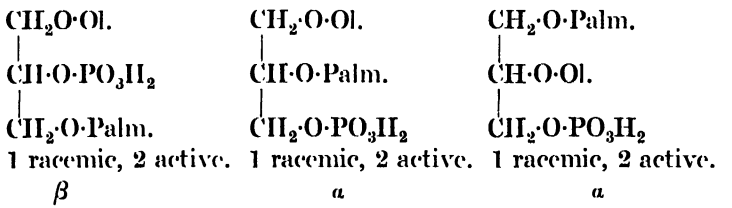
When dealing with such a substance as lecithin, not easily isolated in a pure condition, one must not confuse that which is really and that which is only possibly the desired product. In studying the

[1] The carnithin that frequently accompanies phosphatides should not be confused with carnitine, a hydroxybutyro-betaine; nothing is known of carnithin, except that it contains 28–29% of nitrogen.

possible variations of the constitutional formula for lecithin we do not necessarily expect that substances corresponding to all of them actually exist; but if, by more refined methods, we succeed in isolating substances closely resembling lecithin and having the same empirical formula, we shall know better where we are if we have looked into the theoretical possibilities beforehand. As we have already said, it is not that we are ill-acquainted with natural lecithin, but with the various forms it may take.

Now lecithin is a derivative of glycerophosphoric acid, a compound that had just been synthesised by Pelonze when lecithin was isolated for the first time by Gobley. This information, however, does not take us far. Glycerophosphoric acid may be formulated either as the α- or as the β-derivative.

The α-acid possesses an asymmetric carbon atom and can therefore also exist in two optically active modifications. Thus already there are four possible glycerophosphoric acids, two optically active, two inactive. To these should correspond four lecithins, assuming that in the lecithins there is only one kind of fatty acid, radical, oleic or palmitic, for example. But if, as is certainly the case, the lecithin molecule contains two different fatty acid radicals, then there are nine possible isomerides, *i.e.*, six derived from the α- and three from the β-glycerophosphoric acid:



Then, considering that many other fatty acids besides oleic and palmitic have been isolated from the lecithin mixture, it is easily seen that a huge number of lecithins may exist.

To which class does the lecithin of which we know the most, that from egg-yolk, belong? The results of recent investigation will give us a more or less satisfactory answer to the question.

Willstätter and Lüdecke, and later Levene, showed that by hydrolysing lecithin with cold barium hydroxide solution, an optically active glycerophosphoric acid, rotating the plane of polarisation to the left, could be isolated. The mixture called lecithin must therefore

contain at least one derivative of α-glycerophosphoric acid. But even if the presence of the optically active acid, *i.e.*, the α-acid, were thus demonstrated, that there might also be present derivatives of the β-acid remained an open question. However, most investigators working with lecithin, and examining the salts of the glycerophosphoric acid derived from it, have been struck by the fact that the calcium salt obtained may be separated into at least two fractions, one of which is crystalline, anhydrous and sparingly soluble in cold water (Cousin and Fourneau), whilst the other is amorphous, hydrated, and readily soluble in water. Piettre and Fourneau, in a paper on the alcoholysis of lecithin, drew attention to the fact that two calcium salts, corresponding to two different acids, were obtained, and suggested that this supports the idea that there are two lecithins, one derived from α-glycerophosphoric acid, the other from the β-isomeride; but at that time they were ignorant of the properties of the two glycerophosphoric acids (the two compounds having not yet been isolated) and so were unable to prove their point. Quite recently, however, Bailly has carried out a careful research on the glycerophosphates, and has been successful, not only in preparing both the α- and the β-glycerophosphoric acids, but also, by means of a method to be described later, in demonstrating that the two acids are both present in the products of the hydrolysis of lecithin.

This conclusion is a matter of interest to pharmacists. For some years Messrs. Poulenc have had on the market glycerophosphates as crystallised preparations and these products have attracted a certain amount of attention, and have already been made the subject of several investigations. There is an increasing tendency to prescribe only crystalline glycerophosphates because of their absolute purity, and the question of excluding others from the pharmacopœia is being discussed in France. As the presence of both the α- and the β-isomerides in egg-lecithin has been demonstrated, it seems most rational to continue employing the usual commercial product for pharmaceutical purposes, since it contains the two isomeric acids in just about the same relative proportions as does egg-yolk.

Let us return now to M. Bailly's method of characterising the two isomerides. The underlying principle is that of a colour reaction discovered by M. Denigès, the skilled public analyst of Bordeaux. Denigès showed that polyhydric alcohols containing an adjacent β-keto group, dihydroxyacetone being a particular example, form intensely coloured solutions with certain phenols in presence of sulphuric acid and bromine: thus, with guaiacol, the coloration is blue; with salicylic acid, red; with resorcinol, blood-red.

Now α-glycerophosphoric acid can be converted by oxidation into a derivative of dihydroxyacetone, viz.,

$$CH_2(OH)\cdot CO\cdot CH_2OPO(OH)_2,$$

and so can be made to fulfil the conditions for producing characteristic

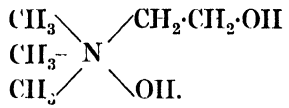
colorations with phenols. But the β-acid, on the other hand, cannot yield a ketone and so cannot react in this way.

Bailly's reaction is carried out in practice as follows: To 10 c.c. of calcium glycerophosphate solution is added 0·25 c.c. of 2·5% bromine water. The mixture is left for twelve hours, then divided into 0·5 c.c. portions in test tubes. For the colour test is to be added either (1) 0·10 c.c. of a 5% solution of resorcinol, followed by 2 c.c. concentrated sulphuric acid; a fine cherry-red colour should appear in a short time, or if the liquid be warmed; or (2) 0·10 c.c. of a 4% potassium bromide solution, 0·10 c.c. of a 5% alcoholic solution of salicylic acid, and finally 2 c.c. concentrated sulphuric acid. A very intense violet-red colour develops when the mixture is warmed in a water-bath. With (3) guaiacol, under the same conditions, the coloration is a most intense blue.

May it be said that the question of the nature of the glycerophosphoric acids of lecithin is almost settled? By no means, because the only lecithin glycerophosphoric acid investigated up to the present has been that derived from that kephalin-lecithin mixture which constitutes the lecithin of egg-yolk.

Let us now turn our attention to the basic portion of the lecithin complex.

Choline is a quaternary base derived from an alcohol, being *trimethyl-hydroxyethyl-ammonium-hydroxide*,



It is generally admitted, nowadays, that the choline in lecithin is combined with the glycerophosphoric acid by its alcoholic hydroxyl, the compound thus being an ester; but there are still those who say that the combination is simply a salt-formation. Several facts, however, indicate that the first explanation is correct. Thus, in the first place, salts of the alkaline earths, those of calcium in particular, give with lecithin precipitates which contain all the lecithin and all its original choline.[1] If the choline were present in salt-like combination, one would expect double decomposition to take place with the calcium salt, precipitating calcium oleo-stearo-glycerophosphate, which would certainly be insoluble, and leaving the choline in solution.

[1] It seems now to be definitely proved that lecithin is a choline derivative. Certainly associated with lecithin there are phosphatides which only differ from it in the nature of the aminoalcohol radical—this may be aminoethyl alcohol itself or its mono- or dimethyl derivatives. But one ought not to call these substances lecithins; it is much better to reserve the name lecithin for esters of choline.

Secondly, ferric and ferrous chlorides, platinic chloride, and cadmium chloride[1] form double salts with lecithin.

Lastly, in the experiments with snake-venom, to be described later, lecithin was found to be converted into a white substance, soluble in warm water; this is *lysocithin*, it contains all the choline originally present in the lecithin but only one fatty acid, oleic acid having been liberated by the action of the enzyme in the venom. Now, if free oleic acid and choline combined as a salt were present in a solution, the choline would certainly distribute itself between the oleic acid and the glycerophosphoric acid and not remain in its original state of combination.

The behaviour of lecithin towards colouring matters should yield valuable evidence as to the ester-like combination of the choline. Unfortunately, it is a difficult matter to carry out such an investigation, and doubtless only hydrogenated lecithins—these derivatives being easily obtained in a colourless crystalline condition—could be used in the undertaking. Above all things, it must not be forgotten how easily and quickly lecithin is hydrolysed.

It only remains now to discuss the fatty acids of lecithin. This means discussing two questions, namely, the nature of the fatty acids and their position in the molecule.

From lecithin there have been isolated palmitic acid, stearic acid, oleic acid (unsaturated, one double bond), linolic acid (two double bonds) and linolenic acid (three double bonds). Let us consider, however, a lecithin containing only the palmitic and oleic radicals, there certainly being such a compound. If, now, analysis shows that only these two acids are present, may one state definitely that they are derived from a single lecithin, *i.e.*, is lecithin always a mixed ester, the molecule of which contains two different fatty acid radicals? And, if these are different, is one always unsaturated, as has been stated to be the case by MacLean, Rollet, and others? Or, alternatively, is the mixture that we isolate as lecithin composed of a di-palmitic lecithin and a di-oleic lecithin, that is, of two homogeneously derived lecithins taken up side by side by solvents?

The general question here is of great importance in relation to the origin of lecithin. We believe that it can be fully answered by submitting lecithin to the action of snake venom (Delezenne and Ledebt). Thus, by this means a crystalline substance, termed by Delezenne and Fourneau **lysocithin,** was obtained from egg-yolk, and was found to be lecithin deprived of one of its fatty acid radicals; it, therefore, contained only one instead of two. Now, the fatty acid radical remaining in the lysocithin is always saturated. It is mostly that of palmitic acid, never of oleic. The oleic acid, or, strictly speaking, all the

[1] It has been shown by Levene, in opposition to what was generally believed, that cadmium chloride does not break up the lecithin; it simply precipitates it, and the precipitate contains nearly all the choline. This could hardly happen if the compound were a choline salt.

mixture of unsaturated acids, can be found in the mother liquors from which the lysocithin has been separated.

If the egg-yolk contained lecithins derived both from saturated acids (di-palmitic or stearopalmitic) and from unsaturated acids (di-oleic), then, when the action of the venom was complete, the mixture should contain oleic lysocithin or unattacked di-palmitic lecithin, but actually not a trace of any lecithin is present, and the lysocithin is always a saturated compound (or, at least, if an oleic ester be present, it is in very small amount). With egg-yolk this is always the case, but a precise statement cannot be made for other lecithins, as our knowledge is too scanty. Other arguments may be quoted. The properties of the lecithin derived solely from saturated fatty acids (palmito-stearic) are now well known, as the compound may be obtained by reducing lecithin. This, a saturated lecithin (hydrolecithin) crystallises well, is sparingly soluble in alcohol, is not hygroscopic, is colourless and quite stable, and so, if it were present in egg-yolk, it should be easily isolated.

Egg-yolk lecithin, therefore, is a mixed ester containing both an unsaturated radical (chiefly that of oleic acid) and a saturated radical (perhaps exclusively that of palmitic acid). As it appears that neutral fats are usually chemically homogenous,[1] that is to say, the individual glycerol esters present in the mixture are derived solely from saturated acids (tri-stearin, tri-palmitin) or from unsaturated acids (tri-olein), lecithin can hardly be derived from that source,[2] unless the fats undergo a profound change.

The positions taken up by the two fatty acid radicals in the lecithin molecule are still undetermined. Of course, in derivatives of β-glycerophosphoric acid, it does not matter how the radicals arrange themselves, as both the free hydroxyl groups are primary; but for the lecithins derived from α-glycerophosphoric acid evidence is, at present, lacking, and so we do not know how the two acid groups are located nor whether they always take up the same position.

From the results described above, an extremely important conclusion is to be drawn, viz., that under certain conditions (see later under Lysocithin) a hydrolytic agent contained in snake venom, a *lecithinase*, in short, has the extraordinary property of being able to split off from lecithin one, and only one, acid radical, this being always that of the unsaturated acid. We cannot carry out by any purely chemical method at present known such a delicate, complete, and specific hydrolysis, even on common fats,[3] and so far attempts in this direction have not given encouraging results. In our opinion it will

[1] A mixed fat has been found in butter.

[2] No serious research has been undertaken with the object of tracing a relationship between the lecithin and the fat in a particular organ.

[3] Quite recently, it has been found possible to split off, one after the other, the three acid radicals in a fat, by a regulated alcoholysis.

be most difficult to avoid displacing the choline radical in the first stage of hydrolysis or alcoholysis.

The general properties of lecithin are well known, as the substance has been employed in therapeutics for about twenty years. Its introduction into pharmacy resulted from the work of Billon, Desgrez, and others. Lecithin is a white substance when pure and freshly isolated. It may be obtained in a dry, friable condition, but is very hygroscopic and unstable. Lecithin dissolves in almost any solvent but acetone ; with water a quite stable opalescent mixture is formed, having a powerful emulsifying action on fats and oils.

The substance is auto-oxidisable ; its iodine number falls rapidly when it is exposed to air as a result of the addition of oxygen at the double bonds. This property of auto-oxidation, due perhaps to the presence of traces of metallic catalysts (iron), is one of the most remarkable characteristics of lecithin. Possibly before it forms an integral part of the lecithin molecule the oxygen exists, for a time, loosely combined in a very active form, the combination sharing, more or less, in the work of transporting oxygen within the organism. Willstätter's experiments on the fixation of oxygen by phosphorised oil make this a plausible hypothesis. Furthermore, it should be noted that this readiness to undergo oxidation makes lecithin a powerful reducing agent.

Lecithin rapidly undergoes alcoholysis with an alcoholic solution of hydrogen chloride, even if this be very dilute. The reaction can be used to determine quantitatively the composition of the substance.

The numerous addition-compounds formed by lecithin provide us with some interesting observations. We may note, for example, that only a part of the lecithin is removed by extracting egg-yolk with ether, the remainder being firmly held by the albumin present. In this connection Erlandsen certainly believed himself justified in stating that it was not a question of an additive compound of phosphatide and albumin, but that the lecithin retained was of a different composition from that dissolved by the ether. However, preliminary treatment with alcohol is known to set all the lecithin free, it all becomes soluble in ether, so Erlandsen's hypothesis is hardly in agreement with the facts. It must, therefore, be granted that lecithin and albumin enter into more or less stable combination—have, at least, an affinity for one another—and very probably such additive compounds play an important part in making homogeneous such a mixture of neutral fats, albumins, salts and lecithin as exists in serum. Additive compounds like these under discussion have been obtained synthetically by Mayer.

Lecithin forms complex combinations not only with albumin but

also with salts,[1] with glucose, and most likely with cholesterol. Even if these are not true compounds they are, at any rate, quite stable adsorption products.

The function of lecithin in the dissemination and effective action of certain drugs, particularly the hypnotics, is still obscure, but its importance is not to be doubted, as the elegant experiments of Overton and Meyer indicate. These investigators state that one of the chief functions of the lipoids, and particularly of the phosphatides, is to control the osmotic action of vegetable and animal cellular membranes. Thus, this is how Overton himself puts the matter: "The brain lipoids, which are composed almost exclusively of phosphatides, form an integral part of the protoplasm of all vegetable and animal cells, and, for the life of the cell, are second in importance only to the proteins. Quite probably, as far as the physical state of the protoplasm is concerned, they are even more important than the latter. Modifications of a physical nature, which the lipoids undergo as a result of absorbing foreign substances (no matter how such modifications influence vital phenomena), provide a common starting point from which the principal action of more than half the organic substances in question begins."

Lastly, there is a point to be particularly insisted on, namely, the huge number of phosphatides that may possibly exist. We have already pointed out that lecithin itself may have very many isomeric forms, even if it contains only two different fatty acid radicals. And when one knows how extraordinarily specific fermentative actions are, or in general, how specific all biological behaviour is, it seems evident that every isomeric or homologous form (if, of course, they actually exist), may have its particular affinity for such substances as combine with the phosphatides or, in fact, for any agent that can modify their structure. We have already seen how snake venom can act, and it is possible that other enzymes, produced by micro-organisms or even formed normally in the cell, may disturb the phosphatides and so bring about profound disorders in the organism.[2]

Lecithin may be regarded as a soap-like compound, yielding, with salts (MCl), such compounds as:

Actually it is nearly always found to contain traces of calcium and the alkali metals. In kephalin these elements seem to occur in a constant proportion. It should not be forgotten that lecithin has a high molecular weight; it is easy to fall into the mistake of thinking that

[1] Even in 1846 Gobley noted that lecithin clings to calcium phosphate and thought that in this way it helped in the introduction and transportation of phosphates in the animal body.

[2] Lysocithin is a poison. Another ester of choline, acetylcholine, recently discovered in ergot, is much more toxic than choline itself.

a metallic component is an impurity, when it may actually be an integral part of the substance. Thus, 1,600 gm. lecithin needs theoretically only 24 gm. magnesium to saturate it ; so in an egg containing about 0·8 to 1·0 gm. lecithin, only 0·01 to 0·015 gm. magnesium would be needed to produce the salt. Similarly for calcium, the presence of 0·02 to 0·03 gm. would be necessary, and just that amount is actually found in egg-yolk. So there is nothing against the notion that the phosphatides may actually be agents in the transport of metallic elements, particularly the more important ones, such as calcium and iron. And it should be noted that small amounts of a metallic element may entirely alter the properties of the phosphatides ; for example, the combination formed by lecithin with an iron salt is quite insoluble in alcohol.

Phosphatides appear to have a great influence on the solubility of certain substances in the fluids of the organism. This effect, chiefly as far as the solubility of fatty acids in the bile is concerned, has been investigated by Parker. A 5% solution of bile salts containing 7% more lecithin than is normally present will take up oleic acid to the extent of four parts to 100 of solution ; pure water dissolves only 0·1% and a bile salt solution without added lecithin only 0·5%. In the same way sodium oleate forms a 5% solution in water, 7·60% in bile salt solution without lecithin, 11·50% with lecithin.

Preparation of Lecithin.—Various methods of isolating lecithin have been recommended (the simplest is described later in the practical section of the book, together with a detailed description of the method of alcoholysis). The product obtained by all these methods is a mixture containing kephalin. To obtain a tolerably pure product, Levene works up the egg-yolk extract that has served for precipitating lecithin. The procedure is as follows :

The eggs are extracted with alcohol, the extract concentrated by evaporation and the residue treated with acetone. The precipitate obtained consists of lecithin containing a considerable proportion of kephalin. Conversely, the acetone solution contains mainly lecithin, and this is precipitated with cadmium chloride. The compound obtained is purified by recrystallisation from a mixture of alcohol and ethyl acetate and then yields pure lecithin when decomposed.

In identifying phosphatides, Levene tends more and more to use the method of reduction with hydrogen and palladium. The hydrogenised phosphatides obtained are unaffected by the air, crystallise readily, and are sparingly soluble in cold solvents. They are stated to be obtained in a condition of almost absolute purity, so further investigation is considerably facilitated.

MacLean obtains satisfactory results by repeatedly precipitating an aqueous emulsion of crude lecithin by acetone and then redissolving it in and reprecipitating it from absolute ether.

Kephalin.—Kephalin always accompanies lecithin, but sometimes,

as in the phosphatides of the brain, it predominates. In the case of egg-yolk, which contains only a small proportion, kephalin may be separated from lecithin by treatment with cadmium chloride. The compound with this reagent is produced by precipitation in an alcohol solution, then recrystallised from a mixture of 80% alcohol and ethyl acetate. Under these conditions the kephalin remains in the solution.

However, kephalin is best prepared from the brain. Fresh brain (preferably dried) is crushed up with acetone, then extracted with petroleum ether. The solution obtained is concentrated at a low temperature and eventually cooled to — 20° to throw out most of the cerebrosides. The clear liquor is decanted off and alcohol added to precipitate the kephalin. The crude kephalin thus obtained is dissolved in ether, precipitated by acetone, again dissolved in ether and again precipitated, the process being repeated several times. Finally the ether solution is treated with absolute alcohol. Kephalin separated in this way contains salts (potassium, calcium), from which it is freed by washing with very dilute hydrochloric acid, and in order to purify it completely, the lead compound, produced by treating a solution of the kephalin in amyl alcohol with lead acetate, is prepared and then decomposed.

Kephalin closely resembles lecithin in its properties, but when nearly pure and very dry is almost insoluble in ether or alcohol.

It was thought that kephalin and lecithin differed only in that one was a derivative of aminoethyl alcohol, whilst the other was a derivative of choline, but, according to Levene, analytical figures show that the difference is more profound, the fatty acid radicals in the two compounds being probably not the same. The percentage of oxygen does not agree with the earlier supposition. And, although it is highly probable that the base is aminoethyl alcohol, if it could be isolated in substance and a characteristic derivative (urethane, acyl compound, etc.) prepared, the conclusion would be sounder. We ought to point out, moreover, that kephalin should differ from lecithin in its chemical properties, or at least, in one of them : aminoethyl alcohol, hydroxyethylamine, is a much weaker base than choline, and so, judging from general experience of the salts of acyl derivatives of aminoalcohols, we should expect kephalin to be acid to litmus.

Sphingomyelin.—All organs that contain lecithin and kephalin also contain sphingomyelin. Like kephalin, it is particularly abundant in nerve tissue, but egg-yolk also contains a small proportion. It is very sparingly soluble in most solvents, so is easily obtained in a pure condition from organs that have been already treated with alcohol and ether to extract the other phosphatides. As it will dissolve in hot pyridine, it is best isolated by taking advantage of this property (Rosenheim and Tebb).

The organs, previously treated with acetone, alcohol and ether, are accordingly extracted with hot pyridine. When the solution cools to

room temperature a precipitate settles out. This impure sphingomyelin is then purified by (i) dissolving it in hot glacial acetic acid, the impurities being thrown down from the cold solution; (ii) to the filtered solution adding acetone, the partly purified sphingomyelin being precipitated; and (iii) dissolving the material in a mixture of alcohol and petroleum ether, filtering, and reprecipitating by adding an excess of alcohol.

Sphingomyelin is a white substance, non-hygroscopic, and unaffected by exposure to light. It is insoluble in alcohol and forms with water a kind of starchy paste. It is optically active, being lævorotatory.

Hydrolysed, sphingomyelin yields phosphoric acid, a C_{24} saturated fatty acid, namely, lignoceric acid, and two bases, choline and sphingosine. Sphingosine is a dihydric alcohol and is also unsaturated (one double bond); it has the formula:

$$CH_3{\cdot}(CH_2)_{11}{\cdot}CH : CH{\cdot}CH(OH){\cdot}CH(OH){\cdot}CH_2{\cdot}NH_2,$$

and presumably is combined with the phosphoric acid by one hydroxy group, the other remaining free, whilst one of the remaining hydrogens of the phosphoric acid is replaced by the choline radical. The lignoceric acid ($C_{24}H_{48}O_2$) forms with the amino group in the sphingosine an amide. Sphingomyelin should, therefore, have the structure here shown (Levene):

$$\begin{matrix} & & O{\cdot}C_{17}H_{32}(OH){\cdot}NH{\cdot}CO{\cdot}C_{23}H_{47} \\ & \diagup & \\ PO{\cdot}OH & & \\ & \diagdown & \\ & & O{\cdot}C_5H_{14}N. \end{matrix}$$

At one time it was supposed that the sphingomyelin complex contained an alcohol [1] (*sphingol*) playing much the same part as an intermediary between choline and the fatty acids as glycerol plays in lecithin, but, as is seen, Levene's formula leaves sphingol out of account.

All the same, it should be pointed out that Levene's formula is not in good agreement with the fact that sphingomyelin is as readily hydrolysed by acids as by bases, and amides are usually more or less resistent to acid hydrolysis.

LYSOCITHIN

This name was given (Delezenne and Fourneau) to a phosphatide derived from lecithin by treatment with snake venom, partial hydrolysis being brought about by an enzyme-like agent (Delezenne and S. Ledebt). Kyes's cobra-lecithide, considered by its discoverer and the Ehrlich school to be a new compound formed between lecithin and

[1] Note that sphingomyelin contains a carbon chain of 17 atoms. If the formula given is truly that of sphingomyelin, this should be derived from a C_{18} amino acid (dihydroxy-oleic acid) by loss of CO_2. It is not impossible that amino acids of very high molecular weight should be discovered some day or another in the organism. Perhaps they have never been looked for.

some unknown principle in the venom, is actually a mixture of lysocithin, unconverted lecithin and venom. Its existence, therefore, as a chemical entity, already shown to be doubtful from the start by the results published by other authors (Ludecke, Dungern and Coca), it being suggested, but not actually demonstrated, that the venom was acting as a ferment, is a hypothesis that must now be definitely rejected. Not only so, but lysocithin is a well-defined compound which can be crystallised and obtained in a highly purified condition. It is a relatively simple phosphatide, and we think it will be useful to describe the method of preparing it and to enumerate its principal characteristics, not only because of the new evidence as to the constitution of lecithin that it has yielded, as has already been mentioned, but also because of its very remarkable physiological properties.

The physiological properties of lysocithin cannot here be considered at length, but nevertheless attention should be directed again to the fact that it possesses a powerful cytolytic action, and that to this should be referred the hæmolytic action exhibited by small doses of venom when added to blood serum or ovolecithin. In fact, as Delezenne and Ledebt have shown, the venom simply carries a ferment which, provided the time is long enough, can, even in minute doses, convert all the lecithin present into lysocithin.

Lysocithin is prepared in the following way: To an emulsion of two egg-yolks and enough physiological saline solution to make up a volume of 100 c.c., one milligram of cobra venom is added. The mixture is set in the oven at 50° for twelve hours, then evaporated to dryness *in vacuo*. The resulting powder is rubbed up with acetone (cold), dried, and extracted with absolute alcohol. To the concentrated extract ether is added, whereupon a voluminous precipitate is thrown down. This is separated with the centrifuge and washed with ether, the white powder so obtained being then recrystallised several times from absolute alcohol. The product is now dissolved in boiling chloroform; on cooling, a mass of crystals separates; they are removed, dissolved in a little absolute alcohol and petroleum ether added to the warm solution until it becomes opalescent. In a moment or so, needle-like crystals appear and soon the lysocithin separates as a shower of brilliant scales.

Prepared in this way, lysocithin is soluble in tepid water or in hot alcohol. It is sparingly soluble in chloroform, almost insoluble in benzene, quite insoluble in ether. It is neutral to litmus. In neutral solution no precipitate is formed with either gold or barium chlorides, nor with lead acetate. The substance also does not give the Florence reaction.

Ultimate analysis and the nature of the decomposition products (alcoholysis) show crystalline lysocithin to be choline palmito-glycerophosphate (ester).

Besides its powerful hæmolytic action, lysocithin also possesses a

well-marked affinity for cholesterol. It has been shown (Sachs and Preston Kyes) that an emulsified aqueous mixture of cobra venom and cholesterol has no hæmolytic power. Actually, cholesterol has no action on the venom (Delezenne and Ledebt), but only on the lysocithin formed; the hæmolysin is neutralised by the cholesterol, and the mixture of these two substances, in definite proportions, has no hæmolytic action. In fact, if a molecular proportion of lysocithin be dissolved in alcohol and two molecular proportions of cholesterol in chloroform, and the two solutions be mixed and evaporated to dryness, a mass is obtained which forms an unstable emulsion with water from which cholesterol in a finely divided form separates. If the emulsion be extracted with ether, exactly *one* molecular proportion of cholesterol is removed, the other remaining combined with the lysocithin. The resulting mixture forms a stable emulsion having no hæmolytic power. It retains water in a surprising way and separation of the latter is extremely difficult. If alcohol be added to the emulsion, treatment with ether will now remove all the cholesterol. One cannot help comparing these phenomena with those observed when, to isolate lecithin, organs are extracted with ether either with or without preliminary treatment with alcohol.

Conclusion.—Our present knowledge of kephalin, lysocithin and sphingomyelin enables us to sum up in this way:

(1) What has hitherto been isolated under the name of lecithin appears to be a mixture of true lecithin, kephalin, and, perhaps, other phosphatides of unknown composition;

(2) Everything suggests that there are at least two mono-amino-phosphatides, the first, viz., choline oleo-palmito-glycerophosphate, being *true lecithin* (possibly this lecithin is derived solely from glycerophosphoric acid), whilst the second, kephalin, is, in a way, analogous to lecithin, but differs from it by having amino-ethyl alcohol in place of choline, and probably also by being derived from a particular fatty acid;

(3) Sphingomyelin, it seems, is well characterised and very little more work should be needed definitely to establish its constitution.

The synthesis of lecithin has yet to be carried out. The great, almost unsurmountable, difficulty lies in the esterification of choline by distearoglycerophosphoric acid. Hundeshagen states that he has prepared the acid in question, but his results have not been confirmed. Further progress has not been realised; Grün and Kade's attempts also gave unsatisfactory results. Maybe it will be better to prepare first the glycerophosphoric ester of choline, as Langheld has done, and then try to combine this with the appropriate fatty acid radicals. But we think that the same obstacles will have to be overcome; to begin with, the esterification of glycerophosphoric acid is a difficult process which has not yet been carried out, and even the conversion

of lysocithin to lecithin, apparently so simple a matter, has not been realised. Yet that these difficulties will be surmounted is not to be doubted, but perhaps the result obtained will come as an unpleasant surprise; the artificial lecithin may have nothing in common with the natural product; and again we shall have to revise our ideas of the nature of this curious substance.

CHAPTER XII

NUCLEIC ACIDS

TRUE nucleic acids are complex esters in which phosphoric acid is combined with carbohydrates and purine or pyrimidine bases. They are of two kinds, namely, plant nucleic acids, in which the carbohydrate is a pentose, *d*-ribose, and animal nucleic acids, which probably contain a hexose. In the organism the nucleic acids are combined with proteins to form nucleins.

Historical.—In 1868 Friedrich Miescher subjected pus to a chemical examination and introduced the term *Nuclein.* Hoppe-Seyler (1871) then prepared a nuclein from yeast and shortly afterwards Miescher separated from the spermatic fluid (*Lachsmilch*) of the salmon the phosphorus-containing portion of the protamine. Piccard (1874) proved that, from this substance, purine bases, viz., guanine and hypoxanthine, could be obtained. Kossel (1891) demonstrated the difference between true nucleins and pseudo-nucleins, the phosphorus-containing part of the latter substances yielding no purine or pyrimidine bases; he also established the relationship between these constituents of the cell-nucleus and the uric acid of the urine. Altmann (1889) succeeded in separating the nucleic acid and the protein from beer yeast, whilst Kossel and Neumann (1894) worked out a practical method for removing the protein and isolated thymus nucleic acid.

Hammarsten (1894) obtained a nucleoprotein from the pancreas and proved that its molecule was built up from a pentose and guanine. Neumann (1896–97) isolated thymic acid, which is formed from thymus nucleic acid by loss of the purine bases. From 1900 to 1912, Levene and his collaborators carried out a series of admirable investigations and succeeded in establishing the constitution of yeast nucleic acid, so that at the present time only a few small questions of detail remain to be cleared up.

Steudel turned his attention to guanylic acid; Hammarsten and Jones investigated the action of enzymes on nucleic acid (1907–14); and the physiological action of its products of hydrolysis was studied by Thannhauser and Dorfmüller (1914–17).

THE DIFFERENT KINDS OF NUCLEIC ACID AND THEIR PROPERTIES

I. Simple Types or Nucleotides

The simplest type of nucleic acid contains a pentose group. The first example to be isolated was **guanylic acid,**[1] $(HO)_2 \cdot OPO \cdot C_5H_8O_3 \cdot C_5H_4N_5O$, which, hydrolysed, yields *guanine.*

[1] Also known as guanosin-phosphoric acid.—*Tr.*

It is obtained by boiling pancreas with water; the soluble nucleoprotein is taken up and may be precipitated by adding acetic acid, but by the action of dilute caustic potash the protein is separated and acetic acid then only precipitates guanylic acid. With lead acetate it forms a precipitate and also yields a crystalline brucine salt.

Inosinic Acid was isolated by Haiser and Wenzel from an extract of muscle. It was precipitated as a lead salt by lead acetate and purified through the barium salt. It differs from guanylic acid only in having *hypoxanthine* in place of guanine.

Uridinic Acid (Thannhauser and Dorfmüller) was prepared by ammoniacal hydrolysis of yeast nucleic acid followed by fractional crystallisation of the brucine salts of the mixture obtained. The base of this nucleotide is a pyrimidine derivative, *uracil*.

Cytidinic Acid (Levene, 1918) was also obtained by ammoniacal hydrolysis of yeast nucleic acid *viâ* the brucine salt. The base here is *cytosine*.

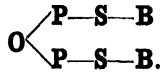
Adenosinic Acid, likewise obtained from the same source by Levene in 1918, did not yield a crystalline salt. *Adenine* forms the base in this substance.

Definitions

A **Nucleotide** is such a combination as guanylic acid, that is to say, one formed by the linking together of a phosphoric acid group and a base by means of a sugar, thus :

P—S—B.

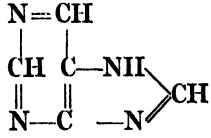
A **Dinucleotide** is the complex formed by union of two nucleotides, *e.g.*, guanylic acid and adenosinic acid, thus :



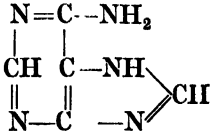
A **Nucleoside** is the glucoside-like compound of a base with a sugar,

S—B.

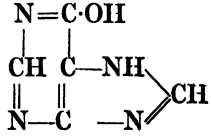
The Bases derived from the Nucleic Acids.—We have just learnt that five bases, namely, *cytosine*, *uracil*, *guanine*, *adenine*, and *hypoxanthine* may be obtained from the simple nucleic acids. Two distinct types of molecular structure are found in these compounds. Cytosine, uracil (and thymine) have a pyrimidine basis, whilst guanine, adenine, and hypoxanthine are derived from purine and are related to uric acid. These relationships are plainly shown by the formulæ given below:—



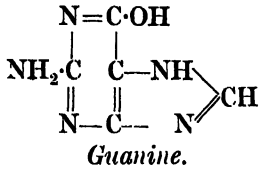
Purine.



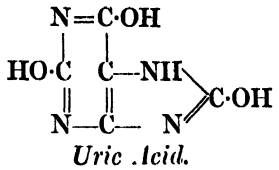
Adenine.



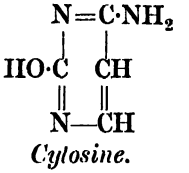
Hypoxanthine.



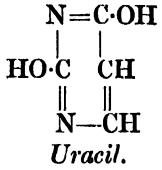
Guanine.



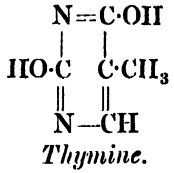
Uric Acid.



Cytosine.



Uracil.



Thymine.

Thymine has, so far, not been found in plants.

II. Compound Types or Polynucleotides

These, the true nucleic acids, are formed by the linking together of the mono-nucleotides just discussed. Of this class the two main representatives are yeast nucleic acid and thymus nucleic acid. We will consider them one at a time.

Yeast Nucleic Acid is prepared from fresh yeast. This is treated with dilute caustic soda to set free the acid from its compounds with proteins. After some time the solution is rendered neutral by acetic acid and alcohol added to throw down the sodium salt of the nucleic acid (the preparation is described in detail in the practical section). The product is insoluble in water or alcohol and gives insoluble salts with heavy metals.

The **constitution** of yeast nucleic acid seems now to be well established. Four nucleotides are united, it is supposed, through the phosphoric acid groups which thus form a pyrophosphoric chain :

		Corresponding Nucleoside.[1]
HO		
O	PO — *sugar* (*d-ribose*) — *guanine* (purine)	**Guanosin(e)**
O	PO — *sugar* (*d-ribose*) — *adenine* (purine)	**Adenosin(e)**
O	PO — *sugar* (*d-ribose*) — *cytosine* (pyrimidine)	**Cytidin(e)**
O	PO — *sugar* (*d-ribose*) — *uracil* (pyrimidine)	**Uridin(e)**
HO		

Two methods of hydrolysis have been employed in investigating yeast nucleic acid, namely, (i) chemical hydrolysis and (ii) biological hydrolysis, *i.e.*, by use of enzymes.

(i) **Chemical Hydrolysis.**—(1) **By 10% sulphuric acid at 125° for four** hours. Complete scission takes place as is represented in the following diagram, all the bases being obtained :

[1] These names should be spelt without the terminal *e*, but some authors use the alternative spelling.—*Tr.*

P—S—Pu

P—S—Pu

P—S—Py

P—S—Py

When the hydrolysis is carried out with 2% sulphuric acid under the same conditions, only the purine bases are split off, and the two pyrimidine mononucleotides are obtained, thus :

P—S—Pu

P—S—Pu

P—S—Py

P—S—Py

Complete hydrolysis is also brought about by treatment with 25% hydrofluoric acid, or with nitric acid (sp. gr. 1·2) in the cold for fifteen days (Steudel), but in the latter process the bases are de-aminated.

(2) Hydrolysis may also be effected in a neutral or ammoniacal solution at a high temperature. Levene first obtained the nucleosides by this method.

(*a*) at 140° :

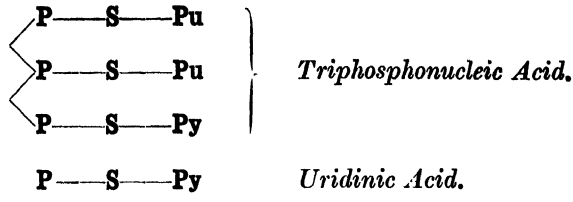
P—S—Pu

P—S—Pu

P—S—Py

P—S—Py

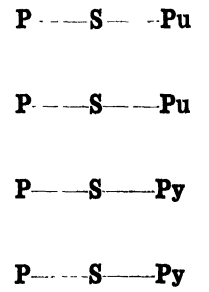
(*b*) With 25% aqueous ammonia at 100° for two hours (Thannhauser) :



Levene isolated cytidinic acid also in carrying out a similar hydrolysis (1918), and so came to the conclusion that triphosphonucleic acid was probably a mixture.

With the same reagent at 115° a mixture of all the mononucleotides is obtained, whilst at higher temperatures scission takes place in the same way as in a neutral medium (see above).

(ii) **Hydrolysis by Ferments.**—Blood serum, laked blood or an extract of the pancreas, are said to have the same action as ammonia at 115°. Intestinal juice effects a similar hydrolysis to that of a neutral medium at 140°. Extracts of intestinal mucous membrane, of the liver or the kidneys, bring about hydrolysis in the manner shown below (Levene, Medigreceanu).



Cobra venom has recently been found to have the same effect (Delezenne and Morel, 1919) if the solution be neutralised as acidity develops; but in a plain solution of the nucleinate the reaction does not go to completion. The hydrolysis is limited by the acidity produced and all the phosphoric acid is not set free, purine bases remaining combined as nucleosides.

The action of the intestinal enzymes needs further investigation. Thannhauser and Dorfmüller state that treatment with duodenal juice produces triphosphonucleic acid and uridinic acid, but their results are not in agreement with those of Levene (the experiments made with juice drawn off from an incision in the duodenum are to be criticised from the physiological point of view).

Nucleosides.—The first nucleoside to be isolated was *guanosin*. Levene and Jacob obtained this compound by subjecting guanylic acid to neutral hydrolysis (1909). It forms well-defined colourless silky crystals, and is, characteristically, soluble in warm water but

thrown completely out of solution again on cooling. With lead acetate in presence of ammonia it forms a precipitate, and it is almost insoluble both in acids and in alkalis. *Adenosin* forms an insoluble picrate, and on this characteristic property the method of preparing and purifying it is based. This nucleoside is more soluble in water than the foregoing, and can be purified by recrystallisation from alcohol. *Cytidin* and *uridin* are not precipitated by ammoniacal lead acetate solution. They may be separated by taking advantage of the insolubility of cytidin picrate.

The compounds may also be separated as nitrates in alcohol solution. From the cytidin mother liquors the uridin may be extracted either by fractional crystallisation from alcohol or as the dibenzoyl derivative.

The pyrimidine nucleosides differ from the purine derivatives in being difficult to hydrolyse, and so in not giving readily the characteristic reactions of pentoses with orcinol and hydrochloric acid.

Levene states that when inosinic acid is hydrolysed by treatment with 1% hydrochloric acid, *d*-ribose phosphoric acid, $C_5H_9O_5 \cdot PO(OH)_2$, is obtained; its barium salt is soluble in acetic acid but precipitated by alcohol.

Recent investigations have thrown doubt on Levene's conclusions regarding the structure of yeast nucleic acid. According to Jones (1916), hydrolysis with ammonia at 115° brings about a scission into two *tetrabasic* dinucleotides, the one being a combination of the guanine and cytosine derivatives, the other an adenine-uracil compound. The union here cannot be through the phosphoric acid radicals. But Levene has shown (1918) that these dinucleotides are simply mixtures of the simpler compounds described above.[1]

Thannhauser and Dorfmüller (1917) prepared an easily soluble trinucleotide by splitting off uridinic acid, as has already been mentioned. Their results ought to be confirmed.

Thymus Nucleic Acids (see also the practical part).—The constitution of these acids is apparently analogous to that of yeast nucleic acid but as one of the hydrolysis products is lævulinic acid, the carbohydrate radical is that of a hexose instead of a pentose. Furthermore, uracil is replaced by *thymine*.

There are two kinds of thymus nucleic acid. Variety A forms a sodium salt which gives gelatinous solutions, whilst the sodium salt of variety B does not possess this property. Variety B seems to be simply an undefined decomposition product of A. When thymus nucleic acid is hydrolysed by treatment with $\frac{2}{3}N$ sulphuric acid for three hours it yields thymic acid, the purine bases being eliminated (Kossel, Neumann, 1896–97). Steudel (1918) states that he obtained an exceedingly pure product by using N/300 sulphuric acid.

[1] See *Chemical Society's Annual Reports*, 1921, XVIII, 170.

The constitution of thymus nucleic acid is not yet established. Although Levene and his collaborators have carried out extensive researches, their conclusions have not been verified. Levene stated that he obtained a nucleotide, namely, *thymine hexose phosphoric acid*, and, by the action of a ferment, the nature of which he did not disclose, a nucleoside, namely, *guanine-hexose*. Moreover, from fish-sperm he prepared a dinucleotide and a thymine-hexose-diphosphoric acid. These results have not been confirmed and have fallen into oblivion. The acid should contain six replaceable hydrogen atoms. Feulgen has recently prepared certain compounds with dyestuffs, and concludes that it is tetrabasic, but that there are two much weaker acid groups not associated with the phosphoric acid radicals. He also is of the opinion that the carbohydrate is not a hexose but a *glucal*. A substance has been isolated by the action of cobra venom (Delezenne and Morel) which is probably an adenine nucleoside, but it seems to be difficult to purify. In short, therefore, the structure of thymus nucleic acid still remains to be elucidated.

Physiological Function of the Nucleic Acids.—Very little is known about the formation of the nucleic acids. Miescher observed that the salmon in the Rhine, which take no food in their journey up the river, lose nearly all their musculature, while their genital organs develop to an enormous size. And it has been found that, although the milk of mammals contains no purine bases, yet the amount of these substances in the body of the animal increases during the suckling period (Burian, 1897).

In the course of their studies of protein metabolism, Osborne and Mendel showed that, while the ingestion of certain amino acids is necessary for the proper growth of an animal, there is no demand for purine bases. It follows, therefore, that the nucleic acids originate from proteins, but of the mechanism of the process we are quite ignorant.

More is known of the decomposition of these substances. In every organ there are enzymes capable of breaking down the nucleic acids more or less completely. The action of the nucleases was concisely described by Salomon in 1881. Iwanoff (1903) showed that in cultures of *Aspergillus niger* on thymus nucleic acid phosphoric acid and purine bases were set free. In the last few years nucleases have been extensively investigated. These ferments are of three kinds, namely:

(1) Those which liberate phosphoric acid and leave the nucleosides untouched;

(2) Those which set free the bases from these glucosides (ribosides);

(3) Those which de-aminate the bases.

The alteration in rotatory power affords a basis for the method that has been chiefly used in investigating these transformations. The various ways in which scission takes place have already been pointed out.

The pyrimidine compounds are particularly resistant, and, apparently, are not broken down by the organism. Levene and Laforge have shown that hydro-uridine is unaffected by ferments, although it is hydrolysed as easily as the purine nucleosides by the action of acids.

Mendel and Myers (1910) demonstrated that free pyrimidine bases undergo no transformation in the organism. Apparently no investigation has yet been made of the metabolism of the pyrimidine nucleosides, but of that of the purine derivatives we are not quite so ignorant. Thannhauser and Dorfmüller (1914) studied the action of guanosin and adenosin after subcutaneous injection, and describe some curious phenomena. Adenine, for instance, is not broken up by the organism and has a pronounced toxic effect on the kidneys, but when injected as adenosin, that is to say, as a riboside, it is converted into uric acid and has no toxic properties. Guanosin, likewise, gives rise to uric acid and, indeed, nearly all of it is oxidised in this way. It should then be very interesting to see whether the pyrimidine bases, which are not attacked in the free state, are affected when they are combined with *d*-ribose. We are not aware that these results have been taken into account in the study of pathological pentosuria; only arabinose is mentioned in connection with these rarities of clinical practice, but as the osazones of ribose and arabinose are identical, it may be asked whether arabinose has actually been identified, or whether the origin of pentosuria should not really be sought for in the metabolism of the pyrimidine nucleosides.

CHAPTER XIII

ALKALOIDS

Definition.—How shall we define the term " alkaloid " ? Here is one definition : Alkaloids are nitrogenous substances with basic properties. This, however, would allow every base in organic chemistry to be included. Yet, if the term be restricted simply to heterocyclic nitrogen compounds, such substances as adrenaline, hordenine, ephedrine, are cut out, and, on the other hand, it then becomes a question whether purine and pyrimidine derivatives, adenine, caffeine, theobromine and the like, should be included. What it comes to is this, that a rigid definition cannot be framed. If the term be applied simply to natural products and closely related synthetic derivatives, its use will be sufficiently limited.

We may therefore say : *An alkaloid is a base of vegetable or animal origin, frequently possessing pronounced and characteristic physiological properties, usually poisonous, forming characteristic precipitates with certain reagents, and containing in its molecule nitrogen atoms which, in the majority of cases, form part of a cyclic structure.*[1]

Extraction.—A knowledge of the general properties of the alkaloids is needed for their proper extraction. Let us say at once that alkaloids exist in the plant as compounds with certain acids, tannins, and so forth. All are set free by treatment with fixed alkalis. Some enter into combination with the alkali, namely, those in whose molecules there are acidic or phenolic groups. Others do not combine ; so a classification into two main groups is immediately possible.

The second of our groups can be subdivided into those alkaloids which are volatile in steam and those which are not. In both these subgroups there are substances soluble in water, others only sparingly soluble or insoluble in ether. Those which are insoluble in ether when once they have been isolated and crystallised are often dissolved just when they are liberated and are kept in solution for some time, but not for long.

Two main principles guiding extraction are based on these general properties. All the alkaloids of the second main group are set free by the action of an alkali, usually lime or ammonia, and extracted by means of an appropriate solvent, that most suitable for laboratory use being ether.

Those of the first group—morphine, cephaeline—are also liberated by treatment with milk of lime, but here the alkaloid cannot be

[1] See also Barger, *Simpler Natural Bases*, pp. 5, 6.—*Tr.*

extracted by ether. The filtered liquor contains the base as a calcium salt, and it may be precipitated by adding the proper amount of an ammonium salt. Morphine is isolated in this way.

Such processes as these are employed on the industrial scale, each firm having its own special methods; yet one may say without fear of contradiction that actually they are merely variations on three themes, viz.:

(1) Application of the methods just described using, according to circumstances, some other solvent (amyl alcohol, heavy oil, benzene, petroleum spirit) instead of ether;

(2) Separation by an acid and precipitation of the acid solution by lime, a method that has the advantage of removing at the start the waste vegetable matter;

(3) Precipitation of the alkaloid from an acid solution by certain special reagents, of which the most important is silicotungstic acid (Bertrand). In this way the base may be separated in a quite tolerably pure condition.

We may summarise the above in the following way:

- (1) Alkaloids not combining with fixed alkalis.
  - Set free by lime or magnesia.
    - Distillation in steam—*Sparteine.*
    - Extraction by an appropriate solvent (ether, amyl alcohol, benzene, etc.)—*Emetine, Atropine.*
  - Set free by acids; filtration.
    - Precipitation of the concentrated liquor by lime, extraction by a solvent—*Quinine.*
    - Precipitation by an alkaloid reagent (sodium silicotungstate)—*Atropine, Cocaine.*
- (2) Alkaloids that combine with lime, soda, etc.
  - Treatment with milk of lime, filtration, precipitation by ammonium chloride—*Morphine.*
  - Treatment with ammonia, extraction with ether, solution in caustic soda, neutralisation of the alkaline solution—*Cephaeline, Cupreine.*

The above methods of extraction yield only crude products. From these pure bases must be separated and isolated as well-crystallised salts. Here it is that difficulties arise and specialists in this field have used all their wits in solving the problems involved.

Derivatives of the Alkaloids.—Once a pure compound has been isolated, a number of derivatives, salts and so forth, must be prepared and their properties carefully noted, so that they may easily be recognised again. The chloroaurates, chloroplatinates and picrates form better defined crystals than other salts, and are the most easily analysed.

Their preparation is not troublesome, yet beginners frequently fail at the first attempt because they usually work with too dilute solutions or with such solvents as hold the double salt in solution. It is best to use concentrated solutions and to employ the calculated amount of the reagent, remembering the formulæ :

$$2R{\cdot}PtCl_4{\cdot}2HCl$$
$$R{\cdot}AuCl_3{\cdot}HCl.$$

If precipitation does not take place, the solution must be concentrated in a vacuum desiccator over strong sulphuric acid.

Another characteristic derivative is the methiodide, the preparation of which will be described later.

Rotatory Power.—Valuable evidences as to the purity of alkaloids may be obtained from polarimetric observations. It must be remembered that frequently both the nature of the solvent and the concentration of the solution are important factors, and so, when one sets out to follow up the purification of an alkaloid with the polarimeter, care must be taken always to work under uniform conditions.

Microscopic examination of the crystalline form is indispensable. In nearly every case alkaloids may be characterised in this way. Either the form of simple salts, such as hydrochlorides or sulphates, may be observed, or microcrystalline precipitates may be prepared on the slide itself.

CHEMICAL PROPERTIES OF THE ALKALOIDS

The first question to present itself in the actual investigation of a base is that of the **degree of saturation** of the molecule. All unsaturated alkaloids, that is to say, those having a double bond in a side chain or in a heterocyclic nucleus, decolourise permanganate in cold dilute acid solution. This preliminary test, which may yield most valuable evidence, is often omitted. Sparteine, for example, although it will not decolourise acid permanganate, was formerly shown in every classical treatise as an unsaturated compound. Chemists beginning work on this alkaloid would then be on the wrong track right at the start.

Hydrogen is taken up at the double bond when the compound is treated with the gas in presence of platinum or palladium. (Example : Hydroquinine.)

Most **alkaloids** are tertiary **bases.** Primary amines are only found in the purine series (Adenine). Secondary bases are, however, less rare ; cicutine, conhydrine, ephedrine, adrenaline may be mentioned.

Physical properties, odour, melting point or boiling point, and so forth, and readiness to undergo oxidation, will already indicate to some extent how the nitrogen is combined, but there are simple methods for definitely settling the question. Besides Hofmann's reaction, which will be discussed later, the reactions with nitrous acid, with

alkaline permanganate, or with acyl chlorides, are employed. Secondary amines yield with nitrous acid nitroso derivatives ; tertiary bases do not react. Acyl chlorides, and phenylisocyanate, also do not interact with tertiary amines, but with primary and secondary amines compounds are formed, *e.g.*, with benzoyl chloride, derivatives of the types :

$$C_6H_5{\cdot}CO{\cdot}NH{\cdot}R,\ C_6H_5{\cdot}CO{\cdot}NR_2,$$

or with phenylisocyanate,

$$C_6H_5{\cdot}NH{\cdot}CO{\cdot}NHR,\ C_6H_5{\cdot}NH{\cdot}CO{\cdot}NR_2.$$

In nearly all saturated alkaloids the third valency of the nitrogen atom is taken up by a methyl group, in fact, this is the only one met with up to the present. The only abnormal saturated bases (from this point of view) are those of the quinine, lupinine, sparteine group ; here the nitrogen atom is attached by its third valency bond to another adjacent carbon atom. We shall see presently how Hofmann's reaction enables such abnormal groupings to be recognised.

Estimation of N-*Methyl Groups.*—Zeisel's method for estimating methyl in methoxy compounds has been so modified by Herzog and Meyer as to render it applicable to substances in which the group $:N{\cdot}CH_3$ is present. The underlying principle is unaltered, but a different apparatus is used, so that the hydriodic acid may act more vigorously. The reaction that takes place is shown by the general equation :

$$R{:}N{-}H \begin{matrix} \diagup CH_3 \\ \\ \diagdown I \end{matrix} = R : NH + CH_3I.$$

The base is heated with hydriodic acid in a special apparatus consisting of two small flasks connected together, the decomposition products being twice subjected to the action of the hydriodic acid before the methyl iodide formed distils over into an alcoholic solution of silver nitrate. The estimation is then completed as in the ordinary Zeisel method.[1]

Groupings containing Oxygen.—Most alkaloids contain oxygen ; this may be combined in various ways, thus :

As part of an **alcoholic hydroxyl** group, primary, secondary or tertiary, in *Quinine, Cinchonine, Tropine, Lupinine, Ephedrine.*
As part of an **ether** grouping, in . . *Quinine.*
,, ,, **ester** ,, ,, . . *Cocaine.*
,, ,, a **phenolic hydroxyl** . . *Morphine.*
,, ,, **carboxyl** group. . . *Ecgonine, benzoyl-ecgonine.*
,, ,, **betaine** grouping . . *Trigonelline.*

Characteristic for **hydroxyl** groups is the reaction with acyl chlorides

[1] See *Chemical Society's Abstracts*, 1895, ii., 296 ; Meyer and Tingle, *Determination of Radicles in Organic Compounds*, 3rd ed., 1908, p. 114 (Wiley, New York) ; Meyer, *Analyse und Konstitutionsermittlung organischer Verbindungen*, 1916, p. 839 (Springer, Berlin).

or anhydrides. When the substance is a free base it is treated with the reagent in warm benzene solution, whilst if it be a hydrochloride, the reaction is carried out in weakly alkaline aqueous suspension, *i.e.*, in the cold with a small excess of 4% caustic soda, as prescribed by Schotten and Baumann.

When hydroxyl derivatives are treated with *dehydrating agents* unsaturated compounds are formed. The best and most widely used reagent for this purpose is a mixture of concentrated sulphuric acid and glacial acetic acid in the proportion of 2 to 1. The reaction mixture is heated at 180° to 200° for ten or twelve hours.

The manner in which *oxidation* takes place will show whether the alcohol is primary, secondary, or tertiary.

Methoxyl groups are characterised and estimated by Zeisel's method. This consists in heating the base with hydriodic acid; methyl iodide is evolved, and when passed into alcoholic silver nitrate gives silver iodide. Scission of a methoxyl group to give the parent hydroxyl derivative may be easily carried out by heating the base with strong hydrochloric acid (or better, hydrobromic acid) in a sealed tube. This has been done with *e.g.*, hydroquinine, emetine.

Alkaloids containing **acidic groupings** are, in general, neutral to litmus like the simple amino acids. They usually form salts with bases and esters with alcohols. The esters are obtained by boiling an alcoholic solution of the alkaloid saturated with hydrogen chloride. The *copper salt* is nearly always a well-crystallised and characteristic derivative, and it, for example, may be prepared by the following simple, but little known, method.

> Neutralise exactly with baryta water a solution of copper sulphate; add the acid in question to the mixture and warm for a few minutes on the water-bath. Filter while hot. The salt will crystallise out when the solution cools or if it be evaporated down. This method has the advantage of bringing the acid in contact with copper oxide in a very finely divided state.

Action of Reagents.—The reactions by which these functional groupings are recognised give only a vague idea of the actual constitution of the alkaloid. Evidently further investigation will need special methods for each substance, and we can hardly consider all of them here. Nevertheless, there are two methods of general application, namely, oxidation and that due to Hofmann.

All oxidising agents do not behave in the same way, and, in any given case, it is impossible to say, *a priori*, which will be the best. Generally, chromic acid is used to oxidise side chains or to disrupt a molecule at a point where there is a double bond, a hydroxyl group, or the like. Permanganate serves in replacing the methyl of a methylamino group by hydrogen, or in oxidising a double bond to form a dihydric alcohol. It acts differently according as the solution is acid or alkaline. Nitric acid is less frequently used. Hydrogen peroxide

has given interesting results in Wolfenstein's hands,[1] but it is not generally useful and produces some odd effects.

Three classical examples of oxidation processes, which will serve as models in almost every case, will be described in detail.

(1) *Oxidation of a N-methyl group. Production of a secondary base by the action of alkaline permanganate solution.*

Conversion of Tropine into Tropigenine (Willstätter).

10 gm. tropine and 5 gm. potassium hydroxide are dissolved in a litre of water, the solution is cooled to 0° and continuously agitated while 22·5 gm. potassium permanganate, also dissolved in a litre of water, is slowly added. The reaction being complete, the filtered liquor is acidified with hydrochloric acid and evaporated to dryness. To the residue a little water and a great excess of solid caustic potash are added and the mixture is extracted with 20 to 30 litres of ether. The ethereal extract is concentrated until its volume is about 220 c.c., and it is then cooled in ice. The tropigenine crystallises out.

(2) *Regulated oxidation of a secondary alcohol. Conversion into a ketone.*

Conversion of Tropine into Tropinone (Willstätter).

To a solution of tropine (25 gm.) in acetic acid (100 gm.), one of chromic acid (12 gm.) is added, drop by drop, agitating meanwhile and keeping the temperature at 60°. At the end the solution is heated for a few minutes on the water-bath. Caustic potash in excess is added and the mixture steam-distilled. Only tropinone passes over with the steam. Yield: 90%.

(3) *Oxidation of an alcohol to an acid.*

Generally, when an alcoholic group has been identified in a base, its nature, whether primary, secondary, or tertiary, is unknown before the oxidation is carried out.

If the group be primary, the product of the oxidation will be an acid containing the same number of carbon atoms as the original alcohol.

If it be secondary, regulated oxidation will yield a ketone, whilst more energetic oxidation will produce a dicarboxylic acid if the hydroxy group be attached to a ring carbon atom, or a monocarboxylic acid if it be a substituent in a straight chain. In the latter case, the place at which oxidation has taken effect will be indicated by the number of carbon atoms that have disappeared.

When lupinine is oxidised lupinic acid is produced; this contains the same number of carbon atoms and only one carboxyl group. Lupinine is therefore a primary alcohol.

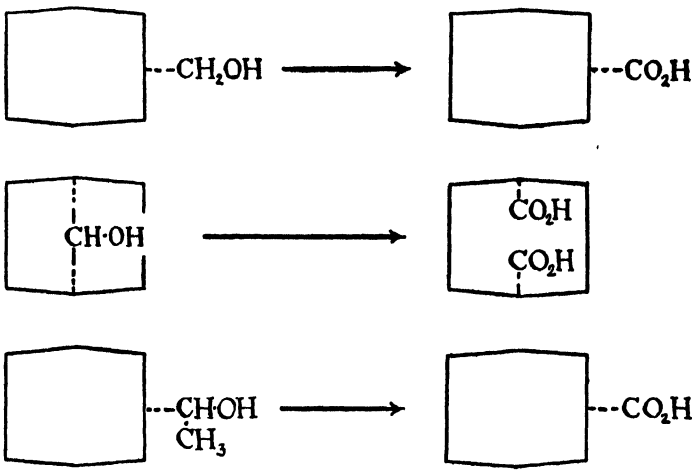
When atropine is oxidised a *di*-carboxylic acid containing the same

[1] See *Chem. Soc. Abstracts*, 1895, i., 479.

number of carbon atoms is obtained: therefore, in atropine the hydroxy group is secondary and is attached to a carbon atom forming part of a cyclic structure.

When conhydrine is oxidised pipecolinic acid is formed. This has one carbon atom less than conhydrine. The latter, therefore, is a secondary alcohol with the hydroxy group in the side chain and attached to the second carbon atom from the end.

These facts may be represented in this way :—



Thus, evidently, the way in which oxidation takes place shows to what class of alcohol the compound in question belongs.

Primary alcohols begin to undergo oxidation even in the cold, and to complete the oxidation enough chromic acid to furnish two atomic proportions of oxygen will suffice.

Oxidation of Lupinine to Lupinic Acid (Willstätter and Fourneau).

50 gm. lupinine is dissolved in 100 c.c. water and 15 gm. sulphuric acid. To the cold solution is added a mixture of 40 gm. chromic acid, 60 gm. sulphuric acid and 800 c.c. water (these quantities correspond with two atomic proportions of oxygen). The oxidation is almost complete in the cold, the temperature rising to 60°. The mixture is boiled for half an hour. When the liquid has a clear green colour with no brownish cast, and addition of a little more chromic acid changes the colour to a brownish yellow, the oxidation is finished. Now to isolate the acid is a somewhat lengthy operation. First, any excess of chromic acid is reduced by bubbling sulphur dioxide into the solution, then excess of the latter is boiled off. The solution is made alkaline by adding ammonia, filtered and evaporated to dryness. The residue, containing the acid and ammonium salts, is extracted with alcohol, the impurities being left behind. The alcoholic solution is

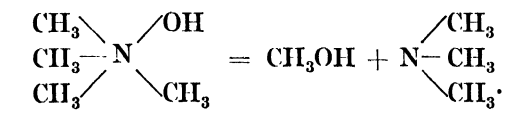
evaporated to dryness and the ammonium salt so obtained dissolved in a large amount of water; an excess of barium hydroxide solution is added, the ammonia is driven out by blowing steam through, and the excess baryta is precipitated by passing carbon dioxide. To the filtered liquor the exactly necessary amount of sulphuric acid is added and the filtered solution of the free acid is evaporated to dryness.

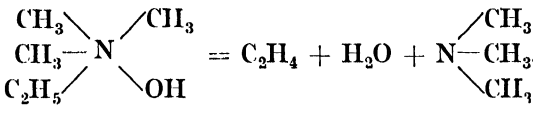
Oxidation of a Double Bond.—An example, in which the acid obtained is sparingly soluble in water, namely, the preparation of quitenine by oxidising quinine, is described in the practical part of the book.

Besides the classical oxidising agents, there are many others that are useful. As an example of an oxidation by nitric acid, that of nicotine (Weidel) may be mentioned. Industrially, electrolytic oxidation has given valuable results in certain cases; the firm of Merck has taken out a patent for converting tropine into tropinone by this means.

Hofmann's Reaction.[1]—We will now turn our attention to this celebrated reaction, the applications of which have been so fruitful and the utility so great that one is justified in saying that, without its help, little progress would have been made in alkaloid chemistry.

The underlying principle is shown by these two equations:



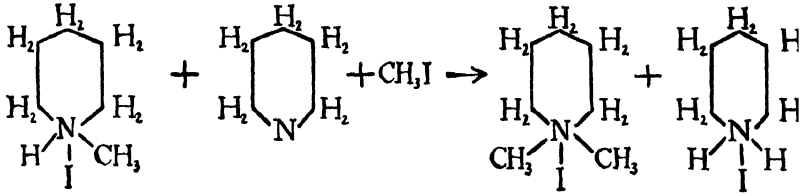


If a quaternary ammonium base containing only methyl groups be heated, it breaks up into trimethylamine and methyl alcohol. But if one or more radicals are homologous to methyl, an unsaturated hydrocarbon, water, and a tertiary amine, containing the remaining alkyl radicals, are obtained. The methyl groups always remain attached to the nitrogen atom.

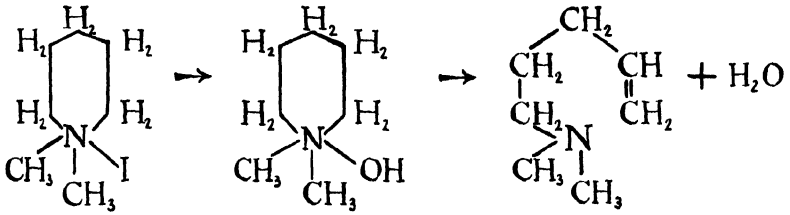
In the alkaloid series, the reaction takes a somewhat different course, because, usually, two at least of the nitrogen valencies are taken up by the two ends of one and the same carbon chain. The classical example of piperidine, studied by Hofmann himself, will illustrate the point.

Piperidine is a secondary base. When it is treated with methyl iodide piperidine methiodide, *i.e.*, N-methylpiperidine hydriodide, is formed, then, with more of the base, piperidine hydriodide and N-methylpiperidine methiodide:

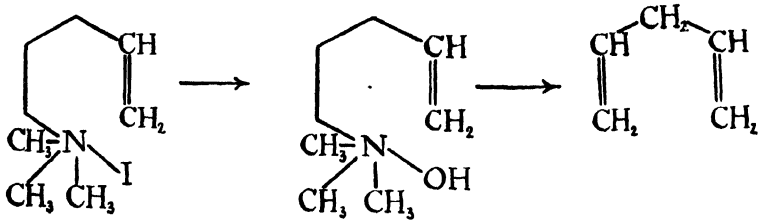
[1] The so-called *exhaustive methylation*.



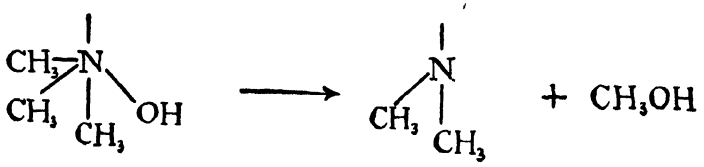
When the latter is treated with silver oxide, the corresponding hydroxide is obtained, and when this is heated it decomposes, water is given off and an unsaturated tertiary amine formed :



This base is what Hofmann incorrectly called dimethylpiperidine. Now, being still a tertiary amine, it will take up methyl iodide, and when the quaternary ammonium iodide formed is heated with potash it breaks up into trimethylamine and a doubly unsaturated hydrocarbon, namely, piperylene :

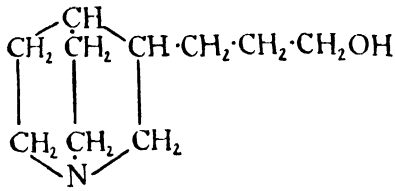


Instead of making the hydroxide separately by causing the salt to interact with moist silver oxide, this time it is heated directly with caustic potash. It is, in fact, most often necessary (as the union between nitrogen and carbon must be broken), to have recourse to this more drastic treatment with caustic alkali. It should be added that the reaction frequently takes a less straightforward course ; it is complicated by the reverse change to the original base, the quaternary hydroxide losing alcohol instead of water, thus :

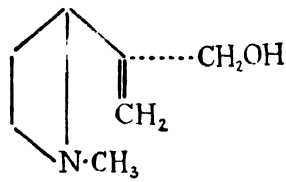


Lastly, the formation of an unsaturated hydrocarbon by this splitting off of the elements of water may take place in so many different ways that it is a rare event for one not to obtain several isomerides at once, particularly when alkaloids more complex than piperidine are undergoing treatment.

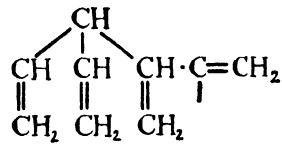
At times the nitrogen atom may form part of a double ring, as in quinine and lupinine. Here Hofmann's reaction takes an abnormal course, because, twice running, a tertiary base is obtained. Lupinine, for example, contains the ring structure :



Here the result of the first treatment with methyl iodide is to disrupt one of the rings, so that again we have the piperidine structure :



The final product is a hydrocarbon of this kind :



Obviously the double bonds may be variously arranged and so a multiplicity of isomerides may be obtained.

Hofmann himself did not discover the true explanation of the reaction ; it was Ladenburg who put out a satisfactory theory, but into the details of the discussions that took place we cannot enter now.

The actual **manipulation** is as follows :

The alkaloid is first treated in a test tube with methyl iodide dissolved in a little methyl alcohol ; whether the reaction is vigorous or not may thus be noted. Sometimes it is very energetic, and when larger quantities of substance are handled it becomes necessary to dilute the solution. Only rarely is heat required, and nearly always the methiodide crystallises out. When the solution is neutral the reaction is complete. The iodide is separated, dissolved in water and a small excess of moist silver oxide is stirred in. The filtered liquor is

evaporated down *in vacuo* on the water-bath. Water passes over, then, when nothing more distils, the residue is heated in an oil-bath. Suddenly the hydroxide decomposes, water distils off, followed by the base. During the distillation of the aqueous solution much froth is formed; this can be kept under control by passing ether vapour over the surface of the liquid.

The base obtained is treated afresh with methyl iodide, silver oxide, and so forth. Sometimes, as has already been pointed out, the last distillation is made over caustic potash. The trimethylamine is collected in dilute hydrochloric acid.

CHAPTER XIV

GENERAL REMARKS ON PHARMACEUTICAL PRODUCTS

In the preceding chapters we have given a more or less detailed description of most groups of medicaments, but some have been left out of account (iodine and silver medicaments, tannin derivatives, anthelmintics, and so forth), either because the particular group was too small to have a chapter to itself; or because it has been found impossible to trace a relationship between constitution and physiological action for the compounds in question; or because we thought that the chemical processes involved in their preparation were not interesting enough from a scientific point of view; or, lastly, because their preparation, in its details, could not be included within the limits we had fixed for ourselves.

So as to fill this gap we now propose to devote a short chapter to a sort of synthesis of the essentials of our knowledge of medicaments and of the relationships that exist between their chemical constitution and their physiological action.

GENERAL CONSIDERATIONS

Little by little precision emerges from the formidable accumulation of observations, the simple recital of which fills a voluminous work (Fränkel's *Arzneimittelsynthese*). Naturally only the more important conclusions can be mentioned here.

I. Open-chain **hydrocarbons** are less toxic than either benzenoid or hydroaromatic hydrocarbons (*cyclo*hexane), and the last named are less toxic than benzene or its homologues.

II. In the **acyclic** series, compounds having unsaturated linkages are more active than the corresponding saturated substances, *e.g.*,

Allyl alcohol—Propyl alcohol.

Acrolein—Propionaldehyde.

But the contrary holds if the compounds be amines, and this difference is still more marked when the cyclic bases are in question.

Allylamine is less toxic than *propylamine*, *pyridine* less than *piperidine*, *naphthylamine* less than *hydronaphthylamine*.

III. Already we see, and later we shall again notice, other factors coming into account, such as particularly the **position of the ethylene linkage** in relation to the other groups or to the nucleus. *Methylvinylamine*, for example, is more poisonous than *allylamine*, *safrole* more than iso*safrole*.

The influence of the double bond depends, therefore, on its position. This must always be kept in mind and, moreover, it must not be

forgotten that derivatives of vinyl alcohol, $CH_2 : CH \cdot OH$, are particularly active.

IV. Certain very **toxic groupings** (the nitrile radical, the nitro group) enter only infrequently into the composition of medicaments (hydrocyanic acid, trinitroglycerine). The remaining classes of compounds may, as a quite general rule, be arranged in the following descending order of toxic potency :

Lactones of the aromatic series (Cantharidine).
Aldehydes, Ketones.
Amines, Heterocyclic bases.
Alcohols, Phenols.
Esters.
Acids.

We must leave the alkaloids out of account, because they do not come under any law of this kind. Why is arecoline, for example, an ester of an amino acid, more toxic than ecgonine methyl ester ? Why is quinine so feebly toxic ? Its molecule contains a quinoline nucleus, a fused piperidine ring, and an unsaturated side chain. We are a long way from being able to answer such questions as these.

V. The introduction of an **acidic group** into any molecule brings about a considerable decrease in physiological activity.

Ethylamine—Glycine.
Tropine—Ecgonine.
Benzene—Benzoic acid.
Phenol—Salicylic acid.

VI. But if the acid be **esterified** some of the activity is regained . . . not always, however, in the same sense.

VII. In some cases **alkylation** of phenols produces a less toxic compound, *e.g.*,

Phenol—Anisole.
Catechol—Guaiacol,

but in others a more toxic product.

VIII. Usually alkylation of amines augments the toxicity. This is particularly the case with Atoxyl and its derivatives (Dystherapeutic influence of the methyl group—Ehrlich).

IX. Amines are rendered considerably less toxic by **acylation** (this has generally meant acetylation).

Aniline—Acetanilide.
Phenetidine—Phenacetine.

X. On the other hand, acylation of a hydroxy group augments the therapeutic activity of the compound, or sometimes completely alters its character, particularly if the molecule contains an amino group.

Ecgonine methyl ester—Cocaine.
Pseudotropine—Tropacocaine.
Salicylic acid—Aspirin.
Choline—Acetylcholine.

Certain acids appear to have a specific influence, viz.,

Benzoic acid in local anæsthetics ;

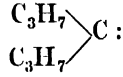
Valeric acid in sedatives.

Acetic acid in all other cases where it is a question of reinforcing an existing property or of diminishing toxicity.

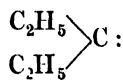
A particular case of acylation is that of the urethanes (carbamic esters), all of which are more or less active hypnotics.

XI. No conclusion of any kind can be drawn as to the influence of the **molecular weight.** In the aliphatic series, where nearly all compounds, not amines, are hypnotics, activity increases with rise in a molecular weight until about the C_6 term is reached.

XII. Ramification of the carbon chain augments the hypnotic potency of the amides, ureas, alcohols, and so forth, of the aliphatic series. The maximum activity seems to be possessed by compounds containing the groupings :



Nevertheless, among urethanes of secondary alcohols, which have been extensively investigated, the grouping (*a*) :

(*a*)
(*b*) 

seems to be more active than the grouping (*b*).

A typical example, exhibiting the influence of molecular weight in a striking manner, is afforded by the esters of choline. Hæmolytic activity appears when the esterifying acid is the C_{15} compound, and increases as the number of carbon atoms rises from 15 to 17, so that stearylcholine is to be placed among the most powerful hæmolytics known (Delezenne, Fourneau).

XIII. The influence of the **position** of the substituent groups may be very great, but an exact rule cannot be formulated.

In the aromatic series no regularity is shown : of the nitrophenols the *para* isomeride is the most toxic, but *o*-nitrobenzaldehyde is more toxic than the *para* compound, and the same may be said of the various phenetidines and phenylene diamines.

Among arylaliphatic amines, of which benzylamine is typical, the β-position with respect to the ring is particularly important. That all β-phenylethylamine derivatives, provided the amino group is primary or secondary, are sympathomimetics has already been pointed out. Tyramine and adrenaline belong to this class.

But not only position isomerism and optical isomerism, stereoisomerism in the other sense also may have an influence, as the example of tropine and *pseudo*-tropine shows. In this connection we may again refer to the difference between *d*-adrenaline and

l-adrenaline, to the effect of stereoisomerism on the taste of amino acids, sugars, and so forth.

XIV. In a general way, methane derivatives are hypnotics, benzene derivatives are antipyretics.

XV. Most reactions of chemical compounds are indicated by their effect on litmus: the beginnings of chemotherapeutics may be found in Ehrlich's admirable investigations on the fixation of acid or basic dyestuffs by the nerve cells.

XVI. The few conclusions as to the influence of chemical constitution on physiological action that seem to be well established are the following:

Benzoylation of amino alcohols always give rise to local anæsthetics.

The β-position in arylaliphatic bases (β-phenylethylamine) is of outstanding importance.

All quaternary ammonium compounds have curari-like properties.

The ethyl group, and particularly the diethylmethylene group (Veronal, Sulphonal, Adaline, etc.), possesses "hypnotic" characteristics.

The introduction of an acidic grouping produces a less toxic compound; acylation of amines has the same effect.

ELIMINATION OF MEDICAMENTS BY THE ORGANISM[1]

The phenomena that take place in the organism are exactly like those which we can bring about in the laboratory, but with this difference, that the reactions occurring in the living cell are infinitely more delicate, diverse and unexpected than those taking place in our flasks and beakers; this is due to a multiplicity of ferments, aided by physical conditions (high osmotic pressure), of which we know very little.

All the same, everything that happens is the result of oxidations, reductions and condensations, with or without separation of the elements of water.

Oxidation.—Fatty acids apparently suffer oxidation at the β-carbon atom, a hydroxy acid being the first product; further oxidation produces a ketonic acid, and eventually the carbon chain is broken, a new carboxyl group being formed. Oxidation of a fatty acid involves, therefore, the removal of two carbon atoms at once.

Amino acids, on the other hand, are usually oxidised to α-keto acids, the amino group being eliminated. Subsequent reduction produces hydroxy acids. In every instance, the ultimate product of the oxidation is an acid with one carbon atom less than the original amino acid. Sometimes, however, more complicated changes take place, glycocoll (glycine) and alanine, for example, being converted into urea.

If an alkyl radical be introduced into the amino group (sarcosine),

[1] "Excretion" in biochemical language.—*Tr.*

the acid is much more, even quite, stable. Acylation (benzoylation) of the amino group has the same effect.

Noteworthy differences in behaviour are observed when the amino group occupies other positions, or according as the organism attacking the amino acid is a yeast, a bacterium, or a mammal, and so on.

Primary and secondary alcohols are readily oxidised, but not tertiary alcohols, nor halogen derivatives. Isopral and trichloroethyl alcohol are eliminated unchanged, in combination with glycuronic acid.

Some ketones pass through the organism without being attacked to any great extent (methyl ethyl ketone); others, such as diethyl ketone, are completely burnt up.

The nucleus in cyclic compounds is usually stable, but side chains are attacked in the same way as aliphatic compounds. Moreover, even if the nucleus is not completely oxidised, it may be subject to some modification. One of the most interesting instances of this taking place is the conversion of aniline into *p*-aminophenol.

Some aromatic acids, such as phenylglycollic acid, would seem to be easily destroyed, but, on the contrary, they are very resistant. Acids with long side chains undergo "β-oxidation" as in the aliphatic series.

Unsaturated acids, *e.g.*, cinnamic acid, reappear as hippuric acid, the intermediate stage being benzoic acid.

Amines are not easily attacked and most often pass through the body unaltered.

Amino acids with the amino group attached to the α-carbon atom, phenylalanine and tyrosine, for example, are completely destroyed. But it is interesting to note that *ortho* substitution, as against *para* (as in tyrosine), renders the molecule less readily oxidised. Introduction of chlorine has the same effect, chlorophenylalanine being hardly attacked. The presence of methyl groups in the nucleus makes no difference.

Benzene itself is oxidised to some extent, and the organism can accustom itself to the work.

Briefly, in parts of the organism, energetic oxidation takes place, and it depends on the constitution of any particular compound whether it be directed towards or away from these centres of oxidation. Furthermore, the oxidation process usually takes place in a predetermined fashion, the molecule is always attacked in the same place, and if this place be protected by substitution, the oxidation does not come to pass.

Reduction.—A well-known instance of reduction by the organism is that of picric acid, which is converted into picramic acid (dinitroaminophenol). Other examples are those of the reduction of certain colouring matters to the corresponding leuco-compounds, the reduction being localised and taking place only in certain cells. This reduction is inhibited by hypnotics.

Chloral yields the corresponding alcohol. According to Ehrlich, arsinic acids, *e.g.*, atoxyl, only act after reduction has taken place, either to the arsenious oxide or to the arseno derivative.

Elimination.—Foreign substances are eliminated from the organism, whether they have been oxidised or reduced, most frequently in combination with other compounds, of which the best known are *glycuronic acid*, $CHO\cdot(CH\cdot OH)_4\cdot CO_2H$, *glycine* (*glycocoll*) [hippuric acid], and *sulphuric acid* [ethereal sulphates].

Essentially it is by converting them into acidic compounds, which are ejected as salts, that the organism rids itself of harmful substances. But there are many other ways in which poisons can be made innocuous: a typical example is the conversion of nitriles into thiocyanates (which apparently takes place at the expense of the amino acid cystine). It has been shown by Chelles that hydrocyanic acid is eliminated in the form of thiocyanates.

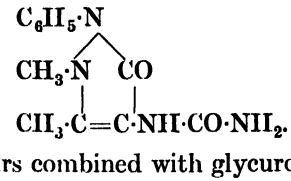
Some other cases may be mentioned:

Phenol is eliminated as potassium phenyl sulphate, $C_6H_5O\cdot SO_3K$; benzoic acid as hippuric acid.

Vanillin is excreted in combination with glycuronic acid after conversion to the corresponding acid:

$$CH_3O\cdot C_6H_3(CO_2H)O\cdot CH\cdot(CH\cdot OH)_2\cdot CH\cdot CH(OH)\cdot CO_2H.$$
$$\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\quad |_____O__|$$

Pyramidon is partly transformed into rubazonic acid, being thus demethylated; in part, also, it forms antipyrylurea:



Antipyrine reappears combined with glycuronic acid, and at times as oxyantipyrine; chloral and tertiary alcohols also form compounds with glycuronic acid.

The organism can avail itself of many other means of getting rid of foreign substances, but those described will illustrate sufficiently the principal ways in which elimination takes place.

PART II—PRACTICAL

SETTING UP APPARATUS—RECOMMENDATIONS TO BEGINNERS

MOST operations in organic chemistry, and indeed all those described in this book, may be carried out in quite simple apparatus. A great variety is not needed. That in frequent use will be described concisely here so as to avoid repetition later.

Manipulation in organic chemistry is chiefly **Distillation per descensum,** and **Distillation per ascensum,** that is to say, with reflux of the condensed vapour; then there are operations needing **Agitation** (shaking or stirring), **Heating under pressure** (sealed tubes or autoclaves), and others of less importance.

Distillation per descensum.—There are three cases to consider, (1) a liquid is to be separated from a solid dissolved in it; (2) two or more liquids of different boiling points are to be separated; and (3) either (1) or (2) must be carried out under diminished pressure.

(1) The three components of an apparatus for **simple distillation** are the still (a flask), the condenser and the receiver. The **flask** is connected with the condenser by means of a bent glass tube fitted with corks or rubber stoppers. The receiver is attached to the lower end of the condenser. Little need be said about the flask except that it should be chosen with care: the neck should be truly circular; the walls uniformly thick, and that they are so may be proved by gently tapping all round with the finger nail.

The **connecting tube** should be bent to a smooth curve and have its ends rounded in the blowpipe flame. Good corks should be selected of such a size that, after being softened by squeezing, they may be pushed into the flask to about a quarter their length. The hole should be drilled with a proper cork-borer of suitable size, first from one end and then from the other. Nothing is more objectionable to a careful chemist than to see a glass tube passing through a cork all askew. The hole should be uniform; it is best to make it at first on the small side and then widen it with a round file until it will just fit the tube tightly.

The **condenser** for liquids more volatile than water should be of either the double surface or Vigreux's pattern; the latter has sharp indentations along the inner tube.

The condenser should follow exactly the line of the tube entering it. Artistry should be shown in practical chemistry. Apparatus

should be fitted up carefully and neatly, even elegantly; the time thus spent will not be wasted.

To the lower end of the condenser is fitted an adapter so as to

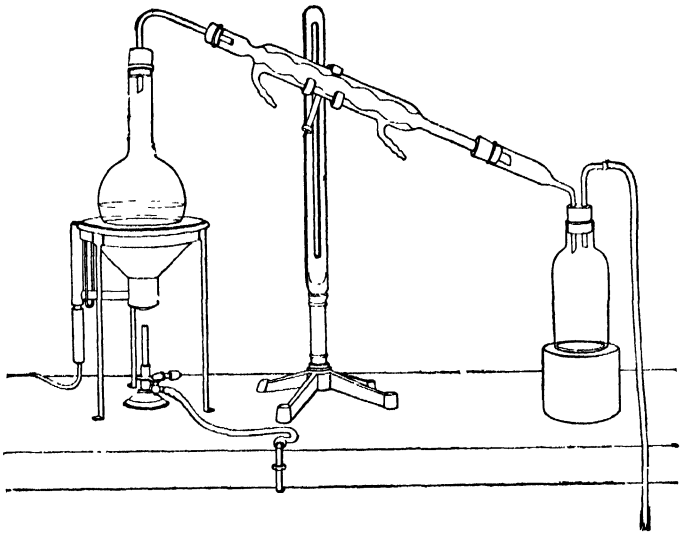


FIG. 1.

connect it with the bottle or flask which serves as receiver. The cork of this flask is bored with two holes, through one of which passes the adapter, while the other carries a bent glass tube ending

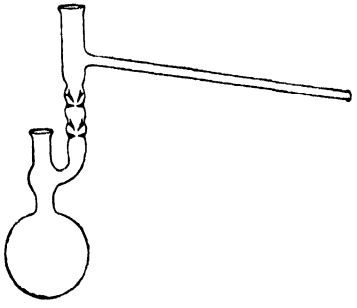


FIG. 2.

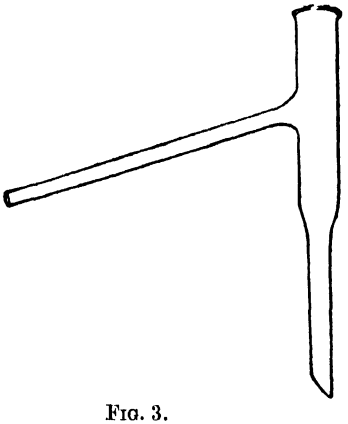


FIG. 3.

in a piece of rubber tube long enough to reach below the level of the bench. In this way, provided the distillation flask does not break, all danger of a fire in distilling a very inflammable liquid is prevented (Fig. 1).

The liquid to be distilled is introduced only after the apparatus has

been satisfactorily fixed up. Then, *before heating*, it is well if a few fragments of unglazed earthenware or broken brick or chips of wood are dropped in to promote steady boiling.

(2) **Fractional distillation,** the theory of which cannot be entered into here, usually involves the use of special flasks. That depicted (Fig. 2) is a convenient form.

The general arrangement is much the same as in the preceding case (Fig. 1), and further description is unnecessary. If the substance to be distilled boils at a temperature above about 140° it is safer to replace the condenser by a wide glass tube with thin walls. The flask should be warmed up gradually if an oil-bath be not used for heating, and the bottom should be wiped with filter paper if condensed moisture

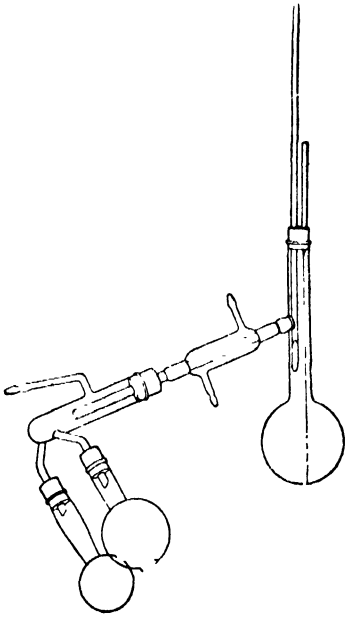


FIG. 4.

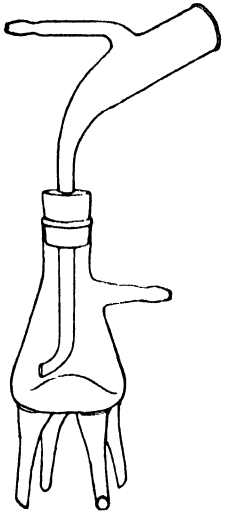


FIG. 5.

from the flame collects. To note down the tare of the receiver should not be forgotten. In place of the somewhat expensive special flasks, distilling heads, such as that figured (Fig. 3), or a fractionating column of Vigreux's or some other pattern, may be used.[1]

(3) **Vacuum Distillation.**—The simplest apparatus consists of two distilling flasks fitted together so that the side tube of the one goes right down the neck of the other, which serves as receiver. It is essential that all joints be quite air-tight. During the distillation a

[1] The English student should consult *Distillation Principles and Processes*, by S. Young and collaborators (London, 1922). Of all branches of laboratory manipulation in organic chemistry, fractional distillation is usually the most unintelligently carried out, shop-made apparatus of traditional, complicated (but ridiculous), design being used.—*Tr.*

stream of minute bubbles of air should pass through the boiling liquid; these are introduced by means of a glass tube drawn out to an extremely

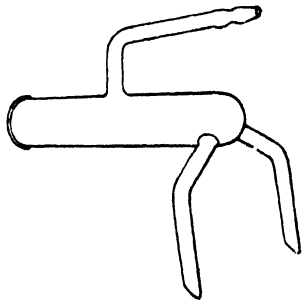


FIG. 6.

fine capillary (Fig. 4). The flask may be heated either directly or in an oil-bath. When it is heated directly the burner must be held in the hand and the flame moved round and round the flask about the

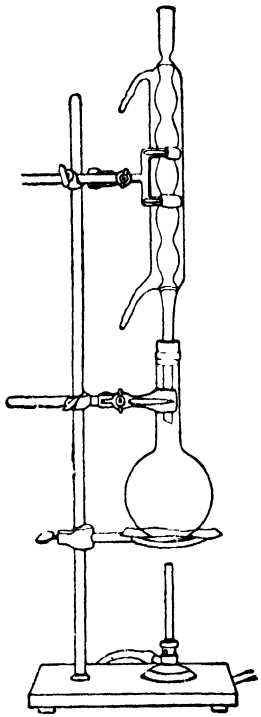


FIG. 7.

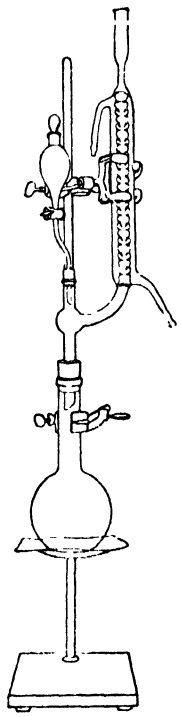


FIG. 8.

level of the liquid; it must never be placed immediately beneath the flask.

One frequently needs to take off several fractions without breaking

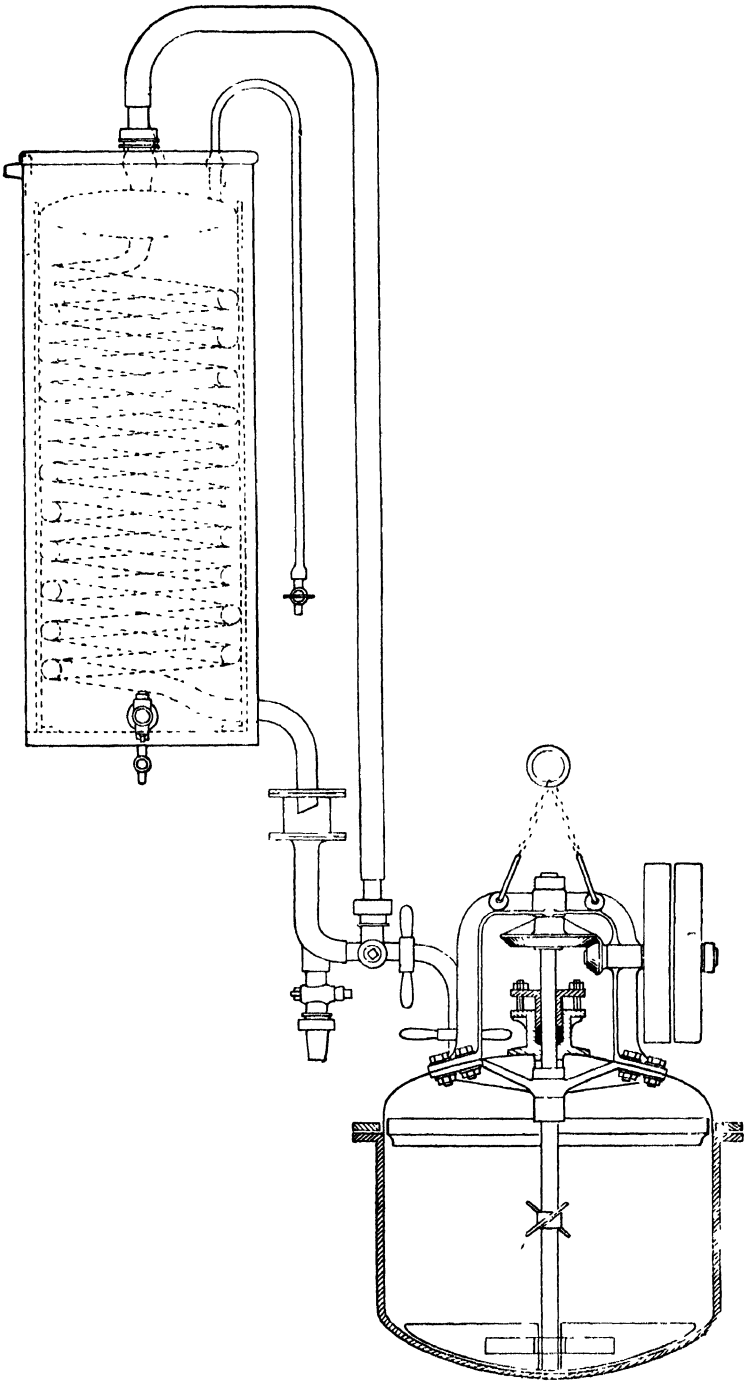


FIG 9.—*Modèle Deroy.*

the vacuum. For this purpose a variety of devices are available, one of which is depicted (Fig. 5). Bertrand's tube is another alternative.

If it suffices to separate two fractions only, a useful piece of apparatus is that shown in Fig. 4, and separately in Fig. 6.

Distillation per ascensum or under Reflux.—In distilling under reflux the vapour from the boiling liquid is condensed by a condenser fitted above the flask and is constantly returned to the latter (Fig. 7). This arrangement of apparatus is in everyday use in an organic laboratory, being, in fact, more frequently used than any other.

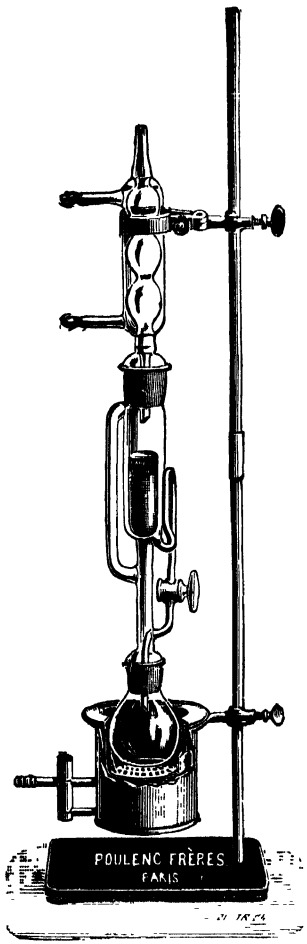


Fig. 10.

Maybe it is necessary to introduce a liquid or a gas whilst reflux distillation is proceeding. In this case the cork carrying the condenser is pierced with a second hole to take either a leading tube for gas or the stem of a dropping funnel. A better plan is to use a specially designed apparatus, such as that shown (Fig. 8). Many processes can be carried out in an arrangement of this kind.

On an industrial scale, apparatus for stirring the liquid, introduction of liquids or solids, reflux distillation and distillation *per descensum* using the same condenser, are all combined in the one plant (Fig. 9).

Distillation under reflux may be adapted to the extraction of soluble components of powdered substances, or of active principles from vegetable or animal matter. Extraction takes place in a special apparatus set between the flask and the condenser (Soxhlet, Vigreux). The powder, contained in a thimble of filter-paper, is thus continuously subjected to the action of fresh solvent, and the extract is drawn off automatically at regular intervals by means of a siphon and returned to the flask (Fig. 10).

Distillation in Steam.—A pure substance may often be separated from a mixture by taking advantage of the property, possessed by many compounds, of being volatile in steam (*e.g.*, *o*-nitrophenol). Fig. 11 shows the arrangement of apparatus used. The flask should be set considerably aslant so as to prevent drops of the violently agitated liquid from being splashed up and carried into the condenser.

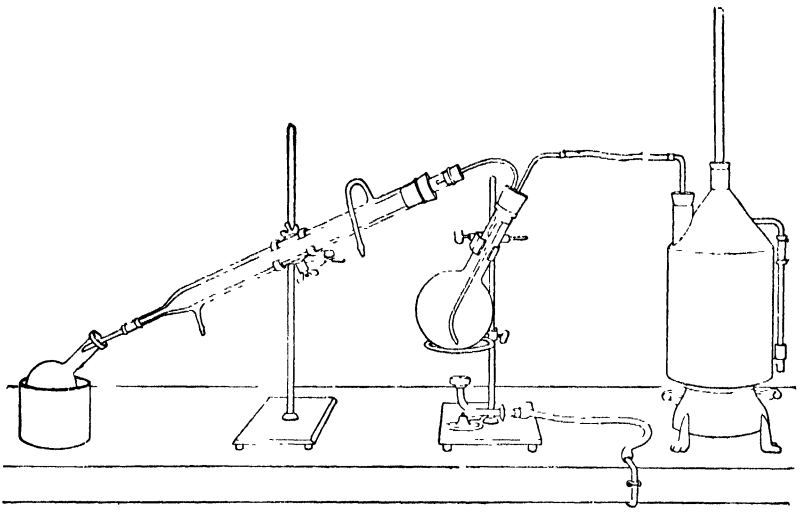


FIG. 11.

For less volatile substances the steam may be superheated by passing it through a hot copper worm.

Purification of Solid Substances.—As volatile compounds are best purified by distillation, so solid compounds are best separated from one another and purified by crystallisation.

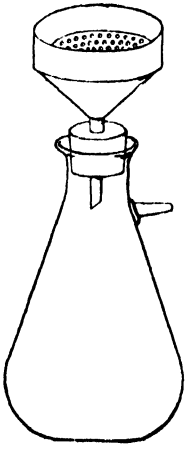


FIG. 12.

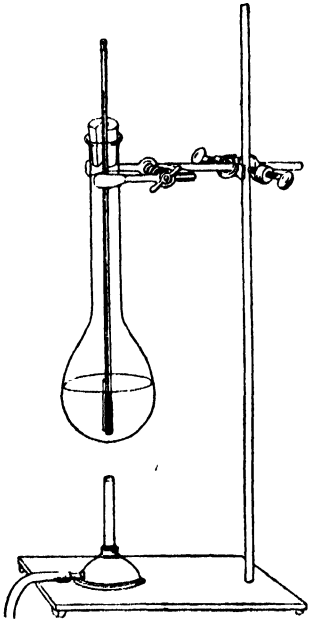


FIG. 13.

With any given product one proceeds as follows :

A small quantity (0·05 to 0·1 gm.) of the substance to be purified is put in a test tube and a few drops of a suitable solvent added. The solid is crushed and stirred up with a rod. If it does not dissolve to any appreciable extent the tube is gently warmed, and then, if it be not completely dissolved, more solvent is gradually added until complete solution results. Similar tests are made with other solvents. When all the test tubes have cooled, one looks for the biggest crop of crystals ; and, by examining with a hand-lens, one also notes in

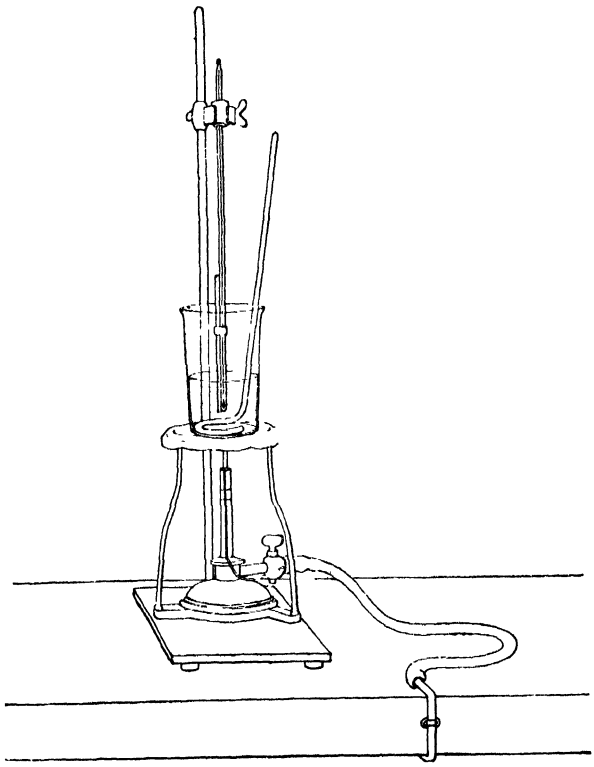


Fig. 14.

which tube the crystals appear most homogeneous. From these observations a decision is made as to the solvent to employ in purifying the bulk of the product.

As far as possible, the use of mixtures of solvents should be avoided ; nevertheless, it sometimes happens that crystallisation, or even any separation at all, will only take place from a mixture.

The bulk of the product being satisfactorily dissolved, the solution is filtered through a pleated filter-paper into a beaker. When crystallisation appears to be complete the recrystallised material is filtered off with the aid of a pump (an excellent glass water-pump is that of

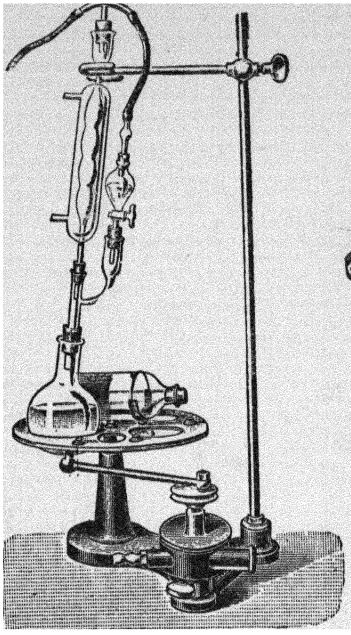


FIG. 15.

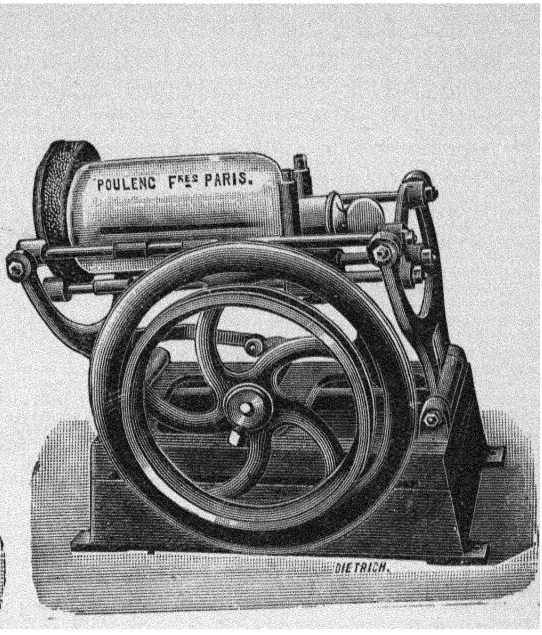


FIG. 16.

the Villiers pattern), in a special funnel (Buchner funnel, see Fig. 12), pressed gently and washed with small quantities of fresh solvent.

Melting Points.—Various arrangements of apparatus may be used in determining melting points. Those depicted are the simplest of all (Figs. 13 and 14).

When the substance has fused, the bath should be allowed to cool

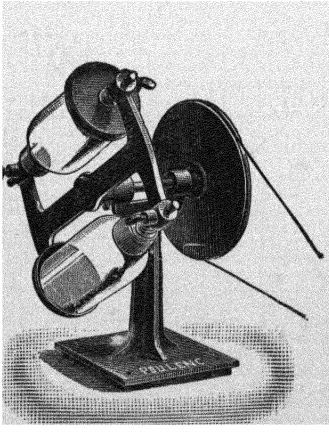


FIG. 17.

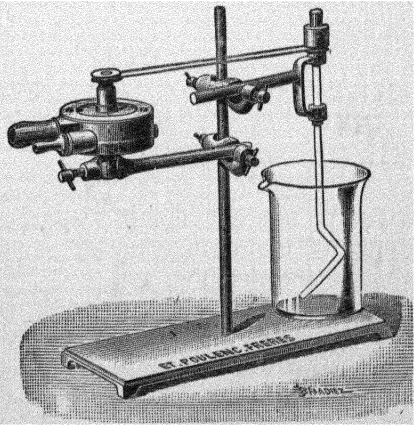


FIG. 18.

a little (2° or 3°), and then another capillary tube containing a fresh sample of the substance to be tested should be introduced and a second observation made. In this way one may avoid erroneous results; many compounds melt at a lower temperature than they should do if they are heated up slowly. The *bloc Maquenne* is a convenient piece of apparatus for use with substances of high melting point or for carrying out a number of determinations one after the other.

Agitation.—If a flask is to be shaken during any process, the best

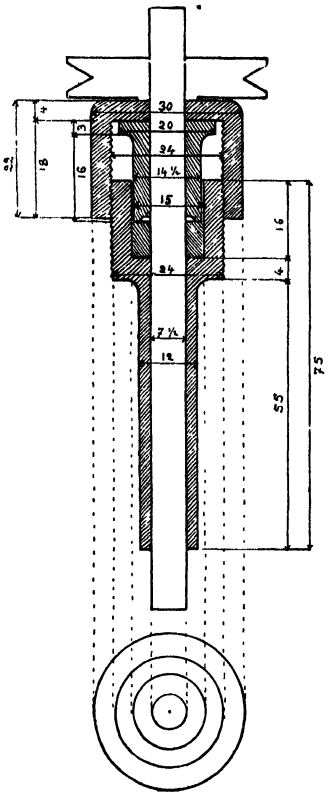


FIG. 19.

machine to use is Grignard's agitator. This piece of apparatus is particularly useful in carrying out the reaction called after that celebrated chemist (Fig. 15).

Either a reciprocating horizontal (Fig. 16), or a rotating vertical (Fig. 17), shaking machine may be used for shaking bottles.

Finally, what is most frequently needed is a contrivance for agitating a liquid in a fixed vessel, such as a beaker or bolt-head. Here there are two cases to consider, as the system may be either open or closed. The first of these alternatives is satisfied by such an

arrangement as that depicted in the figure (Fig. 18); this explains itself. Either a small water turbine or an electric motor may be used as the source of power. In the second case some kind of stuffing box or gland is needed through which to pass the shaft of the stirrer. Fig. 19 shows a laboratory model of a gland such as is used in the works.

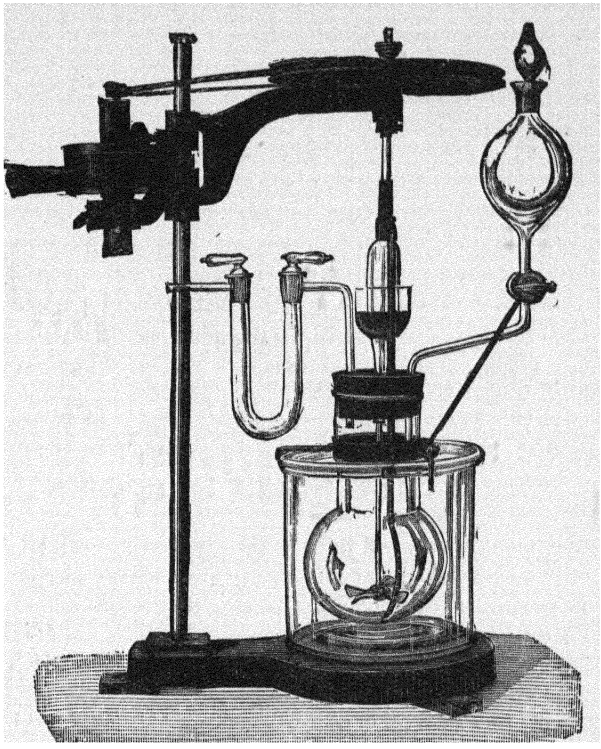


Fig. 20.

The same end is served, to better purpose in some cases, by a mercury sealed joint (Freundler), as is shown in Fig. 20.

Such arrangements of apparatus as are most frequently used in organic chemical preparative work have now been described. It goes without saying that they may be altered and adapted in innumerable ways.

RECOMMENDATIONS TO BEGINNERS

I. In carrying out any chemical preparation proceed as follows:

(1) Write down the equation for the reaction and reckon out how much of each substance to be used is needed;

(2) Fit up the apparatus;

(3) Weigh or measure out the materials.

The only comment that need be made on the first point is that usually an excess of one or another reagent must be used. For example, to improve the yield in an esterification, more alcohol or acid may be added. The chief consideration here is that of cost.

The second point has just been discussed in detail; there only remains, therefore, the third point. Too great emphasis cannot be laid on the need for care in using balances and weights, or, indeed, any measuring apparatus. It will be found useful to have the following rules posted up near the balances:

(1) Do not weigh out any substance directly on the balance-pan, but use either a sheet of paper or some kind of container;

(2) Return the weights to the box after use;

(3) Keep the balance and its neighbourhood rigorously clean.

Similarly, for reagents and other substances, students should observe the following precepts:

(1) Return all materials to the appropriate shelf or store when they are done with, having wiped or dusted the outside of the bottle or tin;

(2) Do not forget to replace stoppers in bottles;

(3) Do not put back an empty reagent bottle; either refill it yourself or tell the laboratory assistant to do so.

II. Order, cleanliness, patience: these should be the chief characteristics of the chemist, or, at any rate, those which he can acquire.

Make it a rule, every evening before leaving the laboratory, to devote half an hour or more to tidying up your bench, straightening up the reagent shelf, writing up your laboratory note-book, labelling the products you have made, and so forth. . . .

Do not drop bits of paper, match-ends, or any sort of liquid on the floor. Do not waste gas or water.

Try and keep the laboratory like a drawing-room. Moissan, one of the cleverest laboratory workers who have ever lived, used to say that the chemist's ideal was to be able to work, and not dirty himself, in his jacket, linen collar and best shoes, on a polished floor.

III. Of accidents, the most to be feared is fire. A fire may easily be caused by sodium or inflammable liquids, or by some mishap to the gas supply, or may break out in various other ways.

Now that yellow phosphorus is hardly ever used, sodium is the most dangerous substance to handle.[1] Residual scraps of sodium should be kept either under kerosene or in a tin box (not too tightly closed), duly labelled. When, say, forty to fifty grams has thus

[1] Nowadays sodamide should be added to the list.

accumulated it should be thrown little by little into waste alcohol in a dish ; it will dissolve without either flame or explosion.

Care should be taken to avoid heating a flask containing sodium as one reagent on a water-bath, and to have the bench on which sodium is being handled quite dry. If a piece inflames in spite of the precautions taken, sand should be thrown on it, inflammable liquids cleared away, and the operator should stand back to escape any explosion.

For combating fire in general, each laboratory should have its own regulations and provisions. The first step in every case should be to turn off the gas. Sand or water, according to the nature of the burning material, should then be thrown on to the fire. A plentiful supply of sand should always be within reach, with a scoop for handling it.

Some chemical operations are of a particularly dangerous nature—for example, nitrations, the preparation of hydrogen and of oxygen the heating-up of autoclaves, the use of certain gases. . . . No matter what precautions are taken, accidents will sometimes happen, and with beginners constant vigilance is necessary.

PREPARATION OF GUAIACOL

Nitration—Methylation—Reduction—Diazotisation.

1. Nitrophenol.
2. Methyl iodide.
3. Nitroanisole, using either methyl iodide or methyl sulphate.
4. *o*-Anisidine.
5. Guaiacol.
6. Guaiacol carbonate.
7. Potassium guaiacol sulphonate.

1. *Ortho-* and *para*-Nitrophenols.

A	Nitric Acid, sp. gr. 1·34	160 gm.
	Water	120 ,,
B	Phenol (molten)	80 ,,
	Water	10 ,,

Make up the solution A in a litre flask and cool it in a bath of ice and water to 5°. Add, little by little, during about three-quarters of an hour, the mixture B, which should be homogeneous. A dark brown coloration will develop. Keep the temperature between 5° and 15°. Leave for three hours, shaking from time to time, then pour into 500 c.c. of ice and water. Decant off the aqueous upper layer and wash the tarry mass by decantation three times with about 50 c.c. water. Steam-distil. Watch the condenser and run as little water as possible through, for if it be too cold, the condensed *o*-nitrophenol may plug it up. Continue the distillation until a sample of the distillate gives no crystalline separation on cooling.[1] When the distillate is cold, filter off the crystals, wash with a little water and dry on filter paper. M.p., 45°. Yield : 25 gm.

Another Method.

Sulphuric acid, sp. gr. 1·84	100 gm.
Water	200 ,,
Sodium nitrate	80 ,,
Phenol	50 ,,
Water	10 ,,

Run the sulphuric acid into the water, add the sodium nitrate, cool and add the mixture of phenol and water, observing the same precautions, and complete as above. Yield : 20 gm.

[1] Preserve the still residues; they contain the *p*-nitrophenol.

2. Methyl Iodide.

A. *Using phosphorus and iodine,* $5CH_3OH + 5I + P = 5CH_3I + H_3PO_4 + H_2O$.

Red phosphorus	10 gm.
Iodine	100 „
Methyl alcohol	30 „

Apparatus: Tubulated retort connected with a two-necked receiver immersed in an ice-bath ; in the second neck of the receiver is fitted a bent glass tube, the end of which dips into iced water (see Fig. 21).

Put the phosphorus and the alcohol in the retort and add the iodine (previously powdered) in small portions during an hour. Set by for twelve hours, then distil. Wash the distillate with a little iced

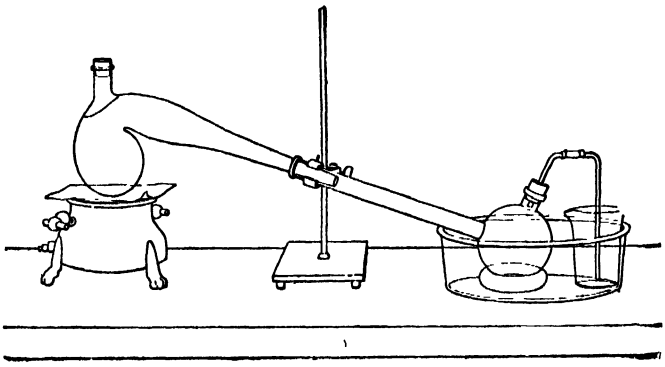


FIG. 21.

sodium hydroxide (10% solution) and dry over fused calcium chloride. Redistil, b.p., 43°–44°.

Calculated yield : from 100 gm. iodine, 110 gm. methyl iodide.

Actual yield : 90 gm., *i.e.* 81%.

B. *Using methyl sulphate and sodium iodide.*

In the retort (apparatus as above), place a solution of sodium iodide (15 gm.) in water (50 c.c.), warm to 90°, and add through a dropping funnel methyl sulphate (15 gm.), regulating the addition by the rate at which methyl iodide distils over. When all is added, raise to the boil for a minute or so. Yield : 90%.

3. *o*-Nitroanisole.

A. *Methylation of o-nitrophenol.*

o-Nitrophenol (as sodium salt) may be methylated by treatment with either methyl chloride or iodide under pressure (autoclave), with or without the addition of methyl alcohol, but the most convenient method for laboratory use employs methyl sulphate. With this reagent, methylation may be carried out in the cold if an excess be

used, whilst, if the temperature be raised sufficiently, almost the calculated amount will serve.

When the reaction takes place at low temperatures, the methyl sulphate loses only one methyl group and sodium methyl sulphate is formed, thus :

$$C_6H_4(NO_2)\cdot ONa + SO_2(OCH_3)_2 = C_6H_4(NO_2)\cdot OCH_3 + CH_3O\cdot SO_2\cdot ONa.$$

As the operation is carried out in aqueous solution, the sodium nitrophenoxide is actually partly dissociated into caustic soda and free nitrophenol :

$$C_6H_4(NO_2)\cdot ONa + H_2O = NaOH + C_6H_4(NO_2)\cdot OH.$$

The caustic soda acts on the methyl sulphate, giving methyl alcohol and sodium methyl sulphate. More soda and more methyl sulphate must therefore be added. It follows, therefore, that it is best to use concentrated caustic soda, so that less of the nitrophenoxide can become hydrolytically dissociated, and to add it to a mixture of methyl sulphate and the nitrophenol.

At higher temperatures, both methyl groups of the methyl sulphate take part in the reaction, theoretically at any rate. In practice, a loss arises through the formation of methyl alcohol. The reaction takes place in two stages, thus :

$$RONa + SO_2(OCH_3)_2 = CH_3O\cdot SO_2\cdot ONa + CH_3OR\ ^1$$
$$CH_3O\cdot SO_2\cdot ONa + RONa = Na_2SO_4 + CH_3OR.$$

Nitrophenol (powdered) . . .	14 gm.
Methyl sulphate	12·6 ,,
Caustic soda, sp. gr. 1·33 . .	18·0 ,,

Mix the nitrophenol and the methyl sulphate, cool in an ice-bath, stir vigorously, and add the caustic soda drop by drop. A red precipitate forms as each drop is added and soon the liquid begins to thicken. Stir for one hour, then keep at the boil for four hours. Pour into 50 c.c. water and extract with ether. Wash the ethereal extract with a little 10% caustic soda and dry over anhydrous potassium carbonate. Remove the ether and distil the residue *in vacuo*. B.p. about 60° at 12 m.m. pressure. Yield : 15 gm.

B. *Direct nitration of anisole, using acetyl nitrate.*

(This preparation should not be carried out by beginners.)

Acetyl nitrate is made as follows : Nitric anhydride, prepared by interaction of fuming nitric acid and phosphorus pentoxide (see below), is mixed with an equal volume of acetic anhydride. Solution takes place without noteworthy rise in temperature. The mixture is distilled under diminished pressure. The product has b.p. 22° at 70 m.m. ; sp. gr. 1·24/15° ; it is easily decomposed by moisture, and should be kept in well-stoppered bottles.

[1] R = phenyl radical.

To anisole, cooled in a freezing mixture, add, carefully, a molecular proportion of the above product. Leave for three hours, pour into water, collect the nitroanisole, and fractionate under diminished pressure. Yield : 90% (Pictet, 1907).

Preparation of Nitric Anhydride (Nitrogen Pentoxide).

Nitric acid (sp. gr. 1·5) is mixed with twice its weight of phosphorus pentoxide. A little heat is developed. The mixture is slowly distilled and nitrogen pentoxide crystallises in the receiver.

4. *o*-Anisidine, $C_6H_4(OCH_3)NH_2$.

Reduction of nitroanisole.

Nitroanisole	80 gm.
Water	100 c.c.
Fine iron filings	100 gm.
Hydrochloric acid, sp. gr. 1·16	10 ,,

Mix, in a flask, the nitroanisole, water, and iron filings. Add the hydrochloric acid little by little. Stir as vigorously as possible for two hours. Heat is evolved, but the mixture need not be cooled unless the temperature rises above 60°. Leave for a day, shaking occasionally. Make alkaline with sodium carbonate and extract with ether or steam distil. Yield : 62 gm. B.p., 225°–226°.

5. Guaiacol.

Diazotisation of anisidine, replacement of the amino by the hydroxy group.

A	Anisidine	61 gm.
	Sulphuric acid (50%)	140 ,,
	Ice and water	400 ,,
B	Sodium nitrite	35 ,,
	Water	100 c.c.
C	Copper sulphate, cryst.	140 gm.
	Water	140 c.c.

Make up the solution A in a thick-walled beaker or crock and cool it in a freezing mixture of ice and salt. Stir it mechanically and run in slowly the solution B.[1] Time : about one hour. Temperature : 0°–5°. While the diazotisation proceeds, fit a 1,500 c.c. flask with a cork, carrying (*a*) a dropping funnel, (*b*) a steam inlet pipe running to the bottom of the flask, and (*c*) a bent tube connected with a good condenser. Make up C in the flask and heat it to boiling, then, passing steam meanwhile, run in the diazo solution as quickly as the frothing will permit. Continue the steam distillation until the distillate has no appreciable odour of guaiacol. Collect the distillate, add enough common salt to give a saturated solution, and extract with benzene. Dry the extract over anhydrous sodium sulphate. Distil

[1] Always test for complete diazotisation by "spotting" on starch-iodide paper. —*Tr.*

off the benzene and fractionate the oily residue. Redistil, collecting that which boils between 175° and 203°. Yield : 35 gm.

Instead of copper sulphate a mixture of the following composition may be used (for 61 gm. anisidine) :

Sulphuric acid, sp. gr. 1·84	550 gm.
Anhydrous sodium sulphate	400 ,,
Water	300 c.c.

6. Guaiacol Carbonate.

Guaiacol	50 gm.
Caustic soda (*N* solution, 2 mol. proportions)	404 c.c.
Phosgene.[1]	

In a flask fitted with leading tubes and a dropping funnel cool the solution of guaiacol in caustic soda to 0°. Pass phosgene into the liquid until 20 gm. (1 mol. proportion) has been absorbed. Run in through the dropping funnel a further molecular proportion of caustic soda (202 c.c.), and again pass phosgene (10 gm. more). The oily precipitate which separates soon crystallises. Shake from time to time for a day and a half. Filter off the white crystals and wash, first with dilute caustic soda, then with water ; dry on filter paper. Recrystallise from spirit : silky needles, m.p., 88°–90°. Yield : 51 gm.

7. Potassium Guaiacolsulphonate.

Guaiacol	80 gm.
Sulphuric acid, sp. gr. 1·84	80 ,,

Mix and heat in a water-bath at 80° for six hours. Pour into 500 c.c. water in a large basin. Add barium carbonate until no more effervescence takes place. Filter. Add to the filtrate a concentrated solution of potassium carbonate until all barium is precipitated, avoiding any excess. Filter, evaporate the liquor until it will crystallise on cooling. Recrystallise the product from aqueous alcohol.

[1] Phosgene, in the form of a solution in toluene, is on the market. The phosgene is given off when the solution is warmed from 30° to 90°. About 120 c.c. of the solution will furnish 30 gm. phosgene. (Compressed phosgene, in cylinders, can also be obtained.—*Tr.*)

PREPARATION OF PHENACETINE

Phenacetine is prepared either by acetylating phenetidine or by ethylating *p*-acetylaminophenol, the former being the better method.

Phenetidine may be obtained either (1) by reducing nitrophenetole or (2) by reducing *p*-azophenetole; nitrophenetole either (1) by ethylating nitrophenol, or (2) by treating *p*-bromo- (or *p*-chloro-) nitrobenzene with sodium ethoxide.

p-Acetylaminophenol is produced either (1) by acetylating aminophenol, or (2) by heating the diazo compound from *p*-acetylaminoaniline (acetyl-*p*-phenylenediamine) with dilute acid. *p*-Aminophenol is obtained by reducing the nitrophenol.

Method I:

p-Nitrophenol—*o*- and *p*-Bromonitrobenzenes—*p*-Nitrophenetole—*p*-Phenetidine.

Method II:

p-Phenetoleazophenol — *p*-Azophenetole — *p*-Phenetidine — Phenacetine.

Method III:

p-Aminophenol—*p*-Acetylaminophenol—Phenacetine.

Other intermediates needed:

Acetyl chloride, acetic anhydride, ethyl bromide.

METHOD I

Preparation of nitrophenetole. To be completed by reduction as for *p*-azophenetole, and acetylation.

p-Nitrophenol

The still residues from the preparation of *o*-nitrophenol (see preceding preparation) contain *p*-nitrophenol as a black mass. This may be purified as follows:

Decant off the aqueous layer and treat the black tar with boiling 15% hydrochloric acid. Decant off the hot solution, decolorise with bone-black, filter and leave to crystallise in a cool place. It is best not to try to dissolve all the *p*-nitrophenol at once, but to treat the mass several times with the solvent until no further extraction takes place, *i.e.*, until a little of the extract gives no crystalline separation on cooling. Another method is to treat the black mass with the least possible amount of warm dilute (5%) caustic soda solution, filter, and precipitate the sodium compound of the *p*-nitrophenol by adding

a great excess of concentrated caustic soda to the filtrate—the phenoxide is only sparingly soluble in strong caustic soda solution. The crystals obtained may be purified by dissolving them again in water and reprecipitating with caustic soda, or the free nitrophenol may be separated by precipitating with hydrochloric acid.

For the tarry mass derived from the nitration of 80 gm. phenol, 125 c.c. caustic soda (5%) will serve for the extraction, and 60 gm. (about 43 c.c.) of more concentrated (36%) for the precipitation. Recrystallise the *p*-nitrophenol from boiling water. It will form almost colourless long needles, m.p. 115°.

Nitrophenetole

p-Nitrophenol	20·0 gm.
Sodium	3·30 ,,
Ethyl bromide	16·0 ,,
Alcohol (absolute)	100·0 ,,

Fit a 500 c.c. flask with a good reflux condenser and introduce the alcohol, then the sodium (in small pieces), and when the sodium has dissolved, the *p*-nitrophenol and the ethyl bromide. Boil under reflux for twelve hours. Carefully distil off most of the alcohol, add a little dilute caustic soda and take up the oil in ether. Wash the ethereal solution again with dilute caustic soda and with water, dry over fused calcium chloride, and distil off the ether. Recrystallise the residue from aqueous alcohol (70%). M.p., 60°; b.p., 283°. Yield: 16 gm.

If the reaction mixture be heated at 140° in a sealed tube for three hours instead of boiling as above, 19 gm. or 81% of the theoretical yield is obtained.

Another method may be merely recalled to mind, as the manipulation is not without risk. In this, *p*-bromonitrobenzene is heated with sodium ethoxide in alcoholic solution. If concentrated solutions in strong spirit be employed, the whole product is dibromoazoxybenzene; to obtain nitrophenetole, one must dilute the ethyl alcohol with its own volume of water.

Acetyl Chloride

$$PCl_3 + 3CH_3{\cdot}CO_2H = 3CH_3{\cdot}COCl + H_3PO_3.$$

Acetic acid (glacial)	125 gm.
Phosphorus trichloride . . .	95 ,,

Fit a distilling flask with a thermometer (to dip into the liquid) and a dropping funnel. Connect it with a condenser. Mix the acetic acid and phosphorus trichloride in the flask, cooling meanwhile, and keep at 40°–50° until two layers separate, say for twelve to fifteen hours. Distil from a water-bath. Fractionate the distillate, collecting that which distils from 45° to 55°; again fractionate, collecting

between 50° and 55°. Finally rectify again over 4 gm. recently fused sodium acetate. B.p., 55°. Yield: 95 gm.

Other methods are represented by the following equations:

$PCl_5 + CH_3CO_2H = CH_3COCl + POCl_3 + HCl.$

$PCl_5 + 3CH_3{\cdot}CO_2Na = 3CH_3{\cdot}COCl + NaPO_3 + 2NaCl.$

$POCl_3 + 2CH_3{\cdot}CO_2Na = 2CH_3COCl + NaPO_3 + NaCl.$

$SOCl_2 + CH_3{\cdot}CO_2H = SO_2 + HCl + CH_3COCl.$

Acetic Anhydride

1st Method: $CH_3{\cdot}CO_2Na + CH_3{\cdot}COCl = (CH_3CO)_2O + NaCl.$

Sodium acetate, recently fused,[1] powdered	60 gm.
Acetyl chloride	50 ,,

Place first the sodium acetate, then the acetyl chloride in a tubulated retort. Cool the retort with water as the action becomes violent. When it has moderated, stir up the mixture with a glass rod, arrange the retort for reflux distillation, and heat on the water-bath until volatile liquid is no longer seen to be condensing. Lower the condenser and distil the mixture from an oil-bath, taking the temperature up to 170°. Rectify over 5 gm. fused sodium acetate, collecting the fraction, 130°–142°: redistil and collect from 135°–140°. B.p., 138°. Yield: 30 gm.

2nd Method:

$4CH_3{\cdot}CO_2Na + POCl_3 = 2(CH_3CO)_2O + NaPO_3 + 3NaCl.$

Sodium acetate, recently fused	66 gm.
Phosphorus oxychloride	30 ,,

Run the phosphorus oxychloride, drop by drop, on to the sodium acetate. Much heat is developed. When all is added, heat on the water-bath for half an hour, then in an oil-bath at 180°–210° for four hours. Distil under diminished (at first not too low) pressure. Redistil the product under atmospheric pressure. Yield: 30 gm., boiling from 132° to 138°.

Ethyl Bromide

1st Method:

$C_2H_5OH + KBr + H_2SO_4 = C_2H_5Br + KHSO_4 + H_2O.$

Sulphuric acid, sp. gr. 1·84	200 gm. (110 c.c.).
Alcohol (95%)	110 ,,
Potassium bromide	100 ,,
Water	75 c.c.

Fit a 1,500 c.c. flask and condenser for ordinary distillation. Introduce first the alcohol, then the sulphuric acid. Allow to stand for three hours; then add the potassium bromide, dissolved in the water.

[1] See footnote on p. 253.

Heat quickly. The receiver should contain 50 c.c. water, into which should dip the end of a bent adapter fitted to the condenser, and should, moreover, be cooled in ice. Wash the product, a heavy oil, with dilute potassium carbonate solution, dry over calcium chloride. To purify from the ether formed as a by-product, treat with 10 c.c. strong sulphuric acid, keeping cold meanwhile. Distil. B.p., 38°–39°. Calculated yield, from 100 gm. KBr : 90 gm. C_2H_5Br. Actual yield: 70 gm.

2nd Method :

Phosphorus (red)	10 gm.
Alcohol	60 ,,
Bromine	20 c.c.

Fit a 300 c.c. distilling flask with a dropping funnel and good condenser. Introduce the alcohol and the phosphorus, then the bromine, drop by drop, keeping the mixture cold by immersing the flask in cold water. Leave for five hours. Distil from a water-bath and finish as above. If the product has a brown colour, wash it with a little sodium carbonate solution. Yield : 60 gm.

o- and *p*-Bromonitrobenzene

Bromobenzene	15 gm.
Nitric acid, sp. gr. 1·42	15 c.c.
Sulphuric acid, sp. gr. 1·84 (= 168° Tw.)	15 ,,

Cool the mixture of the two acids to $-5°$ or $-10°$, stir or shake vigorously and add the bromobenzene slowly in small portions. Warm gently to complete the reaction. Pour into water. Decant the aqueous layer from the crystalline mass (19 gm.). Recrystallise from 50% alcohol. The *p*-isomeride separates almost completely (m.p., 125°), and the *ortho* may be recovered from the mother liquors (m.p., 40°). Yield : 97·7%, viz., 14 gm. *para* and 2·5 gm. *ortho*.

METHOD II

Diazotisation of phenetidine. Coupling with phenol. Ethylation. Reduction of *p*-azophenetole. Acetylation.

p-Phenetoleazophenol

A	Phenetidine	27·4 gm.
	Hydrochloric acid (20%)	75 ,,
	Water	400 c.c.
B	Sodium nitrite	14·0 gm.
	Water	100 c.c.
C	Phenol	19·0 gm.
	Sodium carbonate (anhydrous)	40 ,,
	Water	700 c.c.

Make up solution A in a litre beaker or mug and cool it to 5° by adding crushed ice. Run in solution B, with good stirring, slowly, so as to prevent nitrous fumes being evolved. Stir the completed diazo solution for a few minutes and then pour steadily, with stirring, into solution C, which should have been already cooled to 0°. The azo compound is precipitated forthwith as a tarry mass. Transfer a little to a test tube, add a few drops of ammonia, and grind with a glass rod for a short time : the mass becomes yellow, granular and crystalline, and by adding it to the rest of the product, the whole is obtained in this form. Filter off, wash with water and dry on filter paper. Yield : 40 gm.

A sample of this yellow product may be purified by recrystallisation from benzene (seven times its weight). It then forms brilliant spangles, easily soluble in alcohol and ether, sparingly in water, m.p., 124·5°.

p-Azophenetole

$C_2H_5O \cdot C_6H_4 \cdot N_2 \cdot C_6H_4 \cdot OC_2H_5$.

p-Phenetoleazophenol	10 gm.
Alcohol	50 ,,
Sodium	1·2 ,,
Ethyl bromide	6·0 ,,

Dissolve the sodium in the alcohol in a 125 c.c. flask fitted with a reflux condenser. Add the phenetoleazophenol and the ethyl bromide. Allow to stand without heating for twenty-four hours, then boil under reflux for six hours. On cooling the *p*-azophenetole crystallises out. Filter it off and wash with water. Recrystallise from benzene. Sparingly soluble in alcohol, hot or cold, and in cold benzene. Orange scales, m.p., 160°. Yield : 9 gm.

Phenetidine

Reduction of *p*-azophenetole by stannous chloride.

(i) *Preparation of the reducing agent :*

Tin	20 gm.
Hydrochloric acid, concentrated . .	100 c.c.

Pour over the tin at first only one-third of the hydrochloric acid. When no further evolution of gas takes place, add another one-third and a few drops of 1% solution of platinic chloride, then the rest of the acid. Do not leave the solution exposed to the atmosphere.

(ii) *Reduction :*

p-Azophenetole	10 gm.
Alcohol	6 ,,

Add to the mixture, all at once, 50 c.c. of the stannous chloride solution. Shake and warm on the water-bath until all is decolorised and

in solution. Cool (in an ice-chest or with ice). A copious crystalline separation forms. Filter this off. Treat it with caustic soda in excess. The crystals are those of a tin double salt, and when caustic soda is added hydrated tin oxides are at first precipitated and then dissolved by the excess of soda while the phenetidine separates as an oil. Caustic soda must therefore be added until the oily upper layer no longer increases in bulk. Take this up in ether, dry over anhydrous potassium carbonate and complete as usual. B.p., 195°–200°. Yield : 7 gm.

Azophenetole may also be reduced with sodium hydrosulphite, phenylhydrazine, etc.

Phenacetine

Phenetidine	13 gm.
Water	50 c.c.
Acetic anhydride	13 gm.

Shake all together vigorously. Almost immediately a solid separation forms. Filter off and recrystallise from about 60 c.c. alcohol (30%) or from about 800 c.c. water. M.p., 134°–137°. Phenacetine is soluble in about 80 parts of boiling water.

METHOD III

Ethylation of *p*-acetylaminophenol.

***p*-Aminophenol**

A	*p*-Nitrophenol	13·9 gm.
	Ammonia (sp. gr. 0·880)	50 c.c.
	Water	50 „
B	Ferrous sulphate, cryst.	195 gm.
	Water	450 c.c.

The procedure resembles exactly that for aminobenzoic acid (*q.v.*). Concentrate the liquors to a quarter of their original volume, cool in the ice-chest, filter off the product and wash it with a little water. Yield : 8 gm. Recrystallise from the least possible amount of water (about 20 c.c.). M.p., 184°.

***p*-Acetylaminophenol**

p-Aminophenol, crude product powdered	8 gm.
Water	20 c.c.
Acetic anhydride	8 gm.

Add the acetic anhydride little by little to the aqueous suspension of aminophenol. Solution takes place. Continue shaking for a minute or so, then cool under the tap. The acetyl derivative separates almost quantitatively. Filter it off and recrystallise from 50 c.c. water. Yield : 8 gm. M.p., 169°.

Phenacetine

p-Acetylaminophenol	7·0 gm.
Alcohol	25·0 ,,
Sodium	1·25 ,,
Ethyl bromide	6·0 ,,

Dissolve the sodium in the alcohol, add the acetylaminophenol and the ethyl bromide; leave for three hours, then boil for an hour under reflux. Filter, evaporate off the alcohol. Crush up in water and filter. Recrystallise from 30% alcohol. Yield nearly quantitative. M.p., 137°.

PREPARATION OF ACETANILIDE (*Antifebrine*).

$C_6H_5 \cdot NH \cdot CO \cdot CH_3$.

This preparation entails those of nitrobenzene and aniline.

Nitrobenzene

Nitric acid, sp. gr. 1·43 . . .	21 gm.
Sulphuric acid, sp. gr. 1·84 . .	22·5 ,,
Benzene	22 ,,

Mix the two acids in a beaker placed inside a small dish. Stir vigorously and add the benzene, drop by drop, from a small separating funnel. Allow the temperature to rise to 50° and keep it between 50° and 55° by running a little water, as needed, into the outer dish. When all the benzene is added continue the agitation for a few minutes, then set by for two hours. Separate the oily nitrobenzene, wash it first with water, then with very dilute sodium carbonate solution, until it is free from acid. Dry over calcium chloride and distil under diminished pressure, taking care not to carry the distillation too far. B.p., 95°/17 m.m. Yield : 28 gm.

Aniline

1*st Method of reduction :*

Nitrobenzene	22 gm.
Iron filings	22 ,,
Hydrochloric acid, concentrated . .	2·5 ,,
Water	50 c.c.

Mix the nitrobenzene and the iron filings in a flask with stout walls. Shake or stir vigorously and add the hydrochloric acid, diluted with the water, in three or four portions. The lively reaction will carry the temperature up to about 70°, and the mixture should be cooled if it becomes hotter than this. When the reaction is complete, add 25 c.c. water, and then, gradually, and with care, 5 gm. sodium carbonate. Steam distil : extract the aniline from the distillate with ether. Dry the extract over anhydrous potassium carbonate. Distil off the ether and fractionate. B.p., 183°–185°. Yield : 17 gm.

2*nd Method of reduction :*

Nitrobenzene	25 gm.
Tin, granulated	50 ,,
Hydrochloric acid, concentrated . .	130 ,,

Put the nitrobenzene and about half the tin in a 500 c.c. flask and add about 15 gm. hydrochloric acid. In a few minutes a vigorous

action will begin. Cool under the tap until the effervescence abates, then add more acid. Continue the addition of acid and tin in this way until the action becomes sluggish; it will then be necessary to warm on the water-bath. When nearly all the tin is dissolved dilute with about 60 c.c. water and add 80 gm. caustic soda as a concentrated solution, cooling meanwhile. Steam distil; collect about a litre of distillate, to which add 200 gm. common salt. Extract with ether. Dry the extract over anhydrous sodium carbonate and a few fragments of solid caustic potash. Drive off the ether and distil. B.p., 183°–185°. Yield: 18 gm. to 20 gm.

Acetanilide

Aniline	50 gm.
Glacial acetic acid	40 ,,

Fit a 250 c.c. flask with a cork, carrying a thin-walled glass tube about two feet long, the upper end of which should be bent through 180°, and should dip into a small empty flask. Mix the aniline and acetic acid in the flask and boil for six hours, regulating the heating in such a way that as little aniline as possible distils off and only the water formed in the reaction is collected in the small flask. Distil the residue, b.p., 295°, 58 gm. Recrystallise from hot water, m.p., 112°.

PREPARATION OF ANTIPYRINE

Phenylhydrazine.[1]
Phenylmethylpyrazolone.[1]
Phenyldimethylpyrazolone.

Phenylhydrazine

Aniline	50 gm.
Hydrochloric acid, concentrated . .	125 ,,
Sodium nitrite	37·5 ,,

Dissolve the aniline in the hydrochloric acid diluted with 200 c.c. water, cool with ice, and add the sodium nitrite dissolved in 78 c.c. water. When diazotisation is complete, pour gradually into a well-stirred, ice-cold, 70% solution of sodium sulphite ($2\frac{1}{2}$ mol. proportions are necessary).[2] The mixture becomes coloured first a clear yellow, then orange, then redder still, and soon sodium diazobenzenesulphonate is precipitated. Add water until almost all is redissolved, acidify with acetic acid and treat with zinc dust, continually stirring, until the solution is colourless. Filter warm and run in gradually an amount of strong hydrochloric acid equal in volume to a third of the solution. Phenylhydrazine hydrochloride crystallises out. Filter off and press. The mother liquor yields a second crop on concentration. Dissolve the salt in the least possible quantity of warm water and liberate the base by adding caustic soda. Extract with ether and dry the extract over anhydrous potassium carbonate. Distil, collecting together that distilling between 200° and 240°; rectify and collect the fraction 225°–240°. Yield: 35–40 gm.

Phenylmethylpyrazolone

Phenylhydrazine	100 gm.
Ethyl acetoacetate	125 ,,

Warm together on the water-bath for several minutes; the first action results in the separation of water. Continue to heat for two hours; the oil turns into a resinous mass, which is to be treated with its own volume of acetone and set to crystallise in the ice-chest. A cake of crystals forms. Crush up, filter and wash with water. Recrystallise from water. M.p., 127°.

[1] Cf. Hewitt's *Organic Chemical Manipulation*, 1897 (London: Whittaker). This, like most other English text-books of preparative organic chemistry, gives the stannous chloride method of reducing the diazonium salt; on the manufacturing scale, however, the sulphite process is used.—*Tr.*

[2] This may be prepared by halving a solution of the requisite amount of caustic soda, saturating one half with sulphur dioxide, and adding the other half.—*Tr.*

Phenyldimethylpyrazolone (*Antipyrine*).

(Methylation of phenylmethylpyrazolone.)

Phenylmethylpyrazolone . . .	50 gm.
Methyl iodide	50 ,,
Methyl alcohol (99%)	50 ,,

Mix the reagents and transfer the mixture to thick-walled tubes. Seal up [1] and heat at 115°–125° for ten hours. Distil off the methyl alcohol under diminished pressure. Dissolve the residue in the least possible quantity of water, filter, add caustic soda (36° Bé.) gradually. The quantity added should be just enough to set free an oil, which will collect on the surface, and excess should be carefully avoided. Separate the oily layer and boil it under reflux for two hours with 250 c.c. benzene. Decant the solution and evaporate it down until crystals will form on cooling. Cool in the ice-chest. Filter off the crystalline separation, wash it with a little benzene and dry in the air. Yield : 38 gm.

Antipyrine may be purified by recrystallisation from spirit. For the 38 gm. crude antipyrine, 20 gm. alcohol (50%) is needed. To make the product nearly colourless it will be necessary to recrystallise, using bone-black, at least twice.

[1] The tube for such an operation as this should be sealed up in the manner prescribed for a "Carius tube" (Determination of halogens), in most laboratory handbooks. Hewitt's book, already referred to, gives an excellent description of the operation, but an actual demonstration and a little practice are of more value than any written account.—*Tr.*

PREPARATION OF ACETYLSALICYLIC ACID (*Aspirin*)

Salicylic Acid

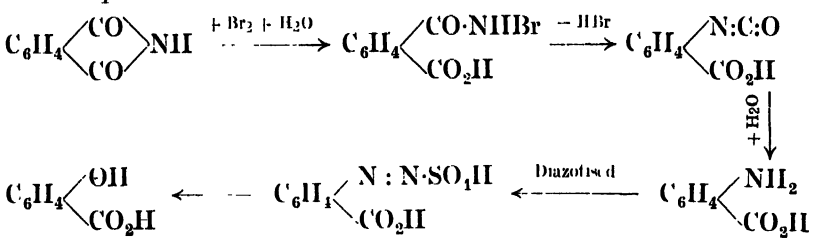
Phenol	28 gm.
Alcohol, absolute	80 „
Sodium	8 „

To the alcohol, in a flask fitted with a reflux condenser, add the sodium, little by little, then the phenol. Distil off the alcohol and carefully dry the residue over a naked flame, keeping the flask moving continuously, until a powdery mass is obtained. Crush and sift rapidly, in as dry a place as possible, and transfer to a 200 c.c. tubulated retort. Heat in an oil-bath. When the temperature reaches 110° start to pass in carbon dioxide (the end of leading tube should be about 1 cm. above the sodium phenoxide). Raise the temperature to 190° in four hours (20° per hour), then to 200° during two hours, shaking frequently to expose fresh material to the gas. Cool, dissolve in water and precipitate the product with hydrochloric acid. Filter off and recrystallise from water. M.p., 158°.

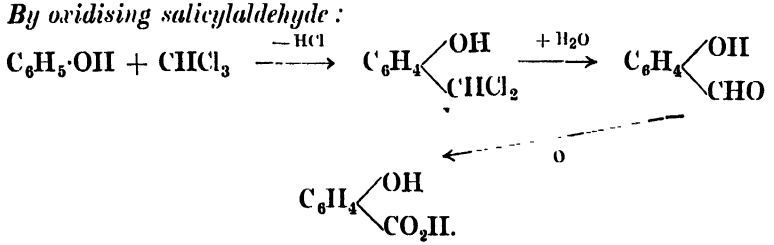
If potassium phenoxide be used, the *p*-carboxylic acid is the main product. Resorcinol will take up carbon dioxide in this way if it is simply heated with sodium bicarbonate in aqueous solution.

Other ways of obtaining salicylic acid are the following :

From phthalimide :



By oxidising salicylaldehyde :



Acetylation of Salicylic Acid using Acetic Anhydride

Salicylic acid	25 gm.
Acetic anhydride	27 „

Mix together in a 125 c.c. flask fitted with a reflux condenser and heat in an oil-bath at 150°–160° for three hours. Distil off the excess of acetic anhydride, and the acetic acid formed in the reaction, *in vacuo*. The distillate should weigh 16 gm. and the residue 31 gm. The latter should be recrystallised from twice its weight of benzene. Yield: 18 gm., purified product, m.p., 135°–137°. If the mother liquors are concentrated, another 10 gm., nearly pure, may be obtained.

Acetylation by means of Acetyl Chloride and Pyridine

Acetyl chloride and salicylic acid will not interact of themselves; a (tertiary) base, such as pyridine, must be present. Pyridine forms with acid chlorides additive compounds, which act as acetylating agents, pyridine hydrochloride being produced.

$$C_5H_5N\begin{matrix}CO\cdot R\\Cl\end{matrix} + R'\cdot OH \longrightarrow R'\cdot O\cdot CO\cdot R + C_5H_5N\cdot HCl.$$

Pyridine	10 gm.
Salicylic acid	14 „
Acetyl chloride	10 „

Dissolve the salicylic acid in the pyridine by gently warming. Cool the solution in a freezing mixture and add the acetyl chloride gradually. As soon as the first drops are added the mixture becomes pasty, then it liquefies but finally again thickens. Warm for ten minutes on the water-bath and pour on to crushed ice, stirring meanwhile. The viscous mass soon solidifies. Crush it up, filter off, wash with water and dry at 60°–70°. Yield, crude product: 13 gm. Recrystallise from benzene as before.

PREPARATION OF STOVAINE

1. Chloracetone.
2. Dimethylamine, from dimethylaniline or ammonia.
3. Methylethylchloromethylcarbinol (*Grignard reaction*).
4. Methylethyldimethylaminomethylcarbinol.
5. Benzoic acid (*Sandmeyer reaction, Grignard reaction*).
6. Benzoyl chloride.
7. Stovaine.

1. Chloracetone

A	Acetone	150 gm.
	Water	15 c.c.
	Marble, crushed to about the size of grains of corn and sifted	30 gm.
B	Manganese dioxide	400 ,,
	Hydrochloric acid	800 ,,

Fit a litre flask with a reflux condenser, leading tube and dropping funnel, introduce the acetone and the marble, warm to 40°–50°. Pass in chlorine (generated from the mixture B), not too quickly, agitating meanwhile, and run in the water (10–15 c.c.), drop by drop, so as to dissolve the calcium chloride as it is formed. Cut off the chlorine supply when nearly all the marble has disappeared—three to four hours. *Caution* If the acetone becomes golden yellow and the marble appears not to be attacked, the current of gas should be stopped and the mixture boiled until the excess chlorine has disappeared. Sometimes the chlorine dissolves in the acetone without actually entering into reaction, and then chlorination suddenly takes place with explosive violence. The colour of the solution must, therefore, never be more than a pale dull yellow, not on any account greenish or golden yellow.

When the reaction is over, the mixture will have separated into two layers. The lower one should be run off and the upper one washed with water (here it will go to the bottom), dried over calcium chloride, and carefully distilled. Acetone first passes over (up to 100°), then the chloracetone (from about 115°–125°). Rectify over a little calcined magnesia. B.p., 119°. Yield: 110 gm.

Chloracetone sharply attacks the eyes. This should be remembered during the distillation, and when the residues are thrown away and the vessels washed out.

2. Dimethylamine. (i) Methyl- and Dimethylaniline

Methylaniline is not used in drug manufacture except for making Exalgine (acetyl methylaniline), the use of which is now

almost abandoned. But from dimethylaniline, dimethylamine is prepared.

Methylation of Aniline.—When aniline is treated with methyl iodide the product consists of a mixture in which unattacked aniline, monomethylaniline, dimethylaniline and trimethylphenylammonium iodide are all present. The salt of the quaternary base is soluble in water so it can be easily separated, and as the boiling point of aniline is about 10° below that of the other two bases, it also may be removed, provided the fractionating apparatus is efficient. But as methyl- and dimethylaniline have almost the same boiling point, efforts must be made so to regulate the reaction that one or the other is produced in a nearly pure state. This may be effected by heating together aniline hydrobromide and the theoretical amount of methyl alcohol.

On the industrial scale the methylating agent is methyl chloride, or simply methyl alcohol together with a certain proportion of aniline hydrochloride or other catalyst. The reactions which then take place are :

$$C_6H_5{\cdot}NH_2{\cdot}HCl + CH_3{\cdot}OH = CH_3Cl + H_2O + C_6H_5{\cdot}NH_2$$
$$CH_3{\cdot}Cl + 2C_6H_5{\cdot}NH_2 = C_6H_5{\cdot}NH{\cdot}CH_3 + C_6H_5{\cdot}NH_2{\cdot}HCl.$$

Thus, aniline hydrochloride always being regenerated, all the methyl alcohol is converted eventually into methyl chloride.

Monomethylaniline.

Aniline hydrobromide	17·0 gm.
Methyl alcohol	3·5 ,,

Heat together in a sealed tube for six hours at 160°. Dissolve the contents of the cooled tube in water, make strongly alkaline with caustic soda and extract with benzene. Dry the extract over anhydrous potassium carbonate, distil off the benzene and fractionate the residue. From 182°–188° 3 gm. aniline distils, from 189°–191° 1 gm. intermediate fraction, from 191°–194° 6·5 gm. methylaniline.

Dimethylaniline.—The procedure is the same as for monomethylaniline, but somewhat more than double the amount of methyl alcohol is used. All the aniline enters into reaction and the product is almost exclusively dimethylaniline, b.p., 192°–193°. When much more methyl alcohol is used a considerable amount of trimethylphenylammonium bromide is formed, and this is thrown out of solution as such, together with the dimethylaniline, when the solution is made strongly alkaline. Being insoluble in benzene, it remains as an oily layer between the aqueous liquor and the benzene extract, and may later be decanted off. If it be set aside in a dish it soon crystallises ; when distilled under diminished pressure it breaks down quantitatively to dimethylaniline.

Industrial dimethylaniline may be bought ; it is a very pure product. For all that, the process for making the compound in the

laboratory is given here because it is a general one, applicable to other alkyl derivatives.

To test for monomethylaniline in dimethylaniline of doubtful purity, dissolve 2 gm. in 20–30 c.c. dilute hydrochloric acid (10%–15%), cool, and add a concentrated solution of sodium nitrite a little at a time. If monomethylaniline be present, a yellow oil, soluble in ether, will separate out. This is methylphenylnitrosamine, $C_6H_5 \cdot N(NO) \cdot CH_3$. The presence of the monomethyl derivative being indicated, the exact amount is determined as follows :

Estimation of Aniline and Monomethylaniline in Dimethylaniline.

Reagents needed :

Acetic anhydride.
Normal solution of sodium hydroxide.

Make a blank titration with the acetic anhydride ; 1·02 gm. should need 20 c.c. *N* sodium hydroxide.

Weigh out accurately, *e.g.*, 2·60 gm. of the sample of crude dimethylaniline, and 1·206 gm. acetic anhydride. Mix and leave in contact for one hour ; stir up with 30 c.c. water, and warm on the water-bath for half an hour ; titrate with the *N* sodium hydroxide. Thus :

1·206 gm. acetic anhydride needed	23·6	c.c. *N* sodium hydroxide.
1·206 gm. acetic anhydride needed after reaction . .	16·8	,, ,, ,,
The acetic anhydride used up was therefore equivalent to .	6·8	,, ,, ,,

I.e., 0·3468 gm. acetic anhydride was used up.

Now 102 gm. acetic anhydride corresponds to $93 \times 2 = 186$ gm. aniline ; so 0·3468 gm. acetic anhydride corresponds to 0·6324 gm. aniline.

That is, in 2·60 gm. crude dimethylaniline there are 0·6324 gm. aniline, or 24·3%.

N.B.—The monomethylaniline also reacts, so actually both this and aniline are estimated and evaluated together as aniline.

Nitrosodimethylaniline

A	Dimethylaniline	200 gm.
	Hydrochloric acid, concentrated .	500 ,,
	Ice, crushed	1,000 ,,
B	Sodium nitrite	130 ,,
	Water	350 c.c.

Make up the mixture A in a 5-litre crock. Stir continuously (mechanically) and add the solution B gradually. Filter off the yellow precipitate of nitrosodimethylaniline hydrochloride, press, wash with a little cold 20% hydrochloric acid and dry on filter paper. Yield : 170 gm. (hydrochloride).

Dimethylamine

Nitrosodimethylaniline hydrochloride	50 gm.
Zinc, granulated	5 „
Caustic soda, 36%	150 „
Water	500 c.c.

Fit a 2-litre flask with a condenser. Connect the condenser with an empty flask, and this by means of a bulb tube with a second flask containing 15% hydrochloric acid (in excess). Mix together in the

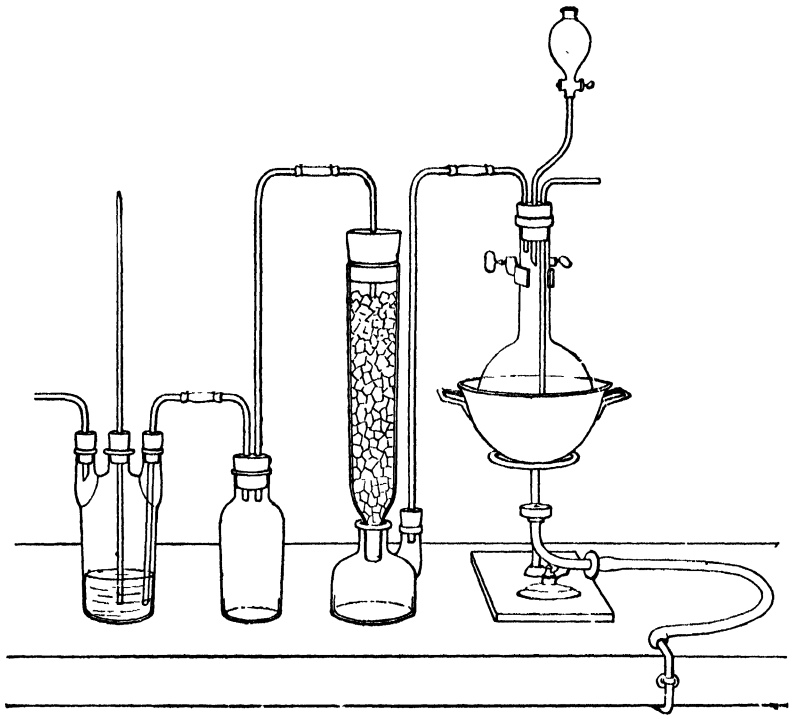


Fig. 22.

distilling flask the caustic soda and the water, add the zinc and 20 gm. nitrosodimethylaniline hydrochloride. Boil until the nitroso compound is decomposed and dissolved. Quickly uncork the distilling flask and add 15 gm. more nitroso hydrochloride. Boil again. Finally add the rest of the nitroso compound and complete. Evaporate the solution of dimethylamine hydrochloride to dryness. About 18 gm. crude product should be obtained, which may be used without further purification for preparing a dry benzene solution of dimethylamine.

This preparation is to be carried out in the apparatus depicted in Fig. 22, by a similar method to that described for trimethylamine (*q.v.*).

(ii) Dimethylamine and Methylamine by Methylating Ammonia.[1]

Formaldehyde (Formalin, 40%)	250 gm.
Ammonium chloride	125 ,,

Fit a 500 c.c. flask with a cork carrying a thermometer to dip into the liquid and an elbow tube connected with an inclined condenser. Another flask will be needed as a receiver. Mix together the formalin and the ammonium chloride in the flask. Heat by means of an oil-bath, first to 50°, then gradually more strongly so that the temperature reaches 104° in two hours; keep it at 104° for two more hours. Leave to cool overnight. In the morning filter off the crystalline separation of ammonium chloride, add 100 gm. formalin and again heat, as before, to 115°. Cool overnight. Filter off the mixture of ammonium chloride and methylamine hydrochlorides and take up the latter in hot alcohol. Filter the alcoholic solution when cold and evaporate it down in a dish, eventually over a free flame, but taking care that the temperature of the concentrated liquor does not rise above 120°. When skin forms on the surface, cool in a desiccator over sulphuric acid; in a few hours the solution will set to a cake. Chop this up and transfer it to a small flask fitted with a reflux condenser, and digest it on the water-bath with 100 c.c. chloroform.[2] Filter warm and distil off 75 c.c. chloroform from the filtrate. Transfer the remaining syrup to a dish and set it in a desiccator over sulphuric acid to crystallise. Yield: 75 gm.

3. Methylethylchloromethylcarbinol

$$CH_2Cl\cdot C(OH)\cdot CH_3.$$
$$\quad\quad\quad

product by adding ice and dilute sulphuric acid (10%). Separate the ethereal layer, dry it over anhydrous sodium sulphate, distil off the ether and fractionate the residue *in vacuo*, collecting that which passes over between 60° and 100°. Rectify under atmospheric pressure. B.p., 150°. Yield : 25 gm.

The product is not quite pure, as it contains some ethylamylcarbinol, produced by a more complicated reaction. This chlorine-free alcohol distils at the same temperature as the chief product, the chlorhydrin.

4. Methylethyldimethylaminomethylcarbinol

Methylethylchloromethylcarbinol . . .	19·5 gm.
Benzene solution of dimethylamine, dry, 25% .	90 ,,

Mix the reagents together. Keep the mixture cool. Transfer to thick-walled tubes, seal up and heat at 125° for a day. Filter off the crystals that have separated and wash them with a little benzene. Unite the filtrate and washings and distil off the excess dimethylamine and some of the benzene. Extract the remaining solution with dilute hydrochloric acid, using enough to take out all the base and give a faintly acid solution. Wash the acid extract with ether. Evaporate it down to small bulk, cool and add 20 gm. concentrated caustic soda, then 40 gm. sodium carbonate. Extract with 100 c.c. ether at twice. The base has b.p. 74°–75°/29 m.m. ; 150°–154° at atmospheric pressure.

Bromobenzene (for 5)

Benzene	225 gm.
Aluminium chloride	12·5 ,,
Bromine	320 ,,

Fit a litre flask with a dropping funnel and a reflux condenser. Connect the upper end of the reflux condenser with a wash bottle containing water, by means of a bulb tube or other device to prevent the water being sucked back. Add the aluminium chloride to the benzene in the flask, warm gently and run in the bromine, drop by drop. As soon as a reaction begins, remove the source of heat. The introduction of the bromine should take from four to six hours. Wash the product three times with water and steam distil, stopping when the distillate contains a crystallisable substance, viz., dibromobenzene. Separate the oil from the distillate, dry over calcium chloride, and distil :

From 90° to 130°, benzene passes over,
130° to 155°, a mixture, about 20 gm.,
155° to 159°, bromobenzene.

Redistil the first runnings and collect all together that distilling from 155°–160°. Yield : 225 gm.

5. Benzoic Acid

(i) *By the Grignard reaction :*

Magnesium (ribbon)	2·3 gm.
Ether, anhydrous	10 ,,
Bromobenzene	16 ,,
Ether, anhydrous	40 ,,

Fit a (dry) 220 c.c. flask with a reflux condenser and dropping funnel. Introduce the magnesium (in small pieces) and *nearly* cover it with the 10 c.c. ether. Run in through the dropping funnel 3 gm. bromobenzene, warm gently, and drop in a minute crystal of iodine. In a few moments, sometimes immediately, reaction begins, little bubbles are formed and the iodine disappears. Add forthwith, drop by drop, the ethereal solution of the bromobenzene, taking care that the action does not become too vigorous. Set aside for four hours. Cool in an ice-bath. Remove the dropping funnel and put in its place a leading tube to dip into the liquid. Pass in a current of well-dried carbon dioxide for about two hours. A nearly solid mass is formed. Add 60 c.c. ether, a few small pieces of ice, and a mixture of 15 c.c. concentrated hydrochloric acid and 60 c.c. water. Shake and continue to extract with ether until all is taken up, then extract the ether with 10% caustic soda in slight excess. Filter the alkaline solution and add an excess of hydrochloric acid. Filter off the precipitated benzoic acid and recrystallise from water. Yield: 10 gm. M.p., 120°.

(ii) *From aniline by Sandmeyer's method :*

$$C_6H_5{\cdot}NH_2 + HNO_2 + HCl = C_6H_5{\cdot}N_2{\cdot}Cl + 2H_2O.$$
$$C_6H_5{\cdot}N_2{\cdot}Cl + KCN = C_6H_5{\cdot}CN + KCl + N_2.$$

Diazotisation.

A	Aniline	75 gm.
	Hydrochloric Acid, concd.	225 ,,
	Water	250 c.c.
B	Sodium nitrite	56 gm.
	Water	225 c.c.

Make up the solution A in a thick-walled beaker or jar of about 2 litres capacity, fitted with a mechanically driven stirrer. Add small lumps of ice until the temperature falls to 4°, then run in from a dropping funnel the sodium nitrite solution (B), little by little. Add more ice from time to time so as to keep the temperature below 8°. Test for completion in the usual way.

Conversion to the Nitrile.

Copper sulphate (crystallised)	320 gm.
Water	800 c.c.
Potassium cyanide	220 gm.
Water	400 c.c.

Fit a 2-litre flask for steam distillation. Make up the copper sul-

phate solution in the flask and add the potassium cyanide solution gradually, keeping the mixture hot on the water-bath. Shake vigorously, and carry out the operation in a good fume cupboard, because cyanogen and hydrogen cyanide are evolved. Cool to 70°, keep on the water-bath, and run in slowly, during about an hour, the above diazo solution. Shake from time to time. When all is introduced, heat on the boiling water-bath for an hour, then steam distil. Separate the benzonitrile, dry over calcium chloride and distil. B.p., 190°. Yield : 47 gm.

Hydrolysis.

Benzonitrile	25 gm.
Sulphuric acid	150 ,,
Water	80 ,,

Mix the reagents in a 500 c.c. flask fitted with a reflux condenser and slowly heat to boiling. A vigorous reaction begins. When it has moderated, boil for five hours. Benzoic acid crystallises out on cooling. Yield : 23 gm.

Benzoic acid may be obtained in other ways, thus :

By oxidising toluene with manganic sulphate ;

By hydrolysing benzotrichloride ;

By heating potassium benzenesulphonate with potassium formate,

$$C_6H_5{\cdot}SO_3K + H{\cdot}CO_2K = C_6H_5{\cdot}CO_2K + KHSO_3 ;$$

(The nitrile) by heating the sulphonate with potassium cyanide,

$$C_6H_5{\cdot}SO_3K + KCN \longrightarrow C_6H_5{\cdot}CN.$$

6. Benzoyl Chloride

$$C_6H_5{\cdot}CO_2H + PCl_5 = C_6H_5{\cdot}COCl + POCl_3 + HCl.$$

Benzoic acid, dry	50 gm.
Phosphorus pentachloride . . .	90 ,,

Rapidly crush the phosphorus pentachloride (in a fume cupboard), mix it with the benzoic acid in a 250 c.c. flask, and immediately attach a reflux condenser. A lively reaction, accompanied by evolution of hydrochloric acid, takes place. When this has ceased, warm on the water-bath for five minutes. Fractionate. Phosphorus oxychloride forms the bulk of the fraction distilling up to 120°. Rectify that which passes over above 150°. Benzoyl chloride has b.p. 200°. Yield : 46 gm.

Other Methods : $3\ C_6H_5{\cdot}CO_2H + PCl_3$,
$C_6H_5{\cdot}CO_2Na + POCl_3$,
$C_6H_5{\cdot}CO_2H + SOCl_2$,
$C_6H_6 + COCl_2$, and a catalyst.

7. Hydrochloride of Methylethyldimethylaminomethylcarbinol Benzoate (*Stovaine*)

A	Methylethyldimethylaminomethylcarbinol	5 gm.
	Benzene	15 „
B	Benzoyl chloride	10 „
	Benzene	10 „

Mix together the solutions A and B in a 125 c.c. flask, without cooling. A lively reaction takes place. All goes into solution, but on cooling a crystalline mass is obtained. Filter off the separation, wash it with benzene and recrystallise from three times its weight of absolute alcohol. M.p., 174°–175°. Yield: 90%.

Test the anæsthetic action on the end of the tongue.

PREPARATION OF ETHYL AMINOBENZOATE (*Anæsthesine*)

p-Nitrotoluene.
p-Nitrobenzoic acid.
p-Aminobenzoic acid.
Ethyl *p*-aminobenzoate.

Nitration of Toluene [1]

Toluene	500 gm.
Nitric acid, sp. gr. 1·5	2,000 ,,

Run the acid, drop by drop, into the toluene, keeping the temperature about 30°. Set by for three hours. Pour into five litres iced water. Separate the oil, dry over calcium chloride, distil carefully, stopping when the temperature reaches 260°. The main fraction is that which passes over between 230° and 255°.

Oxidation of *p*-Nitrotoluene by Potassium Dichromate—*p*-Nitrobenzoic Acid

p-Nitrotoluene	28 gm.
Potassium dichromate	100 ,,
Sulphuric acid	137 ,,
Water	150 c.c.

Mix all together carefully and boil under reflux for about 2½ days, until the colour of the solution has definitely changed to green. Add 200 c.c. water and steam distil off the remaining nitrotoluene. Filter, wash the deposit with hot water, collect the liquors, cool, filter, precipitate by adding hydrochloric acid. Dissolve the filter cake in 10% caustic soda, using a small excess and warming on the water-bath until all is dissolved. Cool, filter, add hydrochloric acid to the filtrate to precipitate the nitrobenzoic acid. Filter off and recrystallise from 70% alcohol. M.p., 238°. Yield: 20 gm.

Potassium permanganate may also be used to oxidise *p*-nitrotoluene.

p-Aminobenzoic Acid

Nitrobenzoic acid	16·7 gm.
Ferrous sulphate	195 ,,
Water	400 c.c.
Ammonia	*q.s.*

Dissolve the nitrobenzoic acid in about 60 c.c. strong ammonia and 50 c.c. water and pour the solution, a little at a time, into the boiling

[1] See Cain's *Manufacture of Intermediates for Dyes.—Tr.*

ferrous sulphate solution. Stir vigorously and add gradually enough ammonia (120–150 c.c.) to keep the mixture faintly alkaline. Filter off and wash the brown precipitate, unite the filtrates, evaporate down on the water-bath to about half the original volume, filter again, precipitate the aminobenzoic acid by adding acetic acid (about 10 c.c.), avoiding any excess. Filter off the precipitate, wash it with the least possible amount of water and dry. The mother liquors may be concentrated and will give a further separation of about 2 gm., making the yield, in all, 13·5 gm. Recrystallise the product from water. Felted needles, sparingly soluble in cold water or alcohol, more readily in warm alcohol and freely in acetone. M.p., 186°.

Ethyl Aminobenzoate Hydrochloride (*Anæsthesine*)

Aminobenzoic acid.	10 gm.
Alcohol (96%)	50 „

Mix, saturate with hydrochloric acid gas in the cold, boil under reflux for half-an-hour ; on cooling, the hydrochloride of the ester will crystallise out almost quantitatively, filter it off and wash with a little absolute alcohol. Yield : 10 gm. M.p., 203° (decomp.).

The product is soluble in alcohol, sparingly so in acetone. When placed on the tongue, it causes an intense sensation of anæsthesia.

Cycloform is butyl aminobenzoate. *Novocaine* is diethylamino-ethyl aminobenzoate.

PREPARATION OF QUIETOL

(Hydrobromide of the valeric ester of propyl dimethylaminohydroxy-*iso*butyrate.)

Chlorohydroxy*iso*butyric acid.
Propyl (and ethyl) chlorohydroxy*iso*butyrate.
Propyl dimethylaminohydroxy*iso*butyrate.
Quietol.

Chlorohydroxy*iso*butyric Acid

$$CH_2Cl\cdot CO\cdot CH_3 \xrightarrow{HCN} CH_2Cl\cdot C(OH)\cdot CN \quad (CH_3)$$

$$\xrightarrow{2H_2O} CH_2Cl\cdot C(OH)\cdot CO_2H. \quad (CH_3)$$

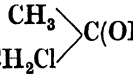
Chloracetone	42 gm.
Hydrocyanic acid (25%) . .	60 ,,
Ammonia	4 drops.
Hydrochloric acid, fuming . .	60 c.c.

Fit a 500 c.c. flask with a reflux condenser. Immerse it in a bath of ice and water and introduce successively the chloracetone and the hydrocyanic acid. When the temperature of the mixture has fallen to 5°, connect up the condenser and run in the four drops of ammonia (down the condenser). A reaction begins. Shake the mixture briskly and leave it for six hours.

Add the fuming hydrochloric acid, then bubble hydrogen chloride gas into the liquid until 42 gm. has been absorbed. Heat at 70° for two hours, boil for four hours, having connected a wash-bottle with the top of the condenser to take up the hydrogen chloride evolved during the boiling. Cool, extract with ether three times, using 150 c.c. ether each time. Dry the extract over anhydrous sodium sulphate. The residue left by distilling off the ether will solidify. Dissolve it in 500 c.c. crystallisable benzene, add 25 gm. anhydrous sodium sulphate; filter hot. As the solution cools the acid will separate in yellowish scales; it may be obtained in a perfectly colourless state by again recrystallising from benzene and treating with bone-black.

Prismatic needles, readily soluble in water and ether, sparingly soluble in cold benzene. M.p., 110°. Yield: 59 gm.

Ethyl Chlorohydroxy*iso*butyrate



Chlorohydroxy*iso*butyric acid	10 gm.
Alcohol, absolute	30 ,,
Hydrogen chloride, gas	5 ,,

Boil the solution under reflux for five hours. Distil off three-quarters of the alcohol, add water to the residue, extract with ether. Wash the extract with sodium bicarbonate solution followed by water, then dry over anhydrous sodium sulphate. Distil the residue left after removing the ether under diminished pressure. B.p., 85°/11 m.m.

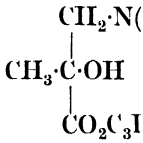
Propyl Chlorohydroxy*iso*butyrate

Chlorohydroxy*iso*butyric acid	115 gm.
Propyl alcohol	345 ,,
Hydrogen chloride, gas	37 ,,

Reflux for three hours. Remove most of the excess of alcohol under diminished pressure, complete as above.

B.p., 100°/13 m.m. Yield: 120 gm.

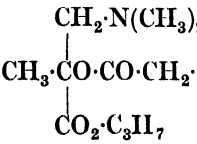
Propyl Dimethylaminohydroxy*iso*butyrate



Propyl chlorohydroxy*iso*butyrate	48 gm.
Dimethylamine	36 ,,
Benzene	120 ,,

Heat at 115° for six hours in sealed tubes. Filter off the dimethylaminehydrochloride and work up the liquor as for the base of Stovaine, using here, however, sodium carbonate to liberate the base, not caustic soda. B.p., 89°–91°/12 m.m.

Valeric Ester of the Above (as Hydrobromide)



A	Propyl dimethylaminohydroxy*iso*butyrate	75 gm.
	Benzene	96 ,,
B	Valeryl bromide[1]	65 ,,
	Benzene	96 ,,

Mix and proceed as for Stovaine. Recrystallise from benzene. M.p., 120°.

[1] See p. 224.

PREPARATION OF ACETOPHENONE (*Hypnone*)

(Friedel and Crafts's Method.)

Aluminium chloride, anhydrous . .	135 gm.
Benzene, dry	180 ,,
Acetyl chloride	80 ,,

Fit a flask with a reflux condenser and dropping funnel. Introduce the aluminium chloride and the benzene and cool the mixture in a bath of ice and water. Run in the acetyl chloride, drop by drop, shaking continuously. Allow to stand for two hours in the cold, pour on ice (care !). Separate, wash the oily layer with water, then with very weak caustic soda, dry over anhydrous sodium sulphate. Distil, collecting the fraction 190°–220°. B.p., 202°; m.p., 20°. Yield: 70 gm.

PREPARATION OF BROMOVALERYLUREA (*Bromural*)

Valeric acid.
Valeryl chloride and bromide.
Bromovaleryl chloride and bromide.

Valeric Acid

A	Amyl alcohol	80 gm.
	Sulphuric acid, concd.	240 ,,
	Water	80 ,,
B	Potassium dichromate, finely powdered	200 ,,
	Water	360 ,,

Add gradually the mixture B to A (in a litre flask with a reflux condenser and dropping funnel) during about 1½ hours. A vigorous reaction takes place, accompanied by lively ebullition. To complete, heat eventually to boiling point. Steam distil. Extract the distillate with ether, then wash the extract with 10% caustic soda, using a small excess. The ethereal solution now contains (i) valeric aldehyde, (ii) amyl valerate, (iii) amyl alcohol, and may be worked up separately. The alkaline aqueous solution, containing sodium valerate, is to be evaporated down on the water-bath, the residue acidified with hydrochloric acid and extracted with ether. Distil. B.p., 171°–175°. Yield: 40 gm.

Valeryl Chloride

Valeric acid	45 gm.
Thionyl chloride	80 ,,

Heat the mixture at 60°–70° in a flask fitted with a reflux condenser and a tube to lead off hydrogen chloride. Raise the temperature gradually to 95° to expel hydrogen chloride and the excess of thionyl chloride. Fractionate. B.p., 112°–115°. Yield: 46–47 gm.

Valeryl Bromide

$$3(CH_3)_2CH\cdot CH_2\cdot CO_2H + P + 5Br = 3(CH_3)_2CH\cdot CH_2\cdot COBr + HPO_3 + 2HBr.$$

Valeric acid	51 gm.
Bromine	80 ,,
Phosphorus, red	10 ,,

Introduce the phosphorus, then the acid, into a flask; shake and run in the bromine, drop by drop. The mixture becomes hot and hydrogen bromide is evolved. Leave overnight. Warm on the water-bath until all hydrogen bromide is driven off (one to two hours).

Decant. Distil from an oil-bath at 170°. Rectify. B.p., 137°–140°. Yield : 72 gm.

Bromovaleryl Chloride

Valeryl chloride	12·3 gm.
Bromine	16·3 „

Fit a 125 c.c. flask with a reflux condenser, a dropping funnel, and a tube to lead away hydrobromic acid gas. Introduce the valeryl chloride, warm to about 60°, and add the bromine, little by little. Gradually (during two hours) heat to 100°. Distil *in vacuo*. B.p., 94°–95°/46 m.m. ; 77°–78°/18 m.m. Yield : 16 gm.

Bromovaleryl Bromide

Valeryl bromide	150 gm.
Bromine	150 „

Run in the bromine, drop by drop, at 80°–90°. Hydrogen bromide is evolved. Heat in a boiling water-bath for two hours, then distil. B.p., 186°, at atmospheric pressure ; 113°/40 m.m. ; 85°/10 m.m. Yield : 190 gm.

Bromovaleryl bromide may be prepared from valeric acid in one operation by using the appropriate amount of bromine in presence of red phosphorus.

Bromovalerylurea (*Bromural*)

Urea	10 gm.
Bromovaleryl chloride	13 „

Mix and warm carefully on the water-bath. Heat more strongly if nothing happens. The reaction usually begins at about 130°–140°, but sometimes earlier, and is accompanied by the evolution of hydrogen chloride. As soon as this takes place, remove the source of heat. Often warming on the water-bath will suffice, and the reaction may become violent ; in this event, the flask should be cooled by immersion in cold water. When the mass, having once liquefied, becomes solid again, interaction is complete. Take up in water and recrystallise rapidly from aqueous alcohol, or better, from toluene. M.p. (after two recrystallisations from toluene), 154°.

PREPARATION OF DIETHYLBARBITURIC ACID (*Veronal*) AND OF BROMODIETHYLACETYLUREA (*Adaline*)

Ethyl malonate.
Ethyl ethylmalonate and diethylmalonate.
Diethylbarbituric acid (Veronal).
Diethylacetic acid.
Diethylacetyl chloride.
Bromodiethylacetyl chloride and bromide.
Bromodiethylacetylurea (Adaline).

Ethyl Malonate

(i) *Calcium malonate.*

Chloracetic acid[1]	200 gm.
Ice	300 ,,
Caustic soda, 33%	500 ,,
Potassium cyanide	138 ,,

There will be needed two 2-litre flasks, one fitted with reflux condenser, and a 10-litre crock.

To the acid add caustic soda until it is exactly neutralised (about 250 c.c. will be needed), introducing ice as needed to keep the solution cool. To the solution, at 40°, add a mixture of the (finely powdered) potassium cyanide and 268 c.c. water. The temperature will rise perhaps to 80°. After an hour has elapsed, warm gradually to 100° and keep at that temperature for another hour. Cool to 20°. Add 250 gm. caustic soda (33%) and boil until no more ammonia is evolved (about five hours). Pour into the crock, add a 25% solution of calcium chloride until no more precipitate is thrown down (about 259 gm. $CaCl_2$ will be needed). After twenty-four hours, filter off the calcium malonate, wash it with water and dry, first on the water-bath, then in an oven at 100°. Yield : 280 gm.

(ii) *Conversion into the ester.*

Calcium malonate	200 gm.
Alcohol, absolute	500 ,,

Suspend 20 gm. of the malonate in the alcohol and pass in hydrogen chloride until it has dissolved, then add another portion of malonate and again pass in hydrogen chloride ; repeat the operations until all the calcium malonate has been taken up ; finally saturate the solution with hydrogen chloride. Set it by for twenty-four hours. Add calcium carbonate cautiously until all the acid is neutralised. Distil off most of the alcohol under diminished pressure. Take up the residue with ether, dry the extract over calcium chloride. Distil off the ether and fractionate. B.p., 197°–198°. Yield : 140 gm.

[1] See p. 248.

Ethyl Ethylmalonate

Malonic ester	16 gm.
Alcohol, absolute	25 ,,
Sodium	2·3 ,,
Ethyl iodide	17 ,,
(or ethyl bromide	12 ,,)

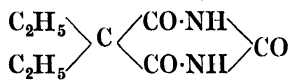
Introduce the alcohol into a 125 c.c. flask fitted with a reflux condenser. Add half the sodium; when that has dissolved, add half the remainder, finally add the last quarter. Connect the flask with the condenser, and introduce the ethyl malonate (down the condenser). The mixture will set almost solid in a few minutes. Add, again down the condenser, the ethyl iodide, by portions. Boil under reflux for 1½ hours. Distil off the alcohol, take up the residue with water and extract with ether. Dry the extract with anhydrous sodium sulphate. Fractionate.

B.p., 206°–208°. Yield: 15 gm.

Ethyl Diethylmalonate

Treat the ethyl ethylmalonate as ethyl malonate has just been treated, using the appropriate quantities. B.p., 218°.

Diethymalonylurea (*Veronal*)



A	Ethyl diethylmalonate	20 gm.
	Alcohol	50 ,,
	Urea	6 ,,
B	Sodium	4·6 ,,
	Alcohol	50 ,,

Make up the solution A in a 250 c.c. flask fitted with a reflux condenser, boil, and add the sodium ethoxide solution B all at once. Continue to boil for four hours, allowing the solution to become gradually more concentrated. Finally heat to 115° in an oil-bath. Neutralise exactly and evaporate off the alcohol. Take up the residue in water, adding a little hydrochloric acid, so that the mixture becomes faintly acid. Set it by in a cold place for twelve hours. Recrystallise the product from 80% alcohol. M.p., 190°. Yield: 8–10 gm.

Diethylacetic Acid

(i) *Diethylmalonic acid.*

A	Ethyl diethylmalonate	21·5 gm.
	Alcohol	40 ,,
B	Potassium hydroxide (stick)	15 ,,
	Water	15 ,,

To the solution A add the caustic potash B, shaking meanwhile;

gradually the ether will dissolve. On the morrow distil off most of the alcohol, take up the residue in water and neutralise exactly with hydrochloric acid. Add calcium chloride, dissolved in a little water, until there is no further precipitate of calcium diethylmalonate. Filter this off, wash with water, stir into more water, and liberate the acid by adding hydrochloric acid. Extract with ether, carefully dry the extract with anhydrous sodium sulphate. Distil off the ether; the residue will crystallise. M.p., 123°. Yield: 13·5 gm. = 84%. A little ester escapes saponification.

(ii) *Diethylacetic acid* (*α-Ethylbutyric acid*).

Heat 37 gm. diethylmalonic acid at 190° for a quarter of an hour in a retort with the beak pointing upwards. Carbon dioxide is evolved. Readjust the retort for distillation and distil. B.p., 192°. Yield: 19·1 gm. = 82%.

Bromodiethylacetyl Bromide

$(C_2H_5)_2CBr{\cdot}COBr$.

(i) *Diethylacetyl bromide* (*α-Ethylbutyryl bromide*).

Bromine	34·5 gm.
Phosphorus, red	4·2 ,,
Diethylacetic acid	25 ,,

Fit a 125-c.c. flask with a reflux condenser (a plain tube) and a dropping funnel. Introduce the phosphorus, add the acid and, without cooling, run in the bromine, drop by drop. The temperature rises a little and hydrogen bromide is evolved. When all the bromine is added, set by for two hours, decant if necessary, and distil.

B.p., 153°–158°. Yield: 34 gm.

(ii) *Bromodiethylacetyl bromide.*

Diethylacetyl bromide	34 gm.
Bromine	31 ,,

Put the diethylacetyl bromide in an apparatus like that used for the first operation, run in the bromine, drop by drop, and heat slowly up to 100°. The bromine is slowly absorbed and hydrogen bromide is evolved. The operation should take from three to four hours. Distil under diminished pressure. Almost all will pass over between 95° and 100° at 26 m.m. Yield: 40 gm.

Diethylacetyl Chloride

Diethylacetic acid	89 gm.
Phosphorus trichloride	45 ,,

Warm the acid to 60° and add the phosphorus trichloride in small quantities, heating gradually to 100°. Separate and distil the product under diminished pressure, taking care to keep the receiver cold. B.p., 67°–75°/30 m.m.; 134°–141° at atm. pressure. Yield: 89 gm.

Bromodiethylacetyl Chloride

Diethylacetyl chloride	50 gm.
Bromine	50 „

Fit a 250-c.c. flask with a reflux condenser and a dropping funnel. Introduce the chloride and heat to 100° on the water-bath. Add the bromine, drop by drop, waiting for decolorisation to take place between each addition. Hydrogen bromide is evolved; when the operation is properly carried out, only very little bromine is carried over, but towards the end it is less readily absorbed and a considerable amount may be lost in this way. The bromination must extend over several hours. Distil under diminished pressure; the product will pass over at 100°/19 m.m. Yield: 50 gm.

Bromodiethylacetylurea (*Idaline*)

Urea (Carbamide)	6 gm.
Bromodiethylacetyl bromide . .	15 „

Mix the reagents, stir, and heat on the water-bath until the mixture solidifies (hydrogen bromide should be evolved). Cool, crush up with water and 1 gm. sodium carbonate. Filter off and recrystallise from hot alcohol (four times the weight of the product), adding, little by little, water up to twice the volume of the alcoholic solution. M.p., 115°–117°. Yield: about 8 gm.

ADRENALINE

PREPARATION OF ADRENALINE FROM THE SUPRARENAL GLAND OF THE HORSE (*G. Bertrand's Method*)

600 gm. of the glands, from which fat has been removed, are finely minced, introduced into a 2-litre bottle or pickle-jar, and mixed with 95% alcohol and 5 gm. oxalic acid. Enough alcohol must be used to fill the vessel brim-full; it is then to be carefully stoppered and shaken at intervals for two days. The contents are then filtered on a framed cloth, the residual mass being well pressed (screw-press), and the filtered extract is concentrated under diminished pressure. When the alcohol has evaporated considerable amounts of coloured substance, lecithin, fats, and the like, separate. The mixture is gently shaken with petrol and transferred to a separating funnel. When it has settled the lower layer is run off and precipitated by adding neutral lead acetate, taking care to avoid excess (if a mistake is made, add sulphuric acid). The precipitate is filtered off and the liquor concentrated *in vacuo* to 200 c.c.; a slight excess of ammonia is added, and the solution set by in a cool place, not exposed to the air. Yield: 0·8 to 1·0 gm.

Purification: The adrenaline is redissolved in about $2\frac{1}{2}$ times its weight of 10% sulphuric acid and an equal volume of alcohol added to the solution; it is filtered and reprecipitated by ammonia.

SYNTHESIS OF ADRENALINE [1]

The steps in this preparation are:

Chloracetocatechol;
Methylaminoacetocatechol;
Racemic adrenaline;
Lævo-Adrenaline.

Chloracetocatechol

Catechol	50 gm.
Monochloracetic acid	50 ,,
Phosphorus oxychloride	50 ,,

Mix the three reagents in a 1,500-c.c. flask, fitted with reflux con-

[1] Only the preparation of methylaminoacetocatechol will be described here. This product yields racemic adrenaline on reduction, but the reduction and separation of the racemic compound into the optical isomerides are not operations that beginners can carry out; moreover, it is only the preparation of chloracetocatechol that is a classical operation, of which an example should be known.

denser and tube to lead away hydrochloric acid gas, and heat for an hour on the water-bath. Add 500 c.c. boiling water to dissolve the mass, and leave the solution in a cool place for two days. Filter off the product and recrystallise from a little hot water, adding bone-black to decolorise the solution. Yield : 30 gm. Yellowish needles, very irritating to the nasal membranes ; m.p., 173°.

Methylaminoacetocatechol

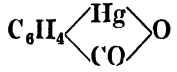
$C_6H_3(OH)_2 \cdot CO \cdot CH_2 \cdot NH \cdot CH_3$.

Chloracetocatechol.	10 gm.
Methylamine (40% aqueous solution) .	20 c.c.

The chloracetocatechol is to be finely powdered and mixed with 5 c.c. alcohol. Cool the mixture in an ice-salt bath, and add the 20 c.c. methylamine solution. The temperature rises ; when the reaction is over, stir for a short time and set by overnight. Filter off the crystalline separation, and dissolve it in a little dilute hydrochloric acid ; filter the solution and add just enough ammonia to precipitate the product. Filter, wash with a little ice-cold water, then with alcohol, finally with ether. Yield : 5 gm. Yellowish crystalline powder.

MERCURIALS [1]

"Mercury Benzoate" (*o*-Hydroxymercuribenzoic Anhydride)

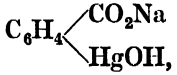


Sodium benzoate	15 gm.
Mercuric acetate	15 „

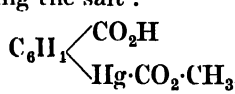
Dissolve the two salts each in 50 c.c. water. Mix the solutions with brisk stirring. Almost insoluble mercuric benzoate separates out. Filter off the precipitate, wash it with water and dry. Recrystallise from chloroform. Needles, m.p., 165°.

This is the (true) mercury benzoate of the pharmacopœias. Treated with soda it yields mercuric oxide, whilst with acids it liberates benzoic acid. When it is heated at 150°–175° the mercury enters the nucleus and *o*-hydroxymercuribenzoic anhydride is formed.

Carefully dry the mercuric benzoate, transfer it to a flask and heat it in an oil-bath at 160°–175°, until a test portion dissolves in caustic soda solution without giving a yellow precipitate of mercuric oxide. (During the heating, free benzoic acid sublimes.) Wash the product with ether to remove benzoic acid. It then forms a white powder with amphoteric properties, being at once a base and an acid. It dissolves in caustic soda, forming the salt :

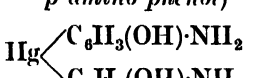


and in acetic acid, forming the salt :



This acetate solution yields a precipitate with sodium chloride (avoid any excess), the chloride being sparingly soluble in water.[2]

MERCURY DIAMINODIHYDROXYDIPHENYL [3] (*Mercuri-di-p-amino-phenol*)



[1] We describe here the preparation of two compounds, neither of which is any longer used in medicine. The two preparations are, however, typical of those which involve the introduction of mercury into the nucleus ; we therefore quote them, being of the opinion that substances of this class will play a more and more important part in the therapeutics of syphilis.

[2] The mercury salicylate employed in pharmacy (in combination with sodium methylarsinate = *Enesol*) likewise contains mercury in the nucleus.

[3] This compound is analogous to Salvarsan. This is a typical example of the preparation of a complex mercurial.

The steps in this preparation are :

p-Nitrophenol ;
Nitrohydroxyphenylmercuric acetate ;
Mercury dinitrodihydroxydiphenyl (Di-*p*-nitromercuridiphenol) ;
Mercury diaminodihydroxydiphenyl.

Nitrohydroxyphenylmercuric Acetate

Sodium nitrophenoxide . . .	16·2 gm.
Mercuric acetate	31·8 ,,
Acetic acid	10 ,,

Dissolve the sodium nitrophenoxide in the least possible amount of water and add a hot concentrated aqueous solution of the mercuric acetate and the acetic acid. Soon the nitrohydroxyphenylmercuric acetate separates, almost quantitatively. Wash with water and dry the product.

Mercury Dinitrodihydroxydiphenyl

Nitrohydroxyphenylmercuric acetate . .	19·6 gm.
Sodium sulphide solution [1] equivalent to .	3·9 ,, Na_2S.

Dissolve the nitrohydroxyphenylmercuric acetate in dilute sodium hydroxide, pass carbon dioxide to saturation into the solution, filter off the mercurial hydroxide which is precipitated, and wash it with water. Suspend it in water and redissolve it by adding the just necessary amount of caustic soda. Filter, make up to 500 c.c. with water, add the sodium sulphide solution (3·9 gm. Na_2S), and heat on the water-bath for three hours. Mercury sulphide is precipitated ; filter hot. The sodium salt of mercury dinitrodihydroxydiphenyl separates on cooling. Yield : 6 gm.

The mercury sulphide precipitate contains some of the desired product. This can be extracted by washing with dilute sodium chloride solution (if this be not used the mercuric sulphide will pass through the filter paper in colloidal suspension), and can be precipitated from the extract by sulphuric acid—2·50 gm.

Mercury Diaminodihydroxydiphenyl

Mercury dinitrodihydroxydiphenyl (sodium salt) .	6 gm.
Sodium hydrosulphite	120 ,,
Sodium carbonate	75 ,,

Dissolve the hydrosulphite and carbonate in 50 c.c. and the phenoxide in the smallest possible amount of water. Mix the solutions and heat at 60° until a test sample gives no yellow colour with sodium hydroxide. Acidify with acetic acid ; a heavy precipitate will form from which the liquid can be decanted. Redissolve the precipitate

[1] A convenient strength for sodium sulphide solution is 10% ; it oxidises fairly rapidly, and should therefore be titrated before use.

in a little dilute hydrochloric acid, filter and add concentrated hydrochloric acid in excess. Mercury diaminodihydroxydiphenyl hydrochloride is thrown down as white needles. Filter these off and wash with a little alcohol. Yield : 4 gm.

An aqueous solution of the hydrochloride yields the free base when sodium carbonate is added. The base is sparingly soluble in water and can be crystallised; it is readily soluble in dilute caustic alkali. The alkaline solution is rapidly oxidised by the air, with separation of metallic mercury.

ARSENICALS

PREPARATION OF SALVARSAN (ARSENOBENZOL, "606")[1]

This preparation includes those of arsanilic acid (*Atoxyl*), hydroxyphenylarsinic acid, and nitrohydroxyphenylarsinic acid.

Arsanilic Acid

Arsenic acid, commercial, 76% . .	200 c.c.
Aniline	280 ,,

Put the arsenic acid in an earthenware jar and heat it in an oil-bath at 120°–140°, until most of the water has been driven off (twelve to fifteen hours). Cool the aniline to 0° and add it, little by little, to the prepared arsenic acid. Stir. Gradually the mixture thickens and forms a granular mass, so that eventually it can be powdered.

Introduce 200 gm. of the powder into an Erlenmeyer flask and heat it at 160° in an oil-bath. At this temperature it will fuse; stir until all is molten, then connect the flask with a reflux condenser. Heat for half an hour at 160°–170°, then for an hour at 180°–185°. Cool a little and add 225 c.c. 25% caustic soda diluted with 225 c.c. water. Part of the material is dissolved, the remainder (aniline) separates out. When cold, separate the lower layer, shake it up with kieselguhr to clarify it; filter. To the filtrate add 100 c.c. 25% hydrochloric acid; then make a number of tests, using 25 c.c. of the solution each time, so as to determine how much more hydrochloric acid should be added to give the maximum precipitation. If this is satisfactorily done, the mixture will set to a mass. Cool for an hour, filter, suspend the precipitate in 200 c.c. water and filter again. Recrystallise from hot water. Yield: 30% of theory.

p-Hydroxyphenylarsinic Acid

$HO{\cdot}C_6H_4{\cdot}AsO_3H_2$.

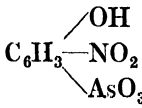
Arsanilic acid	22·0 gm.
Sodium nitrite	7·5 ,,

Dissolve the arsanilic acid in 400 c.c. 5% sulphuric acid in a 2-litre flask, add the sodium nitrite dissolved in a little water and heat on the water-bath until no more nitrogen is evolved. Add barium hydroxide solution to precipitate the sulphuric acid (till no more

[1] The preparation of arsenobenzol, as given here, has been described in great detail by Kober (*J. American Chemical Society*, 1919, *41*, 442), but it must not be supposed that always a usable product will be obtained; although it is relatively easy to produce arsenobenzol, or, at least, a very similar product, yet it is very difficult to prepare a non-poisonous sample.

precipitate is formed), filter, evaporate to dryness *in vacuo*. Extract the residue with boiling 50% alcohol. On cooling, the sodium salt of the hydroxyphenylarsinic acid will separate.

3-Nitro-4-hydroxyphenylarsinic Acid



Sodium hydroxyphenylarsinate	14 gm.
Sulphuric acid	4 c.c.
Mixed acid { Sulphuric acid	4 ,,
Mixed acid { Nitric acid (sp. gr. 1·4)	4 ,,

Cool the sulphuric acid to 0°, dissolve in it the arsinate; stir, add the mixed acid little by little. Let the temperature rise to 10° towards the end. Pour into 225 c.c. water and leave to stand for forty-eight hours. Filter off the nitro-acid. Yield: 65%.

Diaminodihydroxyarsenobenzene (*Salvarsan*)

(Kober, *J. A. C. S.*, 1919, *41*, 442.)

Nitrohydroxyphenylarsinic acid	8·5 gm.
Magnesium chloride	22 ,,
Sodium hydrosulphite	110 ,,
Caustic soda, 40%	6 c.c.

Dissolve the magnesium chloride in 550 c.c. water in a litre flask, add the hydrosulphite, which will also rapidly dissolve.

Make up separately a solution of the nitrohydroxyphenylarsinic acid in 200 c.c. water with the 6 c.c. caustic soda. Mix the two solutions; heat first to 40° until the turbidity settles, filter quickly, and continue heating the filtrate at 50°–60°. The salvarsan separates slowly as a yellow powder. Filter it off and wash with ice-cold water. Transfer it to a porcelain dish, stir up with 40 c.c. water at 0° and add, to dissolve it, 15 c.c. 8% caustic soda. Filter to remove impurities. To the filtered solution add 15 c.c. of a mixture of equal parts of fuming hydrochloric acid and water; the base is at first thrown out of solution, then redissolved. Dilute the solution with 170 c.c. ice-cold water and pour it gradually into 325 c.c. concentrated hydrochloric acid diluted with its own weight of water and cooled to 0°. Salvarsan hydrochloride is sparingly soluble in strong hydrochloric acid, and is therefore precipitated. Filter it off and dry in a vacuum desiccator over calcium chloride. Yield: 75%.

PREPARATION OF TYRAMINE

(*p*-Hydroxyphenylethylamine—$HO \cdot C_6H_4 \cdot CH_2 \cdot CH_2 \cdot NH_2$.)

Benzyl cyanide ;
p-Nitrobenzyl cyanide ;
p-Aminobenzyl cyanide ;
p-Hydroxybenzyl cyanide ;
Tyramine.

Benzyl Cyanide

Potassium cyanide, 90%	60 gm.
Benzyl chloride	100 ,,
Alcohol	100 c.c.

Fit a 500-c.c. flask with a reflux condenser. Dissolve the potassium cyanide in 55 c.c. water, warm to 60°, and add the mixture of benzyl chloride and alcohol gradually down the condenser. Heat for hours on the water-bath. "Bumping" may take place, so the tion will need attention.

The mixture will form two layers, the lower, aqueous, layer containing much potassium chloride. The upper layer should be separated and distilled. The first runnings are mainly alcohol and water; collect the fraction boiling from 210° to 235°. Yield : 75% of theory.

p-Nitrobenzyl Cyanide

Benzyl cyanide	117 gm.
Nitric acid, sp. gr. 1·52	700 ,,

Put the nitric acid in a flask, cool it in an ice-salt freezing mixture, and run in the benzyl cyanide, drop by drop. Shake vigorously and see that the temperature does not rise above 7°. When all is added, set by for half an hour, then pour on much ice. In a few minutes the product solidifies to a mass of crystals. Pure *p*-nitrobenzyl cyanide is obtained by recrystallising these twice from alcohol. M.p., 117°. Yield : 100 gm.

p-Aminobenzyl Cyanide

p-Nitrobenzyl cyanide	16·2 gm.
Tin	22 ,,
Hydrochloric acid, concentrated	100 c.c.
Alcohol	200 ,,

Put the tin, nitro compound and alcohol in a flask and add the hydrochloric acid in small portions, shaking meanwhile and regulating

the addition so that the temperature keeps below 25°. When nearly all the tin has dissolved, warm to 50° and shake until a test portion dissolves completely in caustic soda, giving only a faintly yellow colour. Distil off the alcohol under diminished pressure until the chlorostannate of the base begins to crystallise out. Filter it off and redissolve in a little water, cool, and add a great excess of caustic soda[1]; extract with ether. When the ether is driven off, the amine will crystallise. Recrystallised from alcohol, it has m.p. 46°. Yield: 85%.

p-Hydroxybenzyl Cyanide

p-Aminobenzyl cyanide . . .	6·6 gm.
Sodium nitrite	4·0 „

Fit a litre flask with a three-hole cork, carrying a thermometer and a dropping funnel; the thermometer and the stem of the dropping funnel should both almost touch the bottom of the flask. Introduce 200 c.c. water and 20 c.c. concentrated sulphuric acid; heat nearly to boiling and add the aminobenzyl cyanide. The sulphate of the base is at first precipitated but soon redissolves. Keep the solution just not boiling and run in gradually (through the long-stemmed) the sodium nitrite dissolved in 40 c.c. water. This addition take fifteen minutes, the temperature being 95° to 100°. Nitr n should be steadily evolved, unaccompanied by nitrous fumes. Add now 50 c.c. water and heat to boiling. Add 5 gm. bone-black to decolorise the solution; filter. Cool, extract twice with ether. Wash the extract with 25 c.c. saturated sodium bicarbonate solution, then with 25 c.c. water. Distil off the ether, finish the distillation under reduced pressure. The residue will form a crystalline mass. Purify by distillation *in vacuo*. B.p., 210°/10 m.m. Colourless crystals; m.p., 67°–71°. Yield: 5·5 gm., 83%.

Tyramine Hydrochloride

Hydroxybenzyl cyanide	5 gm.
Sodium	10 „
Alcohol, absolute	90 c.c.

Fit a 500-c.c. flask with a reflux condenser. Arrange it over a tripod and asbestos gauze. Put the cyanide and 50 c.c. alcohol in the flask, boil, and down the condenser drop the sodium, cut into tolerably big chunks, at five-minute intervals. Boil for half an hour, then add 20 c.c. alcohol to help in dissolving the sodium. Boil again for a quarter of an hour and add 20 c.c. more alcohol. When all the sodium has dissolved, reduce the volume to about 50 c.c. by evaporation under diminished pressure. Cool and make acid to litmus with

[1] Such an ether extraction as this is less troublesome if the tin double salt is made

hydrochloric acid. Extract with 200 c.c. ether to remove the cresol formed as a by-product. Make now strongly alkaline with sodium carbonate and extract twice with 150 c.c. amyl alcohol. Dry the extract over anhydrous sodium carbonate, filter, and shake it up three times with 100 c.c. (each time) *N* hydrochloric acid and finally with 100 c.c. water. Evaporate the collected acid extract to dryness under diminished pressure. The residue is almost pure tyramine hydrochloride. Yield : up to 3 gm.

To purify, dissolve the crude product with 1 c.c. concentrated hydrochloric acid in 25 c.c. boiling absolute alcohol. Filter hot. From the solution silky needles will crystallise. Filter and wash with a little absolute alcohol. M.p., 280°.

PREPARATION OF CALCIUM GLYCEROPHOSPHATE

When glycerol is esterified by phosphoric acid several esters are formed, a considerable number being theoretically possible. Most of them are split up again in the process and there remain only the α and β esters, discussed previously in connection with the phosphatides.

Phosphoric acid, crystallised . . .	300 gm.
Glycerine	300 ,,

Heat the mixture for forty hours at about 140°, shaking occasionally. The mass becomes viscous, black and frothy. Add 1,230 gm. water and about 200 gm. chalk, *i.e.*, until no more carbon dioxide is evolved ; filter ; wash the precipitate with a little water. Add milk of lime to the collected filtrate until it is weakly alkaline ; filter. Pass in carbon dioxide to saturation, so that the liquor is faintly acid ; filter. Evaporate down in a dish until an abundant separation begins. Add an equal volume of 80% alcohol to the liquor and filter off the product. To decolorise completely, take up this crude material in enough water to dissolve it, add a few grams of bone-black, filter, and slowly heat to 95° on the water-bath. Filter boiling, and wash with 50% alcohol.

Titration of Glycerophosphates.—Dibasic glycerophosphates, such as the commercial glycerophosphate, are alkaline to methyl orange, neutral to phenolphthalein, faintly alkaline to litmus. Monobasic glycerophosphates are neutral to methyl orange, acid to phenolphthalein. Before titration (of a glycerophosphate or free acid) neutralise, if necessary, exactly to phenolphthalein, then add methyl orange, and determine the remaining alkalinity.

Commercial calcium glycerophosphate is analysed as follows (François) :

Weigh out exactly 0·210 gm. of the product, dissolve it in 500 c.c. water, add a drop of methyl orange, and run in $\frac{N}{10}$ sulphuric acid until the colour change takes place. 0·210 gm. pure glycerophosphate should need exactly 10 c.c. $N/10$ acid. If, for example, only 8 c.c. is needed, the product contains only 80% of the mono-ester.

PREPARATION OF LECITHIN FROM EGG-YOLK

Egg powder, "dried egg" (Layton) [1]	100 gm.
Acetone	600 ,,
Alcohol, 98%	350 ,,
Ether	100 ,,

Put the egg-powder in a wide-necked 1-litre flask, add 260 gm. acetone, cork, shake; set aside for two hours. Drain in a perforated porcelain funnel, mix again with 100 gm. acetone; again drain, and repeat the treatment twice with 100 gm. acetone each time. Dry the powder in the air.

Treat it with 100 c.c. cold alcohol, set aside for three hours; filter, stir the paste again with 75 c.c. alcohol, and repeat this treatment twice. Collect the filtrates and evaporate them to dryness *in vacuo*. Grind the residue up with 50 c.c. acetone. The insoluble portion is the mixture of kephalin and true lecithin, which is generally known as lecithin. It is collected, spread out on a clock-glass, and dried in the vacuum-desiccator over sulphuric acid. Yield: 10–11 gm.

The separation of the lecithin and the kephalin is much too difficult an operation to be included in this course of practical exercises.

Alcoholysis of Lecithin (Fourneau and Piettre, 1912).—Prepare a solution of 25 gm. lecithin and 50 gm. methyl alcohol, cool it to 0°, and saturate with hydrogen chloride. Warm on the water-bath to about 50°, during an hour, until the excess of hydrochloric acid has been driven off. The liquid now forms two layers; the upper contains esters of the lecithin fatty acids, whilst glycerophosphoric acid and choline (chloride) are present in the lower layer. Carefully separate and wash the upper layer with a little water. Unite the aqueous solutions and extract with ether to remove the last traces of fatty substances. Add this ethereal extract to the oil, dry this ethereal solution over anhydrous sodium sulphate, and then over sodium carbonate. Distil off the ether and fractionate the residue under diminished pressure.

The thermometer will rise rapidly to 200° (at 18 m.m.). The fraction 200°–203° amounts to about 9 gm., and forms a clear liquid partly solidifying at about 12°. The temperature will again rise, and between 207° and 210° another fraction of 7·50 gm. may be collected. The last runnings, the temperature rising to 230°, amount to 1·5 gm., whilst the residue in the flask weighs 1·5 gm. Thus, altogether, 18 gm. of mixed esters is obtained.

[1] Or other make as used on the manufacturing scale.

The acid aqueous liquors, containing glycerophosphoric acid and choline, are evaporated *in vacuo* at a low temperature to remove most of the hydrochloric acid. The residue is taken up in 250 c.c. water and 10 gm. bone-black added. To the almost colourless filtrate precipitated chalk is added until no more will dissolve, then milk of lime until a faintly alkaline reaction is obtained; a few drops of very dilute hydrochloric acid are now added, so that the solution is just acid to phenolphthalein. Filter and evaporate the filtrate at a temperature below 55°. Stir the residue with absolute alcohol; this will dissolve the choline and calcium chlorides and leave the calcium glycerophosphate wholly behind. This salt, dried, will weigh 6 gm. Evaporate the alcoholic extract to dryness, dissolve the residue in water, and add just enough sodium carbonate to precipitate all the calcium present. Filter, evaporate to dryness, treat the residue with absolute alcohol. This will dissolve only choline chloride and hydroxyethylamine hydrochloride. These salts may be separated from the solution by evaporation to dryness and then form a crystalline mass, somewhat yellowish in colour, weighing 3·50 gm.

NUCLEIC ACIDS

PREPARATION OF NUCLEIC ACID FROM YEAST

Fresh yeast	100 gm.
Water	140 c.c.
Caustic soda, 36° Bé.[1]	16 gm.

Mix and leave for an hour at 14°.

Add 10 gm. acetic acid (80%) to neutralise, set aside for a day to settle, decant off part of the liquor, filter or centrifuge the rest. Pour the liquor (240 gm.) into a mixture of 250 gm. alcohol and 2 gm. hydrochloric acid. Filter off the precipitate; this in a moist state weighs 6·5 gm. Redissolve in 10 c.c. water and 5 gm. caustic soda solution, centrifuge, wash. Treat the clear solution with 0·80 gm. potassium permanganate and warm to 75°. Filter. Precipitate by adding 66 gm. alcohol (90%). 1·11 gm. sodium nucleinate will be obtained, or at the best, 2·5 gm.

PREPARATION OF NUCLEIC ACID FROM CALF-THYMUS[2]

(Neumann)

1 kgm. very fresh calf-thymus is quickly boiled with water acidified with acetic acid. As soon as the glands are hard enough stop the boiling and mince them in a meat chopper. Mix the mince with 2,000 c.c. water, 100 c.c. caustic soda (33%) and 200 gm. sodium acetate, and heat the mixture on the water-bath. After half an hour, the gelatinous acid, α, is obtained; the β-acid may be obtained by heating for 1½ hours. Neutralise with acetic acid (150 c.c. of 50% acetic acid is needed for 100 c.c. caustic soda), allow the liquor to settle, filter, evaporate down on the water-bath to 500 c.c. and pour into 500 c.c. 96% alcohol. Filter off the precipitate and redissolve it in 250 c.c. water. Warm till the suspended matter clots together; filter. Make a mixture of hydrochloric acid and alcohol in the proportion of 2 c.c. concentrated hydrochloric to 100 c.c. alcohol. Then to obtain the free nucleic acid pour the solution of the sodium salt into thrice its volume of this acid alcohol. Filter off and dry by washing with alcohol and ether. 1 kgm. calf-thymus yields 30 to 35 gm. acid.

[1] 33° Baumé = 1·332 sp. gr. (approx. 30% NaOH). The student is advised to become acquainted with the Twaddell and Baumé hydrometer scales; the first is widely used in technical practice in England and the second equally so on the Continent.—*Tr.*

[2] "Throat-sweetbread."

ALKALOIDS AND GLUCOSIDES

DETERMINATION OF THE NICOTINE-CONTENT OF TOBACCO

(From Bertrand, *Travaux pratiques de chimie biologique*)

The method consists essentially in extracting the alkaloid by boiling the tobacco with dilute acid, precipitating it as silicotungstate, liberating it again by the action of magnesia, and separating it by steam distillation. The distillate is titrated with acid, and so the amount of alkaloid present determined (G. Bertrand and Javillier, 1911).

Carefully weigh out 12 gm. of the tobacco, transfer it to a flask and cover it with twenty-five times its weight of 0·5% hydrochloric acid (say 300 c.c.). Boil gently under a reflux condenser for half an hour. (If a 500 c.c. flask be used, a wide glass tube, sufficiently long, will serve as the condenser.) Cool the flask under the tap, filter its contents through a wad of cotton wool. Measure out 250 c.c. of the filtrate and precipitate the alkaloid by adding silicotungstic acid or potassium silicotungstate (in 10% solution). Collect the dense precipitate by filtration or, better, by use of the centrifuge, suspend it in water containing a few drops of hydrochloric acid and of the precipitating reagent, filter or centrifuge again.

Transfer this washed nicotine silicotungstate to a small long-necked distilling flask. Add 3 gm. magnesium oxide suspended in a little water and steam distil. Care should be taken that the flask contents do not become diluted by condensation; the flask itself should be heated whilst the steam is passing through, so that the liquid becomes more and more concentrated; at the end it should amount to only a few c.c.

100 c.c. water more than suffices to carry over 100–200 mgm. nicotine. The quantity present in the distillate is determined volumetrically. A standard solution of sulphuric acid is used, such that 1 c.c. is equivalent to 10 mgm. nicotine (*i.e.*, 3·024 gm. H_2SO_4 per litre), and as indicator alizarine-sulphonic acid, the colour changing from violet-red to yellow. The result gives directly the nicotine present in 10 gm. tobacco.

PREPARATION OF ATROPINE

Grate 500 gm. fresh belladonna root to as fine a pulp as possible and add 25 gm. dry sodium carbonate. Triturate the mixture thoroughly, transfer it to a flask, and shake it for five minutes with 800 c.c. of a

mixture of 100 c.c. chloroform and 400 c.c. ether. Decant off the liquid, shake it with 50 c.c. 10% hydrochloric acid, wash it with water, and return it to the flask containing the crushed root. Extract three times in this way, using the same ether-chloroform mixture, and carefully preserving the successive acid extracts. Unite these, treat the solution with 3 gm. animal charcoal to decolorise it, filter and evaporate down under diminished pressure at 20°. Add ammonia in very small excess and extract with chloroform. Allow the chloroform to evaporate; the oily residue, 1·3–1·6 gm., will crystallise after a time, or if it be seeded. If it does not crystallise, redissolve it in 5–7 c.c. warm 90% alcohol, add a pinch of bone-black, and filter hot into 30 c.c. cold water. The oily precipitate will soon solidify.

EXAMPLE OF AN OXIDATION IN THE ALKALOID SERIES—

Oxidation of Quinine Sulphate

(Oxidation of an ethylenic side chain, $-CH:CH_2$ to $-CO_2H$.)

According to Bucher and Rudolf [1] four atomic proportions of oxygen are needed, and the reaction takes place thus:

$$R{\cdot}CH:CH_2 = R{\cdot}CO_2H + H{\cdot}CO_2H.$$

Actually, it has been found in various experiments that a better yield is obtained when five atomic proportions of oxygen are used, so the reaction is really in accordance with the equation:

$$R{\cdot}CH:CH_2 = R{\cdot}CO_2H + H_2O + CO_2.$$

Quinine sulphate ($+ 8H_2O$).	44·5 gm.	
Potassium permanganate (for five atoms of oxygen)	52 ,,	(in 5% solution)
Sulphuric acid, H_2SO_4.	16 ,,	(in 10% solution)

Mix the quinine sulphate and the sulphuric acid; all will not dissolve. Add the potassium permanganate solution little by little, keeping the temperature of the mixture about 0° (it should never rise above 10°). Immediate decolorisation takes place. When all the permanganate has been added, set aside for some hours; filter. The filtrate contains only unattacked quinine, the quitenine (chitenine) and MnO_2 remain behind.

Extract the precipitate three times with boiling water, filtering hot. The acid will crystallise as the solution cools. The residue may be extracted again with dilute sodium hydroxide solution and the filtrate exactly neutralised. As an alternative, the extraction may be carried out all at once by using dilute caustic soda. The solution obtained, containing the sodium salt of quitenine, must be acidified (to congo-red) with hydrochloric acid, then neutralised with ammonia, a slight excess of the latter being used. The product will speedily crystallise out in small colourless prisms, readily adhering to the walls of the beaker.

[1] *Monatshefte für Chemie*, 1893, *14*, 598.

PREPARATION OF DIACETYLMORPHINE HYDROCHLORIDE
(*Heroin*)

Morphine	3·5 gm.
Acetic anhydride	7 ,,

Heat in a water-bath at 85° for six hours. Distil off the acetic acid and excess acetic anhydride under diminished pressure. Dissolve the solid residue in 17 c.c. water, decolorise with bone-black, precipitate with ammonia.

Crude diacetylmorphine	4 gm.

Recrystallise from five parts of absolute alcohol. The pure product, 3·5 gm., has m.p. 169°–170°.

Hydrochloride.—Dissolve the diacetylmorphine (3·53 gm.) in five parts of boiling acetone. Filter and add the requisite amount of an ethereal solution of hydrogen chloride (titrated). The hydrochloride will crystallise out; filter it off and wash with acetone. 3·26 gm.

PREPARATION OF DIGITALIN(E) [1] (French *Codex*, 1880)

Digitalis	500 gm.
Water	500 ,,
Neutral lead acetate	125 ,,

Dissolve the lead acetate in the water and grind together the digitalis and the solution. Leave the mixture to stand overnight. Add enough 50% alcohol to make a total volume of 2 litres. Set by again for a day. Transfer the mixture to a percolating apparatus and extract by displacement three times. Final volume of liquid: 4 litres. Add 20 gm. sodium bicarbonate. Evaporate down to 1,200 c.c., then add water to make up the volume to 2 litres; set by for two days. Siphon the clear liquid away and filter off the precipitate. (This will take a long time, and it is better to use a centrifuge if one is available.)

Suspend the precipitate in 500 gm. 80% alcohol; boil. Leave overnight and boil up again. Add 5 gm. neutral lead acetate, and 10 gm. bone-black. Boil up again and filter, washing the residue with alcohol. The solution obtained has a deep greenish-brown colour. Distil off the alcohol, adding, before it has all evaporated, 28 gm. powdered wood charcoal. The cold charcoal residue so obtained is then ground up with a little water, filtered, washed, dried and extracted for a day in a Soxhlet apparatus with chloroform. The chloroform is distilled off.

The *residue from the chloroform extract* is to be dissolved in 50 gm.

[1] Nomenclature is discussed in the article "Digitalis" in Thorpe's *Dictionary of Applied Chemistry*, II (1921). This article should be consulted.—*Tr.*

90% alcohol. Add 0·50 gm. lead acetate, 0·50 gm. bone-black, and boil for ten minutes. Cool, filter, evaporate the filtrate to dryness. Take up again in 5 gm. alcohol; add 2·5 gm. ether, 7·5 gm. water, and seed the mixture with a morsel of isolated digitalin.

Gradually the product will separate, accompanied at first by a little oil. Filter it off on the following day, wash first with ether (the oil will pass through the filter-paper), then with water. . . . . 0·28 gm.

The ether-alcohol mother liquors (50 c.c.) yield, on standing overnight, another small crop, adhering to the walls of the vessel. At the bottom of the vessel will be found some gummy substance. Filter off the digitalin . . . . . 0·09 gm.

Total yield: 0·37 gm., equivalent to 0·74 gm. per kgm. of leaves.

Redissolve this product in twenty times its weight of chloroform, 0·08 gm. remains undissolved. Evaporate to dryness, take up the residue in 3 gm. alcohol, and reprecipitate with ether and water, as before. The product now weighs 0·21 gm.

Treatment of the crude digitalin from the first alcohol-ether precipitation.—Dissolve the powdered substance in twenty times its weight of chloroform (a considerable proportion is insoluble), filter, evaporate to dryness *in vacuo*. Redissolve the residue in twenty parts of hot 90% alcohol, treat with bone-black; filter, wash. Evaporate down to 20 c.c., add an equal volume of ether, then of water. Set aside for some hours. Part of the digitalin will separate. Add more water and more ether, little by little, to make forty "parts" in all, the volumes of the ethereal and aqueous layers being about equal. Filter after two days, wash with ether and water; dry. Redissolve in twenty parts of chloroform, filter off the insoluble portion, evaporate to dryness. Take up again in—

10 parts alcohol,
5 parts ether,
10 parts water.

A homogeneous mixture, not two layers, will be obtained. Set aside for two days to precipitate.

PREPARATION OF BETAINE HYDROCHLORIDE (*Acidol*)

Chloracetic acid.
Ethyl chloracetate.
Trimethylamine.
Betaine hydrochloride.

Chloracetic Acid

Acetic acid, glacial	150 gm.
Sulphur	35 ,,

Fit a 500-c.c. flask with a rubber bung carrying (i) a leading tube for chlorine; (ii) a safety-tube also projecting into the liquid; (iii) an upright condenser to which is connected a tube to lead the evolved gases into milk of lime. Heat the mixture of acetic acid and sulphur gently, not above 100°, and pass in chlorine (from a laboratory generator or from a cylinder of compressed gas). The apparatus should be exposed to sunlight or to an electric light of at least 50 c.p. Continue to pass chlorine until a drop of the liquid in a test tube will solidify when cooled. According to the strength of the illumination, the operation will take from six hours to two days. Distil, collecting that which passes over from 150° to 195°. Set this to crystallise in the ice-chest. Filter quickly. Redistil the filtrate, collecting the fraction 170°–180° this time; repeat the operations, making a final fractionation at 180°–190°.

Chloracetic acid has b.p. 186°. Yield: 80–125 gm.

Ethyl Chloracetate

Chloracetic acid	150 gm.
Absolute alcohol	100 ,,
Sulphuric acid, sp. gr. 1·84	15 ,,

Fit a 500-c.c. flask with a reflux condenser, mix the chloracetic acid and the alcohol together in the flask and add the sulphuric acid gradually. Heat on the water-bath for three hours. Cool, add 450 c.c. water, separate, wash the oil with 200 c.c. water, dry it over calcium chloride. Distil. B.p., 144°–144·5°. Yield: 140 gm.

Trimethylamine

(1) *Hydrochloride.*

Ammonium chloride	100 gm.
Trioxymethylene	265 ,,

Mix the reagents in a 500-c.c. flask, attach a reflux condenser, and heat the mixture at first on the water-bath, then in an oil-bath, so

that the temperature rises gradually to 130°. Keep there for two hours. Carbon dioxide begins to be evolved at about 125°, but eventually the evolution of gas ceases. Cool; the flask contents will set to a crystalline mass. The reaction is almost quantitative. The product may be purified by dissolving it in half its weight of hot water. Cool in ice and filter quickly. Do not wash the crystals.

(2) *Benzene Solution of Trimethylamine.*

Caustic soda in small lumps	200 gm.
Trimethylamine hydrochloride	100 „
Water	100 „

Fit a 1,000-c.c. flask with a dropping funnel and reflux condenser; connect the upper end of the condenser, *viâ* a small empty flask, with a drying bottle containing sticks of caustic soda; attach to the exit of the drying bottle first another empty flask, then a three-neck Woulff's bottle; fit the second neck of the bottle with a safety tube, the third with a tube dipping into a small flask. Put 200 c.c. dry benzene in the Woulff's bottle and a small quantity, to cover the end of the leading tube, in the last small flask (*cf.* Fig. 22).

Immerse the flask, containing the 200 gm. caustic soda, in a pan of boiling water, and run into it, drop by drop, the solution of trimethylamine hydrochloride. Some trouble may be caused by sucking back, so care should be taken to add the solution in a steady stream.

The benzene solution of trimethylamine may be titrated, like an alkali, with *N* sulphuric acid. 10 c.c. *N* acid are equivalent to 0·59 gm. trimethylamine.

Ethyl Dimethylaminoacetate Methochloride

Benzene solution of trimethylamine (at 20% = 6 gm. trimethylamine)	30 c.c.
Ethyl chloracetate	10 gm.

Mix the reagents in a 150-c.c. bottle fitted with a screw or clip stopper (*e.g.*, a beer bottle) *at a low temperature.* Quickly stopper the bottle. The mixture rapidly becomes hot and a white precipitate separates. After some hours the contents of the bottle will have formed a crystalline mass. Wrap the bottle in a stout cloth and heat it for an hour in a water-bath to carry the reaction to completion. Cool, filter, and wash the product with ether. A theoretical yield of ethyl dimethylaminoacetate methochloride is thus obtained.

This product is hydrolysed by treatment with boiling 20% hydrochloric acid.

Betaine Hydrochloride

Ethyl dimethylaminoacetate methochloride	15 gm.
Hydrochloric acid, 20%	100 c.c.

Boil together under reflux for three hours. Evaporate to dryness

under diminished pressure. Take up the residue in boiling alcohol. Splendid colourless crystals separate on cooling. Yield : 93%.

This hydrochloride, the trade name of which is *Acidol*, is acid to litmus and may be titrated like a strong acid. It is chiefly used mixed with pepsin in the form of tablets (*Acidol-pepsin*).

PREPARATION OF SODIUM CINNAMATE (*Hetol*)

Benzaldehyde.
Benzylidene-acetone.
Cinnamic acid.

Preparation of Benzaldehyde (*Sommelet*)

The process is based on the reaction between benzyl chloride and hexamethylenetetramine in aqueous-alcoholic solution. The formation of benzaldehyde is to be regarded as taking place in two stages: the first stage corresponds with the combination, molecule for molecule, of benzyl chloride and hexamethylenetetramine, a quaternary ammonium salt, the benzochloride of the base, being formed, thus,

$$C_6H_5{\cdot}CH_2Cl + C_6H_{12}N_4 = C_6H_{12}N_3 : N\begin{matrix}CH_2{\cdot}C_6H_5\\ Cl.\end{matrix}$$

In the second stage this salt undergoes drastic hydrolysis, benzylamine and other bases, the molecules of which contain the groupings $\cdot CH_2 : N\cdot$ or $\cdot CH_2\begin{matrix}N :\\ N :\end{matrix}$, being formed. The latter, under the conditions of the reaction, dehydrogenate the benzylamine, converting it into an imino base which is immediately hydrolysed to benzaldehyde and ammonia, thus:

$$C_6H_5{\cdot}CH_2\begin{matrix}\\ Cl\end{matrix}\!\!>N : C_6H_{12}N_3 \longrightarrow C_6H_5{\cdot}CH_2{\cdot}NH_2 + CH_2\begin{matrix}N =\\ N =\end{matrix}$$

$$\longrightarrow C_6H_5{\cdot}CH{=}NH \longrightarrow C_6H_5{\cdot}CHO + NH_3.$$

Hexamethylenetetramine . .	35 gm. ($\frac{1}{4}$ mol.)
Benzyl chloride	32 ,,
Alcohol, 80%	300 c.c.

Fit a litre flask with a reflux condenser, introduce the reagents and boil on the water-bath for four hours. Distil off most of the alcohol (the distillate is alkaline). The residuum in the flask separates into a lower aqueous layer and an upper oily layer containing the benzaldehyde. Add about 100 c.c. water and extract several times with ether. Unite the ethereal extracts and wash them with a little 10% sulphuric acid to remove a small amount of basic substances. Remove the ether by distillation and shake the oil with an excess of a concentrated solution of sodium bisulphite; a voluminous crystalline separation will form. Set aside for several hours, filter the product off and

wash with a little alcohol. Decompose the bisulphite compound by treating it with a small excess of sodium carbonate or dilute sulphuric acid. Extract the aldehyde with ether. B.p., 178°–180°. Yield: about 70% of the theoretical.

An alternative, perhaps simpler, procedure is to add 10% sulphuric acid in excess to the residue in the flask after the alcohol has been distilled off, then to steam distil. Extract the distillate with benzene, remove the benzene and purify the crude aldehyde through the bisulphite compound as above.

Cinnamic Acid

1st Method.

(i) **Benzylidene-acetone** (Methyl styryl ketone), $C_6H_5 \cdot CH : CH \cdot CO \cdot CH_3$.

A	Benzaldehyde	15 gm.
	Water	1,350 ,,
	Acetone	30 ,,
B.	Caustic soda, 10%	15 ,,

Agitate the mixture A vigorously and add the caustic soda B gradually. Leave together for four days, shaking frequently. Extract with ether, dry the extract over calcium chloride, distil. After removing the ether, continue the distillation under diminished pressure. Collect that which passes over up to 160°; the residue is dibenzylidene-acetone.

Benzylidene-acetone has b.p. 151°–153°/25 m.m.; m.p., 41°–42°. Dibenzylidene-acetone, $CO(CH : CH \cdot C_6H_5)_2$, has m.p. 112°.

(ii) **Oxidation of the Benzylidene-acetone.**

Benzylidene-acetone	5 gm.
Bleaching powder, with about 30% available chlorine	12 ,,
Anhydrous sodium carbonate	15 ,,

From these reagents prepare an approximately 5% sodium hypochlorite solution, warm to 80°–90°, and add the benzylidene-acetone. Shake vigorously in a stout flask, unstoppering from time to time to allow carbon dioxide and chloroform to escape. Keep the temperature at 80° and continue to shake until all the oil is dissolved. Cool, add dilute sulphuric acid to precipitate the cinnamic acid. Filter, wash, recrystallise from water. M.p., 133°.

Yield: 4 gm., including that extractible from the mother liquor by ether.

2nd Method (*Application of Perkins' Reaction*).

Benzaldehyde, redistilled . . .	25 gm.
Sodium acetate, fused [1] . . .	12·5 ,,
Acetic anhydride	37·5 ,,

Heat the mixture under reflux for eight hours at 180°. Pour into cold water. Steam distil to remove unchanged benzaldehyde and to convert the excess of acetic anhydride into acetic acid. Add sodium carbonate and decolorise with bone-black. Filter and precipitate by adding hydrochloric acid. Yield: 15 gm.

To prepare *Hetol*, dissolve cinnamic acid with the exactly necessary amount of sodium hydroxide (titrate) and evaporate the solution to dryness.

[1] Fused sodium acetate is prepared by heating the crystalline salt ($3H_2O$) in a dish (a small enamelled iron bowl is a suitable vessel.—*Tr.*) over a naked flame. First the crystals melt in their water of crystallisation, then, as this evaporates, a white granular mass remains; on further heating (the mass should be stirred continuously) the whole fuses. It is then poured out on to a plate or sheet of enamelled iron and quickly powdered.

PREPARATION OF ALLYLTHIOUREA (*Thiosinamine*)

Allyl iodide.
Allyl *iso*-thiocyanate (Mustard oil).
Thiosinamine.

Allyl Iodide

Iodine	105 gm.
Glycerine	420 „
Phosphorus, red	38 „

Fit a 2½-litre tubulated retort with a dropping funnel and arrange it so that the beak projects well into a long-necked flask cooled by a stream of water. Put the glycerine and the phosphorus in the retort, warm gently and shake to distribute the phosphorus in the liquid. Add 15 gm. iodine and shake again. Warm carefully over a naked flame and distil over about 25 gm. of liquid. Add this distillate to the remainder of the iodine in a small flask and transfer the saturated solution of iodine so obtained to the retort through the dropping funnel. Repeat the operations until the whole of the iodine has been introduced. Distil the mixture as far as possible. The distillate will form two layers, allyl iodide being at the bottom with a mixture of allyl alcohol and water above. Separate the lower layer, wash it with a little dilute caustic soda, and dry over calcium chloride. Rectify; b.p., 101°–102°. Yield: 91 gm.

The upper layer may be distilled and the allyl alcohol used to prepare allyl bromide, from which the mustard oil may equally well be obtained.

Mustard Oil

Allyl iodide	16 gm.
Ammonium thiocyanate	8 „
Alcohol, 97%	35 „

Boil under reflux for half an hour. Add 100 c.c. water. Extract with 100 c.c. ether. Wash the extract with 40 c.c. water, dry over anhydrous sodium sulphate. Distil; nearly all passes over at 145°–150°. Yield: 8 gm.

Thiosinamine

Allyl *iso*thiocyanate	8 gm.
Ammonia, 20% aqueous solution	30 c.c.

Shake together and warm gently until a homogeneous solution is formed. Evaporate down at a low temperature. The residue will crystallise, particularly readily if it be seeded. Recrystallise the crude product, after it has been carefully dried, from a mixture of ethyl acetate and benzene. M.p., 78°.

PREPARATION OF *p*-IODOANISOLE

(Replacement of the group $\cdot NH_2$ by I—Sandmeyer.)

$$CH_3O\cdot C_6H_4\cdot N_2Cl \xrightarrow{KI} CH_3O\cdot C_6H_4\cdot I + N_2 + KCl.$$

A	*p*-Anisidine	12	gm.
	Hydrochloric acid, concd.	90	,,
	Water	150	,,
	Ice	*q.s.*	
B	Sodium nitrite	8	,,
	Water	50	,,
C	Potassium iodide	25	,,
	Water	50	,,

A thick-walled glass beaker or earthenware mug of about 500 c.c. capacity, fitted with a mechanically-driven stirrer.

Make up the solution A in the mug, adding enough ice to give a temperature of 5°. Stir vigorously and run in the solution B slowly. The mixture turns violet in colour, then yellow, eventually a pale brown. When a drop on starch-iodide paper gives definitely the blue spot indicating free nitrous acid, add no more nitrite.

Transfer the diazo solution to a litre flask and add the solution C all at once. A precipitate forms, but no other reaction takes place. Set by for three hours, then heat up gradually on the water-bath. Nitrogen is evolved, and oily globules separate. Make the liquor weakly alkaline, cool. Filter off the crystals, dry them in the air and recrystallise from low b.p. petrol. M.p., 51°–52°. Yield: 20 gm.

p-Iodoanisole is used to prepare *p*-iodoxyanisole—*Isoform*,

$$CH_3O\cdot C_6H_4\cdot IO_2.$$

This compound is explosive and so its preparation will not be included in our list.

BIBLIOGRAPHY [1]

SOME references to the literature are given in the text; the abbreviations used should be self-explanatory. In most cases, even if only a date is given, the *Abstracts* section of the *Journal of the Chemical Society* will either give enough additional information or show where it can be found.[2]

Many of the works referred to below contain copious references, and both for these and for further general information the various articles in Thorpe's *Dictionary of Applied Chemistry*, 1921–24 (London: Longmans), may be consulted.

Valuable summaries of progress made during each year will be found in the *Annual Reports of the Progress of Applied Chemistry*, issued by the Society of Chemical Industry (Vol. I, 1916), Fine Chemicals, Medicinal Substances and Essential Oils Section (see references below).

General.—A general account chiefly from the chemical point of view will be found in *The Chemistry of Synthetic Drugs*, by P. May, 3rd edition, 1921 (London: Longmans); the manufacturing aspect of the subject is treated in *Organic Medicinal Chemicals (Synthetic and Natural)*, by M. Barrowcliff and F. H. Carr, 1921 (London: Ballière, Tindall and Cox); and the medical side may be approached through Martindale and Westcott's *Extra Pharmacopœia* (London: Lewis).

Chapters I–IV.—See *Applied Chemistry Reports*, I, 276, 279; II, 474; III, 436; IV, 500. The industrial preparation of chlorobenzene, chloronitrobenzene, *o*-nitroanisole, *p*-aminophenol, salicylic acid, dimethylaniline, etc., is described in J. C. Cain's *Manufacture of Intermediate Products for Dyes*, 2nd edition, 1919 (London: Macmillan).

Chapter V.—*Applied Chemistry Reports*, VII, 498, 500; VIII, 526, 531, and earlier volumes.

Chapter VI.—*Applied Chemistry Reports*, I–VI, and VIII, 528.

Chapter VII.—*Applied Chemistry Reports*, VI, 505, 506; VIII, 533.

Chapter VIII.—*The Organic Compounds of Arsenic and Antimony*, by G. T. Morgan, 1918 (London: Longmans, Monographs on Industrial Chemistry), *Applied Chemistry Reports*, VI, 530; VIII, 533, etc.

Chapter IX.—*Organic Compounds of Mercury*, by F. C. Whitmore, 1921 (New York: Chemical Catalog Co., Inc., A. C. S. Monographs), *Applied Chemistry Reports*, V, 503; VII, 507.

[1] Translator's addition.

[2] The patent and technical literature is not completely covered.

Chapter X.—*The Simpler Natural Bases*, by G. Barger, 1914 (London: Longmans, Monographs on Biochemistry); see also *Applied Chemistry Reports*, II, 492; V, 492. For Thyroxin, see *ibid.*, III, 451.

Chapters XI, XII.—*Lecithin and Allied Substances*, by H. Maclean, 1918 (London: Longmans, Monographs on Biochemistry); *Nucleic Acids*, by W. Jones, 2nd edition, 1920 (in the same series); see also *Applied Chemistry Reports*, VII, 494; VIII, 520.

Chapter XIII.—See all *Applied Chemistry Reports*, the *Annual Reports of the Chemical Society*, and Henry's *Plant Alkaloids* (London: Churchill).

Chapter XIV.—The chapter (XV) on Protective Synthesis in Parsons' *Fundamentals of Biochemistry*, 1923 (Cambridge: Heffer), is of interest.

Practical Work: The following books, among others, will be found useful:

Preparation of Organic Compounds, by E. de B. Barnett, 2nd edition, 1920 (London: Churchill).

Organic Syntheses (*an annual publication of satisfactory methods for the preparation of organic compounds*), by R. Adams, J. B. Conant, H. T. Clarke, and O. Kamm, Vols. I–III, 1921–23 (New York: Wiley);

and a vast number of recipes is given in:—

L. Vanino's *Handbuch der präparativen Chemie, II, Organischer Teil*, 1923 (Stuttgart: Enke).

In experimental work on a larger scale than is usual in an organic chemistry laboratory, Mason's translation (or the original) of Fierz-David's *Fundamental Processes of Dye-Chemistry*, 1921 (London: Churchill), will be helpful.

INDEX

The Index is alphabetical: nitrohydroxyphenylarsinic acid, for example, is put under N. The Chemical Society's nomenclature is used except for, *e.g.*, nitraniline, where the shorter form is well established; or for, *e.g.*, diethylacetic acid, where confusion would arise. On the pages indicated by figures in heavier type practical details will be found.

Acetals, 45
Acetanilide, 19, **205**
Acetic anhydride, **199**
Acetophenone, 46, **223**
Acetotoluidide, 20
Acetyl chloride, **198**
,, nitrate, **194**
Acidol, **248**
Acoine, 55, 64
Acriflavine, 79
Adaline, 47, **229**
Adenine, 154
Adenosinic acid, 154
Adrenaline, 117, 119, **230**
,, , physiological action, 122
Afridol, 114
Airol, 78
Aldogen, 75
Algolan, 32
Alkaloids, definition, extraction, etc., 161
Allyl iodide, *iso*-thiocyanate, etc., **254**
Aluminium amalgam, reduction with, **11**
Alypine, 62
p-**Aminobenzoic** acid, **219**
p-**Aminobenzyl** cyanide, **236**
5-**Amino-2** : 4-**dihydroxyphenylarsinic** acid, **107**
p-**Aminophenol**, 11, **202**
Amylene glycol, 43
" **Amylene** hydrate," 43
Amylurea, 47
Anæsthesine, 55, 62, **220**
Anæsthetics, 42
,, , local, 68
Analgen, 28
Analgesine (*see* Antipyrine).
Aniline, **204**
o-**Anisidine**, 1, **195**
Antifebrine (*see* Acetanilide).
Antipyretics, action of, 15
Antipyrine, 18, 33, **207**
Antiseptics, 70
Antodyne, 24
Apolysin, 22
Aponal, 47
Apothesine, 64
Aristol, 75
Aristoquinine, 27
Arrhenal (*see* Methylarsinic acid).
Arsacetine, 96
Arsalyte, 99
Arsanilic acid (*see* Atoxyl).
Arsenicals, experimental chemotherapy of, 99
Arsenobenzoylquinine, 27
Arseno compounds, 90
Arsenobenzol (*see* Salvarsan).
Arsenophenylglycine, 96
Arsinic acids, 87
Arterenol, 121, 122
Asciatine, 46
Aseptol, 73
Aspirin, 18, 31, **209**
Asurol, 113
Atoxyl, 86, 95, **235**
Atropine, **244**
Autan, 75
Azophenetole, **201**

" **Bayer** 205," 74, 80
Benzaldehyde, **251**
Benzoic acid, **216**
Benzosal, 9
Benzosalin, 32
Benzoyl chloride, **217**
Benzyl alcohol, 64
,, cyanide, **237**
Benzylidene-acetone, **252**
Betaine (*see* Acidol).
Betol, 31
Bismal, 78
Bornyval, 46
Brilliant green, 79
Bromobenzene, **215**
Bromodiethylacetyl chloride, **229**
,, urea (*see* Adaline).
Bromonitrobenzene, 3, **200**
Bromovaleryl chloride and bromide, **225**
,, urea (*see* Bromural).
Bromural, 47, **224**
Butyn, 69

Cacodyl, 82
Cacodylic acid, 83
Carbolic acid, 72
Carnaubon, 137
Catechol, 5
Chavasol, 73
Chinosol, 74
Chloracetic acid, **248**
Chloracetocatechol, **230**
Chloracetone, **210**

Chloracetyl chloride, use of, 9
Chloral hydrate, 45
Chloralose, 46
Chloramine T, **77**
Chloretone, 44
Chlorohydroxy*iso*butyric acid, **221**
„ „ „ ethyl and propyl esters, **222**
4-Chloro-2-methylphenylarsinic acid, **106**
Chloronitrobenzene, 3, 4
o-**Chlorophenol**, 7
Choline, 142
Cinnamic acid, **252**
Cocaine, 55, 56
Collargol, 78
Copper salts (of alkaloids), **165**
Coryfin, 78
Cosaprin, 21
Cotarnine, 134
" **Coupling** " (of diazo compounds), **14**
Creoline Pearson, 73
Cryogenin, 17, 18, 25
Crystallisation, **185**
Cuorin, 137
Cupreine, 26
Cytidinic acid, 154
Cytosine, 154

Dermatol, 78
Dial, 48
Diaspirin, 32
Dichloramine T, **77**
Diethylacetic acid, **227**
Diethylmalonic acid, **227**
Diethylmalonylurea (*see* Veronal).
Digitaline, **246**
2 : 4-Dihydroxyphenylarsinic acid, **106**
Dimethylamine, **213**, **214**
Dimethylaniline, **211**
Dioform, 43
Diplosal, 32
Distillation, **179**
Dormiol, 46
Dulcin, 22
Duotal (*see* Guaiacol carbonate).

Eau de Javel, 76
Eccaine, 56, 64
Ecgonine, 56, 57
Elimination, by the organism, 8, 20, 177
Enesol, 113
Ephedrine, 131, 135
Epicarin, 73
Epinine, 122
Ergamine (*see* Iminazolylethylamine).
Ether, 44
Ethone, 45
Ethyl bromide, **199**
„ diethylmalonate, **227**
„ dimethylaminoacetate (methochloride), **249**
„ ethylmalonate, **227**
Eucaines, 59, 60
Eucupin, 27
Eudoxine, 78
Euphorine, 21
Euphtalmin, 60
Euquinine, 27
Europhen, 76
Exalgine, 21
Excretion (*see* Elimination).

Formalin, 75
Formanilide, 20

Geosote, 9
Glycerophosphoric acid, 140, **240**
Glycine, 12
Glycosal, 32
Grignard's reaction, 44, 62, **214**, etc.
Guaiacol, 1, 6, **195**
„ carbonate, 9, **196**
„ *o*-sulphonic acid, 9, **196**
Guaiacophosphal, 9
Guaiasanol, 9
Guanine, 154
Guanylic acid, 153

Halazone, 80
Hectine, 96
Hedonal, 47
Hermophenyl, 114
Heroin, **246**
Hetol, **251**
Histamine (*see* Iminazolylethylamine).
Hofmann's reaction, 168, **170**
Homorenon, 122
Holocaine, 55, 64
Hordenine, 126, 130
Hydrastine, 133
p-**Hydroxybenzyl** cyanide, **238**
4-Hydroxy-2-carboxyphenylarsinic acid, **107**
p-**Hydroxyphenylarsinic** acid, **235**
Hypnal, 39, 46
Hypnone (*see* Acetophenone).
Hypnotics, action of, 49, 51
Hypoxanthine, 154

Ichthyol, 77
β-**Iminazolylethylamine**, 128
β-**Indolethylamine**, 130
Inosinic acid, 154
Intramine, 77
p-**Iodoanisole**, **255**
Iodofan, 76
Iodoform, 75
Isoform, 76, **255**
Isopral, 44

Jecorin, 138

Kairin, 28
Kairolin, 28
Kephalin, 147
Kharsivan (*see* Salvarsan).

Lactophenin, 17, 22
Laudanosine, 134
Lecithin, 136, 139, 145, **241**
Lodal, 134
Losophan, 76
Luargol, 99
Ludyl, 99
Luminal, 48
Lupinic acid, **167**
Lupinine, 167, 170
Lysocithin, 143, 149, **150**
Lysol, 73

Malachite green, 79
Malonic ester, **226**
Maretin, 25
Melubrin, 25, 39
Mercuration, 112
Mercurials, 108, **232**
Mercuridipropionic acid, 111
" **Mercury** benzoate," **232**
Mercury diaminodihydroxydiphenyl, 115, **232**
Mesotan, 32
Methylaminoacetocatechol, 119, **231**
Methylaniline, **211**
Methylarsinic acid, 83
Methylchloroform, 43
Methylene chloride, 43
Methylethylchloromethylcarbinol, **214**
Methylethyldimethylaminomethylcarbinol, **215**
Methyl iodide, **193**
,, salicylate, 32
Methylmercuric iodide, 111
Metol, 12
Microcidin, 73
Monotal, 9
Morphine, 42, **243**

Narceine, 134
Narcotine, 134
Neosalvarsan, 98
Neottin, 137
Neuronal, 47
Nicotine, determination of (in tobacco), **244**
Nirvanine, 63
Nirvanol, 47
o-**Nitraniline**, 2
p-**Nitraniline**, 10
Nitration, rules of, 3
,, with acetyl nitrate, 5, **195**
o-**Nitroanisole**, 1, 4, **193**
Nitrobenzene, **204**
p-**Nitrobenzyl** cyanide, **237**
4-**Nitro-2-carboxyphenylarsinic** acid, **106**
5-**Nitro-2 : 4-dihydroxyphenylarsinic** acid, **106**
3-**Nitro-4-hydroxyphenylarsinic** acid, **236**
5-**Nitro-2-methylphenylarsinic** acid, **105**
Nitrohydroxyphenylmercuric acetate, **233**
p-**Nitrophenetole**, 1, 10, **198**
o-**Nitrophenol**, 1, **192**
p-**Nitrophenol**, 1, 10, **197**
Nitrosodimethylaniline, **212**
Nitrotoluene, **219**
Nosophen, 76
Novarsenobenzol, 98
Novaspirin, 32
Novocaine, 56, 63
Nucleic acids, 153, **243**
,, ,, , physiological function, 159
Nucleosides, 157

Optochin, 27
Osarsan, 97
Orthoform, 55, 63
Overton-Meyer theory, 52
Oxidation in the organism, 175
Oxychinaseptol, 74

Pental, 43
Peruol, Peruscabin, 75
Phenacetine, 1, 10, 17, 21, **202**
p-**Phenetidine**, 11, 13, **201**
p-**Phenetoleazophenol**, 13, **200**
Phenocoll, 22
Phenol, 72
Phenolsulphonic acids, 6
β-**Phenylethylamine**, 128
Phenylglycine, 20
Phenylhydrazine, 24, **206**
Phenylmethylethoxypyrazole, 37
Phenylmethylpyrazolone, 37, **206**
Phosphatides, 136
Pinacones, 44
Profiavine, 80
Proponal, 48
Propyl dimethylaminohydroxy-*iso*-butyrate, **222**
Protagon, 138
Protargol, 78
Providoform, 74, 77
Psicaine, 69
Pyramidon, 18, 39
Pyrazolone, 33
Pyrocatechol (*see* Catechol).

Quietol, **221**
Quinine, 26, 162, **245**
Quinotoxin, 27
Quitenine, **245**

Reduction, in the organism, 176
Rivanol, 80
Rodinal, 12

Salacetol, 32
Salen, 32
Salicylic acid, 29, 30, **208**
Salipyrin, 39
Salol, 31
Salophen, 31
Salvarsan, 97, **238**
Scopolamine, 42
Solveol, 73
Soneryl, 48
Sozoiodol, 73, 76
Sphingomyelin, 139, 148
Spirosal, 32
Stovaine, 55, 60, **218**
Stovarsol, 105
Stypticine, 134
Sulphonal, 48
Sympathomimetic action, 124

Tetralol, 73
Tetronal, 49
Thalline, 28
Thermodin, 23
Thermopuncture, 16
Thiocol (*see* Guaiacol-*o*-sulphonic acid).
Thymus nucleic acid, 158, **243**
Thiosinamine, 77, **254**
Tolylarsinic acid, **105**
Triacetonamine, 59
Trigemin, 46
Trimethylamine, **248**
Trional, 49
Tropacocaine, 55, 58
Tropigenine, **166**
Tropinic acid, 57
Tropinone, 57, **166**
Trypaflavine, 79
Tyramine, 126, 128, **238**

Ulmaren, 32
Uracil, 154
Urethane, 21
Uridinic acid, 154
Urotropin, 75

Valeric acid, 46, **224**
Valeryl chloride and bromide, **224**
Validol, 47
Veratrole, 6
Veronal, 48, **227**
Vesalthin, 137
Viferral, 46
Vinyldiacetonamine, 59
Vioform, 76
Vuzin, 27, 72

Xeroform, 77

Yeast nucleic acid, 155, **243**

Zeisel's method, 164, 165

CORRECTION.

P. 71, last line but one:—For "charcoal" read "anthrax" (Fr. *charbon*). Under the conditions referred to the virulence of the anthrax bacillus becomes attenuated.

THE WHITEFRIARS PRESS, LTD., LONDON AND TONBRIDGE.

No. 4.

J. & A. CHURCHILL

Selected Books for Students and Practitioners of Medicine.

LONDON

7, Great Marlborough Street.

SEPTEMBER, 1924

INDEX.

PAGE

2 Anatomy. Physiology.

3 Materia Medica. Pharmacy.

4 Hygiene. Bacteriology.

5 Pathology. Psychology. Electro-Therapeutics. Dictionaries.

6 Medicine.

7 Medicine.

8 Surgery.

9 Surgery. Anæsthetics.

10 Neurology. Urinary Disorders. Tropical Diseases.

11 Midwifery. Gynæcology.

12 Medical Jurisprudence. Ophthalmology. Dermatology.

13 Otology. Pædiatrics. Dentistry.

14 Chemistry.

15 Chemistry. Physics.

16 Physics. Microscopy. Miscellaneous.

FREE ON . . . APPLICATION.

1. Complete Illustrated Catalogue.
2. Catalogue of Science Books.

Anatomy & Physiology

The Principles of Anatomy as Seen in the Hand. By F. Wood Jones, D.Sc., M.B., B.S., Professor of Anatomy, University of Adelaide. 125 Illustrations. 15*s.* net.

The Anatomy of the Human Skeleton. By J. Ernest Frazer, F.R.C.S., Professor of Anatomy, University of London. Second Edition. With 219 Illustrations, many in Colours. 28*s.* net.

Synopsis of Anatomy. By T. B. Johnston, M.B., Professor of Anatomy, Univ. of London. 11 Illustrations. 12*s.* 6*d.* net.

Anatomical Terminology, with Special Reference to the B.N.A. By L. F. Barker, M.D. With Illustrations. 5*s.* net.

A Manual of Practical Anatomy. By the late Prof. A. W. Hughes, M.B. Edited by Arthur Keith, M.D. In three parts. Part I, 12*s.* 6*d.* net. Part II, 10*s.* 6*d.* net. Part III, 12*s.* 6*d.* net.

Heath's Practical Anatomy: a Manual of Dissections. Edited by J. E. Lane, F.R.C.S. 9th Edition. 321 Engravings. 15*s.* net.

Clinical Applied Anatomy; or, The Anatomy of Medicine and Surgery. By Charles R. Box, M.D., Physician, St. Thomas's Hospital, and W. McAdam Eccles, M.S.Lond., F.R.C.S.Eng., Surgeon, St. Bartholomew's Hospital. 45 Plates. 12*s.* 6*d.* net.

Essentials of Surface Anatomy. By C. R. Whittaker, F.R.C.S. Third Edition. With 17 Plates. 7*s.* 6*d.* net.

Text-Book of Anatomy and Physiology. By E. R. Bundy, M.D. 5th Edition. With 266 Illustrations. 11*s.* 6*d.* net.

Human Osteology. By Luther Holden. Eighth Edition. Edited by Charles Stewart, F.R.S., and Robert W. Reid, M.D., F.R.C.S. 59 Lithographic Plates and 74 Engravings. 18*s.* net.

Principles of Human Physiology. By Ernest H. Starling, C.M.G., M.D., F.R.C.P., F.R.S., Jodrell Professor of Physiology, University College, London. Third Edition. 579 Illustrations. 25*s.* net.

Practical Physiology. By G. V. Anrep, M.D., D.Sc., and D. T. Harris, M.B., B.S. With Introduction by Prof. E. H. Starling, C.M.G., F.R.S. With 197 Illustrations. 10*s.* 6*d.* net.

Synopsis of Physiology. By Ffrangcon Roberts, M.D., M.R.C.P., Fellow and Lecturer in Physiology, Clare College, Cambridge. With 73 Illustrations. 10*s.* 6*d.* net.

An Introduction to Biophysics. By D. Burns, D.Sc., Professor of Physiology, University of Durham. With 85 Illustrations. 21*s.* net.

The Method of Enzyme Action. By J. Beatty, M.A., M.D., D.P.H. 5*s.* net.

The Cell as the Unit of Life, and other Lectures: An Introduction to Biology. By the late Allan Macfadyen, M.D., B.Sc. Edited by R. Tanner Hewlett, M.D., F.R.C.P., D.P.H. 7*s.* 6*d.* net.

Materia Medica & Pharmacy

Applied Pharmacology. By A. J. Clark, M.C., M.D., F.R.C.P., Professor of Pharmacology, University of London. With 46 Illustrations. 15*s.* net.

A Text-Book of Pharmacology and Therapeutics. By A. R. Cushny, M.A., M.D., F.R.S., Professor of Pharmacology in the University of Edinburgh, etc. Eighth Edition. 73 Illustrations. 24*s.* net.

Materia Medica, Pharmacy, Pharmacology, and Therapeutics. By Sir W. Hale-White, M.D., F.R.C.P., Consulting Physician, Guy's Hospital. Eighteenth Edition. 10*s.* 6*d.* net.

Synopsis of Pharmacology. By D. V. Cow, M.D. With 15 Illustrations. 7*s.* 6*d.* net.

Synopsis of Materia Medica, with Notes on Prescription Writing. By J. Burnet, M.D. 4*s.* 6*d.* net.

A Text-Book of Materia Medica for Students of Medicine. By C. R. Marshall, M.D. 127 Illustrations. 10*s.* 6*d.* net.

Southall's Organic Materia Medica. By John Barclay, B.Sc.Lond. Eighth Edition by E. W. Mann. 12*s.* 6*d.* net.

A Text-Book of Materia Medica. By Henry G. Greenish, F.I.C., F.L.S., Professor of Pharmaceutics, Pharmaceutical Society. Fourth Edition. With 270 Illustrations. 25*s.* net. **The Microscopical Examination of Foods and Drugs.** Third Edition. 209 Illustrations. 18*s.* net. **An Anatomical Atlas of Vegetable Powders.** 138 Illustrations. 12*s.* 6*d.* net.

The Book of Pharmacopœias and Unofficial Formularies. By E. W. Lucas, C.B.E., F.I.C., F.C.S., and H. B. Stevens, O.B.E., F.I.C., F.C.S. 7*s.* 6*d.* net.

ALSO

The Book of Receipts: containing a Veterinary Materia Medica, a Pharmaceutical Formulary, a Photographic Formulary, a Synopsis of Practical Methods employed in the Examination of Urine, Milk, Potable Waters, Sputum, etc. Twelfth Edition. 10*s.* 6*d.* net.

First Lines in Dispensing. 2nd Edition. 97 Illus. 6*s.* net.

Practical Pharmacy. 3rd Edition. 224 Illus. 27*s.* net.

ALSO BY E. W. LUCAS.

The Book of Prescriptions (Beasley) with an Index of Diseases and Remedies. Tenth Edition. 7*s.* 6*d.* net.

Materia Medica, Step by Step. By A. W. Nunn. 3*s.* 6*d.* net.

Medical and Pharmaceutical Latin for Students of Pharmacy and Medicine. By R. R. Bennett. 3rd Edition. 10*s.* 6*d.* net.

A Companion to the British Pharmacopœia. By Peter Wyatt Squire, F.L.S., F.C.S. Nineteenth Edition. 25*s.* net. **Pocket Edition.** Second Edition. 12*s.* 6*d.* net. **The Pharmacopœias of Thirty of the London Hospitals.** Ninth Edition. 12*s.* 6*d.* net.

The Pharmaceutical Formulary. By Henry Beasley. Twelfth Edition by J. Oldham Braithwaite. 6*s.* 6*d.* net.

Hygiene & Bacteriology

The Health of the Industrial Worker. By E. L. Collis, M.D., Professor of Preventive Medicine, Welsh National School of Medicine, and Major Greenwood, M.R.C.P., M.R.C.S., Medical Officer, Ministry of Health. With Introduction by Sir George Newman, K.C.B., M.D. 30*s.* net.

The Principles of Preventive Medicine. By R. Tanner Hewlett, M.D., F.R.C.P., D.P.H., Professor of Bacteriology, Univ. of London, and A. T. Nankivell, M.D., D.P.H., Med. Officer of Health, Hornsey. With 12 Charts and 5 Diagrams. 18*s.* net.

Synopsis of Hygiene. By W. W. Jameson, M.D., D.P.H., Med. Officer of Health, Finchley, and F. T. Marchant, M.R.San.I., Assistant, Dept. of Hygiene, University College. With 18 Illustrations. 18*s.* net.

Sanitation in War. By Lt.-Col. P. S. Lelean, C.B., F.R.C.S., D.P.H., Assistant Professor of Hygiene, Royal Army Medical College. Third Edition. 68 Illustrations. 7*s.* 6*d.* net.

Elementary Hygiene for Nurses. By H. C. R. Darling, M.D., F.R.C.S. Second Edition. 45 Illustrations. 4*s.* 6*d.* net.

A Simple Method of Water Analysis. By John C. Thresh, M.D.Vic., D.Sc.Lond. Ninth Edition. 3*s.* net. **The Examination of Waters and Water Supplies.** Third Edition. 25*s.* net.

ALSO, WITH ARTHUR E. PORTER, M.D., M.A.CANTAB.

Preservatives in Food and Food Examination. 8 Plates. 16*s.* net.

Foods and their Adulteration. By Harvey W. Wiley, M.D., Ph.D. Third Edition. With 11 Coloured Plates and 87 other Illustrations. 27*s.* net. **Beverages and their Adulteration.** With 42 Illustrations. 21*s.* net.

Text-book of Meat Hygiene. By R. Edelmann, Ph.D. Translated by J. R. Mohler, A.M., V.M.D., and A. Eichhorn, D.V.S. Fourth Edition. With 161 Illustrations and 5 Plates. 24*s.* net.

A Manual of Bacteriology, Clinical and Applied. By R. Tanner Hewlett, M.D., Professor of Bacteriology, Univ. of London. Seventh Edition. 31 Plates and 68 Text-figures. 21*s.* net. **Serum and Vaccine Therapy, Bacterial Therapeutics and Prophylaxis, Bacterial Diagnostic Agents.** Second Edition. 32 Figures. 8*s.* 6*d.* net.

Immunity: Methods of Diagnosis and Therapy. By Dr. J. Citron. Second Edition. 40 Illustrations. 14*s.* net.

Clinical Diagnostic Bacteriology, including Serum- and Cyto-diagnosis. By A. C. Coles, M.D., D.Sc. 2 Plates. 8*s.* net.

Lessons in Disinfection and Sterilisation. By F. W. Andrewes, M.D., F.R.C.P. Second Edition. 31 Illustrations. 3*s.* 6*d.* net.

Dairy Bacteriology. By Dr. Orla-Jensen. Translated by P. S. Arup, B.Sc., F.I.C. With 70 Illustrations. 18*s.* net.

Pathology • Psychology Electro-Therapeutics • Dictionaries

Pathology, General and Special, for Students of Medicine. By R. Tanner Hewlett, M.D., F.R.C.P., D.P.H., Professor of Bacteriology, Univ. of London. 48 Plates and 12 Illustrations in Text. Fifth Edition. 18s. net.

Clinical Pathology. By P. N. Panton, M.B., Clinical Pathologist and Director of Hale Clinical Laboratory, London Hospital. With 13 Plates (11 Coloured) and 45 Illustrations in the Text. 14s. net.

A Manual of General or Experimental Pathology. By W. S. Lazarus-Barlow, M.D., F.R.C.P., Director of the Cancer Research Laboratories, Middlesex Hospital. Second Edition. 21s. net. **The Elements of Pathological Anatomy and Histology for Students.** 24s. net.

Surgical Pathology and Morbid Anatomy. See p. 8.

Post-mortem Manual. By C. R. Box, M.D., Lecturer on Applied Anatomy, St. Thomas's Hospital. Second Edition. 22 Illustrations. 10s. 6d. net.

The Pathologist's Handbook: a Manual for the Post-mortem Room. By T. N. Kelynack, M.D. 126 Illustrations. 4s. 6d. net.

Psychological Medicine. By Sir M. Craig, M.D., Physician, Mental Diseases, Guy's Hospital. Fourth Edition. *In the Press.*

ALSO BY SIR M. CRAIG.

Nerve Exhaustion. 6s. net.

Mental Diseases: Clinical Lectures. By Sir T. S. Clouston, M.D., F.R.C.P.Edin. Sixth Edition. 30 Plates. 16s. net.

Unconscious Therapeutics; or, The Personality of the Physician. By Alfred T. Schofield, M.D., Hon. Physician to Friedenheim Hospital. Second Edition. 5s. net. **The Management of a Nerve Patient.** 5s. net.

The Journal of Mental Science. Published Quarterly, by Authority of the Medico-Psychological Association. 7s. 6d. net.

Practical Electro-Therapeutics and Diathermy. By G. B. Massey, M.D., Late President, American Electro-Therapeutic Association. 157 Illustrations. 21s. net.

Lang's German-English Dictionary of Terms used in Medicine and the Allied Sciences. Third Edition. *Reprinting.*

Medicine

The Practice of Medicine. By the late Sir FREDERICK TAYLOR, M.D., F.R.C.P. Twelfth Edition. Revised by Drs. E. P. POULTON, C. P. SYMONDS, and H. W. BARBER, Physicians, Guy's Hospital. 87 Illustrations. 30*s.* net.

Endocrine Diseases: Their Diagnosis and Treatment. By W. FALTA (Vienna). Translated by M. K. MYERS, M.D., with a Foreword by Sir A. E. GARROD, K.C.M.G., M.D. 104 Illustrations. 36*s.* net.

Medical Diagnosis. By ARTHUR LATHAM, M.D., F.R.C.P., Physician and Lecturer on Medicine, St. George's Hospital, and JAMES TORRENS, M.B., M.R.C.P., Assistant Physician, St. George's Hospital. With 74 Illustrations, 19 in colour. 15*s.* net.

Pulmonary Tuberculosis: its Diagnosis, Prevention, and Treatment. By W. M. CROFTON, M.D., Lecturer in Special Pathology, University College, Dublin. With Coloured Frontispiece and 20 Illustrations. 6*s.* net. **Therapeutic Immunisation: Theory and Practice.** 7*s.* 6*d.* net.

A Text-Book of Medicine. Begun by the late C. HILTON FAGGE, M.D.; completed and re-written by P. H. PYE-SMITH, M.D., F.R.S. Fourth Edition. 2 vols. 42*s.* net.

The Primary Lung Focus of Tuberculosis in Children. By Dr. ANTHON GHON. Translated by D. BARTY KING, O.B.E., M.D. 2 Plates and 74 Text-figures. 10*s.* 6*d.* net.

Studies in Influenza and its Pulmonary Complications. By D. BARTY KING, O.B.E., M.D. 7*s.* 6*d.* net.

A Short Practice of Medicine. By R. A. FLEMING, M.D., F.R.C.P.E., F.R.S.E., Lecturer on Medicine, School of Royal Colleges, Edinburgh. Third Edition. With 64 Illustrations. 21*s.* net.

The Practice of Medicine. By M. CHARTERIS, M.D., Professor of Therapeutics, University of Glasgow. Ninth Edition. Edited by F. J. CHARTERIS, M.D., Ch.B. Illustrated. 9*s.* 6*d.* net.

Digestion and Metabolism: The Physiological and Pathological Chemistry of Nutrition. By A. E. TAYLOR, M.D. 18*s.* net.

Text-Book of Medical Treatment. By N. I. C. TIRARD, M.D., F.R.C.P. 15*s.* net.

A Manual of Family Medicine and Hygiene for India. By Sir W. J. MOORE, K.C.I.E., M.D. Ninth Edition, edited by Col. C. A. SPRAWSON, C.I.E., M.D., I.M.S. 69 Engravings. 10*s.* 6*d.* net.

The Blood: how to Examine and Diagnose its Diseases. By ALFRED C. COLES, M.D., D.Sc., F.R.S.Edin. Third Edition. 7 Coloured Plates. 10*s.* 6*d.* net.

Lectures on Medicine to Nurses. By HERBERT E. CUFF, M.D., F.R.C.S. Seventh Edition. 29 Illustrations. 7*s.* 6*d.* net.

On Alcoholism: Its Clinical Aspects and Treatment. By FRANCIS HARE, M.D., Medical Superintendent of the Norwood Sanatorium, Beckenham. 5*s.* net.

Venereal Disease, its Prevention, Symptoms and Treatment. By H. WANSEY BAYLY, M.C. Second Edition. With 55 Illustrations. 7*s.* 6*d.* net.

Medicine

Recent Advances in Medicine. Clinical: Laboratory: Therapeutic. By G. E. BEAUMONT, M.D., Assistant Physician, Middlesex Hospital, and E. C. DODDS, M.R.C.S., L.R.C.P., Chemical Pathologist, Middlesex Hospital. 37 Illustrations. 10s. 6d. net.

Massage: its Principles and Practice. By JAMES B. MENNELL, M.A., M.D., B.C., Medical Officer, Physico-Therapeutic Dept., St. Thomas's Hospital. With 167 Illustrations. 21s. net.

Massage and Medical Gymnastics. By Dr. EMIL KLEEN. Translated by MINA L. DOBBIE, M.B., B.Ch., Med. Officer, Chelsea Physical Training College. 2nd Edition. With 182 Illustrations. 32s. net.

ALSO TRANSLATED AND EDITED BY DR. MINA L. DOBBIE.

Notes on Diseases Treated by Medical Gymnastics and Massage. By Dr. J. ARVEDSON. 8s. 6d. net.

Treatment of Tuberculosis. By ALBERT ROBIN. Translated by Dr. LÉON BLANC and H. de MÉRIC. 21s. net.

Researches on Rheumatism. By F. J. POYNTON, M.D., F.R.C.P.Lond., and ALEXANDER PAINE, M.D., B.S.Lond. With 106 Illustrations and a Coloured Frontispiece. 15s. net.

Vicious Circles in Disease. By J. B. HURRY, M.A., M.D. Third Edition. With Illustrations. 15s. net. **Poverty and its Vicious Circles.** Second Edition. 15s. net. **The Vicious Circles of Neurasthenia and their Treatment.** 3s. 6d. net. **Vicious Circles in Sociology and their Treatment.** 2s. net. **The Ideals and Organisation of a Medical Society.** 2s. net.

Physical Signs in the Chest and Abdomen. By A. J. JEX-BLAKE, M.D., F.R.C.P. 27 Illustrations. 9s. 6d. net.

Ulcer of the Stomach and Duodenum. By SAMUEL FENWICK, M.D., F.R.C.P., and W. SOLTAU FENWICK, M.D., B.S. 55 Illustrations. 10s. 6d. net. **Cancer and other Tumours of the Stomach.** 70 Illustrations. 10s. 6d. net.

The Schott Methods of the Treatment of Chronic Diseases of the Heart. By W. BEZLY THORNE, M.D., M.R.C.P. Fifth Edition. Illustrated. 5s. net.

The Clinical Examination of Urine. By LINDLEY SCOTT, M.A., M.D. 41 original Plates (mostly in colours). 15s. net.

Electro-Therapy: Its Rationale and Indications. By J. CURTIS WEBB, M.B., B.C. With 6 diagrams. 5s. net.

Electricity: Its Medical and Surgical Applications. By C. S. POTTS, M.D. With 356 Illustrations and 6 Plates. 21s. net.

Uric Acid as a Factor in the Causation of Disease. By ALEXANDER HAIG, M.D., F.R.C.P., Physician to the Metropolitan Hospital. Seventh Edition. 75 Illustrations. 14s. net. **Uric Acid in the Clinic.** 5s. net. **Uric Acid, an Epitome of the Subject.** Second Edition. 2s. 6d. net.

Malay Poisons and Charm Cures. By JOHN D. GIMLETTE, M.R.C.S., L.R.C.P. Second Edition. 8s. 6d. net.

Surgery

Surgery. Edited by G. E. GASK, C.M.G., D.S.O., F.R.C.S., and HAROLD W. WILSON, M.S., M.B., F.R.C.S., Surgeons, St. Bartholomew's Hospital. With 39 Plates, 20 in colour, and 467 Text-figures. 30*s.* net.

The After-Treatment of Wounds and Injuries. By R. C. ELMSLIE, M.S., F.R.C.S., Special Mil. Surg. Hosp., Shepherd's Bush; Surgeon, St. Bartholomew's Hospital. 144 Illustrations. 15*s.* net.

A Textbook of Surgery. By R. WARREN, M.D., F.R.C.S., Assistant Surgeon, London Hospital. With 504 Original Illustrations. 2 vols. 27*s.* net.

Surgical Pathology and Morbid Anatomy. By Sir ANTHONY A. BOWLBY, F.R.C.S., Surgeon to St. Bartholomew's Hospital, and Sir F. W. ANDREWES, M.D., F.R.S., Lecturer onPathology, St. Bartholomew's Hospital. Seventh Edition. With 210 Illustrations. 30*s.* net.

A Manual of Surgery. By F. T. STEWART, M.D., Professor of Clinical Surgery, Jefferson Medical College. Third Edition. 571 Illustrations. 16*s.* net.

Ovariotomy and Abdominal Surgery. By HARRISON CRIPPS, F.R.C.S., Surgical Staff, St. Bartholomew's Hospital. 25*s.* net.

BY THE SAME AUTHOR.

On Diseases of the Rectum and Anus, including the Sixth Edition of the Jacksonian Prize Essay on Cancer. Fourth Edition. With 14 Plates and 34 Illustrations. 10*s.* 6*d.* net.

Cancer of the Rectum, especially considered with regard to its Surgical Treatment. Jacksonian Prize Essay. Sixth Edition. With 13 Plates and several Engravings. 5*s.* net.

Diseases of the Rectum, Anus, and Sigmoid Colon. By F. SWINFORD EDWARDS, F.R.C.S., Senior Surgeon to St. Mark's Hospital for Fistula and other Diseases of the Rectum. Third Edition. 102 Illustrations. 10*s.* 6*d.* net.

Minor Surgery and Bandaging. Eighteenth Edition. (Heath, Pollard and Davies.) By GWYNNE WILLIAMS, M.S., F.R.C.S., Surgeon, University College Hospital. 239 Engravings. 10*s.* 6*d.* net.

Injuries and Diseases of the Jaws. By CHRISTOPHER HEATH, F.R.C.S. Fourth Edition. Edited by H. P. DEAN, M.S., F.R.C.S., Assistant Surgeon, London Hospital. 187 Illustrations. 14*s.* net.

BY THE SAME AUTHOR.

Clinical Lectures on Surgical Subjects delivered at University College Hospital. First Series, 6*s.* net; Second Series, 6*s.* net.

Surgical Nursing and After-Treatment. By H. C. RUTHERFORD DARLING, M.D., F.R.C.S., Surgeon, South Coast Hospital, Sydney. Second Edition. With 138 Illustrations. 8*s.* 6*d.* net.

BY THE SAME AUTHOR.

Elementary Hygiene for Nurses. Second Edition. With 45 Illustrations. 4*s.* 6*d.* net.

Surgery & Anæsthetics

The Operations of Surgery. Sixth Edition of JACOBSON. By R. P. ROWLANDS, M.S.Lond., F.R.C.S., Surgeon, Guy's Hospital, and PHILIP TURNER, M.S., F.R.C.S., Surgeon, Guy's Hospital. 2 vols. 797 Illustrations; 40 in Colour. 50*s.* net.

Surgery in War. By A. J. HULL, F.R.C.S., Lieut.-Col., R.A.M.C. With 210 Illustrations. 25*s.* net.

Operative Surgery of the Head, Neck, Thorax and Abdomen. By E. H. TAYLOR, F.R.C.S.I., University Professor of Surgery, Trinity College, Dublin. With 300 Original Illustrations, many in colour. 32*s.* net.

Synopsis of Surgery. By IVOR BACK, F.R.C.S., Surgeon, St. George's Hospital, and A. T. EDWARDS, F.R.C.S., Assistant Surgeon, Westminster Hospital. 12*s.* 6*d.* net.

Synopsis of Surgical Diagnosis. By W. H. C. ROMANIS, M.B., M.C., F.R.C.S. 8*s.* 6*d.* net.

Synopsis of Surgical Pathology. By ERIC PEARCE GOULD, M.D., F.R.C.S. 6*s.* net.

Inguinal Hernia, the Imperfectly Descended Testicle, and Varicocele. By PHILIP TURNER, M.S., F.R.C.S., Assistant Surgeon, Guy's Hospital. With 22 Illustrations. 10*s.* 6*d.* net.

Hare-Lip and Cleft Palate, with Special Reference to the Operative Treatment and its Results. By JAMES BERRY, B.S., F.R.C.S., and T. PERCY LEGG, M.S., F.R.C.S. *Reprinting.*

War Surgery of the Abdomen. By CUTHBERT WALLACE, C.M.G., F.R.C.S. With 26 Illustrations. 10*s.* 6*d.* net.

Practice and Problem in Abdominal Surgery. By ALFRED ERNEST MAYLARD, M.B.Lond. and B.S., Senior Surgeon to the Victoria Infirmary, Glasgow. With 39 Illustrations. 8*s.* 6*d.* net.

BY THE SAME AUTHOR.

Abdominal Tuberculosis. 57 Illustrations. 12*s.* 6*d.* net.

Clinical Essays and Lectures. By HOWARD MARSH, F.R.C.S., Prof. Surgery, Univ. Cambridge. 26 Figures. 7*s.* 6*d.* net.

Hernia, its Cause and Treatment. By R. W. MURRAY, F.R.C.S., Surgeon, David Lewis Northern Hospital. Second Edition. 62 Illustrations. 6*s.* net.

Modern Bullet-Wounds and Modern Treatment. By Major F. SMITH, D.S.O., R.A.M.C. 3*s.* net.

Surgical Emergencies. By PAUL SWAIN, F.R.C.S. Fifth Edition. 149 Engravings. 6*s.* net.

Chloroform: a Manual for Students and Practi- tioners. By EDWARD LAWRIE, M.B.Edin. Illustrated. 5*s.* net.

Urinary Disorders
Neurology & Tropical Diseases

An Epitome of Mental Disorders. A Practical Guide to Ætiology, Diagnosis, and Treatment. By E. Fryer Ballard, M.B., B.S., Capt., R.A.M.C., Medical Officer in Charge of Observation and Mental Block, 2nd Eastern General Hospital. 3 Illustrations. 7*s.* 6*d.* net.

A Text-Book of Nervous Diseases. By W. Aldren Turner, M.D., F.R.C.P., Physician and Lecturer on Neurology, King's College Hospital, and T. Grainger Stewart, M.B., M.R.C.P., Assistant Physician, National Hospital for Paralysed. 188 Illustrations. 18*s.* net.

Paralysis and other Nervous Diseases in Childhood and Early Life. By James Taylor, M.D., F.R.C.P., Physician National Hospital for Paralysed. 74 Illustrations. 12*s.* 6*d.* net.

BY SIR W. R. GOWERS, M.D., F.R.S.

Subjective Sensations of Sight and Sound, Abiotrophy, and other Lectures on Diseases of the Nervous System. 6*s.* net. **Epilepsy and Other Chronic Convulsive Diseases, their Causes, Symptoms and Treatment.** Second Edition. 10*s.* 6*d.* net. **The Borderland of Epilepsy, Faints, Vagal Attacks, Vertigo, Migraine, Sleep Symptoms, and their Treatment.** 4*s.* 6*d.* net.

Selected Papers on Stone, Prostate, and other Urinary Disorders. By R. Harrison, F.R.C.S. 15 Illustrations. 5*s.* net.

BY E. HURRY FENWICK, F.R.C.S., SURGEON TO THE LONDON HOSPITAL.

Atlas of Electric Cystoscopy. 34 Coloured Pls. 21*s.* net.

Obscure Diseases of the Urethra. 63 Illus. 6*s.* 6*d.* net.

Operative and Inoperative Tumours of the Urinary Bladder. 39 Illustrations. 5*s.* net.

Tumours of the Urinary Bladder. Fasc. I. 5*s.* net.

Ulceration of the Bladder, Simple, Tuberculous, and Malignant: a Clinical Study. Illustrated. 5*s.* net.

The Malarial Fevers of British Malaya. By Hamilton Wright, M.D. Map and Charts. 3*s.* net. **The Etiology and Pathology of Beri-Beri.** With Map and Charts. 3*s.* net.

Midwifery & Gynæcology

The Difficulties and Emergencies of Obstetric Practice. By COMYNS BERKELEY, M.D., F.R.C.P., and VICTOR BONNEY, M.D., F.R.C.S., Obstetric and Gynæcological Surgeons, Middlesex Hospital. Third Edition. With 309 Original Illustrations. 36s. net.

Manual of Midwifery. By T. W. EDEN, M.D., C.M.Edin., F.R.C.P.Lond., Obstetric Physician and Lecturer on Practical Midwifery, Charing Cross Hospital. 5th Ed. 5 Plates and 369 Illustrations. 24s. net.

BY THE SAME AUTHOR AND C. LOCKYER, M.D, F.R.C.P., F.R.C.S.

Gynæcology. Second Edition. 513 Illustrations and 24 Coloured Plates. 36s. net.

Practical Midwifery. By GIBBON FITZGIBBON, M.D., F.R.C.P.I. With 175 Illustrations. 16s. net.

A Short Practice of Midwifery, embodying the Treatment adopted in the Rotunda Hospital, Dublin. By HENRY JELLETT, M.D., B.A.O.Dub., Ex-Master, Rotunda Hospital, Dublin. Ninth Edition. 4 Coloured Plates and 263 Illustrations. 18s. net.

BY THE SAME AUTHOR.

A Short Practice of Midwifery for Nurses, with a Glossary of Medical Terms, and the Regulations of the C.M.B. Sixth Edition. 4 Coloured Plates and 172 Illustrations. 9s. net.

ALSO

A Practice of Gynæcology. With 374 Illustrations (some coloured). 21s. net. **A Short Practice of Gynæcology.** With 318 Illustrations (many in colour) and 10 Plates. Fifth Edition. 18s. net.

Manual of Obstetrics. By O. ST.JOHN MOSES, M.D., C.M., D.Sc., F.R.C.S. With 136 Illustrations. 21s. net.

Obstetric Aphorisms. By the late J. G. SWAYNE, M.D. Revised by W. C. SWAYNE, M.D., B.S.Lond., Professor of Obstetrics, University of Bristol. Eleventh Edition. With 29 Illustrations. 3s. 6d. net.

A Manual for Midwives. By C. J. N. LONGRIDGE, M.D., and J. B. BANISTER, M.D., F.R.C.S., Physician to Out-patients, Queen Charlotte's Hospital. Third Edition. With 51 Illustrations. 7s. 6d. net.

A Short Manual for Monthly Nurses. By CHARLES J. CULLINGWORTH, M.D., F.R.C.P. Sixth Edition. 1s. 6d. net.

A Clinical Manual of the Malformations and Congenital Diseases of the Fœtus. By Prof. Dr. R. BIRNBAUM. Translated and Annotated by G. BLACKER, M.D., F.R.C.P., F.R.C.S., Obstetric Physician to University College Hospital. With 66 Illustrations. 15s. net.

Outlines of Gynæcological Pathology and Morbid Anatomy. By C. HUBERT ROBERTS, M.D.Lond., Physician to the Samaritan Free Hospital for Women. 151 Illustrations. 21s. net.

Sterility in Woman: Its Causes and Treatment. By R. A. GIBBONS, M.D., F.R.C.S.Ed. With 44 Illustrations. 12s. 6d. net.

ALSO BY DR. R. A. GIBBONS.

A Lecture on Dysmenorrhœa. 2s. 6d. net. **A Lecture on Sterility: its Ætiology and Treatment.** 2s. 6d. net. **A Lecture on Pruritus Vulvæ: its Ætiology and Treatment.** 2s. 6d. net.

Medical Jurisprudence Ophthalmology & Dermatology

Medical Jurisprudence: its Principles and Practice. By Alfred S. Taylor, M.D., F.R.C.P., F.R.S. Seventh Edition, by Fred. J. Smith, M.D., F.R.C.P., Physician to, and Lecturer on Forensic Medicine at, the London Hospital. 2 vols. 20 Engravings. £3 3s. net. Also by Dr. F. J. Smith. **Law for Medical Men, containing Extracts** from Acts of Parliament interesting to Medical Men. 10s. 6d. net.

Medical Ophthalmology. By R. Foster Moore, O.B.E., F.R.C.S., Assistant Ophthalmic Surgeon, St. Bartholomew's Hospital. 80 Illustrations. 15s. net.

Medical Ophthalmoscopy: A Manual and Atlas. Fourth Edition. By Sir W. R. Gowers, M.D., F.R.S., and Marcus Gunn, M.B., F.R.C.S. Autotype Plates and Woodcuts. 14s. net.

Refraction of the Eye: a Manual for Students. By Gustavus Hartridge, F.R.C.S., Consulting Surgeon to the Royal Westminster Ophthalmic Hospital. Sixteenth Edition. 110 Illustrations, also Test-types, etc. 7s. 6d. net. **The Ophthalmoscope: a Manual for Students.** Sixth Edition. 65 Illustrations and 4 Plates. 6s. 6d. net.

Diseases of the Eye: a Manual for Students and Practitioners. By Sir J. H. Parsons, D.Sc., M.B., B.S., F.R.C.S., F.R.S., Ophthalmic Surgeon, University College Hospital; Surgeon, Royal London (Moorfields) Ophthalmic Hospital. Fourth Edition. 326 Illustrations and 19 Coloured Plates. 19s. net. **Elementary Ophthalmic Optics, including Ophthalmoscopy and Retinoscopy.** 66 Illustrations. 6s. 6d. net.

The Principles of Ophthalmoscopy and Skiascopy. By G. F. Alexander, M.B., C.M. 31 Illustrations. 5s. net.

The Ophthalmoscope and How to Use It, with a Chapter on Diplopia. By A. Freeland Fergus, LL.D, M.D. Second Edition. 17 Illustrations. 3s. 6d net.

Refraction of the Eye, including Elementary Ophthalmic Optics. By C. B. Goulden, O.B.E., M.D., F.R.C.S., Ophthalmic Surgeon, London Hospital and Royal London Ophthalmic Hospital. 180 Illustrations. 10s. 6d. net.

Sight Testing made Easy, including Chapter on Retinoscopy. By W. W. Hardwicke, M.D., M.R.C.P. Fourth Edition. 12 Engravings. 5s. net.

Ophthalmological Society of the United Kingdom. Transactions. Vol. XLIII. 30s. net.

A Text-Book of Diseases of the Skin. By J. H. Sequeira, M.D., F.R.C.P., F.R.C.S., Physician to the Skin Department, London Hospital. Third Edition. With 52 Plates in Colours and 257 Text-figures. 38s. net.

The Diagnosis and Treatment of Syphilis. By Tom Robinson, M.D.St.And. Second Edition. 3s. 6d. net. **The Diagnosis and Treatment of Eczema.** Second Edition. 3s. 6d. net.

Otology • Pædiatrics • Dentistry

The Labyrinth of Animals, including Mammals, Birds, Reptiles, and Amphibians. By ALBERT A. GRAY, M.D.(Glas.), F.R.S.E., Surgeon, Victoria Infirmary, Glasgow. Vol. I, with 31 Stereoscopic Plates. 21*s.* net (including Stereoscope). Vol. II, with 45 Stereoscopic Plates, 25*s.* net.

Manual of Diseases of Nose and Throat. By C. G. COAKLEY, M.D. Sixth Edition. With 145 Illustrations and 7 Coloured Plates. 18*s.* net.

The Pharmacopœia of the Hospital for Diseases of the Throat, Nose, and Ear. Edited by C. A. PARKER, F.R.C.S., and T. JEFFERSON FAULDER, F.R.C.S. Seventh Edition. 2*s.* 6*d.* net.

Diseases of the Ear. By T. MARK HOVELL, Senior Aural Surgeon to the London Hospital. Second Edition. 128 Engravings. 21*s.* net.

Premature and Congenitally Diseased Infants. By JULIUS H. HESS, M.D. With 189 Illustrations. 18*s.* net.

The Diseases of Children. By the late SIR JAMES F. GOODHART, Bart., M.D., F.R.C.P. Edited by G. F. STILL, M.D., F.R.C.P., Professor of the Diseases of Children, King's College. Eleventh Edition. 60 Illustrations. 32*s.* net.

The Wasting Diseases of Infants and Children. By EUSTACE SMITH, M.D., F.R.C.P. Sixth Edition. 6*s.* net.

An Introduction to Dental Anatomy and Physiology, Descriptive and Applied. By A. HOPEWELL-SMITH, L.D.S.Eng., Professor of Dental Histology, University of Pennsylvania, Philadelphia. With 6 Plates and 340 Illustrations. 21*s.* net. **The Normal and Pathological Histology of the Mouth.** Vol. I, Normal Histology. Vol. II, Pathological Histology. With 658 Illustrations. 2 vols. £2 2*s.* 0*d.* per set.

Dental Anatomy, Human and Comparative. By CHARLES S. TOMES, M.A., F.R.S. Eighth Edition. Edited by H. W. MARETT TIMS, O.B.E., M.D., F.L.S., with assistance of C. BOWDLER HENRY, L.R.C.P., M.R.C.S., L.D.S.(Eng.). 325 Illustrations. 18*s.* net.

A System of Dental Surgery. By Sir JOHN TOMES, F.R.S. Revised by C. S. TOMES, M.A., F.R.S., and WALTER S. NOWELL, M.A.Oxon. Fifth Edition. 318 Engravings. 15*s.* net.

An Atlas of Dental Extractions, with Notes on the Causes and Relief of Dental Pain. By C. EDWARD WALLIS, M.R.C.S., L.R.C.P., L.D.S., Assistant Dental Surgeon, King's College Hospital. Second Edition. With 11 Plates. 6*s.* net.

A Manual of Dental Metallurgy. By ERNEST A. SMITH, Assay Office, Sheffield. Fourth Edition. 37 Illustrations. 12*s.* 6*d.* net.

Synopsis of Dentistry. By A. B. G. UNDERWOOD, M.B., B.S., L.D.S.Eng. With 10 Illustrations. 9*s.* 6*d.* net.

Handbook of Mechanical Dentistry. By J. L. DUDLEY BUXTON, L.D.S., Dental Surgeon, University College Hospital. With 168 Illustrations. 12*s.* 6*d.* net.

Chemistry

Inorganic and Organic Chemistry. By C. L. BLOXAM. Eleventh Edition. By A. G. BLOXAM, F.I.C., and S. JUDD LEWIS, D.Sc., F.I.C. 310 Illustrations. 36*s.* net.

A Text-Book of Practical Chemistry. By G. F. HOOD, M.A., B.Sc., Chemistry Master, Nottingham High School, and J. A. CARPENTER, M.A., late Science Master, St. Paul's School. With 162 Illustrations. Price 21*s.* net.

Explosives. Their Manufacture, Properties, Tests, and History. By ARTHUR MARSHALL, A.C.G.I., F.I.C., F.C.S. Second Edition. 2 vols. With 158 Illustrations. £3 3*s.* net. **A Short Account of Explosives.** 7*s.* 6*d.* net. **A Dictionary of Explosives.** 15*s.* net.

Introduction to Qualitative Chemical Analysis. By C. R. FRESENIUS. Seventh Edition, translated by C. A. MITCHELL, M.A., F.I.C., Editor of "The Analyst." With 57 Illustrations. 36*s.* net.

Treatise on Applied Analytical Chemistry. Edited by Prof. V. VILLAVECCHIA. Translated by T. H. POPE, B.Sc. Vol. I. With 58 Illustrations. 21*s.* net. Vol. II. With 105 Illustrations. 25*s.* net.

Treatise on General and Industrial Chemistry. By Dr. ETTORE MOLINARI. Second English Edition. Translated by T. H. POPE, B.Sc., F.I.C. Vol. I.—**Inorganic.** With 328 Illustrations and 2 Plates. 42*s.* net. Vol. II.—**Organic.** Pt. I. 254 Illustrations. 30*s.* net. Pt. II. 303 Illustrations. 30*s.* net.

Bricks and Artificial Stones of Non-Plastic Materials. By ALFRED B. SEARLE. 65 Illustrations. 10*s.* 6*d.* net.

The Chemistry of Cyanogen Compounds, and their Manufacture and Estimation. By HERBERT E. WILLIAMS. 12*s.* 6*d.* net.

The Preparation of Organic Compounds. By E. DE BARRY BARNETT, B.Sc. Second Edition. With 54 Illustrations. 10*s.* 6*d.* net. **A Text-Book of Organic Chemistry.** With 15 Illustrations. 10*s.* 6*d.* net.

Organic Medicaments and their Preparation. By M. FOURNEAU. Translated by W. A. SILVESTER. Illustrated. net.

The Plant Alkaloids. By T. A. HENRY, D.Sc. Second Edition. 8 Plates. 28*s.* net.

Quantitative Analysis in Practice. By J. WADDELL, D.Sc., Ph.D. 4*s.* 6*d.* net.

Industrial Organic Analysis. By PAUL S. ARUP, B.Sc., A.C.G.I. Second Edition. 25 Illustrations. 12*s.* 6*d.* net.

A History of Chemistry. By the late J. CAMPBELL BROWN. Edited by H. H. BROWN. Second Edition. With 120 Illustrations. 21*s.* net. **Practical Chemistry.** Sixth Edition. Edited by G. D. BENGOUGH, D.Sc. 2*s.* 6*d.* net. **Essays and Addresses.** With 23 Illustrations. 5*s.* net.

Microbiology for Agricultural and Domestic Science Students. Edited by C. E. MARSHALL. Third Edition. With 186 Illustrations. 21*s.* net.

Cocoa and Chocolate: their Chemistry and Manufacture. By R. WEYMPER. Second Edition. With 16 Plates and 10 Figures. 42*s.* net.

Laboratory Text-Book of Chemistry. Part I. By V. S. BRYANT, M.A., Assistant Master at Wellington College. 4*s.* net.

Modern Methods of Steel Analysis. By J. A. PICKARD, A.R.C.Sc. 5*s.* net.

The Synthetic Use of Metals in Organic Chemistry. By A. J. HALE. 6*s.* net.

Reagents and Reactions. By E. TOGNOLI. Trans. by C. A. MITCHELL. 7*s.* 6*d.* net.

Colloid Chemistry of the Proteins. By Prof. Dr. W. PAULI. Translated by P. C. L. THORNE, M.A. With 27 Diagrams. 8*s.* 6*d.* net.

The Formation of Colloids. By THE SVEDBERG. 22 Illustrations. 7*s.* 6*d.* net.

Laboratory Manual of Elementary Colloid Chemistry. By E. HATSCHEK. With 20 Illustrations. 7*s.* 6*d.* net.

Practical Physiological Chemistry. By P. B. HAWK, M.S., Ph.D. Eighth Edition. With 6 Coloured Plates and 197 Text-figures. 24*s.* net.

Chemistry & Physics

Elementary Qualitative and Volumetric Chemical Analysis. By W. Caldwell, Sc.D., Professor of Chemistry, Royal College of Surgeons in Ireland. Introduction by Prof. C. S. Gibson, O.B.E. 10*s.* 6*d.* net.

A Systematic Handbook of Volumetric Analysis. By Francis Sutton. Eleventh Edition, revised by W. Lincolne Sutton, F.I.C., and A. E. Johnson, F.I.C. 35*s.* net.

Quantitative Organic Microanalysis. By F. Pregl. Translated by E. Fyleman. 42 Illustrations. 12*s.* 6*d.* net.

The Fundamental Processes of Dye Chemistry. By H. E. Fierz-David. Translated by F. A. Mason, Ph.D. With 45 Illustrations, including 19 Plates. Price 21*s.* net.

A Junior Inorganic Chemistry. By R. H. Spear, M.A. With 93 Illustrations. 6*s.* 6*d.* net. Also **Part I** (up to Atomic Theory). 3*s.* 6*d.* net.

Chemical Combinations among Metals. By Dr. M. Guia. Translated by G. W. Robinson. With 207 Illustrations. 21*s.* net.

Ammonia and the Nitrides. With Special Reference to their Synthesis. By E. B. Maxted, Ph.D., B.Sc. 7*s.* 6*d.* net.

The Analyst's Laboratory Companion. By A. E. Johnson, B.Sc., F.I.C., Fifth Edition. 10*s.* 6*d.* net.

Commercial Organic Analysis: a Treatise on the Properties, Modes of Assaying, Proximate Analytical Examination, etc., of Organic Chemicals and Products. By A. H. Allen, F.I.C. Fifth Edition, in 9 vols. Vols. 1 and 2, 30*s.* net; Vols. 3-9 In Preparation.

Volumetric Analysis for Students of Pharmaceutical and General Chemistry. By C. H. Hampshire, B.Sc., F.I.C. Third Edition. 7*s.* 6*d.* net.

Valentin's Practical Chemistry. By Dr. W. R. Hodgkinson, F.R.S.E. Tenth Edition. 97 Illustrations. 12*s.* 6*d.* net.

Quantitative Analysis. By Frank Clowes, D.Sc.Lond., and J. B. Coleman, A.R.C.Sci.Dub. Twelfth Edition. 136 Engravings. 18*s.* net. **Qualitative Analysis.** Ninth Edition. 84 Engravings. 12*s.* 6*d.* net. **Elementary Practical Chemistry.** Part I. Seventh Edition. General Chemistry. 76 Engravings. 6*s.* net. Part II. Ninth Edition. Analytical Chemistry. 20 Engravings. 6*s.* net.

Researches on the Affinities of the Elements. By Geoffrey Martin, B.Sc.Lond. Illustrated. 16*s.* net.

Elementary Practical Metallurgy. By J. H. Stansbie, B.Sc., F.I.C. 25 Illustrations. 5*s.* net.

TEXT-BOOKS OF CHEMICAL RESEARCH AND ENGINEERING. Edited by W. P. Dreaper, O.B.E., F.I.C.

Clouds and Smokes. The Properties of Disperse Systems in Gases. By W. E. Gibbs, D.Sc. 30 Illustrations. 10*s.* 6*d.* net.

The Theory of Emulsions and Emulsification. By W. Clayton, D.Sc. Foreword by Prof. F. G. Donnan, F.R.S. With 22 Illustrations 9*s.* 6*d.* net.

Catalytic Hydrogenation and Reduction. By E. B. Maxted, Ph.D. B.Sc., F.C.S. With 12 Illustrations. 5*s.* net.

Surface Tension and Surface Energy and their Influence on Chemical Phenomena. By R. S. Willows, M.A., D.Sc., and E. Hatschek. Third Edition. With 25 Illustrations. 6*s.* 6*d.* net.

Molecular Physics. By J. A. Crowther, M.A., Sc.D., F.Inst.P. Third Edition. With 32 Illustrations. 7*s.* 6*d.* net.

Notes on Chemical Research. By W. P. Dreaper, O.B.E., F.I.C. Second Edition. 7*s.* 6*d.* net.

An Introduction to the Physics and Chemistry of Colloids. By Emil Hatschek. Fourth Edition. With 17 Illustrations. 7*s.* 6*d.* net.

Catalysis and its Industrial Applications. By E. Jobling, A.R.C.Sc., B.Sc., F.C.S. Second Edition. With 12 Illustrations. 7*s.* 6*d.* net.

Physics
Microscopy Miscellaneous

Elementary Physics. By G. STEAD, Reader in Physics, Guy's Hospital Medical School. 288 Illustrations. 10s. 6d. net.

A Handbook of Physics and Chemistry for the Conjoint Board. By H. E. CORBIN, B.Sc.Lond., and A. M. STEWART, B.Sc.Lond. Fifth Edition. 200 Illustrations. 12s. 6d. net.

A Treatise on Physics. By ANDREW GRAY, LL.D., F.R.S., Vol. I. Dynamics and Properties of Matter. 350 Illustrations. 18s. net.

A Text-book of Physics. Edited by A. W. DUFF, D.Sc., Fifth Edition. 609 Illustrations. 16s. net.

The Conduction of Electricity through Gases and Radio-activity. By R. K. MCCLUNG, D.Sc. 8s. 6d. net.

Physical Measurements. By A. WILMER DUFF, D.Sc., and A. W. EWELL, Ph.D. Second Edition. 78 Illustrations. 7s. 6d. net.

The Principles of Radiography. By J. A. CROWTHER, Sc.D., F.Inst.P. With 55 Illustrations. 7s. 6d. net.

Critical Microscopy. How to get the best out of the Microscope. By A. C. COLES, M.D., D.Sc. 8 Illustrations. 7s. 6d net.

The Microscope and its Revelations. By the late WILLIAM B. CARPENTER, C.B., M.D., F.R.S. Eighth Edition, by the Rev. W. H. DALLINGER, F.R.S. 23 Plates and 800 Wood Engravings. 36s. net; or, in two vols., sold separately, cloth, 20s. net each. Vol. I. The Microscope and its Accessories. Vol. II. The Microscope, its Revelations.

The Microtomist's Vade-Mecum. By ARTHUR BOLLES LEE. Eighth Edition. Edited by J. BRONTË GATENBY, D.Sc. 28s. net.

A Text-book of Botany, for Medical and Phar- maceutical Students. By J. SMALL, D.Sc, F.L.S., Professor of Botany, Queen's University, Belfast. 1350 Illustrations. 21s. net.

The Horticultural Record. By REGINALD CORY. With 117 Plates in Colour and 71 Black and White Illustrations. 42s. net.

The Story of Plant Life in the British Isles. By A. R. HORWOOD. In 3 vols. With Illus. 6s. 6d. net per volume.

Plant Anatomy. By W. C. STEVENS, Professor of Botany in the University of Kansas. Third Edition. 152 Illustrations. 15s. net.

A Text-book of Mycology and Plant Pathology. By J. W. HARSHBERGER. With 271 Illustrations. 21s. net.

The Mechanical Principles of the Aeroplane. By S. BRODETSKY, M.A., Reader in Applied Mathematics, University of Leeds. 119 Diagrams. 21s. net.

The Principles of Aeroplane Construction. By RANKIN KENNEDY, C.E. 51 Illustrations. 6s. 6d. net.

Therapeutic Electricity and Practical Muscle Testing. By W. S. HEDLEY, M.D. 110 Illustrations. 8s. 6d. net.

A Manual for Hospital Nurses. By E. J. DOMVILLE, Surgeon, Devon and Exeter Hospital. Ninth Edition. 1s. net.

The Mothercraft Manual. By M. LIDDIARD, S.R.N., Matron, Mothercraft Training Society. Second Edition. 34 Illustrations. 3s. 6d. net.

J. & A. CHURCHILL
LONDON: 7 GREAT MARLBOROUGH STREET