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Proceedings of the

WOOD CHEMISTRY SYMPOSIUM

Comptes rendus du

COLLOQUE SUR LA CHIMIE DU BOIS

UNION INTERNATIONALE DE
CHIMIE PURE ET APPLIQUÉE

SECTION DE CHIMIE APPLIQUÉE
DIVISION DE LA PÂTE, DU PAPIER ET DU CARTON

LA CHIMIE DU BOIS

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COLLOQUE SUR LA CHIMIE DU BOIS

à Montréal, Canada

Du 9–11 août, 1961

LONDRES

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1962

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WOOD CHEMISTRY

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SCIENTIFIC PROGRAMME

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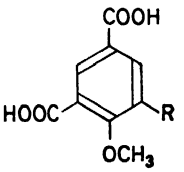
BIOGENESIS AND CONSTITUTION OF LIGNIN

K. FREUDENBERG

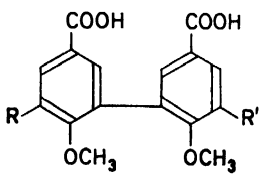
Universität Heidelberg, Deutsche Bundesrepublik

In recent years, the investigation of lignin has begun to take on a more definite form[1–7]; it is of value to present reviews, from time to time, expounding the latest position of research in this field. I take the liberty of placing the work done in our Heidelberg laboratory in the forefront of my discussion.

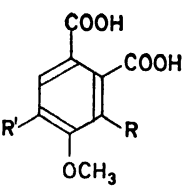
After achieving a biosynthesis of a lignin-like material by dehydrogenation of coniferyl alcohol, extensive efforts were devoted to the comparison of this synthetic product with milled wood lignin, isolated from conifers according to the Björkman procedure. The identity is as good as can be reasonably expected between high molecular weight substances, in which the sequence of their monomeric constituent units is not subject to a stringent pattern. One new proof of their identity is provided by the



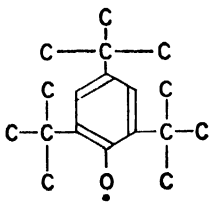
(I) R=OCH_3· Isohemipinic acid
(VII) R=H· 4-Methoxyisophthalic acid
(VIII) R=COOH: Methoxytrimesic acid



(II) R,R'=OCH_3: Dehydrodiveratric acid
(IX) R,R'=H: Dehydrodianisic acid
(X) R=H, R'=OCH_3: 2,3,2'-Trimethoxydiphenyl-5,5'-dicarboxylic acid



(III) R=H, R'=OCH_3· Metahemipinic acid
(IV) R=OCH_3, R'=H: Hemipinic acid
(V) R,R'—OCH_3· Trimethoxyphthalic acid
(VI) R,R'=H: 4-Methoxyphthalic acid



(XI) Tri-t-butylphenoxyl radical

numerous methylated phenolic acids formed from both natural and synthetic lignins. The lignin is methylated, treated with hot alkali to hydrolyse ether linkages, re-methylated and oxidized. Apart from the veratric acid, isohemipinic acid (I), dehydrodiveratric acid (II), and metahemipinic acid (III) previously detected, degradation of natural lignin also yields hemipinic acid (IV), trimethylgallic acid, 3,4,5-trimethoxyphthalic acid (V), benzenepentacarboxylic acid, succinic acid and tricarballylic acid (propane-1,2,3-tricarboxylic acid). In addition, anisic acid, the related 4-methoxyphthalic acid (VI), 4-methoxyisophthalic acid (VII), methoxytrimesic acid (VIII),

dehydrodianisic acid (IX), and 2,3,2′-trimethoxydiphenyl-5,5′-dicarboxylic acid (X) are also found (24 in all).

If a dehydrogenation polymer formed from coniferyl alcohol is treated similarly, the acids cited above, from veratric to tricarballylic, are obtained in approximately the same relative proportions as from wood, with the exception of trimethoxyphthalic acid (V), the presence of which is dubious. If the synthetic lignin has been produced from 80 per cent coniferyl alcohol, 14 per cent *p*-coumaric and 6 per cent sinapic alcohol, the same acids are obtained together with anisic acid and its congeners (VI–X). The number and type of acids obtained from the natural and synthetic products are the same and, in our estimation, even the relative quantitative proportions are much the same.

Although these results primarily provide further evidence for the identity of the natural and biosynthetic preparations, the acids also allow further deductions regarding the constitution of lignin.

The occurence of trimethylgallic acid as a degradation product of a dehydrogenation polymer made from coniferyl alcohol alone is remarkable. Under equivalent degradation conditions, vanillic acid does not yield any gallic acid derivative. The trimethylgallic acid is formed from the dehydrogenation products of coniferyl alcohol. This conversion and the formation of benzene pentacarboxylic, tricarballylic and the phthalic acids will be discussed later. The same degradation products are obtained from both artificial and natural lignin—albeit in lower yield—when acetic anhydride plus a trace of perchloric acid is used to open the ether bonds instead of hot alkali.

Dehydrogenation of coniferyl alcohol occurs at the phenol group. This reaction is completely unspecific. Plants utilize laccase and peroxidase, and the process can be duplicated *in vitro* using these enzymes. However, dehydrogenation with cupric salts or manganese or lead dioxides produces the same result. Even Eugen Müller's blue 2,4,6-tri-t-butylphenoxyl radical (XI) effects the same operation. If the dehydrogenation is conducted so that approximately one hydrogen atom is abstracted per molecule of coniferyl alcohol, many new substances—at least forty in number—are formed; these are all phenols and can be distinguished by their coupling reaction using diazobenzene sulphonic acid. More than 20 of these substances have been isolated[7] and their constitutions elucidated, among them those which are formed in largest quantities.

From a knowledge of the oligomers of dehydrogenated coniferyl alcohol, it is not difficult to deduce that a radical R_α (XII) is first formed which is a mesomer of both R_β (XIII) and R_γ (XIV). These various radical forms saturate themselves mutually forming dimeric intermediates, such as dehydrodiconiferyl alcohol (XV), DL-pinoresinol and DL-epipinoresinol (XVI*a* and *b*), and *bis*-dehydroconiferyl alcohol (XVII). However, the most important dimeric product of coniferyl alcohol is the quinone methide (XIX) formed by combination of R_β with R_α. All the above dimeric transformation products of coniferyl alcohol are formed *via* quinone methides which can be stabilized by intramolecular prototropy. This cannot occur with (XIX). This substance is formed just like all the others under the dehydrogenation conditions (0·2 per cent aqueous coniferyl

alcohol solution at 20° and pH 5·5–6·0, mushroom laccase and oxygen (air), or peroxidase and hydrogen peroxide). It can be identified in 90 per cent dioxan by its characteristic quinone methide absorption using a Cary apparatus; here its half-life is about 1 hour. By addition of water, it is converted into guaiacylglycerol-β-coniferyl ether (XX), the most abundant intermediate of lignin formation. A monolactone corresponding pinoresinol (XVI) has also been isolated.

Aldehydes are also found among the intermediates mentioned above. Those isolated are coniferyl aldehyde itself and the aldehydes of substances (XV) and (XX). These aldehydes are also formed in the absence of oxygen using the blue aryloxy radical (XI); hence they are formed by hydrogen transfer to the aryloxy radicals, which in this case attack the carbinol hydroxyl of the cinnamyl alcohols.

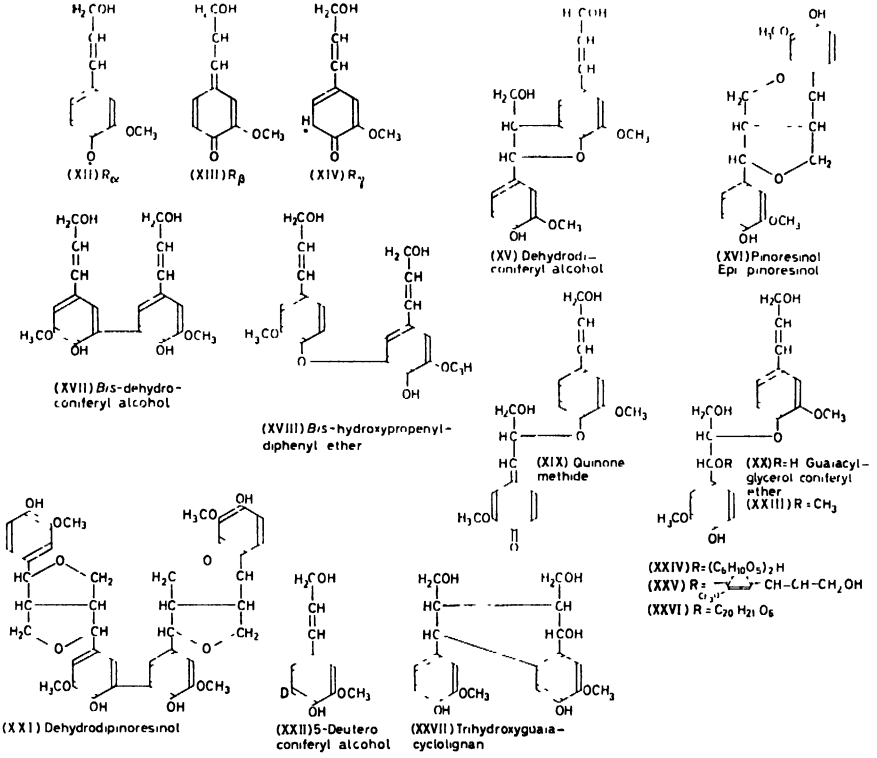
Furthermore, probably two other dimers are formed: the aryl ether (XVIII) and a cyclolignan for which we propose the preliminary formula (XXVII). These hypothetical dimers will be discussed below.

All the dimeric products of coniferyl alcohol cited above are phenols, which themselves form radicals, either by repeated dehydrogenation or radical transfer, and which can combine either with monomeric radicals to give trimers or with each other giving tetramers. All these substances and their derivatives are true lignification intermediates and give valid information concerning the constitution of the natural material, since the latter is identical with synthetic lignin. The starting materials—*p*-hydroxycinnamyl alcohols—are the same for both the synthetic and natural products. Hence the mode of formation—which can be investigated using the synthetic material—is the same for the natural product. This is confirmed by the following experiment. Trunks of spruce felled at the end of May during the growth season were immediately stripped of their bark, and the cambial layers exposed on both trunk and bark were washed down with formalin solution to inactivate the enzymes. The sap contained coniferin and very small quantities of coniferyl alcohol, *p*-gluco-coumar alcohol and the main intermediates also found in the *in vitro* lignification process.

Growth beyond the dimers is confirmed by substance (XXI), dehydrodipinoresinol, also one of the intermediates. Undoubtedly, substances (XV) and (XX) can also " dimerize " similarly through a diphenyl linkage, and (XV), (XVI*a*), and (XX) can form mixed diphenyl derivatives. Recently, 5 new trimeric dehydrogenation products were isolated. The substances isolated so far represented more than 80 per cent by weight of the sum total of the intermediates.

From the discussion so far, formation of larger entities by progressive dehydrogenation can be envisaged. This is the first lignin growth principle.

It soon becomes apparent to anyone attempting to construct a scheme for the massive, branched lignin molecule with the aid of progressive dehydrogenation alone that this will give unsatisfactory results. On the average, each coniferyl alcohol unit would have to lose two atoms of hydrogen to form a bifunctional intermediate. A glance at the units etherified at the phenol group in the dimeric building stones (XV) and (XX) reveals that many side arms incapable of further dehydrogenation are formed. The elementary composition of lignin suggests that, on conversion into lignin,



the coniferyl alcohol loses approximately two hydrogen atoms, simultaneously incorporating 0·3–0·5 molecules of water. However, not every carbon or oxygen atom deprived of its hydrogen takes part in a linking process. For example, if a primary alcohol group becomes aldehydic, two hydrogen atoms are removed without effecting a condensation. The number of hydrogen atoms dislodged without producing a condensation can be estimated at 0·2–0·3 hydrogen atom per unit. Accordingly, the number of hydrogen atoms whose displacement leads to new bonding is reduced to 1·7–1·8 atoms per unit. This implies that, apart from progressive dehydrogenation, some other growth principle associated with no loss of hydrogen, and hence based on a simple addition, must occur to some extent in lignification.

We have found examples of such a second growth principle centred upon the unstable quinone methide (XIX). It has already been stated that this is capable of adding on water. If the enzymatic or chemical dehydrogenation of coniferyl alcohol is carried out in a solution containing 30 per cent methanol, a new substance occurs in quantities which can be isolated, alongside the numerous known dehydrogenation products. This was separated and identified as the methyl ether (XXIII) of guaiacylglycerol-β-coniferyl ether. Hence, despite the excessive amount of water present, here the methanol adds onto the quinone methide. The ether can be split by very dilute acids into methanol and substance (XX). If the dehydrogenation of

coniferyl alcohol is carried out in a concentrated solution of sucrose, the latter behaves just like the methanol. Two combinations of cane sugar with lignin units occur, the more abundant being an adduct (XXIV) of cane sugar onto the quinone methide (XIX). This is not a glycoside but an ether of the sucrose with the carbinol group of compound (XX). This product can also be hydrolysed easily, giving (XX), glucose and fructose. It is a phenol, and just like any other, can take part in further lignification processes. We believe that this reveals the principle of the bonding of lignin onto the polysaccharides of wood and thus provides the mechanism whereby lignin is grafted onto polysaccharides. Alcohols which, unlike cinnamic alcohols, are not unsaturated (*e.g.* substances (XV) and (XX)) occur in sufficient amounts among the dimeric lignin building stones. It can be assumed that in all probability, such primary alcohols add onto the quinone methide (XIX) in the same way as methanol does in (XXIII). We have not yet succeeded in adding the primary carbinol of cinnamyl alcohols onto the quinone methide (XIX).

If an acid such as very dilute acetic acid is present during the dehydrogenation of coniferyl alcohol, a labile acetate can be identified in the mixture of primary products. If phenols are present, *e.g.* phenol, guaiacol or symmetrical pyrogallol dimethyl ether, adducts of these phenols onto the quinone methide (XIX) are detectable on paper chromatograms. This process yields $\beta\gamma$-*bis*-aryl ethers of guaiacylglycerol. The γ-aryl ether linkage is very easily hydrolysed. Such $\beta\gamma$-*bis*-aryl ethers of guaiacylglycerol also occur among the normal conversion products of dehydrogenated coniferyl alcohol; for example, the substance designated as guaiacylglycerol-*bis*-coniferyl (ether XXV). Here coniferyl alcohol acts as phenol and adds onto the quinone methide (XIX). Dilute acid splits the compound into coniferyl alcohol and substance (XX). A second example of this group occurs in substance (XXVI), which is an adduct of preformed substance (XV) onto the quinone methide (XIX). Both are hydrolysed within a few days, even in aqueous solution and disappear rapidly from the bulk of the dehydrogenation products of coniferyl alcohol on continued dehydrogenation. We do not doubt that a portion of these products are incorporated directly into the lignin but we believe that they are still reactive after inclusion and may undergo rearrangement. I have only spoken so far of *one* quinone methide, substance (XIX), because this has been identified. However, it is obvious that other analogous quinone methides occur as intermediates. Addition onto quinone methides is the second growth principle of a lignin macromolecule of any considerable size. A third principle recognized recently is the polymerization of the quinone methides.

The discussion so far is based on isolated intermediates of lignin synthesis of known constitution.

Numerous other intermediates have not yet been isolated and identified and may harbour some surprises. Two types can be forecast with a high degree of certainty, even if not yet isolated. One of them is represented by formula (XVIII). The corresponding trimethoxydiphenylether dicarboxylic acid has been isolated. As mentioned above, when treated with strong alkali, methylated and oxidized, the dehydrogenation product of coniferyl alcohol

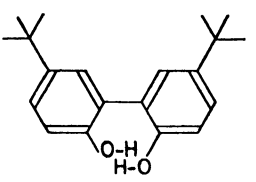
yields trimethylgallic acid among other acids. A dimer (XVIII), originating from R_α and R_γ, would explain its formation. The other dimer is the substance from which benzenepentacarboxylic, tricarballylic and the *o*-phthalic acids are derived. It must be of the isolignan type; it might be better to term these cyclolignans[5]. (XXVII) shows a trihydroxyguaiacyclolignan which might occur as an intermediate. Its formation can be explained as entailing the double quinone methide formed by combination of two R_β radicals; after addition of two moles of water, this could condense intramolecularly to give (XXVII). Such cyclolignans are tetralin derivatives which can be aromatized and which yield benzenepentacarboxylic acid on oxidation. Tricarballylic acid can also be portioned off and the *o*-phthalic acids can be explained in the same fashion. It is, therefore, permissible to include such a cyclolignan in a lignin structure scheme containing a sufficiently large number of units.

So far, I have stressed the synthesis of lignin and the intermediates in this synthesis. Besides oxidation reference has been made to analytical results, such as the hydrogen balance. Recently we have studied the participation of carbon atom-5 of the coniferyl alcohol in the dehydrogenation.

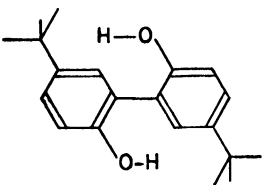
The 5-position in coniferyl alcohol, *i.e.* the unsubstituted position next to the phenolic hydroxyl, is involved in three different functions in the intermediates mentioned above: in substance (XV) it is linked by dehydrogenative condensation directly to the β-carbon atom of the side chain; in substance (XVIII) by etherification with one phenol oxygen; and in (XVII) and (XXI) in diphenyl bonds. The number of units reacting in position 5 can be measured by converting 5-deuteroconiferyl alcohol (XXII) into its dehydrogenation polymer. This was done in both ordinary and heavy water. Normal coniferyl alcohol was also dehydrogenated in heavy water and absorbed a few per cent of deuterium in the process. The results of these experiments indicate that about 45 per cent of all coniferyl alcohol units react in the 5-position during lignification.

Analysis in lignin chemistry will now be discussed, albeit incompletely for the sake of brevity, as the data are extensive and have been gleaned from the work of many laboratories, particularly Swedish.

It is only since it became possible to isolate a considerable proportion of the lignin from wood without chemical interference, as Bjorkman's milled wood lignin, that an analytical investigation of lignin became feasible.



(XXVIII)



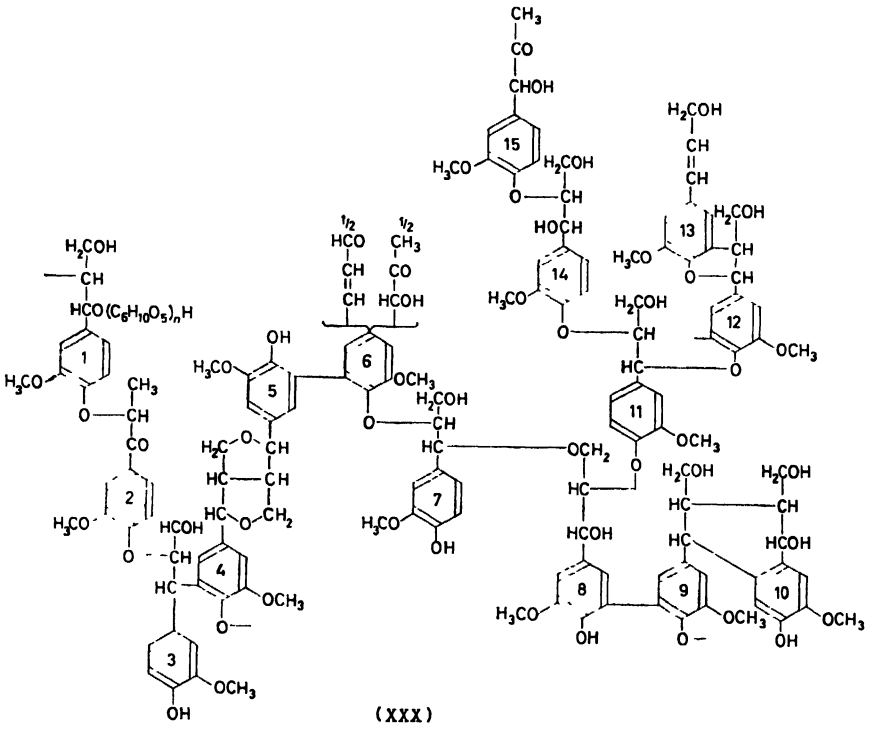
(XXIX)

This comprises aromatic and aliphatic hydroxyl, keto and aldehyde groups, alkyl and aryl ether groups, free, unsubstituted guaiacyl groups, *etc.* Both chemical and optical methods have been used. The principal advances in this field are due to Adler, Marton and their collaborators[4]. In most cases, the chemical and optical assay methods were tested on simple model

compounds and then applied to lignin. However, here, the influence of unsurveyable topochemical and conformational effects makes analysis more complicated. For example, diphenyls bearing two *o,o'*-hydroxyls or one hydroxyl and an ether function will behave differently if the oxygen atoms can approach each other as in (XXVIII) or when, after rotation, they lie on opposite sides of the diphenyl axis as in (XXIX). In a small molecule, one definite conformation will be preferred, but in the massive lignin structure, the conformation of such a bond is determined by quite different factors. Even crosslinking may be possible.

If reactions and kinetic studies carried out on small molecules be applied to a macromolecule containing the corresponding groups, it is to be expected that side reactions occurring with different groups in the larger molecule can scarcely be avoided, and that the products of such reactions may remain occluded within the molecular framework. Optical methods, in particular, may lose some of their precision in this way. However, evaluated with caution, comparative measurements using models provide very valuable data.

About a year ago, a first attempt was made by the author and Sidhu[6] to construct a tentative scheme illustrating the constitution of lignin and comprising 15 coniferyl units. The time is ripe for such attempts to be earnestly considered. They illustrate the concepts recognized so far and, as has already become evident, they form a suitable basis for discussions. Such a draft is made up of the intermediates identified from studies of lignin



(XXX)

synthesis. The growth pathways disclosed above have formed the basis for the combination of the individual building stones. Any such draft must be tailored to fit the number and type of hydroxyls, the number of condensations in the 5-position, the number and type of ether bonds, carbonyl groups, olefinic linkages, and much more.

The second draft shown here (XXX) is an adaptation of the first[6], incorporating the results of the intervening year, particularly the results of the oxidative degradation, studies on the intermediates, and analytical data. Other alterations involving the side chains are unimportant. Naturally, an illustration of this type is necessarily imperfect. Bjorkman lignin and dehydrogenation polymers have molecular weights of the order 8,000 and, hence, consist of more than 40 units. Lignin in spruce wood and sufficiently dehydrogenated synthetic lignins, have even higher molecular weights. Thus, it is impossible to include all possible forms in a scheme with only 15 units. Moreover, as stated above, many intermediates are still unidentified and may encompass forms completely unaccounted for by both the synthetic and analytical modes of approach. Such a tentative scheme is, therefore, only an approximation based on the status of current knowledge, and future embodiments will have to be adapted step by step to conform with new information. This was in fact done when, thanks to a useful symbiosis of synthesis and analysis, *Draft 2* (XXX) was evolved from *Draft 1*. Thus, as shown, the second draft is also only intended as a stepping stone. This concept will now be discussed in detail.

Unit 1 has been formed from one half of the quinone methide intermediate (XIX); a polysaccharide has been added to this. After loss of its phenolic hydrogen, the aryloxy radical of unit 1 combines with a radical R_β (XIII), which is converted into a β-substituted coniferyl alcohol and undergoes an allylic rearrangement. After dehydrogenation of 2 to its aryloxy radical, unit 3 is added in its R_β form. The linkage between the quinone methide and the unit 4 involves one of the addition reactions[7]. It is feasible that the quinone methide at 3 reacts directly with carbon atom-5 of the dehydrogenated unit 4. The phenol group of unit 4 is etherified forming a branching point. Units 4 and 5 both pertain to a pinoresinol molecule which is joined by a diphenyl bond through carbon-5 of unit 5 to the corresponding position in unit 6. The side chain in unit 6 represents half a cinnamic aldehyde, half a ketol. The conformation of the diphenyl bond linking units 5 and 6 is represented in such a way that the phenol hydroxyl of 5 and the ether group of 6 assume a type of *trans* configuration (*cf.* XXIX). The bulk of the entangled aggregates 1–5 and 6–15 prevents normal free rotation. This completes the chain from 1 to 6. The discussion will now be continued starting from unit 15.

Together, 15 and 14 correspond to substance (XX) with the difference that the cinnamyl alcohol group in 15 has undergone rearrangement and oxidation. After removal of its phenolic hydrogen, unit 14 combines with an R_β radical, which first forms a quinone methide as unit 11, onto which the phenol group of substance (XV) is added in the form of units 12 and 13. The 5-position in unit 12 is also condensed. Unit 11 loses its phenolic hydrogen and adds on an R_β radical as unit 8, which then adds on water giving it initially three hydroxyl groups. Formation of a diphenyl bond

then links it up to the cyclolignan composed of 9 and 10. The formation of the oligomer 8 through 15 is now complete.

The entities 1–6 and 8–15 now combine as follows. Unit 6 loses its phenolic hydrogen either by direct dehydrogenation or radical transfer; an R_β radical couples with the aryloxy radical forming unit 7, at present still in the form of a quinone methide. This quinone methide can combine with the primary hydroxyl of unit 8. This completes the whole chain from 1 to 15. It can be seen how larger, prefabricated sections of the lignin chain are welded together by the bifunctional role of the R_β radical.

The units 3 and 11 have a similar function. Unit 11 forms one of the branching points; branching also occurs at units 4, 8, and 12. The bond between units 3 and 4, and the side chains with β-carbonyl groups (in units 6 and 15), are still only hypothetical. All other types of bonds between units, and the various modifications of the coniferyl monomer, are based on experimental results. However, the sequence of the constituent units and their relative proportions are arbitrary. Dimers are included in the forms of pinoresinol (XVI), dehydrodiconiferyl alcohol (XV) and the cyclolignan (XXVII) respectively.

The construction of this scheme is based on an idealized lignin, composed entirely of coniferyl units and of composition:

$$C_9H_7O_2(H_2O)_{0\cdot37}(OCH_3)$$

In comparison with coniferyl alcohol, $C_9H_9O_2(OCH_3)$, this artificial lignin has lost two atoms of hydrogen and gained 0·37 molecule of water. In reality, in a formula entailing 15 units, about 2·1 units of *p*-coumaryl alcohol and 0·9 unit of sinapic alcohol should be entailed with 12 of coniferyl alcohol. The composition of this polymer would be $C_9H_{7\cdot08}O_2(H_2O)_{0\cdot37}(OCH_3)_{0\cdot92}$ and hence would agree favourably with that of conifer lignin.

One open question is the presence of C-methyl groups at the ends of side chains. It is possible that these are formed mainly from cinnamyl alcohols during the estimation procedure; however, they are also found to a small extent when a cold chromic acid treatment precedes the determination. Adler has kindly called our attention to the absence of a guaiacylcarbinol in *Draft 1*. This can be included easily as a (XX)-type substance, but in a scheme entailing only 15 units, all requirements cannot be satisfied. The same applies to the too high proportion of free to etherified hydroxyls.

Investigation of beechwood lignin has indicated that the proportion of syringyl to vanillyl components is in the ratio of 11 : 12; otherwise, there is no significant difference between the condensing principles involved in spruce or beech lignin. Judging by their methoxyl content, deciduous wood lignins never contain more than 50 per cent sinapic component. A dehydrogenation polymer cannot be made from sinapic alcohol alone. If coniferyl alcohol is added, even with an excess of the sinapic component, its content in the biosynthetic lignin cannot be increased. Although it is plausible that inclusion of sinapic alcohol alters the sequence and form of the units, it is indeed remarkable that, in our formula, there is space available on 8 out of 15 units (or 53 per cent) for inclusion of a second methoxyl

group; as in beechwood lignin, sinapic alcohol constitutes less than 50 per cent of the monomers, it seems that coniferyl alcohol occupies some positions which could be held by sinapic alcohol.

All intermediates, as well as synthetic and natural lignins, are optically inactive, despite the presence of numerous asymmetric carbon atoms. This is so because no asymmetric form is produced by the simple abstraction of the phenolic hydrogen, and all the subsequent processes occur automatically without the participation of enzymes. Since optically active lignans, particularly (+)-pinoresinol, occur in resinous extrusions from spruce, some different type of enzymatic mechanism must be involved in resin metabolism.

It is worthwhile comparing lignin with other macromolecules. In the limiting case of linear polymerization, the chain is lengthened by unitary addition of monomeric units. All the units are identical and the bonding is of a uniform type. Lignin is just the opposite from this. Apart from the fact that three different *p*-hydroxycinnamyl alcohols react together in admixture, hardly a single unit is the same as another although the majority originate from the same monomer, as can be seen from the formula. Initially, oligomers of great diversity are formed, including even some bound to polysaccharides.

The foundations for constructing such a formula are given by the type and quantity of the intermediates occuring in the lignin biosynthesis, together with the constructional principles deduced. Furthermore, limitations are set by the need to conform to the type and number of functional groups, such as hydroxyl, ether linkages, *etc.* In constructing a formula representation based on these considerations, further limitations are encountered as only a few forms are compatible with the previous ones. The freedom with which one can proceed is so restricted that it is not at all easy to draw up a scheme which satisfies all the numerous conditions, and it is surprising how similar various drafts starting from different points become in the end.

With no other known substance is a knowledge of the constitution so dependent upon the clarification of its mode of genesis as with lignin. All the experiments and considerations reported above depend upon the assumption that in the biosynthesis of lignin, *p*-hydroxycinnamyl alcohols, principally coniferyl alcohol, are the ultimate monomolecular C_6–C_3 compounds, and only after their generation in the plant does polymerization to larger molecules occur. Another opinion occasionally expressed is that the characteristic features of these starting materials, such as the methoxyl or primary alcohol groups, are formed at a later period, after commencement of the condensation. This conjecture is invalidated by everything that has been ascertained about the formation of lignin from *p*-hydroxycinnamyl alcohols.

It is another question as to whether other extraneous phenols, which can also be dehydrogenated by laccase or peroxidase and which may be present in the cambium and surrounding tissues, may be copolymerized into the lignin. For this reason, we have examined " cambial sap " closely, trying to detect foreign phenols or their glycosides, but have found no indication of their occurrence in significant amounts. If they are not

present, obviously they are not incorporated. The presence of hydroxymatairesinol[8] is of particular interest. This occurs in the acetone-soluble fraction of spruce wood in the remarkable quantity of 0·3 per cent of the wood, corresponding to about 1 per cent of the lignin content. It is a precursor of the conidendrin found in spent sulphite liquor. In " cambial sap " as we prepared it, hydroxymatairesinol occurs in noticeable quantities. Perhaps it originates from another vessel system, *e.g.* the resin ducts. Hydroxymatairesinol is optically active, in contrast to the lignin intermediates. This indicates that it does not belong to the lignification system. On the other hand, the sap contains small but readily identifiable amounts of coniferyl alcohol and the most abundant dimeric lignification intermediates. Besides large quantities of various sugars, there are considerable amounts of coniferin in the sap, but so far the glycoside of sinapic alcohol (syringin) has not yet been observed in cambial sap from spruce. However, a small amount of the glycoside of *p*-coumaric alcohol has been found accompanying coniferin.

The following experiment with tracers illustrates once more the ability of coniferyl alcohol and phenylalanine to form lignin in spruce. By treatment with hot concentrated mineral acids, a synthetic lignin produced from coniferyl alcohol labelled at the end of its side chain yields formaldehyde which is radioactive, and which therefore originates from the α-carbon atom. Identically labelled coniferin produces in spruce a radioactive lignin which also yields radioactive formaldehyde.

Coniferyl alcohol labelled at the central carbon atom of the side chain gives a dehydrogenation polymer from which radioactive isohemipinic acid is obtained, *e.g.* from substance (XV). Isohemipinic acid derived from radioactive lignin obtained after administration of β-^{14}C-phenylalanine to spruce was also radioactive.

An experiment with radioactive glucovanillin demonstrated the fact that phenols which do not belong to the *p*-hydroxycinnamyl alcohol class can be incorporated into lignin. Introduced by our shoot immersion procedure, the radioactivity of the glucovanillin distributed itself in the tiny spruce stem mainly in the lignin directly above the shoot. However, when the radioactive lignin was extirpated and degraded according to Hibbert to give the well known group of C_6–C_3 ketones, these proved to be almost inactive. This means that only a small amount of the vanillin from the glucovanillin is built up to a C_6–C_3 compound corresponding to a lignification monomer which would be capable of yielding the ketones. The major part of the vanillin is incorporated into lignin in some form other than C_6–C_3. Similar observations were made by Kratzl[1] when he applied *p*-creosol to spruce. A small part of it was transformed into C_6–C_3 lignin units.

The *p*-hydroxycinnamyl alcohols are the main intermediates in the biogenesis of lignin. As mentioned above, coniferyl alcohol occurs in minute amounts in cambial sap. However, the majority is present as the glucoside coniferin. I am inclined to believe that the coniferyl alcohol can be converted directly into lignin together with the accompanying small amounts of *p*-coumaric and sinapic alcohols, and that only an excess of coniferyl alcohol is stored in reserve as coniferin. The aglycone of radioactive D-coniferin is transformed very readily in spruce into genuine lignin

which yields radioactive Hibbert ketones. By contrast, radioactive coniferin synthesized from L-glucose is not metabolized. The explanation is due to the presence, in spruce, of a tissue bound β-glucosidase in the layers of young cells undergoing lignification. It can, therefore, be concluded that both free *p*-hydroxycinnamyl alcohols, and their D-glucosides are suitable lignin progenitors.

Thus, the problem of lignification is divided into two main sections: the biochemical formation of *p*-hydroxycinnamyl alcohols, and their conversion into lignin. The pathway followed in converting *p*-hydroxycinnamyl alcohols into lignin has been discussed above. That from glucose to the *p*-hydroxycinnamyl alcohols has been largely explored by the work of many authors. This route leads *via* shikimic and prephenic acids to phenylpyruvic acid, which is in biochemical equilibrium with phenyl-lactic acid, cinnamic acid and phenylalanine. One or more of these acids is converted into the *p*-hydroxycinnamyl alcohols in several reaction stages, about whose sequence little is known. The results so far obtained are somewhat inconclusive since the various authors have experimented with different kinds of plants, and the plants behaved differently. This is illustrated by the fact that *p*-hydroxyphenylpyruvic acid, for example, is converted into lignin readily in Gramineae but hardly at all in spruce.

What is the fate of lignin when dehydrogenases attack it further? If coniferyl alcohol is shaken in extremely dilute aqueous solution with air or oxygen and laccase at pH 5·5–6·0, oxygen is absorbed as long as the laccase remains active. After the addition of fresh enzyme, the oxygen consumption proceeds further, even after the coniferyl alcohol has been converted into lignin. Bjorkman lignin brought into extremely fine suspension by dissolution in acetone and reprecipitation with water is also oxidized by laccase and air, some of the lignin going into solution in the process. Some also assumes humin-like character. The reaction leading to lignin, therefore, destroys it again on proceeding further. This does not occur in the healthy plant since the enzyme activity ceases on completion of lignification. However, if dehydrogenating enzymes come into contact with the wood from external sources, such as is the case with wood rotting fungi, the lignin is attacked. The laccase in the mycelia of common mushrooms, which are not lignified, enables them to degrade lignin and phenols in general for their own assimilation.

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BIOGENESIS OF CARBOHYDRATES IN WOOD

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It is believed that the primitive earth had on its surface only simple carbon compounds. It is generally agreed that the atmosphere was a reducing one and that most of the carbon was in the form of methane and carbon dioxide, with methane predominating. There was much hydrogen and water. Calvin has shown that irradiation of a mixture of water and carbon dioxide in a cyclotron yielded formic acid, acetic acid, succinic acid and other derivatives[1]. If carbon dioxide was replaced by methane and ammonia was added the more rapid formation of these acids was observed and, in addition, glycine and other amino-acids were produced[2, 3].

Formic acid on decomposition yields carbon monoxide and this with hydrogen could yield methanol and formaldehyde. Many years ago it was shown that formaldehyde could be polymerized by basic solutions, such as lime water, lead carbonate or an aqueous suspension of calcium carbonate, to yield a mixture of sugars called *formose*[4]. This mixture has been analysed and the mechanism of its formation has been investigated extensively. It contains mainly pentulose and hexulose sugars together with traces of heptuloses and substances with fewer carbon atoms[5]. The mixture of sugars has been reduced and the acetylated materials were examined in the gas chromatographic apparatus. The major products were identified as

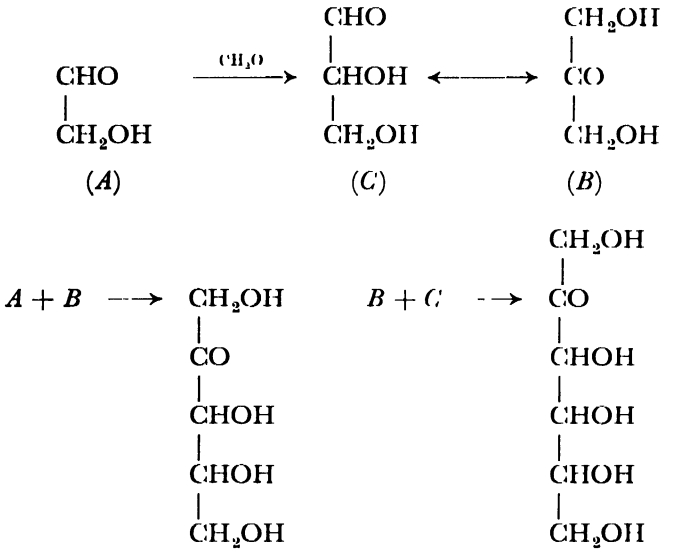


Figure 1. Formose synthesis

penitol and hexitol acetates[6]. These sugars are produced by mechanisms shown in *Figure 1*.

The key compound is glycollic aldehyde (*A*) which, it has been suggested, catalyzes the reaction according to the following scheme: It reacts with formaldehyde to yield DL-glycerose (*C*) which isomerizes to dihydroxypropanone (*B*), which in turn condenses with a molecule of formaldehyde to yield DL-erythrulose (*D*), which isomerizes to a tetrose (*E*), the tetrose then splits into two molecules of glycollic aldehyde (*A*) and the reaction continues[7] (*Figure 2*).

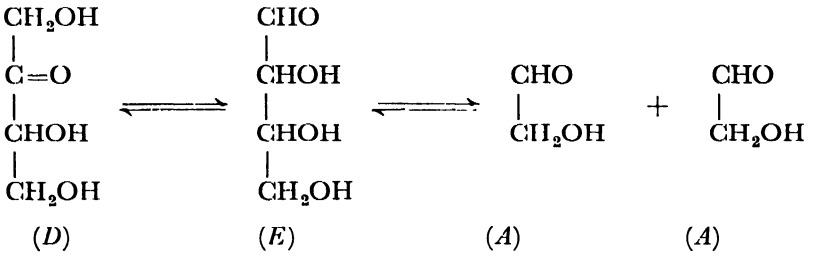


Figure 2

The ketoses which are first produced are converted in part to aldoses under the influence of basic catalysts. It is very interesting that the major substances produced in the formose reaction are those which are commonly encountered in natural products, *e.g.* ribulose, xylulose, fructose and heptulose. None of the products produced in the formose reaction is optically active. Optical activity may have developed as a result of irradiation by polarized light, from the moon for example or resolution may have occurred by chromatography over optically active materials such as quartz or even by zone melting or by some other mechanism. Why the D-form of glucose rather than the L-form should be favoured is unknown at present.

The development of plants took place some 400,000,000 years ago. The consequent conversion of carbon dioxide to complex polysaccharides takes place on a very large scale, an example is illustrated in *Table 1*.

Table 1. Comparison of the amount of food produced by photosynthesis and the amount of wood produced by stands of several tree species

Species	*Annual net photosynthesis* (kg carbon per hectare)	*Annual net production of wood* (kg carbon per hectare)
Douglas Fir	18,500	7,000
Beech	13,500	3,700
Spruce	11,100	4,100
Larch	10,700	3,100
Birch	8,900	2,300
Oak	6,300	2,800
Scots Pine	4,700	2,900

The mechanism of conversion of carbon dioxide to sugars and to polymers has been studied by Hassid[8], a pioneer in this work, and by Krotkov[9], Calvin[1, 2], Neish[10] and by many others. ^{14}C-labelled compounds have generally been used to follow the interconversion of the sugars. While much is known of the origin of sugars in plants, relatively little information is available on carbohydrate metabolism in trees. Thus, many of the suggested metabolic pathways result from analogy with known sources in plants and fungi.

Some sugars occur in the free form in plants and trees, *e.g.* D-glucose, L-arabinose, L-fructose, L-rhamnose, vitamin C, hexitols, heptitols, octitols and sucrose and its derivatives. The following are some sugars which are found combined in polysaccharides:

D-Glucose

D-Galactose (and its 3-*O*-methyl ether)

D-Mannose

D-Fructose

L-Arabinose

D-Xylose (and its 2-*O*-methyl ether)

L-Rhamnose

L-Fucose (and its 2-*O*-methyl ether)

D-Glucuronic acid (and its 4-*O*-methyl ether)

D-Galacturonic acid

It appears that all these sugars arise from carbon dioxide *via* triose fragments (*H*) and D-fructose-1,6-diphosphate (*I*) (*Figure 3*). It has been shown

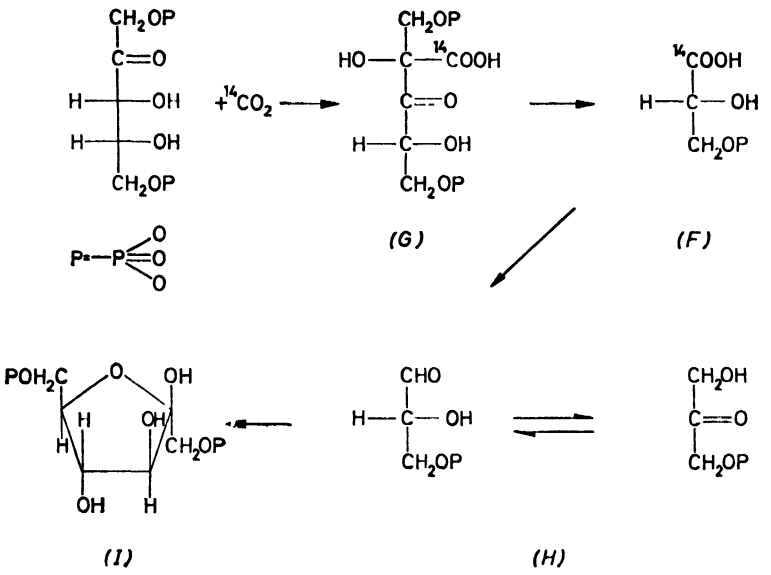


Figure 3. Origin of hexoses

that carbohydrates may be phosphorylated preferentially on the primary alcohol groupings by heating them with phosphoric acid or with its salts[12]. Perhaps this is how the vital sugar phosphates were first produced long ago.

Calvin and his co-workers[1] have shown that the first product of photosynthesis is 3-D-phosphoglyceronic acid (*F*) which arises from a D-erythro-pentulose diphosphate (*G*) derivative.

Krotkov, Nelson, and Shiroya[13] of Queen's University have kindly allowed me to use some of their results to illustrate the photosynthetic activities of pine trees. When a seedling of *Pinus strobus* is allowed to photosynthesize for eight hours in light and in an atmosphere of $^{14}CO_2$, sucrose and raffinose are the major primary water-soluble products.

The photosynthesized material is translocated mainly as non-reducing saccharides, as is illustrated in *Table 2.*

Table 2. 2500 FT-c photosynthesis

	Shoot (%)	*Stem* (%)	*Root* (%)
Pentose	—	—	—
Fructose	11·2	1 5	1·3
Glucose	10·0	1 4	2·3
Sucrose	74·3	94 6	94·2
Disaccharide	0 9	0·8	—
Raffinose	1 2	1·7	1 7
Trisaccharide	0·1	—	—
Neutral substance	1·8	—	—
Organic acid	0·5	—	—
Total	100 0	100 0	100·0

The small amounts of acids which are produced are of particular interest as they contain shikimic and quinic acids which may be utilized to make lignin—an aspect of carbohydrate chemistry which was discussed earlier by Freudenberg[11] (see *Table 3*).

Table 3. Distribution of ^{14}C in organic acid fractions as percentage of total activity spotted on chromatogram

Organic acid	*Expt 11A* (%)	*Expt. 11B* (%)
Quinic acid (overlapped with oxalic acid)	39·5	23 9
Shikimic acid	7·5	5·4
Citric acid	9·8	16·3
Malic acid	19 4	28·5
Succinic acid	2 5	3·1
Fumaric acid	0·8	1·3
Unknown I (Hexose phosphate?)	12·8	14·5
Unknown II (Phosphoglyceronic acid?)	5·7	4·4
Unknown III	2·0	2 6
Total	100 0	100·0

Sucrose is a common major component of the fluid in the sieve tube but higher molecular weight sugars are also found[15] (*Table 4*).

Table 4. Sugars in sieve-tube exudate of some American trees

Family	*Name of tree*	*Sucrose* (%)	*Raffinose* (%)	*Stachyose* (%)	*Verbascose* (%)
Salicaceae	Aspen	>10	Trace	Trace	-
Fagaceae	Beech	>10	Trace	—	—
	Chestnut	>10	Trace	—	—
	White Oak	>10	—	—	—
	Chestnut Oak	>10	—	—	—
	Red Oak	>10			-
Ulmaceae	Elm	2–10	2–10	2–10	-
Magnoliaceae	Tulip Tree	>10	Trace	Trace	—
Rosaceae	Black Cherry	>10	Trace	—	—
Leguminosae	Black Locust	>10		—	-
Aceraceae	Striped Maple	>10	Trace	—	
	Sugar Maple	>10	—	—	—
Rhamnaceae	Buckthorn	>10	Trace	Trace	—
Tiliaceae	Basswood	>10	0·5–2	0·5–2	
Nyssaceae	Black Gum	>10	Trace	-	-
Oleaceae	White Ash	2–10	2–10	10	Trace

These are derivatives of sucrose and contain α-linked D-galactopyranose residues (*Figure 4*). Free D-galactose does not appear in the sieve tube fluid. It is of interest that α-linked D-galactopyranose residues are found in some wood polysaccharides, (*e.g.* glucomannans)[16].

Maple sap besides containing sucrose also contains fructosylsucrose and a polysaccharide which is composed of L-arabinose, D-galactose and L-rhamnose[17]. The problem is to explain the origin of these and other sugars from carbon dioxide.

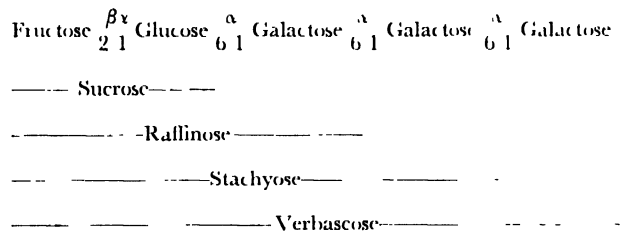


Figure 4. D-Galactose containing oligosaccharides

Mannitol, a common constituent, is believed to arise from D-mannose by reduction, the hexose sugar itself arising from D-fructose-6-phosphate by isomerization. Sorbitol is formed by the reduction of D-fructose, this is a reversible reaction.

As we saw earlier, the D-fructose, 1,6-diphosphate (*I*) originates from the condensation of two triose fragments under the influence of the enzyme aldolase. D-Fructose-6-phosphate is split by the enzymes *transaldolase* and *transketolase* in different ways as is illustrated in *Figure 5* [19].

Transketolase and transaldolase require thiamine pyrophosphate as a *co-factor*. These two enzymes between them can cause the formation of pentose residues with no great change of energy and no formation of carbon dioxide as was suggested by Neish a few years ago. The net result is that

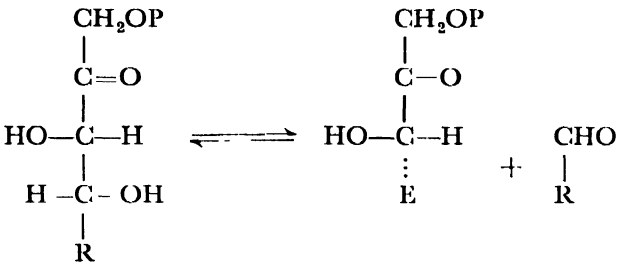


Figure 5(a) Transaldolase reaction

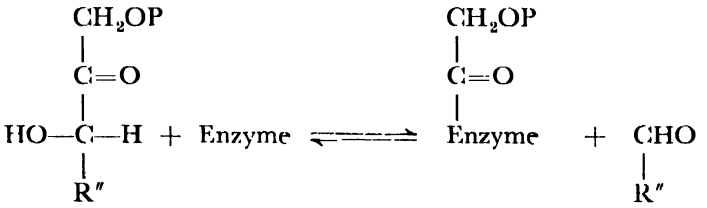
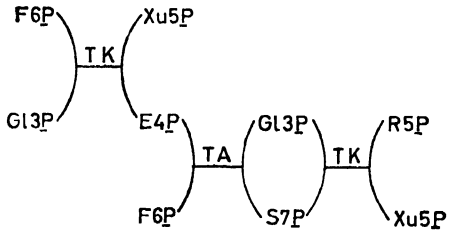


Figure 5(b). Transketolase reaction

two moles of D-fructose-6-phosphate and one mole of D-glycerose-3-phosphate are converted to 3 moles of pentose-5-phosphate[10] (*Figure 6*).



F6P D-Fructose-6-phosphate
Gl3P D-Glycerose-3-phosphate
Xu5P D-Xylulose-5-phosphate
E4P D-Erythrose-4-phosphate
S7P D-Sedoheptulose-7-phosphate
R5P D-Ribose-5-phosphate
TK Transketolase
TA Transaldolase

Figure 6. Origin of pentose sugars

Thus a possible route to pentoses from hexoses can be advanced.

A second method (see *Figure 7*) of pentose formation is *via* the aldolase reaction. For example, D-fructose-1,6-diphosphate is split by aldolase to give dihydroxypropanone phosphate and D-glycerose-3-phosphate (*H*). The enzyme is specific for the ketose only and the triose-aldehyde can be replaced by a diose *e.g.* glycollic aldehyde when D-xyulose-1,5-diphosphate results or with D-erythrose when D-sedoheptulose-1,7-diphosphate is obtained[18].

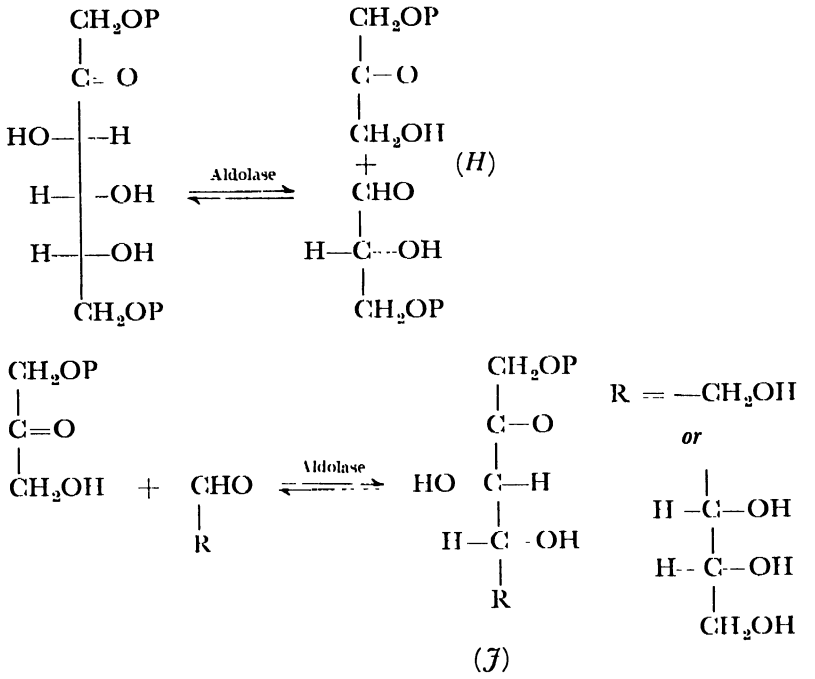


Figure 7. Origin of pentose and heptose sugars

The hydroxyl groups on C-3 and C-4 possess the D-*threo* configuration, *i.e.* they are *trans* to one another. However, if the product (*J*) of enzyme action existed initially in the zig-zag conformation, the two hydroxyl groups would be *cis* to one another as might be expected for a reaction on an enzyme surface (*Figure 8*).

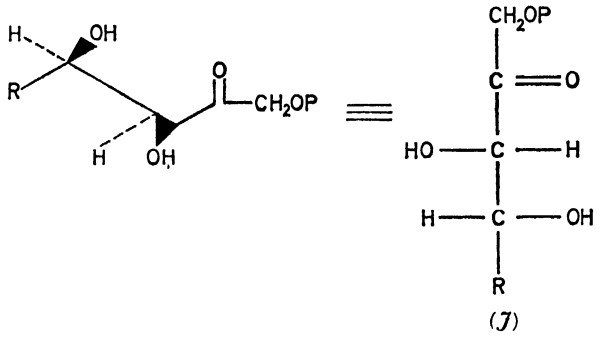
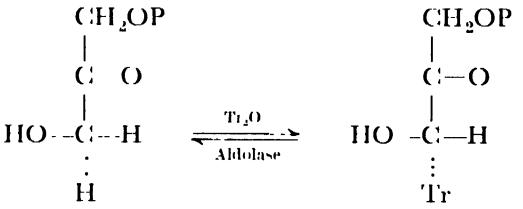
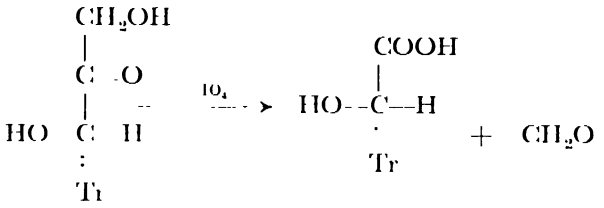


Figure 8

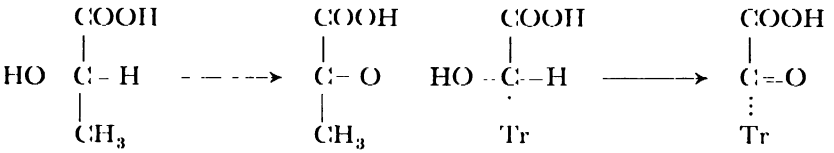
It has been proved conclusively by Rose that only one hydrogen of the dihydroxypropanone phosphate is involved in this reaction in the following way. The substance was incubated in tritiated water with aldolase when one hydrogen was substituted by tritium:



The tritiated product was dephosphorylated and oxidized with periodate:



The glycollic acid was then dehydrogenated with L-lactic acid dehydrogenase. Hydrogen only was removed[20]:



Yet a third method of obtaining the pentose residues which are so commonly found in trees is the decarboxylation process first suggested by the French chemist Maquenne and later amplified and discussed by Hirst[21]. As a result of much work mainly by Hirst[22], Hassid[8] and Neish[10], it has been shown that a derivative of D-glucose can be oxidized to D-glucuronic acid and then decarboxylated to a D-xylose derivative. This transformation occurs at the monosaccharide level.

It has been shown recently that the uridine diphosphate derivatives are the key compounds in these transformations. When D-mannose or L-galactose is the sugar transformed, guanosine or thymidine may be the heterocyclic system involved[23a, b].

The mechanism of formation of uridine diphosphate glucose (UDPG) is pictured in *Figure 9*.

Uridine diphosphate glucose is also involved in the formation of sucrose from D-glucose-1-phosphate and very probably it is responsible for the synthesis of starch from D-glucose-1-phosphate. The rarer sugars, L-fucose and L-rhamnose, probably arise from purine-diphosphate-glucose derivatives without fragmentation of the sugar molecule, but the exact sequence of events is unknown. However, it is known that in L-rhamnose synthesis the hydroxyl group at C 6 is probably first converted to CH_3, perhaps, first by elimination of the elements of water followed by reduction and then the

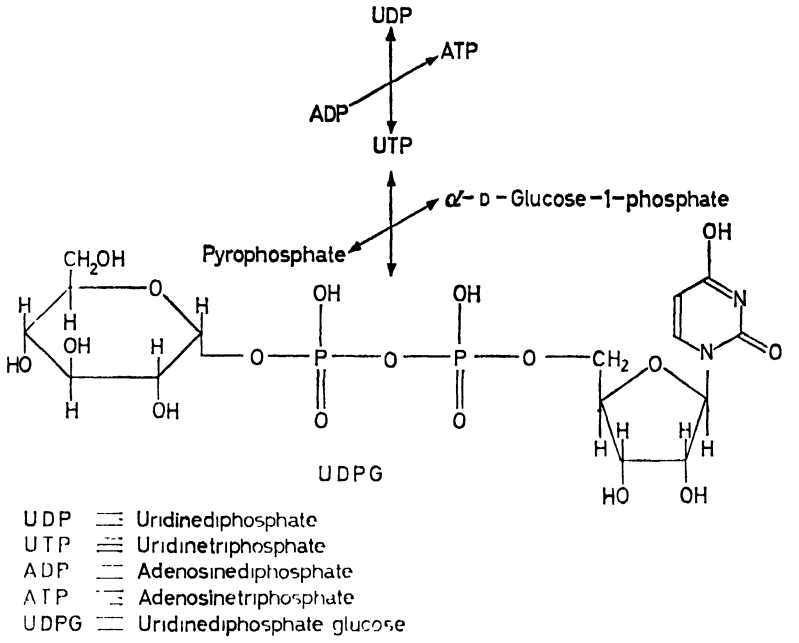


Figure 9 Formation of uridine diphosphate glucose

hydroxyl group at C 1 is oxidized to a keto group[21] (*Figure 10*). (The last two steps may occur simultaneously.)

D-Glucose (100 per cent 1-^{14}C) → L-Rhamnose (89 per cent 1-^{14}C)

as *as*

Thymidine diphosphate D-glucose Thymidine diphosphate L-rhamnose

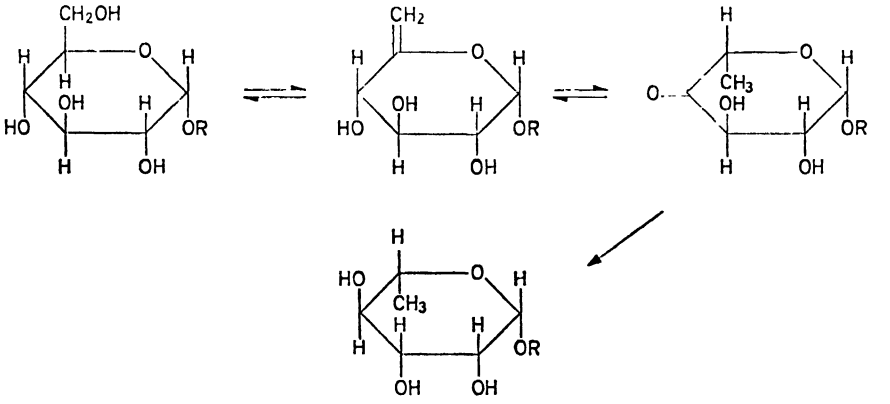
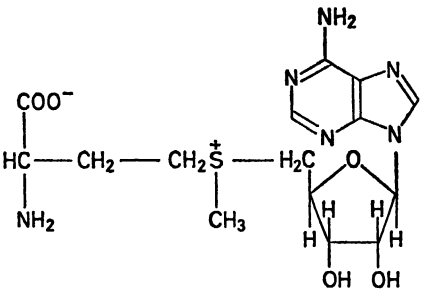


Figure 10 Formation of L-rhamnose

The methoxyl grouping which is found on D-galactose at C-3, on D-glucuronic acid at C 4, on C-2 of D-xylose and L-fucose, on the carbomethoxy group in pectin and which is present as an ether in lignin probably

originate from methionine ($CH_3SCH_2CH_2CHNH_2COOH$) *via* an adenine derivative.



Vitamin C which is found in the needles of spruce and pines may originate from D-glucose* by the following series of reactions as it does in cress seedlings[25].

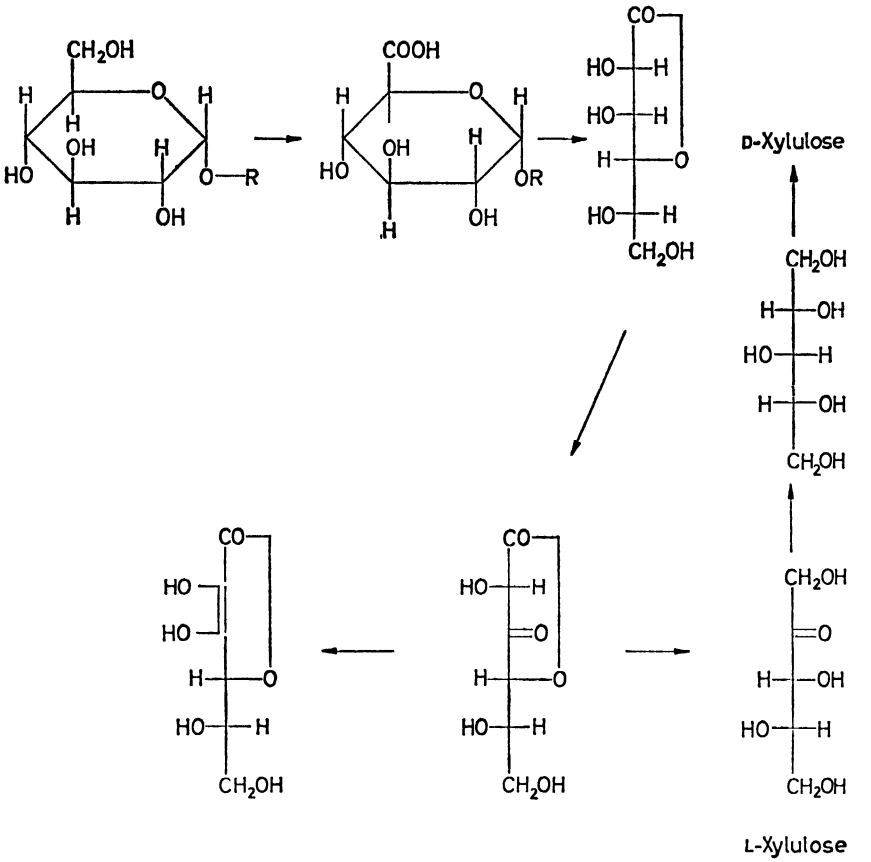


Figure 11. Vitamin C biosynthesis

* Vitamin C may also originate from D-glucose without inversion of the molecule[26] (Loewus; personal communication from Dr A. G. Neish).

The synthesis of some of these sugars from carbon dioxide may be illustrated by showing some of the results of Andrews and Hough[27] who have examined the photosynthesis of sugar in plum leaves. I wish to thank these authors for their kind permission to reproduce these results. They allowed leaves, which had been kept in the dark for 24 hours to photosynthesize in the presence of $^{14}CO_2$. After 5 hours, when all the $^{14}CO_2$ had been absorbed, photosynthesis was allowed to continue in the presence of $^{12}CO_2$. Samples of leaves were removed at intervals and

Table 5. Leaf fractions as percentage of ^{14}C supplied in $^{14}CO_2$

Period of photosynthesis (h)	1	1·5	3	6	15·5	21	48
Activity:							
in leaf extract	18·4	24·5	53 8	58·4	56·3	52·3	49·0
in insoluble leaf residue	6·2	9·5	25 0	39·9	40·7	38·7	32·3
Total	24·6	34 0	78·8	98 3	97 0	91 0	81·3

Table 6 Relative distribution of ^{14}C between leaf extracts

Leaf extract	75	72	68	59 5	58	57 5	60
Insoluble leaf residue	25	28	32	40·5	42	42 5	10

Table 7. Polysaccharide sugar carbon (%) originating from $^{14}CO_2$ after 48 hours photosynthesis

	Water soluble	*Soluble in cold* N NaOH	*Soluble in hot* 2·5N NaOH
D-Glucose		21 6	18·5
D-Galactose	1·66	—	1·72
L-Rhamnose	0·94	—	1 12
L-Arabinose	1·58	0 90	0 74
D-Xylose	0·21	0 30	0 49
D-Galacturonic acid	0·08	—	—

examined. *Tables 5* and *6* show the uptake of $^{14}CO_2$ at various time intervals. The total activity in leaf residues did not exceed that of the leaf extracts. It is evident that the rate of incorporation of $^{14}CO_2$ into metabolically inert polysaccharides is slow under these experimental conditions. Whistler has shown that hemicelluloses (xylan) once formed are not broken down.

The percentage of sugars of the three fractions which are labelled is given in *Table 7.*

Each monosaccharide was crystallized, sometimes with the addition of a carrier, and converted to a crystalline derivative before combustion and assay as ^{14}C-barium carbonate. D-Galactose was converted to mucic acid, L-arabinose to its benzhydrazide, D-xylose to the tosylhydrazone, and D-galacturonic acid was assayed as $NaCa(C_6H_9O_3)_3 \cdot 6H_2O$.

Table 8. 48 h Plum leaves[27]

Monosaccharides from polysaccharides which are:	*Monosaccharide constituent*	*Weight isolated* (mg)	*Specific activity of undiluted sugar* (μc/milli-atom-equiv of C)
Water soluble	D-Galactose	16·0	0 338
	D-Galacturonic Acid	9 3	0·017
	L-Arabinose	14·0	0·320
	D-Xylose	2 6	0 043
	L-Rhamnose	5 5	0 190
Soluble in cold N NaOH	L-Arabinose	5·4	0·182
	D-Glucose	41 0	4·40
	D-Xylose	8 0	0 062
Soluble in hot 2 5N NaOH	D-Galactose	19·7	0 350
	L-Arabinose	31·0	0·149
	D-Glucose	45·3	3·76
	D-Xylose	26·5	0·100
	L-Rhamnose	8 1	0 227

Table 9. Incorporation of 1-(^{14}C)-D-glucose into plum leaf polysaccharides

Period of incorporation (h)	*Polysaccharide fraction*	*Monosaccharide constituent*	*Weight isolated* (mg)	*Specific activity of sugar carbon* (μc/mA)	*Distribution of ^{14}C along carbon chain of monosaccharide* (%) C_1	C_2	C_3	C_4	C_5	C_6
5	Hot water soluble	D-Glucose	9·6	0 53	64·6		13 7			21·7
		D-Galactose	11 6	0 031	77·3		6·7			16·5
		L-Arabinose	15·4	0 027	91·4		6·2			2·4
		D-Xylose	2·9	0 020	65 0		28·8			6·2
		L-Rhamnose	3·7	0 022	56·4		7·8		1·9	33·9
5	Hot alkali soluble	D-Glucose	40·3	0·815	59 9		12·8			27 3
		D-Galactose	26 0	0·035	68·5		12·9			18·7
		L-Arabinose	95 3	0 017	85·0		9·1			7·1
		D-Xylose	42·1	0·016	89·0		7·0			4 0
		L-Rhamnose	19·0	0·022	78·5		3·0		1·1	17·4
5	Cellulose	D-Glucose	138·6	0·0094	71·8		6 6			21·6
20	Cellulose	D-Glucose	115·5	0·0072	77·8		1·8			20·4

The activities of the sugars varied from fraction to fraction as is illustrated in *Table 8.* This effect has been noted in starch metabolism by Perlin[29], Porter and others. The very small activity in D-galacturonic acid suggests that pectic acid is not a very active metabolite and that once D-galacturonic acid is produced it is decarboxylated to L-arabinose. Note also that the rate of conversion of D-glucose to D-xylose is not very high because starch is a major product which is metabolized. It suggests that xylan is not an active metabolite[30].

It is interesting that no sucrose was detected—it was probably present during the early stages of photosynthesis.

Table 10. Incorporation of 6-(^{14}C)-D-glucose into plum leaf polysaccharides

Period of incorporation (h)	*Polysaccharide fraction*	*Monosaccharide constituent*	*Weight isolated* (mg)	*Specific activity of sugar carbon* (μc/mA)	*Distribution of* ^{14}C *along carbon chain of monosaccharide* (%) C_1	C_2	C_3	C_4	C_5	C_6
5	Hot water soluble	D-Glucose	24·1	0·21	28·6			1·8		69·5
		D-Galactose	19·9	0 024	21 6			3·2		75·2
		L-Arabinose	16 4	0 0029[a]	66 6			24 9		8·3
		D-Xylose	4·9	0·0058[a]	78 0			32·0		0·0
		L-Rhamnose	7·2	0·017	20·0			4 0		2·9 73
5	Hot alkali soluble	D-Glucose	65·1	0·17	27·1			10·7		62·2
		D-Galactose	10·7	0·015	19 5			15·0		65·5
		L-Arabinose	21·3	0·0023[b]	51·0			26·5		22·5
		D-Xylose	20·0	0 0014[b]	40 0			14 0		16 0
		L-Rhamnose	1·1	0 0062	10 8			13·0		[c] 76·3
5	Cellulose	D-Glucose	46·8	0 0034	23·1			1·9		75·0
20	Cellulose	D-Glucose	116·6	0·0065	21·5			6 0		72·5

[a] Activities determined to standard deviation of 4%.

[b] Activities determined to standard deviation of 10%.

[c] C_6 lost during preparation of sample.

Tables 9 and *10* illustrate the changes in radioactive polysaccharides during photosynthesis. Cellulose metabolism is not reversible and the intermediates are slowly converted to cellulose.

In conclusion, the conversion of sugars to aromatic compounds may be illustrated. These pathways are known to occur in fungi and are very probably present in plants and trees according to the following scheme.

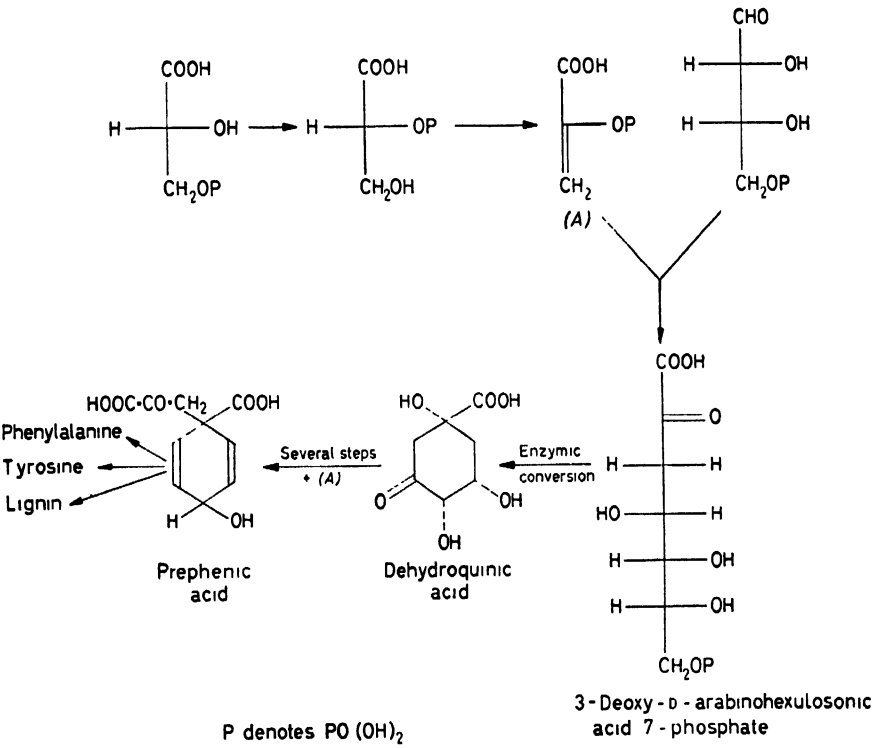


Figure 12. Biosynthesis of phenylalanine, tyrosine and (?)lignin

It will be noted that D-glycerose-3-phosphate and D-erythrose-4-phosphate are intermediate in an aldol-like condensation which results in the formation of a 3-deoxy-2-keto-D-arabinohexulosonic acid which then undergoes conversion to an intermediate cyclohexane derivative[31].

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CHEMICAL AND MORPHOLOGICAL ASPECTS OF THE FINE STRUCTURE OF WOOD

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In former days, wood chemists were mainly interested in obtaining information on the over-all chemical composition of trees. In recent years, however, wood anatomists, fibre morphologists, and wood chemists have started to co-operate with each other and to correlate the physical with the chemical aspects. This correlation has resulted in a much better understanding of the chemical complexity of wood.

In the present paper an outline will be given of fibre morphology and fibre chemistry as well as of the chemistry of anatomically distinct elements in wood.

The scope of the morphology of wood fibres, *i.e.* softwood tracheids and libriform fibres from hardwoods as well as other cell-elements in wood, was initially more-or-less restricted to the description of the outer shape of these cells and the arrangement of the pits. In 1926, Ambronn and Frey-Wyssling[1] showed, with the aid of the polarizing microscope, that a step forward could be taken from *microscopic* to what they called *submicroscopic* morphology. Somewhat later, X-ray methods were also applied to the investigation of the submicroscopic morphology of fibres and ten years ago electron microscopy became the ultimate tool in the field. It was then possible to see bundles of some hundred cellulose molecules, the so-called microfibrils, and it was recognized that high polymer compounds could arrange themselves into molecular aggregates peculiar to a certain molecular species.

Of the three main components in the cell walls of wood fibres, *i.e.* cellulose, hemicellulose and lignin, the cellulose has been studied most thoroughly with regard to submicroscopic structure. There has been much argument over the fine structure of the cellulose microfibrils. Mühlethaler[2], with the aid of a new electron microscopic preparation technique, the so-called " negative contrast " technique, as opposed to the shadowing technique, has now shown that the narrowest fibrils, which he calls elementary fibrils after Frey-Wyssling, have a diameter of 35 Å. These elementary fibrils, each containing 40 cellulose chains, aggregate to form larger elements, microfibrils, with a diameter which is always a multiple of 35 Å, *e.g.* 70, 105, 140 *etc.*, up to about 350 Å.

Consequently, the following cellulosic structural elements are present in the native cell wall: the cellulose molecule, the elementary fibril, the microfibril, the macrofibril, and the lamellar membrane. The macrofibrils and the lamellar membranes have no definite sizes and can be seen in the light microscope.

The submicroscopic structure of the *hemicelluloses* in wood fibres is rather obscure. Most textbooks say that the hemicelluloses are present as amorphous material between the cellulose microfibrils. However, no proof has ever been given for this statement. Some years ago, we studied the mannan hemicelluloses from dates and ivory nuts[3]. There were two mannans present, termed mannan *A* and *B*. The low molecular weight mannan *A* with a D.P. of 16 was perfectly crystalline and was present in the cell walls in the form of small granules. The high molecular weight mannan *B* with a D.P. of 80 was amorphous, but was present in the cell walls in the form of microfibrils very similar to the cellulose microfibrils in wood fibres (*Figures 1* and *2*). This indicates the following possibilities for the wood hemicelluloses: they may lie *between* the cellulosic microfibrils *either* as an amorphous or as a crystalline granular material, *or* they may form their own microfibrils which may or may not have crystalline regions. A precipitated glucomannan which we had extracted from spruce holocellulose showed no crystallinity on examination with X-rays. After mild hydrolysis, the same glucomannan gave a crystalline X-ray pattern analogous to that of ivory nut mannan *A*. This result, of course, does not shed any light on the submicroscopic structure of the glucomannan in the *native* state in the wood fibre, but it shows that if the native glucomannan molecules do crystallize, then they obviously can do so only with difficulty. If, however, their D.P. is reduced, possibly with debranching in the process, they can build up a crystalline lattice more easily. As all the hemicelluloses in wood are heteropolysaccharides, no pure mannan or pure xylan having been found, and as some are also partially acetylated, the probability that they are crystalline in the native state is not very high. Yet, they need not be completely amorphous. Most of them are only slightly branched *chain* molecules and they must, therefore, have a strong tendency to assume a parallel arrangement. In fact, Liang and co-workers[4] have recently shown, by using the polarized beam technique of infra-red spectroscopy, that the glucuronoxylan and glucomannan chains in softwood, as well as hardwood fibres, are probably arranged parallel to each other. However, the question of whether the glucuronoxylans and glucomannans form their own elementary fibrils or not has not yet been solved. In addition to ivory nut mannan, the mannans and xylans of certain algae have been found to be present in the cell walls in the form of microfibrils[5] and it is, therefore, not unlikely that the same is the case for the wood hemicelluloses, or at least for some of them.

There is not much to be said of the submicroscopic structure of lignin. Lignin differs from the polysaccharides in wood, which are almost entirely long chain molecules, in being a three dimensional macromolecule. This and the heterogeneous structures of the lignin molecules make the formation of a crystal lattice or of any other form of ordered molecular aggregate very difficult and, in fact, lignin does seem to be a completely amorphous material. The wood cells which have been studied most thoroughly with respect to fine structure are hardwood fibres and softwood tracheids. Those of most wood species are very similar. The cell walls usually consist of five layers: middle lamella (M), primary wall(P), transition layer or outer layer of the secondary wall (S_1), central layer of the secondary wall (S_2), and tertiary wall or inner layer of the secondary wall (S_3).

The two layers which are formed first in the cambium are the middle lamella and the primary wall. The middle lamella in the cambial cells is very thin but becomes thicker later when the lignin is deposited. After the delignification of transverse sections of wood, usually no microfibrils are found in the region of the middle lamella which, therefore, can be regarded as being free from cellulose.

The thin primary wall consists of an irregular network of microfibrils (*Figure 3*). In its native state this network is very loose and can easily undergo plastic deformation. The cell walls can grow and enlarge their surface as long as no other wall layers are deposited apart from the middle lamella and the primary wall. When the period of surface growth of a fibre finishes, the deposition of the secondary wall layers begins. The layer S_1, which is deposited directly on the primary wall, has also been called the transition layer because its structure is intermediate between the structures of the primary wall and the secondary wall. The microfibrils run more-or-less parallel to each other at an angle of about 50° to the fibre axis. Electron microscope studies have shown that the S_1-layer consists of two or more lamellae with a crossed fibrillar structure (*Figure 4*). The large angle which the microfibrils make with the fibre axis in the S_1-layer is responsible for its strong double refraction in transverse sections through wood, and is also of great importance in the physical behaviour of delignified wood fibres. The fibrils swell at right angles to their longitudinal axis. The S_1-layer when treated with swelling agents, therefore, shows a tendency to elongate the fibres. The S_2-layer, on the other hand, with the microfibrils running almost parallel to the fibre axis, swells at right angles to the axis and breaks open the framework of the S_1-layer, producing the well-known " balloons ". There is no doubt that the S_1-layer is a severe obstacle to the extensive swelling of fibres. In swelling agents, such as copper ethylenediamine, the swelling forces of the S_2-layer are strong enough to break open the S_1-layer. In less powerful swelling agents, *e.g.* water, the S_1-layer must be mechanically damaged before the fibres can swell without restraint.

In the S_2-layer, the so-called central layer of the secondary wall, the microfibrils are closely parallel and run in a steep helix, which forms an angle of 10 to 20° to the fibre axis (*Figure 5*). Thus, in a thin transverse wood section the S_2-layer shows only a slight double refraction in the polarizing microscope. The contact between S_1 and S_2 is very loose and in transverse sections made with a poor blade it can often be seen that S_2 has become detached from S_1. The abrupt transition between S_1 and S_2 is in striking contrast with the gradual transition between the primary wall and S_1. It is very intriguing to speculate as to why and how the cytoplasm brings about this sudden change in microfibrillar orientation in S_2.

Quantitatively, the S_2-layer is the most important layer of the fibre. Its thickness can vary from about 0·5 microns in spring wood tracheids up to about 4 microns in summer wood tracheids and is a most important factor in the stiffness of the fibres. Delignified spring wood tracheids are very flexible, while summer wood tracheids are stiff. Paper sheets which are made of summer wood tracheids only have a high tearing strength, but low tensile and bursting strength.

In the final layer of the cell wall, the tertiary wall or inner layer of the secondary wall, which borders the lumen, the microfibrils usually run in a flat spiral and, thus, this layer, like S_1, is also birefringent in transverse sections (*Figure 6*). The tertiary wall is usually rather thin, and its inner side adjacent to the cell lumen often has a surface with wart-like structures. The microfibrils are not as closely parallel as in the S_2-layer and their angle to the fibre axis can vary within wide limits as Bucher[6] and Liese[7] have shown.

Special features of the fine structure of fibres are the pits. In the tracheids of coniferous woods they are almost exclusively localized on the radial cell walls and are more numerous at the ends of the tracheids than in the middle parts. They are abundant in spring wood tracheids and relatively infrequent in summer wood tracheids, which means that the impregnation of summer wood is more difficult than impregnation of spring wood. In pine tracheids there are three different kinds of pit: the ordinary bordered pits which connect vertical tracheids (*Figure 7*), the small bordered pits between vertical tracheids and radial ray tracheids, and the large window pits between vertical tracheids and radial ray parenchyma cells (*Figure 8*). The window pits are gaps in the cell wall where the three secondary wall layers S_1, S_2 and S_3 are absent and the window consists of primary wall only. The bordered pits are more complicated. Their structure is best understood from their development. The first sign of the formation of a pit in the young tracheid can be seen in the primary wall at the stage when its surface growth has finished. The network of microfibrils becomes looser at certain places in the primary wall and, at about the same time, new microfibrils are deposited in a circle around the areas of looser network. The microfibrils deposited later in the S_1, S_2 and S_3 layers " flow " around the pits. In the centre of the pit, the structure termed the torus is deposited by secondary thickening of the primary wall. The pits are always formed in pairs on adjacent cell walls. The mechanism which induces the two neighbouring cells to build a pit at the same place and at the same time is not known but there must obviously exist some protoplasmic connection between two adjacent cells. Usually, the bordered pits are functional only for a limited number of years. They are finally blocked by the torus fastening across the opening on one side.

The *chemistry* of the fibre wall has been studied by spectrophotometric as well as by microchemical methods. For the identification of cellulose the electron microscope has also been used. The presence of microfibrils has usually been attributed to the presence of cellulose. It must, however, be pointed out that this conclusion is scarcely permissible as long as it is not known if the hemicelluloses in wood can also form microfibrils.

The distribution of lignin through the fibre wall has been studied most thoroughly. The following techniques have been used: staining reagents, dissolution of the non-lignin compounds, microdissection and chemical analysis, ultra-violet and fluorescence microscopy. All the techniques gave similar results, and it has now been known for some time that between 60 and 90 per cent of the lignin in the wall is localized in the region of the middle lamella and the primary wall. For softwoods, the lignin content lies somewhere near the lower figure and for hardwoods, near the higher

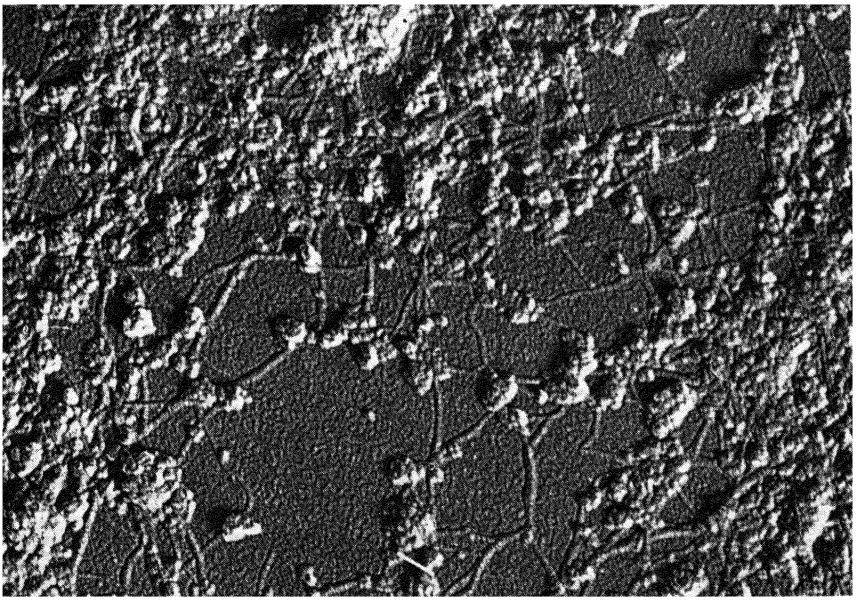


Figure 1. Date endosperm after Turmix and ultrasonic treatment before extraction of mannan .1 (magnification 45,000)

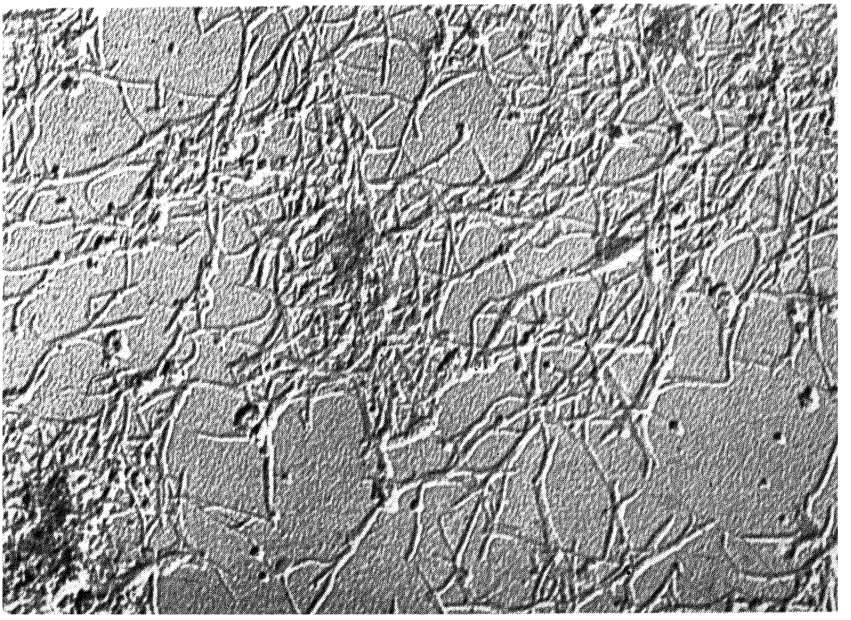


Figure 2. The same as in *Figure 1* but after extraction of mannan .1 (magnification 37,000)

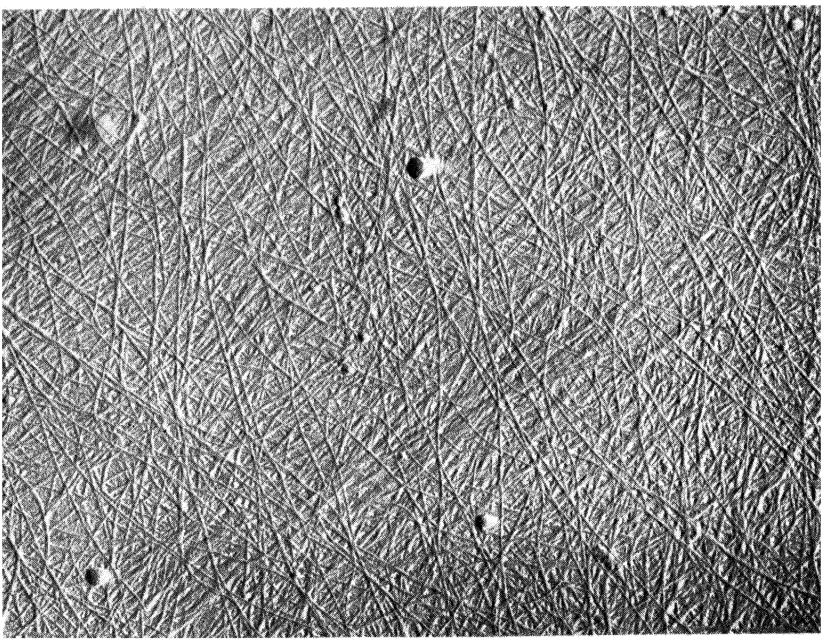


Figure 3 Part of a delignified primary wall (magnification 25,000)

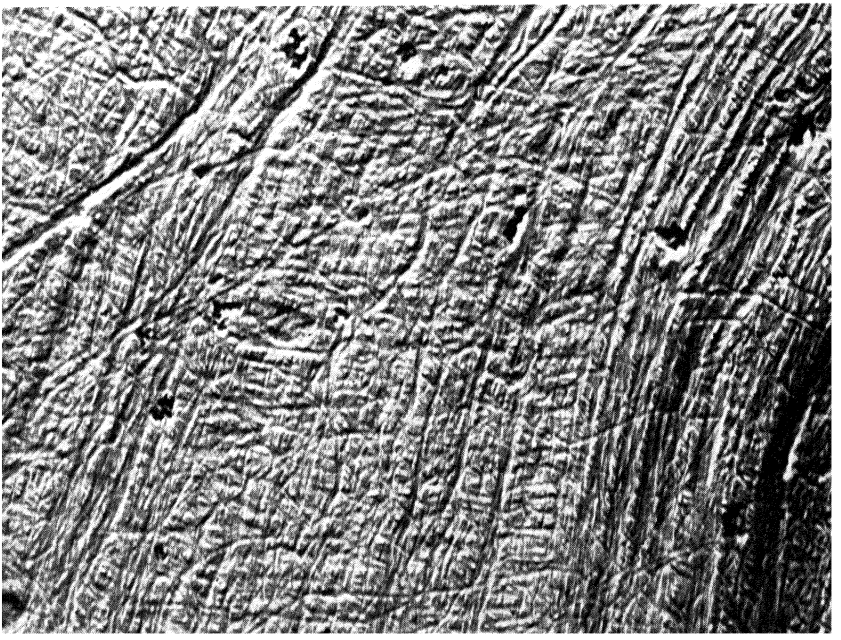


Figure 4. Part of a delignified S_1-layer (magnification 20,000)

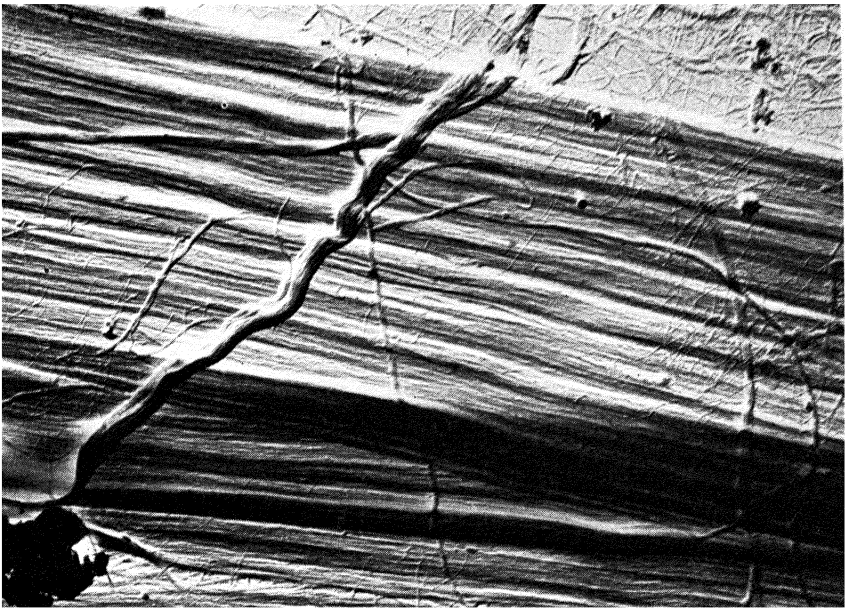


Figure 5. Part of a delignified S_2-layer (magnification 20,000)

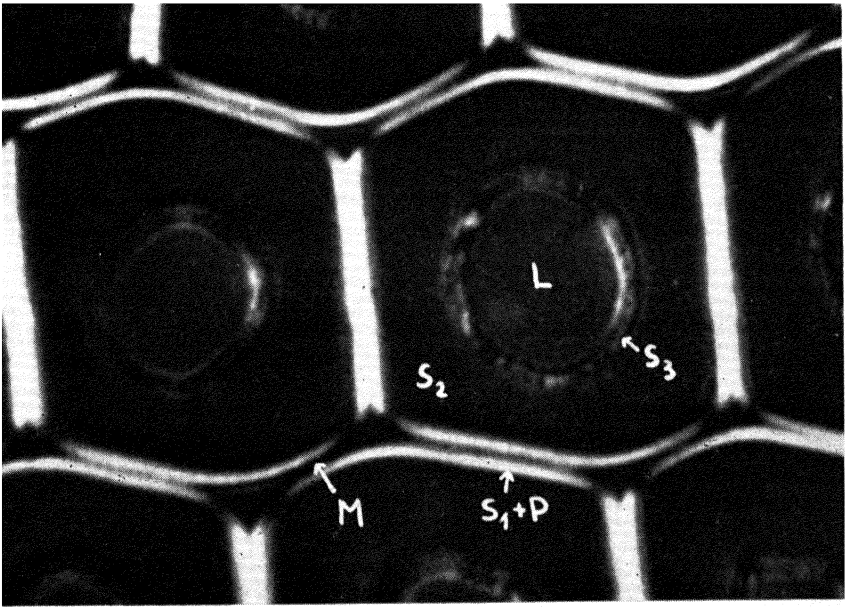


Figure 6. Transverse section through pine wood in polarized light: *L* lumen, *M* middle lamella, S_1 – outer layer of the secondary wall, S_2 – central layer of the secondary wall, S_3 tertiary wall (magnification 3,100)

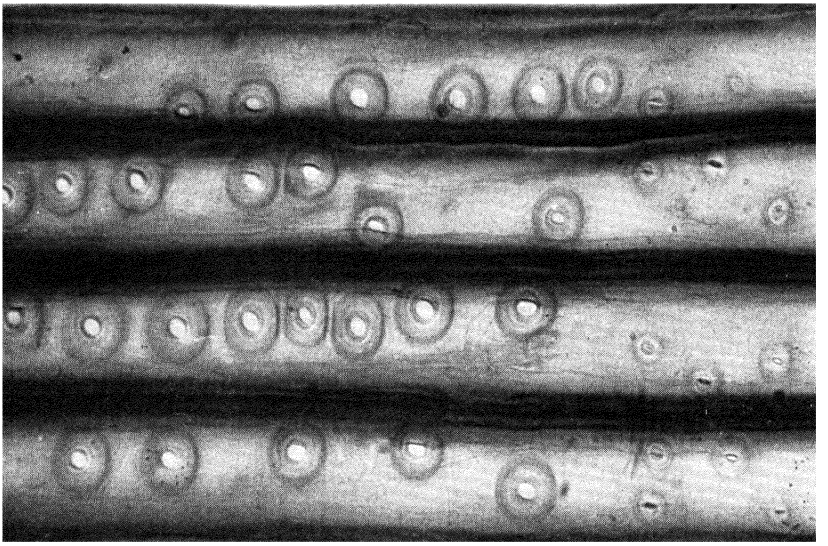


Figure 7 Pine tracheids with bordered pits (magnification 100)

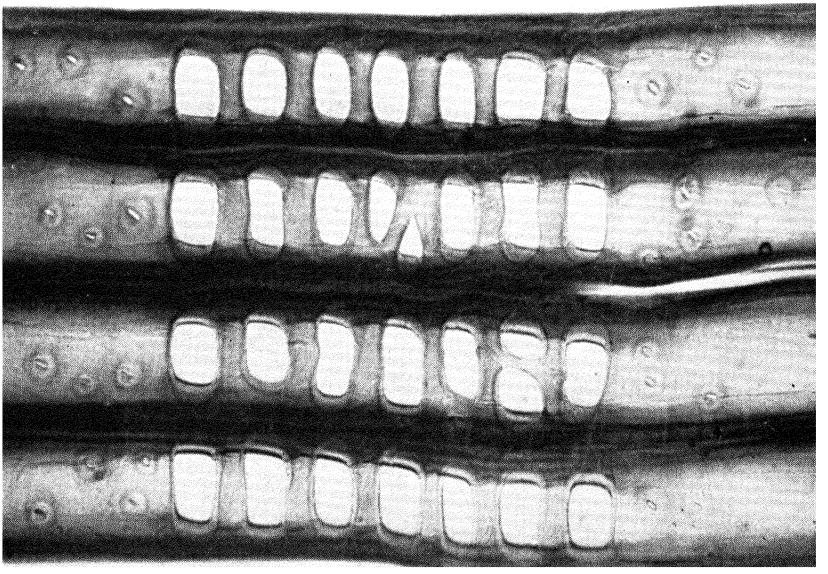


Figure 8 Pine tracheids with large window pits and small bordered pits (magnification 400)

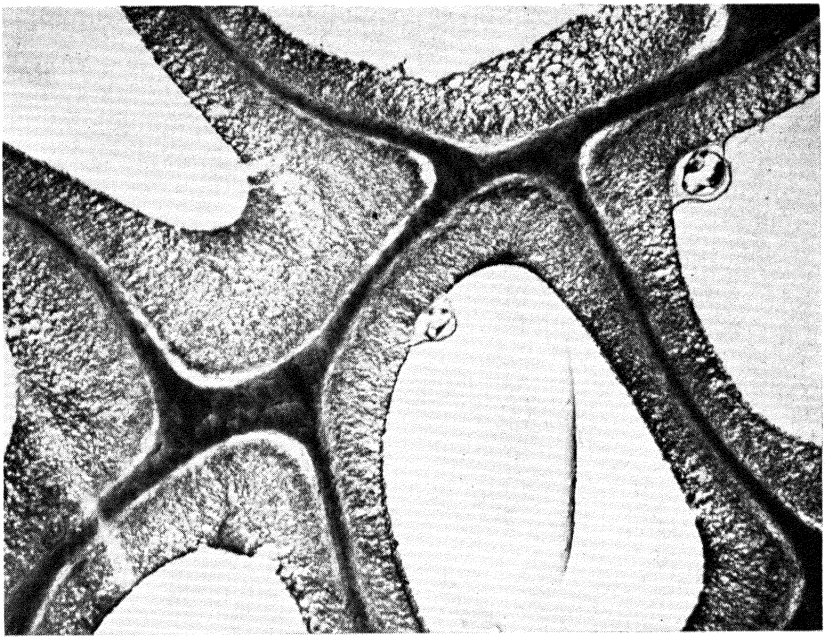


Figure 9. Transverse section through spruce wood, the cellulose has been dissolved to a great extent by a brown rot fungus, the lignin skeleton is left (magnification × 4,500)

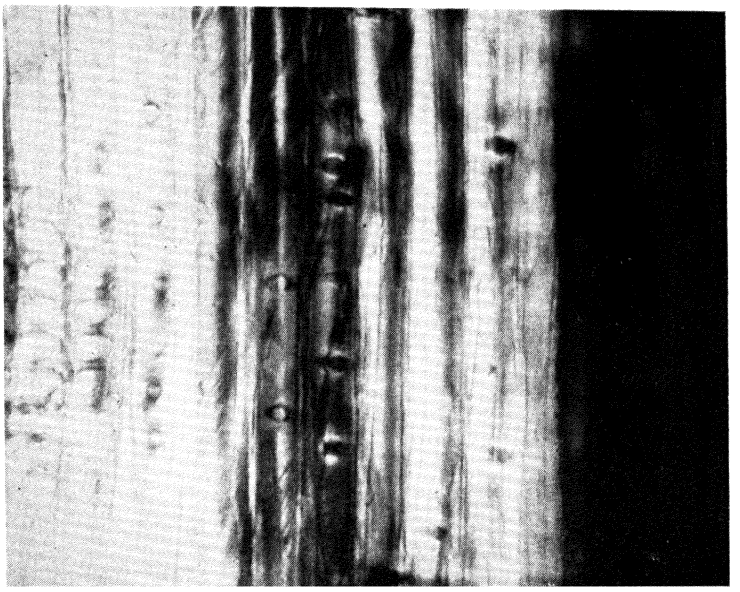


Figure 10. Radial section through the outermost annual ring of the xylem of pine in polarized light; tracheids in different stages of maturation exhibit different optical behaviour (magnification · 380)

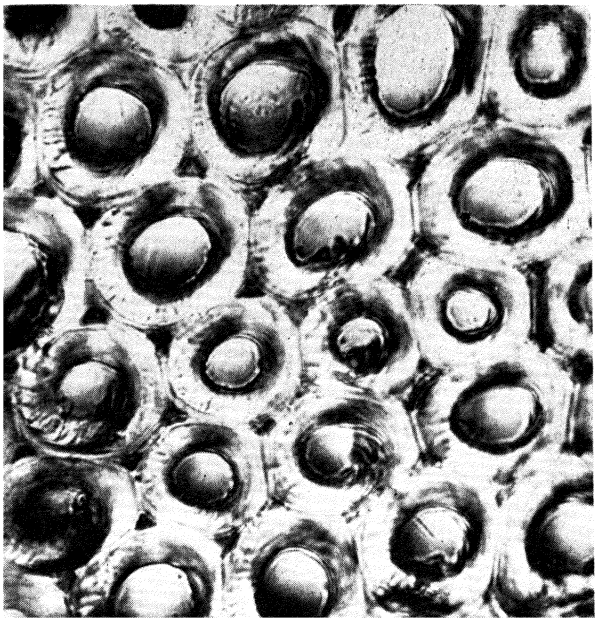


Figure 11 Transverse section through compression wood of pine (magnification × 700)

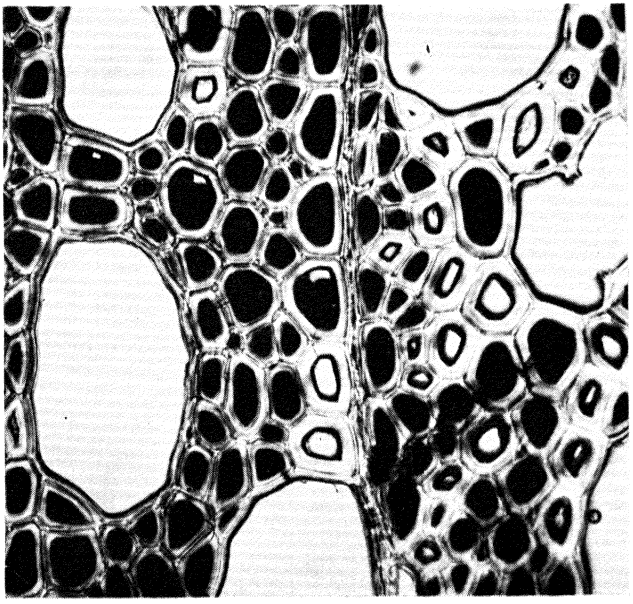


Figure 12. Transverse section through tension wood of aspen, the gelatinous inner layer of the cell walls is swollen and stained with chlor-zinc-iodide (magnification × 350)

one. *Figure 9* gives a rough idea of the lignin distribution in spruce tracheids. The precise location of the border of the heavily lignified middle lamella area and the less lignified secondary wall area is not quite clear. It might lie on the outside, within or on the inside of the S_1-layer.

The location of the *cellulose* and *hemicelluloses* in the fibre wall is more difficult than that of lignin. Kallmes[8] was able, by beating unbleached spruce sulphite fibres, to unravel the $(P + S_1)$-layers and to get two fractions, one being rich in $(P + S_1)$-fragments and the other being rich in $(S_2 + S_3)$-fragments. He found that both fractions differed appreciably from each other in their lignin content but were very similar with respect to their polysaccharide composition. This result, however, does not give much information on the polysaccharide distribution in a *native* fibre. Asunmaa and Lange[9] sought to obtain such information by a microspectrographic method. This, however, involved various drastic pre-treatments of the fibres and the results must, therefore, be interpreted with care. Furthermore, the method suffered from the drawback that it did not enable differentiation between the various types of hemicelluloses. The most reliable technique for studying their location would be the microdissection of single fibres into the single wall layers and the analysis of them after hydrolysis for their sugar composition. Bailey[10], some 25 years ago, isolated the middle lamella of the tracheids of Douglas fir in that way and estimated the lignin microchemically. However, up to now, no-one has succeeded in isolating each of the fibre cell wall layers completely. An alternative approach was, therefore, required and has been devised at the Swedish Forest Products Research Laboratory [11, 12]. During the aging of the young fibre, successive polysaccharide layers are deposited next to one another. Fibres have now been isolated and analysed at various stages of maturation. From a knowledge of the polysaccharides found to be present in these fibre fractions an attempt has been made to deduce the nature of the cell-wall layers in the mature fibre. This approach to the problem entails the assumption that, once a cell-wall layer has been deposited, its polysaccharide composition does not subsequently alter. This is probably true, as the polysaccharides, unlike lignin are deposited on the surface of the protoplast in successive layers.

The young fibres in radial sections through the outermost part of the wood xylem were separated into four fractions: $(M + P)$-fibres, $(M + P + S_1)$-fibres, $(M + P + S_1 + S_{2\ \text{outer part}})$ fibres and $(M + P + S_1 + S_2 + S_3)$-fibres. This separation was carried out under the polarizing microscope where the different layers exhibit different optical properties (*Figure 10*). Each fibre fraction was hydrolysed and the sugars were estimated quantitatively. The results were then used to calculate the polysaccharide composition in each of the four fractions.

It is thus possible to deduce very roughly the proportions of the individual polysaccharides in each cell-wall layer. Micrographs of transverse sections taken with the polarizing microscope and with the electron microscope gave a certain volume fraction for each layer in the fibre wall. The density is certainly not the same in all the different layers, but, as a first approximation, the volume percentages can probably be taken as weight percentages. It must, however, be emphasized that this estimate is very tentative and the

Table 1. Percentages of polysaccharides in the different layers of the fibre wall

Polysaccharide	*ML + P**	S_1	S_2 outer part	S_2 inner part + S_3
Birch				
Galactan	16·9	1 2	0·7	0 0
Cellulose	41·4	49 8	48·0	60·0
Glucomannan	3·1	2·8	2·1	5·1
Arabinan	13·4	1·9	1 5	0·0
Glucuronoxylan	25·2	44 1	47·7	35 1
Spruce				
Galactan	16·4	8 0	0·0	0·0
Cellulose	33·4	55·2	64 3	63·6
Glucomannan	7·9	18 1	24·4	23·7
Arabinan	29·3	1·1	0·8	0 0
Glucurono-arabinoxylan	13 0	17·6	10 7	12·7
Pine				
Galactan	20 1	5·2	1·6	3·2
Cellulose	35·5	61 5	66 5	47·5
Glucomannan	7·7	16 9	24·6	27·2
Arabinan	29 4	0·6	0 0	2·4
Glucurono-arabinoxylan	7·3	15·7	7 4	19·4

* Contains also a high percentage of pectic acid.

carbohydrate composition of the different layers as presented in *Table 1* can, therefore, show only the main trends.

For all three woods investigated, the cellulose content is lowest in the (*ML* + *P*)-layer which contains a high percentage of pectic material (galactan, arabinan and pectic acid). There is, however, a marked difference between birch and the two coniferous woods in the arabinan content of the (*ML* + *P*)-layer; in birch it is only half as high as in pine. For the xylan content it is the reverse. In *birch*, the cellulose content is highest in the inner part of the S_2-layer and in the S_3-layer. From the present investigation, however, no conclusion can be drawn about whether S_3 *and* the inner part of S_2 are equally rich in cellulose or whether the cellulose is enriched only in one of them. The S_1-layer and the outer part of the S_2-layer have a very high content of glucuronoxylan.

In spruce and pine the composition of the different cell wall layers is similar. There are some minor differences but these cannot be regarded as characteristic either of spruce or of pine, since they may be due to the use of spring wood tracheids in the case of spruce and of summer wood tracheids in the case of pine. In pine summer wood tracheids the outer part of the S_2-layer is richest in cellulose. The glucomannan content increases steadily from the outer parts of the cell walls to the inner parts. The glucurono-arabinoxylan content is very high in the S_3-layer. The same was found by Bucher[13], who made a controlled hydrolysis of the tertiary wall in spruce wood. There seems to be practically no arabinan in the secondary wall layers of spruce and pine. The galactan value in these layers may be due to the presence of a galacto-glucomannan.

The uneven distribution of the chemical compounds through the single fibre wall also reflects on the chemical differences in distinct anatomical elements in wood. This is clearly shown in spring wood and summer wood. Spring wood fibres and summer wood fibres differ mainly from each other by the much thicker S_2-layer in the latter. The lignin-rich middle lamella makes up a larger portion of the spring wood than of the summer wood, which, therefore, has a somewhat lower lignin content than the former. Another consequence, as is shown in *Table 2*, is that summer wood has a higher glucomannan and a lower glucuronarabinoxylan content than spring wood, where the glucomannan-rich S_2-layer makes up a lower percentage and the glucuronarabinoxylan-rich S_3-layer a higher percentage than in summer wood.

Table 2. Relative percentages of polysaccharides in spring wood and summer wood from pine (*P. Silvestris L.*)

Polysaccharide	*Spring wood*	*Summer wood*
Galactan	3·4	3·1
Cellulose	56·7	56·2
Glucomannan	20 3	24·8
Arabinan	1·0	1·8
Glucurono-arabinoxylan	18·6	14·1

Besides the vertical tracheids and fibres, the ray cells are an important anatomical entity in both softwoods and hardwoods. Their volume percentage is a few per cent in softwoods rising to some 30 per cent in certain hardwoods. Perilä and co-workers[14] have studied the chemical composition of ray cells from spruce, pine and birch. In spruce and pine they found that the ray cells contained about the same amount of total hemicellulose as the vertical tracheids, but the total hydrolysate of the tracheids was found to be richer in mannose and poorer in xylose than the hydrolysate of the ray cells. The ray cells from birch had an entirely different composition from the fibres and the vessels. Whereas the glucuronoxylan content in the latter is about 2/3 of the cellulose content, the ray cells contain almost four times as much glucuronoxylan as cellulose.

Most stems, hardwood as well as softwood, contain a smaller or larger amount of reaction wood, *i.e.* wood which has grown under tension or compression. In leaning stems or in stems which grow on places which are very much exposed to wind, the percentage of reaction wood can be rather high. It has been known for some time that reaction wood fibres differ morphologically as well as chemically from normal wood fibres. *Figure 11* shows a transverse section through compression wood from pine. This is characterized by rounded tracheids with well-developed intercellular spaces and with fissures in the cell wall. The lignin content in compression wood from pine is as high as 40 per cent. The carbohydrate composition is shown in *Table 3*. The very high galactose content which was found to originate from a β-1,4-linked galactan with some galacturonic acid residues linked to it[15] is most characteristic.

Table 3. The carbohydrate composition of normal wood and of compression wood from spruce (*Picea Abies Karst.*)

	Galactose	*Glucose*	*Mannose*	*Arabinose*	*Xylose*
Normal wood	2·1	70·7	17 2	1·3	8·7
Compression wood	19 6	55·6	11·5	2·6	10·7

The tension wood fibres in hardwoods are characterized by a gelatinous cell wall layer bordering the lumen (*Figure 12*). This layer seems to consist almost exclusively of cellulose and is obviously responsible for the higher cellulose, the lower lignin and the lower glucuronoxylan content of tension wood, as is seen in *Table 4.*

Table 4. The carbohydrate composition of normal wood and of tension wood from beech (*Fagus Silvatica L.*)

	Galactose	*Glucose*	*Mannose*	*Arabinose*	*Xylose*
Normal wood	1·6	57·4	4·9	1·0	35·1
Tension wood	6·6	73·5	- -	2·6	17·3

Our knowledge of the physical and chemical fine structure of wood has greatly increased in recent times. The biological processes which produce these structures are, however, still rather obscure, and it appears that it will need considerable efforts to throw some light on this side of the subject. But we may hope that the research, which up to now has been directed very much to the investigation of the mature, dead wood fibres, will, in the future, be directed more and more to the cambial region of the stem, *i.e.* to that region where things happen.

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THE CHEMICAL STRUCTURE OF THE HEMICELLULOSES

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The cellulose fibrils in the walls of plant cells which have undergone lignification are embedded in an amorphous mass of material consisting mainly of a mixture of lignin and various polymeric carbohydrates. For want of a better name it is at present convenient to refer to these polysaccharides as hemicelluloses, but when detailed knowledge of their chemical structures becomes available it will be practicable to refer to them by more appropriate chemical names. To some extent that possibility has now been reached and, as the result of intensive investigations carried out in many laboratories over the past few years, it has become clear that the greater portion of the hemicellulose group of substances consists of materials belonging to a small number of distinct and clearly recognizable families of polysaccharides. These include the large and very varied families of the xylans, the glucomannans and the arabogalactans, with the possibility that other family groups such as the β-glucans of barley and oats, some mannans and some galactomannans may also be regarded as hemicelluloses. In some instances, the distinctions between hemicelluloses on the one hand and plant gums and mucilages on the other have become vague and indefinite and it will be necessary to restrict the present discussion mainly to the typical hemicelluloses of wood. Considerable progress has been made in the determination of the main structural features of these materials, most of which appear to be present in the cell wall as mixtures of polysaccharides, the individual molecules of which have a similar general structure but differ amongst themselves in their degree of polymerization and in a wide variety of minor features, such as the nature, number and position of the side chains attached to the main chain of the molecular structure. This recognition of the occurrence of many plant polysaccharides as families of closely related substances, rather than as a collection of exactly identical molecules, has had an important bearing on the development of ideas in polysaccharide chemistry, and in many ways, therefore, the present time is appropriate for a review of structure in the hemicellulose group. The present account must, however, be restricted to general conclusions and to indications of gaps in our knowledge. Fuller accounts of the experimental evidence with detailed references are already available in recently published articles[1].

Interest in these materials is of long standing. Xylan or Wood Gum was described by Thomsen and by Wheeler and Tollens and used as a source of the xylose which it yielded on hydrolysis[2]. A similar material can be obtained from esparto grass and, in view of the convenience of this source,

much of the early work centred on the xylan obtained from esparto by alkaline extraction. Application of the methylation method revealed that the main structural feature is a chain of (1——→4)-linked β-D-xylopyranose units[3] (*Figure 1*), a conclusion which was substantiated later by the isolation of a series of oligosaccharides consisting of 1— →4-linked xylose residues[4].

In subsequent investigations, attempts were made to determine the chain-length by the end-group procedure, but difficulties were at once encountered in that two kinds of non-reducing terminal groups were found to be present, namely D-xylopyranose and L-arabofuranose. The amount of the L-arabofuranose end group varied from fraction to fraction as the crude xylan was purified and fractionated by use of its copper complex.

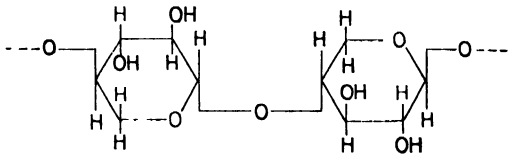


Figure 1. Main chain in xylan

One of the main points at issue at this stage was the question whether there were present two distinct polysaccharides—a true xylan in admixture with a true araban—or a single polysaccharide in which both xylose and arabinose residues were united in one molecular structure. That true xylans did exist was proved by examination of arabinose-free material obtained by exhaustive fractionation of the xylan from esparto. The main structural features of this are indicated in *Figure 2* which depicts a main chain of (1——→4)-linked β-D-xylopyranose units, probably with one branch chain of similarly combined xylose residues, the molecular size being of the order of 80–100 xylose residues.

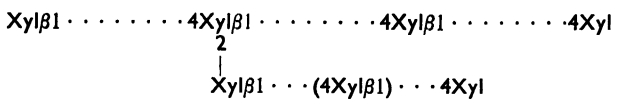


Figure 2

On the other hand, fractions containing arabinose were readily obtainable from esparto and from other sources, and Perlin[5] isolated from wheat flour a xylan fraction which contained almost as much arabinose as xylose. A study of this by the methylation procedure and by other methods clearly showed that single L-arabofuranose residues were attached to a main chain of xylose residues, in many instances substituting the hydroxyls at both C–2 and C–3 of the same xylose residue. These arabofuranose residues were readily removed by gentle hydrolysis with acid, leaving a xylan similar to the arabinose-free material from esparto mentioned above. The general molecular structure of wheat flour xylan is, therefore, the one shown in *Figure 3* and, by analogy, the arabinose-containing fractions from esparto may be expected to have a generally similar structure but with a smaller proportion of arabinose residues.

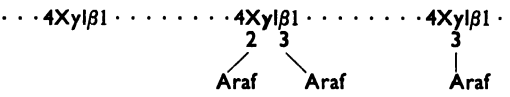


Figure 3

Direct proof of the attachment of L-arabofuranose residues to a xylose unit in the main chain has been obtained in two different ways. In one instance, a wheat straw xylan was subjected to partial hydrolysis by means of a cellulolytic enzyme present in the mould *Myrothecium verrucaria*[6]. This gave rise to a series of oligosaccharides containing both xylose and arabinose residues, one of these being the trisaccharide *O*-L-α-arabofuranosyl-(1——→3)-*O*-β-D-xylopyranoysl-(1——→4)-D-xylopyranose (see *Figure 4*). The mode of action of the enzyme on the original xylan is indicated in the same figure.

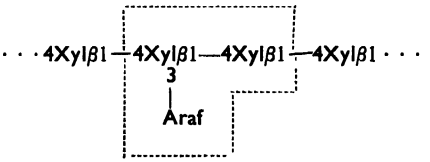


Figure 4

Another method, developed by Aspinall[7] and used in the course of work on rye-flour xylan, is to oxidize selectively by a catalytic procedure the primary alcoholic groups of the terminal L-arabinose residues to carboxylic acid groups. The arabofuranosyl linkage then becomes comparatively resistant to hydrolysis and, after partial hydrolysis of the oxidized polysaccharide, an aldobiouronic acid (L-arabofuranosyl uronic acid)-(1——→3)-D-xylose was obtained (*Figure 5*) and proof was thereby provided of the mode of attachment of the arabinose residue to the xylan chain.

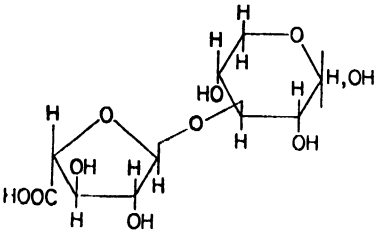


Figure 5

The arabinose residues in xylans are usually terminal units attached glycosidically to C-3 of a xylose residue. Frequently, however, non-terminal L-arabofuranose residues are encountered, as in the xylan of barley husks and in the complex group of hemicelluloses present in corn (maize) cobs, maize fibre and maize hulls. The xylan from barley husks may be taken as an example[8]. The structural evidence here clearly points to the presence of side chains containing a β-xylopyranose unit linked to C-2

of an L-arabofuranose residue (D-Xylβp1-L-Araf1 . . .) probably attached to the main chain as indicated in *Figure 6.*

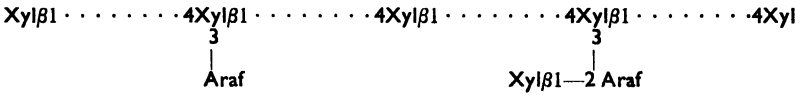


Figure 6. General features of structure of barley husk xylan

Other widely distributed units which are found to be attached directly to the main chain, usually but not invariably, *via* the C-2 of a xylose residue are D-glucuronic acid and its 4-*O*-methyl derivative (see *Figure 7*). These acidic residues were first recognized in a xylan from the pear (fruit). They occur in some wheat straw xylan fractions (where both 1–3 and 1–2 branch point linkages have been reported), although with wheat straw as also with wheat flour, barley flour, rye flour and esparto, fractions can be obtained which are devoid of acidic residues. In most of these instances it seems likely that both D-glucuronic acid and its 4-*O*-methyl ether are present in the xylan, whereas residues of 4-*O*-methyl D-glucuronic acid are typical of the xylans present in both soft and hard woods, where the uronic acid contents may be as high as 20 per cent and 15 per cent respectively. Normally, these residues are linked top osition 2 of a xylose residue in the interior of the main chain, but occasionally, as for example in the xylan from oak heart-wood, it is likely that a uronic acid residue of this type is present as the terminal unit of the chain.

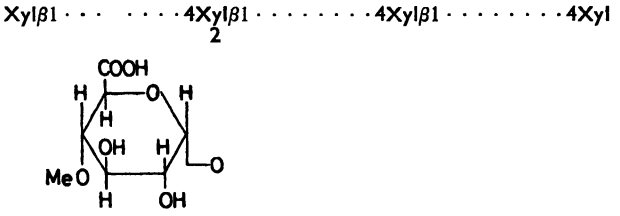


Figure 7

The question of the possible occurrence of branching in the main chain of xylose residues is still undecided in most instances, largely owing to difficulties in the accurate determination of molecular weights in the region (D.P. *ca* 50–150) in which most of the xylans occur. There is some evidence of a single branch point (see *Figure 2*) in the molecular structure of the arabinose-free esparto xylan mentioned previously. Furthermore, the xylan from Loblolly pine must contain a branched structure since there are three xylose end groups per molecule. Similar branched structures may occur in the xylans from North American beechwood, jute, milkweed floss and kapok[1].

It is evident from this summary that xylans from a wide variety of sources conform to the same general structural pattern characterized by a main chain, which may be branched, of β-1,4-linked xylopyranose residues, with side chains of L-arabofuranose, D-glucuronic acid (or its 4-*O*-methyl

derivative) and of L-arabofuranose carrying a xylopyranose residue attached at the C–2 position. From one xylan to another, frequently even in different fractions of a xylan from one plant source, the proportions in which these side chains are present may vary widely—an observation of special interest in connection with the mechanism of biogenesis of the xylans. In general, however, all these materials can be regarded as coming within one family group of polysaccharides, the main structural feature of which is the backbone of 1,4-linked xylose residues. This structural unit is a characteristic feature of the hemicelluloses of woody substances from both monocotyledonous and dicotyledonous plants, but it is clear that this mode of occurrence of xylose is by no means universal in the plant world. Xylans are prominent amongst the polysaccharide components of many algae and, in at least two cases which have been examined in some detail, β-xylopyranose residues linked through positions 1 and 3 play an important part in the structure. In the xylan from the red seaweed *Rhodymenia palmata*, both 1,3-and 1,4- links are present in the proportions of approximately 25 per cent and 75 per cent respectively (*Figure 8*)[9].

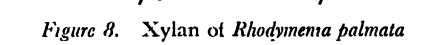


Figure 8. Xylan of *Rhodymenia palmata*

On the other hand, in the xylan present in the green alga *Caulerpa filiformis* the main chain is undoubtedly composed entirely of 1,3-linked β-xylose residues[10]. The principal structural features of this xylan, which may possess a branched xylose backbone, are indicated in *Figure 9*.

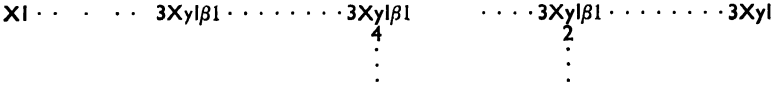


Figure 9 Xylan of *Caulerpa filiformis*

The question may be raised as to whether these algal xylans are to be regarded as hemicelluloses in the sense in which the word is usually employed, and structural comparisons may, therefore, be inappropriate. Nevertheless, they may well be constituents of the cell wall of the plants concerned and may be more nearly related to the hemicelluloses of the land plants than to the plant mucilages and wound gums which frequently contain xylose residues as components of extremely complex molecular structures. These, however, are outside the scope of the present discussion. It is probable also that the xylans which are found in the so-called "gum fractions" of the flour and grain of cereals have a biological function different from that of the wood hemicelluloses, although they show marked resemblances in their structural features, namely a backbone of β-1,4-linked xylose residues with side chains of arabofuranose units. Some of these may be highly complex polysaccharides resembling the true gums, with other sugar residues present also, *e.g.* galactose and mannose[11].

Many points of detail in the chemistry of the hemicelluloses still remain obscure. Reference has already been made to the need for more information concerning the molecular weight distribution of the various members

of the group, which appear to be, in general, both polymolecular and polydisperse despite their comparatively small D.P. values. This problem, at one time an extremely difficult one, would appear to be capable of resolution by the techniques now available. There are, however, many other still more formidable problems, one or two of which may be mentioned at this stage.

It will be evident from the descriptions and the formulae given above that no detailed information is available concerning the positions along the chain of the xylose residues which carry the side chains. The relative proportions in which these substituents are present can be determined, but, except in one or two special cases, *e.g.* Perlin's wheat flour arabinoxylan[12], neither the order in which they occur, nor their placing along the main chain has been ascertained. One of the difficulties here stems from the extreme lability of the furanoside linkage which joins the arabinose residues to the main chain. The usefulness of the method of linkage analysis by partial hydrolysis of the polysaccharide is thereby much restricted. Attempts to overcome this are now being made by taking advantage of the fact that the glycosidic link in uronic acids is much stronger than that of the corresponding sugar. If, therefore, the arabinose residues can be oxidized at C–5 giving a —COOH group in place of the original $-CH_2OH$ group (*Figure 5*), the rate of removal of the arabinose residue is greatly diminished and it may be feasible to make use of the method of partial hydrolysis to gain knowledge of the distribution of side chains in the molecule (see above). At present it is not known whether the side chains (Aral—, GpAl—, 4Me-GpAl—, Xyll——→2Aral) are attached randomwise to the main chains or are present in ordered sequences.

Another problem is raised by the apparent stability of xylans to alkali. Many of them, indeed, are isolated by extraction with alkali under far from mild conditions. There is some evidence from molecular weight measurements that, in some cases, xylans which have been extracted without use of alkali undergo slow degradation when treated with aqueous alkali. The breakdown is of the type which would be expected if the first residue in the chain possessed a free reducing group (*Figure 10*).

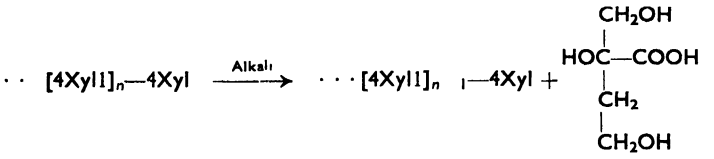


Figure 10

By contrast, xylans which have been acted upon by sodium borohydride to eliminate any free reducing group are stable to alkali. It seems likely that the peeling off reaction would be stopped by the presence of a 2-substituted xylose residue close to the end of the chain. On the other hand, when the main chain carries a substituent at C 3, the ready elimination of the side chain is not necessarily followed by rearrangement of the xylose residue to an alkali-stable saccharinic acid unit (*Figure 11*), but a second

elimination may occur, thus allowing the peeling reaction to continue as indicated in *Figure 10* [13].

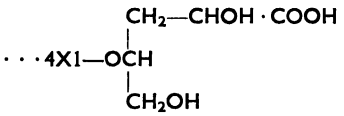


Figure 11

Another possible reason for the comparatively small reducing power of xylans as they are usually encountered may be that the chlorite treatment, often used for delignification during the preparation of the holocellulose, results in oxidation of the first residue to xylonic acid.

Mention of holocellulose raises yet another unsolved problem in hemicellulose chemistry, namely the mode of occurrence of the xylans in the cell wall itself. The marked difficulty with which many of these materials are separated from the other constituents has prompted suggestions, for which there is some supporting evidence, that in the cell wall the xylans are chemically combined in much larger macromolecular structures, perhaps as esters of their glucuronic acid residues with other carbohydrates and perhaps also through ether links to lignin. An indication of the complexity of the problem is revealed in Timell's work on the effect of the normal delignification procedures in reducing the molecular size of the xylan components of the cell wall[13a].

The vigorous methods used in the past for the isolation of hemicelluloses would inevitably result in the hydrolysis of ester groups attached to the polysaccharides. That such groups, particularly *O*-acetyl, may be present has been revealed by the use of mild reagents such as dimethyl sulphoxide for the extraction of glucuronoxylans from holocellulose preparations[14]. In this way, xylan fractions containing up to 17 per cent of acetyl groups have been isolated. It has been reported that some glucomannans from coniferous woods also contain acetyl groups and the question arises as to whether the presence of acetyl groups in natural xylans is general. Attempts have been made to determine the location of these acetyl groups by the methylation procedure using methyl iodide and silver oxide in dimethyl formamide. Difficulties are encountered owing to de-acetylation during the reaction and to the possibility of migration of the acetyl groups in the presence of silver oxide. In more recent studies[15], the free hydroxyl groups were protected by formation of the phenylcarbamoyl derivative (use of phenyl isocyanate), the acetyl groups removed and the product methylated. Control experiments indicated that, under the conditions employed, migration of phenylcarbamoyl groups did not occur, and the protecting group could be removed readily by reductive fission. Some idea of the distribution of the acetyl groups in a birch wood xylan is given by the following figures. The xylan contained some 13 per cent of acetyl apportioned in such a way that 58 per cent of the xylose residues were free from acetyl groups, 24 per cent were acetylated at C–3, 12 per cent at C–2, whilst 6 per cent had acetyl groups on both C–2 and C–3. The uronic acid residues were apparently not acetylated.

The materials considered up to this point have all been xylans in respect of their main structure. Xylose residues occur also in the much more complicated polysaccharides present in corn cobs, maize fibre, maize hulls, and in other cereal brans. Amongst these, a wide variety of sugar residues is encountered, these being present in all probability in a complex mixture of individual polysaccharides, some of which closely resemble the plant gums and mucilages in their properties. There are indeed indications that polysaccharides of these types are widely distributed in nature, and their inclusion in the hemicellulose fraction may be due in part at least to the difficulty of separating cell wall materials from those present in the cell sap.

As an example, brief reference may be made to the hemicellulose of maize fibre[16]. This does contain material similar in its main structure to the glucurono-xylans mentioned above, but whether the remaining residues are present exclusively as side chains, or partly in this way and partly as independent polysaccharides, is not known with certainty. This material is unusual amongst land plants in containing residues of both L- and D-galactose which are here present solely as end groups. Application of the methods of methylation and linkage analysis has revealed the presence of the following residues as component parts of larger molecules. The products containing reducing arabinose residues probably arose by fission of furanosyl linkages between the side chain and a xylan backbone. D-Gal1-; L-Gal1-; α-D-GpA-(1–2)-D-Xylp1-; α-D-GpA-(1–4)-D-Xylp1-; α-D-GpA-(1–4)-β-D-Xylp-(1–4)-D-Xylp1-; α-D-Xylp-(1–3)-L-Ara1-; L-Galp-(1–4)-D-Xylp-(1–2)-L-Ara1-; β-D-Galp-(1–4)-D-Xylp1-; and β-D-Galp-(1–5)-L-Araf1-.

The presence of galactose and arabinose in maize fibre hemicellulose indicates a resemblance between this material and the plant mucilages. A still closer relationship is evident in materials found in many conifers in association with polysaccharides of the xylan type. In illustration of this reference may be made to recent work on the hemicelluloses of *Pinus pinaster*[17] and to the group of galactoarabans typified by the ε-galactan of the European larch[18]. From *Pinus pinaster* on partial hydrolysis followed by examination of the aldobiouronic acids so liberated, *O*-(α-D-glucosyluronic acid)-(1–2)-D-xylose and *O*-(4-*O*-methyl-α-D-glucosyluronic acid)-(1–2)-D-xylose were obtained, characteristic of the glucuronoxylans of woody materials, but, in addition, several other acidic fragments were also isolated. These included the aldobiouronic acid *O*-(α-D-galactosyluronic acid)-(1–4)-D-xylose, which appears to be present in many woods, both of angiosperms and gymnosperms, but is absent from cereal straws and from chlorite holocelluloses prepared from wood. It was accompanied by *O*-(β-D-glucosyl-uronic acid)-(1–6)-D-galactose, a trisaccharide containing the residue β-D-GpA-(1–6)-D-Galp1 . . ., and the aldobiouronic acid *O*-(α-D-galactosyluronic acid)-2-L-rhamnose. All of these are characteristic of gums and mucilages rather than of cell wall materials and may not arise from the hemicellulosic portion of the wood. In this connection it may be noted that maple sap[19], as distinct from maple wood, has been found to contain polysaccharides composed of residues of galactose, arabinose and rhamnose (see below), whilst the combination of galactose, galacturonic acid and rhamnose occurs in some pectic materials[20] and also in various mucilages (linseed, *Ulmus fulva*, *Plantago ovata*, Cholla gum, Okra mucilage *etc.*).

As indicated above, angiosperm wood xylans contain only small proportions, if any, of arabinose residues. The situation is different however with the gymnosperms. In the xylan of the European larch, for example, arabinose residues are definitely present, and this may be true also of the Norway Spruce[21]. The mode of attachment of the arabinose units to C–3 of the xylose residues is normal, but in the case of the larch xylan it is not yet certain whether these arabinose units are directly linked to the main chain or whether to side chains of 1,4-linked xylose residues. Larch wood differs again from the hard woods in its composition in that it contains considerable proportions of an arabinogalactan or perhaps a mixture of such polysaccharides. Early work showed that fractionation of the crude ϵ-galactan from European larch could be effected and more recent work has definitely revealed the heterogeneity of some arabino galactans from larch. On the other hand, some samples of ϵ-galactan fail to reveal evidence of heterogeneity when examined in the ultracentrifuge. Structural examination of this apparently homogeneous material revealed in the first place that, on mild hydrolysis, the disaccharide 3-*O*-β-L-arabinopyranosyl-L-arabinose could be isolated. This disaccharide residue (L-Arapβ1-3-Ara**f** . . .) occurs also in gum arabic and in other plant gums. Further examination by the methylation procedure showed that the arabinose residues in the polysaccharide could be accounted for almost completely as disaccharide units, which are linked as such to a main structure consisting of D-galactose residues. The results of linkage analysis and methylation experiments show that this galactose framework is highly branched and consists of D-galactose residues linked together by 1,3- and 1,6-linkages. Further experiments, including the application of F. Smith's reductive procedure to periodate oxidized ϵ-galactan, and linkage analysis investigations following catalytic oxidation of unsubstituted —CH_2OH groups in the galactan. The sum total of the evidence so obtained favours a structure for ϵ-galactan depicted in *Figure 12* in which the side chains R^1 and R^2 are L-Arap1-3L-Araf1 and D-Galp1-6D-Galp1- respectively, in proportions such that 1 in 12 of the D-galactose residues of the ϵ-galactan carried an arabinose side chain. This structure consists of a main chain containing 1,3-linked galactose residues with branches of 1,6-linked D-galactose residues united to the main chain at C–6 positions, and it now appears that at any rate the majority of the arabinose side chains are attached directly to the main chain and not to the side chains of galactose residues[22].

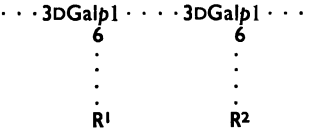


Figure 12

Studies on other arabinogalactans carried out in Canada[23] and in Sweden[24] reveal structural features similar to but not identical with those indicated for larch ϵ-galactan. There is strong reason, therefore, for the view that the arabinogalactans form a family of closely related polysaccharides possessing a highly branched structure of D-galactose residues joined by 1,3- and

1,6-linkages, to which are attached, in varying proportions, residues of L-arabopyranose and L-arabofuranose, some of the arabinose units being in the form -3L-Araf1-3L-Araf1- and some in the disaccharide unit previously mentioned. In Western larch arabinogalactan--and possibly in others it is probable that some of the galactose residues are linked through the 1,4- positions in addition to 1,3- and 1,6-linkages[23] and it is clear that much further work will be required before unique molecular structures can be assigned. There is an obvious analogy with the structure of the main chain in gums of the Acacia type (chains of 1,3-linked galactose residues with branches of galactose side chains linked through the 1,6-positions). A backbone of 1,3-linked D-galactose residues with side chains of L-arabofuranose and L-rhamnopyranose units occurs also in the arabinogalactan present in maple sap[19]. It is of interest that up to the present no indication has been obtained of the presence in hemicellulose fractions of separate arabans of the kind often found in pectic materials. It must be remembered, however, that difficulties of separating the mixed polysaccharides are great and that it may well be the case that some of the materials studied come from the cell sap as well as from the cell wall. The complexity of the situation may be further illustrated by reference to a galactan present in large amounts in spruce compression wood, which shows a high degree of lignification. Although full details of structure are not yet available it is already known that part of the galactan consists of a chain of β-1,4-linked D-galactose residues, and is similar in structure to the galactan which is normally present in pectic materials (*Figure 13*)[26].

Galβ1 · · · · 4Galβ1 · · · 4Gal

Figure 13. Galactan in spruce compression wood

The similarity extends even further in that D-galacturonic acid residues are present, probably in the form of a polygalacturonic acid of the pectic acid type. It is possible, therefore, that on occasions pectic materials may constitute a considerable proportion of the carbohydrate materials in a wood cell wall.

It is clear that wood hemicelluloses, particularly those from soft woods, contain a large variety of polysaccharides but, in view of their importance, reference must be made to yet another group, namely the glucomannans, or galactoglucomannans, which are present in considerable amounts in coniferous woods, to a lesser extent in some hard woods, and are found also in the seeds of many land plants. True mannans are also known, for example, the mannans present in ivory nuts, the molecular structure of which contains chains of (1–4)-linked β-D-mannopyranose residues[27]. A true mannan of similar structure is present in the marine alga *Porphyra umbilicalis*[28], but claims that mannans containing no other type of sugar residue can be isolated from spruce sulphite pulp and spruce holocellulose require confirmation. On the other hand, there is no doubt that the glucomannans which comprise up to half the hemicellulose content of some coniferous woods contain units of both mannose and glucose in their molecular structure[29,30] and this is true also of the Iles mannan of some species of *Amorphophallus*[29], and of the glucomannans from lily bulbs and iris seeds[30].

Separation of the glucomannan portion of wood hemicellulose from other components is by no means easy. The glucomannans are on the whole less readily extracted than the xylans, and in isolating them advantage may be taken of this fact and of their ready precipitation by barium hydroxide[31]. Purification may also be effected by precipitation of their copper complexes and fractionation of their acetates. Glucomannans from coniferous woods give, on hydrolysis, D-glucose and D-mannose in proportions ranging between 1:3 and 1:4, and some samples give rise also to small amounts of D-galactose. The galactopyranose residues are present as terminal units and it is not yet possible to say whether the galactose is part of a glucomannan polysaccharide or whether it occurs in a separate galactomannan of the type already known in guaran[32], which consists of chains of 1,4-linked β-mannose residues with galactose side chains consisting of single galactose residues linked to C–6 positions of the mannose chain (*Figure 14*).

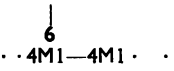


Figure 14. Guaran

Considerable attention has been given recently to structural investigations in the glucomannan of woody materials. Materials studied include polysaccharides from Loblolly pine[33], Western red cedar[34], Norwegian spruce[35], Sitka spruce[36], European larch[37], Red maple[38], Western Hemlock[39], Scots pine[40], Jack pine[41], White birch[42], Eastern white pine[43], and Aspen[44].

In some instances, materials free from galactose have been isolated and examined[45], whilst in others both galactose-containing and galactose-free fractions have been obtained from the same holocellulose. The glucomannans so far examined have revealed a striking similarity in molecular structure in that they consist essentially of linear molecules composed of residues of D-glucose and D-mannose linked through their 1,4-positions. On linkage analysis they give rise to the disaccharides 4-*O*-β-D-mannopyranosyl-D-mannose (Mβ1–4M), 4-*O*-β-D-glucopyranosyl-D-mannose (Glucβ1–4M), cellobiose (Glucβ1–4Glu) and 4-*O*-β-D-mannopyranosyl-D-glucose (Mβ1–4Glu). It is clear, therefore, that the molecular structure involves both glucose and mannose residues and there can be no question of a mixture of a mannan and a glucan. Oligosaccharides have also been identified after partial hydrolysis. These include several trisaccharides *e.g.* the substances Mβ1–4Mβ1–4M and Glucβ1–4Glucβ1–4M. From Norwegian Spruce no less than five of the eight theoretically possible trisaccharides containing mannose and glucose were isolated. Some caution in interpretation of the nature of the glycosidic links is necessary in view of the remarkable anomerization which mannosyl linkages can undergo in the presence of acids. The evidence as it stands, based partly on linkage analysis, partly on methylation and partly on periodate oxidation followed by degradation of the oxidized polysaccharide by F. Smith's method (borohydride reduction followed by hydrolysis), is in favour of the general structure for these glucomannans shown in *Figure 15*. In this structure

there is a random arrangement of the mannose and glucose residues, which may occur either as single units or as blocks of two, three or more of the same

$$\cdots\cdot 4Glu\beta 1\cdot\cdot\cdot\cdot 4M\beta 1\cdot\cdot\cdot\cdot$$

Figure 15. General features of molecular structure of glucomannans

kind in succession. It is just possible that the glucomannan of Loblolly pine may be an exception in that the glucose residues may be present as single units. The glucomannan from *Amorphophallus sp.* (*Iles mannan*) has the same general structure and, although smaller proportions of mannose-containing polysaccharides occur in the hard woods, the available evidence indicates that in these also (*e.g.* red maple[38], aspen[44], white birch[42]) the same type of structure is present. Ferns also appear to contain glucomannans (or galactoglucomannans) of the same general type in their hemicellulose fractions[46]. The question of branching in the main chain of some of these polysaccharides is still under investigation and further information is needed concerning the part played by the galactose residues when they are present. The latter give tetramethyl-D-galactopyranose after methylation of the polysaccharide and hydrolysis and must, therefore, occur as end groups. In the case of Norwegian Spruce hemicellulose, partial hydrolysis of the galactoglucomannan gives rise to the disaccharide Galα1–6M and the trisaccharide Galα1–6Mβ1–4M, indicating a mode of linkage which is found in guaran, but the evidence at present available does not permit any decision to be made as to whether or not polysaccharides of the guaran type are present in these hemicelluloses in admixture with glucomannans or galactoglucomannans. There is, however, evidence obtained during a study of the enzymic hydrolysis of a Jack Pine glucomannan that xylose residues present in side chains are constituent parts of the molecule. In the general patterns of their structures there would seem to be a close similarity between the xylan family and the glucomannan family. Like other polysaccharides of the hemicellulose group the glucomannans have comparatively small molecular weights (D.P. in the region of 100 in those cases where measurements have been made).

The greater part of cell wall materials in wood, other than cellulose itself, are included in the family groups which have been referred to above, namely the xylans (including arabinoxylans and glucuronoxylans), the galactoarabans and the glucomannans. It is evident that other polysaccharides also are present in many woods to a greater or lesser extent, and with improved methods of extraction and fractionation it should soon be possible to characterize these and determine the general features of their structures. Even so, however, only a beginning will have been made in the chemical study of the hemicelluloses and it will still be necessary to deal with the detailed structure of these polysaccharides and the way in which variations in structural detail occur within the family groups of molecules present in a single cell wall. Knowledge of the organization of the hemicelluloses within the cell wall and of the nature of their association with the cellulose and lignin components is at present scanty in the extreme and enquiry is needed into the possibility that certain members of the hemicellulose group may occur both in the cell wall and in the cell sap. There

remains, therefore, despite the rapid progress made during the past few years, a long programme of work awaiting both chemists and biochemists before our knowledge of the structure, function and biosynthesis of the hemicelluloses could be regarded as adequate.

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RECENT ADVANCES IN METHODS OF ISOLATING AND PURIFYING HEMICELLULOSES

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It is one of the main objectives of wood chemists to study the chemical structure of the polymers cellulose, hemicellulose and lignin, the main wood constituents. Such studies must begin with the isolation and purification of these components. The isolation of natural products of low molecular weight is usually quite simple, provided there is no shortage of biological material and the percentage of the desired products is not too low. Often these products can be obtained crystalline and analytically pure. In the case of polymers, the situation is quite different. Drastic methods, which may be needed for the extraction of the product from the biological material, can result in degradation and chemical modification of the polymer. The fractionation and purification of extracted polymers is often difficult, and it is only recently that methods of more general application have been developed in this field. The present discussion will be concerned with some recent advances in the isolation and purification of the wood hemicelluloses.

A considerable part of the xylan in hardwoods may be extracted from the milled wood by alkali, while softwoods require a prior delignification before significant amounts of any polysaccharide can be extracted. Larch woods are exceptional among softwoods as the arabinogalactans, often constituting 10 per cent or more of the wood, may be extracted with cold water from the milled wood. The difference between hardwoods and softwoods in this respect is due to the higher lignification of the secondary wall in the softwood fibres, which renders the polysaccharides less accessible to extraction.

The delignification procedures usually employed are the chlorite method, first described by Jayme[1] and modified by Wise and co-workers[2], and the chlorine–ethanolamine method, devised by van Beckum and Ritter[3]. According to the former method, the wood is treated repeatedly with chlorite solutions of pH 4·7 at 60–80°. According to the latter, the wood is delignified by alternate treatments with chlorine in ice-water and ethanolamine in boiling ethanol. Both methods are oxidative, affecting not only the lignin, but also the polysaccharides to some extent. This results in a partial depolymerization (decreased molecular weight) of the cellulose and hemicelluloses, an increased number of carbonyl and carboxyl groups in these polysaccharides, and—when the degradation is severe—the presence of degraded cellulose in the hemicellulose fractions. Timell and Jahn[4] found that the degradation of polysaccharides caused during the chlorite method was more severe than that caused during the chlorine–ethanolamine method, these results being confirmed in a later investigation by Glaudemans

and Timell[5]. Softwoods, however, are more difficult to delignify by the chlorine–ethanolamine method, unless the number of treatments is high; hence softwoods are generally delignified with chlorite. Successful delignifications with chlorine–ethanolamine have, however, been reported. Occasionally it is not realized that a considerable part of the wood polysaccharides may be dissolved during the treatment with chlorite. Thus, on delignification of wood meal from spruce compression wood, more than 10 per cent of the wood polysaccharides were dissolved and could be recovered from the delignification liquors[6]. Most of this material was a

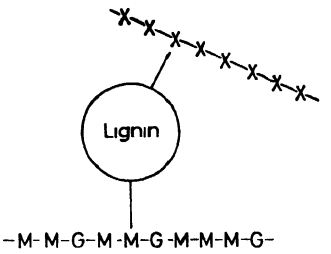


Figure 1

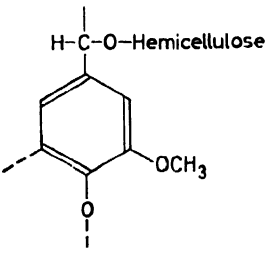


Figure 2

galactan, typical for compression wood. Xylans and glucomannans are also dissolved, however. There is an important difference between wood meal and intact wood fibres, the material being less accessible in the fibres and consequently extracted to a lower extent during the delignification.

The holocelluloses may contain 1–2 per cent lignin, most of which is dissolved, together with the hemicelluloses, during an alkaline extraction. We have observed that a fractionation of the hemicelluloses sometimes is difficult or impossible to achieve unless this lignin is removed by delignification of the hemicellulose in solution[7]. The results may be explained by assuming a lignin hemicellulose linkage, the existence of which is still a very live problem. If, for example, some xylan and glucomannan molecules are linked to the same lignin molecule, as indicated in *Figure 1*, a fractionation into pure components is of course impossible but may readily be achieved after complete delignification. Recent results of Freudenberg make the existence of lignin–hemicellulose linkages of the benzyl ether type probable (*Figure 2*). Methylenequinones are formed as intermediates in the biosynthesis of lignin and these react easily with any alcoholic groups present, including the hydroxyl groups of the hemicelluloses. (For references, see Freudenberg's contribution to this Symposium, p. 9.) Another possibility, that these linkages are glycosidic, appears less probable.

Another delignification method which appears to have some merit was devised by Poljak[8] and involves treatment with peracetic acid in aqueous sodium acetate at 70°. It has recently been investigated by Leopold[9] who found that few carboxyl groups but a great number of carbonyl groups were introduced into the holocellulose during this treatment; accordingly, he modified the method to involve a final treatment with borohydride. A holocellulose from pine wood prepared by this method was compared with a chlorite and a chlorine–ethanolamine holocellulose from the same wood.

The peracetic acid and chlorine–ethanolamine holocelluloses had somewhat similar properties and were considerably less degraded and modified than the chlorite holocellulose. It is, however, most probable that a great portion of the carbonyl groups introduced (0·2 mmole per g) are ketogroups, the reduction of which might give sugar residues which are artefacts and thus atypical for the native polysaccharides (*Figure 3*).

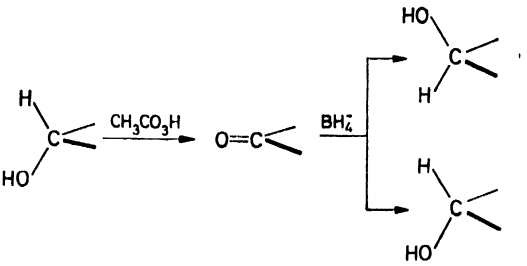


Figure 3

Hemicelluloses can be extracted from holocellulose by alkali but part of them, especially glucomannans, are exceedingly resistant to extraction. It was an important advance, therefore, when Jones and co-workers[10] devised a technique, by which these polysaccharides could be extracted. Borate forms strong complexes with cyclic α-*cis*-glycol groupings, such as are present in the mannose residues in a glucomannan (*Figure 4*), and by addition of borate to the alkali, these polysaccharides could be extracted as borate complexes. As most of the other hemicelluloses can be extracted by alkali alone, fairly pure glucomannans can thus be obtained. This method, of course, has a wide application and may be extended, not only to other biological materials and complexing agents, but, possibly, also to other types of natural polymers.

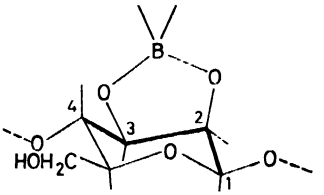


Figure 4

Even after exhaustive extraction of a holocellulose, it usually contains some percentage of hemicellulose components, which are probably inaccessible and therefore not extracted. By dissolving the whole sample, *e.g.* in copper ethylenediamine and regenerating it (thus destroying the biostructure completely), further amounts of hemicelluloses may be extracted by conventional methods.

The result of an extraction depends both on the type of base employed and its concentration; by a suitable extraction scheme, a considerable fractionation of the hemicelluloses can be achieved. These aspects are, however, considered by Kyle Ward in another paper (p. 77).

It has been known for a considerable time that wood contains *O*-acetyl groups. As such groups were also found in the derived holocellulose, it

was believed that they were linked to the hemicelluloses. When these are isolated by alkaline extraction, any ester groups present are simultanously saponified. Some years ago we demonstrated[11] that part of the hemicelluloses were extracted from the holocellulose in their native, acetylated form, by dimethylsulphoxide. From a birch holocellulose prepared by the chlorine–ethanolamine method, an almost pure xylan containing 17 per cent *O*-acetyl groups and representing about 50 per cent of the total xylan, could be isolated[12]. In a similar manner, a fraction of natively acetylated hemicelluloses could be prepared from softwood[11]. This fraction was more complex and its fractionation offered special problems which I shall discuss later. The acetylated hemicellulose components which could be isolated by this procedure, represent only a part of the total hemicelluloses. It may be assumed, however, that they are representative of the total hemicellulose fraction, and the *O*-acetyl content of the wood calculated on the basis of this assumption agrees fairly well with that actually found (*Table 1*).

Table 1. *O*-Acetyl content of some holocelluloses and isolated polysaccharides

Birch wood	Holocellulose	OAc 5·8%
	Total xylan —	35% of holocellulose
	Isolated acetylated xylan	OAc 17%
Calculated OAc content of holocellulose	17 · 0·35 = 6 0%	
Pine wood	Holocellulose	OAc 1·1%
	Total glucomannan =	20% of holocellulose
	Isolated acetylated glucomannan	OAc 6·0%
Calculated OAc content of holocellulose	6 · 0·2 — 1·2%	

The study of these products has not only theoretical interest, but has also added to our knowledge of the chemical reactions which occur during the pulping processes. The partially acetylated hemicelluloses, in analogy to the cellulose derivatives, have higher solubilities and a lower tendency to "crystallize", than the unsubstituted products, and their physical properties are, therefore, changed to a great extent when these groups are removed, as may occur during pulping reactions. Öhrn and Croon[13] have shown that a considerable part of the *O*-acetyl groups survive a normal, acid sulphite cook. In a two-stage sulphite cook of softwoods, an improved yield of pulp is obtained which is due to reduced dissolution of the glucomannans. This "glucomannan stabilization" has been studied by Annergren *et al.*[14] who have come to the conclusion that it is probably achieved because the *O*-acetyl groups are removed in the first, neutral or alkaline step, rendering the glucomannans less accessible to a hydrolytic attack during the second, acid step.

We have seen that it is sometimes possible to obtain reasonably pure hemicellulose fractions by extraction only. It is, however, more usual that the extracts consist of mixtures of polysaccharides, which must be subjected to fractionation prior to a structural investigation. It is often difficult to decide whether or not a polysaccharide fraction is homogeneous and even the definition of homogeneity offers some difficulty. A molecular weight distribution in a homogeneous fraction must be accepted. If the different

monomer residues, *e.g.* in a glucomannan, are arranged at random and not according to a regular pattern (which is an open question but does not seem improbable), the significance of the term *homogeneous* is further reduced. Polysaccharides of the same general type but with different proportions of monomer residues have been isolated from the same wood sample. One example is the glucomannans, or, more correctly, galactoglucomannans, from softwoods, various fractions of which may differ considerably in their galactose content—from almost nil to about 10 per cent or more. There are also indications that the acidic xylans can be divided into fractions with different percentages of acidic groups, and it may be that all possible structures between some extreme limits are represented.

A fractionation should preferably be followed by as many independent analytical methods as possible. Carbohydrate composition and determination of the optical rotation are generally the first analyses performed. Electrophoresis in borate and other buffer systems, either in cells, on columns or on glass fibre sheets[15] may often reveal inhomogeneity. The last method is especially convenient as the technique is simple and the equipment quite inexpensive. Other methods, such as ultra-centrifugation studies, ultra-filtration, use of molecular sieves, infra-red absorption studies and specific precipitations with antisera, may often give further valuable information.

Precipitating agents of some selectivity are often used to purify polysaccharides. Fehling's solution has been used for the precipitation of hardwood xylans and of mannose containing polysaccharides. The latter and also some galactans are precipitated almost quantitatively by barium hydroxide[16] and several useful separations in the field of wood hemicelluloses have been reported with this agent.

Meier[17] recently made a systematic investigation of the precipitation of some polysaccharides using metallic salts as complexing agents (*Table 2*).

Table 2. Precipitation of some polysaccharides as metal complexes

Precipitant / *Polysaccharide*	*Fehling's solution*	$Ba(OH)_2$ 0·03 M	$Ba(OH)_2$ 0·15 M	$Pb_2(OAc)_3OH$	$Pb(OAc)_4$
Galactomannan (guaran)	+	+	+	+	+
Glucomannan (softwood)	+	+	+	+	+
Galactan (compression wood)	—	(+)	+	+	+
Arabinoglucuronoxylan (softwood)	—	—	—	+	—
Glucuronoxylan (hardwood)	+	—	+	+	+
Acetylated glucuronoxylan (hardwood)	— → +	—	— → +	+	—
Arabinogalactan (larch)	—	—		—	-

He studied Fehling's solution, barium hydroxide at two concentrations, basic lead acetate and lead acetate, the latter being the only neutral precipitating agent. The galacto- and glucomannans were precipitated by all reagents. The galactan from compression wood, with 1,4-β-linked galactose residues and some uronic acid residues, was not precipitated with Fehling's solution (no *cis*-glycol groupings) and only partially with barium

hydroxide of low concentration. There are interesting differences between the xylans, an arabinoglucuronoxylan from softwood, glucuronoxylan from hardwood and the same polysaccharide, isolated under mild conditions when the *O*-acetyl groups are preserved. The deacetylated hardwood xylan is precipitated by all agents except the dilute barium hydroxide. The softwood xylan, in which the xylan chain is more heavily substituted, is precipitated only with basic lead acetate. The partially acetylated xylan is obviously deacetylated in the alkaline solutions because the precipitate is formed slowly and not immediately as with the other polysaccharides. The highly branched arabinogalactans are not precipitated with any of these agents.

One of the most important recent advances in this field is the introduction of quaternary ammonium salts, with a long alkyl chain, as precipitating agents, cetyltrimethylammonium bromide (I) or cetylpyridinium bromide (II) being the most commonly used.



(I) (II)

The method has been studied and applied by Stacey and co-workers[18] and by Scott[19]. The most extensive and systematic studies, chiefly on animal polysaccharides, have been conducted by Scott, who has also summarized the work in this area[20]. Negatively charged polyelectrolytes, *e.g.* acidic polysaccharides, combine with the base to give precipitates. The terminal ends of the long alkyl chains probably form micells (*Figure 5*), and it is significant that the longer the chains, the more efficient is the precipitating agent. It is essential therefore that the alkyl derivatives used as precipitating agents are pure and not mixtures of a homologous series, otherwise a fractionation not of the polymer but of the precipitating agent may result. Commercial products of high purity are now available. The precipitate may be dissolved by increasing the acidity, when the carboxyl ions are transformed into neutral carboxylic groups. Strongly acidic polymers (*e.g.* polysaccharide sulphate esters), those of medium acidity (carboxylic acids) and neutral polymers are easily separated, the former being precipitated also in strongly acid solutions and the latter not at all. The precipitates are dissolved when the ionic strength of the solution is increased, because of the competition between the metal ions and the organic base at the charge sites of the polyelectrolyte. Not only very different types of polysaccharides, but also closely related polysaccharides may be separated, either by gradual precipitation or by extraction of a precipitate with solutions of increasing ionic strength. The latter technique can be performed by column chromatography with gradient elution.

Another technique is to precipitate the polysaccharide mixture completely, dissolve the precipitate in a solution of sufficiently high ionic strength, and effect fractionated precipitation by simply diluting the solution. By using cetyl-pyridinium bromide as precipitating agent, the course of the precipitation can easily be followed by determining the ultraviolet adsorption of the supernatant liquid in a spectrophotometer. Finally,

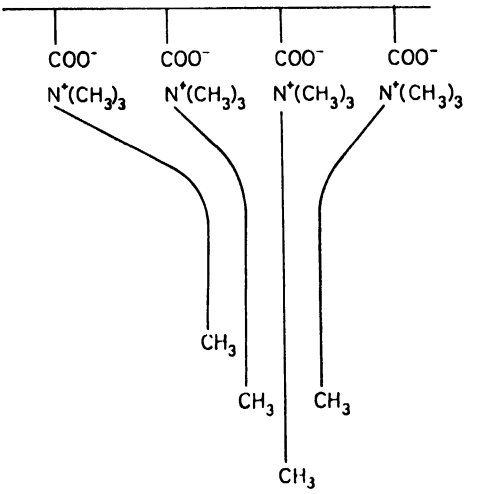


Figure 5

the precipitation of acidic polysaccharides has been developed into a method for the quantitative determination of acidic groups.

Table 3. Fractionation of 40 g (= 250 mmoles of sugar residues) arabinogalactan in aqueous 0·25 M boric acid (1000 ml)

CTA–OH* *added* (mmoles)	NaOH *added* (mmoles)	*Weight of fraction* (g)	*Components*
10	0·25	14·6	A
10	0·50	11·6	A + B
10	0·50	5·9	B
5	1	1·5	B
Residue		2·1	B

	% Arabinose	% Galactose	$[\alpha]_D^{20}$	M_w	M_t value (glass fibre sheet)
A	19	81	7°	100 000	0·76
B	21	79	10°	16 000	0·63

* Cetyltrimethylammonium hydroxide.

The method can be extended to neutral polysaccharides[18b] which give borate complexes. In order to keep the ionic strength as low as possible, it is advantageous to use boric acid and the free, quaternary base in these precipitations. We were able to separate the larch arabinogalactan into two components by this procedure[21] (*Table 3*). These fractions had similar compositions and optical rotations, but different molecular weights, immunochemical reactivities and electrophoretic mobilities in borate buffer. The old problem of the homogeneity of this wood polysaccharide has now been brought to a definite solution.

A further possibility for purifying a polysaccharide in a mixture is by selective enzymatic degradation of the other components. This technique has so far been of little value in wood chemistry because of the lack of highly pure enzymes. One example of this technique is the removal of starch from a mixture of polysaccharides from the inner bark of spruce[22].

Chromotographic fractionation of polysaccharide mixtures on different column materials such as aluminium oxide, carbon, and ion-exchange resins, has been attempted but has not proved very successful. The active surfaces of these highly porous materials are probably accessible to the polymers only to a limited extent (*Figure 6*). Furthermore, a molecular sieve effect is superimposed on the fractionation due to absorption and complicates the results. Better results may possibly be obtained by the use of pure molecular sieves which are now commercially available, such as dextrans, with a low degree of crosslinking, and which form highly swollen gels.

The introduction of a new type of ion-exchange material has changed this situation completely. When cellulose is oxidized with nitrogen dioxide, a material is obtained which is sometimes called celluronic acid or nitrogen dioxide oxocellulose. It contains a high percentage of glucuronic acid residues, and has been used for a considerable time in the pharmaceutical industry for the fractionation of biologically active proteins. Its superiority to other ion-exchange materials probably depends upon the fact that the

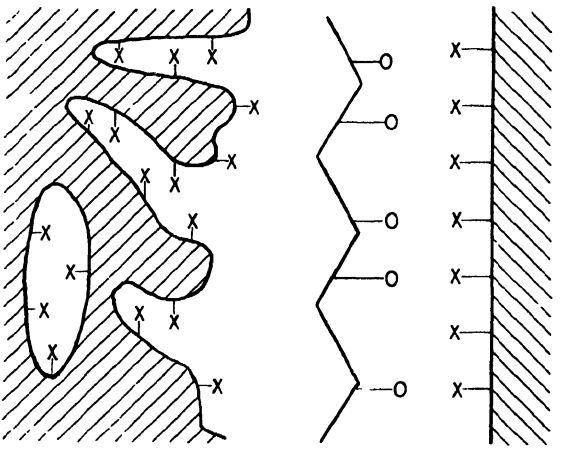


Figure 6

active groups, carboxyl functions, are situated on the surfaces of crystalline regions, and the polymers are more easily adsorbed on these surfaces than on those of the highly porous synthetic ion-exchange resins. Cellulose derivatives with all types of ionic substituents may be prepared, some are commercially available. Several successful fractionations of polysaccharides on these materials have been reported[23]. An example from our laboratory[17] is the polysaccharide mixture, referred to earlier, which was obtained on extraction of a softwood holocellulose with hot water after a dimethyl sulphoxide treatment. The mixture contained, beside the galactoglucomannan, some 15 per cent glucuronoarabinoxylan and had 4·5 per cent *O*-acetyl groups, which should have been removed by precipitation with Fehling's solution or barium hydroxide. The acidic xylan could be

adsorbed on a column of diethylaminoethylcellulose (III) in the phosphate form while the neutral galactoglucomannan was not adsorbed.

$$\text{Cellulose—OCH}_2\cdot\text{CH}_2\text{N}(\text{C}_2\text{H}_5)_2$$

(III)

It was found that this neutral polysaccharide contained all the *O*-acetyl groups. The xylan could be eluted only by alkaline solvents. By precipitation of another portion of the original mixture with cetyltrimethylammonium bromide, a xylan devoid of *O*-acetyl groups was prepared under conditions where such groups should be stable. Fractionation of the sensitive *O*-acetylated polysaccharides was thus enabled by these modern techniques. The result—that in softwoods only the glucomannan and not the xylan is acetylated—is somewhat unexpected as the hardwood xylans contain a high percentage of *O*-acetyl groups.

The only problem discussed in this address which is unique to hemicellulose chemistry is that of delignification. All the other problems are common to polysaccharide chemistry in general and in some respects also to the chemistry of other natural polymers. Consequently the recent advances discussed are products of a joint effort by chemists working in these fields. Hence hemicellulose chemistry does not stand alone, and advances in any area of polysaccharide chemistry should be closely followed by the wood chemist.

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FRACTIONAL EXTRACTION AND PROPERTIES OF HEMICELLULOSES

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After the excellent presentations which you have just read on the hemicelluloses, you may wonder with reason whether there is anything left to discuss. We should, therefore, like to state the basis, somewhat arbitrary at best, of the present review. The first limitation is the use of the restricted definition of hemicellulose, which excludes starch, pectins and water-extractable polysaccharides, except those freed for extraction by strong swelling agents, such as liquid ammonia or dimethyl sulphoxide. The second limitation is consideration only of experiments in which the hemicellulose mixtures were precipitated so that we know something of the yield, at least, of the product. While work on softwoods is emphasized, some pertinent papers dealing with other materials have been included.

The separation of polymers must, in its very nature, be a fractionation procedure and there are two main types of fractionation, precipitation and solution. In general, precipitation procedures are preferred, but there are some special reasons which make solution fractionation of particular interest to the chemist interested in hemicelluloses. Most important, perhaps, is the fact that separation of the hemicelluloses from the more inert and insoluble cellulosic component of the wood has to be done by solution anyway. Closely associated with this is the consideration that the avoidance of as many different manipulations in purifying as possible makes the material balance more accurate. If one is interested in determination of structure and high purity, this is less important, but if one is interested in the composition of the original wood, it becomes very important. Similarly, the simplification of purification procedures and elimination of steps makes it more likely that the hemicellulose has been isolated unchanged, a goal rarely if ever attained.

Extraction techniques are made especially difficult in this field by the nature and structure of wood. The morphology of the fibre, the varying accessibility of the components, and the strong sorptive forces exerted by cellulose, all combine to prevent as complete separation as one would wish. Nevertheless, the same series of scientific breakthroughs that have accelerated progress in the whole field of hemicelluloses have also given fresh impetus to the investigation of fractional extraction.

In 1946, Partridge[1] demonstrated the applicability of paper chromatography to the separation of sugar mixtures. This made possible investigations of polysaccharides that were previously out of the question; in particular, one could analyse extracted hemicellulose fractions readily and reasonably accurately. In 1947, Yan[2], working with Purves, investigated

liquid ammonia as an extractant. This was the first use in fractional extraction of a nonaqueous solvent. In 1956, Hägglund, Lindberg, and McPherson[3] explored a second such solvent, dimethyl sulphoxide. This exhibited a certain specificity in its action, as will be discussed later. Still more specific was the use of alkali borate extractions, introduced the same year by Jones, Wise, and Jappe[4]. It was also this year that Anthis[5], working with Wise, demonstrated unequivocally the existence of a chemical linkage between mannose and glucose in the cell-wall polysaccharides of spruce, which led to the establishment of glucomannans as a class. Finally, in 1957, Hamilton and Quimby[6] demonstrated the superiority of sodium hydroxide over potassium hydroxide for extracting hemicelluloses.

These developments changed the rate of progress tremendously, but they also changed the direction. Prior to this, successively extracted fractions showed a gradual change in properties. The structure of the hemicelluloses was not well understood and fractionation according to molecular size, as in the case of homologous polymers, might have been expected. Now, as we learned more of the complexity of the hemicellulose mixtures, it became clearer that fractionation according to structure was within our reach.

Naturally enough, everyone studies his own particular problems and uses extraction procedures chosen from his viewpoint. This variety of procedures has made it hard to compare results, but in this review we shall try to trace the development of fractional extraction from the start.

We shall not discuss the simplest case of fractional solution, repeated extraction with liquid of the same composition. The omission is not serious, however, for it is well known that the bulk of the extraction is done by the first charge. The composition of the extracted hemicellulose has been found to change with later repetitions, but the small amounts involved make this a rather trivial observation for most purposes.

The first real fractional extractions used the same liquid at different temperatures. Preece[7] extracted " free hemicellulose " from boxwood with 4 per cent sodium hydroxide at room temperature and then " combined hemicellulose " with boiling 4 per cent sodium hydroxide. We know now that the material extracted with hot alkali, and the residue as well, have been greatly altered by the treatment and the procedure is not satisfactory for most purposes, but Preece's work shows two important points, first, that fractional extraction of wood hemicelluloses is possible (his yields were 7·7 and 6·4 per cent respectively, of the dry wood) and, second, that it might be incomplete (there was still " xylan " in the residue). Of course, the principle of fractional extraction of the hemicelluloses was established much earlier when Cross, Bevan, and Smith[8] showed that alkali extracts only part of the furfuroids.

The only other application of hot alkali extraction to the fractional solution of wood hemicelluloses is the work of Mitchell, Rogers, and Ritter[9,10] on maple holocellulose. It has been used, however, on such materials as corn cobs and cottonseed hull bran[11].

Sands and Nutter[12] used a different fractional extraction to separate " free " and " combined " hemicelluloses from mesquite wood. The wood was extracted with 10 per cent sodium hydroxide, then delignified, and, finally, a second fraction was extracted with 10 per cent sodium hydroxide.

Anderson and his co-workers used this technique on locust, lemon, birch[13], cottonwood[14], and white pine[15]. Rogers, Mitchell, and Ritter[16] and Clermont[17] used modified variants on maple sawdust and on spruce holocellulose, respectively. Rogers, *et al.*[16] used an interesting procedure—steeping the material first in 20 per cent sodium hydroxide and then diluting to 3 per cent before filtering. In none of the above cases is there much difference in the nature of the extracts, only a slight increase in mannose in the second extraction for softwoods and a correspondingly slight decrease in xylose and uronic anhydrides. Opinions are divided as to the effect of delignification. Some believe that a chemical bond is actually broken, others that lignin simply inhibits swelling to the extent that complete removal of the hemicelluloses is hindered, but there is no conclusive evidence. In any case, delignification is helpful in any extraction, indeed it is quite essential for softwoods, as Nelson and Schuerch have shown[18].

Swelling is also a factor in achieving fractionation by extraction sequences with increasing alkali concentration where the swelling increases correspondingly. It should be remembered, however, that Schuerch[18] has shown, in extracting hemicelluloses from hardwoods, that there is an optimum swelling concentration above which the swelling of the cellulose inhibits the removal of the pentosans.

Successive extractions with alkali of increasing concentration were introduced very early. In 1931, Anderson[11] extracted corn cobs with 4 per cent sodium hydroxide, followed by a second extraction with 17 per cent sodium hydroxide. Many groups of chemists have used some type of concentration sequence (*Nables 1* and *2*).

Table 1. Sequences of varying concentration: KOH

Alkali concentration (%)	*Investigators*
5–16	Wise, *et al.*[20–22]
5–16	Brasch and Jones[23]
5 24	Wise, *et al*[19, 24]
5 24	Runkel and Luthgens[25]
5–24	Karnik[26]
5–16–24	Wise and Ratliff[27]
1–4–7–16	J. O. Thompson, Swanson, and Wise[22]
1-4–7–16	Most[28]

Table 2. Sequences of varying concentration: NaOH

Alkali concentration (%)	*Investigators*
1–5	Millett and Stamm[29]
1-2–3–4–6–12	Nelson and Schuerch[18]
0·5 ·1 2 -12	Roudier[30]
1–4–10	Aspinall and McKay[31]
5–10–18	Painter and Purves[32]
4–17·5	Laidlaw, *et al.*[33]
4–10	Laidlaw, *et al.*[33]

Some fractionation occurs in these extractions as seen in *Table 3*, but it is not very great; the glucuronoxylan group of hemicelluloses is, in general, more easily extracted than the glucomannan group and fractionation is, therefore, more apparent in the softwoods where there is more of the latter. Really efficient separation only came when specific action was attained by the use of different compounds for extraction.

Table 3. Fractionation by varying alkali concentration slash: pine holocellulose*

KOH *concentration* (%)	*Percentage of total*	*Mannose units* (%)	*Pentose units* (%)	D P.	*Uronic acid* (%)
1	42	5·4	49 5	100	28·0
4	17	10·2	56·1	105	20·4
7	19	15 2	48·9	120	16·8
16	22	22·6	27·8	130	12·8

* Data from Thompson, Swanson and Wise[22] except last column from Most[28].

The first experiments describing the use of different materials did not exhibit any very specific action, nor, indeed, were they used for this purpose. For instance, Voss, Bauer, and Pfirschke[31] extracted beechwood with 0·2 per cent sodium hydroxide, followed by extraction with a solution containing 5 per cent sodium hydroxide and 3 per cent sodium chloride. Actually, this is an extension of studies begun much earlier by Schmidt. The first paper by Schmidt on cellulose and its incrustants was published in 1921[35], before the work already discussed, but he never seems to have isolated his very soluble xylan and less soluble xylan from a single source. In all his work on beechwood, which was summarized in 1937[36], he drew his conclusions from weight losses and properties of the residues. Both Schmidt[37], and later Voss[34], added salt to the alkali to hinder solubility of β-cellulose, not to increase the solubility of the non-celluloses.

Other early experiments in different extractants were aimed merely at altering the pH of the alkali used. Such a sequence was Bennett's extraction of cornstalk holocellulose[38], which used 2 per cent sodium carbonate, 4 per cent sodium hydroxide and 10 per cent sodium hydroxide. This resembles Mitchell, Rogers, and Ritter's earlier sequence used on maple[9,10], except that Bennett did not mention the use of hot alkali. Separations achieved in this way are not very startling and, as the yields in Thomas' study of aspen show[39], most of the extraction is done by the stronger alkalis (*Table 4*).

It might be argued that the use of liquid ammonia is another example of this type, but this is only superficially so. Ammonia is a weak base, but liquid ammonia is a very strong swelling agent and facilitates subsequent extraction. It was first used by Yan and Purves[2, 40] as an extractant for lignin and, later, carbohydrates and it was shortly after used by Bishop and Adams[41] in a fractionation of wheat straw holocellulose. This does not strictly belong in a discussion of wood chemistry, but it is the first use of liquid ammonia in a fractionation sequence and also the first use of paper partition chromatography for evaluating hemicelluloses fractionally extracted. Both techniques were applied to wood the same year by Bjorkqvist

and Jörgensen[42]. The fractionating capacity is probably best shown by the recent paper of Milford, Watts, and Purves[43] in which spruce holocellulose was extracted first with liquid ammonia at 20°, then with water at 75°, and finally with 10 per cent sodium hydroxide at 5°. The ammonia deacetylates the acetylated derivatives and removes lignin and a little carbohydrate. The warm water removes what is mainly a glucuronoarabinoxylan. There is glucose and galactose in the mixture, but no mannose.

Table 4. Extraction of aspen holocellulose

Extractant	*Yield, on holocellulose*
Water at 20°	3·9
Water at 100°	9·1
5% $KHCO_3$	0·6
5% K_2CO_3	1·6
1·4% KOH	1·3
5·6% KOH	10·8
13·9% KOH	6·3
23·8% KOH	1·1
Residue	65·0
Total	99·7

The sodium hydroxide removes a mixture of which over half is glucomannan. The extracted pulp still contains 9 per cent of mannose residues. This was reduced to 3 per cent by extraction with 18 per cent sodium hydroxide, but the extract was not worked up.

The thesis of Schoettler[44] (The Institute of Paper Chemistry, 1952) has not been published as such. It dealt with the resistant xylan of kraft pulp and included some data of interest here. When aspen kraft pulp was treated with 17·5 per cent sodium hydroxide, a complex mixture of hemicelluloses is extracted, of course, but Schoettler found that with 24 per cent potassium hydroxide, he could get a yield of 5·7 per cent of a pure xylan from the residual α-cellulose (90·5 per cent xylose residues, 8·9 per cent ash). Glucuronic acid residues were probably removed during the kraft cook. This preferential action of the two alkalis in the case of aspen is remarkable, since it has since been shown, in the case of softwoods, that sodium hydroxide is the better extractant. In 1957, Hamilton and Quimby[6] studied the action of the two alkalis at various concentrations on Western hemlock. In the same year, Aspinall, Laidlaw, and Rashbrook[45] used an extraction sequence on partially delignified Sitka spruces of 4 per cent sodium hydroxide—24 per cent potassium hydroxide—delignification—10 per cent sodium hydroxide. The last fraction was largely glucomannan. Later work by Hamilton and N. S. Thompson[46, 47], Dutton and Hunt[48], and Murray, Morak, and Ward[49–51] has adequately demonstrated the value of extracting softwood hemicelluloses with potassium hydroxide and then sodium hydroxide. It has also emphasized again the importance of swelling. In order to remove everything soluble in a given extractant, the structure must be as available as possible to the extractant, which can only be achieved at high states of swelling. Even then, sorption phenomena will

prevent absolutely complete removal. The difference between the behaviour of aspen and of other woods is still unexplained.

Jones, Wise, and Jappe[4] introduced the use of alkali borate in 1956 and Jones, Merler, and Wise[52] used it in fractionally extracting aspen wood the next year. The sequence (water, 1 per cent potassium hydroxide, 10 per cent potassium hydroxide, 20 per cent potassium hydroxide, and 20 per cent potassium hydroxide + 4 per cent boric acid) gave mainly xylan polymers in the first four and mainly glucomannans in the final fraction. The value of this reagent was immediately recognized and it has been used by many groups since. The same is true of the use of dimethyl sulphoxide, also introduced in 1956 by Hägglund, Lindberg, and McPherson[3] as a solvent for hemicelluloses. The list of those who have made use of extraction sequences containing one or both of these solvents includes Jones, Merler, and Wise[52], Lindberg and Meier[53, 54], Croon[55], Mian and Timell[56–62], Nelson[63], Leopold[61], Murray, Morak, and Ward[49–51]. Koshijima[65] described an interesting variant last year; he extracted pine holocellulose with dimethyl sulphoxide acidified with acetic acid, then dimethyl sulphoxide acidified with boric acid, then water, and finally a more acid dimethyl sulphoxide mixture with boric acid. The pH values of his dimethyl sulphoxide extractants were 6·8, 5·2, and 4·3 respectively. Decreasing amounts of pentose and increasing amounts of glucomannan were evident in his fractions.

To sum up the present status of fractional extraction of softwood hemicelluloses, the problem is to effect as good a separation as feasible of the xylan-containing components and of the glucomannans and related compounds during extraction. We can come nearer this with some of the recent techniques described, but we are not near enough. We can remove almost all the xylan-containing components, but some of the galactoglucomannan group are so readily available that they are also removed in the extract. By removing the xylan, however, it is possible in a later extraction to get a nearly pure glucomannan. It will not represent the entire mannose content of the tree, however. *Table 5* shows the composition of this fraction by different methods. In this table, Lindberg and Meier[53] used a sequence of dimethyl sulphoxide—hot water—14 per cent potassium hydroxide—14 per cent potassium hydroxide + 3 per cent boric acid—24 per cent potassium hydroxide + 3 per cent boric acid. This represents

Table 5. Crude glucomannan fractions

Source	*Yield*	*Composition*					*Investigator*
		Gluc	*Gal.*	*Man.*	*Arab.*	*Xyl.*	
Spruce holo.	7·6*	[17·7]		81·0	—	1·3	Lindberg and Meier
Pine holo.	2·9	[24·4]		72·6	1·2	1·8	Meier
Pine holo.	2·3	[24·7]		70·3	2·3	2·7	Meier
Slash pine holo.	2·0	[24·5]		70·2	Minor	5·3	Nelson
Spruce holo.	9·6	36·3	1·2	60·3	0·6	1·6	Morak and Ward
Spruce holo.	5·1	21·6	1·9	75·9	—	0·6	Morak and Ward

* Total amount extracted.

the last but one fraction. Meier[54] used the same sequence. The results shown are for the water extraction and for the 24 per cent potassium borate. Nelson[63] used a series of sodium hydroxide extracts of varying strength, followed by a sodium borate hemicellulose which is that given. In the case of the two examples from Morak and Ward[50], the xylans were first removed with potassium hydroxide. The final extractant was then sodium hydroxide in the first case and potassium borate in the second.

Let us discuss briefly some of the properties of these unpurified extracted hemicelluloses. The reasons for my interest in the unpurified fractions have been indicated; let me review a few of the results.

The sequence studied most thoroughly is essentially in three stages—potassium hydroxide, potassium borate, and sodium hydroxide—although several successive extractions were actually made with each to get exhaustive extraction with each reagent. The analyses in *Table 6* are for the alcohol-insoluble precipitate from the first extract with each reagent.

Table 6. Comparison of fractions from spruce holocellulose

Extractant	*Yield*	*Composition of hydrolysate*				
		Galactose	*Glucose*	*Mannose*	*Arabinose*	*Xylose*
KOH	21·02	14 0	9·9	21·7	7·9	46·5
KBO_2	5·26	3·6	19·8	75·4	—	1 2
NaOH	4·90	2·0	54·5	41·3	—	2 2

The first fraction is the usual mixture obtained with potassium hydroxide. The second is a practically pure galactoglucomannan. We are not sure yet of the sodium hydroxide fraction. It is a mixture and probably contains the same glucomannan as the preceding along with some degraded glucans from cellulose, but this has not yet been proved.

To get optical rotations (*Table 7*) we dissolved each fraction in the extractant used (incidentally, this is not always easy), but we cannot draw any important conclusion other than that there are differences between the hemicelluloses from holocelluloses, sulphite pulp and kraft pulp, which we already knew from the chromatograms. A study of the rotations of the borate complexes would be needed to evaluate this series thoroughly.

Table 7. Optical rotations

Source	KOH *hemi.*	KBO_2 *hemi.*	NaOH *hemi.*
	$[\alpha]_D^{21}$	$[\alpha]_D^{21}$	$[\alpha]_D^{20}$
Chlorite holocellulose	−40·7	−65·8	−78·4
Chlorited sulphite pulp *A*	−77·4	−72·7	−21·3
Chlorited sulphite pulp *B*	−78 9	−74·4	−22·6
Chlorited kraft pulp	−108·2	−79·6	−46·4

X-ray diagrams were obtained of all the precipitated hemicelluloses and of all the residues at each stage. I am repeating here (*Figure 1*) the diagrams

of the three hemicellulose fractions extracted from spruce sulphite pulp combined for convenient comparison. Kraft pulp and holocellulose furnish similar sets of diffractograms[51]. There is no doubt that the fractions are crystalline, in the sense that polymers are crystalline. There is no doubt that they are different from each other, but the difference between the borate and the sodium hydroxide hemicelluloses is not great. The peaks at about 12·5 and about 20·5 are similar in both. They are also similar to Cellulose II. The patterns of the potassium hydroxide hemicelluloses correspond roughly to those of purified glucuronoxylans, but the main peak is at an intermediate value between the strong reflection for xylan and xylan hydrate of Marchessault and Timell[66]. The pattern of the borate fraction strongly resembles that of the hydrolysed glucomannan described by Lindberg and Meier[53].. However, we shall at this time heed the warning of Bjorkqvist, Jorgensen, and Wallmark[67] and draw no conclusions as to identity.

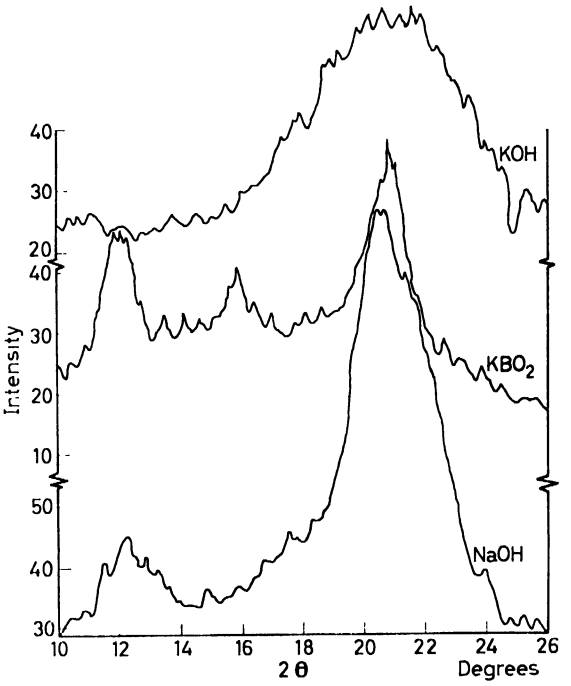


Figure 1 X-ray diffraction of hemicellulose of sulphite pulp (S_3)

The crystallinity was also investigated by means of the electron microscope. The general similarity of the crystals from the potassium hydroxide hemicellulose to the xylan crystals of Yundt[68, 69] and of Marchessault[70] is striking (*Figure 2*). This is, we believe, ample evidence that such crystals are *not* artefacts produced during the purification procedures.

Finally, we should like to discuss the molecular size of these fractions. We examined the potassium hydroxide extract of the holocellulose in detail.

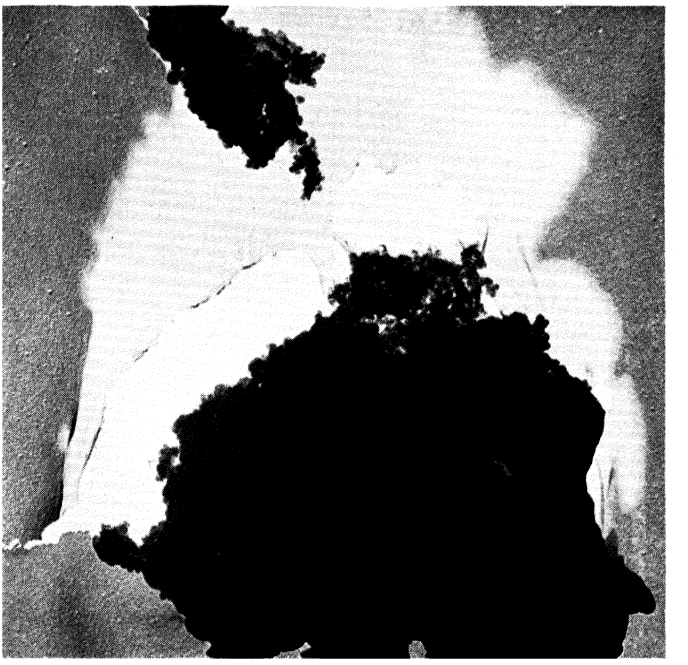


Figure 2 Spruce wood hemicellulose extracted from chlorite holocellulose with 3·9 N KOH; chromium shadowed

Light-scattering measurements gave a molecular weight of 45,000 and ultra-centrifuge measurements 42,000. The intrinsic viscosity in cupric ethylene-diamine was 0·52. When we used the calculated monomer weight of 145·8, this gave us a ratio of P_w (light scattering)/$[\eta]_{CED}$ of 592. This is in reasonable agreement with similarly calculated ratios for 4-*O*-methyl-glucuronoxylans from the data of Goring and Timell[71]: 561 and 562. This is, of course, mostly fortuitous, for both they and we have shown that this ratio varies with the source of the hemicellulose.

In spite of all the uncertainties involved, we have used the value of 592 in calculating the D.P.'s in *Table 8*. We recognize that there is error in going from one hemicellulose to another and that the relation of D.P. to intrinsic viscosity may not be so nearly linear for hemicelluloses as for cellulose, but we wanted some approximation and we prefer to use a weight-average value rather than a number-average and a value derived on hemi-celluloses rather than one derived from celluloses. In spite of the fact that these hemicelluloses were obtained from chlorited materials, the molecular weights are higher than most of those in the literature, except those of Goring and Timell[71] who were careful to avoid degradation. Some pre-liminary work at The Institute of Paper Chemistry indicates that our calcu-lations for the borate fractions may be much too low.

Table 8. D.P. of extracted hemicelluloses

Chlorited holocellulose from	KOH *hemi.*	KBO_2 *hemi.*	NaOH *hemi.*
Sprucewood	308	254	355
Sulphite pulp *A*	201	148	361
Sulphite pulp *B*	201	148	319
Kraft pulp	396	201	373

Summing up briefly, it is now possible, for softwoods at least, to make a preliminary fractionation of hemicelluloses during the extraction process and to obtain the galactoglucomannan series in practically pure form. In the sequence used by us, this is the second of three fractions. It should be stressed that while we can reduce the number of steps in the sequences dis-cussed, we are not obtaining our hemicelluloses unchanged so long as we use alkaline extractions. Acetyl groups and, perhaps, galactose units have been removed. It may be possible to devise a sequence which will separate the xylose group from the mannose group in the first extraction. Dimethyl sulphoxide looks promising but we have not been able to obtain the separations reported by Meier[51]. We are still investigating the material in our final fraction.

Let me close by giving credit to my co-workers at the Institute, Mr Swenson for the light scattering, Dr Kurath for the ultra-centrifuge, Dr Williams for the X-ray diagrams, Miss Smith and Mr Dearth for the electron micrographs, and Dr Wise, Dr N. S. Thompson, and Mr Dickey for valuable advice and occasionally comfort and condolence.

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MODERN METHODS OF DETERMINING CRYSTALLINITY IN CELLULOSE

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INTRODUCTION

The existence of three-dimensional order in celluloses is shown conclusively by X-ray and electron-diffraction diagrams of such materials as ramie fibres[1], *valonia ventricosa* cellulose[2] and Fortisan fibres[1]. These diagrams exhibit many sharp reflections which can only be accounted for in terms of three spatial co-ordinates. All the reflections from regenerated celluloses which possess the cellulose II structure can be accounted for in terms of a monoclinic unit cell of approximate dimensions[3] $a = 7{\cdot}92$ Å, $b = 10{\cdot}34$ Å, $c = 9{\cdot}08$ Å, $\beta = 62{\cdot}7°$. Similarly the reflections of ramie (cellulose I) can be accounted for by a monoclinic unit cell of approximate dimensions[3] $a = 8{\cdot}17$ Å, $b = 10{\cdot}34$ Å, $c = 7{\cdot}85$ Å, $\beta = 83{\cdot}6°$. The electron-diffraction diagrams[2] of *valonia ventricosa* cellulose however suggest a unit cell of triclinic symmetry for this material in which the a and c axes are double those quoted for ramie cellulose.

Although these observations show the existence of three-dimensional crystalline order they do not prove that all the regions which contribute to the sharp reflections of the diffraction diagrams show the same high degree of three-dimensional order. Such a conclusion would be warranted only if good agreement between calculated intensities for an ordered crystal structure model and the observed intensities could be obtained. This has not yet been shown to be possible[4]. Indeed observations made by Hermans and Weidinger[5] suggest that different degrees of ordering may exist along different directions in cellulose II crystallites. In using the term crystalline to discuss cellulose structure it should be remembered, therefore, that this does not necessarily imply the existence of full three dimensional order and that there is an element of ambiguity in the use of the term.

The non-crystalline portion of celluloses is frequently called amorphous, and a similar ambiguity is attached to this term which is often used to mean complete lack of order. Although the nature of the non-crystalline or amorphous regions is still largely a matter for speculation, it is clear that there can be no sudden transition from three-dimensional order to complete absence of order, since the long chain cellulose molecules are known to pass through both crystalline and amorphous regions. The possibility of the existence of a continuous range of degrees of order in celluloses must be admitted.

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The use of the terms *crystalline* and *amorphous*, or the division of a cellulose into crystalline and non-crystalline regions, provides a valuable way of picturing cellulose structure in spite of these theoretical objections and the lack of precision attached to the terms. The simple crystalline–amorphous concept of cellulose structure implies that structure can be pictured in terms of only two types of regions of widely different properties, one highly ordered and the other highly disordered, these regions having the same structure and properties in different celluloses. The extent to which this simple picture provides an adequate approximation to the true structure of celluloses is a matter for experimental investigation. Whatever the answer to this question, however, it is reasonable to try to determine the fraction of a cellulose which is crystalline towards a particular experimental technique.

DETERMINATION OF CRYSTALLINITY

Many different techniques have been used to determine crystallinities of celluloses and it has been found that results from different methods are rarely in agreement[6]. The significance of this fact is not clear however, since most of the methods measure properties of cellulose which are connected with crystallinity in a rather indirect way, *e.g.* the rates or extents of chemical reactions and physical properties such as moisture sorption.

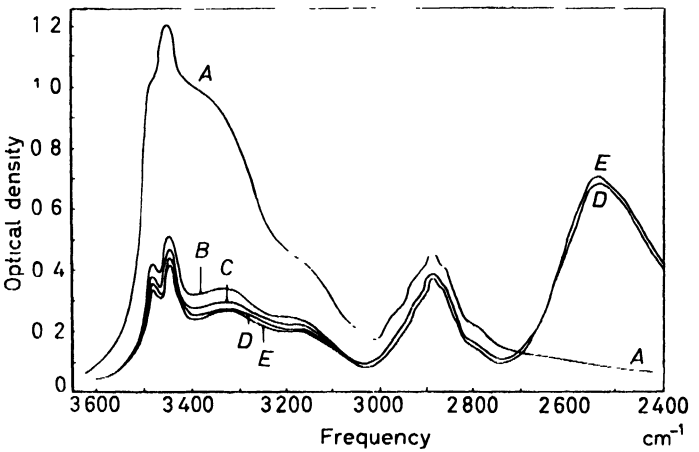


Figure 1. Viscose film—exchanged with D_2O vapour: *A:* dry viscose undeuterated; *B:* Deuterated for 5 min, *C:* deuterated for 30 min; *D:* deuterated for 60 min; *E:* deuterated for 120 min

Since the phenomenon of crystallization is due to the orderly arrangement of molecules relative to one another, the most significant measurements of crystallinity are those obtained by physical techniques which study order in a rather direct way and are based on principles which are thoroughly understood. It is only in terms of the results obtained by such techniques that the significance of measurements by other methods can be understood.

The two properties of cellulose which are most closely related to the presence of intermolecular order are its X-ray diffraction pattern and its infra-red spectrum. X-ray diffraction must be regarded as the basic

method for all crystallization studies because of the direct relation between the diffraction pattern and order. The infra-red spectrum of a material is less closely connected with the presence of order, and its importance in the case of cellulose lies in the great sensitivity of bands due to stretching of OH groups to the local environment around such groups.

INFRA-RED STUDIES OF CRYSTALLINITY

When D_2O vapour is passed over a thin film of cellulose there is a rapid exchange reaction in which OH groups are converted into OD groups. This reaction produces changes in the infra-red spectrum[7] which are illustrated in *Figure 1* for a regenerated cellulose. The band due to stretching of OH groups, which extends over the frequency range 3500–2950 cm^{-1}, is reduced in strength and there is a corresponding increase in absorption in the 2500 cm^{-1} region caused by the appearance of a band due to stretching

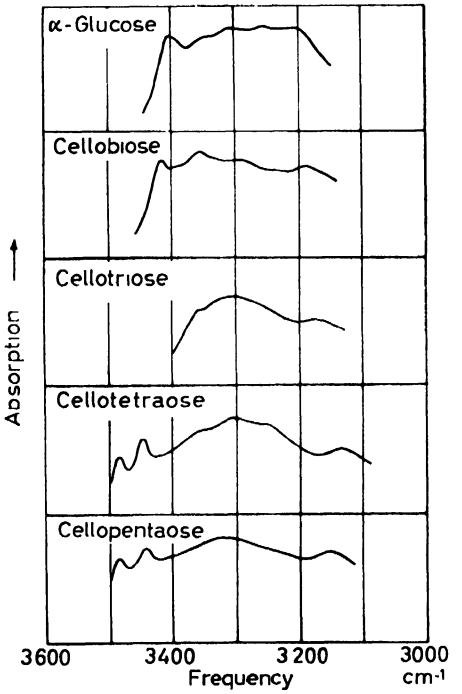


Figure 2. Infra-red spectra of oligosaccharides

of OD groups. The reduction in the optical density of the OH band is due to the removal of a broad diffuse absorption similar in shape to the OD band. As the reaction proceeds the reduction in the optical density of the OH band continues; after about 1 hour, the broad OH band has disappeared completely, leaving four well-resolved absorption bands. Further deuteration leads to a very slow decrease in the optical density of all four bands with no change in their relative peak heights.

These four resolved bands are due to OH groups in crystalline regions which are hydrogen bonded in a regular manner. This can be shown from the spectra of the oligosaccharides illustrated in *Figure 2.* Cellotetraose and cellopentaose show the same four bands as cellulose, and X-ray diffraction diagrams prove that these materials are fully crystalline and have monoclinic unit cells of similar dimensions to the cellulose II lattice of regenerated cellulose[3].

Since it is known from X-ray diffraction that the regenerated cellulose contains only one type of crystal lattice, the broad diffuse absorption band which is removed by deuteration must correspond to absorption by OH groups in non-crystalline regions. This is consistent with our knowledge of absorption by hydroxyl groups in liquids and amorphous solids. The diffuseness of the absorption band is presumably due to the fact that the hydroxyl groups are hydrogen bonded in an irregular manner. This leads to a continuous range of hydrogen bond strengths and, consequently, to a series of absorption bands of different frequencies which overlap and are unresolved. In contrast, only a limited number of different hydrogen bond strengths exists in crystalline regions, leading to discrete, resolved absorption bands.

Quantitative measurements of the decrease in the optical density of the OH band with time have shown that all the OH groups in non-crystalline regions can be deuterated before appreciable deuteration of OH groups in crystalline regions has occurred. At this stage of the reaction therefore, the ratio of the optical densities of the OH and OD bands of the dry film gives a measure of crystallinity; determination of this ratio for different

Table 1

Cellulose	*Crystallinity* (%)
Saponified acetate	25
Viscose rayon	26
Precipitated cellulose	32
Viscose treated with NaOH	33 5
Mercerized bacterial cellulose	33
Bacterial cellulose	70

celluloses gives relative crystallinities. Absolute values of crystallinity can be obtained by estimating the deuterium content of the cellulose. This can be done by extracting the deuterium from the cellulose with a known amount of H_2O and estimating the deuterium content of the water by measuring its refractive index[8]. Absolute crystallinities obtained in this way can then be used to calibrate the infra-red method so that absolute crystallinities on further samples can be determined by infra-red spetroscopy[9].

The method can also be used to determine crystallinities of native celluloses which possess the cellulose I crystal lattice. A separate calibration of the infra-red method is required for these celluloses, however, since the bands due to stretching of OH groups in crystalline regions are different from those of regenerated celluloses[7].

Some crystallinity values determined by the infra-red method are given in *Table 1.* These crystallinities will not necessarily be the same as crystallinities measured on the same samples by other techniques, and it is important to bear in mind what the infra-red method measures. The crystallinity measured by the infra-red method described above is the fraction of *OH* groups which are hydrogen bonded in the regular manner characteristic of crystalline material. It can be readily appreciated that the X-ray method, for example, measures a somewhat different quantity, ideally the fraction of *molecules* which are arranged in a regular repeating pattern.

X-RAY STUDIES OF CRYSTALLINITY

The existence of crystallinity in celluloses was first revealed by X-ray diffraction[10] and this technique must still be regarded as the basic method for studying crystallization. However, despite the excellent pioneering work of Hermans[11] and many subsequent investigations, it must be admitted that we still cannot determine absolute crystallinities of celluloses in a completely objective way by this method. It is even doubtful whether reliable relative crystallinities can be obtained.

The basic step in determining the crystallinity of any polymer from its X-ray diffraction pattern is to separate the total scattering curve into scattering from crystalline material and scattering from non-crystalline material. This is a simple matter with polymers such as natural rubber where only the amorphous regions contribute to the scatter at lower angles. In the case of cellulose, however, the scattering from crystalline and non-crystalline material overlap seriously and it is doubtful whether there is any angle at which accurate measurements can be made where only one type of material scatters. In these circumstances, an objective separation of the total curve into its component parts can be made only if the precise forms of *both* scattering curves are known.

Following the work of Hermans and Weidinger[11], ball-milled cellulose has frequently been used as a standard for amorphous cellulose in X-ray work. The suitability of this material as a standard has been reinforced by observations of Ellefsen *et al.*[12] on lichenin and β-methyl cellobioside glass and by recent work carried out at the British Rayon Research Association[13].

The monomer of cellulose, glucose, and its polymers, cellobiose to cellohexaose have been obtained in the amorphous state by freeze drying their aqueous solutions. The X-ray diffraction curves of these substances are shown in *Figure 3* together with the curve for ball-milled cellulose. The latter curve was found to be the same within experimental error for ball-milled cellulose prepared from cotton (cellulose I) or from Fortisan H (cellulose II). It can be seen that the scattering curves are all very similar, particularly in the position of the maximum and the shapes of the high angle sides of the curves. The main difference between cellulose and its lower homologues is the shoulder exhibited by cellulose on the low angle side of the maximum. However, this feature fits in with a trend observed in the shapes of the oligosaccharide curves. The curve of glucose is convex to the 2θ axis on the low angle side of the maximum and the curves of the higher members of the series become progressively less convex until with cellohexaose this part of the curve is almost linear. This comparisor

suggests, therefore, that ball-milled cellulose is a suitable standard for amorphous cellulose.

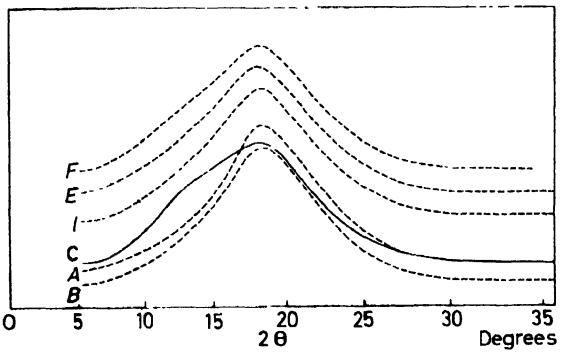


Figure 3. Scattering curves of amorphous oligosaccharides and ball-milled cellulose; curves displaced along intensity axis: *A:* glucose and cellobiose; *B:* cellotriose; *I:* cellotetraose, *E:* cellopentaose; *F:* cellohexaose; *C:* Ball-milled cellulose

The scattering curves for ball-milled cellulose and some cellulose derivatives which have been subjected to ball-milling or quenched from the melt also form a regular series, as can be seen from *Figure 4.* All the cellulose derivatives show two maxima, the positions and shapes of the higher angle maxima being very similar to that of cellulose. The maxima at lower angles vary in position however, there being a trend towards higher angles as the size of the substituent decreases. The shoulder of the cellulose curve fits in with this trend.

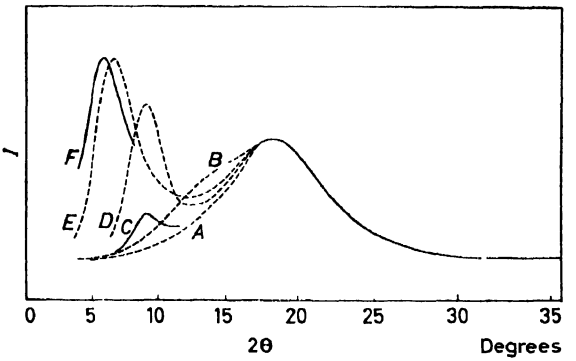


Figure 4. Scattering curves of ball-milled cellulose and its derivatives: *A:* amorphous glucose; *B:* cellulose; *C:* sodium carboxymethyl cellulose; *D:* methyl cellulose; *E:* ethyl cellulose; *F:* cellulose tributyrate

Since no part of the non-crystalline scattering from a partially crystalline cellulose can be measured with certainty free from overlap by crystalline scattering, an objection can be raised against the use of ball-milled cellulose as a standard for crystallinity determinations. The objection is that there is no proof that there are regions in a partially crystalline cellulose which

have the same structure and scattering curve as ball-milled cellulose. For this reason the X-ray diagram of highly oriented Fortisan H has been examined[13].

The crystalline reflections from this material are observed as short arcs because of the high orientation. On photographs exposed for long times a weak halo can be seen which is strongest near the equator where it merges with the 002 and $\bar{1}01$ crystalline reflections and which extends outwards towards the meridian. This halo can be photometered without too great an interference from crystalline reflections and the resultant curve is shown in *Figure 5.* It can be seen that the form of the curve is generally similar to that of ball-milled cellulose. It seems reasonably certain, therefore, that there are regions in partially crystalline cellulose which scatter X-rays in a way similar to ball-milled cellulose.

Unfortunately, we do not possess similar information concerning the shapes of the X-ray reflections from crystalline regions and it follows, therefore, that we cannot obtain an absolute crystallinity from the X-ray diagrams of celluloses. It is possible to fix a minimum for the absolute crystallinity, however, by carrying out the separation of the total scattering curve according to the procedure used by Ellefsen *et al.*[12]. It is obvious that the amorphous scattering cannot be larger than the total scattering at any point on the curve. Thus, if the separation is carried out in such a way

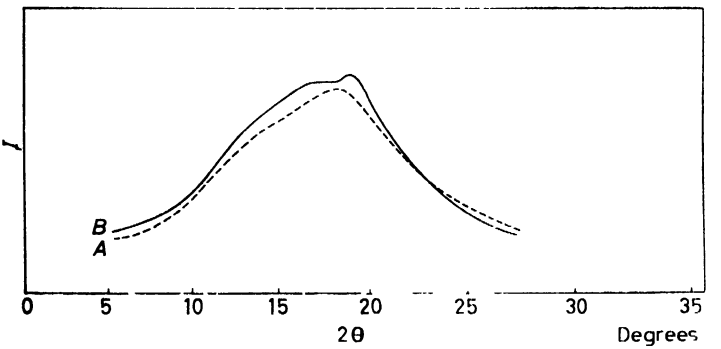


Figure 5. A: ball-milled cellulose; *B:* Fortisan halo

that the amorphous scattering curve touches the total scattering curve at some point, it follows that this must represent a maximum for the amorphous scattering. If ball-milled cellulose is used as a standard for completely amorphous cellulose, a maximum of amorphous content *i.e.* a minimum crystallinity can be obtained. This is the only quantity which can be determined without making any assumptions. It is worth noting that the minimum crystallinity determined in this way represents an absolute quantity which is valid even if celluloses do not possess a simple crystalline-amorphous structure, but have a structure of varying degrees of order as suggested by Howsmon[14]. It would also be valid even if celluloses possess no highly disordered amorphous regions, a situation which Hosemann's theories concerning the X-ray diagrams of cellulose would allow[15]. For

this reason, the term *minimum crystallinity* is to be preferred to Ellefsen's term *amorphicity* for the quantity obtained as described above.

Norman and co-workers[16] have shown that the X-ray diagram of a given cellulose leads to a much higher crystallinity if the not unreasonable assumption is made that the shapes of crystalline reflections are Cauchy distributions. The Cauchy distribution has long tails which lead to appreciable overlap between different crystalline reflections in the X-ray diagram. In consequence, the amorphous scattering has to be drawn in much lower if this assumption is made than is the case in the minimum crystallinity method. *Table* 2 shows values of minimum crystallinity and crystallinity values assuming the Cauchy distribution as determined by Norman and co-workers[16]. The difference between the two crystallinities is particularly striking with the cellulose I samples, since overlap between different crystalline reflections is greater with cellulose I than with cellulose II.

Table 2

	Minimum crystallinity (%)	*'Cauchy' crystallinity* (%)
Acetate grade wood pulp	53	95
H_2SO_4 treated surgical cotton	62	95
Acetate grade wood pulp mercerized in 18% NaOH	43	60
Acetate grade wood pulp treated with 39% HCl	49	60

Further progress in the determination of absolute crystallinities from X-ray diffraction patterns must depend on experimental determinations of the shapes of crystalline reflections. This is not a simple matter however, since it is difficult to be certain whether any of the reflections which can be observed free from non-crystalline scatter are in fact produced by only one type of crystallographic plane[1]. Until decisive evidence is obtained on this point it must be admitted that the X-ray diffraction pattern of a cellulose is consistent with a range of crystallinities.

COMPARISON OF X-RAY AND INFRA-RED CRYSTALLINITIES

In an attempt to throw light on the structure of celluloses and the significance of crystallinity measurements, a comparison has recently been made of the minimum X-ray crystallinity and the infra-red–deuteration crystallinity of a regenerated viscose rayon film[13].

The film was studied in an unoriented form and in a form showing high planar orientation; both ball-milled cellulose and amorphous cellotetraose were used as 100 per cent amorphous standards. The diffraction curves of these materials are shown in *Figures 6* and *7* standardized to amount of X-radiation scattered per unit weight of irradiated material per unit amount of incident X-radiation. Crystal reflected monochromatic radiation was used and the X-ray beam was monitored with a Goppel camera[17]. The radiation was recorded photographically in a vacuum camera. The

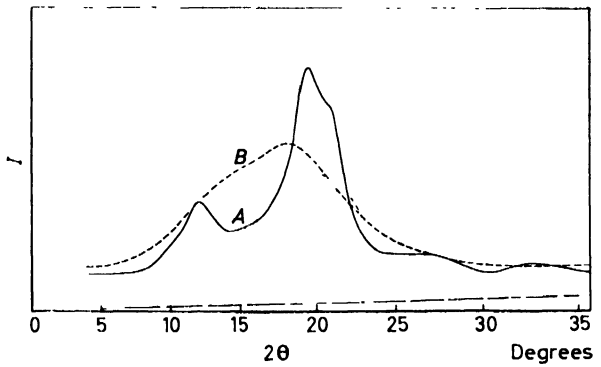


Figure 6. Normalized scattering curves: *A:* unoriented viscose rayon; *B:* ball-milled cellulose

weight of the samples per unit area was the same within 5 per cent and no correction was made for absorption of the X-ray beam. The level of incoherent radiation shown in *Figures 6* and *7* was assessed from the theoretical calculations of Ellefsen *et al.*[12]. The determination of the infra-red crystallinities has already been described[9].

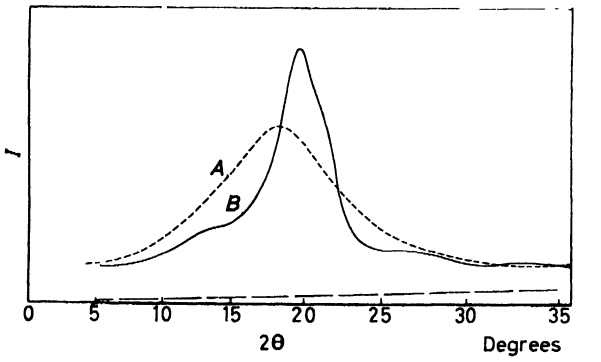


Figure 7. Normalized scattering curves: *A:* planar oriented viscose rayon, *B:* amorphous cellotetraose

The results of the determinations are shown in *Table 3.* The agreement between the figures for minimum X-ray crystallinity obtained with the two amorphous standards suggests that this crystallinity is not very sensitive to the precise shape of the scattering curve assumed for non-crystalline material. It can also be seen that the minimum crystallinity is not very sensitive to the degree of planar orientation present. The significance of this is doubtful, however[13].

The difference between the minimum X-ray crystallinity of 40 per cent and the infra-red crystallinity of 26 per cent is large and implies that although a minimum of 40 per cent of the cellulose chains is arranged in a crystalline way only 26 per cent of the OH groups are hydrogen bonded in the regular crystalline manner.

Now OH groups which show the characteristic absorption pattern of crystalline material deuterate only slowly and this must imply that they are not easily accessible to D_2O; the deuteration reaction is extremely rapid and the rate of deuteration is determined by the rate at which D_2O molecules are able to reach OH groups. It must be assumed, therefore, that such OH groups are protected from easy access by D_2O by being in the interior of crystallites. OH groups which lie in the surfaces of crystallites are presumably readily accessible to D_2O molecules and will deuterate rapidly.

Table 3

	X-ray minimum crystallinity		
	Ball-milled cellulose as standard (%)	*Amorphous cellotetraose as standard* (%)	*Infra-red crystallinity* (%)
Unoriented film	43	40	26
Planar oriented film	43	42	26

Since the OD band of deuterated cellulose shows no trace of structure, the vast majority of surface OH groups must be hydrogen bonded in an irregular amorphous fashion. This conclusion provides an obvious qualitative explanation of the crystallinity results and approximate calculations can be made to test this explanation quantitatively.

It is well established that cellulose II crystallites are tabular in shape with the crystallographic 101 plane as the plate face[18,19]. In addition, our knowledge of the crystal structure shows that the planes of the glucose units are roughly inclined in the crystallographic $\bar{1}01/002$ planes[4]. From this information it can be calculated that for crystallites 29 Å by 65 Å in cross-section 35 per cent of the OH groups will lie in the surfaces of crystallites. The crystallinity measurements show that at least 35 per cent of the OH groups of crystalline chains are hydrogen bonded in an irregular manner. Thus the difference between the minimum X-ray crystallinity and the infra-red crystallinity can be accounted for in terms of crystallites of cross-section 29 Å by 65 Å. The size of this crystallite is consistent with our present knowledge of crystallite size obtained from electron microscopy[18,20] and low-angle X-ray scattering[21].

NATURE OF INFRA-RED AMORPHOUS MATERIAL

Although the difference between the minimum X-ray crystallinity and the infra-red crystallinity seems to be consistent with our knowledge of crystallite size, this is only so if *all* the OH groups which lie in the surface of crystallites are hydrogen bonded in an irregular manner. Now the two sharp infra-red bands of crystalline cellulose at 3485 and 3445 cm^{-1} are due to intramolecular hydrogen bonds which join successive glucose units of the cellulose chain[22] and, therefore, characterize molecular form. The fact that there seem to be no OH groups in the surface of crystallites which

show these bands suggests that the material which is crystalline towards X-ray diffraction but amorphous to infra-red spectroscopy does not possess the precise molecular form characteristic of crystalline chains. The extent of this departure from the perfect crystalline molecular form is uncertain but may not be great in view of the sensitivity of O—H stretching frequencies to the hydrogen bond length[23].

This line of reasoning suggests that the infra-red amorphous material can be divided into two parts. The first is amorphous material of unknown structure and the second chains in crystalline regions which are somewhat distorted from the characteristic crystalline form. It might be expected, therefore, that the structure of the infra-red amorphous material would vary from one cellulose to another, *e.g.* with varying crystallinity. One aspect of the structure of the amorphous material which can be studied is the distribution of hydrogen bond strengths. This can be examined by measuring the shape of the amorphous OD band, since, as was mentioned earlier, this is related to the distribution of hydrogen bond strengths (strictly deuterium-bond strengths).

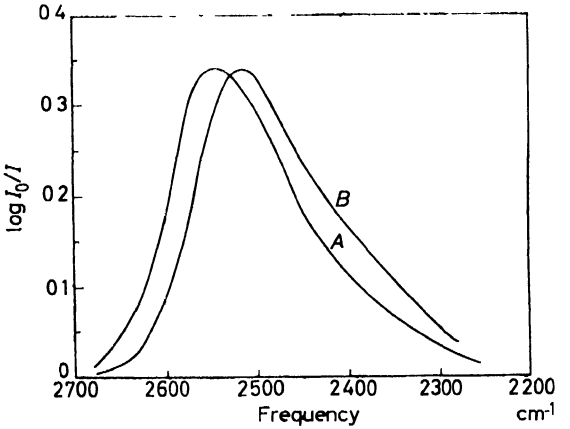


Figure 8. OD band of celluloses: *A:* regenerated cellulose; *B:* bacterial cellulose

The amorphous OD band has been carefully measured for five regenerated and one native cellulose[13]. The bands of the regenerated celluloses were found to be the same within the experimental error but the OD band of bacterial cellulose was found to be significantly different, as shown in *Figure 8.* Although the shape of the band is very similar to that of the regenerated cellulose bands, the whole band seems to be shifted towards lower frequencies, the maximum being some 20–30 cm^{-1} lower than with the regenerated celluloses. This difference shows conclusively that the infra-red amorphous material in bacterial cellulose is different in structure from the infra-red amorphous material of the regenerated celluloses, and suggests that the average hydrogen bond strength in bacterial cellulose is greater than in the regenerated celluloses.

The fact that no differences were found between the regenerated celluloses studied may simply be due to the fact that they did not vary greatly

in crystallinity. The bacterial cellulose differs from the regenerated celluloses not only in its crystal lattice but also in having a very much higher crystallinity[8].

The difference between the OD band of bacterial cellulose and the bands of regenerated cellulose suggests that the absorption bands of the material which is crystalline to X-rays but amorphous to infra-red may be blurred versions of the crystalline absorption bands. This could account for the bacterial cellulose band being at lower frequencies, since the strongest crystalline band of this material is lower in frequency than the corresponding band of regenerated celluloses.

In a highly oriented cellulose the material which is crystalline to X-rays but amorphous to infra-red must be highly oriented. If the absorption bands of this material are in fact blurred versions of the crystalline absorption bands, the amorphous OD band should exhibit dichroism when studied with plane-polarized radiation. Dichroism of this band in a highly oriented regenerated cellulose was observed several years ago during work with plane-polarized radiation[22], but the results were not reported since their interpretation was not clear. In the frequency region corresponding to the two parallel type bands of crystalline material which characterize molecular form, the OD band showed parallel dichroism; however, there were no signs of any absorption peaks. This observation now seems to provide support for the view that part of the infra-red amorphous material shows bands which are a blurred version of the crystalline bands, at least in the high frequency region. It is reasonable to identify this material as that which is crystalline to X-rays but amorphous to infra-red.

NATURE OF CRYSTALLINE MATERIAL

The crystallinity measurements show that 26 per cent of the viscose rayon film is crystalline towards both X-ray diffraction and infra-red spectroscopy. Study of the deuteration reaction shows, however, that this material must possess varying degrees of order.

It is known[7-9] that although this crystalline material deuterates slowly only a limited fraction can be deuterated, the remainder being extremely resistant towards deuteration. Work by Heritage at B.R.R.A. has shown[24] that OH groups in the crystalline oligosaccharides which are hydrogen bonded in the same way as the OH groups in crystalline regions of cellulose deuterate rapidly. The actual deuteration reaction seems to be extremely fast even with OH groups involved in a strong intramolecular hydrogen bond and in a non-polar environment, *e.g.* salicylic acid in carbon tetrachloride[24]. The slow rate of deuteration of crystalline OH groups in cellulose is a measure, therefore, of the rate of diffusion of D_2O molecules into the crystals. Since a substantial fraction of these OH groups cannot be deuterated at all, however, it would seem that the deuteration must occur at regions of imperfection. Thus, the existence of varying degrees of perfection in the crystalline regions seems to be established by these observations, and a study of the detailed course of the deuteration of crystalline regions may throw further light on these imperfections.

Published measurements[8,9] suggest that between a quarter and a third of the infra-red crystalline material in viscose rayon films can be deuterated.

With bacterial cellulose this figure was found to be about 15 per cent. Recent work by Jeffries[25] at B.R.R.A., however, suggests that the fraction of the infra-red crystalline material which can be deuterated in viscose rayon films is higher than reported in the previous work, namely about 60 per cent.

ORDER IN CELLULOSES

The measurements by X-ray diffraction and infra-red spectroscopy which have been outlined above show that four types of material which differ in the degree of orderly arrangement of molecular chains can be distinguished in celluloses. This statement applies to both regenerated and native celluloses, though a complete set of quantitative measurements has only been made on one viscose rayon film[13]. The four types of material are outlined below with reference to this rayon and the percentages of OH groups which show the different types of order.

(i) Material which is crystalline towards both X-ray diffraction and infra-red spectroscopy and which is inaccessible to water. This is the most perfectly ordered material and constitutes approximately 8 per cent by weight of the viscose rayon film.

(ii) Material which is crystalline to both X-ray diffraction and infra-red spectroscopy and is accessible to water. This may be termed imperfect crystalline material and constitutes approximately 17 per cent by weight of the viscose rayon film.

(iii) Material which is crystalline to X-ray diffraction and amorphous to infra-red spectroscopy. This material can be reasonably termed material of intermediate order, *i.e.* material which shows a degree of order intermediate between that of crystalline material and amorphous material. The molecular chains of this material are arranged in a regular crystalline manner, though the chains are somewhat distorted from the form characteristic of crystalline material and form hydrogen bonds of a continuous range of strengths. This material constitutes *at least* 13 per cent of the viscose rayon film, and our present knowledge of crystallite size is consistent with all this material lying in the surfaces of crystallites.

(iv) The remainder of the material is of unknown structure and may tentatively be called amorphous. The hydrogen bonds formed by this material are irregular and cover a continuous range of strengths. It is possible that some of the chains of this fraction may be arranged in an orderly way. This fraction constitutes *at most* approximately 62 per cent of the viscose rayon film.

These facts prove that the simple crystalline–amorphous concept of structure does not provide a close approximation of the true structure of celluloses. Four different degrees of order can be clearly distinguished and quantitatively measured by physical methods. This seems to provide strong support for the ideas on cellulose structure put forward by Howsmon[14].

It has been stated earlier that since the long chain molecules pass through both crystalline and non-crystalline regions there cannot be an abrupt change from a high degree of order to complete disorder. Howsmon's suggestion is that in celluloses this transition is very gradual and a significant

proportion of the total material lies in the transition region. The curve relating quantity of material and degree of order is visualized as being continuous with two peaks, one relating to the highly ordered or crystalline material and one relating to the poorly ordered or amorphous material. The positions and shapes of both peaks are visualized as varying from one cellulose to another. Howsmon and Marchessault[26] have further suggested that the degree of order within the disordered or amorphous material can be described in terms of a distribution of hydrogen bond strengths.

The present measurements provide striking confirmation of two of Howsmon's predictions, namely the existence of material of intermediate order and differences between the distributions of hydrogen bond strengths of infra-red amorphous material in different celluloses. It should be remembered, however, that these facts can be accounted for in terms of the chains lying in the surfaces of crystallites, whereas it is an essential feature of Howsmon's view of structure that the material of intermediate order is located between the crystalline and amorphous regions. A closer estimate of X-ray crystallinity than is afforded by the minimum crystallinity method seems desirable in this respect.

VALUE OF CRYSTALLINITY MEASUREMENTS

The validity and usefulness of measurements of crystallinity on celluloses is called into question by the results which have been discussed. The order present in celluloses cannot be characterized by a single quantitative measurement of crystallinity; indeed it must be admitted that even the term crystallinity cannot be defined in a unique way.

The usefulness of crystallinity measurements by a particular method depends entirely on how well we understand what aspect of structure it measures. The X-ray and infra-red methods which have been discussed at length must be regarded as the basic methods to which other techniques can be referred. The X-ray diffraction method is applicable to all types of material but demands expensive equipment and very careful experimental work. The infra-red method suffers from the severe limitation that it can be easily applied only to film samples. Of the other techniques which have been applied to cellulose, acid hydrolysis and water sorption seem, to the writer, to be the most useful.

Many variants of the acid hydrolysis technique have been described in the literature. The method of Sharples[27] in which the initial rate of hydrolysis is measured is probably the most fundamental of these. The quantitative results obtained from this method seem to be very similar to the crystallinities determined by X-ray diffraction using the assumption of a Cauchy distribution for the shape of crystalline reflections[16].

The value of the water sorption method lies in its simplicity and in the fact that water sorption seems to be related to infra-red crystallinity[28,29]. It must be emphasized that an exact quantitative relation has not yet been established between these two quantities but there can be no doubt that water sorption and infra-red crystallinity show a parallel trend.

There can be little doubt that measurements of crystallinity will continue to play an important part in the study of cellulose structure and its relation to properties. The simple crystalline–amorphous concept also has its part

to play, despite the fact that the true structure of cellulose is more complex than this concept allows. Its value lies in its simplicity and it is unlikely to be abandoned until it is shown that another concept of structure is capable of giving a better *quantitative* account of cellulose properties.

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APPLICATION OF INFRA-RED SPECTROSCOPY TO CELLULOSE AND WOOD POLYSACCHARIDES

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INTRODUCTION

The interpretation of the infra-red spectra of small molecules is one of the important triumphs of molecular physics. These spectroscopic studies have led to precise information concerning the rotational and vibrational properties of molecules and consequently their structure and shape. The spectra of polyatomic molecules, even symmetric ones, offer considerably greater difficulty due to the fact that there are *several* internuclear distances, *several* force constants and so on, which must be determined simultaneously. In the case of polymers, their very great asymmetry and polymorphism lead to complex selection rules and make it all but impossible to calculate the normal vibrations. As a result, the observation made over a decade ago[1] " . . . *the most that can be stated about a large organic molecule or polymer is that it does or does not contain certain chemical groups and even this statement has frequently to be hedged about with certain qualifications* " is still somewhat true.

As polymers go, polysaccharides are among the more difficult to study because the predominance of OH and CH groups with their similar absorption frequencies leads to ill-defined absorption maxima. The presence of several OH groups is in itself a sufficiently complicating factor but to this must be added the numerous hydrogen bonding possibilities which compound the number of different OH frequencies. However, recent reviews[2, 3] show clearly that the situation is far from hopeless. In the synthetic polymer field we are systematically approaching the ultimate goal of the polymer spectroscopist: to establish (i) band assignments, (ii) chain orientation, (iii) chain conformation, (iv) degree of crystallinity and (v) crystal structure, through the infra-red approach. The approach is mainly empirical basing itself on well-established properties of polymers and the known spectroscopic characteristics of the small molecules that constitute the polymer. Some of the foregoing goals are closely related to results which are generally derived from the X-ray diffraction method. There is ample evidence that the two techniques do indeed complement each other and examples of this in the polysaccharide field will be discussed below.

BAND ASSIGNMENT

Before structural or analytical information can be obtained from the infra-red spectrum of a polymer, accurate knowledge must be had of the relation between a given absorption band and the chemical group from

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which it arises. The achievement of this aim by theoretical analysis is out of the question for anything but the simplest polymer molecules. The spectra of the component crystalline carbohydrates are likewise of little help because the higher symmetry, greater crystallinity and different crystal structure of these small molecules often gives rise to much greater spectral complexity. There are times, however, when the spectra of low molecular weight water-soluble fragments (oligosaccharides), which crystallize with the same structure as the parent polymer, yield useful information. Reference will be made to this approach below.

The most straightforward approach to the problem of band assignments is through the familiar group frequencies. Empirically it has been found that, to a first approximation, a good part of the spectrum of a polymer

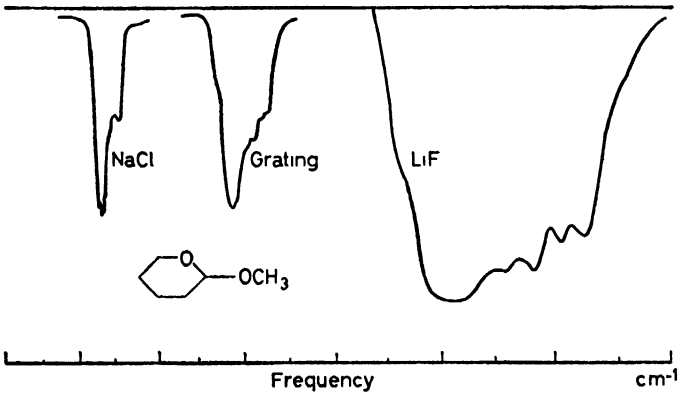


Figure 1. The spectrum of 2-methoxytetrahydropyran in the 2800 to 3000 cm^{-1} region with (*a*) NaCl prism, (*b*) grating of Perkin Elmer 221G instrument and (*c*) LiF prism

can be considered as a composite of the various bands which arise from the component groups within the monomer unit. Most simply this means that vibrational modes due to OH, NH, C=O stretching and CH_3 and CH_2 stretching and bending modes, *etc.*, have been found to be easily recognizable by their characteristic intensities and frequencies. This is not to say that there are never bands that do not admit of such a simple explanation. In particular, it is known that complex vibrational frequencies arise from combinations of the so-called internal group frequencies and the skeleton modes of polymers. The spectral characteristics which are related to the configurational and conformational properties of carbohydrates are likewise as yet uninterpreted. These are problems which remain to be solved. As the solutions are found we can expect a rapid application of the knowledge to the polymer field.

The group frequency approach can be illustrated by considering the spectra of model compounds based on tetrahydropyran. These spectra illustrate how confidence is acquired in the assignment of absorption bands to given chemical groups.

Pyran derivatives

The molecular skeleton of the most important carbohydrates of wood chemistry is the tetrahydropyran molecule. By examining the infra-red

spectrum of simple methyl and hydroxylated derivatives of this molecule it is possible to approach in stepwise fashion some of the characteristics of the spectrum of carbohydrates and polysaccharides in the 3 to 8μ region. The lack of crystallinity and liquid state of these molecular species also gives to their spectra characteristics which one associates with the spectra of very large molecules rather than those of well-crystallized small ones. The fact that these molecules readily undergo conformation changes makes them unsuitable, however, for identifying the ring vibrations and configurational characteristics of carbohydrate spectra which occur in the region beyond 8μ. (The whole field of conformational analysis of the simple pyran derivatives remains to be explored. The recent efforts[4, 5] of the Birmingham school along these lines as well as their pioneering efforts[6] in correlating configurational properties of carbohydrates with infra-red characteristics is recommended as first reference in this area. Some of the useful spectral differences between wood polysaccharides were first reported by them[6].)

By the use of a lithium fluoride prism, or a grating, much greater resolution is attainable in the C—H stretching region than one gets with simple rock-salt optics. The resulting spectra allow separation and assignment of specific bands to the various absorbing groups. An example of this increased resolution is shown in *Figure 1* where the spectrum of 2-methoxytetrahydropyran in the C—H stretching region is shown at three different degrees of resolution. It is evident that when more than one



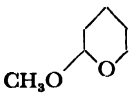
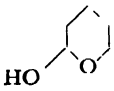
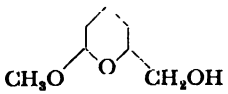
Figure 2 High resolution spectrum in the C—H region of 2-methoxytetrahydropyran, 2-hydroxytetrahydropyran and tetrahydropyran

alkane grouping is present in the molecule, sodium chloride optics are inadequate to resolve the various bands. The grating on the P.E. Model 221G is satisfactory for the spectrum in question but for highest accuracy the lithium fluoride prism is recommended for this purpose. When polarized beam techniques are used important conformational data can be derived from high resolution spectra in this region as we shall see further on.

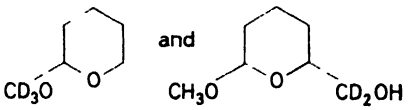
At least six separate absorption frequencies are detectable in the high resolution spectrum of 2-methoxytetrahydropyran. Four of them are at about the expected frequencies for the symmetric and antisymmetric modes of the methylene and *O*-methyl groups. Removal of the methyl group does indeed cause the disappearance of the 2835 and 2970 cm^{-1} bands, commonly associated with the *O*-methyl group, and perhaps of another band at 2893 cm^{-1} as may be judged from *Figure 2*. There is apparently a still further simplification of the spectrum on removal of the hydroxyl group. The two principal bands in tetrahydropyran are at the expected frequencies for the symmetric and antisymmetric stretching modes of a methylene group.

From spectra of the type shown in *Figure 2* the data of *Table 1* listing the frequencies assigned to the CH_2 modes (either on the ring or on the hydroxy methyl group) and the CH_3 modes of the methoxy group were compiled. It is worth noting as shown originally by Henbest and co-workers[20] that the low frequency absorption of the *O*-methyl is at 2835 or lower. This is quite a different frequency than the equivalent absorption band of the *C*-methyl group and important to know since the methoxyl grouping is a frequently occurring one in carbohydrates.

Table 1. Methylene and methyl group frequencies for tetrahydropyran and some of its derivatives

Compound	$\nu_s(CH_2)$	$\nu_a(CH_2)$	$\nu_s(CH_3)$	$\nu_a(CH_3)$
				
	2845	2935		
	2854	2935	2830	2980
	2853	2940		
	2850	2940	2835	2970

To confirm some of the assignments shown in *Table 1* we have made some deuterated analogues of the pyrans shown in the table, *viz.*



The benefit of such compounds in band assignment depends on the fact that the heavier deuterium atoms lead to lower stretching frequencies without affecting the remaining C–H group frequencies. The observed effect is shown in *Figure 3* where the two principal absorption frequencies of the CD_3 group are clearly visible. From the position of these bands it is possible to calculate the theoretical positions of the corresponding CH_3 modes. Generally it is more reliable to observe the high resolution spectra of the two compounds and observe the positions of the missing bands.

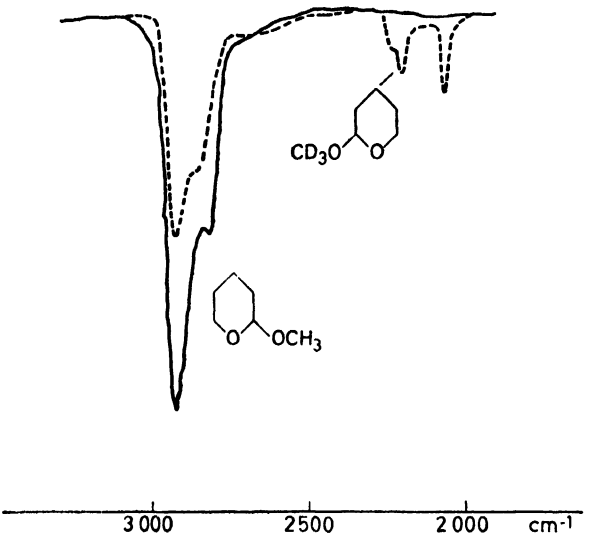


Figure 3. Spectrum of 2-methoxy-d_3-tetrahydropyran in the 2000 to 3000 cm^{-1} region, NaCl optics

One of the principal factors limiting resolution of polysaccharide spectra is the overlap of the CH and OH bending modes between 1500 and 1200 cm^{-1}. This problem is especially acute in the low crystallinity polysaccharides where multiple hydrogen bonds lead to multiplicity of OH modes. In such cases the deuteration technique, *i.e.* conversion of OH to OD, leads to a considerable sharpening of the spectrum in the region in question. As an example the problem is well illustrated by the spectrum of 2,3-dihydroxy-tetrahydropyran shown in *Figure 4.* Conversion of all OH to OD leads to the appearance of bands which are quite different from the ones in the original spectrum. It appears that the resolution of the latter was so poor

that little or no confidence can be placed in the recorded spectrum in this region. The absorption frequencies of the C—H bending modes can be determined from the spectrum of the deuterated material.

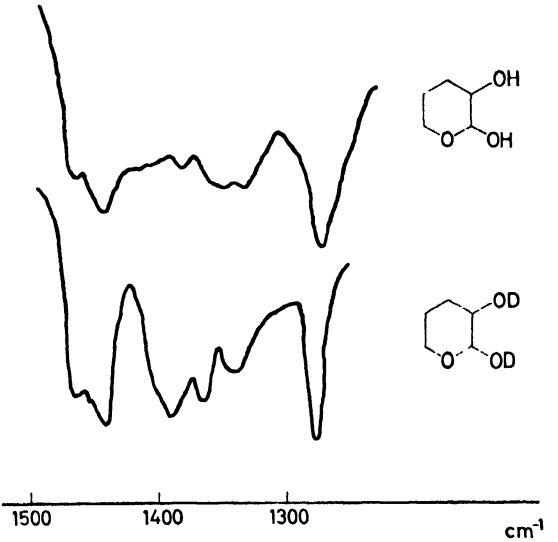


Figure 4. Spectrum of 2,3-dihydroxytetrahydropyran and the deuteroxy analogue in the 1200 to 1500 cm^{-1} region

Oligosaccharides

It is relatively easy to understand why the spectra of some neutral polysaccharides have been of little use for analytical or identification purposes. As was shown in the previous section, when one or more OH groups are attached to the pyran ring the C-H bending and O-H bending region suffers a considerable loss of resolution. The situation can be illustrated in the polysaccharide series by the spectrum of xylopentaose which is shown in the 1600 to 1200 cm^{-1} region in *Figure 5*. It is worth noting that the spectrum is identical to that of the "dry form" of the crystalline neutral xylan from esparto grass[7].

The spectrum of the original crystalline sample was recorded on a KBr disc. The band multiplicity and overlap is such as to make the spectrum useless in the 1500 to 1200 region. The situation is considerably improved by deuteration as shown in the same figure. The peak at 1465 is the expected CH_2 symmetric bending mode of the pyran ring and the blank in the spectrum from this region until the 1380 peaks confirms that this is indeed the region of strong O—H in-plane bending absorption. The shape of the band which fills in between the two peaks at 1465 and 1380 in the partially deuterated spectrum of *Figure 5* is probably characteristic of the O—H in-plane bending mode for low crystallinity polysaccharides in general, the breadth being due to the variety of environmental and hydrogen bonding possibilities.

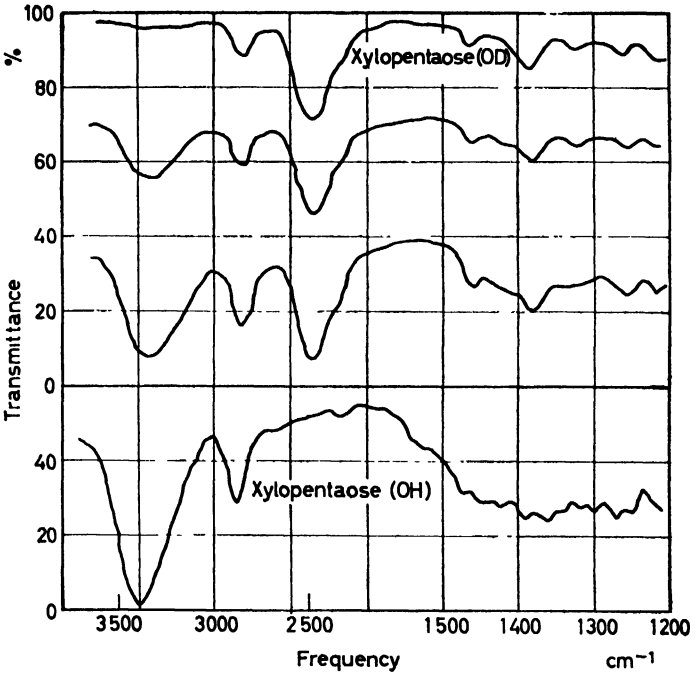
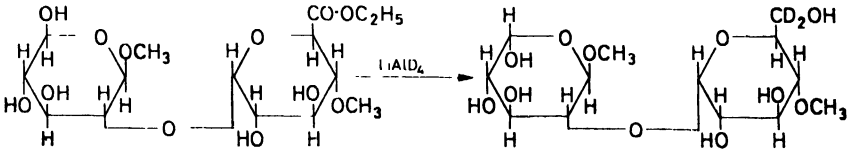


Figure 5. Spectrum of xylopentaose before and after treatment with deuterium oxide

The foregoing example illustrated one of the reasons why the oligosaccharides can be valuable aids in the interpretation of the polysaccharide spectra. The neutral xylan from esparto grass which is chemically equivalent to xylopentaose is considerably more difficult to deuterate. Another advantage of water-soluble oligosaccharides is that they are readily obtained in the completely amorphous form when freeze-dried from dilute aqueous solution.

The water solubility of oligosaccharides often allows chemical transformations which would be difficult or impossible with the parent polymer. For example, the transformations shown in equation (1) below:



could be readily performed on the aldobiuronic acid from xylan but only with difficulty and incompletely on the xylan itself. This transformation separated the methylene stretching frequencies due to the hydroxymethyl group of glucose from those of the xylopyranose ring. All these transformations are by way of establishing unequivocally the particular group responsible for a given absorption band.

Sometimes it is the property of high chemical purity combined with high crystallinity which makes a particular oligosaccharide valuable. The crystalline mannotetraose, for example, was particularly useful in establishing the exact positions of the absorption frequencies of the crystalline glucomannans which give poorly resolved spectra. Thus, only after deuteration (*cf. Figure 6*), does the infra-red spectrum of a crystalline mannan show the same resolution as that of the completely crystalline mannotetraose. Furthermore, deuteration of the mannan reveals an absorption band at 1600 indicating some impurity containing a carboxylic acid salt which is not present in the oligosaccharide. Finally, the contamination of the spectrum due to adsorbed water is almost negligible in the crystalline oligosaccharides. When using an oligosaccharide as a crystalline standard of a polysaccharide it is important that it should have the same unit cell as the parent polymer. This is the case for the regenerated mannan film and the crystalline mannotetraose used to obtain the spectrum shown in *Figure 6*.

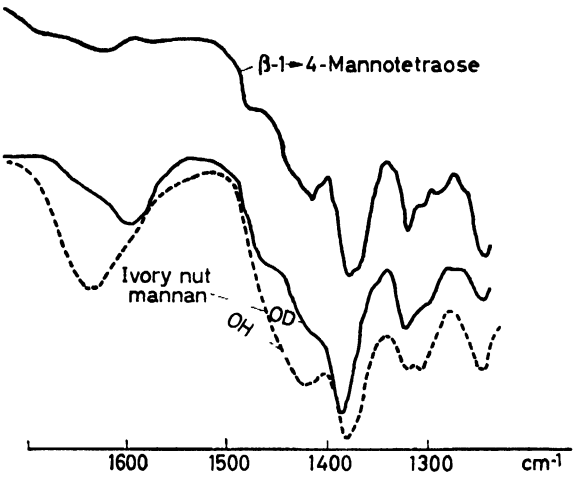


Figure 6. The spectrum of crystalline regenerated ivory nut mannan before and after deuteration compared to the spectrum of crystalline mannotetraose

Finally, if a particular oligosaccharide is not crystallizable and/or yields a rather ill-defined spectrum it is still sometimes possible to remedy the situation by conversion to the methyl derivative. *Figure 7* shows the spectra of methyl-2-*O*(4-*O*-methyl-α-D-glucopyranosyl)α,β-D-xylopyranoside and methyl-2-*O*(2,3,4-tri-*O*-methyl-α-D-glucopyranosyl)3-*O*-methyl-α,β-D-xylopyranoside in the 1500 to 1300 region. The effect of methylation is to remove the pronounced overlap of CH and OH bending modes. Since the CH_3 modes at 1450 and 1380 are easily identified, this treatment makes visible the shoulder at 1410 which most probably represents the methylene vibrations of the glucopyranosyl. The proper assignment of this band is important in the interpretation of the polarized spectrum of cellulose.

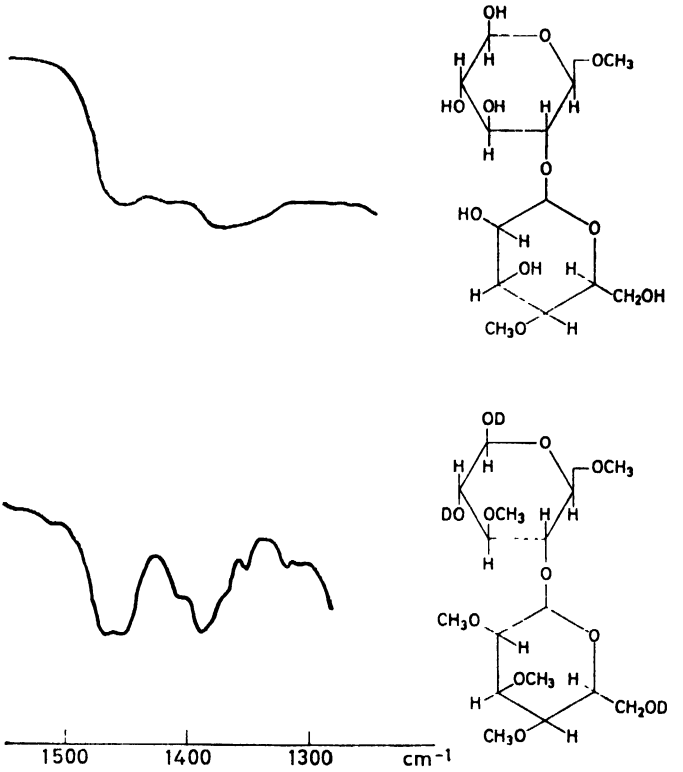


Figure 7. The spectra of methyl-2-*O*-(4-*O*-methyl-α-D-glucopyranosyl)α,β-D-xylopyranoside and methyl-2-*O*-(2,3,4-tri-*O*-methyl-α-D-glucopyranosyl) 3-*O*-methyl-α,β-D-xylopyranoside in the 1500 to 1300 cm^{-1}; the latter was treated with D_2O to convert residual OH to OD

Wood polysaccharides

We come now to the actual spectra of the polysaccharides which occur in wood. A list of the component sugars and the important polysaccharides which we may expect to encounter is shown in *Table 2.* From the table one deduces that a total of no less than six different sugars can be expected to contribute significantly to the spectrum of the holocellulose and, in the case of wood, a number of bands due to lignin must be added. A typical wood spectrum is shown in *Figure 8.* Obviously, if some use other than

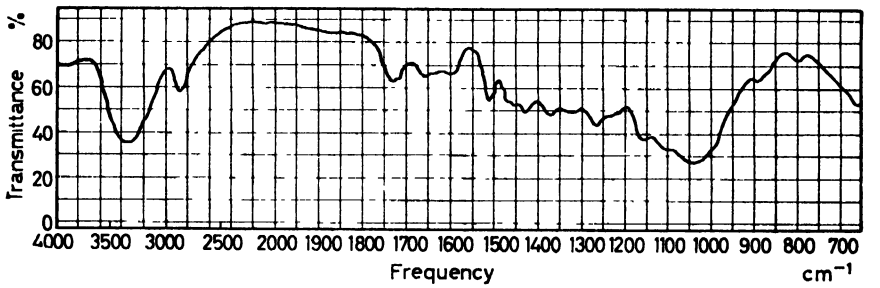


Figure 8 Infra-red spectrum of wood powder from white spruce in KBr disc

simple " fingerprinting " is to be derived from such a spectrum it will be necessary to study in detail the spectra of the component polysaccharide so that the absorption bands may be associated not only with a given component but with a given group in this component. Up to now two of the wood polysaccharides have been studied in detail: cellulose[8, 9] and xylan[10]. An outline of the semi-empirical approach which is used to make a full assignment of the absorption bands in a polymer has been given[3]. The following section will illustrate some aspects of the approach as it applies to wood polysaccharides.

Xylans—Xylans are one of the two most important hemicelluloses; they constitute about 25 to 33 per cent of the weight of hardwoods. To study

Table 2. Principal wood components and characteristic absorption bands in infra-red spectrum

Polymer	*Component monomer*	*Characteristic absorption* (cm^{-1})
Cellulose I	β-D-glucose	1425, 663
Xylan	β-D-xylose 4-*O*-methyl-α-D-glucuronic acid β-L-arabinose	890, 1735
Glucomannan	β-D-mannose β-D-glucose	805, 870
Water-soluble Polysaccharides	β-D-galactose β-L-arabinose	768
Lignin	Phenylpropane	1505, 1595

the spectrum of this material sample films were prepared by autoclaving aqueous slurries of alkali-extracted xylans to obtain solutions which were then air-dried on a glass plate. The chemical structure of a typical xylan is shown in *Figure 9.* The actual spectrum before and after deuteration is shown in *Figure 10.* The spectrum bears a good deal of similarity to that of cellulose as one might expect from the similarities between xylose and glucose. The 4-*O*-methyl glucuronic acid group leaves a characteristic

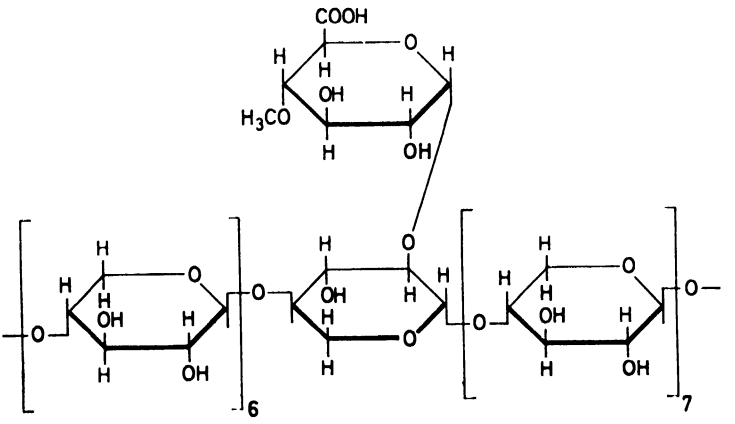


Figure 9. Chemical structure of 4-*O*-methylglucuronoxylan from milkweed

imprint on the spectrum contributing the strong absorption at 1600 and 1425. Since the moisture uptake of xylan is particularly large, the crystalline part itself being a hydrate[7], deuteration of all OH in the sample is both simple and valuable. From such a spectrum a fairly complete assignment of the CH and OH frequencies can be made[10]. Similar studies on other xylans from hard and softwoods have led to the same results despite small differences in chemical composition.

One may then ask what practical application can be made knowing the relation between a given absorption band and the chemical group from which it springs. The analytical applications are far from elegant at present. One can tell whether the carboxyl group is present as free acid or salt, depending on whether the absorption is at 1730 or 1600. In order to see the 1600 band clearly, deuteration is advisable. The intensity of these bands is related to the amount of carboxyl as may be judged from the relative intensities of the 1600 bands in *Figure 10.* Milkweed xylan is

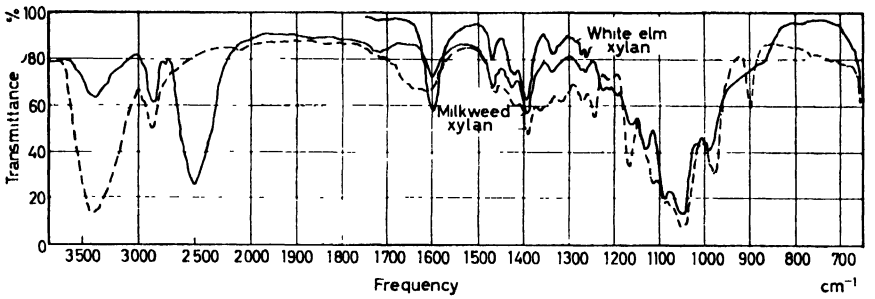


Figure 10. Infra-red spectra of 4-*O*-methylglucuronoxylan salts; deuterated —; — · — rehydrogenated

known[11] to have about half as much glucuronic acid/xylose as white elm. Acetylation is easily detected when both the 1730 and the 1250 bands are strong. These applications are all fairly obvious and it is discouraging to think that there is no spectral characteristic whereby one can even recognize the presence of arabinose groups, for example, which occur to fairly high percentage in softwood xylans.

Cellulose—Band assignments are greatly facilitated when a polarized spectrum of the sample is available. For such a spectrum a uniaxially oriented polymer sample is required. The spectrum is then recorded with the incident infra-red beam polarized: (*A*) parallel to the fibre axis and (*B*) perpendicular to the fibre axis. Bands which show greater absorption in one case than in the other are said to be dichroic. If absorption is greater in *A* than in *B* then the band is parallel polarized (π band). If the reverse is true then the band is perpendicularly polarized (σ band). The ratio of the two optical densities is called the dichroic ratio.

A polarized spectrum of native cellulose crystallites[9] in the C—H stretching region is shown in *Figure 11.* Remembering that the C—H bonds of each glucose residue are approximately at right angles to the axis of the cellulose molecule it is evident that the absorption bands corresponding to

these vibrational modes must show maximum absorption when the incident beam is polarized perpendicular to the fibre axis assuming there is cylindrical symmetry about the molecular axis. With this in mind we readily assign the central σ bands at about 2900 to the C—H stretching modes. The

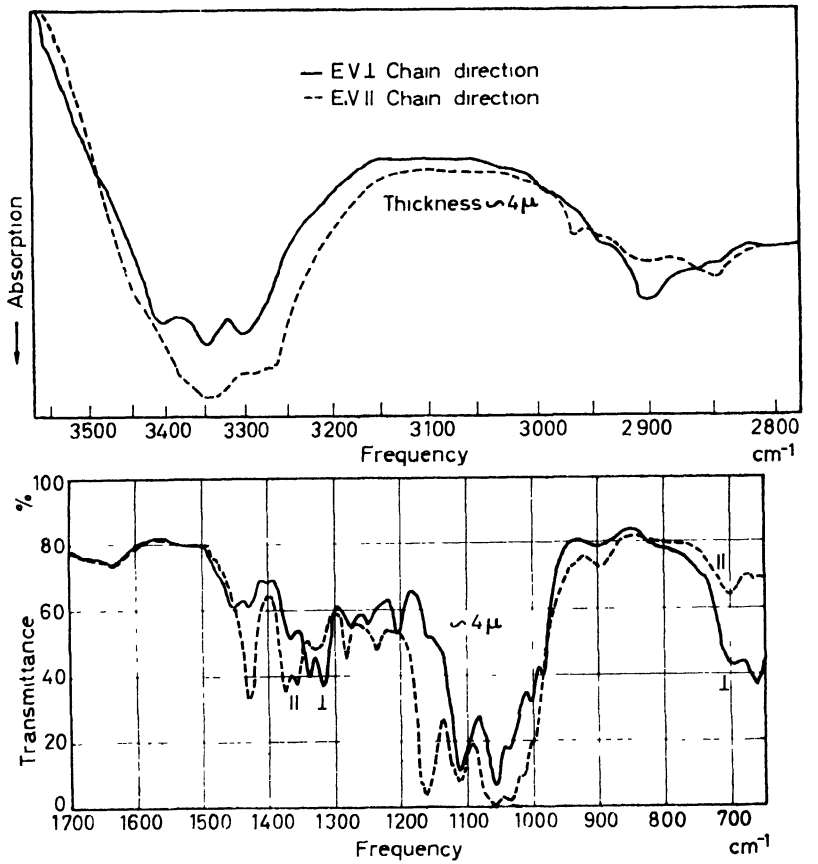


Figure 11. Polarized infra-red spectrum of ramie cellulose crystallites[9], the spectrum was recorded with a LiF prism in the 3μ region

assignment is further strengthened by our experience with the pyrans whereby substitution on the ring tended to increase absorption in the central part of the C—H stretching region.

In *Figure 11* one also recognizes absorption at the frequencies characteristic of the CH_2 symmetric and antisymmetric stretching modes. Since these frequencies are so characteristic of this grouping one can usually rely on this assignment. Having made the assignment it is now the observed dichroism which assumes significance. The parallel polarization of the CH_2 symmetric stretching band at 2853 requires a similar polarization for the CH_2 symmetric bending mode which should be found somewhere

between 1460 and 1400. In the polarized spectrum of native cellulose in the 7 to 15μ region, shown in *Figure 11*, the only parallel band in the stipulated region is at 1430. Since its intensity is not greatly affected by deuteration the assignment to the CH_2 symmetric bending mode is reasonable and consistent.

Other polysaccharides in wood—With this illustration of how a detailed study is made of the spectrum of a given polysaccharide we may briefly consider the other important wood components and compare them with one another. Such a comparison is made in *Figure 12* which includes

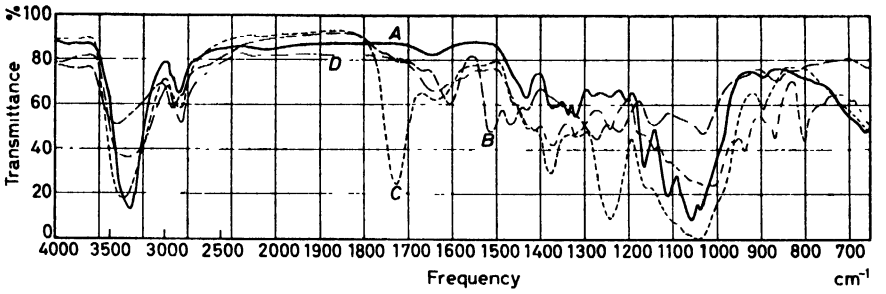


Figure 12. Infra-red spectra of (*A*) cellulose crystallites from prehydrolyzed cellulose wood pulp; (*B*) Klason lignin from red cedar; (*C*) *O*-acetyl-4-*O*-methylglucuronoxylan from white birch; (*D*) ivory nut mannan

spectra of an acidic *O*-acetyl xylan, wood cellulose crystallites, Klason lignin and ivory nut mannan. One notes the happy situation whereby the three non-cellulosic components have at least one strong band which occurs at a position corresponding to a " window " for all the others. This is fortunate for it means that acid xylan, mannan and lignin can be identified in the presence of the other wood components. The characteristic band for each polysaccharide is listed in *Table 2.*

Besides the ivory nut mannan spectrum, part of which is shown in *Figure 6*, the spectra of a series of glucomannan from hard and softwoods has been studied. These samples varied in their mannose glucose ratio from 20 to 3. However, except for the resolution of the spectra which decreased with increasing sugar heterogeneity there was no feature which was characteristic of the glucose component.

Included in *Table 2* is the arabogalactan polysaccharide which has been found characteristic of water-soluble wood polysaccharides from wood[13]. The particular sample examined was from Western Larch.

From a detailed study of the infra-red spectra of the various wood components as well as the spectra of both hard and softwoods it is possible to tabulate the principal absorption bands occurring in wood and make some proposals as to their origin. Such a compilation has been made in *Table 3.* It is very approximate, as it must be, since many of the absorption bands in the wood spectrum have contributions from several different components. The polarizations are the ones actually observed for wood sections[14].

Table 3. Infra-red spectrum of wood

Frequency (cm^{-1})	*Relative intensity*	*Polarization*	*Assignment*
3600–3200	s		H-bonded OH stretching
2970	sd		CH stretching
2945	sd	⊥	CH_2 antisymmetrical stretching
2914–2870	s	⊥	CH stretching
2850	m		CH_2 stretching
1730–1725	s	⊥ or u	C=O stretching of acetyl or carboxylic acid
1670	w	u	Lignin
1635	m	u	Adsorbed water
1600	s	u	COO^- ion
1595	m	u	Lignin
1500	m	u	Lignin
1460	m	⊥	Lignin and CH_2 symmetrical bending on pyran ring
1455–1400	m		OH in-plane bending
1430	s		CH_2 symmetrical bending mode of hydroxymethyl
1425	s		Carboxylic acid and COO^- vibration
1380	s		CH bending
1335–1315	w	⊥	CH_2 wagging
1270	m or sd		Lignin
1240	m		C–O of acetyl
1162	s		Antisymmetrical bridge oxygen stretching
1125–895	s		C–O stretching and ring vibrational modes
895	w		Characteristic of β-link
875	sd		Mode due to glucomannan
800	w	⊥	Mode due to glucomannan
768			Mode due to arabogalactan
700–650	m		OH out-of-plane bending

m medium, s. strong; sd. shoulder, w. weak

SPECTRA OF WOOD SECTIONS

The composite nature of the infra-red spectrum of wood is shown by the series of spectra in *Figure 13.* These data were collected by recording the spectra of wood sections lightly impregnated with Nujol[14] after the section had been given the treatments shown in *Table 4.* As is well known, the gross morphology of the wood is maintained after the listed treatments and it was also shown that Nujol affected principally the intensity of the 1465 band.

The chemical treatments in *Table 4* were essentially of two kinds: (*a*) delignification, (*b*) removal of hemicellulose. Reference to *Figure 12* allows us to identify almost every band in the wood spectra with either cellulose, xylan, glucomannan or lignin. Delignification produces the very characteristic removal of the absorption bands at 1595 and 1505 and of a weaker band at 1270. In both hard and softwood one is left, after delignification, with a broad band at 1240 which is characteristic of an acetylated polysaccharide. Hardwood xylan is known to be acetylated but this is a good

clue that either the mannan or xylan in softwood also carry acetyl groups[21]. In both softwood and hardwood the C=O peak due to xylan is readily removed by treatment with alkali. It is not legitimate to conclude, how-

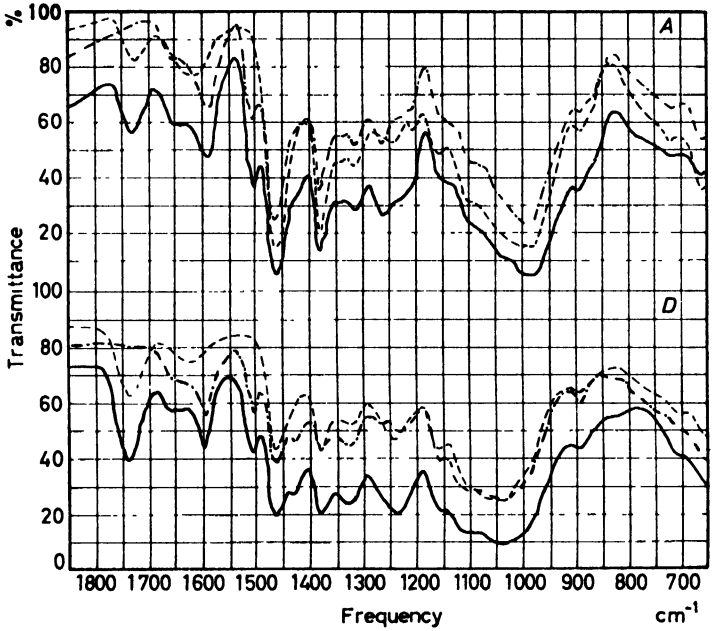


Figure 13. Infra-red spectra of wood cross-sections of western red cedar (*A*) and radial section of red maple (*D*) —: untreated; — — —: delignified; — — · —: treated for removal of hemicellulose, all sections contain Nujol

ever, that all carboxyl-containing polysaccharides have been removed unless the samples are given an acid (ion exchanging) wash prior to recording the spectrum since the alkali treatment also causes a shift in the carbonyl frequency, due to salt formation.

The spectrum of the untreated Western Red Cedar (C–1) shows absorption shoulders at 875 and 800 which is characteristic of mannan. These

Table 4. Treatment of wood sections whose spectra are recorded in *Figure 13*

Wood	*No.*	*Section*	*Thickness* (μ)	*Treatment*
Western Red Cedar	C–1	Cross	30	Untreated
	C-2	Cross	30	Delignified*
	C 5	Cross	30	Treated with 8% NaOH, 20 min; H_2O
Red Maple	M–2	Radial	20	Untreated
	M–4	Radial	20	Delignified*
	M 8	Radial	20	Treated with 22% KOH, 20 min, 0·1N HCl, H_2O

* Cross and Bevan procedure repeated twice

bands are somewhat weakened by the alkali treatment and are essentially absent in the spectra of hardwoods. They constitute, therefore, spectral characteristics for distinguishing between hard and softwoods. Another such characteristic is the shape of the absorption band from 1275 to 1200. Softwoods have their maxima at 1270 while hardwoods have theirs at 1240.

Obviously the foregoing data could have been recorded on powdered samples of wood mixed with potassium bromide although spectral resolution is superior with the wood sections. The real reason for working with wood sections was as a preliminary step to developing (hopefully) a micro-spectrophotometric method of studying the distribution of polysaccharides in the cell wall by direct spectroscopy. This goal is not yet achieved. (For a review of the situation with respect to infra-red microspectroscopy *cf.* ref. 15.) Wood radial sections have a special merit, independent of microspectroscopy, in that they provide an opportunity to answer the question of whether or not the wood hemicelluloses are oriented. *Figure 14* shows the polarized infra-red spectrum of a radial section from red maple and the same region recorded for an amorphous but oriented *O*-acetyl film of xylan from white birch. The similar polarizations for the two " xylan " bands, at 1725 and 1465, leaves little doubt that the xylan within the wood structure is oriented. The same conclusions can be made about the glucomannan fraction from the spectra of radial sections of Western Red Cedar and Douglas Fir[14]. This is an important piece of structural information for the wood chemist and is only obtainable *via* the infra-red approach.

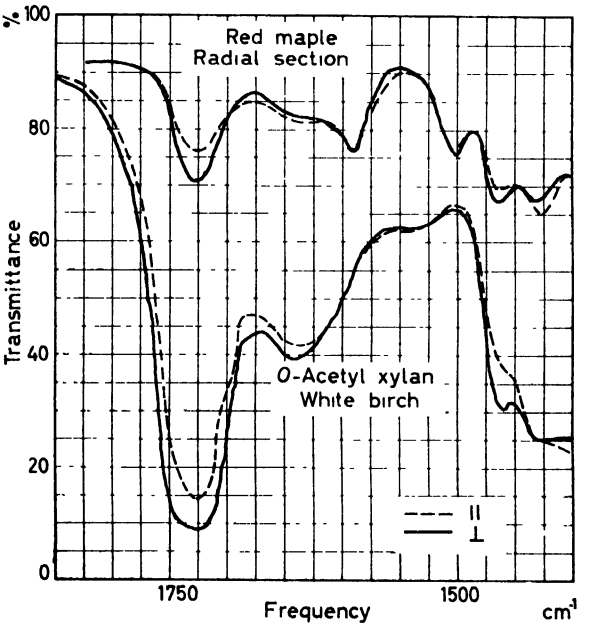


Figure 14. Polarized infra-red spectrum of radial wood section of Red Maple and oriented film of *O*-acetyl-4-*O*-methylglucuronoxylan from White Birch

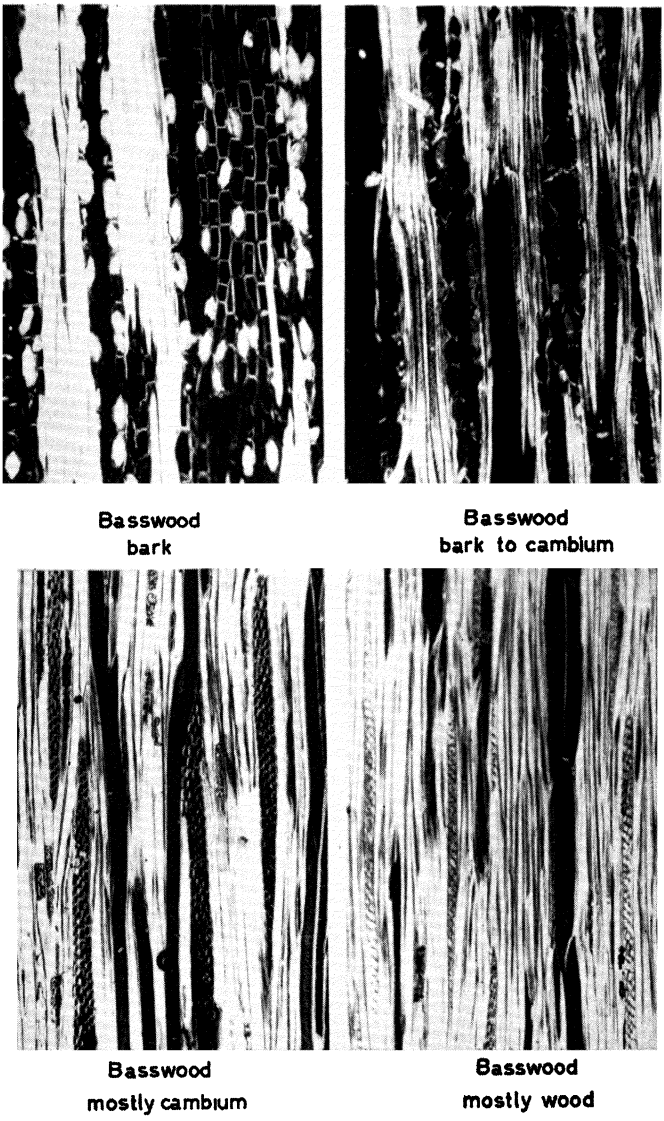


Figure 15. Photomicrographs of members of a series of tangential sections of basswood taken from bark - mature wood

The infra-red spectrum of a wood section can be a very valuable adjunct to microscopic examination of wood sections. For example, if the infra-red spectra of serial tangential sections of wood from the bark through the cambium are recorded, and at the same time photomicrographs are taken, an interesting correlation can be made between morphology and chemical composition. Examples of the type of data which might be obtained is illustrated for basswood sections (*tillia Americana*) and are shown in *Figures 15* and *16*.

The photomicrographs of *Figure 15* were taken between crossed Nicol prisms with the fibre axis at 45°. The interpretation of these photomicrographs depicting the transition from the bark area containing the hexagonal calcium oxalate crystals to the area of low birefringence constituting the cambial zone to, finally, the mature wood with maximum birefringence is well known. The transition represents the various stages of laying down carbohydrate and ligneous material for building up the cell and wood structure. Changes in the infra-red spectrum should reflect these additions to the primary cell wall and wood structure. Thus, the change in optical density of the carbonyl frequency at 1730, in *Figure 16*, can be taken as a

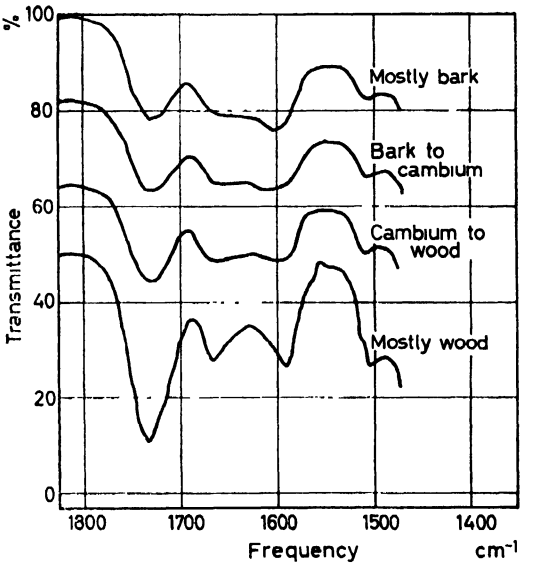


Figure 16. Infra-red spectra of representative spectra from a series of tangential sections of basswood

measure of the increasing density of uronide hemicellulose in the cell wall, since the thickness of each section was held constant. When the technique is perfected it will afford a convenient measure of wood constituents as a function of radial distance in the cambial zone which in turn can be correlated with the various cell wall layers visible in the microscope. It is doubtful if this instrumental technique can ever equal in precision the classical microchemical technique developed by Meier[16] but it does have the advantage of convenience.

MOLECULAR CONFORMATION AND HYDROGEN BONDING

So far we have touched only briefly on the more classical applications of infra-red spectroscopy to the polymer field. Most generally this approach has involved polarized spectra from which valuable information concerning the " backbone " conformation and orientation of pendant groups relative to the polymer " backbone " could be deduced. The latter application has been particularly fruitful in the case of cellulose where the classical X-ray approach has fixed the " backbone " conformation as that of a twofold screw but failed to provide the details of structure concerning hydrogen bonding and the arrangement of the hydroxymethyl group.

As stated above, the dichroism of the CH_2 symmetric stretching and bending modes observed[9] for doubly oriented films of cellulose I crystallites is parallel and that of the antisymmetric stretching is perpendicular. This means that the transition moment for the ν_s (CH_2), which is a vector bisecting the H—C—H bond angle, must make an angle less than 45° with the fibre axis. Examination of molecular models leads one to the conformation shown in *Figure 17* wherein the valence bond C_6–O_6 is *trans* to the C_5–C_4 bond. No other position is conformationally as stable and yields the correct dichroism. It is noteworthy that the conformation shown in the Newman diagram of *Figure 17* is that which would be predicted on the basis of simple conformational analysis assuming that hydrogen bonds would be of equal strength in any position. The position wherein C_6–O_6 is *trans* to C_5–O_5 is considered less stable than the one shown in *Figure 17* because of the additional conflict between O_6 and O_4 in this position analogous to a Peri conflict.

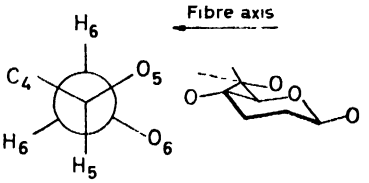


Figure 17. Schematic diagram showing orientation of CH_2OH in native cellulose relative to pyran ring as deduced from infra-red dichroism of methylene group

X-ray studies[17] lead to the conclusion that the latter position for O_6 best fits the data or possibly a combination of the two foregoing positions for O_6. There is, therefore, a net disagreement, at present, between the interpretation of the data from these two approaches. This is reflected in the various hydrogen bonding schemes which have been proposed for native cellulose[9, 17, 18] which will be discussed below.

The fixing of the hydroxymethyl group leads naturally to the question of hydrogen bonding in the cellulose I crystal. The polarized spectrum of ramie crystallites indicates the presence of five distinct hydrogen bonds, of which at least two show strong parallel dichroism *cf. Figure 11.* This means that any hydrogen bonding scheme for native cellulose must (*a*) be in agreement with the general tenets of the X-ray data, (*b*) have the correct CH_2 dichroism relative to the fibre axis and (*c*) have at least two hydrogen

bonds with a preferred orientation in the direction of the fibre axis (parallel hydrogen bonds). Two of the many proposed bonding schemes for cellulose I are shown in outline in *Figure 18.* The upper one is to be criticized on the grounds that it accounts for only a single parallel hydrogen bond. The lower scheme on the left does have two different parallel hydrogen bonds but would yield the incorrect dichroism for the CH_2

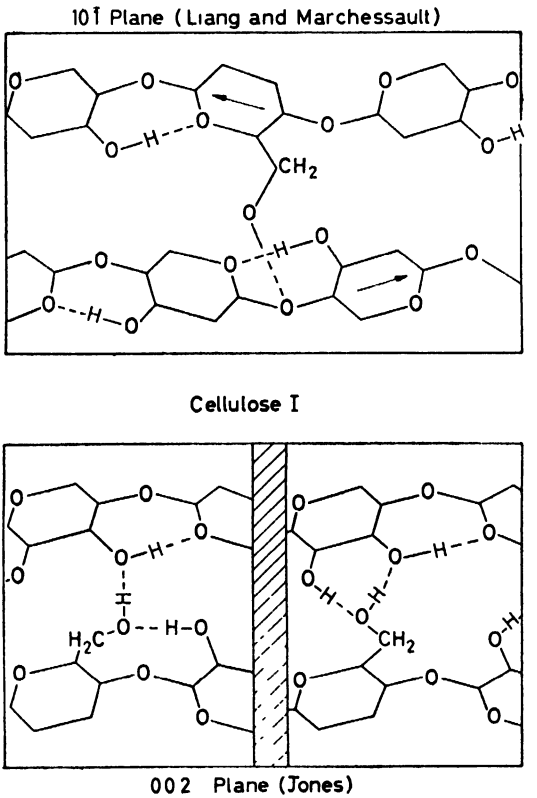


Figure 18. Three different hydrogen bonding schemes for native cellulose which encompass some of the requirements of the infra-red data and X-ray data

modes. The scheme on the right does have the required dichroism for the CH_2 modes and two different parallel hydrogen bonds (one of which is not included in the scheme as shown). It would appear, therefore, as the likeliest candidate to account for the hydrogen bonding in native cellulose. Unfortunately it gives very imperfect agreement with observed 0*k*0 X-ray intensities[17].

Although certainty still eludes us as to the hydrogen bonding scheme in native cellulose it is clear that considerable progress has been achieved by the alliance of spectroscopy and crystallography if only that the very important hypothesis of intramolecular hydrogen bonding has been given some experimental backing.

NEAR AND FAR INFRA-RED SPECTRUM OF CELLULOSE

Two important spectroscopic regions of the infra-red spectrum of cellulose have not received attention as yet. These regions, the " near " and " far " infra-red, are generally considered to range from 1 to 3μ and 15 to 40μ respectively. The " near " infra-red is the easier part of the spectrum to investigate and interpret since absorption is usually due to overtones and combination modes. Another reason for interest in this region of the spectrum is the fact that much thicker samples can be used than in the infra-red. This is the result of the weaker absorption usually shown by overtone and combination bands compared to fundamentals.

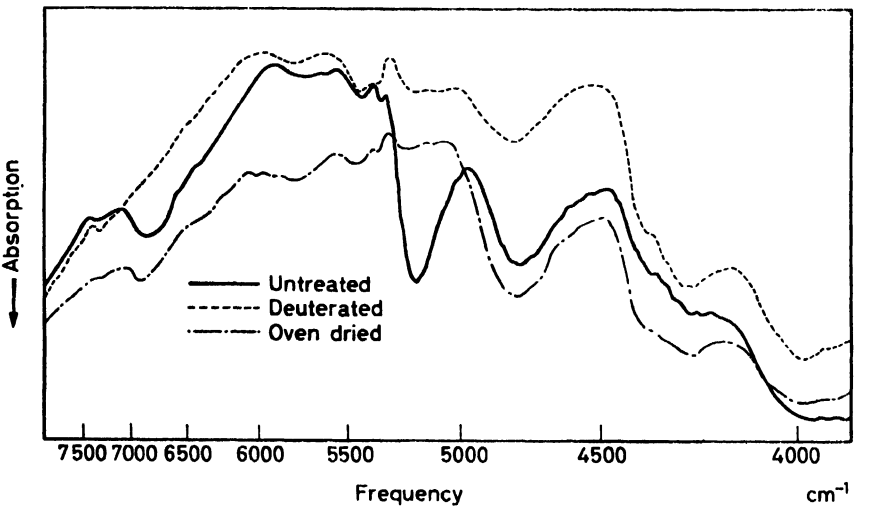


Figure 19. Near infra-red spectrum of an dry, oven dry and deuterated tyre cord rayon

Where 6 to 10 micron samples are required for the infra-red and the " far " infra-red it is possible to use $\frac{1}{4}$ inch fibre bundles in the " near " infra-red when a suitable immersion medium (CS_2 + CCl_4) is used.

Except for thep ioneering work on hydrogen bonding by Ellis and Bath[18], nothing has been published on the " near " infra-red spectrum of cellulose. *Figure 19* shows the spectrum in the " near " infra-red of conditioned tyre cord rayon, the dry material and finally the deuterated material[22]. It is evident that the 5190 band is exclusively due to bound water while the other bands appear to arise from the various combinations shown in *Table 5.* The dichroism was not pronounced for any of the bands when the polarized spectrum was recorded using a silver chloride plate polarizer. On the other hand, the 6770 band, which is the first overtone of the OH stretching fundamental, could be useful in hydrogen bonding[19] and derivative studies since this region of the spectrum is readily accessible on modern broad range ultra-violet spectrophotometers.

The " far " infra-red spectrum of polysaccharides contains a number of well defined bands with distinct dichroism. These bands probably arise

Table 5. Near infra-red spectrum of cellulose

Frequency (cm^{-1})	*Assignment*
3970 3990	CO stretching + CH and CH_2 stretching
4235	OH and CH deformational modes + CH and CH_2 stretching
4365	CO stretching + OH stretching CH_2 bending + CH_2 stretching
4560	Cellophane only
4780	OH and CH deformation modes + OH stretching
5190	Absorbed H_2O
5310–5480	Background
6770	2 × OH stretching from cellulose and absorbed H_2O
7265	Background

from various modes of the pyran ring and may prove useful in conformation studies.

Acknowledgement is made to Dr T. E. Timell who kindly provided many of the samples used in our studies. Figure 18 was provided by Dr H. Marrinan. The writer is indebted to his colleague Dr C. Y. Liang who has collaborated in most of these studies and to Professor W. Cote, of the State College of Forestry, Syracuse, New York, for assistance in the preparation and examination of the wood sections. Technical assistance in these studies was received from Mrs J. Watson, Miss Joan Koch and Miss P. Fay.

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CELLULOSE AS A VISCOELASTIC MATERIAL

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INTRODUCTION

The complex problem of viscoelasticity can be treated from a purely mathematical point of view. Gross[1] has given a concise summary on this type of analysis. Such an approach is not without merit, even for those whose interests transcend the mathematical techniques, since it clarifies our understanding of the behaviour of viscoelastic materials when subjected to various types of stress. However, the molecular interpretation of the mechanical properties of cellulose, and of all other materials, must remain the primary aim of physical chemists. To obtain such interpretations from the various phenomenological functions (*e.g.* the distribution of relaxation times) used to describe viscoelastic behaviour can be extremely difficult.

The viscoelasticity of cellulosic materials is known to be the product of many factors, interacting in complicated and, as yet, not completely understood ways; there is much evidence to indicate that these mechanical properties are strongly influenced, or even governed, by hydrogen bonds, particularly those bonds which exist in the amorphous regions of cellulosics. It is reasonable to expect that a molecular theory based on the *known* properties of the hydrogen bond would be a logical first approximation to the viscoelastic behaviour of cellulose. The authors believe that such an approach is intrinsically more desirable than an attempt to translate the somewhat abstract nature of a distribution of relaxation times into molecular reality. Nevertheless, some aspects of the phenomenological problem and their interpretation should always be borne in mind since any attempt to predict viscoelastic behaviour from a molecular model must not contradict these findings.

Most attempts to treat the viscoelastic behaviour of cellulose quantitatively have been primarily concerned with the phenomenological aspects of the problem. It is well known that a single Maxwell element cannot describe, adequately, the relaxation of cellulose. Thus, many investigators have been concerned with the problem of obtaining a spectrum of relaxation times for cellulosic materials. Such an approach is tantamount to assuming that a large or infinite number of relaxation units exist in cellulose, each with a characteristic relaxation time. For example, Passaglia and Koppehele[2] used a modified, strain-dependent distribution function of relaxation times in order to solve the problem of non-linear viscoelastic behaviour exhibited by isotropic and oriented cellulose filaments.

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The effects of humidity and frequency on the dynamic mechanical properties of viscose rayon have been studied by Price, McIntyre, Pattison, and Dunell[3]. These investigators found that the dynamic moduli of rayons were constant with frequency (5–100 c/s) at constant relative humidity, while the dynamic viscosities varied with frequency, but were unaffected by changes in relative humidity. Theoretically, it is possible to relate dynamic moduli and viscosities to a distribution of relaxation times, but the non-linearity of cellulose viscoelasticity makes such an interpretation of doubtful significance.

If we choose to explain cellulose viscoelasticity in terms of the phenomenological theory, then we must explain the non-linear effects observed experimentally. Such effects can be interpreted in two ways: (i) cellulosics *cannot* be considered as composed of linear springs and Newtonian dashpots; (ii) if the linear element model is to be used, then we must explain the non-linear effects as due to a continuous variation in the distribution of relaxation times induced by structural changes that occur on straining cellulose. There does not appear to be, at present, any means by which to distinguish between the two interpretations given above.

A notable attempt to describe the non-linear behaviour of polymeric and fibrous systems in terms of a semi-molecular model is that by Eyring and Halsey[4]. They propose a model consisting of two Hookean springs in combination with a viscous element obeying the Eyring hyperbolic-sine flow law. This model has been successful in describing viscoelastic phenomena in cellulosic systems (see Wakeham[5], and also Meredith[6]). However, there is still a phenomenological point of view involved in the Eyring and Halsey model, namely, the Hookean springs, and also in the analytical separation of *elastic elements* from a *viscous element.*

MORE DEFINITELY MOLECULAR APPROACHES

Macroscopic deformation of cellulose may involve several of the following microscopic deformations: valence bond length and angle deformation, secondary bond deformation, re-orientation of macromolecules in amorphous regions, re-orientation of crystalline regions, and configurational entropy effects. Generally, when discussing the effects of these molecular processes on mechanical properties, it is convenient to classify them in somewhat arbitrary ways and consider gross changes in these properties rather than small variations with strain. Thus, the over-all effects of crystallinity, orientation (or stretch), side group additions, degree of polymerization, *etc.*, are used to classify the behaviour of cellulosic materials, bearing in mind that one or several of these properties may be changing during the straining process. It is, therefore, not always possible to assign an unequivocal value to all of these factors for a material undergoing strain. Meredith[6] has summarized the mechanical properties of natural and regenerated cellulose and discusses various effects which supermolecular structure have on these properties, as do Tripp, Orr, Ziifle, and Conrad[7] for native and chemically modified cottons.

Many investigators of the mechanical properties of cellulose and its derivatives have recognized the importance of the hydrogen bonds which exist in cellulose and of their profound effect on the behaviour of cellulosic materials.

A study of the infra-red region of dry cellulose by Marrinan and Mann[8] indicates that there are *no* free hydroxyl groups in cellulosic materials. Since each cellobiose unit is capable of forming six hydrogen bonds, it seems reasonable to postulate that any viscoelastic phenomenon is accompanied by a breakdown, re-arrangement, bending, and stretching of these hydrogen bonds. There is ample evidence to support this belief.

In a summary of the mechanical properties of cellulose, Wakeham[5] has pointed out that the hydrogen-bonding capabilities of cellulose play an important rôle in determining the properties of cellulose. Thus, the adsorption of water by cellulose leads to a marked decrease in the dynamic elastic modulus.

The effect of temperature on the elastic modulus of wet rayon is due to the number and lifetime of crosslinks which exist between cellulose chains, as postulated by Roseveare and Poore[9]. Rubber elasticity theory cannot account fully for the entropy decrease which occurs on stretching wet and room air-conditioned (50 per cent R.H.) rayon because of the observed decrease of modulus of elasticity with temperature. Roseveare and Poore postulate that the entropy decrease may be due to the increase in equilibrium moisture content which occurs on stretching rayon, as reported by Treloar[10].

It should be noted that Gabrail and Prins[11] have analysed force, temperature, and length data for rayon, using the rigorous forms of the various thermodynamic relationships involved. These investigators point out that correction of constant pressure data to constant volume is of more importance than generally considered, and that energy effects on the stresses which are observed on stretching rayon are by no means negligible, even in the wet state. These authors suggest that the short range elasticity of dry cellulose may be due to interchain hydrogen bonding.

The effects of temperature on degree of swelling, extension at break, breaking load, and relaxation of wet viscose rayon have been studied by Clark and Preston[12]. These investigators found that the variation of these properties with temperature changes markedly in the range 50–60°. Clark and Preston postulate that these changes are the net result of several reaction mechanisms, namely, the dissociation of cellulose–cellulose crosslinks, the formation of cellulose–water bonds, and the dissociation of cellulose–water bonds; each of these reactions has a characteristic reaction rate.

The effects of humidity, crystallization, and drawing on the dynamic mechanical properties of fibres have been studied by Tokita[13], who found that the disperson of the loss factor for rayon shifted to lower temperatures with increase in humidity. Tokita suggests that these dispersions originate in the amorphous regions since these regions are most sensitive to humidity changes. It was also noted that increased drawing tended to raise the dynamic moduli, probably because of orientation effects, while the loss factor remained independent of degree of crystallinity and of orientation.

The dispersion of mechanical properties of viscose rayon at low temperatures (0 to −80°) in the frequency range 8–80 c/s has been studied by Dunell and Price[14]. They found that the dynamic modulus and loss factor are independent of frequency in this range of frequencies. From the maximum slopes of the dynamic modulus and loss factor *v.* temperature

curves, the activation energies for these viscoelastic parameters were calculated to be 400 and 1100 cal/mole, respectively. Dunell and Price note that Mark's estimate of the molar cohesion energy for the cellulose chain is 6200 cal/mole per 5 Å of chain length, based on a co-ordination number of 4 for each chain. Thus, they conclude that the low activation energies found for viscose rayon suggest that the relaxation unit is a single glucose unit in the amorphous regions with co-ordination number much less than 4, or a CH_2OH side chain whose flow motion does not involve the breaking of a hydrogen bond.

The effects of degree of polymerization and molecular orientation on the ultimate mechanical properties of rayon monofilaments have been studied comprehensively by Cumberbirch and Harland[15]. They explain the increase of tenacity with degree of polymerization, at constant orientation (measured by birefringence), in terms of the fringe-micellar theory. It is postulated that, as the degree of polymerization increases, the probability that a given cellulose chain will connect two crystalline regions increases. Cumberbirch and Harland showed that the tenacity approaches a nearly constant value as the degree of polymerization increases beyond a certain value. Also, the increase of tenacity and the increased ratio of wet to dry strength due to higher orientation, at constant degree of polymerization, is considered to be due to an increase in the number of hydrogen bonds in the amorphous regions and in the number of minor crystallites formed. In a later paper Cumberbirch[16] has investigated the effects of distribution of degree of polymerization on the mechanical properties of rayon monofilaments, and finds that a simple average D.P. is not sufficient to characterize the fibre.

Cumberbirch and Mack[17] have investigated the tenacity and breaking extension of cellulose monofilaments and have developed a quantitative theory based on the concepts of statistical chain configurations. It is postulated that in the completely wet state the cellulose chains connecting crystallites are mobile due to the adsorption of water on the hydrogen bonding sites of the glucose units. The tenacity of the filament is then calculated in terms of the rupture energy of a cellulose chain and the number and distribution of chain lengths connecting adjacent crystallites.

The tenacity of wet and dry filaments correlate with degree of polymerization, as predicted by the Cumberbirch and Mack theory. The explanation given is that the increased tenacity of dry filaments, which was found to be greater by a constant amount than the tenacity of the wet filament with the same degree of polymerization, is due to the energy required to rupture the hydrogen bonds existing in the amorphous regions. Cumberbirch and Mack postulate that the degree of hydrogen bonding may be nearly independent of degree of polymerization, at constant orientation, as indicated by the constant difference of dry and wet tenacity with degree of polymerization.

The various relationships which exist between humidity (*i.e.* accessibility) and relaxation, breaking strength and length, dynamic modulus, stress-strain curve, *etc.*, indicate that the hydrogen bond is of even more importance in determining the viscoelasticity of cellulose than was previously considered. In this connection it is interesting to note that there is a positive

increase in the entropic force component with water content for various cellulosic materials, as reported in a summary by Wakeham[5]. The fact that the modulus of elasticity decreases with temperature, and also that the entropic force component remains negative and smaller in value than the energy force component, leads to the belief that even in the wet state, cellulose deformation is primarily controlled by inter- and intra-chain forces of energic origin. If this is a valid deduction then some modification may be necessary in the finer details of Cumberbirch and Mack's theory. The principal conclusions, however, appear to contribute a major advance in the understanding of the problem.

H. Krässig and W. Kitchen[18] discuss quantitatively the simultaneous influences of DP, of DP_L (the effective length of the morphological units expressed in number of glucose units), of crystallinity index and of an orientation parameter on the tensile strength of wet and air conditioned yarns and fibres. They demonstrate effectively that in previous work it was a mistaken assumption that these parameters could be changed singly: changing one inevitably changed the others. By considering them simultaneously, the true relations became clear.

It will be noticed that in both of these extensive and quantitatively checked works—by Cumberbirch and Mack[17], and by Krässig and Kitchen[18]—the morphological units are of great significance to the ultimate properties of the fibre. It is no criticism of this approach to state that it should prove of some interest and possible value to look at even a lower level than the interconnections of the morphological units, *i.e.* to look at individual intermolecular bonds as the ultimate basis of mechanical properties. While the proper approach to a bridge design is in terms of the strengths, numbers, and geometrical relationships of the struts, beams and other morphological units which, in the aggregate, make up the bridge, it is still profitable to look at the crystallography and molecular arrangements which give the struts their peculiar properties. This is particularly true if we do not concentrate our attention on ultimate properties, but endeavour to learn the mechanism of elastic and plastic yielding at small strains.

Cellulosic materials are governed by a number of bond types: covalent, hydrogen, Van der Waals' and possibly ionic. However, if we choose a region of rigidity and yielding which is neither too high nor too low, and if we select materials which are not too highly crystalline nor too well oriented, *i.e.* if we limit ourselves to regenerated cellulose of low draw-ratios and to paper, the H-bond appears to predominate.

THE APPROACH THROUGH THE H-BOND

There is a large amount of evidence which indicates that the hydrogen bond may be the primary parameter governing the viscoelastic behaviour of cellulosic systems. Hence it appears justified to choose a *specific* model based on the properties of this bond as the logical, molecular starting point from which to elucidate the viscoelastic behaviour of cellulose. Undoubedly, in dry regenerated cellulose with high amorphous ratio, and in strong paper, the hydrogen bond is of greater significance than in weak paper, where Van der Waals' forces are significant, or in highly crystalline or oriented

cellulose such as cotton, flax, and ramie where valence bonds are a major factor. It is likely that the hydrogen bond is of varying significance even in regenerated cellulose, depending on the degree of crystallinity, orientation, and regain. Nevertheless, it will be instructive to consider the viscoelastic dictates of a system which is completely controlled by hydrogen bonding effects, realizing that such a system is probably a first approximation to reality for some cellulosic materials, while for other cellulosics (*e.g.* regenerated cellulose) it may prove to be a very good approximation.

In a first approach to the hydrogen bond problem, Nissan[19, 20] made several simplifying assumptions to arrive at a working model for the viscoelastic properties of hydrogen-bonded solids. Nissan assumed that (*a*) the macroscopic body was isotropic and non-interacting, *i.e.* that one-third of the hydrogen bonds in the body acted along each of three principal axes, (*b*) no re-orientation of bonds occurred as a result of straining, (*c*) configurational entropy effects could be neglected.

Nissan assumed that the potential energy (U) of the hydrogen bond could be described adequately by the Morse Function*

$$U = \frac{\Delta H}{N}\left[1 - \exp\left(-a\right)(r - r_e)/r_e\right]^2 \qquad (1)$$

where ΔH is the heat of dissociation of one mole of hydrogen bonds, N is Avogadro's number, a is a constant obtainable from spectroscopic data, and r and r_e are the H··O distances at energy U and at equilibrium, respectively. The force on a hydrogen bond is given by dU/dr, and this force acts on an area l^2 where $3/l^3$ is the *effective* hydrogen bond density, *i.e.* the number of hydrogen bonds being strained per unit volume. (The details of the derivation have been presented in a paper by Sternstein and Nissan[21] and only the results will be given here.) The final equation relating the macroscopic, unidirectional stress (σ) to the macroscopic strain (ϵ) is as follows:

$$\frac{Nr_e l^2 \sigma}{2a\Delta H} = \left[1 - \exp\left(\frac{-al\epsilon}{r_0}\right)\right]\left[\exp\left(\frac{-al\epsilon}{r_0}\right)\right] \qquad (2)$$

where r_0 is equal to r_e $(1 + K_2/K_1)$ and K_2 and K_1 are the force constants of the H··O and O—H bonds, respectively. Equation (2) is plotted in *Figure 1* and labelled " one-dimensional theory ". Before considering the applications and interpretation of the hydrogen bond theory as applied to stress-relaxation, it will be useful to remove one of the limiting assumptions made by Nissan.

In any real body, the hydrogen bonds of the system will exist in various orientations with respect to the principal axes. If a unidirectional stress is applied to the body, then some distribution of strains (or stresses) will exist among the hydrogen bonds of the system. We shall refer to this effect as " interaction ". In a treatment of the interaction effect for isotropic systems[21, 22], Sternstein and Nissan have deduced the stress–strain law for hydrogen-bonded networks which do not exhibit re-orientation or

* See p. 145 for full nomenclature.

configurational entropy effects. The problem was couched in terms of generalized stress–energy relationships, and the distribution of stresses was obtained by using the *Principle of Least Work* in conjunction with the calculus of variations. The resultant stress-strain law for unidirectional *applied* stress is shown in *Figure 1*, and is labelled " three-dimensional theory". It is evident that the effect of interaction is to increase the stress for any given strain; the increase in stress is due to the increased energy stored in the system due to the distribution of stress among the randomly-oriented hydrogen bonds. For example, those bonds at right angles to the direction of macroscopic strain will be in compression—the energy associated with these bonds is neglected in the one-dimensional theory.

The three-dimensional theory is not an *ad hoc* addition to the hydrogen bond theory but rather, as the authors believe it to be, the solution to the hydrogen bond network problem within the stated limitations of *isotropy*, absence of re-orientation of bonds and configurational entropy effects. The hydrogen bond theory has been checked against experimental evidence in

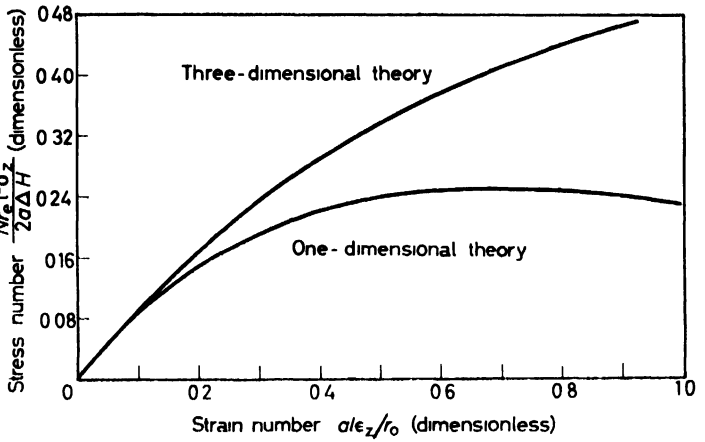


Figure 1

a number of ways and the reader is referred to other papers[19-22] for a review of these verifications. It is our purpose here to demonstrate the application of the theory to stress relaxation as an example of viscoelasticity.

In view of the effect hydrogen bonding is believed to have on the viscoelasticity of cellulose, it is plausible to expect that a time-dependent phenomenon, such as stress-relaxation, can be studied by following the process of hydrogen bond breakdown. Thus, a strained solid will undergo a time-dependent change in the *effective* number of hydrogen bonds which results in a decay of stress; those hydrogen bonds which reform do so in a state of no stress. This interpretation is not in disagreement with the Eyring treatment if one accepts the postulate that movement of a flow unit must involve the destruction of one or several hydrogen bonds, and that there is a fundamental relationship between the number and rate of flow of these units and the number and rate of breakdown of effective hydrogen bonds in the system.

A divergence of opinion occurs when one compares the basic hypothesis presented here with the accepted principles of phenomenological viscoelasticity. We postulate that the primary elastic elements in the system are the hydrogen bonds in the amorphous regions; hence, the elastic elements *cannot* be considered Hookean in nature. Secondly, we need not postulate the existence of separate viscous elements to account for time-dependent phenomena but, instead, we consider the possibility that a change in the number of elastic elements can occur with time.

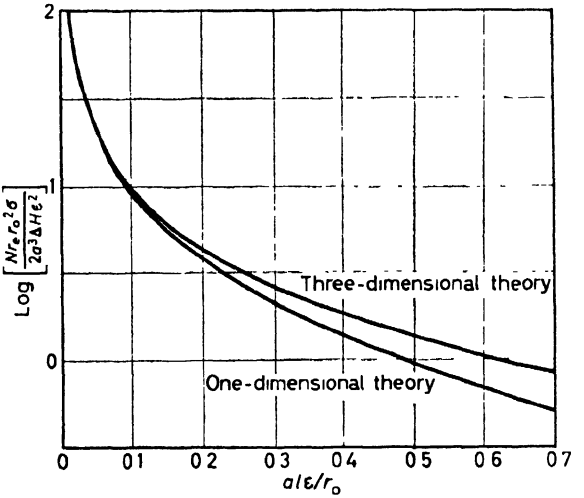


Figure 2

For convenience in dealing with stress relaxation we shall define a dimensionless quantity called the relaxation number (Σ) which is defined as follows:

$$\Sigma = \frac{Nr_e l^2 \sigma}{2a\Delta H}\left(\frac{r_0}{al\epsilon}\right)^2 \tag{3}$$

$$\Sigma = \frac{3\sigma}{2K\epsilon^2} \tag{3a}$$

where

$$K = \frac{3a^3\Delta H}{Nr_e r_0^2} \tag{3b}$$

The relaxation number is plotted *vs.* strain number ($al\epsilon/r_0$) in *Figure 2* for the one and three-dimensional theories. In a stress-relaxation experiment the relaxation number varies because of changes in l and we have the following relationship:

$$\Sigma/\Sigma_0 = \sigma/\sigma_0 = f(al\epsilon/r_0)/f(al_0\epsilon/r_0) \tag{4}$$

where f is the functional relationship indicated graphically in *Figure 2*. Making use of known values of the molecular constants in equations (3) and (4), it is possible to construct a plot of *effective* hydrogen bond density

($n = 3/l^3$) *v.* time, given a stress-relaxation curve. From this plot, a molecular interpretation of the stress-relaxation process is obtained.

A stress–relaxation curve for rayon at 25° and 65 per cent R.H. over a time interval 10^{-1} to 10^{16} sec has been obtained by Meredith[6] by use of the superposition principle. This curve has been transformed to a plot of hydrogen-bond density *v.* time by Nissan[19], with the use of the one-dimensional theory. Nissan's results (in slightly modified form) are plotted

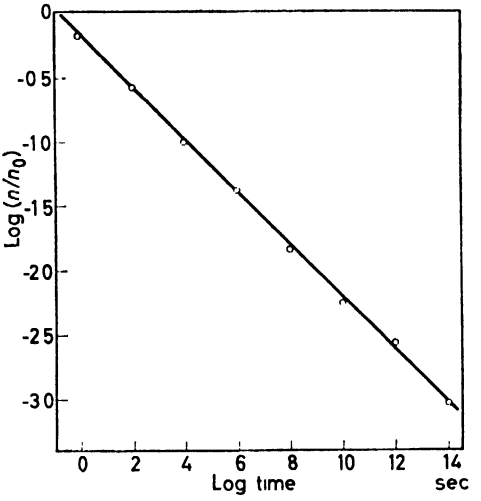


Figure 3

in *Figure 3* as log (n/n_0) *v.* log t where n_0 is the effective hydrogen bond density at time zero, and t is time. To analyse *Figure 3*, we proceed as follows:

If we assume that a co-operative phenomenon is occurring whereby α hydrogen bonds must break simultaneously, or co-operatively, in order for a relaxation unit to pass from a stress supporting state to an unstressed state, then

$$-\frac{\mathrm{d}(n/n_0)}{\mathrm{d}t} = k(n/n_0)^{\alpha} \tag{5}$$

where k is a reaction rate constant and α is a co-operative index. Integration of equation (5) gives

$$(n/n_0)^{1-\alpha} - 1 = k(\alpha - 1)t \tag{5a}$$

The co-operative index α is assumed to be greater than unity; hence, for all values of n/n_0 less than a particular value, the following condition is fulfilled (note that $0 \leqslant n/n_0 \leqslant 1$):

$$(n/n_0)^{1-\alpha} \gg \text{unity} \tag{5b}$$

Equation (5a) may then be written:

$$\frac{\mathrm{d}\log(n/n_0)}{\mathrm{d}\log t} = \frac{1}{1-\alpha} = \beta \text{ (a constant)} \qquad (5c)$$

Thus, we see that Nissan's results obey equation (5c), and since $\beta = -0{\cdot}2$, we calculate α to be 6·0. A co-operative index of six is not unacceptable, but is in fact corroborated by other considerations. A study of regain phenomena, in terms of the hydrogen bond theory, led Nissan[19] to conclude that, at 65 per cent R.H., 5·8 hydrogen bonds, on the average, are grouped together in a structural unit. Thus, the regain and relaxation mechanisms predicted by the theory are in agreement as to the structure of the amorphous, or low order, regions in regenerated cellulose.

It should also be noted that Burleigh and Wakeham[23] have analysed stress–relaxation data for cellulose in terms of the Eyring and Halsey model. Assuming that the average distance through which a flow unit moves was equal to the length of a cellobiose unit, Burleigh and Wakeham were able to obtain reasonable results for the number of flow units per unit cross section of sample. Since a cellobiose unit is capable of forming *six* hydrogen bonds, the results obtained by Nissan appear to be compatible with these findings.

The interesting results obtained by Nissan have prompted further investigation by the authors along similar paths. Stress–relaxation experiments were carried out on regenerated cellulose (cellophane) at various temperatures under dry conditions (dew point *ca.* — 100°F). The experiments were performed on an Instron tester specially fitted with a sample conditioning chamber through which dry air at the desired temperature could be passed. The stress–relaxation experiments were performed on samples 1 inch wide and 7 inches long cut on a precision sample cutter to ensure smooth and parallel edges. The samples were mechanically conditioned by cyclic loading and stress–relaxation under dry conditions at the highest temperature at which they were to be tested. Some of the experimental results are shown in *Figure 4*, for a sample which was mechanically conditioned at 41°.

The stress–relaxation curve can be converted to a curve of n/n_0 *v.* time by means of equations (3) and (4), using the measured values of

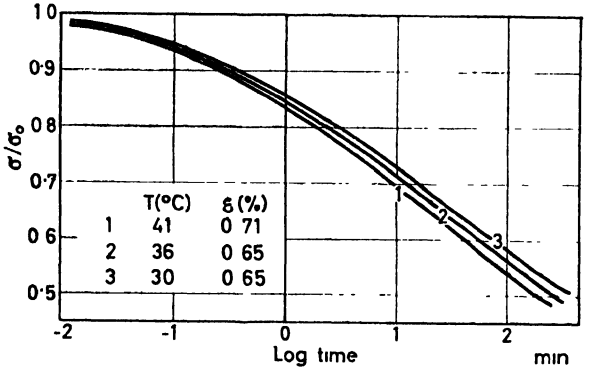


Figure 4

initial stress (σ_0), strain (ϵ), and a value of K, the relaxation constant, equal to $1{\cdot}1 \times 10^{12}$ dynes/cm^2. This value of K is the same as the value for the " second coefficient of elasticity ", as defined by Nissan[19]. Thus, for an initial stress of approximately 6×10^8 dynes/cm^2, Σ_0 is *ca.* 16. Inspection of *Figure 2* reveals that the one- and three-dimensional theories are indistinguishable for this value of Σ; hence, the following results are not dependent on the model used. For further discussion of the significance of the one- and three-dimensional theories at large strains, the reader is referred to papers by Sternstein and Nissan[21], and Sternstein[22].

The results of the calculations for experiment 1 are shown in *Figure 5*; it is seen that the rate equation given by equation (5c) is not applicable for

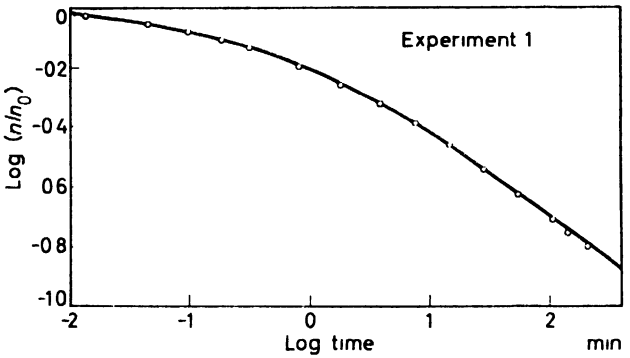


Figure 5

n/n_0 values near unity as can also be seen by inspection of equation (5c) for values of time approaching zero. A correct test of the applicability of the postulated rate mechanism, equation (5), is given by a plot of equation (5a) on logarithmic co-ordinates. This test is shown in *Figure 6* and it is seen that for a value of α — 3, a straight line is obtained, but *not* of unit slope as predicted by equation (5a). (*Figure 6* also shows the effect on n/n_0 values as calculated by the one- and three-dimensional theories.)

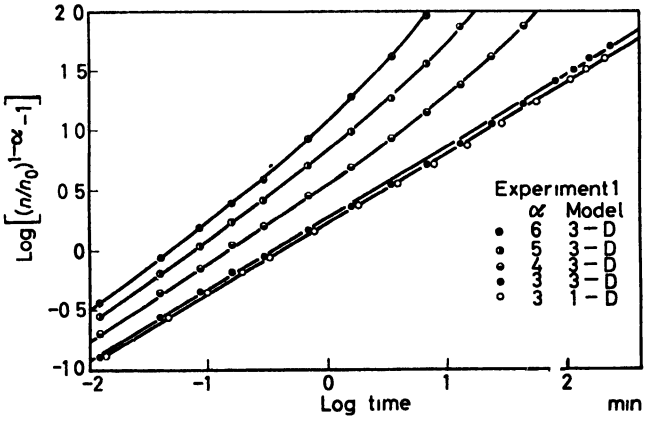


Figure 6

Thus, from *Figure 6*, we have that

$$\frac{\mathrm{d}\log[(n/n_0)^{1-\alpha}-1]}{\mathrm{d}\log t} = m = 0{\cdot}59 \tag{6}$$

Equation (6) can be written as follows:

$$-\frac{\mathrm{d}(n/n_0)}{\mathrm{d}t} = k'(n/n_0)^{\alpha}[(n/n_0)^{1-\alpha}-1]^{\frac{m-1}{m}} \tag{7}$$

where

$$k' = m[(n/n_0)^{1-\alpha}-1]_1^{1/m}/(\alpha-1)$$

and $[(n/n_0)^{1-\alpha}-1]_1$ is to be evaluated at $t =$ unity.

Equation (7) reduces to equation (5) if $m =$ unity. Expanding the above equation in a Taylor series gives

$$\begin{aligned}-\frac{\mathrm{d}(n/n_0)}{\mathrm{d}t} = k'\Bigg\{&(n/n_0)^{\alpha_1} + \frac{1-m}{m}(n/n_0)^{\alpha_2} \\ &+ \frac{1-m}{2!m^2}(n/n_0)^{\alpha_3} + \frac{(1-m)(1+m)}{3!m^3}(n/n_0)^{\alpha_4} \\ &+ \cdots + \frac{(1-m)\prod_{q=0}^{p-2}(1+qm)}{m^p p!}(n/n_0)^{\alpha_{p+1}}\Bigg\}\end{aligned} \tag{8}$$

where

$$\alpha_1 = \frac{m+\alpha-1}{m}, \qquad \alpha_2 = \frac{m\alpha+\alpha-1}{m},$$

$$\alpha_3 = \frac{m(2\alpha-1)+\alpha-1}{m}, \qquad \alpha_4 = \frac{m(3\alpha-2)+\alpha-1}{m},$$

and

$$\alpha_{p+1} = \frac{m(\alpha-p+1)+\alpha-1}{m}$$

The experimental results indicated in *Figure 4* are plotted according to equation (6) in *Figure 7*, and it is seen that a value of $\alpha = 3$ and a slope $m = 0{\cdot}59$ describe all the experimental results reasonably well. Thus the "multiple index mechanism" given by equation (8) (or equation (7)) appears to be a valid representation of the stress–relaxation mechanism in dry cellulose. We shall attempt to give a possible explanation of the significance of equation (8) in terms of the energetics of the breakdown mechanism.

The reduction of the multiple index mechanism to a mechanism controlled by a single co-operative index for a value of m equal to unity, *or* for n/n_0 values somewhat less than unity (*ca.* $n/n_0 < 0{\cdot}8$), can be seen

by inspection of equation (8). Thus, the single co-operative index mechanism, equation (5), is a special case of the more general multiple index mechanism, equation (8). Whether or not the relaxation data on rayon at 65 per cent R.H., as used by Nissan[19] for construction of *Figure 3*, would indicate a multiple index mechanism for very small times is a matter of conjecture; however, Nissan's treatment of the data definitely establishes the single co-operative index effect over many decades of time. The data for cellophane as presented in *Figures 4* and *5* confirm Nissan's findings for larger times, and in addition, lead to a generalization of the stress relaxation process in terms of a distribution of co-operative indices for small times (see *Figure 7* and equation (8)).

The primary co-operative index predicted by equation (8) is given by $\alpha_1 = (m + \alpha - 1)/m$ and is equal to 4·4 for the cellophane studied ($\alpha = 3$, m (slope) $= 0{\cdot}59$; see *Figure 7*). This is the limiting co-operative

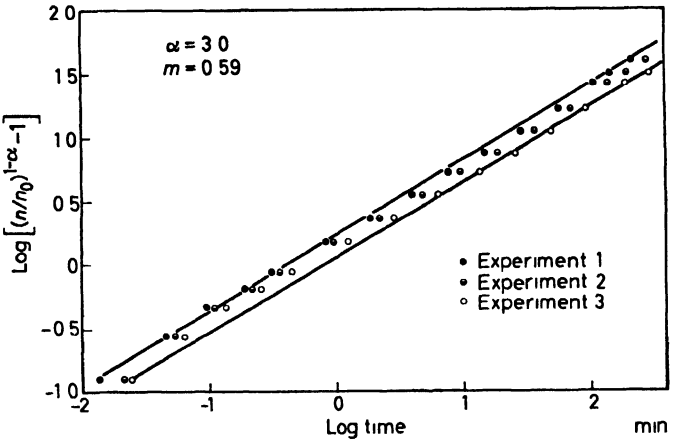


Figure 7

index which governs the hydrogen–bond relaxation process at large times (small values of n/n_0).

An apparent correlation between the co-operative index obtained from stress–relaxation studies and a parameter obtained from regain studies has been given by Nissan[19]. Nissan finds that for low or zero regain, in cellulosics, the co-operative index should approach unity. This is not an unreasonable assumption since it might be expected that, at zero water content, the hydrogen bonds act individually. At higher water content, the cellulose in the amorphous regions of regenerated cellulose may behave as if groups of various sizes (and hence of varying number of hydrogen bonds linking the group to the surrounding structure) are present, thus resulting in a higher co-operative index or a *spectrum* of indices.

The primary index of 4·4 found for the cellophane studied may be due to the fact that a small amount of plasticizer is present in the cellophane. It is reasonable to expect that the primary co-operative index will vary with regain, plasticizer content, density or any other factor which affects the degree of association in the amorphous regions.

Inspection of equation (8) reveals that, for $0{\cdot}5 < m < 1$ and $\alpha > 1$, the co-operative index *increases* for each term in the series, and the coefficient of each term *decreases*. Thus, the reaction rate constant associated with the index α_1 is k', and the constant associated with α_2 is $k'(1 - m)/m$, which is less than k'. If the free energy of activation for k' is calculated from equation (7), *Figure 7*, and the Eyring absolute reaction rate theory, a value of 21·2 kcal per mole of relaxation units is obtained (at 41°), each relaxation unit containing α_1 hydrogen bonds. The energies of activation for the reaction rate constants of all other terms in equation (8) will be *larger* than 21·2 kcal, as can be verified by inspection of equation (8) and the absolute reaction rate equation.

At very small times, *i.e.* at n/n_0 near *unity*, there are a large number of active hydrogen bonds in the system, and the stress on the system is high. In this condition, it is possible for groups with high co-operative indices, *i.e.* a large number of hydrogen bond links to the surrounding structure, to " relax " by a co-operative hydrogen bond breakdown. This is only possible when the strain energy in the system is high. As the number of stress-supporting hydrogen bonds decreases, the stress on the system also decays. The system then approaches the breakdown mechanism whereby a *minimum* free energy of activation is required per *co-operative group* — this corresponds to the primary reaction order of the system (α_1), where the value of α_1 depends on the degree of association in the amorphous regions, as discussed above. All other mechanisms require a higher free energy of activation which becomes increasingly more difficult (less probable) to obtain since the stored strain-energy in the system has decreased.

The basic hypothesis on which interpretation of stress relaxation given above is based, is that first advanced by Nissan[19]—that the viscoelastic properties of hydrogen-bond controlled solids can be interpreted in terms of a single non-Hookean element, the hydrogen bond. Thus, viscous effects are a result of a time-dependent change in the active number of hydrogen bonds, and are not caused by elements which are purely viscous in their behaviour.

While the basic hypothesis of the hydrogen bond theory is markedly different from the concepts of phenomenological viscoelasticity theory, there are some interesting, but not completely understood, consonant findings which result from the application of the two theories.

The energy of activation found for the primary co-operative index mechanism of equation (8) is 21·2 kcal, and this result is comparable to the values of 25 kcal as quoted by Meredith[6] for stress–relaxation of dry viscose filaments. The value quoted was obtained from an analysis of the stress–relaxation process in terms of the Eyring theory of viscoelastic behaviour.

The apparent distribution of co-operative indices found in this investigation may be considered analogous to the distribution of relaxation times obtained from linear viscoelasticity theory. Thus, if in equation (8), m is unity, a single co-operative index describes the relaxation process, and if $m = 1/2$, all of the coefficients in equation (8) are equal—this results in a " box " distribution of reaction rate constants. Further experimental and theoretical investigation is required to establish a quantitative link between

the hydrogen bond theory and the phenomenological approach—however, the hydrogen bond theory appears to present a logical, self-consistent approach to the viscoelasticity of regenerated cellulose, and although an interpretation of the theory in terms of the well-established phenomenological approach is highly desirable, the absence of a quantitative link between the two methods does not detract from the plausible *molecular* interpretation of viscoelasticity afforded by the hydrogen bond theory.

Nomenclature

a	Morse function constant	n	Effective hydrogen bond density, $3/l^3$
ΔH	Energy of dissociation per mole of hydrogen bonds	n_0	Effective hydrogen bond density at zero time of relaxation
K	Relaxation constant, $3a^3\Delta H/Nr_e r_0^2$	r	Hydrogen bond length, H · · O
K_1	O—H force constant	r_e	Hydrogen bond length at equilibrium
K_2	H · · O force constant	r_0	$r_e(1 + K_2/K_1)$
k	Reaction rate constant	t	Time
k'	Reaction rate constant, multiple co-operative index mechanism	U	Potential energy of a hydrogen bond
l	Hydrogen bond repeat distance	α	Co-operative index
l^3	Volume containing 3 non-interacting hydrogen bonds	β	Slope, equation (5c)
m	Slope, equations (6, 7, 8)	ϵ	Strain
N	Avogadro's number	Σ	Relaxation number
		σ	Stress

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CHEMICAL MECHANISMS IN THE GRAFTING OF CELLULOSE

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INTRODUCTION

The detailed mechanism of the various processes used in grafting synthetic polymers onto cellulose is not always known. The suggestions that have been made are based on a comparison with what is known of similar reactions with low molecular weight model compounds, and are of necessity highly speculative. The discussion will be restricted to polymerization and grafting initiated by radicals.

Before discussing mechanisms of grafting, some general remarks are in order.

Physical conditions affecting grafting

Until comparatively recently, most attempts to achieve grafting onto cellulose gave poor results. The reason for the greater success of later work is frequently related to the physical rather than the chemical conditions. Most of the work has been done with fibres and films, and it is now realized that swelling is an important pre-requisite to render the system accessible to monomer. Although cellulose does not swell spontaneously in most organic liquids, it can be made to hold large amounts of these liquids if water is stepwise removed, first by alcohol–water mixtures of gradually increasing alcohol content and, subsequently, by the organic liquid in question (*solvent-exchange*).

However, essential though this swelling may be for the penetration of the monomer, this is not the only factor of importance. Swollen films or fibres are systems in which the cellulose concentration is very high. As a result, the cellulose can compete successfully with monomer or with other compounds to give reactions that lead to grafting. It is believed, for example, that the reaction between cellulose and hydroxyl radicals falls in this category (see below).

The problem of swelling and accessibility brings up another question which sometimes may be a determinative factor in grafting experiments: the rôle of diffusion. This has not been studied systematically, but there are indications that under certain conditions diffusion can be rate controlling[1].

The possible rôle of swelling and diffusion will often render the quantitative interpretation a very difficult task. Even if none of the reactions is diffusion controlled, the composition of the fibre or film changes gradually as grafting proceeds, and the solubility of the compounds participating in the process

will likewise change. From this point of view, studies in homogeneous solutions are preferable, but in practice this is not always possible.

Another conclusion which may be drawn is that the process of grafting onto paper is not necessarily wholly comparable to that of grafting onto swollen cellophane or rayon. The grafting onto paper fibres may be much more in the nature of a surface phenomenon.

Proof of grafting

When the reaction has taken place in a homogeneous solution, proof of chemical grafting must often be based on time-consuming fractionations to separate the graft copolymer from the homopolymer. In this respect, the use of fibres or films has an advantage. One extracts with a liquid which is a solvent for the homopolymer, then dries, and considers the weight increase as a measure of grafting. It is clear, however, that this is permissible only when all the homopolymer can be removed by extraction. This is seldom true, and it is probably less true when the weight increase is larger than when it is slight.

A number of qualitative tests can be applied, however, to show that the product contains graft copolymer, although they do not give us quantitative information about the ratio of graft to homopolymer. In these tests it is recommendable, and sometimes essential, to compare the properties of the product believed to be a graft, with those of a physical mixture of the two types of macromolecules. This mixture should have the same over-all composition as the graft and, if at all possible, the average molecular weight of the polymer grafted on to the cellulosic material should be the same as that of the homopolymer in the physical mixture. One attempt to meet this last requirement is to extract the homopolymer from the fibre or film onto which grafting took place; however, even this may not always be sufficient guarantee that the desired goal has been reached, because if the extraction of the homopolymer is incomplete it is likely that the molecules with relatively low molecular weight were extracted in preference to those of high molecular weight.

Easily accessible properties of the graft copolymer which may be compared with those of a physical mixture are the intrinsic viscosity and the concentration dependence of the viscosity[2]. Any difference between the two products is indicative of grafting. Another property is the solubility. By way of example, mention is made of the following experiments[3].

Acrylamide was grafted onto cellophane. The product was dissolved in cuprammonium, and the solution acidified. The amount of precipitate formed was always larger than the amount of cellulose present, and infra-red absorption showed the presence of polyacrylamide. When the same experiment was performed with a physical mixture of cellulose and polyacrylamide of the same composition, only the cellulose was precipitated. The experimental data are shown in *Figure 1.*

Sometimes the grafting process itself shows features which are incompatible with the assumption that only homopolymer is formed. For example, if the rate at which the weight of the film increases becomes gradually lower with increasing time while the homopolymerization outside the film shows no such tendency, this does not constitute a proof, but it can sometimes be

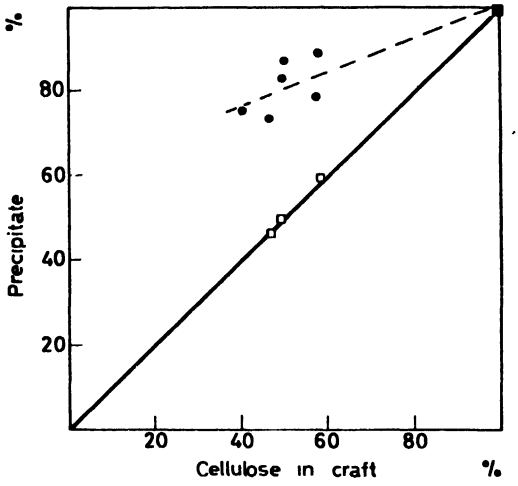


Figure 1. Weight of precipitate after acidifying a cuprammonium solution of cellulose–polyacrylamide, as a function of the amount of cellulose in the system; ●: graft copolymer; □: physical mixture

considered an indication of grafting. In one case[2] the failure to observe a weight increase in the presence of acetone could be considered as evidence for grafting in the absence of acetone, because the homopolymerization takes place in either case.

Molecular weight of grafted polymer

Data concerning the chain length of the polymers grafted onto cellulose are very scarce. The polystyrene grafted onto mercapto-ethylated cellulose acetate films through chain transfer was shown to have a molecular weight of more than 1 million[4], which means that even when the weight increase is 100 per cent and is entirely due to grafting, only 1 out of more than 5,000 glucose units carries a polystyrene side chain. Similar conclusions can be drawn from the results obtained by Rapson[5] and by Kobayashi[6] in their experiments on γ-ray-induced grafting. This discouraging result raises the question of whether the product was a graft copolymer in the usual sense. It could mean that grafting occurred only at the ends of cellulose chains, in which case the product should, more properly, be called a block copolymer.

A possible explanation of this high molecular weight is a reduction in the rate of termination due to the low mobility of the polystyrene chain radicals inside the film or fibre.

HIGH ENERGY IRRADIATION

Gamma irradiation has been applied to a very large number of polymer–monomer systems. Applications to cellulose were not very successful at first, owing to the degradation inflicted on the cellulose. Recent work has shown, however, that this degradation can be suppressed to a considerable degree if care is taken that the cellulose is accessible to the monomer[5–10].

This is a typical example of the rôle of swelling and solvent-exchange mentioned in the introduction.

Judging from what is known of the effect of high energy irradiation on other polymers, it may be assumed that hydrogen atoms are abstracted from the cellulose. If this takes place in the presence of a monomer, grafting can occur immediately. It is also possible, however, to use the pre-irradiation technique: irradiation in the absence of monomer, followed by immersion in a solution of the monomer[6]. This shows that some of the radicals formed on the cellulosic backbone are long-lived; the pre-irradiation method, however, is more likely to cause degradation.

It should also be mentioned that to the author's knowledge no proof has so far been given that the products obtained by γ-irradiation of cellulose are true grafts, although this may be considered as very likely.

Although direct hydrogen abstraction is a likely process, it must still be considered an hypothesis, and it is almost certain that, in the presence of water, hydroxyl radicals play a prominent part. Leavit[11] has observed crosslinking of cellulose derivatives in aqueous solutions when exposed to γ-irradiation, and has formulated the hypothesis that the first step in this process is the production of OH radicals from water. It is believed that these OH radicals subsequently abstract hydrogen from the cellulosic macromolecules. The same mechanism has been postulated to explain the effect of γ-irradiation on gelatine[12], which produces crosslinks in the presence, but not in the absence, of water.

This raises a question in connection with the effect of OH radicals produced by the reaction between ferrous ions and hydrogen peroxide (see below). It may be expected that these, too, give rise to some crosslinking, although evidence for this has not been found.

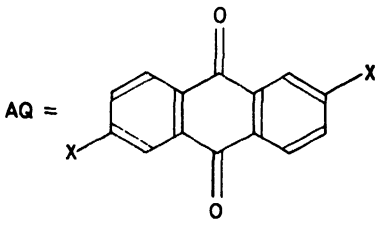
PHOTOSENSITIZERS

Polymerization may be initiated by low energy photons if the irradiation takes place in the presence of a photosensitizer. Photosensitizing action has been found in well-known radical producing catalysts such as 2-azo-*bis*-isobutyronitrile[13] and benzoylperoxide[14]. Most studies have been done, however, with dyes, in particular with dyes that have phototendering activity, *i.e.* they cause degradation of cellulose fibres when these fibres are exposed to light. It is now fairly generally accepted that this phototendering action is due to radical reactions. A wealth of interesting data can be found in the proceedings of a symposium[15]. A recent discussion was given by Bridge[16].

The mechanism of phototendering has been elucidated to a large extent by work with low molecular weight model compounds[17]. The reaction between the photo-excited dye molecule and an alcohol produces radicals, which may then initiate the polymerization of a monomer. If the alcohol is itself a polymer, this polymerization may produce a graft.

Application of photosensitizers to polymerization and grafting

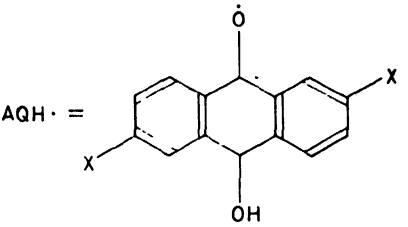
Detailed studies of this type of polymerization and grafting have been carried out by Geacintov[3, 18], using anthraquinoid dyes of the type



where X was SO_3Na when a water-soluble dye was desired, but CH_3 in the case of organic solvents. This latter compound will be abbreviated MeAQ. The reaction between the excited dye molecule and an alcohol is[17]

$$AQ^* + RH \longrightarrow AQH\cdot + R\cdot \qquad (1)$$

where



It is usually assumed that the hydrogen abstraction takes place from the carbon atom carrying the OH group. For example, with ethanol,

$$AQ^* + CH_3CH_2OH \longrightarrow CH_3\dot{C}HOH + AQH\cdot$$

It is of some interest to note, however, that the rate of photopolymerization of styrene in benzene, in the presence of MeAQ, was increased to an even greater extent by t-butanol than by s-butanol, n-butanol or ethanol[3]. Since no α-hydrogen is present in the t-butanol, this suggests that the $(CH_3)_3CO\cdot$ radical is formed, which perhaps decomposes subsequently to acetone and the methyl radical[19]. This hydrogen abstraction from the oxygen atom may be the normal process if the anthraquinone, *via* its carbonyl groups, complexes with the hydroxyl group of the alcohol. If α-hydrogen is present, the radical formed may be expected to rearrange readily:

$$RCH_2O\cdot \longrightarrow R\dot{C}HOH$$

Some studies were made with carbon-14 labelled ethanol to measure the amount of alcohol in the polymer produced. The monomer was methylmethacrylate (MMA) or styrene (Sty) in benzene, to which varying amounts of radioactive alcohol were added. The photosensitizer was the dye MeAQ, and the light-source a GE–H–400 E–1 400 Watt mercury lamp, of which only the light of wavelengths larger than 320 mμ was allowed to reach the reaction vessel. By way of comparison, the photosensitizer, in some experiments, was replaced by azo-*bis*-isobutyronitrile (ABN).

Table 1. Photo-initiated polymerization of methylmethacrylate (MMA) and of styrene (Sty) in the presence of 20 vol per cent ethanol; *N is the number of polymer chains per ethanol molecule in the polymer*

Monomer	*Initiator*	*t* (h)	*Conversion* (%)	*D.P.*	*N*
MMA	MeAQ	3	24	80	5
MMA	ABN	3	26	120	220
Sty	MeAQ	20	15	70	6
Sty	ABN	20	18	70	53

It was found that the photosensitized polymerization of MMA was four to five times faster in the presence of 20 vol. per cent ethanol than in the absence of ethanol. No accelerating effect of the alcohol was observed when ABN was used as initiator.

The degree of polymerization of the polymer was measured, and the number of polymer chains per alcohol molecule found in the polymer was calculated from the radioactivity, with the results shown in *Table 1.* The small amount of ethanol found in the polymer when initiating with ABN is probably due to chain transfer from polymer radicals to ethanol, with subsequent initiation by the alcohol radical.

$$M_nM\cdot + CH_3CH_2OH \longrightarrow CH_3\dot{C}HOH + \text{Polymer} \qquad (2)$$

$$CH_3\dot{C}HOH + M \longrightarrow \text{Polymer} \qquad (3)$$

The poly-MMA formed by MeAQ-sensitized photo-initiation contains only one molecule of alcohol for every 5 chains. Taking into account that for this polymer 75 per cent of the termination is by disproportionation and 25 per cent by combination, it follows from a simple calculation that one out of about 6 chains is initiated by addition of an alcohol radical. This neglects the possible termination by alcohol radicals. In the polymerization of styrene, termination is mainly by combination and one must conclude that one out of about 12 chains is initiated by addition of alcohol. Although these figures are considerably lower than expected, the results show convincingly that alcohol radicals are formed and play a rôle in the initiation.

It has not been possible to describe the kinetics of the process in a satisfactory manner. If the rate of polymerization is represented by an equation of the form:

$$-\mathrm{d}M/\mathrm{d}t = kI^mD^nA^pM^q, \qquad (4)$$

where I is the light-intensity, D the dye concentration, A the alcohol concentration and M the monomer content, it is found that the exponent n is about 0·3 at low alcohol content but decreases to half this value when A becomes large. The value of m is only 0·25, p is about 0·55 and q about 1·3. This behaviour suggests that the kinetics are complex. Kinetic schemes can be developed that predict $m = 0{\cdot}5$, $n = 0{\cdot}5$, p between zero and 0·5, and

$q = 1$. Other schemes lead to $m = 0$, $n = 0$, $p = 1$ and $q = 2$. The actual behaviour may be due to a mixture of mechanisms, but the details are as yet so speculative that it is considered premature to enter into this matter any further.

If the behaviour of ethanol is indicative of that of cellulose, reaction (3) may lead to a graft copolymer. The kinetics of this grafting were found to be as complicated as that of the homopolymerization. They were further complicated by the fact that grafting took place in a film of cellophane or cellulose acetate immersed in the reaction mixture. Evidence of grafting was obtained by the techniques described in the introduction.

REDOX SYSTEMS

The reaction (1) between the dye and the alcohol is an example of an oxidation-reduction reaction that involves radical species. Many oxidation–reduction reactions are of this type, and some of them have been used to initiate grafting.

Persulphate initiation

Grafting on to cellophane has been achieved through initiation by persulphate ions. Some experiments were done with cotton in acrylonitrile vapour by Haydel *et al.*[20], but these authors did not investigate whether real grafting occurred and referred to the process as deposition in and coating of the cotton. There is little doubt that the product obtained by Chaudhuri[21] who used cellophane instead of cotton was a graft copolymer; this was proved by some of the methods described in the introduction.

It is believed that the conditions for grafting in Chaudhuri's experiments were particularly favourable. The cellophane was soaked in an aqueous solution of persulphate, and the wet film immersed in hot acrylonitrile. The monomer diffuses into the film, but very little of the catalyst diffuses into the monomer because the solubility in the monomer is low. Consequently, little if any homopolymer is formed outside the film. Inside the film the concentration of cellulose is high, which is likely to favour reaction of cellulose with persulphate radicals (or with hydroxyl radicals if these are formed).

Mechanism of the initiation by persulphate

Kolthoff and Miller[22] have shown that persulphate ions in aqueous solution, when heated, decompose according to two mechanisms, only one of which is believed to produce radicals:

$$S_2O_8^{(2-)} \xrightarrow{k_1} 2SO_4^{-}\cdot \qquad (5)$$

This is a first-order reaction. The sulphate radicals formed may react with water to produce hydroxyl radicals and, finally, oxygen,

$$SO_4^{-}\cdot + H_2O \longrightarrow HSO_4^{-} + HO\cdot$$

$$2HO\cdot \longrightarrow H_2O + \tfrac{1}{2}O_2$$

Polymerization may thus be initiated either by $SO_4^-\cdot$ or by $OH\cdot$. That SO_4^- radicals do take part in the initiation is shown by the fact that sulphur occurs in the polymer[23, 24].

Simultaneous with process (5) there takes place a decomposition of the persulphate which does not produce radicals. The rate of this second decomposition is proportional to the concentration of hydrogen ions and is markedly affected by ionic strength. The total rate constant for the decomposition is, therefore,

$$k = k_1 + k_2(H^+)$$

When studying the mechanism of oxidation or initiation by persulphate, it is convenient to keep the hydrogen ion concentration below 10^{-4}M, because under these conditions $k_2(H^+)$ is negligible compared with k_1.

The decomposition of persulphate has been studied also in the presence of alcohols. As early as 1903 Marie and Bunel[25] published data to show that methanol accelerates the persulphate decomposition considerably. This has been confirmed by Kolthoff, Meehan and Carr[26], who proved that the reaction is not between the persulphate ion and methanol, but between the sulphate ion radical and methanol,

$$S_2O_8^{(2-)} \longrightarrow 2SO_4^-\cdot; \quad SO_4^-\cdot + CH_3OH \longrightarrow HSO_4^- + \cdot CH_2OH$$

This is proved by the fact that the rate of decomposition of persulphate is reduced to the same low value as when allyl acetate is added in the absence of alcohol. The accelerating effect of the alcohol is, therefore, attributed to a reaction with the CH_2OH radicals,

$$\cdot CH_2OH + S_2O_8^{(2-)} \longrightarrow HCHO + HSO_4^- + SO_4^-\cdot \qquad (6)$$

On addition of allyl acetate, the SO_4^- radical reacts with the monomer rather than with the alcohol, so that no CH_2OH radicals are formed and reaction (6) can no longer take place. This scheme is confirmed by the fact that formaldehyde is formed in the absence, but not in the presence of, allyl acetate.

Likewise, there is no direct reaction between the persulphate and the monomer. It should be noted, however, that in this respect Kolthoff's results are not in agreement with those of Bartlett and Nozaki[24], who found that the decomposition of persulphate is accelerated by allyl acetate.

It is clear that if cellulose reacts in the same manner as methanol, no grafting would take place, because the SO_4^- radicals would react with monomer rather than with cellulose. Chaudhuri's experiments[21], which indicate that grafting did occur, would thus appear to constitute a discrepancy. However, several factors must be borne in mind.

(i) The results obtained by Kolthoff *et al.* suggest that the result of the competition between monomer and cellulose will decide whether grafting takes place. If cellulose is present in excess—which may be the case in a swollen film or fibre—a substantial amount of $SO_4^-\cdot$ (or $OH\cdot$ if $SO_4^-\cdot$ reacts with water) may attack the cellulose.

(ii) For the same reason the result of the experiment will depend on the nature of the monomer.

(iii) We have no guarantee that all alcohols react the same way. Preliminary results recently obtained at Syracuse in a study of persulphate decomposition in the presence of pinacol appear to indicate that the rate of this decomposition is independent of the pinacol concentration, although oxidation of the pinacol does take place. It is obvious, therefore, that further experiments are needed.

Initiation by hydroxyl radicals

A well-known example of an oxidation–reduction reaction in which radicals are produced is the decomposition of hydrogen peroxide by ferrous ions, for which Haber and Weiss[27] developed the following scheme:

$$H_2O_2 + Fe^{2+} \longrightarrow HO\cdot + HO^- + Fe^{3+} \qquad (7)$$

$$HO\cdot + Fe^{2+} \longrightarrow HO^- + Fe^{3+} \qquad (8)$$

This is accompanied by a chain reaction in which HO radicals decompose H_2O_2

$$HO\cdot + H_2O_2 \longrightarrow H_2O + \cdot HO_2\cdot \qquad \cdot HO_2\cdot + H_2O_2 \longrightarrow HO\cdot + H_2O + O_2$$

but, according to Kolthoff and Parry[28], this takes place to any appreciable extent only if H_2O_2 is present in large excess over ferrous ion.

Baxendale, Evans and Park[29] in a classical study showed that the HO radicals produced in reaction (7) can initiate polymerization. As the monomer concentration is increased, reaction (8) is suppressed to an ever-increasing extent until, finally, at high monomer content, all the OH radicals react with the monomer to produce polymer.

It is been claimed[30] that the reaction between H_2O_2 and Fe^{2+} in the presence of cellulose and a monomer produces a graft copolymer, and the reaction which is believed to be responsible for this is abstraction of hydrogen:

$$\text{Cell H} + OH\cdot \longrightarrow \text{Cell}\cdot + H_2O \qquad (9)$$

This is based on known reactions with low molecular weight alcohols. According to Kolthoff and Medalia[31], for example, OH radicals in the presence of hydrogen peroxide are capable of oxidizing ethanol in a chain reaction, as follows:

$$HO\cdot + CH_3CH_2OH \longrightarrow CH_3\dot{C}HOH + H_2O \qquad (10)$$

$$CH_3\dot{C}HOH + H_2O_2 \longrightarrow CH_3CHO + H_2O + HO\cdot \qquad (11)$$

Here again it was shown that acrylonitrile can capture all the OH radicals, so that in the presence of this monomer not only reaction (8) but also (10) is suppressed. Hence, if cellulose gives essentially the same reaction as ethanol, *i.e.* reaction (10), it will have to compete with the monomer while, at the same time, for grafting to occur, the cellulosic radical formed must be able to react with monomer. It is clear that under these circumstances

the yield of graft copolymer must depend to a large extent on the conditions of the experiment. The best technique appears to be to first expose the cellulose to a solution of a ferrous salt. The ferrous ions are adsorbed by the cellulose, and the subsequent reaction with hydrogen peroxide in the presence of a monomer will, therefore, take place in close proximity to cellulose molecules at a large concentration of these molecules. It is to be expected that factors favouring the adsorption of the ferrous ions (for example the presence of carboxyl groups in the cellulose) will affect the grafting. For the same reason, one may expect that more polymer will be deposited in (and/or grafted on to) the cellulose in neutral media than in acid media.

Salts of cerium (IV)

An oxidizing agent that has received much attention in recent years is the ceric ion. The reaction by which this ion produces radicals when reacting with certain organic compounds is believed to be

$$Ce^{4+} + RH \longrightarrow Ce^{3+} + H^+ + R\cdot$$

Quantitative studies appear to have been made mainly with alcohols, although many other organic compounds are likewise susceptible to this oxidation. There is much evidence[32] that the oxidation of alcohols proceeds through the disproportionation of a co-ordination complex. The evidence for this is partly from absorption spectra and partly from the kinetics of the oxidation itself. Either method has been used, also, to obtain the instability constants of the complexes involved. Ardon[33], for example, described the oxidation of ethanol in aqueous solutions of ceric perchlorate by the reactions

$$Ce^{4+} + EtOH \underset{}{\overset{K}{\rightleftharpoons}} \text{Complex}$$

$$\text{Complex} \xrightarrow{k} Ce^{3+} + H^+ + \text{oxidation product}$$

The alcohol was present in excess. If the complexing equilibrium is established rapidly, one should expect that the concentration c of the complex obeys the equation

$$c = KA([Ce] - c), \text{ or } c = (1 + KA)^{-1}KA[Ce]$$

where A represents the alcohol concentration and [Ce] the total ceric content. The rate at which ceric is reduced will be kc or k^1[Ce], where

$$k^1 = kKA(1 + KA)^{-1}$$

This means that k and kK can be derived from a plot of $1/k^1$ *v.* $1/A$. The linearity of this plot was confirmed by experiment, and the composition of the complex was confirmed independently by spectrophotometric measurements.

Similar studies with other systems, in which the ethanol was replaced by other compounds[32, 34] or the cerium (IV) by other metals[35] have led to similar results.

However, the analysis of the data is not always simple. This is due to the fact that the aqueous solutions of ceric salts, even in the absence of alcohol, are quite complex, owing to the strong tendency of ceric ions to form complexes with anions. In aqueous solutions of ceric sulphates the following species have been postulated[36–38]: $CeSO_4^{2+}$, $Ce(SO_4)_2$ and $Ce(SO_4)_3^{2}$, and the stability constants of these complexes have been estimated. Ceric nitrate solutions show less complexing, and the perchlorate anion does not complex with Ce(IV) to any appreciable extent. This does not mean, however, that aqueous solutions of ceric perchlorate contain free cerium (IV) ions. According to Hardwick and Robertson[36], Sherrill[39] and Offner[40], the ceric ions in these solutions, even at quite low pH, are complexed with hydroxyl ions to $CeOH^{3+}$ and $Ce(OH)_2^{2+}$.

A further complication arises from the fact that dimers occur and—depending on pH and on concentration—trimeric, tetrameric and other polymeric ions.

A few examples may illustrate the situation. The stability constant of the ceric–ethanol complex[33] depends on pH. This dependence was explained by Ardon[33] on the basis of the equilibrium

$$Ce^{4+} + H_2O \rightleftharpoons CeOH^{3+} + H^+$$

This same equilibrium was used by Hargreaves and Sutcliffe[34] to explain the effect of $HClO_4$ content on the rate of oxidation of formaldehyde by ceric perchlorate. It leads to the prediction that the reciprocal of the rate constant for this oxidation is a linear function of the reciprocal of the acid concentration, in accordance with the experimental results. Addition of sodium perchlorate to ceric perchlorate solutions has only a small effect on the rate of oxidation, indicating that perchlorate ions do not complex with Ce(IV). Addition of neutral sulphate to ceric sulphate solutions, however, reduces the rate of oxidation. On the other hand, addition of sulphuric acid to ceric sulphate may lead to *increased* oxidation rates. Hargreaves and Sutcliffe[34] explained this by postulating a special complexing scheme involving both sulphate ions and hydrogen ions. It will be clear from these data that the quantitative results in any one series of experiments may depend strongly on the concentrations of the various species present.

Initiation by cerium (IV) ions

A study which is of great interest in the understanding of grafting is the oxidation of pinacol by ceric sulphate, which was published by Mino, Kaizerman and Rasmussen[41]. The ceric salt concentration varied from about 2×10^{-3} to 10×10^{-3} M, the pinacol concentration from 0·03 to 0·16M. The rate at which Ce(IV) was reduced was proportional to both the ceric and the pinacol content, and 2 moles of acetone were produced for every 2 moles of Ce(IV) reduced. The reactions describing this are:

$$Ce^{4+} + (CH_3)_2\overset{HO}{\overset{|}{C}}—\overset{OH}{\overset{|}{C}}(CH_3)_2 \longrightarrow (CH_3)_2CO + (CH_3)_2\dot{C}OH + H^+ + Ce^{3+}$$

$$Ce^{4+} + (CH_3)_2\dot{C}OH \longrightarrow (CH_3)_2CO + H^+ + Ce^{3+}$$

in which the first of the two reactions is rate determining.

In the presence of (excess) acrylamide, only one mole of acetone was produced per mole of Ce(IV) reduced. Since it is known from kinetic studies of the polymerization, that the termination step involves reaction with a ceric ion, the following reactions are believed to take place (M = monomer):

Initiation $(CH_3)_2\dot{C}OH + M \longrightarrow RM\cdot$

Propagation $RM\cdot + M \longrightarrow RM_n\cdot$

Termination $RM_n\cdot + Ce^{4+} \longrightarrow$ Polymer $+ Ce^{3+} + H^+$

This termination could conceivably consist of oxidation to an hydroxyl end group in the polymer chain.

It is clear that if the initiation step involves addition of the alcohol radical, and assuming that cellulose reacts similarly, one may expect grafting[42]. The method has been used to graft a number of monomers onto paper, and both the tensile strength and the burst strength of the paper were shown to be improved considerably[43]. At the same time the rate of water absorption was decreased, although the equilibrium water absorption per unit weight of cellulose did not change.

CHAIN TRANSFER

Growing polymer radicals are capable of transferring the radical character to certain compounds called chain transfer agents. The process is called chain transfer; it terminates the polymer chain and leaves the chain transfer agent in the form of a radical that may subsequently initiate another polymerization reaction. In many cases chain transfer takes place by abstraction of a hydrogen or halogen atom, *e.g.*:

$$-CH_2\overset{\overset{H}{|}}{\underset{\underset{R}{|}}{C}}\cdot + CCl_4 \xrightarrow{k_a} -CH_2-\overset{\overset{H}{|}}{\underset{\underset{R}{|}}{C}}Cl + \dot{C}Cl_3 \qquad (12)$$

$$-CH_2\overset{\overset{H}{|}}{\underset{\underset{R}{|}}{C}}\cdot + XH \xrightarrow{k_a} -CH_2-CH_2R + X\cdot \qquad (13)$$

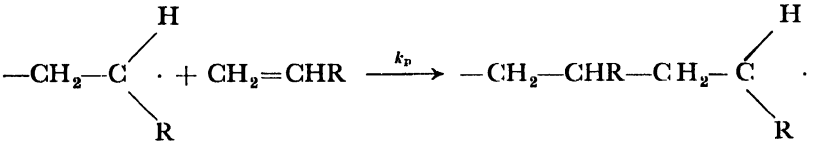
but in chain transfer to monomer, hydrogen donation is more likely than hydrogen abstraction:

$$-CH_2\overset{\overset{H}{|}}{\underset{\underset{R}{|}}{C}}\cdot + CH_2=CHR \xrightarrow{k_a} -CH_2=CHR + CH_2-\overset{\overset{H}{|}}{\underset{\underset{R}{|}}{C}}\cdot \qquad (14)$$

Since in a polymerization process the chain transfer agent has to compete with the monomer, the quantity that determines the relative frequency of chain transfer is the " chain transfer constant "

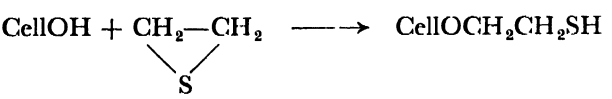
$$C_s = k_s/k_p \qquad (15)$$

where k_s is the rate constant for the bimolecular reaction of type (12), (13) or (14) and k_p the propagation rate constant for the bimolecular addition reaction between growing polymer radicals and monomer molecules:



Clearly, if the chain transfer agent is itself a polymer, the polymerization, initiated by the radical formed in the chain transfer process, may lead to a graft copolymer. In the earliest attempts to apply this to cellulosic compounds, halogen-substituted cellulosics[44] were used, but the yields were very low. As will be discussed below, chain transfer with subsequent grafting takes place to some extent even in the absence of halogen. The failure to produce graft from the halogen-substituted cellulose must therefore be attributed to the conditions used in the experiments.

Mercaptans are known to show particularly high chain transfer constants, and this has led to several attempts to introduce SH groups in polymers. The methods proposed[45] were complicated, but recently Chaudhuri[2] succeeded in preparing mercapto-ethylated films of cellophane and cellulose acetate by an addition reaction with ethylene sulphide:



This reaction takes place readily when the cellulose acetate is swollen in benzene. To achieve the same result with cellophane, it is necessary to first solvent-exchange the water, *via* alcohol, by benzene After washing the mercapto-ethylated film with benzene, styrene was added and its polymerization initiated by azo-*bis*-isobutyronitrile at 65°. The rate of grafting was of the order of 10 weight per cent per hour.

Kinetics of grafting through chain transfer

A quantitative treatment of the kinetics of grafting by chain transfer in a homogeneous system was given by Voeks[46] and by Fox *et al.*[47]. Assuming that the polymerization is initiated by the decomposition of some radical-producing catalyst, *C*, the main processes to be considered are (M = monomer):

Initiation	C	$\longrightarrow 2R_0\cdot$		(k_i)
Propagation	$R_0\cdot + M$	$\longrightarrow R_0M\cdot \longrightarrow$	Homopolymer	(k_p)
Transfer	$R_0M_n\cdot + XH$	$\longrightarrow X\cdot + M_nH$		(k_s)
Propagation	$X\cdot + M$	$\longrightarrow XM\cdot \longrightarrow$	Graft	(k_p)

Termination, by combination (k_{tc}) and/or disproportionation (k_{td})

In the case under consideration, XH is the cellulosic macromolecule. The usual approximation, *i.e.* that the rate at which the various radicals react with monomer, chain transfer agent and each other is independent of the nature of the radical, is made. Under these conditions the total steady state concentration of radicals is found to be the same as in the absence of chain transfer agent:

$$[R_{tot}\cdot] = a = (k_i f c/k_t)^{\frac{1}{2}} \tag{16}$$

where c is the catalyst concentration and f the efficiency of initiation; $k_t = k_{tc} + k_{td}$. The rate of monomer consumption is therefore likewise independent of the presence of chain transfer agent:

$$-\mathrm{d}M/\mathrm{d}t = k_p a M \tag{17}$$

while the chain transfer agent is consumed at a rate:

$$-\mathrm{d}[XH]/\mathrm{d}t = C_s k_p a [XH] \tag{18}$$

It follows from these equations that both M and [XH] decrease exponentially with increasing time. The weight at which the weight G of graft copolymer increases is found to be:

$$\frac{\mathrm{d}G}{\mathrm{d}t} = m M_0 k_p a \exp(-k_p a t) \frac{x}{1+x}\left[1 - \frac{k_{tc}/k_t}{(1+x)^2}\right] \tag{19}$$

where

$$x = \frac{k_s[XH]_0}{2k_t a}\exp(-k_s a t) \tag{20}$$

m is the molecular weight of the monomer and the subscript zero indicates zero time.

In Chaudhuri's grafting experiments, the constant $k_s[XH]_0/2k_t a$ in equation (20) was very large compared to unity, so that the initial rate of grafting becomes

$$(\mathrm{d}G/\mathrm{d}t)_0 = m M_0 k_p a = B M_0 f^{\frac{1}{2}} c^{\frac{1}{2}} \tag{21}$$

B being a constant which is independent of M_0 and c. To compare equation (21) with the experimental data, it must be taken into account that the efficiency of the initiation, f, changes somewhat with the monomer concentration M_0. It is assumed that this change was (relatively) the same as that outside the cellulose film. The result of the calculation is given in

Figures 2 and *3*. It could further be shown[4] that the way in which grafting proceeds at later times is also in surprisingly good agreement with the theory. In fact, the agreement is better than could have been expected, and it is believed that this is partly fortuitous and due to a number of factors that partially compensate each other:

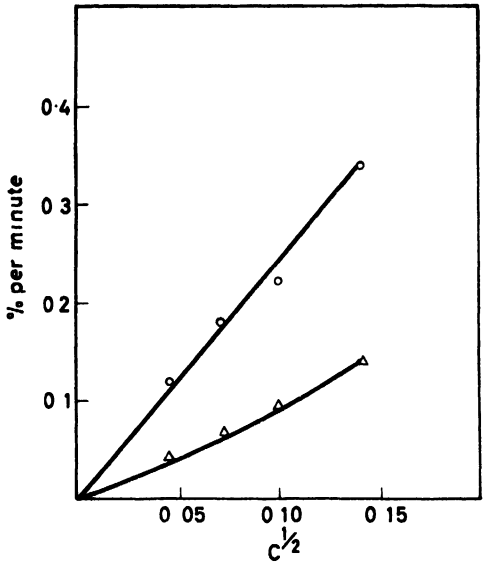


Figure 2. Initial rate of grafting due to chain transfer as a function of the square root of initiator concentration; ○: grafting; ∧: homopolymerization

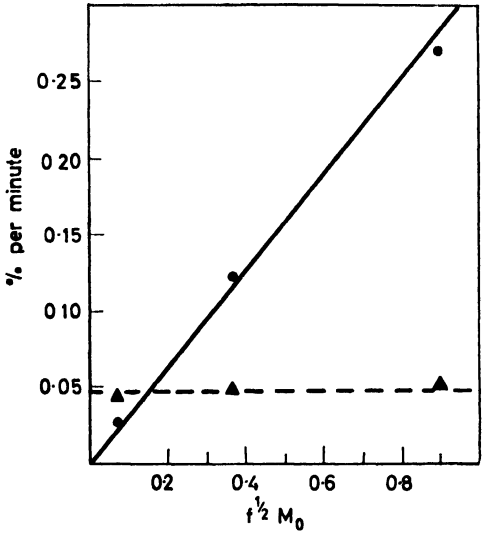


Figure 3. Initial rate of grafting due to chain transfer as a function of $M_0 f^{\frac{1}{2}}$ where M_0 is the monomer concentration and f the efficiency of the initiation; ●: grafting; ▲: homopolymerization

(i) The theory assumes that M_0, $[XH]_0$ and all the rate constants can be treated as independent of time. In reality, as grafting proceeds, the composition of the film changes and the film material will become more and more compatible with both styrene and benzene.

(ii) The amount of grafting was identified with the weight increase after thorough extraction with benzene. Although the evidence for grafting was quite convincing, it is always accompanied by some homopolymerization, and it is doubtful whether all the homopolymer can be removed by extraction in those cases where the weight increase is large.

(iii) Perhaps most important of all is the observation which was made in later experiments. A substantial amount of grafting onto cellulose acetate film took place also in the absence of SH groups. Under the conditions of the experiments this amounted to about 20 per cent of the total grafting, and one must conclude that the chain transfer constant to be used in the equations is an average over different atoms.

On the basis of this result it is believed that the failure to observe grafting by chain transfer to halogen-substituted cellulosics[44] was due to unfavourable experimental conditions.

RADICAL PRODUCING SUBSTITUENTS IN THE CELLULOSIC MOLECULE

Other methods of producing radicals attached to cellulosic macromolecules have been suggested, and although they are less likely to become of practical interest than are the previously discussed procedures, they possess some very interesting features.

Ceresa[48] applied the process of mastication to cellulose derivatives. The cellulosic material is mixed with a monomer and then masticated under high shearing forces. In this process the cellulosic molecules are torn apart, and the newly formed chain ends have radical character. These ends may initiate polymerization, thus forming a block copolymer.

Whereas Ceresa's method produces radicals by mechanical forces, it is possible to achieve this also by chemical decomposition of radical-producing substituents. This has been done with peroxides and hydroperoxides of cellulose ethers[49], but it was found that these led to excessive homopolymerization and very little grafting. Kargin, Usmanov and Aikhodzhaev[50] reported on peroxidation of rayon by ozone, followed by grafting of styrene or acrylonitrile.

Richards[51] used a method in which the number of radical-producing groups can be chemically controlled and easily determined. This consists of introducing amino groups through reaction of a halogenated amine with either a cellulosate or the sodium salt of carboxymethylcellulose.

$$\text{CellONa} + \text{ClCH}_2\text{COC}_6\text{H}_4\text{NH}_2 \longrightarrow \text{CellOCH}_2\text{COC}_6\text{H}_4\text{NH}_2$$

$$\text{CellOCH}_2\text{COONa} + \text{ClCH}_2\text{COC}_6\text{H}_4\text{NH}_2 \longrightarrow$$
$$\longrightarrow \text{CellOCH}_2\text{COOCH}_2\text{COC}_6\text{H}_4\text{NH}_2$$

The second product is an ester, the first an ether. By diazotizing, a diazonium salt is obtained which, on heating, produces radicals. A possible scheme for this is:

$$RC_6H_4N_2^+Cl^- \longrightarrow RC_6H_4\cdot + Cl\cdot + N_2$$

If this takes place in the presence of a monomer, one may obtain both homopolymer (initiation by Cl·) and graft copolymer (initiation by $RC_6H_4\cdot$). This is interesting as it opens a possibility of suppressing the homopolymerization by the addition of ferrous ions which will react as follows:

$$RC_6H_4N_2^+Cl^- + Fe^{2+} \longrightarrow RC_6H_4\cdot + Fe^{3+} + Cl^- + N_2$$

Another interesting aspect of the method is the ease with which the grafted polymer chains can be removed by hydrolysis, if the grafting has been done through ester linkages. This has advantages for fundamental studies concerning the molecular weight of the grafted chains and the effect of the grafting on the molecular weight of the cellulose.

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RECENT DEVELOPMENTS AND PRESENT TRENDS IN SULPHITE PULPING

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INTRODUCTION

Ninety-four years ago the American chemist Benjamin Tilghman was granted the world's first patent on sulphite pulping. Seven years later the world's first sulphite pulp mill was started. No one can deny that great progress has been made since this time, although this has been mainly in the technological field. Chemically, the sulphite pulping is still, with a few exceptions, the same acid one-stage process it was ninety years ago, the pH of the cold cooking liquor being fixed at between 1·5 and 2 by the use of calcium base. In spite of this, the chemists and chemical engineers have skilfully found out how to use variables other than pH, such as chemical-to-wood ratio and temperature, in order to produce a variety of pulp qualities from greaseproof to dissolving pulp.

In the competition between the calcium-base sulphite pulping and the somewhat older kraft pulping method, the sulphite process had predominated for a long time. During the 1920's, however, the alkaline process became more important, and in recent decades the growth of the kraft industry has been rather spectacular, especially in the U.S.A. One of the reasons for this is the fact that pine, and to some extent hardwoods, are not very suitable for calcium base sulphite cooking, but are readily pulped by the kraft process. Another reason is the improvements in bleaching, by which the kraft pulp can now be bleached to the same brightness as sulphite pulp. Some years ago, the slow death of the sulphite pulping method was predicted. But, the last few years have given the sulphite pulp producer a new lease of life, and the sulphite process is now recovering from its decline. New modifications of the process and new methods, which are of considerable importance for the sulphite industry, have been developed. There are now some optimists who even believe that the recent developments will slowly make the sulphite method once again the dominant process.

In order to realize the significance of the developments, some of the disadvantages, and also the advantages, of the conventional sulphite process over the sulphate process must be summarized:

Disadvantages:

Limited use of pulp wood species
Expensive pulp wood, *e.g.* spruce
Low strength properties, especially tear
Lower opacity
Water pollution problems
Longer cooking time

Advantages:

Higher yield at the same *Kappa*-number
Higher unbleached brightness
Better bleachibility
Better beatibility

All these factors are economically important and, where industrial development is concerned, only improvements which imply better economy will count.

With this background I will discuss how the recent developments can change the picture, how some disadvantages of the old sulphite process can be cancelled, and how some advantages can be further improved.

The developments, which I have in mind are:

(i) Soluble-base cooking
(ii) Improved chipping
(iii) Improved bleaching
(iv) Continuous digestion.

SOLUBLE-BASE COOKING

Soluble-base cooking with sodium, magnesium and ammonium bases is by no means, generally speaking, a new concept. Tilghman worked with soluble bases and the first sulphite mill used a magnesium base. However, in the last decade or two there has been a renewed interest, and considerable developments have been achieved. Soluble-base cooking has been a most popular subject for both pure and applied chemical research during recent years. The change from calcium to sodium, magnesium or ammonium will make it possible to change the sulphite process so that almost all the disadvantages, and also the advantages, can be influenced favourably. Of course, all these improvements do not occur simultaneously.

The great advantage of soluble bases is that they allow the pH to be used as an independent variable. It might, therefore, be convenient to discuss soluble-base sulphite cooking from the variable pH point of view.

The pH plays a most important rôle in chemical reactions, and sulphite pulping is no exception. It must be further considered that there are numerous reactions taking place during a sulphite cook, and that these are influenced in different ways by the change of pH. To take some examples: the sulphonation of lignin and the different behaviour of the so-called X-, Y- and Z-groups; the dissolution or hydrolysis of lignin, the hydrolysis of carbohydrates; the splitting off of different substituents of hemicelluloses, such as acetyl and glucuronic acid groups, and of sidechains, which will change the physico-chemical behaviour of the hemicelluloses. It is, therefore, easily understood that pH is a most important variable, and that by controlling this at one or several levels in a sulphite cook, new types of pulps and improvements of different types can be expected. A great number

of different pH combinations, especially in two- and three-stage cooking is possible, but so far only a few have been extensively studied and discussed.

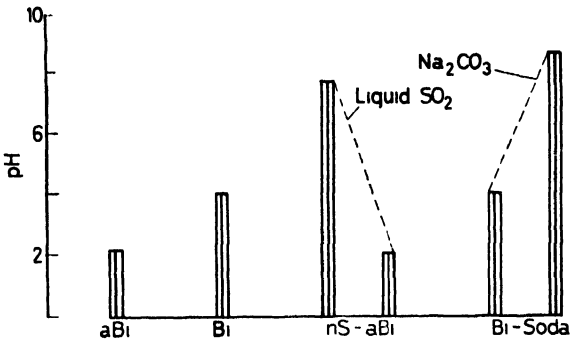


Figure 1. Some soluble base sulphite cooking methods: aBi: acid bisulphite; Bi: bisulphite; nS: neutral sulphite

Figure 1 shows graphically the new sulphite cooking methods. To the left is the conventional acid bisulphite cooking, but with soluble bases; however, this method is neither new nor very exciting. Then there is the bisulphite cooking at about pH 4. The Magnefite and Arbiso processes belong to this group. The next bars on the graph indicate a two-stage cook of the Stora type with the pH between 5 and 8 in the first stage and then, after addition of liquid sulphur dioxide, an acid stage. A modification of this method starts with pH 8, but the second stage is carried out at pH 4. Finally there is an example of the Sivola two-stage sulphite cooking starting with pH 4 and then, after addition of sodium carbonate, ending at pH 9 in the final stage. Some of the merits and limitations of these methods will now be mentioned.

(a) One-stage acid bisulphite cooking

The mere substitution of a sodium, magnesium or ammonium base for a calcium base without changing the pH, does not imply much of a change in pulping. Sodium or ammonium bases give shorter cooking times than calcium and magnesium bases in paper pulp cooking, which means a higher capacity. In making dissolving pulp, sodium and ammonium will give a lower *Kappa*-number, *i.e.* lignin content, at a given D.P. or pulp viscosity with somewhat less chemical consumption in bleaching. The same bases also give less shives, compared with calcium or magnesium base cooking with good cooking acid impregnation. Furthermore, species, like Douglas Fir and larch, containing taxifolin will be more easily pulped by soluble bases.

(b) One-stage bisulphite pulping (pH 4)

The one-stage bisulphite cooking at pH 4 constitutes a new development, although some of the merits of this method were mentioned as early as 1926, when Hagglund found that sodium bisulphite pulps have exceptionally

high strength and also are easy to defibrate in high yields. The cooking acid contains pure bisulphite without excess of sulphur dioxide. At a pH of about 4 and higher the interference of pinosylvin in pine heartwood on the sulphonation of the lignin is negligible. Therefore, all one- or multi-stage sulphite cooking methods with an initial pH of 4 or higher make it feasible to pulp pine. The feasibility of pulping pine is of economical importance to the industry, because pine is cheaper than spruce in many pulp producing areas, and the cost of the wood is often in the order of 50–60 per cent of the production cost of the pulp. This also explains why increased yield, even if very small, is of great importance.

It has been claimed that bisulphite cooking at pH 4 gives higher yields than the conventional sulphite process. If one means higher yield at a given degree of delignification there is only a minor difference between the two types of pulping, as shown in *Figure 2.* Both in the acid bisulphite

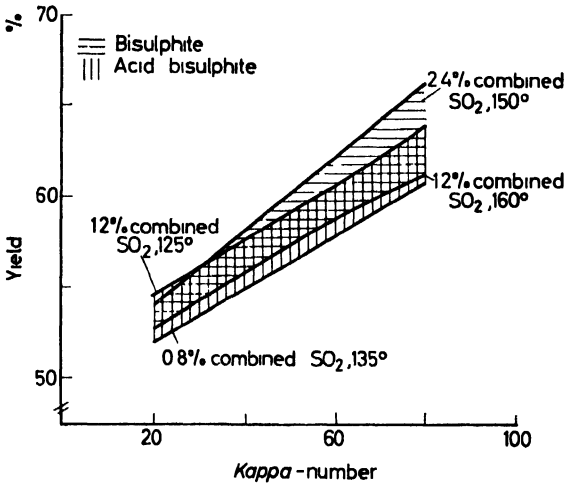


Figure 2. Unbleached pulp yield *v.* lignin-number for bisulphite and acid bisulphite softwood pulps

and the bisulphite cooking, the yield at a given *Kappa*-number can be varied somewhat by changing the cooking temperature or the chemical charge. Decreased temperature, or increased chemical charge, will increase the yield by higher hemicellulose retention in relation to the lignin dissolution. Recent investigations have shown, that this increase in hemicellulose content with higher chemical charge is almost entirely due to an increase in glucomannan and only slightly due to an increase in xylan (*Figure 3*). No explanation can be given for these observations so far.

In another sense, however, the bisulphite cooking can give higher yields. At high *Kappa*-numbers the delignifications seems to be a little more selective, especially at a high chemical to wood ratio. A high chemical charge also gives a higher yield at the *defibration point*, which might be interpreted as a more specific delignification of the middle lamella. This point, which

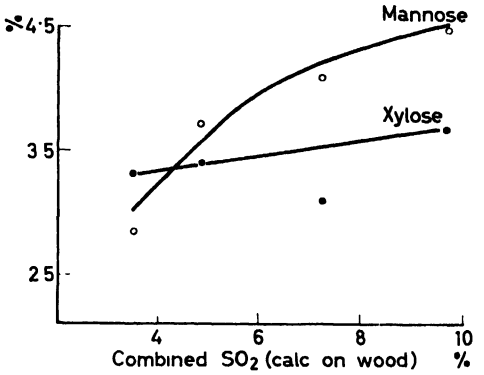


Figure 3. Hemicellulose content of bisulphite pulps *v.* chemical charge in pulping

roughly indicates the maximum yield of what can be denoted as chemical pulp that does not need any mechanical defibration, is in the order of 60 per cent for bisulphite pulp compared with 56 per cent for a good acid bisulphite pulp. Also bisulphite pulps in higher yields, 65–75 per cent, are surprisingly easy to refine compared with conventional sulphite or kraft pulps.

Another attractive advantage of the bisulphite pulps, especially those with sodium base, is their higher quality. The strength properties are somewhere between those of the conventional sulphite and kraft pulps as illustrated in *Figure 4.* Here the tear is plotted against tensile strength, to give the best over-all picture of the strength properties. The upper,

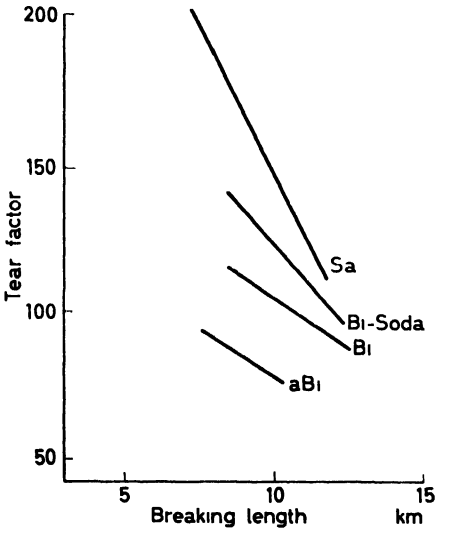


Figure 4. Strength properties of various types of bleached chemical spruce pulps in high yields: aBi: acid bisulphite; Bi: bisulphite; Sa: sulphate

left hand end point of each curve represents the unbeaten stage and the lower, right hand point approximately 550° SR freeness*. The low tear of the sulphite pulp, which really is a pronounced disadvantage in many cases, can be increased by going over to bisulphite pulps, without losing any tensile strength. The high tear is partly due to the greater bulk of the bisulphite pulp over the conventional acid sulphite. Moreover, the tensile strength of these pulps is the same as, or sometimes even greater than, that of kraft pulps made from the same soft wood. It is, furthermore, of considerable technical importance that the initial, unbeaten tensile and burst strength of the pH 4-pulps is very great, and that these pulps, therefore, require very little beating. They are also very easily drained, a property which is of great significance for the modern fast running paper-machines, the speed of which is continuously increasing. The opacity, which also is an important pulp property for the paper-maker, is significantly higher for the pH 4-pulps than for the conventional sulphite pulps.

Some of the merits of the bisulphite pulps, especially the tensile and burst strength, are more pronounced in the higher yield range of 55–60 per cent. The lignin content is, therefore, rather high and, in many cases, too high to allow the pulps to be bleached economically in the conventional way. However, as in the case of other high yield and semi-chemical pulps, peroxide bleaching or other non-delignifying types of bleaching should be applicable. The stability of the brightness will probably not be very good. The problem of making lignin white and stable white has yet to be solved.

The results so far quoted have been obtained using softwoods. For hardwoods, at least for birch and aspen, the bisulphite cooking has been reported to give an even greater increase in strength properties, compared with the conventional acid bisulphite pulping. Also, the opacity of the product is higher. There is a limitation, however, namely that chemical hardwood pulps, at least birch pulps, can be obtained only within a very narrow *Kappa*-number range. The birch bisulphite pulps first become defibrable below a *Kappa*-number of about 40–45. On the other hand it is difficult to get pulps with lower *Kappa*-number than about 25–30 due to decomposition of the cooking liquor, resulting in a loss of bisulphite and the production of dark pulps.

It might be a little too early to state that there is a trend at present towards bisulphite cooking, but there are two mills in Norway, two in Sweden and at least one other in the United States making chemical magnesium base bisulphite (Magnefite) pulp. A new sulphite mill under construction in Sweden will also produce a similar magnesium base bisulphite pulp of both soft- and hardwood.

The bisulphite process will probably also be of great interest for semi-chemical pulps, as has been pointed out by Husband. Some years ago he showed that semi-chemical pulps of a quality at least equal to neutral sulphite semi-chemical (NSSC) pulps can be pulped in a considerably shorter time, and with less consumption of chemicals, with sodium bisulphite at pH 4 than with neutral sulphite. The pulps are brighter than the NSSC-pulps. In Russian papers, semi-chemical bisulphite pulp has also

*SR — Schopper-Riegler : °SR defined by the construction of the equipment, which measures the ability of the pump to be drained.

been stated to be suitable both for liner† and for the corrugated medium. It would, not be surprising, therefore, if there were to be a development of semi-chemical bisulphite pulps. This is also in line with the suggestion from the Abitibi Company.

(c) Two-stage sulphite cooking with second stage acid

Another, and perhaps stronger, trend is towards two-stage sulphite cooking. In 1953 the Soråker Mill (belonging to the Swedish Cellulose Company), in 1956 the Kramfors, and in 1958 the Ortivken Mill, all changed to two-stage pulping with sodium sulphite at pH 8 in the first stage and normal acid calcium base sulphite at pH 1·5–2 in the second stage. One reason for this was that it made the pulping of bark-damaged wood feasible. In 1955 the Stora Company Mill in Skutskar started to pulp pine wood in two stages with a pH between 4 and 8 in the first stage and pH 1·5–2 in the second stage. This is done entirely with sodium base, and with recovery of the chemicals. A new sulphite mill in Nova Scotia, which is going to pulp in the same way, is now under construction. The Mo and Domsjo Sulphite Mill, which normally runs an acid sodium bisulphite dissolving pulp, has also made plans to switch to the same two-stage process at any time. Weyerhaeuser also announced last year that their Cosmopolis mill had changed to a similar two-stage cooking process with a magnesium base.

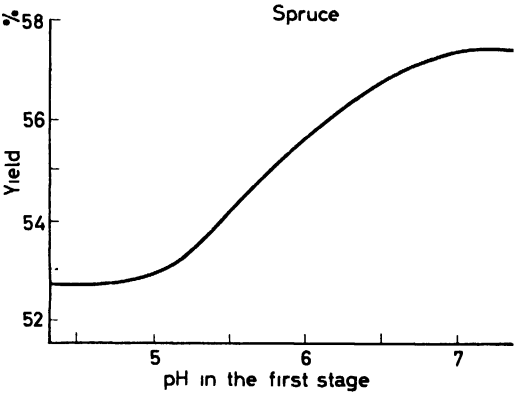


Figure 5. Unbleached yield after two-stage sulphite cooking to *Kappa*-number 20 *v.* pH in the first stage

What are the factors which make this type of two-stage cooking so attractive? It has already been pointed out that any method starting at pH 4 or higher will give a free choice of pulp wood, and that pine can be readily pulped by this process.

A greater advantage of this particular process is, however, the increase in pulp yield, as compared with that achieved in the conventional sulphite process, by the proper choice of pH in the first stage. This increase in yield at a given *Kappa*-number is due to retention of glucomannan, which is controlled by the pH in the first stage (*Figure 5*). Rydholm has shown

†Paper for surface layers of corrugated board.

that the glucomannan retention is most probably a function of the acetyl content. The softwood glucomannan has no substituents apart from the acetyl groups. By splitting these off, the straight glucomannan chains will more easily be attached to the cellulose by hydrogen bonds. The conditions during the first stage, and especially the pH, will determine the amount of residual acetyl groups, and thus also the glucomannan retention. With a sodium base the optimum glucomannan rentention will easily be attained. It has recently been shown that with a magnesium base the same glucomannan retention can also be achieved.

For softwood pulping this two-stage process obviously has great advantages in so far as the yield of paper pulps is concerned. In the manufacture of dissolving pulp with high α-cellulose content, no hemicellulose retention is desired. Hardwoods, on the other hand, do not contain any glucomannan worth mentioning, and therefore no yield increase is obtained by high pH in the first stage.

Because of their high hemicellulose content these softwood two-stage pulps with high pH in the first stage are very easy to beat. They are especially suitable for the manufacture of greaseproof paper, but are useful also for other types of paper, especially if the maximum yield increase is purposely not achieved and the beating is carefully done. Under these conditions the pulp has approximately the same strength properties as a conventional acid bisulphite pulp from the same wood.

A modification of the method suggested by Rydholm involves cooking at pH 8 in the first stage, but pH 4 in the second. This would result in the same yield increase as if the second stage were more acidic, but the resulting pulp will show somewhat better strength characteristics, fully comparable with the conventional acid bisulphite pulp.

(d) Two-stage sulphite cooking with an alkaline second stage

There is also another type of two-stage sulphite cooking, which was worked out some years ago by Sivola and others, and practised industrially since about 1954 in the Finnish Rauma Mill. The last carbonate stage, at about pH 10, is typical of this process, often called the Sivola process. The preceding stage or stages are chosen according to the type of pulp to be produced and the pulpwood (*Figure 6*). For paper pulp the first stage is usually carried out at pH 4. For dissolving pulp the preceding stage is either an acid bisulphite stage, or a pH 4-stage followed by an acid bisulphite one. The latter type is of course necessary if pine has to be used. In all cases the pH increase in the last stage is accomplished by the addition of sodium carbonate. This stage is equivalent to a separate hot alkali refining stage, and from the standpoint of the quality of the pulp produced the carbonate stage can be a separate one as well. By including it as a part of the cooking process some advantages are obtained: the sodium carbonate will be a part of the chemicals in the recovery system, and, thus, the cost for chemicals will be less than with the separate stage; the organic substance dissolved in the alkaline stage will be collected in the waste liquor and burnt together with the other dissolved wood substance from the first stage, and will thus not contribute to water pollution; also the heat economy will be better.

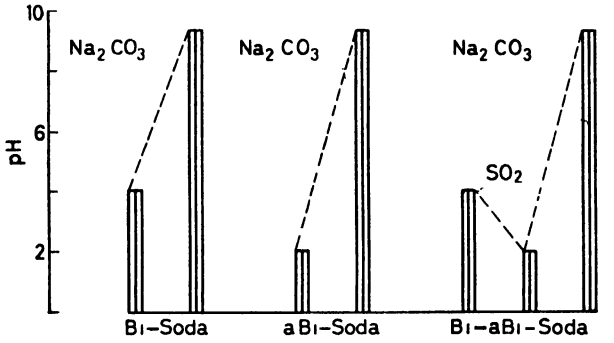


Figure 6. Various types of Sivola pulps: aBi: acid bisulphite; Bi: bisulphite

A separate hot alkali refining stage, which is a part of the bleaching sequence for dissolving pulps, will break down the hemicelluloses by peeling and dissolve them. The action of the carbonate stage in a Sivola cook is similar. The dissolution of hemicelluloses is controlled by the amount of carbonate, time and temperature. Alpha cellulose contents as high as 97%, which also is the maximum for conventional hot alkali refining, can be reached.

The two-stage bisulphite–soda cook will give paper pulps of exceptionally good strength properties, approaching those of kraft pulp. By courtesy of the Billerud Company some of their data has been made available. *Figure 4* demonstrates that the bisulphite soda pulp has an intermediate position between the kraft and bisulphite pulps as far as strength is concerned.

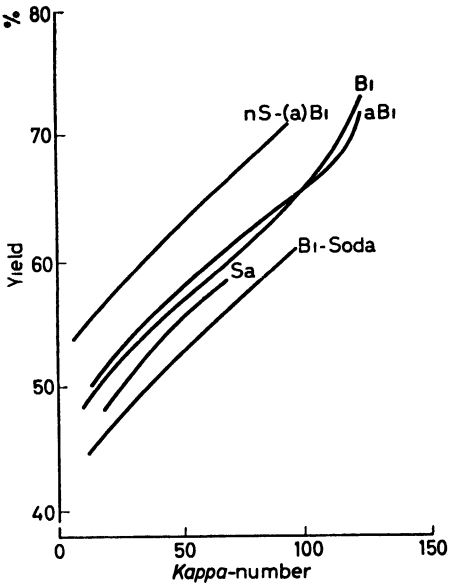


Figure 7. Unbleached yield of various types of spruce pulps *v.* lignin-number: aBi: acid bisulphite; Bi: bisulphite; nS: neutral sulphite; Sa: sulphate

The pulp is rather bright, and as easy to bleach as a sulphite pulp. But, unfortunately, there is a sacrifice to be made in order to get this pulp, and that lies in the low yields obtained. Bisulphite soda pulps are shown in *Figure 7* to have an even lower yield at a given *Kappa*-number than the kraft pulps. The relative order of pulps will still be the same after bleaching. By choosing milder conditions in the carbonate stage the yield of the carbonate pulp can be increased to that of a kraft pulp, or even higher, but, at the same time, the strength properties will be lower. The strength properties of the pulps shown in *Figure 4* were those obtained after the bleaching of the pulps shown in *Figure 7*.

By the introduction of the soluble bases, and by the utilization of the possibilities they offer, especially in making the pH an independent variable, the sulphite process can be made highly flexible with regard to the choice of pulp wood and product produced. The economically important pulp yield, which for the conventional sulphite pulp is already higher than for kraft, can be further increased, with a simultaneous increase, in some cases, in tensile strength, tear, and opacity.

(e) Recovery

The soluble bases are expensive and the chemicals must of necessity be recovered. The recovery in magnesium-base sulphite pulping is well established and simple. For sodium bases several recovery methods have been worked out, but so far only a limited number are used commercially. These are more complicated than the magnesium recovery process, and require a rather high capital investment. It is, therefore, out of the question for a small mill making chemical pulp to install a sodium recovery system; the smallest size has a capacity of the order of 50–60,000 tons per year. However, in several countries, there is extensive work going on in order to develop simple methods for sodium base recovery. There is hope, that we, in a few years time, will develop new methods, more simple than the present ones and requiring less capital investment. Sodium bases are generally preferred to magnesium bases for greater flexibility in the cooking process and easier delignification. Interest seems at present to be divided equally between sodium and magnesium bases, but if simpler recovery methods are worked out for sodium bases they might command the greater interest.

IMPROVED CHIPPING

It is a well-known fact that the conventional chippers damage the chips by exerting a pressure parallel to the fibres. The fibres are corrugated in the damaged area. Probably disorientation occurs where the fibres are bent. Then, during the sulphite cook, fibres are weakened by hydrolytic attack at the points of disorientation in the fibre wall. This weakening does not occur in kraft pulping. The damaged sulphite fibres will easily break into fibre fragments on beating. The freeness will be low and the strength properties of the pulp, tear, tensile, and burst, will be some 10–15 per cent lower than the pulp from undamaged hand-made chips. Logan at Anglo-Canadian has now succeeded in his development of a new type of drum chipper which will not cause any fibre damage. In industrial

operation this has increased the strength properties of the sulphite pulp by about 5–15 per cent. This is a most valuable contribution to the development of the sulphite pulp quality. It can be predicted that this basic idea in chipper design will be adopted by the sulphite industry, especially by the market pulp mills.

IMPROVED BLEACHING

A trend which is more clearly visible for market pulp mills, as least in Scandinavia, than for integrated mills is worth mentioning briefly. The competition in the market has created an abnormal demand for high brightness, which is not always based on a demand for this high brightness from the ultimate consumers of the end products of the pulp. The use of chlorine dioxide in the final stage in sulphite bleaching has, therefore, become more common. Even typical kraft pulp bleach sequences have been adopted by the sulphite industry. One Swedish sulphite pulp mill has a five-stage operation, including two chlorine dioxide stages, and a new sulphite mill will employ a similar system.

CONTINUOUS SULPHITE COOKING

Although this is a wood chemistry symposium I will mention a recent technical development which, however, does not yet constitute a trend. The Billerud Company in Sweden has solved the technical and chemical problems involved in continuous sulphite digestion. Their experimental continuous digester, with a capacity of about 10 tons a day, can now produce an acid bisulphite pulp of the same quality as the normal batch pulp. The sulphite liquor decomposition is not a problem in the continuous acid bisulphite pulping, but is troublesome in the continuous bisulphite pulping to low *Kappa*-numbers. The experimental digester is a twin tower arrangement, in which a variety of two-stage cooks has also been carried out successfully.

CONCLUSION

Finally, I should like to mention that all these developments are the result of fundamental and technical research. It has been facilitated by the free exchange of ideas and results which prevails in the pulp and paper industry. However, further results are needed to get a better knowledge of the chemistry of the wood components and a better understanding of the chemistry of the technical processes. On this basis further developments will be made.

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NEW INFORMATION ON THE REACTIONS OF LIGNIN IN KRAFT PULPING

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EARLIER WORK ON THE PART PLAYED BY SULPHUR IN KRAFT PULPING

The part played by sulphur in kraft pulping has been investigated extensively, *i.e.* by the Swedish chemists Klason, Holmberg and Hägglund. It has been found to be very complicated. My co-workers and I have investigated one of these experimentally which holds that the main effect is caused by the reducing properties of sulphide or hydrogen sulphide. In fact, the redox potential E_h, measured after the digestion at pH 11, in a kraft liquor has a value of about $-0{\cdot}30$, whereas the corresponding soda or sodium hydroxide liquor shows a value of about $-0{\cdot}15$. It is well known from the work of Schuerch and co-workers[1], among others, that pulping of wood can be achieved by hydrogenation in alkaline solution in the presence of catalysts. In our experiments[2] we adjusted the redox potential of an alkaline cooking to the same value as for a kraft liquor, with the aid of a reducing reagent liquor other than sodium sulphide; we used pyrocatechol and other reducing reagents. The effect of the cooking was, however, still about the same as with sodium hydroxide alone. Thus, the main part of the rôle played by sulphur in kraft pulping cannot be explained by the reduction theory.

Modern theories about the kraft process are founded on the assumption that some active group in lignin reacts chemically with a sulphide ion, *i.e.* with a sulphide or hydrogen sulphide ion. During the digestion an amount ranging from about one-fifth to one-half of the sulphide is consumed, depending on the concentration of sulphide used[3]. The sulphur consumed is mainly found organically bound in the kraft or thiolignin. The latter is dissolved during the digestion and can be precipitated if the liquor is acidified after the cooking. That kraft lignin contains, as a rule, about 2·5 per cent of organic sulphur. It is, however, not homogeneous[4] but contains fractions of very different molecular weight, varying from about 7,000 according to Gralén[5], to about 1,000 or less. The low molecular, dialysable fraction seems to contain more sulphur, about 4 per cent, the high molecular part less, about 2 per cent or less[6].

Research carried out by Hägglund, my co-workers and myself in Stockholm and Helsinki proved that lignin can be sulphurized to thiolignins which have sulphur contents varying between about 2 and 21 per cent, and very different solubilities, stabilities, reactivity and spectroscopic properties. The scheme in *Figure 1* was published 7 years ago[7] and shows the various types of thiolignins. If spruce wood is digested in neutral or almost neutral sodium hydrogen sulphide solution of pH 7 or 8·5 there

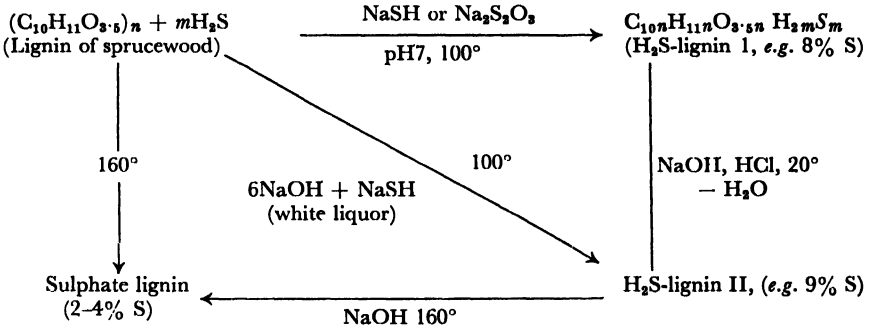


Figure 1. Scheme for the formation and reactions of thiolignins (By permission *Tappi* **37**, 350 (1954))

is addition of the elements of hydrogen sulphide. A hydrogen sulphide lignin I is formed, with, for instance, 8 per cent sulphur content. At first it remains in the wood, but microphotographs in ultra-violet light show that it melts and flows out in the open spaces, the lumina, of the wood fibre cells[8]. This thiolignin can be dissolved easily in alkaline solutions, and also in neutral organic solvents, such as 75 per cent alcohol, alcohol–acetone or dioxan. If carefully prepared at 80°, it shows a sharp ultra-violet spectrum of about the same type as the lignosulphonic acids. It is, however, unstable. On dissolution in an alkaline solution at room temperature and reprecipitation with hydrochloric acid, it loses the elements of water. Thus, it forms hydrogen sulphide lignin II, which has a higher sulphur content, *i.e.* 9 per cent, and a rather flat ultra-violet absorption curve. This spectral change is probably caused by the formation of carbonyl groups, or other groups with double bonds, in conjugation with the benzene nucleus[9]. By heating with sodium hydroxide solution at 160°, hydrogen sulphide lignin II loses half or more of its sulphur in the form of sodium sulphide. Thus the lignin is changed to a product very like kraft lignin. Also at the beginning of a usual kraft digestion, at 100°, a lignin, often rich in sulphur, which closely resembles the hydrogen sulphide lignin II, is formed. Comparative studies of the dissolution of lignin from wood at different temperatures with kraft and sodium hydroxide liquors have been made[2,10] In those studies, the greatest difference has been observed at early stages of the digestion, at 130 or 140°. At this temperature, the dissolution of lignin in the kraft cooking proceeds continuously. In contrast, digestion with sodium hydroxide leads, at first, to the same rapid dissolution of the lignin as in the kraft pulping, but when about a third of the lignin has been dissolved, practically no more lignin is soluble in sodium hydroxide at 140°. This shows that the explanation of the part played by sulphur in kraft pulping is to be sought in the early stages of the digestion, and in the thiolignins which are formed as intermediates. Further, a great number of widely varied sulphurizations of spruce wood have shown that only those thiolignins which have a sulphur content of not less than 4 per cent can be dissolved from wood by neutral or alkaline solvents at low temperatures. The facts point to a kind of cyclic process comprising an addition and

subsequent elimination of hydrogen sulphide, which can then react again with new molecules of lignin. The sulphide ions thus react as a kind of catalyst. The final kraft lignin in the black liquor, with its sulphur content of about 2 per cent only, is a product formed by rather deleterious side reactions, a kind of catalyst poisoning, in which utilizable sulphur has been bound to stabilized, non-reactive sulphur compounds, and can no longer be removed by alkali.

Then we have the question of which specific groups in lignin react with sulphur. Moilanen and I[11] have made a series of sodium hydrogen sulphide digestions at 100° and pH 6–9 as has Gierer in Stockholm[12]. The results make it seem very probable that it is the so-called *X*- groups which react with sulphide ions (*Figure 2*). The *X*-groups are *p*-hydroxybenzylalcohol

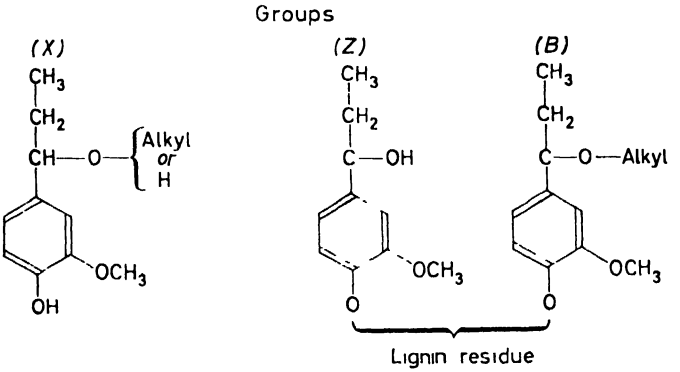


Figure 2. *X*-, *Z*- and *B*-groups of lignin

or corresponding benzyl alkyl ether groups. The sulphur attacks the carbinol carbon atom, and splits the benzyl alkyl ether group. In the case of the *Z*- and *B*-groups, the phenolic hydroxyl group is etherified and no sulphur is introduced. This contrasts with the sulphite digestion, where *Z*-groups also react directly with the reagent, and *B*-groups react after hydrolysis.

Measurements of the contents of free phenolic hydroxyl groups have been carried out by Freudenberg and Adler and their co-workers[13], and also by my co-workers and me[14]. They show that milled wood lignin, prepared according to the method of Björkman, and probably also the protolignin of wood, contains about one free phenolic group in every group of three phenylpropane units. The hydrogen sulphide lignins contain somewhat more, *i.e.* about one free phenolic hydroxyl group for every two phenylpropane units. Thus, in protolignin only some of the benzyl ether bonds can be cleaved by sulphidic ions. There are also other types of bonds in lignin, for instance phenol ether bonds and carbon—carbon bonds, especially in the 5-position. There is good reason to believe that many new carbon—carbon bonds are formed in this position during alkaline pulping[15]. Hence, it is not surprising that it is impossible to make pulp from wood by digesting with pure sodium hydrogen sulphide solutions. However, in kraft pulping we have also hydroxyl ions; they are, in fact,

present in large amounts. I have already proposed the theory that the dissolution of lignin in kraft pulping is a result of concerted action by hydrogen sulphide ions and hydroxyl ions. The hydroxyl ions would cleave phenol ether bonds, and thus set new phenolic hydroxyl groups free. Then the hydrogen sulphide ions would split benzyl alkyl ether bonds. An interesting paper read by Gierer at the lignin symposium in Helsinki in June 1961[16] throws new light on this hypothesis. On the basis of model experiments, Gierer concludes that the hydroxyl ions are probably only able to cleave those phenol ether bonds in which the benzene nucleus has its phenolic hydroxyl group etherified, as in the *Z*- and *B*-groups. Thus, we should have a clear division of functions: the sulphidic ions splitting the H-groups and the hydroxyl ions splitting *Z*- and *B*-groups, thus creating new *X*-groups. Those can then in their turn be cleaved by sulphidic ions. This hypothesis could be called the *ether cleavage* hypothesis. Until now important questions in connection with this hypothesis have been left open for discussion. Firstly, it is of interest to determine the extent to which facts obtained experimentally are valid for the kraft pulping reaction, which proceeds in strongly alkaline solution. Secondly, it is highly probable that lignin contains many alkyl aryl ether bonds in the β-position. We should like to know what happens to these bonds during sulphurization.

The form in which the sulphur in the kraft liquor reacts with lignin has been the subject of some discussion. The hydrogen sulphide molecule, the hydrogen sulphide ions and the sulphide ions have all been advocated. Quantitative studies of the formation of alcohol-soluble thiolignin from spruce wood on cooking with sodium hydrogen sulphide solutions of varying pH have been made[8]. These studies have shown that the increase in the net yield of thiolignin in the range pH 5·5–8·5 follows the rise of the hydrogen sulphide ion concentration, but is inversely proportional to the concentration of undissociated hydrogen sulphide. From pH 8·5 upwards the yield remains roughly constant, in spite of the fact that at higher pH values part of the sulphur occurs as the bivalent sulphide ion. Thus, it seems that hydrogen sulphide ions and sulphide ions alike contribute to the formation of thiolignin, in contrast to the undissociated hydrogen sulphide molecule.

It is, however, probable that the thiolignin formation occurs in a somewhat different manner with hydrogen sulphide ions than with sulphide ions, *i.e.* in almost neutral as compared with strongly alkaline solutions.

SULPHATE DIGESTIONS OF THE LIGNIN MODEL VANILLYL ALCOHOL

There have been some discrepancies in the results of the sulphurization of the lignin model vanillyl alcohol at 100° and pH $\sim$ 7. Moilanen and I obtained vanillyl disulphide[11], whereas Mikawa and co-workers got a mixture of vanillyl disulphide and vanillyl monosulphide[17] After digestion of wood with sodium hydrogen sulphide at pH 7, some disulphidic sulphur was found by iodometric titration[7,18], in contrast to the results of analogous titrations of commercial kraft lignin. It can be assumed that the

vanillyl disulphide and the disulphidic sulphur in the sulphurized wood have been formed from the corresponding mercapto groups by aerial dehydrogenation. This has probably happened not so much during the digestion itself but during the isolation and purification of the products. This will be illustrated by the results of Hastbacka, at Helsinki[19]. He carried out kinetic studies of the reaction of vanillyl alcohol at 75–95° in strongly alkaline solutions which contained sodium hydroxide and, in most cases, sodium sulphide, corresponding to the composition of commercial soda or kraft white liquors. He rigorously excluded the air and succeeded in determining the reaction products by quantitative paper chromatography and colorimetry to an accuracy of about ±1 per cent. This is much better than the accuracy achieved in most earlier chromatographic experiments. Some of his results are presented in *Figure 3.*

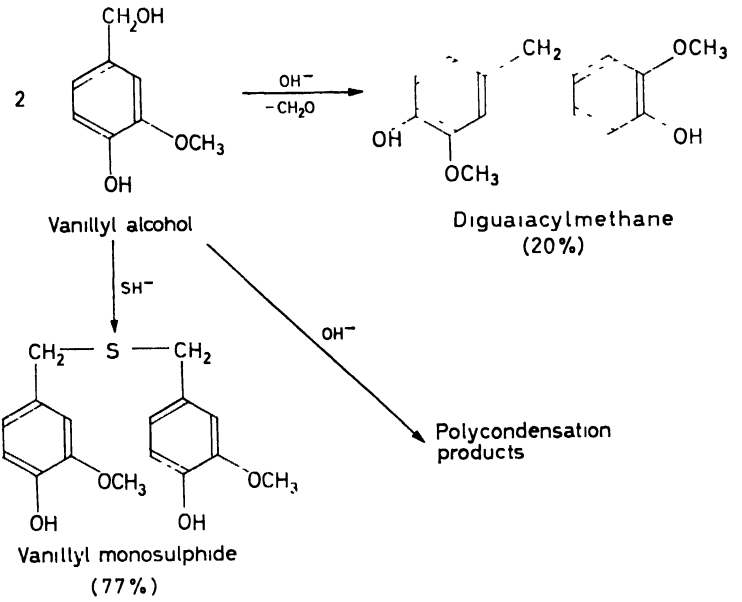


Figure 3. Reactions of vanillyl alcohol

This shows that in the presence of sulphide ions the main product was vanillyl monosulphide, with 77 per cent of the theoretical yield. No vanillyl disulphide or vanillyl mercaptan could be detected. The sulphurization reaction was found to be first order; the reaction rate of the consumption of vanillyl alcohol in sodium sulphide solution, as well as in sodium hydroxide solution, is decreased on increasing the concentration of the inorganic reagent. Hästbacka explains this behaviour by assuming that the rate-determining step in both cases is the formation of a quinone methide as an intermediate, and that this quinone methide then adds to vanillyl mercaptan or vanillyl alcohol. The formation of disulphide in the experiments by Moilanen and me might then perhaps be explained

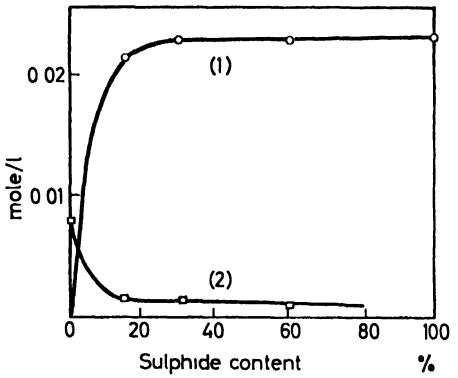


Figure 4. The yield of vanillyl monosulphide (1) and diguaiacyl methane (2) as a function of the sulphide content after a reaction time of 8 h at 95·6° C; initial concentration of vanillyl alcohol: 0·0648 mole/l; active alkali content: 13·4 g Na_2O/l

as a kind of " catching reaction ", what the Germans call " Abfangreaktion ", for the intermediate vanillyl mercaptan by the oxygen of the air.

From the first line of *Figure 3* we see that on digestion of vanillyl alcohol with sodium hydroxide solution, diguaiacyl methane was obtained in 20 per cent yield in addition to polycondensation products. This is a polycondensation reaction very similar to that which proceeds in the reaction of phenols with formaldehyde during the formation of plastics of the bakelite type.

These results of Hästbacka have a bearing on an important aspect of the part played by sulphur in the kraft process, namely the hypothesis—or as we might now call it—the theory of the blocking effect. This theory was originally proposed by Hagglund in 1941[20]. It holds that the main effect of the sulphide ions would be a blocking by sulphurization of reactive groups in lignin, especially phenol alcohol groups. Thus, condensation

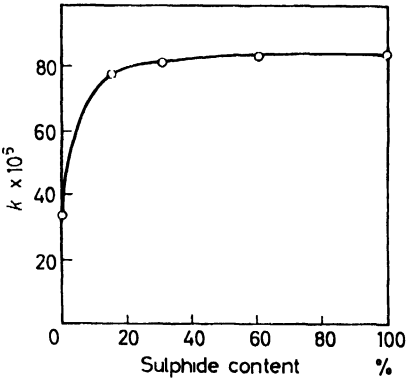


Figure 5. The effect of sulphide content on the rate of consumption of vanillyl alcohol at 95·6° C; concentrations, see *Figure 4*

reactions of the type involved in the formation of phenol–formaldehyde plastics are prevented. The results of Hästbacka make it seem most probable that this theory of the blocking effect holds true, at least in the case of vanillyl alcohol. Furthermore, his experiments with various sulphide concentrations show that the formation of vanillyl monosulphide predominates over the formation of diguaiacyl methane and polycondensation products at sulphide contents as low as 20 or 15 per cent (*Figure 4*). The rate of consumption of vanillyl alcohol was roughly twice as fast in the presence of sulphide ions as with pure sodium hydroxide. It is quite clear that this effect also holds for sulphide concentrations down to about 15–20 per cent (*Figure 5*). Thus, the influence of the sulphide content is very much the same as in kraft digestions where sulphide concentrations of 15–20 per cent in the white liquor suffice to bring about the favourable effects of kraft pulping. It is an interesting fact that the simple lignin model vanillyl alcohol can imitate so closely the effects of sulphidity concentration at the kraft digestions, although all kinds of carbohydrate compounds are absent in the digestions.

THE DEGRADATION OF SPRUCE LIGNIN BY SULPHIDE IONS TO ACETOGUAIACOL, PROPIOGUAIACOL AND CONIFERYLALDEHYDE

The results obtained by Hastbacka also, in a way, provide an argument against the ether cleavage theory. They prove that sulphide ions, do not instigate degradation as assumed by the ether cleavage theory, but can also condense a monomeric lignin model, vanillyl alcohol, to a dimeric product, vanillyl monosulphide, of higher molecular weight. However, in May 1961 another of my co-workers, Ashorn, produced results[21] which can be said to develop the ether cleavage theory.

Ashorn detected that spruce wood, on digestion with a sodium hydrogen sulphide solution of pH 8·5 for a period of 18 days at 100° in a rotating autoclave of stainless steel, formed an ether-soluble phenolic fraction in an

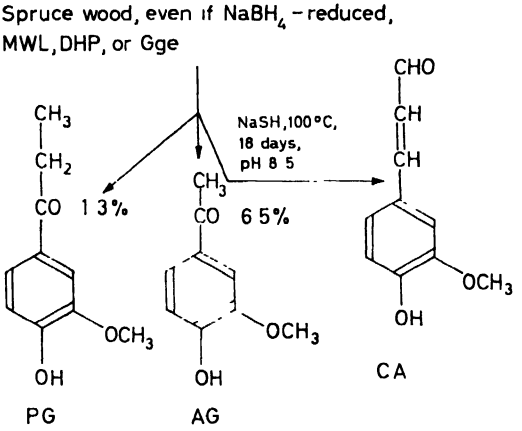


Figure 6. Formation of acetoguaiacol, propioguaiacol and coniferylaldehyde from spruce wood and other materials

amount of about 10 per cent calculated with reference to the lignin of the wood. The main substances in this fraction were (*Figure 6*) acetoguaiacol (AG) 6·5 per cent, its homologue propioguaiacol (PG) 1·3 per cent, and, further, coniferyladehyde (CA). Neither of the ketones could be obtained by a corresponding digestion of spruce wood with a sulphur-free phosphate buffer solution of pH 8·5, and the yield of coniferylaldehyde was much less in the sulphur-free digestion. Similar results were obtained on comparing digestions of milled wood lignin (MWL) prepared according to the method of Bjorkman, and also from the Freudenberg enzymatic dehydrogenation polymer (DHP) of coniferyl alcohol. In this respect this dehydrogenation polymer also resembles the protolignin of spruce wood. Curiously enough, the lignin model of Adler, the guaiacylglycerol-β-guaiacyl ether (Gge), also gave the same products. This must be the result of complicated rearrangement reactions. Even spruce wood which had been treated with sodium borohydride in order to reduce any carbonyl groups in the lignin, gave the same reaction products. Thus, their formation is not dependent on any pre-formed carbonyl groups in the wood.

The yield of coniferylaldehyde after 24 hours was only 0·08 per cent of the lignin obtained from unreduced spruce wood, and 0·25 per cent from sodium borohydride reduced wood. On prolonged digestion it was still lower. *Figure 7* shows that coniferylaldehyde is not stable on digestion with sodium hydrogen sulphide solutions. It is thus possible that fairly large amounts of the lignin are degraded to coniferylaldehyde which then reacts further, probably forming resinous products.

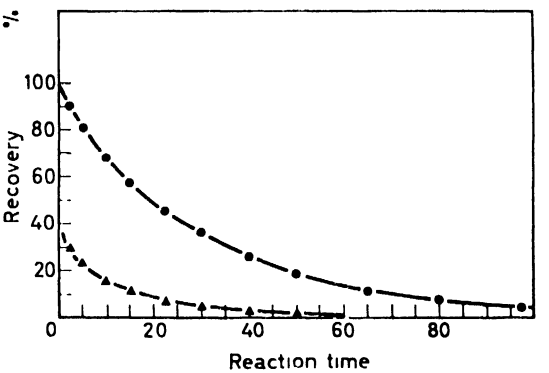


Figure 7. The stability of coniferylaldehyde to digestion with sodium hydrogen sulphide (—▲—) and sulphur free (- -●—) buffer solutions at 110° and pH 8·5

Figure 8 shows some further results by Ashorn obtained in similar digestions with sodium hydrogen sulphide or phosphate buffer solutions. We see that acetoveratrol is easily demethylated to acetoguaiacol even by the buffer solution. This probably has something to do with the fact that acetoveratrol is a vinylogue of methyl acetate and is thus easy to hydrolyze. Hydrochloric acid lignin gives acetoguaiacol and propioguaiacol, but several model substances for lignin, including guaiacylglycerol, do not.

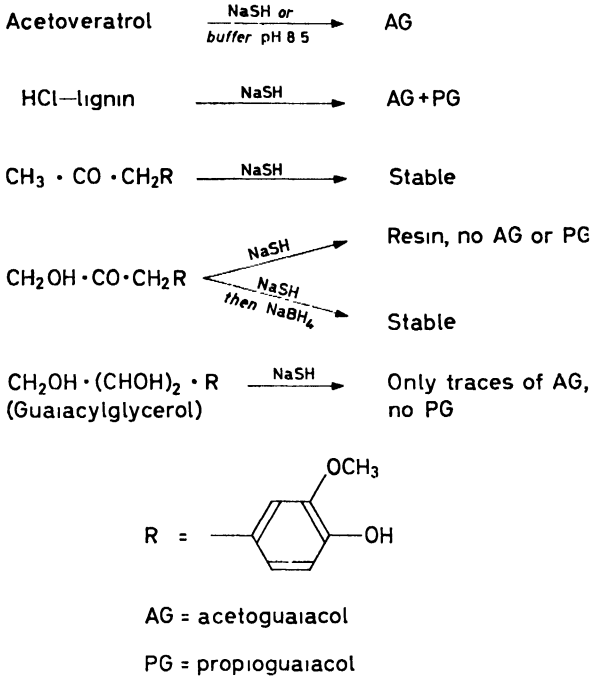


Figure 8. Sodium hydrogen sulphide digestions of lignin models and hydrochloric acid lignin

Thus, the formation of acetoguaiacol, propioguaiacol and coniferylaldehyde under these conditions would appear to be very specific.

Figure 9 shows the behaviour of the lignin model ω-hydroxypropioguaiacol. With sodium hydrogen sulphide this substance gives an organic mono-

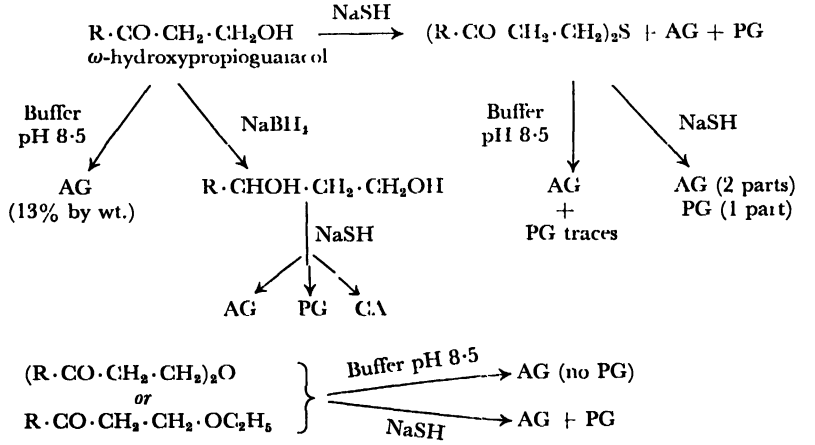


Figure 9. Reactions of ω-hydroxypropioguaiacol and some of its ethers

sulphide, which on further digestion is split into acetoguaiacol and propioguaiacol. ω-Hydroxypropioguaiacol was reduced with sodium borohydride to the corresponding diol. On sodium hydrogen sulphide digestion the latter gives acetoguaiacol and propioguaiacol, as well as coniferyl aldehyde. Also alkyl ethers of ω-hydroxypropioguaiacol give acetoguaiacol and propioguaiacol on sulphurization.

It may be mentioned that Oksanen, at Helsinki has obtained strong chromatographic evidence in four different systems[22] indicating that free ω-hydroxypropioguaiacol occurs in the bark of pine. Free guaiacylglycerol also seems to be present. After spruce wood had been digested with sodium hydrogen sulphide Ashorn found a chromatographic spot which could be due to ω-hydroxypropioguaiacol.

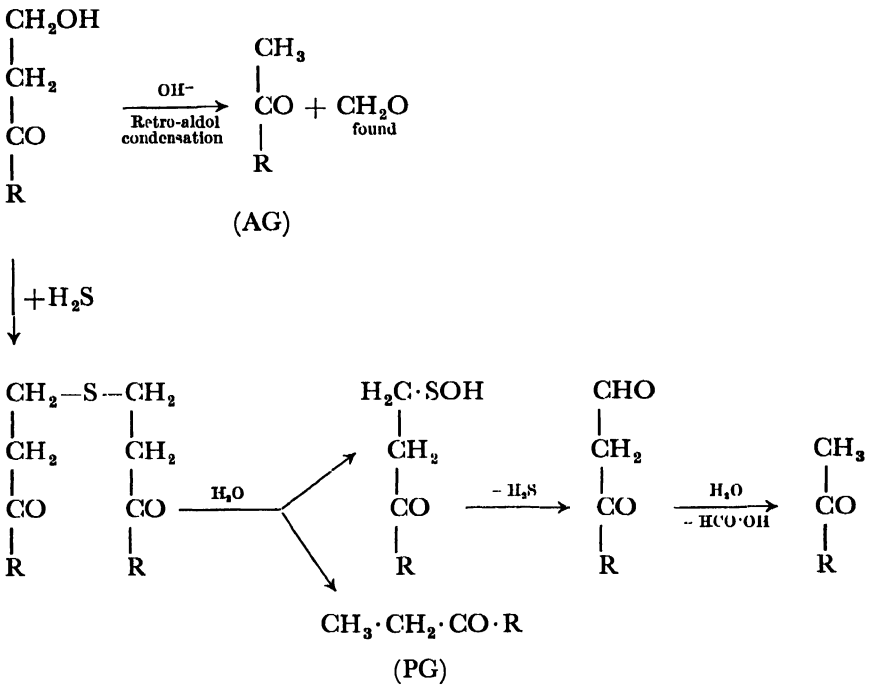


Figure 10. Formation of acetoguaiacol and propioguaiacol from ω-hydroxypropioguaiacol

Figure 10 gives a possible reaction mechanism for the formation of acetoguaiacol with an alkaline buffer solution in the absence of sulphur; the retro-aldol reaction would lead to the formation of formaldehyde, which was, in fact, found in the solutions. The figure also indicates mechanisms for the sulphurization, for the conversion of the sulphide to propioguaiacol, and, *via* a sulphenic acid, to acetoguaiacol.

At this stage I might mention that acetoguaiacol[23] in particular, but also propioguaiacol, have been found in considerable amounts in kraft black liquor, and also that acetoguaiacol is formed on further heating of

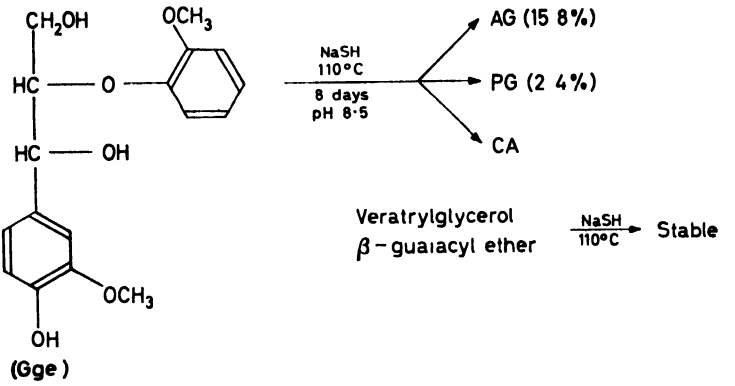


Figure 11. Yields of acetoguaiacol and propioguaiacol from guaiacylglycerol-β-guaiacyl ether

isolated kraft or even soda lignin with sodium hydroxide solution at 170°[23,24]. Acetoguaiacol can thus be formed, even without any sulphur containing reagent, but its formation is very much stimulated by the presence of sulphide ions.

It would be tempting to assume that the formation of acetoguaiacol, propioguaicol and coniferylaldehyde from lignin on digestion with sodium hydrogen sulphide proceeds *via* ω-hydroxypropioguaicol. In Ashorn's opinion it is not impossible that ω-hydroxypropioguaiacol could be formed from guaiacylglycerol β-guaiacyl ether (Gge) and analogous groups in lignin (*Figure 11*). With sodium hydrogen sulphide this substance gives a considerable yield of acetoguaiacol, and also some propioguaiacol and coniferylaldehyde. If the phenolic hydroxyl group in Gge is etherified we arrive at veratrylglycerol β-guaiacyl ether, which, however, is stable under the sulphurization conditions. This coincides well with the

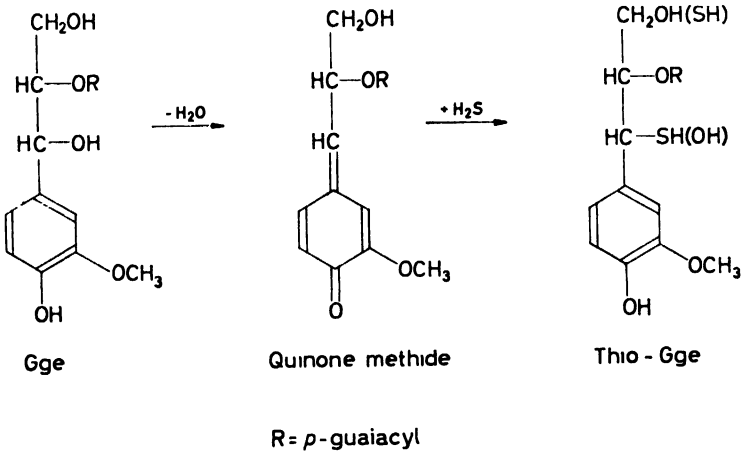


Figure 12. Sulphurization of guaiacylglycerol β-guaiacylether

assumption that the reaction (*Figure 12*) involves a quinone methide intermediate. The latter cannot be formed if the hydrogen atom of the free phenolic hydroxyl group has been substituted with a methyl group, or a group larger than methyl. By addition of hydrogen sulphide the quinone methide can give thioguaiacylglycerol β-guaiacyl ether. Later, I will give experimental evidence which makes it probable that the thiocompound and the corresponding group in hydrogen sulphide lignin have their sulphur partly in the γ-position. This is indicated by the parentheses in the formula. Then (*Figure 13*) the phenol ether group OR is split off under the influence of the sulphur atom, for instance by formation of an episulphide intermediate. This would be similar to epoxides known in analogous fissions of phenolglycosides and saccharides. Epoxides are also supposed by Gierer to be intermediates in the digestion of lignin with sodium hydroxide. Such an episulphide would then by hydrolysis and rearrangement eventually give ω-hydroxypropioguaiacol. If treated with more hydrogen sulphide ion or mercaptan the episulphide would give a dithiol or a thiol ether. This would then be rearranged into a model for a hydrogen sulphide lignin, with a high sulphur content. *Figure 14* again shows the way from ω-hydroxypropioguaiacol to acetoguaiacol and propioguaiacol, and, further, how coniferylaldehyde can be formed from the thioguaiacylglycerol

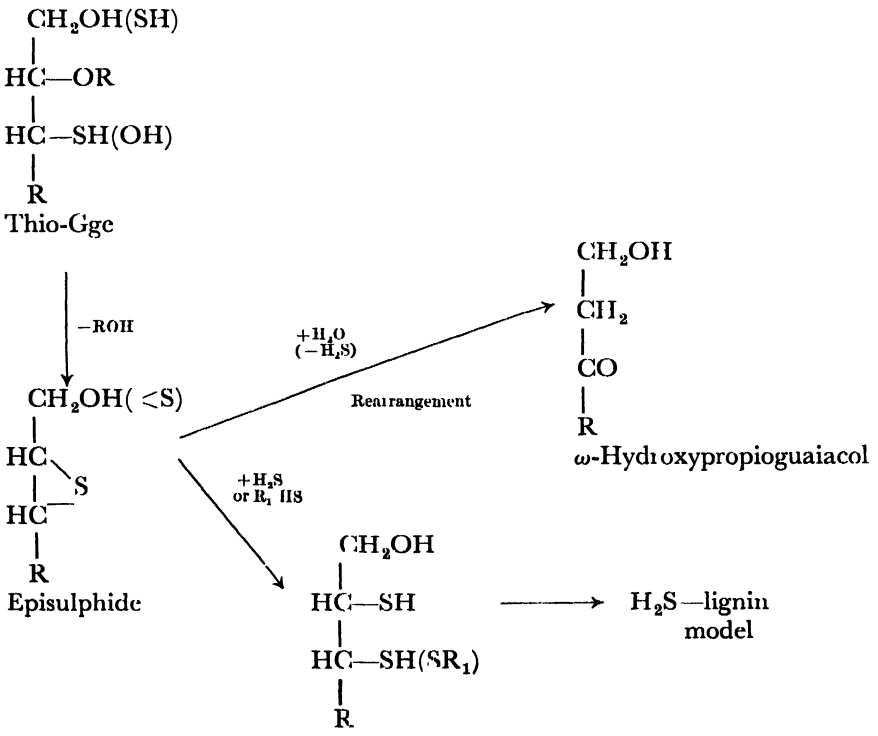


Figure 13. Formation of ω-hydroxypropioguaiacol and a hydrogen sulphide lignin model *via* a hypothetical episulphide; R_1 = alkyl group

β-guaiacyl ether and analogous groups in lignin. The ratio of acetoguaiacol to propioguaiacol obtained, and also the rate of the formation of acetoguaiacol are considerably greater in the sulphurization of guaiacylglycerol β-guaiacyl ether than in the corresponding treatment of ω-hydroxypropioguaiacol. Therefore, Ashorn assumes that at least one other reaction mechanism, involving an unknown intermediate, is at work at the same time, forming about two thirds of the acetoguaiacol. The remaining third of the acetoguaiacol, and the whole of the propioguaiacol, could be formed *via* ω-hydroxypropioguaiacol.

These reaction mechanisms and possible intermediates are, of course, to a great extent hypothetical. But, independently of the nature of the intermediates, there remains the experimental fact that, on treatment with sodium hydrogen sulphide solutions, lignin and dimericlign in models, such as guaiacylglycerol-β-guaiacyl ether give considerable yields of the monomeric products acetoguaiacol, propioguaiacol and coniferylaldehyde. This proves that the hydrogen sulphide ion can split not only dialkyl ethers but also alkyl aryl ethers as well, provided that the phenolic hydroxyl group is free. This gives strong support to the ether cleavage theory.

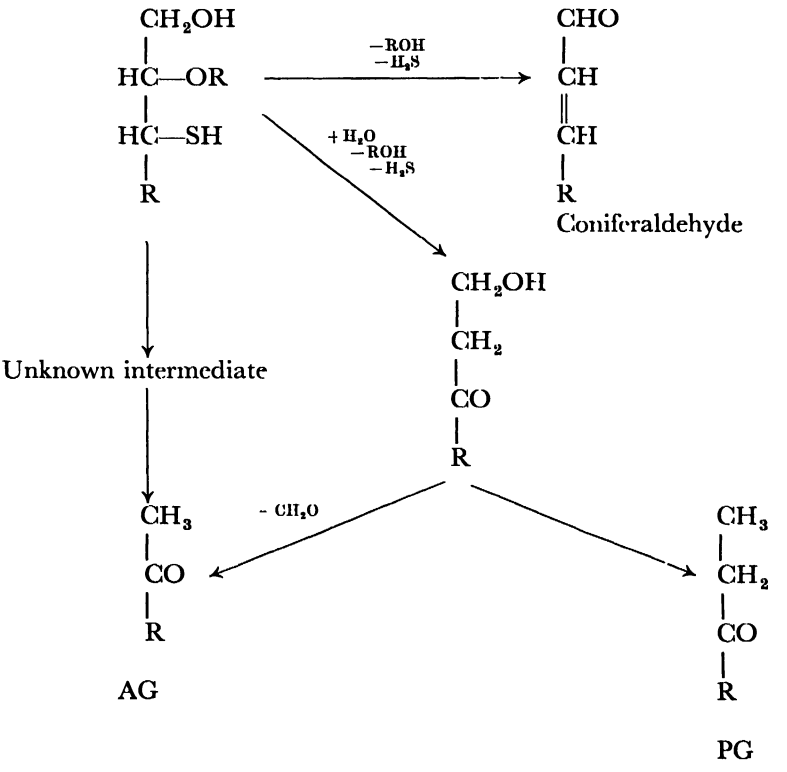


Figure 14. Scheme for the formation of acetoguaiacol, propioguaiacol and coniferylaldehyde from sulphurized guaiacylglycerol β-guaiacyl ether

OTHER REACTIONS

Figure 15 contains a scheme for the results obtained by Hästbacka on hydrogenation of hydrogen sulphide lignin with Raney nickel and boiling ethanol. He got a high yield, about 40 per cent, of ether-soluble products, which he then investigated by chromatography. It had previously been discovered, *e.g.* by model experiments by Juslén in Helsinki[25], that Raney nickel in such a procedure replaces sulphur by hydrogen. *p*-Propylguaiacol, 3-guaiacyl- and 1-guaiacylpropanol were detected chromatographically. In an analogous hydrogenation of the monosulphide of ω-hydroxypropioguaiacol, *p*-propylguaiacol and 1-guaiacylpropanol were obtained. The fact that thiolignin in addition gave also 3-guaiacylpropanol makes it probable that it contains sulphur bound in the 3-(or γ-) as well as in the 1- (or α-) position, as indicated earlier by the parentheses in *Figure 13*.

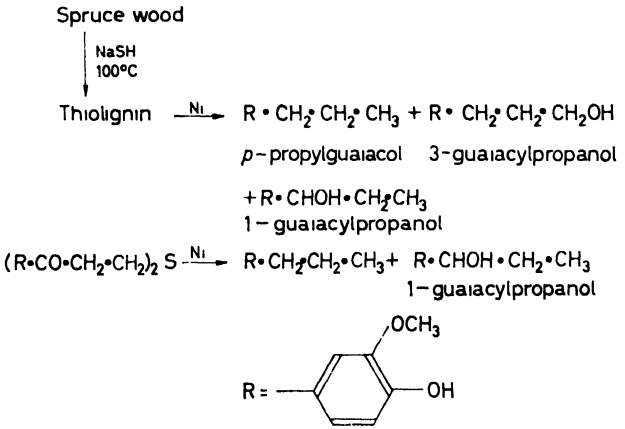


Figure 15. Desulphurization of thiolignin with Raney nickel

In addition to the ether cleavage and blocking reactions described, other reactions also occur with lignin in kraft pulping. This can be proved, *e.g.* by chromatography of the ether-soluble phenolic and acidic fractions of kraft black liquor. The amount of ether-soluble phenolic fraction alone is about 4 per cent of the organic matter, or 8 per cent of the original lignin. The ether-soluble acidic fraction is still greater, but it contains much material of carbohydrate origin. My co-workers and I have studied these fractions, and also the interesting products which are formed on heating kraft black liquors under pressure with sodium sulphide–sodium hydroxide solutions at 250–290°/65 atm. This reaction, in a way, is a continuation of the kraft pulping reaction after the cellulose has been removed. At this elevated temperature the sulphide ion has a still stronger ether splitting effect, cleaving also the simple methoxyl groups of lignin and forming methyl sulphide. This is now a commercially important reaction of which I will give a more detailed description at the *Tappi* Lignin Symposium in Chicago.

Here I will first describe some of the chromatographic methods used for the investigation of the ether soluble fractions.

CHROMATOGRAPHIC INVESTIGATIONS

The ether-soluble phenols and acids of the original and alkali-heated spent liquors have been investigated, mainly by the use of column and paper chromatography, but partly with the aid of new systems specially developed by Halmekoski. He uses, among other things, papers impregnated with sodium molybdate or sodium tungstate[26]. In paper chromatography we have worked extensively with the Freudenberg-Lehmann system II[27], *i.e.* using papers impregnated with formamide, with xylene and methylethylketone as developing solvents. Interesting variations of this system which have been tried out in collaboration with Räisänen are the use of formamide-impregnated paper and chloroform, or a mixture of light petroleum with 2 per cent xylene. Other systems which we have used are simply 2 per cent acetic acid, ethanol–ammonia, butanol on phosphate buffer impregnated paper of various pH, *etc.* For column chromatography we use columns filled with polycaprolactam or Perlon powder, and water with rising contents

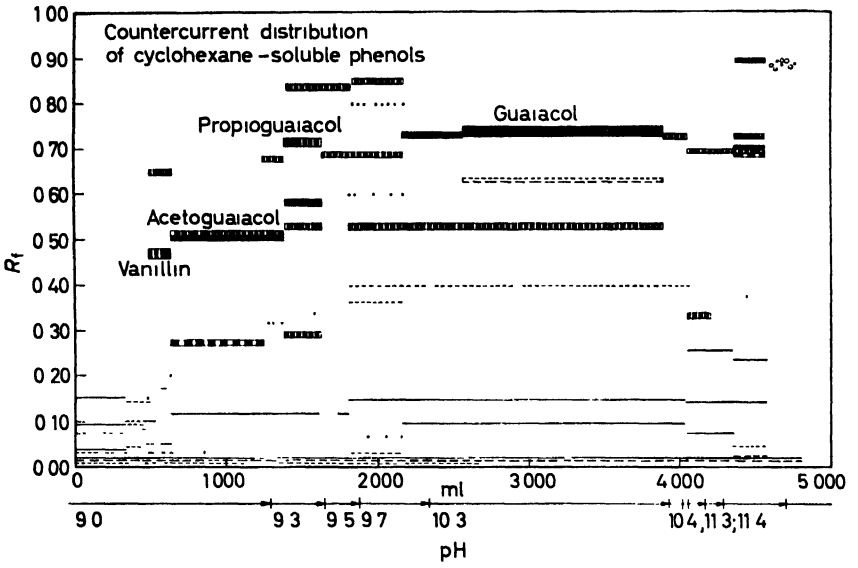


Figure 16. Countercurrent distribution of cyclohexane-soluble phenols of kraft black liquor in butanol-phosphate buffer and paper chromatography of the fractions in the Freudenberg-Lehmann system

Spots coloured with diazotized *p*-nitroaniline ———————— ———— ——

Spots fluorescing in ultra-violet light — — — — — — — — — — —

Spots fluorescing in ultra-violet light after treatment with ammonia only . . . . . .

of methanol as solvent according to Grassmann *et al.*[28]. Better results are obtained with polycaprolactam powder impregnated with formamide, followed by elution with chloroform or other solvents insoluble in formamide. Electrophoresis, thin layer chromatography according to Stahl[29], and countercurrent distribution by means of apparatus developed in the Biochemical Research Institute in Helsinki by Hietala[30] have also been used.

The spots containing phenolic substances can be coloured by diazotized sulphanilic acid or diazotized *p*-nitroaniline which give characteristic colours for various types of compounds.

Figure 16 summarizes the results of countercurrent distribution of the original spent kraft liquor phenols soluble in cyclohexane, with butanol–phosphate buffer systems with pH values rising from 9 to 11·5, and paper chromatography of the fractions. Throughput in ml are put on the abscissa and the R_f values in the Freudenberg-Lehmann system on the ordinates. Various colourations in ultra-violet light, or after spraying with soda–alkaline diazotized *p*-nitroaniline, are marked by a dotted line. In this way every line represents one substance and the thickness of the lines represents, very roughly, the amounts. We see that this rather simple phenolic fraction contains at least 50 substances, most of which, however, are present only in small amounts. However, the system has proved to be very effective as the homologues vanillin, acetoguaiacol and propioguaiacol have been completely separated from each other, without any overlapping.

In original kraft black liquor (R — *guaiacyl*, *p*):

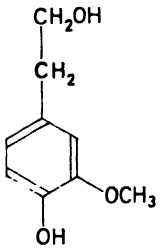
C_6H_5OH, $C_6H_4(OH)_2$ (*o*), RH, $(HO)_2C_6H_3\cdot CHO$ (*p*), $R\cdot CHO$, $R\cdot COOH$, $R\cdot CO\cdot CH_3$, $R\cdot CO\cdot C_2H_5$, $R\cdot CH_2CH_2OH$, $R\cdot CH_2\cdot COOH$ $R\cdot CH_2\cdot CH_2\cdot CH_2OH$, $R\cdot CH_2\cdot CH_2\cdot COOH$, $R\cdot CH{=}CH\cdot R$, $HOC_6H_4\cdot COOH$ (all *p*), methylcyclopentenolone.

In Na_2S*-pressure heated kraft black liquor*, 255 *or* 285°C:

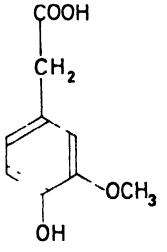
$C_6H_4(OH)_2$, $CH_3\cdot C_6H_3(OH)_2$, $C_2H_5\cdot C_6H_3(OH)_2$, RH, $(HO)_2C_6H_3\cdot CHO$, $R\cdot CHO$, $(HO)_2C_6H_3\cdot CO\cdot CH_3$, $(HO)_2C_6H_3\cdot CH_2\cdot COOH$ (all *p*) Homoprotocatechuic acid

Figure 17. Formulae of ether-soluble substances found in original and pressure heated kraft black liquor; underlined formulae belong to substances found in considerable amounts

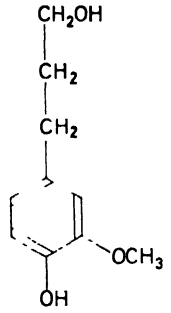
Figure 17 shows some of the ether-soluble substances which my co-workers and I found in the original kraft black liquor, and in the same liquor after pressure heating with sodium sulphide at about 280° and 65 atm pressure.



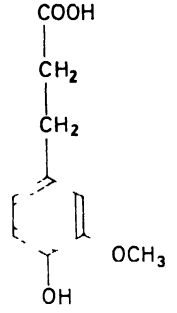
2-guaiacylethanol



Homovanillic or 2-guaiacylacetic acid



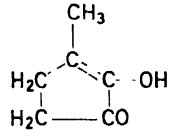
3-guaiacylpropanol



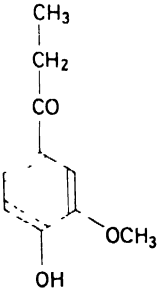
3-guaiacylpropionic acid

Figure 18. Formulae of pairs of phenol alcohols and phenol carboxylic acids found in original kraft black liquor

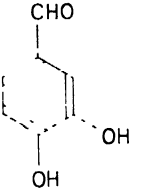
Most of the substances have been identified chromatographically, usually in four different systems. The formulae underlined belong to substances which have been found in considerable amounts. We see that, in the original black liquors, phenol alcohols occur together with the corresponding phenolcarboxylic acids, for instance guaiacylethanol and homovanillic acid, guaiacylpropanol and guaiacylpropionic acid (*Figure 18*). This



Methylcyclopentenolone



Propioguaiacol



Protocatechualdehyde

Figure 19. Formulae for 3-methyl-cyclopent-2-en-2-ol-1-one, propioguaiacol and protocatechualdehyde

indicates, perhaps, disproportionation reactions of the Cannizzaro type. Guaiacol, vanillin, vanillic acid and acetoguaiacol are all found to be present in considerable amounts, from 2 to 5 pounds per ton of pulp.

The occurrence of phenolcarboxylic acids in black liquor is of interest in view of the fact that commercial kraft and soda lignin also contain carboxylic groups to the extent of about one carboxylic group for every five phenylpropane units. This has been shown by Ekman in Helsinki[9]. Since the carboxyl groups occur also in soda lignin, it is possible that the Cannizzaro-type reactions are mainly due to the hydroxyl ions, and not to the sulphide ions.

The methylcyclopentenolone (*Figure 19*) is an alicyclic compound that we have shown to be formed from galactose, and probably other simple sugars, and also from spruce wood with a 4 per cent sodium hydroxide solution at 100° [31].

Most of the substances found in the pressure-heated black liquors are products of strong demethylation. We have found that pyrocatechol, to the extent of about 2 pounds per ton of pulp, and also some protocatechualdehyde (*Figure 19*) occur in the original kraft black liquor. This seems to show that the demethylation reactions start to some extent in the kraft digestion (*cf.* 32, 33).

The question open to discussion is whether some of the ether-soluble substances found in commercial kraft black liquor originate from lignans or other simple compounds occurring in the wood, or even in the bark. Oksanen, in Helsinki, provided chromatographic evidence for the occurrence of the following substances, among others, in the bark of pine: *p*-hydroxybenzoic, vanillic, ferulic, protocatechuic, caffeic, 3-guaiacylpropionic acid, 1-guaiacylpropanol, ω-hydroxypropioguaiacol and guaiacylglycerol. A series of pressure heatings of lignin models with sodium sulphide solution at 250–290° may help to answer this question. Among other things, pyrocatechol was derived from methylcatechol and guaiacol, and pyrocatechol and methylcatechol were obtained from pinoresinol, dehydrodi-isoeugenol and dihydrodehydrodi-isoeugenol. The two latter compounds gave propylguaiacol and propylcatechol as well[34]. Thus, there can be no doubt that heating with sodium sulphide at elevated temperatures can break carbon-carbon bonds. For instance, the pyrocatechol in the kraft black liquor might have been formed by a reaction which involves cleavage of carbon–carbon bonds. To what extent this is an effect of hydroxyl ions or of sulphidic ions is still an open question.

CONCLUSION

To summarize, it seems that the favourable effect of sulphur in kraft pulping depends partly on sulphurization of reactive groups in lignin, probably phenolic alcohol groups. These are thus prevented from reacting by condensation. Another important effect is the fission of dialkyl- and arylalkyl ether bonds in phenylpropane units carrying a free phenolic hydroxyl group. Fissions of carbon—carbon bonds, and oxidation–reduction reactions of the Cannizzaro type, also occur during kraft pulping, but it is quite possible that these reactions are mainly due to the hydroxyl

ions of the liquor. Many questions are still open, but the development of new qualitative and quantitative chromatographic and other methods offer improved facilities for further research.

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THE BEHAVIOUR OF WOOD CARBOHYDRATES IN TECHNICAL PULPING PROCESSES*

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INTRODUCTION

The predominant carbohydrate polymer in wood is cellulose and, irrespective of whether it is isolated from a softwood or a hardwood, it is very similar in chemical and physical properties[1]. It contains both amorphous and crystalline areas and has a degree of polymerization in the range of 5,000 to 10,000. Cellulose comprises approximately 40 to 50 per cent of the weight of the tree[2,3]. The hemicelluloses rank second to cellulose as the most abundant naturally occurring organic material and are mainly associated with the cell wall, although minor quantities of hemicellulosic and pectic material and other water-soluble carbohydrate polymers occur in the middle lamella[2]. The hemicelluloses, which comprise 20 to 35 per cent of the wood weight, have been considered as amorphous materials of low degree of polymerization (50 to 200) interspersed primarily in the cell walls. Isolated hemicelluloses have been obtained in crystalline form, and recent results based on a study of delignified wood sections indicate an orientation of these polymers in the cell structure[4]. Thus, the hemicelluloses, or some members of this family of polysaccharides, may exist with both amorphous and crystalline areas. Lignin, which exists mainly in the middle lamella and to a lesser degree in the cell wall, occurs to an extent of about 17 to 25 per cent in most hardwoods and about 25 to 32 per cent in the softwoods. The lignin from hardwoods has a higher methoxyl content (20 to 22 per cent) than softwood lignin (14 to 16 per cent). The so-called " extractives " differ markedly in amount and nature in both softwoods and hardwoods. The average extractive content of wood is in the order of 2 to 8 per cent. These " minor " constituents are frequently of prime importance in determining the ultimate properties and end uses of the wood itself.

The common sugar and sugar acid constituents present in wood belong to the D-series with the exception of L-arabinose, L-fucose, and L-rhamnose. Cellulose and the glucomannans are joined predominantly, if not exclusively, by β-glycosidic bonds. The xylose family of polymers contains a β-linked backbone of anhydroxylose units to which are attached, in nearly all instances, an occasional 4-*O*-methyl-D-glucuronic acid unit by an α-glycosidic bond. Certain members of this family contain, in addition to the occasional above-mentioned uronic acid, an infrequent arabofuranose unit linked by glycosidic bonds of unknown configuration. There is some

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indirect evidence that this bond may be of the β-configuration[5]. The galactoglucomannans, arabogalactans, other galactose-containing polymers, and the complex " extracellular " polysaccharides of the middle lamella contain even more complex arrangements involving both α- and β-glycosidic bonds. Starch and pectin are each joined predominantly by bonds of α-configuration.

In the case of hardwoods, the average hemicellulose content is 20 to 35 per cent, the main constituent being D-xylose. Softwoods, on the other hand, average 20 to 30 per cent hemicellulose, the main constituent of which is D-mannose, with smaller amounts of D-xylose. The major xylose-containing polymer in softwoods is the 4-*O*-methylglucuronoaraboxylan which has a lower ratio of D-xylose to 4-*O*-methyl-D-glucuronic acid (about 6 to 1) than the predominant 4-*O*-methylglucuronoxylan type of polymer of hardwoods (about 10 to 1)[2].

It should be emphasized that each family of wood polysaccharides undoubtedly exists as a spectrum of closely related polymers differing subtly in degree of polymerization, degree of branching (if branched), ratio of constituent sugars in the case of heteropolymers, and in many other properties.

The polymeric carbohydrate fraction of a tree accounts for approximately 60 to 75 per cent by weight of the wood. It is these materials, especially the cellulose portion, which are desired in the highest possible yield and in the least degraded form from the technical delignification processes commonly termed " pulping ".

The action of technical pulping processes on the above complex carbohydrate polymeric systems is widely varied and is dependent upon many factors[6]. The pH, temperature, time, chemical concentrations of liquors, the nature of the constituent building blocks of the polysaccharide, the carbon atoms involved in the glycosidic linkage, the configuration of the glycosidic bond, the presence or absence of side chains, the degree of substitution and location of acyl groups (formyl, acetyl), the polarity of the polymers, the position, type, location, and amount of the uronic acid and the degree of polymerization are some of the more important parameters and properties which contribute to the type and amount of degradation.

The purpose of this paper is to discuss the behaviour of cellulose and the various hemicelluloses during technical delignification processes. Because of the many excellent books and papers describing the various pulping methods[7,8] no details on these processes will be given in this paper.

THE TECHNICAL PULPING PROCESSES

Chemical pulping may be defined as the treatment (cooking) of wood with chemical reagents to a point where fibres can be easily separated from each other (*i.e.*, pulped). The cooking process is often described as delignification; however, there is a small amount of lignin or lignin-like material remaining with the fibres (pulp). A large amount of carbohydrate material is dissolved with the lignin and some is destroyed during the pulping procedure. In most instances nearly all of the " extractives " are removed during this stage.

The two main pulping processes which are currently being used to produce

nearly all of the pulp in the world today are the highly acid sulphite process and the strongly alkaline kraft process. Many other processes, such as soda pulping, various two-stage sulphite modifications, neutral sulphite semichemical, and partial sulphite-kraft and *vice versa*, to mention but a few of the possibilities, are also currently being used in industry, but at the present time they produce a small percentage of the total tonnage of pulp.

In order to obtain pulps of high yield and high strength with the best over-all papermaking properties, pulping conditions modified from the present normal sulphite and kraft-type processes are required. Those hemicelluloses which confer upon a wood cellulose the most desirable properties for a specific end use must be retained rather than dissolved and destroyed (for the most part) as in the present delignification processes. As wood supplies become more scarce and competition in world markets increases, the various subtleties that are evident in wood celluloses produced from modified pulping conditions, especially where pH changes are involved, will become much more important and more economically feasible.

Historically, the sulphite pulping industry is based on the use of calcium bisulphite and sulphur dioxide ("combined acid plus free acid"). This was followed by ammonium base pulping and still later by the use of magnesium for the base. The present trend is toward the use of the soluble base pulping processes. The temperatures of the digestions in the above-mentioned processes are about 120 to 150°. The pH is in the range of 1 to 2. In this process the lignin is made soluble by sulphonation, giving rise to lignosulphonic acids. Modifications of this process in which cooking liquors having pH's ranging from 2 to 4·5–5 are also commercially employed. The economic success of the magnesium and sodium systems is contingent upon satisfactory recovery systems. The future of the sulphite industry looks very bright indeed from the point of view of soda base pulping over a wide range of pH.

The second of the two main processes currently employed is termed the sulphate or kraft process. The kraft process is an improvement on the older soda process which employs sodium hydroxide as the cooking chemical. The addition of sodium sulphide to a soda-type cooking liquor and the carrying out of the digestion at temperatures in the range of 150 to 175° (conventional kraft process) facilitates preferential lignin removal by the formation of low molecular weight thiolignins and thus decreases the fibre damage. Consequently, the lesser damaged kraft pulps produce stronger pulps than does the soda or sulphite process. The soda process, however, actually preceded the sulphite and kraft processes.

Wood celluloses (cellulose plus hemicelluloses) of varying degrees of refinement may be prepared by delignification processes in which the pH level ranges from the values of 1 to 2 for the sulphite process (and other acid processes, some of which are below pH 1) through *various intermediate levels and combinations thereof* (*e.g.*, two stage sulphite, neutral sulphite semichemical, Sivola type), to the values of 12 to 13 for the kraft process. These wood celluloses, even when derived from the same wood, differ quite markedly in most instances in chemical and physical properties and in chemical composition. These differences are the result of characteristic degradative effects which occur during the delignification processes. Some

of these differences are probably due to subtle changes in the cellulose itself and to the presence of varying amounts of short chain glucans (β-cellulose). *The main differences in the wood celluloses, however, are due to changes in the structure, composition, degree of polymerization, solubility, yield, and the location of the hemicellulose components. The properties of the various hemicelluloses and their effect, individually and collectively, upon the ultimate end-use properties of the pulp has not been given the consideration it deserves. Certain types of hemicellulose have a much greater effect on the final properties of a pulp than other types of hemicellulose.* This is true whether the pulp was originally made for the dissolving industry (acetate, viscose) or whether it was specifically tailored for the paper industry. The latter pulps are obtained in higher yield and retain, or should retain, large amounts of certain of the hemicelluloses.

A comparison of the carbohydrate analyses of typical pulps prepared from western hemlock wood is given in *Table 1.* It is similar to those that have already appeared in the literature[6]. A detailed description of the carbohydrate composition of sulphite, kraft, and other pulps has been given by Hamilton, Partlow and Thompson[9], and also by Jorgensen[10].

Table 1. A comparison of the carbohydrate contents of the residues obtained from western hemlock wood by various pulping processes

	Acid sulphite	*Bisulphite*	*Chlorine dioxide holocellulose*	*Modified kraft*	*Conventional kraft*	*Prehydrolized kraft*
Galactose (%)†	0*	0*	4·6	0·6	0·6	0*
Mannose (%)†	8·1	9·8	20 3	8 1	9·3	1·6
Arabinose (%)†	0	0	1 0	0·5	0·5	0
Xylose (%)†	2·2	3·2	4 7	4·7	5·7	1·1
Rhamnose§	0	0	P	0	0	0
4-*O*-Methyl-glucuronic acid§	P	P	P	P	0	0
Galacturonic acid§	0	0	P	0	0	0
pH	1 1 5	2 5–4·0	7 8	10 11	12-13	(3 4) 12 13
Temperature(°C)	130 150	150–170	25	160–175	160 175	150-180 160-170
Carbohydrate yield (%)‡	47	46	65	42	42	35
Cellulose yield (%)‡	41	38	43	35	34	34

*Trace amounts noted.
†Based on oven dry residue
§As determined by paper partition chromatography.
P — Present
‡Based on oven dry wood, corrected for lignin and alcohol–benzene extract.

The carbohydrate content of the chlorine dioxide holocellulose of western hemlock may be considered to be a close approximation to that of the actual carbohydrate portion of western hemlock wood. Also included are the analyses of the sulphite, conventional kraft, and prehydrolyzed kraft pulps of western hemlock, as well as a modified sulphite (pH 3–4) and a modified kraft (pH 10-11). It is interesting to note the complete absence

of rhamnose and galacturonic acid from all pulps prepared by commercial techniques, the absence of galactose and arabinose from the sulphite pulp (although the former does occur in trace amounts) and the absence of 4-*O*-methylglucuronic acid, in nearly all kraft pulps. The prehydrolyzed kraft pulp lacks the acid-labile arabinose and contains only minute amounts of galactose. The mannose and xylose contents are appreciably lowered (as compared with conventional kraft pulps), and the alkali-labile 4-*O*-methylglucuronic acid is absent in nearly all prehydrolyzed kraft pulps. The reasons for these variations have been the subject of a number of investigations over the last few years[6].

Under sulphite pulping conditions the main sugars generally appear in the liquor in the following order: arabinose, galactose, xylose, mannose, and glucose[11]. The order of appearance can vary with the actual pulping conditions employed.

For convenience, the behaviour during pulping of each of the more common wood polysaccharides will be treated separately.

THE BEHAVIOUR OF CELLULOSE DURING TECHNICAL PULPING PROCESSES

Cellulose is a polydisperse polymer of high molecular weight comprised of long chains of D-glucose units joined together by β-1,4-glucosidic bonds. The possibility that a very small percentage of linkages other than β-1$\rightarrow$4 exists[12]. The presence of trace amounts of sugars other than D-glucose linked by bonds other than β-1$\rightarrow$4 is not excluded, although if such sugars and linkages occur they exist to only a very minor extent. The cellulose microfibril chains exhibit a very strong tendency to assemble themselves in bundles or areas of lesser or greater degree of order. The lateral order distribution function takes into consideration the gradual transition from an area of complete order (inaccessible area or crystallite) to an area of complete disorder (accessible or so-called amorphous area). In addition to orientation within the fibrils, the fibrils themselves exhibit a regular orientation which frequently is different for each layer of the cell wall. The degree of orientation influences the mechanical properties of cellulose in certain of its end uses. The concepts of lateral order distribution and orientation allow the satisfactory correlation of the supermolecular structure and technical behaviour of cellulose[13].

As has been mentioned previously, cellulose is the most important product obtained from a technical pulping process. It is desired in the highest possible yield and with the minimum amount of degradation. Unfortunately, however, in all present pulping processes, the cellulose is degraded in chain length and is modified, to a certain extent, in structure.

Under acidic pulping conditions, the disordered areas in the cellulose microfibril chains (amorphous areas) are more susceptible to attack and the cellulose chains are hydrolyzed preferentially in the amorphous areas with a resulting loss in degree of polymerization (and ultimately in yield[3,8]). Damage of the wood chip during the chipping operation prior to entering the digester leads to more extensive damage (and yield loss) to the cellulose due to the fact that the cellulose microfibrils are exposed to the high temperature acid conditions for a longer period[14]. During the earlier period

of cooking the lignin protects the cellulose; however, in the case of excessive chip damage this early protection is lost. In general, the greatest loss in cellulose and in carbohydrate yield occurs during the latter part of the pulping process.

The delignification processes carried out at pH's in the range of 2·5–4·5, in order to be effective, require higher temperatures than do the normal acid sulphite processes (pH 1–1·5). This increase in temperature partially offsets the more moderate pH conditions, thus, although the conditions are not so acidic, considerable hydrolytic degradation takes place due to the higher temperature[15]. The total carbohydrate yield (cellulose plus hemicellulose), depending, of course, upon actual pulping conditions, is slightly less than for delignification processes carried out at lower temperatures and at pH's in the range of 1–1·5. The actual cellulose yield is also slightly lower.

If, in the cellulose molecule as it exists in nature, there are carboxyl groups, carbonyl groups, linkages other than β-1→4 and sugars other than D-glucose, the attack of the cooking acid will not necessarily be random. This, in part, will be due to the electron-attracting ability of the carboxyl and carbonyl groups or due to the different conformation and stereochemistry of the anomalous sugars [*i.e.*, mannose, xylose (no C–6)]. In the case of carboxyl and carbonyl groups this results in a partial positive charge on the C–1 glycosidic oxygen which would repel the approach of the hydrated hydrogen ion, thus increasing the resistance of this bond toward acid hydrolysis[6,16–18]. The increased rates of hydrolysis which have been observed in oxidized cellulose have also been explained by this same inductive effect operating through the C–4 position of the carbonyl-containing anhydro sugar unit.[19]

The behaviour that cellulose undergoes under alkaline conditions at elevated temperatures is markedly different to that observed under the acidic pulping conditions. At temperatures up to about 150° the main attack on the cellulose molecule is from the reducing end by a stepwise degradation. This stepwise degradation of the terminal reducing group proceeds *via* a β-alkoxy carbonyl elimination reaction. The major acidic components from such a system have been shown to be gluco-isosaccharinic acids, although considerable quantities of other acids containing a lesser number of carbon atoms also are present. The original mechanism was postulated by Isbell[20] and, with minor modifications, has been shown to be correct by extensive studies carried out at the British Rayon Research Association[16,21].

The end-group degradation of cellulose does not continue indefinitely. The cellulose becomes very resistant to further attack due to the occurrence of a so-called " termination " reaction. The curtailment of the end-group degradation is brought about by the formation of glucometasaccharinic acids (rather than gluco-isosaccharinic acids) as the terminal (formerly reducing) group[16] (*Figure 1*). Some evidence has been presented indicating that other termination reactions (in addition to metasaccharinic acid) are also operative[22].

The effect of the end-group degradation reaction on the weight average D.P. of the cellulose is not very great. The shorter chain cellulose frag-

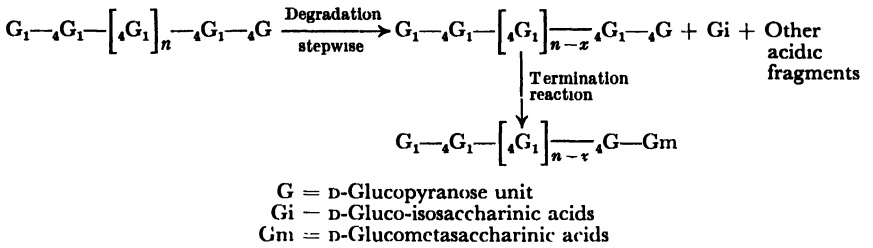


Figure 1

ments will be rapidly degraded to soluble fragments while higher D.P. material is reduced only by 40-50 D.P. units. At temperatures in the range of 150° a second type of degradation becomes important. This degradation is due to the cleavage of the glycosidic bonds by the hydroxyl ions. It has been indicated that reactions in alkaline media are similar at temperatures extending from 120–170°. However, as expected, the reaction velocities differ greatly. This cleavage naturally results in the formation of shorter fragments with reducing groups which, in turn, are degraded by the Isbell β-alkoxyl carbonyl reaction to acidic fragments.

The mechanism of alkaline hydrolysis is as yet unsolved. Evidence for the formation of anhydro sugars as intermediates in the alkaline hydrolysis of model compounds is being accumulated[23].

The temperatures which are used in most alkaline pulping processes are in the range of 140-180°. Thus the loss of carbohydrate material, by the above mechanisms, is considerable. Depending upon the pH, temperature, and time, the average D.P. of the cellulose is usually lower from an alkaline pulping process than from an acid pulping process. The chain length uniformity, however, is greater for the alkaline pulping process. The actual yield of cellulose is less from a kraft pulp than from a sulphite pulp. At similar lignin contents, the former will contain less total carbohydrate material, less cellulose, but more hemicellulose than the latter[6,9,10].

The above stepwise degradation, termination, and chain scission are the main reactions; however, other degradation reactions undoubtedly occur during alkaline pulping which also give rise to carboxyl groups and carbonyl groups at various positions in the chain. Prior reduction of the existing terminal reducing groups and reduction of such groups immediately upon their formation (due to alkali chain scission) would lead to greater recovery of the theoretical amount of cellulose from the tree[24].

THE BEHAVIOUR OF THE XYLOSE-TYPE HEMICELLULOSES DURING TECHNICAL PULPING PROCESSES

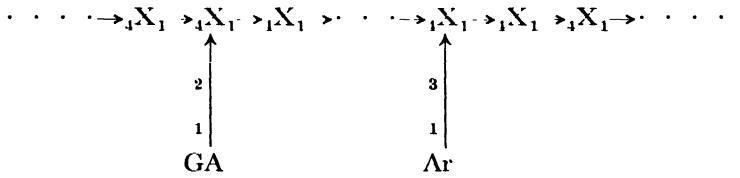
The general behaviour of the xylose-containing hemicelluloses derived from coniferous and deciduous woods during various pulping procedures has been the subject of considerable investigation[2,9,25,26]. Some of the properties of this family of polymers derived from various *pulps* prepared from western hemlock and western red alder are shown in *Table 2* [6].

Table 2. A comparison of the xylose-containing carbohydrates from a softwood and a hardwood

Source	Delignification technique	$[\alpha]_D^{23}$	Intrinsic viscosity*	Molar ratio of:— Arabinose	Xylose	4-*O*-Methyl-D-glucuronic acid
Western hemlock	ClO_2 holocellulose	54·5°	0·91	0·8	5·2	1·0
	Sulphite	− 61°	0·23	0	8·0	1·0
	Conventional kraft	− 105°	0·77	1·0	17·3	0
	Prehydrolyzed kraft	− 125°	0·33	0	100%	0
Western red alder	ClO_2 holocellulose	− 72°	0·80	0	8	1
	Sulphite	− 78°	0·21	0	8	1
	Conventional kraft	− 108°	0·65	0	100%	0
	Prehydrolyzed kraft	− 110°	0·30	0	100%	0

* Measured in molar cuprietylenediamine hydroxide and expressed in deciliters per gram.

The main xylose polymer present in an alkaline extract of western hemlock holocellulose was shown to be a 4-*O*-methylglucurono-araboxylan[27] and is represented by *Figure 2*. This same type of polymer is also present in southern pine[9].



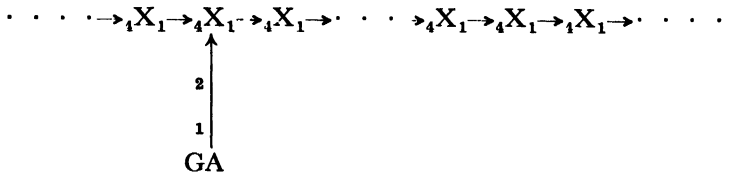
GA — 4-*O*-methyl-D-glucuronopyranose unit
Ar — L-arabofuranose unit
X — D-xylopyranose unit

Figure 2

When xylose-containing polymers are extracted from a holocellulose with non-hydrolytic solvents, it has been found that they have a fairly high acetyl content. The acetyl groups were, for the most part, attached to the 3-position of the anhydroxylose units[28,29]. Birch sulphite pulps have been reported to contain a partially acetylated 4-*O*-methylglucuronoxylan[30]. Other work involving enzymatic hydrolysis of hemicellulose from Jack pine gave rise to a xylosidoglucose[31]. A complex glucoxylan has never been isolated previously; however, the spectrum of xylose-containing polymers may be broader than suspected.

After the sulphite cooks of western hemlock or southern pine, the only xylose polymer subsequently isolated was a 4-*O*-methylglucuronoxylan. The polymers in southern pine and western hemlock sulphite *pulps* were characterized by conventional methylation studies[9,32].

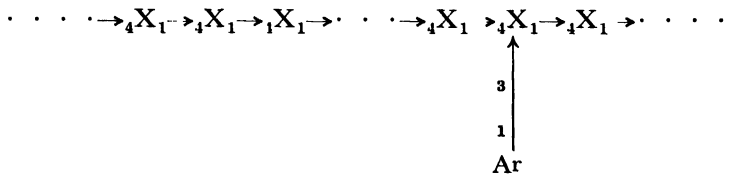
These results may be explained by the acidic hydrolytic conditions which occur during the sulphite cook. The acid-labile arabofuranose unit and most acetyl groups are cleaved from the 4-*O*-methylglucurono-araboxylan producing a 4-*O*-methylglucuronoxylan (*Figure 3*).



GA = 4-*O*-methyl-D-glucuronopyranose unit
X — D-xylopyranose unit

Figure 3

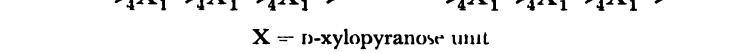
This reaction was verified by a model experiment using the actual isolated polymer from the holocellulose[9]. In addition, this artefact polymer is also degraded by acid hydrolysis to a much shorter chain length. This hydrolysis of the xylose–xylose glycosidic bonds is, of course, not random but is directed to the extent that the resistance of the glycosidic bond increases for those bonds closer to the uronic acid branch[9,17]. It is assumed that the electron-attracting ability of the carboxyl group is transmitted through the pyranose oxygen of the ring to the glycosidic bond. This produces a partial positive charge on the glycosidic oxygen which would tend to repel the approach of the hydrated hydrogen ion.



Ar — L-arabofuranose unit
X — D-xylopyranose unit

Figure 4

The conventional kraft *pulps* of western hemlock and southern pine wood have been shown to contain araboxylans[9]. The araboxylan which was isolated from the southern pine pulp was characterized by conventional chemical studies and is represented by *Figure 4*. When the readily soluble 4-*O*-methylglucurono-araboxylan of western hemlock *holocellulose* was subjected to a kraft cook for 1 hour at 160° a much less soluble polymer was isolated which was composed of arabinose and xylose[6]. This araboxylan was identical to the araboxylan isolated from the conventional kraft pulp. The prehydrolyzed kraft pulps of both southern pine and western hemlock were found to contain xylans. The xylan from the southern pine prehydrolyzed kraft pulp was characterized by conventional chemical techniques and found to have the structure represented by *Figure 5*[9].



X = D-xylopyranose unit

Figure 5

Hardwoods, such as western red alder, have been shown to contain 4-*O*-methylglucuronoxylans of high D.P. which are structurally similar to the 4-*O*-methylglucuronoxylan of the sulphite pulps of hemlock and

southern pine (*Figure 3*)[2]. These polymers differ in the ratio of uronic acid to xylose present and also in the degree of polymerization. The glycosidic linkages involved in all these polymers are to all intents and purposes identical. This polymer from alder was converted to a short chain 4-*O*-methylglucuronoxylan (with very large losses) by the sulphite pulping procedure and to a xylan by both the conventional kraft and prehydrolyzed kraft pulping procedures. The xylan from the latter pulp is present in small amount and has a low D.P.

Initial studies on polymeric 4-*O*-methylglucuronoxylan indicated the selective removal of 4-*O*-methylglucuronic acid under alkaline cooking conditions. A time–temperature plot has been presented which denotes the points of complete uronic acid disappearance[6,9]. More recent work[33,34] has verified the original findings, but with some correction of the previously noted points of complete removal of 4-*O*-methyl-D-glucuronic acid. For example, traces of 4-*O*-methylglucuronic acid were still present after 30 minutes at 170°.

The actual mechanism of the uronic acid removal during the alkaline high temperature pulping processes is not clearly understood. Other mechanisms to explain this "uronic" acid removal are currently under investigation[33].

In kraft cooked pulp which has been prepared using an alkaline stage in the range of 165–180° most, if not all, of the uronic acid portions of the various polyuronides are destroyed[6,9,35,36]. The pulp will, of course, have carboxyl groups present due to other degradation mechanisms.

In certain cases, particularly in some hardwoods, a small amount of 4-*O*-methylglucuronic acid is observed following the kraft digestion[6,9,35]. This may perhaps be explained by the location of the hemicelluloses in the wood and lack of penetration of the alkali, due perhaps to too low an alkali concentration, too low a digesting temperature, or too short a time at temperature*.

It should also be noted that some of the arabofuranose residues are also removed by the high temperature alkaline digestions indicating the relative degree of stability at this 1 >3 glycosidic bond.

As can be seen from *Table 2*, the D.P. of xylose-containing polymers as indicated by viscosity is much lower for the pulps prepared by acidic pulping conditions[37] or subjected to an acid stage prior to an alkaline delignification step. Some pulping processes carried out at intermediate pH levels give rise to xylose hemicelluloses of increasing D.P. approaching, in some cases, the D.P. of those obtained from holocellulose.

The adsorption (orientation, crystallization) of xylan polymers has been shown in kraft pulping[36]. This may be related to the removal or partial

* Since this paper was completed (August 1, 1961) and presented, an article by Croon and Enstrom has been published[58] which confirms our findings on the importance of pH. Their ordinary sulphate cooks, which were carried out at pH's of 10·55 and 11·05, showed the presence of 4-*O*-methylglucuronoxylans. This is in agreement with the results presented in this paper where cooks carried out at pH of 10–11 showed the presence of 4-*O*-methylglucuronic acid, whereas conventional kraft cooks at pH's of 12–13 did not. (See *Table 1*.) The problem of what constitutes an "ordinary sulphate cook" or a "conventional kraft cook" is a problem of semantics; however, no such problem exists with respect to the effect of pH.

For more recent papers confirming the importance of pH in pulping see Refs. 59 and 60.

removal of (*a*) " uronic " acid, (*b*) removal or partial removal or translocation of acetyl groups, (*c*) removal or partial removal of the arabinose residues. *The extent of the above degradation, the D.P., and the location of the ultimate xylose-containing hemicellulose are also highly important.*

The presence of formyl and acetyl groups in wood celluloses has been recognized for some time[39]. The presence of the acetyl groups on the native xylose-containing hemicellulose has been more recently disclosed and the position of the acetyl and the average degree of substitution has been determined in certain instances[28,29]. The rôle which these groups play in the delignification of the wood has not been clearly demonstrated. The xylose hemicelluloses would be less hindered sterically by removal of the acetyl groups, and the adsorption by hydrogen bonds to one another, to other hemicelluloses, or to cellulose would probably be enhanced. Much more work must be done before this fascinating problem can be definitely solved.

THE BEHAVIOUR OF THE MANNOSE-CONTAINING HEMICELLULOSES DURING PULPING

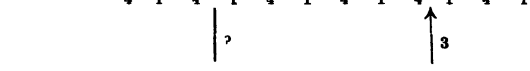
The mannose-containing polymers of conifers comprise approximately one-half to three-quarters of the total hemicellulosic material in the wood[2,40,41]. In deciduous trees the mannose-containing polymers make up a very small percentage of the total hemicellulose[2,42]. Although it was thought for some time that wood cellulose contained large amounts of mannose, it was shown, in 1956, that a very large percentage of the mannose in sulphite pulp occurred as a true diheteropolymer glucomannan[43]. Further studies in 1957 showed that extraction of pulps or holocelluloses with high strength sodium hydroxide (about 10 per cent), in conjunction with mechanical action and slight acid hydrolysis, gave residues in high yield (about 30 per cent based on dry wood) of high degree of polymerization in which the total hemicellulosic material was reduced to less than 0·25 per cent for softwoods and less than 0·1 per cent for hardwoods (based on wood)[2,44]. Rapson, in more recent studies, has shown even lower values for residual hemicelluloses[45]. These results indicate that the residual extraction-resistant non-cellulosic cell wall polysaccharides, which remain associated with cellulose, as shown by hydrolysis (*i.e.*, mannose and xylose), are not part of the cellulose (except perhaps in trace amounts), but are much more probably due to inaccessible or highly insoluble polymers intimately associated with the cellulose microfibrils[2].

The mannose-containing polymers of conifers appear to be made up of at least two main families of polymers, the glucomannans and the galactoglucomannans[2,6,9,46]. The possible presence of other polymers, such as a true mannan and galactomannan, is not excluded, although in general these polymers will probably not be present in large amounts[2,40,44]. As a family of polymers or two closely related families, it is possible that the glucomannans vary not only within themselves in chain length and glucose-to-mannose ratio and sequence, but also contain varying amounts of galactose as a part of their structure[40]. The number average molecular weight generally falls in the range of 70 to 130, although some depolymerization undoubtedly occurs in isolation. For the present it will be assumed that

both glucomannans and galactoglucomannans exist as separate but closely related families of polymers (with sub-families), each with its own spectrum of subtle changes in molecular architecture. These changes will have a variable effect on the physical and chemical properties of the polymers (*i.e.*, solubility, ability to orient, stability).

In certain instances polymers which are high in mannose, but contain also glucose and galactose, are most readily removed by low strength sodium hydroxide (below swelling strength) whereas, in most cases, extraction with high strength caustic (8–18 per cent) following lower strength extractions gives rise to polymers high in mannose and low in glucose (glucomannans with 1:3 or 1:4 ratio of glucose to mannose). In most instances, however, small amounts of galactose have also been shown to occur here as well as at lower caustic strengths[40]. This follows the general rule that the more highly branched the polysaccharide, the greater will be its solubility.

As mentioned previously, the predominant sugar in hydrolyzed coniferous wood, apart from glucose, is mannose. Most of the mannose of softwoods is a part of the glucomannan and galactoglucomannan mixture in which the glucose:mannose ratio is approximately 1:3 or 1:4 (see *Figure 6*). Partial hydrolysis of the polymeric chain indicates that random distribution of the individual monosaccharide constituents is most probable, at least for the " backbone " portion of the polymers. Glucomannans have been isolated from a variety of conifers, as indicated in a recent review[47]. Slightly different glucomannans having glucose:mannose ratios in the range of 1:1 to 1:2 have been isolated from western red alder[2] and other hardwoods[42]. Glucomannans have also been detected in western hemlock sulphite pulp and Norwegian spruce sulphite pulp. As with *all* polysaccharides isolated from pulps, the properties and structure are not necessarily the same as in the native wood. The majority of the glucomannans reported appear to be essentially linear or slightly branched polymers. In only a few instances has the position of the branch point been inferred[41] or actually stated (position-3 of glucose)[48].



G = Glucopyranose unit
M = Mannopyranose unit

Figure 6

Table 3 lists the properties of the glucomannans which have been isolated from western hemlock and western red alder wood pulps. The persistence of the glucomannan in the sulphite pulp indicates a relative stability towards acid hydrolysis. Actual graded acid hydrolysis experiments suggest that the glucomannans are more resistant to acid hydrolysis than are the xylose-containing family of carbohydrates, although in certain instances the glucomannans appear to be more resistant to hydrolysis than cellulose[33]. Glucomannans were also detected in the conventional kraft pulps of western hemlock and southern pine. Due to the properties of the araboxylan contaminant, it is often difficult to obtain a pure glucomannan;

Table 3. A comparison of glucomannans of western hemlock and western red alder

Wood	*Delignification technique*	$[\alpha]_D^{23}$	*Cuene I.V.*	*Molar ratio of:*	
				Glucose	*Mannose*
Western hemlock	ClO_2 holocellulose	− 35°	0·56	1	2·6
	Sulphite	− 37°	0·29	1	3·0
	Conventional kraft	*	—	1	2·5
Western red alder	ClO_2 holocellulose	− 25°	—	2·0	3·0
	NSSC wood cellulose	− 32°	—	2·0	3·0

* This polymer was not pure but contained an araboxylan contaminant.

however, separation can be achieved by fractionating the glucomannan acetate of kraft pulps. Anomalous glucomannans have also been isolated from western hemlock sulphite pulp[41].

The alkaline degradation of the predominantly straight chain 1→4 β-linked glucomannans is very similar to that of cellulose. Using model experiments and isolated glucomannans of markedly different intrinsic viscosities only about 15 to 20 per cent yield of the glucomannans was obtained following kraft cooks at 160° [6].

During the alkaline pulping of glucomannans, the stepwise end-group degradation and termination reactions occur in conjunction with the high temperature chain scission reaction due to the hydroxyl ion. This is true both of the hardwood and softwood glucomannans. Reduction of the terminal end groups using sodium borohydride gives a higher carbohydrate yield as in the case of cellulose. The alkaline degradation of glucomannans is much more rapid than xylose-type hemicelluloses and seems to be more closely related to cellulose.

The presence of acetyl groups in wood has been mentioned previously and it is known that some of the acetyl groups are associated with the mannose-containing polymers[25,49] (*e.g.*, glucomannans, galactoglucomannans). Pulping in the presence of sodium borohydride[38] or by a two-stage process in which there is first a slightly alkaline pH followed by a lowering of the pH and completion of the cooking cycle, gave a much higher carbohydrate yield and nearly all of the increased yield could be accounted for in the increased amount of glucomannan associated with the pulp[25,49]. Annergren and Rydholm[25,49] theorize that this may be due to deacetylation which then enhances the opportunity for sorption of the glucomannan on the cellulose microfibrils. This stabilization of the glucomannan presumably results from the prevention (by adsorption and screening, complex formation) of the end-group β-alkoxycarbonyl degradation reaction. In the case of sodium borohydride-containing kraft cooks the fact that the amount of galactose does not increase proportionately is perhaps indicative of the existence of both glucomannans and galactoglucomannans.

It is interesting to note the marked stabilization of the glucomannan in comparison with the poor stabilization of the xylose hemicelluloses both in sodium borohydride pulping and in two-stage sulphite cooks (xylan content of the borohydride-reduced cooks was found to decrease.) The removal of some galactan residues may have an effect similar to, or perhaps

greater than, the removal of acetyl groups in allowing greater hydrogen bonding (as this results in less steric hindrance).

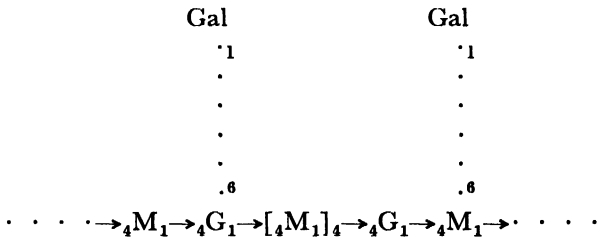
A number of workers have reported the probable presence of galactoglucomannans from a variety of sources. The following table, although by no means complete, indicates the specific rotation and sugar ratios of a number of galactoglucomannans from softwood holocelluloses (western hemlock) and three softwood conventional kraft pulps. The presence of a galactoglucomannan in wood was first demonstrated by Adams[50]. Recently, Timell has reported the isolation of galactoglucomannans from a number of conifers[47].

Table 4. A comparison of the galactoglucomannans from various wood sources

Wood source	*Delignification technique*	$[\alpha]_D^{23}$	*Molar ratio of:*		
			Galactose	*Glucose*	*Mannose*
Western Hemlock	ClO_2 holocellulose	$-10°$	0·8	1·5	5·0
	Conventional kraft	$+2$ to $-2°$	1·0	1·5	5·0
Southern Pine	Conventional kraft	$+12·5°$	1·2	1·0	3·0
Redwood	Conventional kraft	$+17·5°$	1·0	1·0	3·0

It is interesting to note that the specific rotation of the polysaccharides isolated from holocelluloses is lævorotatory, whereas those from the pulps are dextrorotatory. This would indicate changes occur in the polymer during delignification.

The first characterization of such a triheteropolymer was carried out on the galactoglucomannan obtained from kraft wood cellulose produced from southern pine[46]. Although a unique solution to the structure of this polymer is not possible on the basis of the data presented to date, a simplified structure consistent with the facts so far ascertained is shown below. It is understood that a mixture of a galactomannan and glucomannan would give similar results from methylation studies. All chemical methods failed to give any evidence of mixed polymers. Data by Timell[47] and Bishop[51] using electrophoresis and/or ultracentrifugation on galactoglucomannans from other wood sources showed homogenity of the polymer.



G = Glucopyranose unit
M = Mannopyranose unit
Gal = Galactopyranose unit

Figure 7

Galactoglucomannans are not found in sulphite or prehydrolyzed kraft pulps to any appreciable extent[6]. The low positive optical rotation of the galactoglucomannan indicates a predominance of β-glycosidic bonds but it does not preclude the possibility that some of the bonds may be, and probably are, of the α-configuration. The behaviour of the polymer during acid pulping conditions and during graded hydrolysis strongly indicates that most of the galactose residues are joined to the polymer by a labile linkage[41,46]. Recent studies by Meier report the isolation of a galactosyl mannose and a galactosyl mannobiose from a spruce "glucomannan"[52]. The galactose residue of these oligosaccharides was linked by an α-glycosidic bond. Meier considers that these oligosaccharides are derived from a mixture of glucomannan and galactomannan.

The removal of galactose residues could, as predicted previously, leave a type of glucomannan[41]. It is possible that part of the glucomannan left following an acidic cook may be derived from this source. However, the availability to hydronium ions of the branched soluble galactoglucomannans, as compared to the highly "resistant", insoluble and inaccessible glucomannans, would indicate that the former would probably be destroyed to a great extent during the acid cooks. In model experiments using 0·25N oxalic acid at 100° for 6 hours, the galactoglucomannan was found to be very labile to acid hydrolysis[46]. Thus, very little mannose would be derived from this triheteropolymer in sulphite or prehydrolyzed kraft pulps.

The relative stability of the galactoglucomannans to alkali was initially considered to be unusually high[6]. Subsequent investigations, however, have demonstrated a marked dependence on the polymer source, and intrinsic viscosities[33]. Structurally, the galactoglucomannan has been shown to be a chain of 1→4 linked glucose and mannose anhydro units to which are attached branches, by 1→6 glycosidic bonds, terminated by galactopyranose units. The presence of branches on the 6-position should not affect the end group degrading mechanism. If, however, the molecule was constructed such that short branches were linked by 1→4 bonds and the main chain of the polymer was linked by both 1→4 and 1→6 bonds, greater alkali stability might be expected. Alternatively, the presence of some α-glycosidic bonds in this polymer would result in less degradation due to the greater stability of these bonds to alkaline hydrolysis. Previous studies have shown that amylopectin, which has α-1→4 and α-1→6 linkages when cooked under similar conditions, was isolated in 43 per cent yield[6].

Comparison of the alkali stability of galactoglucomannans with glucomannans isolated from the same source indicates the existence of two different polymers. Significantly dissimilar yields were obtained for these hemicelluloses in alkaline media for identical time temperature relationships. Since yield results also appear to be dependent on the previous history of hemicelluloses, the rate of alkaline degradation may be related in part to the initial intrinsic viscosities.

It should be noted that traces of xylose have been reported present in hydrolyzates of many glucomannan fractions. This could indicate incomplete separation of the xylose hemicelluloses or perhaps xylose as a constit-

uent of a glucomannan or galactoglucomannan. The former possibility appears the more probable. Mannose–xylose linkages could, of course, occur and glucose–xylose linkages have been reported[31].

THE BEHAVIOUR OF THE MISCELLANEOUS CARBOHYDRATE FRACTIONS OF WOOD DURING PULPING

Very little is known of this complex mixture which, upon hydrolysis, can give rise to galactose, arabinose, xylose, glucose, mannose, rhamnose, and uronic acids. Polygalacturonic acid has been detected in a number of woods by Anderson and co-workers, while starch, arabogalactans and unusual mannose-containing polymers have been detected by others[2]. When a commercial pectin and an algin were cooked with kraft liquor, all the uronic acid components were destroyed in less than 30 minutes, and the small amount of carbohydrate material which was isolated appeared to be neutral araban and galactan contaminants (in the case of pectin) and glucan and xylan (in the case of the algin), which had been present in the original samples. It would appear, therefore, that pectin, which is present in wood, but never present in wood pulps, has been removed from these pulps and degraded by the hydrolytic action of the cooking liquors[6].

Starch has also been shown to be present in wood. This component undoubtedly varies in concentration with time of year. Starch is very susceptible to acid hydrolysis and, hence, would be degraded and removed during the sulphite cook. Starch was found to be quite resistant to alkaline degradation, but its marked solubility in hot alkali or hot water, together with its location in an accessible region, suggests it is dissolved and diffuses out of the kraft wood pulps. Pure amylose was recovered in 25 per cent yield from a 60-minute kraft cook at 160°, while amylopectin was recovered in 43 per cent yield from a similar cook[6].

Other complex carbohydrate polymers exist in wood. These have not been isolated and characterized. Their properties in certain instances, however, resemble those of the various plant gums such as cherry gum, gum arabic, and Shasta fir gum[53]. A comparison of the yield losses of cherry gum, gum arabic, and the 4-*O*-methylglucuronoxylan during a kraft cook at 160° showed that the cherry gum was rapidly degraded to non-isolatable products. Shasta fir gum behaved in a similar manner. The gum arabic was degraded by alkali in a manner quite similar to 4-*O*-methylglucuronoxylan. Such a behaviour might be predicted from the postulated complex structure of this polymer[51]. Hydrolysis of the polymer before kraft cooking indicated the presence of galactose, arabinose, rhamnose, and glucuronic acid. Following the kraft cook, it was found that the polymer was now composed of galactose and arabinose only (no glucuronic acid was observed chromatographically). Such behaviour would be explained if the anhydrorhamnose units on the branch of the tentative structure were further removed from the main chain of the polymer than the uronic acid. It could also be explained by variations which undoubtedly occur in the structure of various samples of gum arabic.

While the above polymers are associated chiefly with the cambium and inner bark, they are probably representative of the many " extracellular " carbohydrates. The alkaline degradation, as noted above, is not entirely

related to end group cleavage as cherry gum does not reduce Fehlings solution. The behaviour of Shasta fir gum and cherry gum may be explained if uronic acids are incorporated in the main chain of the polymer as well as in the branches, as was the case for gum arabic. In this manner cleavage of the polymer at the uronic acid position will result in a large number of short chain molecules with terminal reducing groups susceptible to the action of hot alkali. These fragments would be rapidly degraded to non-carbohydrate constituents.

An arabogalactan has been isolated from the water-soluble extracts of many woods, especially western larch. The most recent work has been the isolation and characterization of the arabogalactan of Monterey pine by Brasch and Jones[55]. These authors showed their polymer to be a highly branched galactan joined through positions 1, 3 and 6. The branches were terminated by arabofuranose units. This type of polymer would not be particularly sensitive to alkaline hydrolysis. The lack of arabogalactan in kraft pulps is probably the result of the dissolving of the polymer and its diffusion into the cooking liquor. The lack of arabogalactan in the sulphite pulp is undoubtedly the result of its lability towards acid hydrolysis.

CONCLUSION

Pulping procedures carried out at various pH levels introduce important chemical and physical changes in the resulting wood cellulose. The most obvious and most important chemical differences are those which arise in the " non-cellulosic cell wall polysaccharides " or, as they are commonly termed, " hemicelluloses ". The absence (except in trace amounts in some instances) of such sugars as galactose, arabinose, rhamnose, and galacturonic acid in the sulphite and prehydrolysed kraft pulps arises from the acid-catalysed hydrolysis of the polymers containing these sugars or the selective removal of the sugars from the polymers. In general, these carbohydrate polymers are degraded to short chain fragments which dissolve in the cooking liquor, diffuse out of the chips and are thus removed from the final pulp. Pectin, starch, arabogalactans, galactoglucomannans (except for trace amounts), as well as small amounts of unidentified polymers, behave in this manner.

Certain sugars, particularly arabinose, are attached to various polymers as an acid-labile terminal unit. These sugars are rapidly and selectively cleaved from these polymers during acidic cooks. This behaviour accounts for the conversion of the 4-*O*-methylglucurono-araboxylan of softwoods to a 4-*O*-methylglucuronoxylan. This new polymer, as an artefact in softwood pulps and as the predominant original polymer in the hardwoods, is degraded by acid hydrolysis into molecules of lower D.P. Many, but not all, of these polymers dissolve in the cooking liquor and diffuse out of the pulp. The glucomannans of softwoods and hardwoods behave in the same manner under acidic pulping conditions, with the loss by solution in the liquors, of the shortest molecules.

Most of the galactose from the galactoglucomannan is readily removed under strongly acidic pulping conditions and the molecule is degraded

such that most, if not all, of the galactoglucomannan appears to be removed from the final pulp. Galactoglucomannans will occur, however, in modified sulphite, modified kraft and kraft pulps.

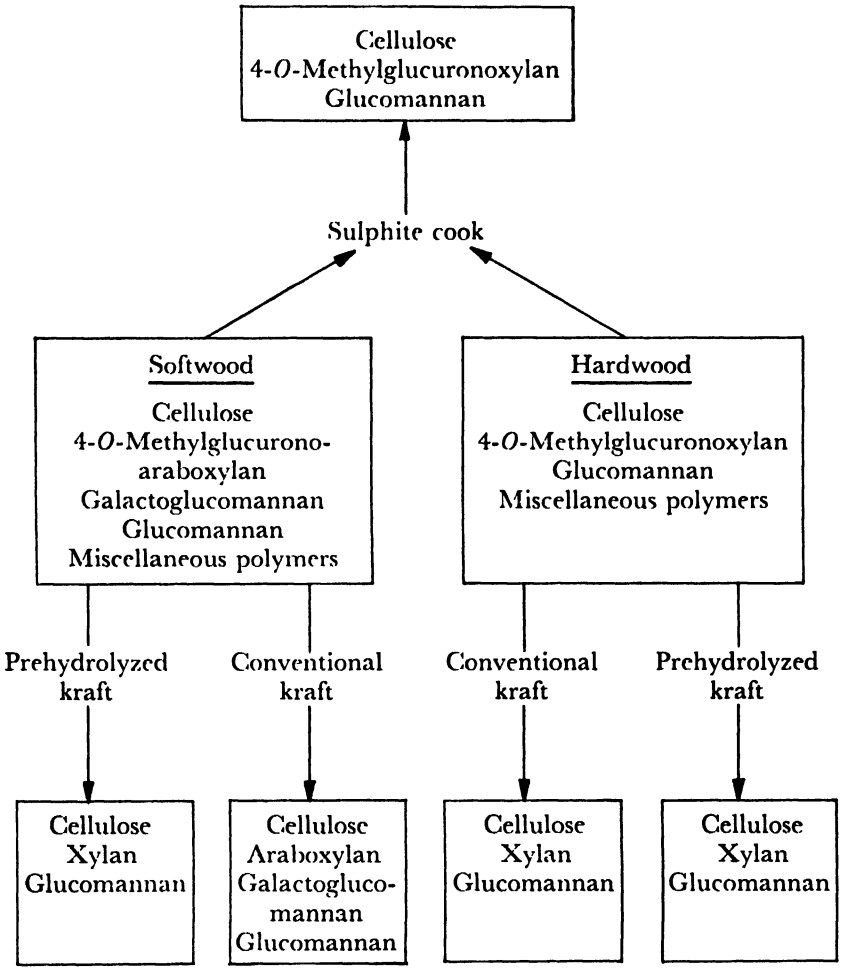
In the presence of the hydroxyl ion during the kraft and soda cooking procedures, different types of degradation occur. The best understood reaction is the end group degradation reaction resulting from an attack on the reducing group of a carbohydrate polymer by the hydroxide ion. In the case of cellulose, Samuelson and Wennerblom[56] have estimated that 40 to 50 glucose units are cleaved to isosaccharinic acid fragments, which in the process of formation, must be removed from the chain, exposing a new reducing group to attack. After some 40 to 50 anhydro units have been cleaved, on the average, a metasaccharinic acid is formed which remains attached to the polymer as an alkali-stable glycoside of the final reducing sugar, thus terminating the end group-alkoxy-carbonyl degradation.

Undoubtedly the xylose, glucomannan and galactoglucomannan polymers behave in a similar manner to the degradation of the cellulose. There will obviously be many modifications but, in principle, the degradation will proceed analogously. A substituent attached to a 3-position such as is present in the 4-*O*-methylglucurono-araboxylan which, in turn is converted under high temperature alkaline cooking conditions into an araboxylan, will cause a termination of the end group degradation. The presence of α-1$\rightarrow$6 glycosidic bonds will probably also result in the increased stability of certain polysaccharides such as the galactoglucomannans which exist in modified kraft and kraft pulps.

At these elevated temperatures, however, other complex reactions occur. The degradation studies on 4-*O*-methylglucurono-araboxylans, pectin and plant gums indicate that the presence of a uronic acid carboxyl group confers alkaline instability to the glycosidic bond which joins the uronic acid to the rest of the molecule. This instability results in the rapid removal of most, if not all, of the uronic acid when such a polymer is treated with alkali at temperatures in the range of 160 to 170°.

These complex reactions which occur during the kraft cook result in the production of araboxylan and xylans in the pulps from softwoods and hardwoods, respectively. The polymers have a somewhat lower D.P. than the original polymer in the wood due to the hydrolysis of xylose xylose glycosidic bonds. The decrease in uronic acid content results in a *drastic decrease* in the solubility of the xylan and araboxylan in caustic. Polymers such as arabogalactans and pectin are rapidly dissolved in the alkali. In the case of pectin, the degradation would be to non-identifiable products. Substances similar to cherry gum and Shasta fir gum would also be rapidly degraded to non-identifiable products due to the presence of uronic acid in the main chain of the polymer. These conclusions are summarized in a very general manner in *Figure 8.*

Undoubtedly those hemicelluloses which are in contact with the pulping chemicals at high temperature for the longest time are highly degraded. The extent of degradation will, of course, be dependent upon the conditions and the structure of the polysaccharides themselves. The highly resistant polysaccharides (those requiring caustic of a solution strength



Note. The yield of polysaccharide, the degree of polymerization, structure and properties are different and are contingent upon the type of wood and pulping process. For simplification, only extremes of pH are considered.

Figure 8. A simplified scheme which indicates the general behaviour of some carbohydrate polymers of wood during technical pulping processes

of 8 per cent or higher to remove them from the pulp) will probably be degraded to a lesser degree.

The differences in the properties of pulps produced at different pH's [acid sulphite, bisulphite, neutral sulphite semichemical (similar to holocellulose preparation), modified kraft, conventional kraft, prehydrolyzed kraft, and all of the many possible combinations of acid, acid–base, base–acid] are due primarily to the amount, type, structure, chain length, location and general chemical and physical properties of the hemicelluloses. Some of the differences are due to subtle changes in the cellulose structure and chain length as well as in the amount and chain length of the short chain glucans (β-cellulose) which are always present.

Conditions must be selected such that the desired type of hemicellulose is left or deposited (adsorbed or crystallized) in the appropriate place in order that they will confer upon the ultimate wood cellulose those properties which will give it the maximum beneficial effects for a specific given end use.

A thorough understanding of the physical and chemical properties and o the location across the cell wall of the various hemicelluloses present in the different woods and from the different wood celluloses derived from these woods is essential before we can hope to utilize our wood supplies to their ultimate potential. Meier has carried out basic studies in this field[57]. Such studies will lead to a thorough understanding of the type and extent of the degradation which each hemicellulose undergoes under technical pulping processes and will eventually lead to modified processes where the degradation is essentially eliminated (if possible) or kept to a minimum.

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THE CHEMISTRY OF DELIGNIFICATION IN PULP BLEACHING

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INTRODUCTION

The commercial bleaching of pulps represents a chemical process by which the residual non-carbohydrate constituents, principally lignin, are removed from the pulp. The colour improvement in the process is due to the fact that the chromophoric groups responsible for the discoloration are chemically combined with the latter constituents and are effectively removed from the pulp when these are dissolved in the bleach liquor.

Obviously, an effective delignifying bleaching chemical has to fulfil two essential conditions as far as its chemical characteristics are concerned. Firstly, it must react rapidly with lignin, degrading it to water and/or alkali-soluble fragments, and, secondly, its action on carbohydrates, particularly cellulose, must be extremely slow compared with the rate of reaction with lignin.

The same requirements are valid for reagents which are used for the preparation of holocellulose and various other cellulose preparations from extracted wood meal. It is interesting to note that for each commercial bleaching procedure, there exists a method for the preparation of holocellulose, or various other " cellulose " preparations, that utilizes the same agent and a largely similar set of conditions. This parallelism,

Table 1. Bleaching methods compared with holocellulose preparation methods

Reactant	*Pulp bleaching method*	*Counterpart holocellulose method*	*Ref.*
Molecular Cl_2	Acid chlorination	Cl_2–ethanolamine–holocellulose	2
HOCl	Hypochlorite bleaching	Norman-Jenkins–cellulose	3
ClO_2	ClO_2 bleaching	Schmidt-holocellulose	4
$NaClO_2$	Chlorite bleaching	Wise-holocellulose	5
Peracetic acid		Poljak–holocellulose	6
Nitric acid		Kurschner-Hoffer–cellulose	7

illustrated in *Table 1*, is by no means obvious, if one takes into account that the residual lignin in pulps differs from wood lignin by the pulping and, sometimes, by the pre-bleaching operations.

Some of the delignifying agents listed, however, have not found application as industrial bleaching agents. Nitric acid, although an excellent delignifying agent, reacts too quickly with cellulose to be utilized for pulp

bleaching. The new bleaching agent peracetic acid shows considerable promise as a bleaching chemical and is at present being studied in a number of laboratories.

In the bleaching process, the pulp constituents cellulose, hemicelluloses and lignin all compete for the consumption of the bleaching agent. A situation where the rate of the lignin reaction is very high in comparison with the rate of cellulose degradation is most advantageous for a successful operation. A convenient method for studying this ratio of reactivities was developed by Paulson[1] and consists of plotting the fluidity of the pulp against the consumption of the bleaching agent. The method forms an excellent basis for comparison between different bleaching agents and conditions, and is limited only by the fact that the action of the bleaching agent does not consist of one but several parallel and consecutive reactions.

A more thorough understanding of bleaching phenomena would involve the chemical pathways by which lignin is degraded to soluble compounds. Work in this area presents several difficulties which cannot easily be solved by studying actual bleaching systems. Some of these difficulties can be avoided by studying the reactions of wood lignin itself, or those of various lignin preparations, with bleaching agents. Furthermore, studies on lignin model compounds are of considerable help in evaluating these complex reactions.

One of the outstanding questions in bleaching chemistry is concerned with the site of the primary attack by the reactant. The agent, depending on its chemical nature, may attack the guaiacyl propane units either at the aromatic nuclei or, alternatively, it may react with the three carbon side chain of these units. *A priori*, the former possibility would seem to be the more likely one, particularly if the bleaching reagent has strongly electrophilic characteristics, because the reactivity of the aromatic nucleus is enhanced by the phenolic hydroxyl, phenol ether and side-chain substituents.

In testing the reactivity of the aromatic nuclei and that of side-chain α-carbon atoms in lignin, simple model compounds, such as vanillyl and veratryl alcohols and their derivatives, are particularly useful. It is well known that model studies on the compounds mentioned have made an important contribution to the understanding of pulping reactions. The problem of elucidating the reactivity of β- and γ-carbon atoms is a more difficult one and no well-defined approach for solving the problem is available at present.

In the following, a brief review is presented on the current ideas of the mechanism of degradation of lignin by various bleaching agents.

CHLORINE–WATER SYSTEMS: CHLORINATION AND HYPOCHLORITE BLEACHING

In aqueous chlorine systems, chlorine can exist in three different forms, depending on the pH of the solution. These are molecular chlorine (Cl_2), hypochlorous acid (HOCl) and its anion OCl^-. In the region up to pH 5, a concentration-dependent equilibrium exists between molecular chlorine and hypochlorous acid, while at higher pH values hypochlorous

acid and its anion are both present in proportions directly determined by the pH of the solution.

Two pH regions are of particular interest for bleaching chemistry. The pre-chlorination of pulp is carried out at pH 2, and the purpose of this operation is to make the bulk of the residual lignin soluble in both water and alkali. No colour improvement of pulp is accomplished at this stage, nor in the following alkali-wash, but the subsequent hypochlorite bleaching stage is greatly facilitated because of the decreased residual lignin content. The optimum region for the hypochlorite bleaching is pH 8–9, and the effect of the bleaching at this pH differs significantly from that of pre-chlorination.

The difference between the effects of pre-chlorination and hypochlorite bleaching is probably due to the difference in the nature of the reacting chlorine species. At the lower pH, molecular chlorine, a species much more reactive than hypochlorous acid, directs the reaction, while in the hypochlorite bleaching, hypochlorous acid, or a reactive intermediate derived from it, probably plays the rôle of the primary attacking species.

The effect of molecular chlorine on lignin and lignin models has been subject to more extensive studies than the hypochlorite oxidation. Several

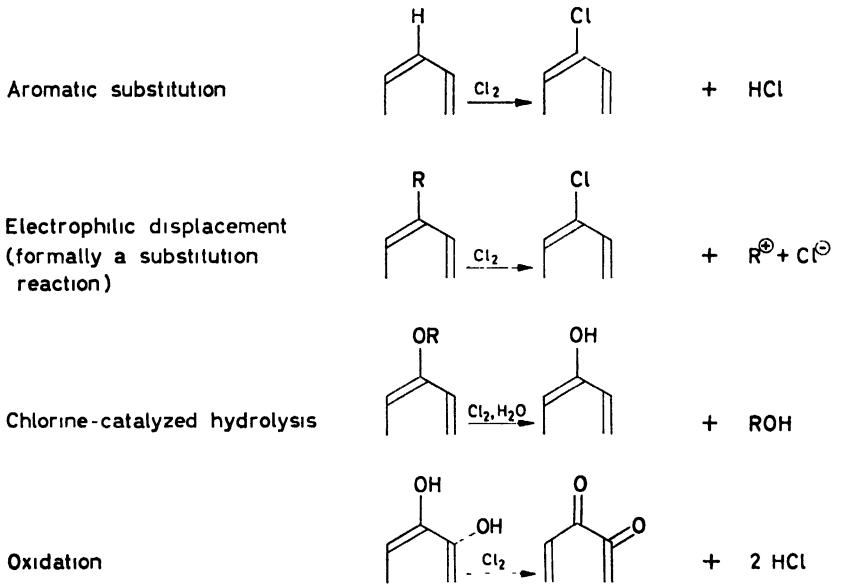


Figure 1. Competing reactions in the chlorination of aromatic compounds

reactions take place which can be classified in four classes illustrated in *Figure 1.* The reactions are shown in the approximate sequence of decreasing reaction rates, although it should be borne in mind that considerable variation in rate exists in each class of reactions.

(a) Chlorine substitution

The rate of this reaction is immeasurably fast in the guaiacyl propane units and takes place at the 6- and 5-positions of the aromatic nucleus. The rate constants at these positions depend on the presence or absence of a free phenolic hydroxyl group at the 4-position as well as on the nature of the aliphatic side chain at position 1. This reaction does not contribute to the degradation of lignin.

(b) Side-chain displacement

This reaction is formally analogous to the substitution reaction, except that the leaving group is a carbonium ion instead of a proton. In etherified

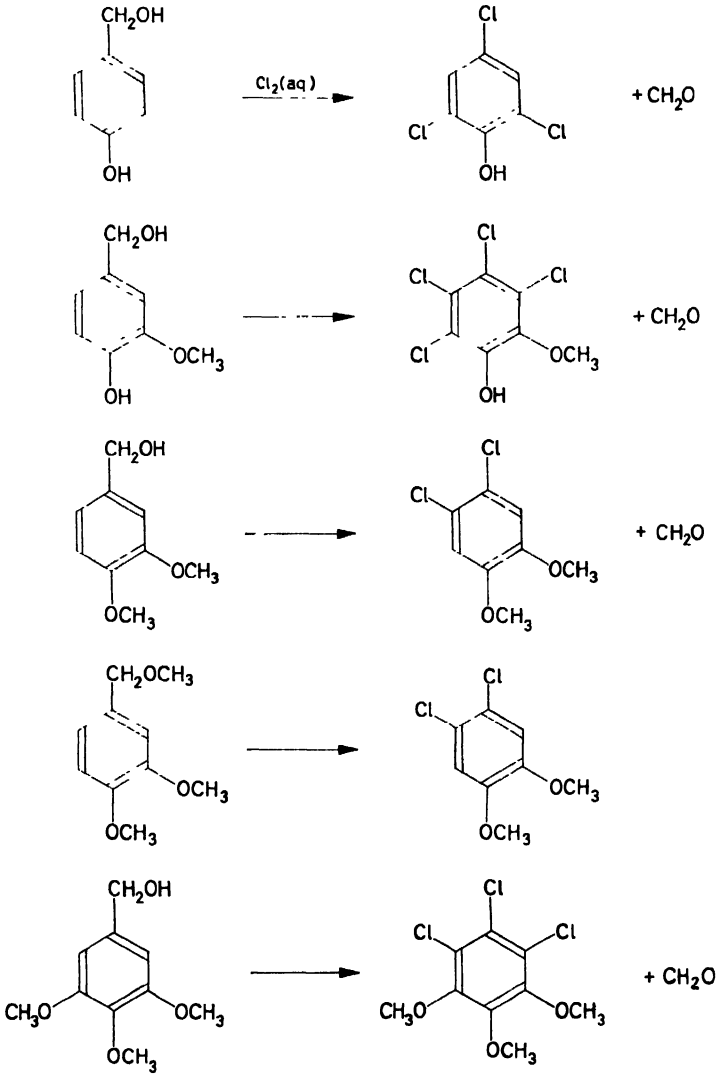


Figure 2. Reactions of primary *p*-hydroxylbenzyl alcohols and their ethers with chlorine

units, at least α-carbinol and α-ether groups can be displaced easily. The rate of the reaction appears to be of the order 1/10 of the rate of corresponding substitution reaction in the case of a primary carbinol group[8].

A free phenolic hydroxyl group in position *para* to the side-chain exerts an enhancing effect on the displacement reaction. In addition to carbinol and carbinol ether groups, also α-carbonyl and carboxyl groups can be displaced rapidly.

(c) The hydrolysis of aromatic ether groups by the catalytic action of molecular chlorine

The relative ease of demethylation of lignin in the chlorination reaction has been known for a long period of time[9]. As has been recently demonstrated[10] the methoxyl loss is due to the formation of methanol, which can be recovered from the bleach liquor and is quite stable to the action of molecular chlorine. Model compound studies suggest that the methoxyl groups are hydrolysed to free phenolic hydroxyl groups and methanol by the catalytic action of molecular chlorine[8]. A similar hydrolysis of ether linkages between monomeric units in lignin appears likely.

(d) Oxidation of chlorinated catechol groups to various quinone structures

These reactions are known to occur but are not advantageous for the bleaching process because the chlorinated quinones easily polymerize to larger molecular aggregates which can be removed only with difficulty.

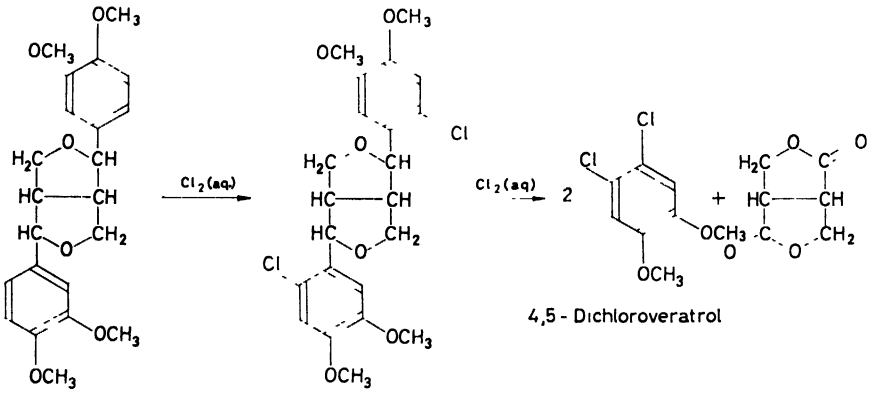


Figure 3. Chlorination of the dimethyl ether of pinoresinol

Some examples of the electrophilic displacement reactions on various model compounds are given in *Figures 2* and *3*. The occurrence of the displacement reaction in the end units of wood lignin has been demonstrated by converting them to tetrachloroguaiacol by chlorination in glacial acetic acid (*Figure 4*).

Model compound studies have shown that the hydrolysis of aromatic ether groups is subject to two inhibitive effects. Firstly, no de-alkylation reactions occur in the complete absence of water, not even in glacial acetic

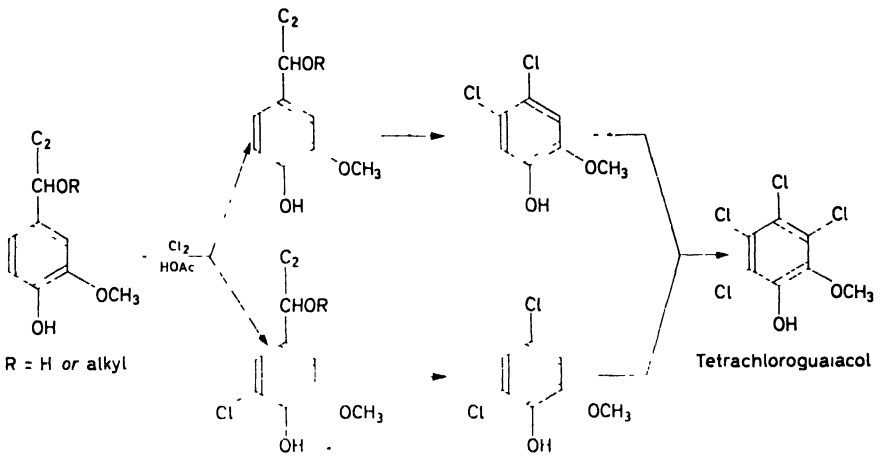


Figure 4. Reaction sequence for chlorination of end unit of spruce lignin *in situ*

acid. Secondly, the hydrolysis of veratryl-type model compounds is inhibited by two chlorine substituents *ortho* to the methylether groups. A similar inhibition effect has been observed in pyrogallol trimethylether derivatives. The effect is probably steric in nature. It is absent, however, in many compounds containing free phenolic hydroxyl groups, such as guaiacyl and syringyl derivatives.

Studies carried out on the chlorination of wood lignin and various lignin preparations suggest that both methoxyl groups and aromatic ether links are extensively hydrolysed by the catalytic action of chlorine. The reaction does not, however, reach completion, because of competitive chlorine substitution at positions 2 and 5. *Figure 5*, which presents the amount of liberated methanol in the chlorination of various lignins, illustrates the fact that only about 75 per cent of methoxyl groups in spruce wood and lignosulphonic acid are hydrolysed to methanol. The resistance of the remaining methoxyl groups is probably due to steric inhibition effects described above.

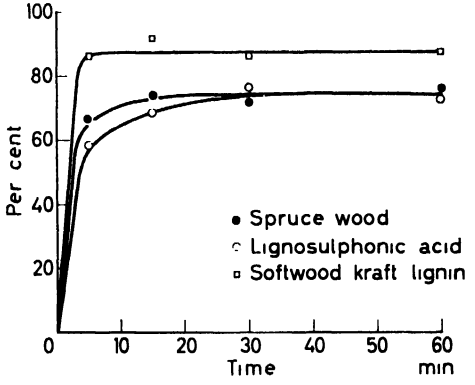


Figure 5. Production of methanol by chlorination

Figure 6 summarizes the reactions that occur in a guaiacyl propane unit during the chlorination. The reaction to occur first is chlorine substitution at either the 6- or 5-position of the nucleus. To what degree the next reaction, the electrophilic displacement of the side chain, takes place depends largely on the number of free phenolic groups and on the nature of the functional group bonded to the α-carbon of the side chain. Unfortunately, no adequate analytical means exist to determine the extent

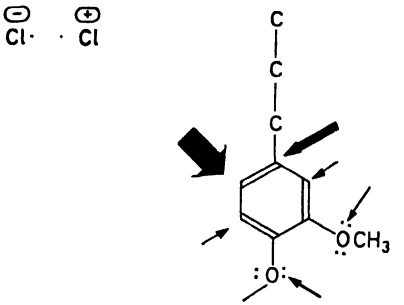


Figure 6. Sites reactive towards chlorine in a guaiacyl propane unit of lignin

of this reaction in various lignins. Since carbinol and carbinol ethers are displaced with particular ease, it would seem likely that this reaction is most extensive in the chlorination of native wood lignin and less extensive in the sulphite pulp lignin which contains sulphonic acid groups attached to the α-carbon in a substantial number of guaiacyl propane units. The hydrolysis of the methoxyl groups and 4-ether linkages proceeds to a stage that is limited by the amount of chlorine substitution at the positions 2 and 5.

It should be pointed out that side chains condensed in the 5-position are not likely to undergo electrophilic displacement reaction. Secondarily formed condensed structures can consequently be expected to resist the degradative action of chlorine.

The question of the reactivity of the side chain in the chlorination still remains open. In the past, efforts have been made to determine the aliphatically bound chlorine using alkaline hydrolysis data. However, since the chlorine atoms in chlorinated quinones are easily hydrolysed by alkali, estimations of this sort are hardly reliable. Since all reactions which occur at the aromatic nucleus are very rapid, the contribution of side-chain reaction in acidic chlorination is likely to be relatively insignificant.

Hypochlorite bleaching

As pointed out earlier, the primary reactant in the hypochlorite oxidation is probably hypochlorous acid, HOCl. The information available for evaluating the degradative mechanism of hypochlorite oxidation is rather limited, and the reaction in general is probably more complicated than acidic chlorination.

In the same way as molecular chlorine, hypochlorous acid reacts as a substituting agent and scattered pieces of information in the literature[12–14] indicate that it can cause electrophilic displacement reactions as well.

Some of the current ideas on the action of HOCl on phenols and phenol ethers are summarized in *Table 2.* According to de la Mare and co-workers[15], HOCl itself is rather unreactive in acidic solution, but the chloronium ion formed from the reaction between hypochlorous acid and hydronium ion is a very effective substituting agent. In reactions with reactive phenols and their ethers the rate-determining step appears to be

Table 2. Hypochlorous acid as substituting agent

(1) *Acidic solution*

Rate = k [H^+] [HOCl] (Reactive phenol ethers)
Reactive species Cl^+ (or H_2OCl^+)
o/p ratio higher than in case of Cl_2

(2) *Neutral and alkaline regions*

Phenol ethers unreactive
Free phenols: rate = k [$C_6H_5O^-$] [HOCl]; optimum pH = $\frac{1}{2}(pK_{C_6H_5O^-} + pK_{HOCl})$
Carbohydrates: rate probably 2nd order in [HOCl]
Decomposition to ClO_3^- and Cl^-: rate 2nd order in [HOCl]

the formation of chloronium ion, and the reaction is consequently zero order with respect to the substrate. The *o/p* ratio in substitution reactions is generally higher than in chlorinations with elemental chlorine, probably for reasons of steric nature.

In the neutral and alkaline regions, hypochlorous acid itself acts as a reactive species, but is only capable of reacting with negatively charged ions. This is indicated by the fact that most phenolic ethers are practically unreactive while free phenols are rapidly substituted according to the expression[16]:

$$\text{Rate} = k[C_6H_5O^-][HOCl]$$

As can be shown by a simple calculation, a maximum rate is obtained at a pH equal to the arithmetic mean of the pK values of hypochlorous acid (7·4) and the phenol. Ordinary phenols usually have a pK value of about 10. Consequently, the maximum rate region should be close to pH 9 or slightly below. With very reactive phenols, substitution reactions can be accomplished at as high a pH as 12.

These results would suggest that the etherified units in lignin might be quite unreactive in hypochlorite oxidation while the units with free phenolic hydroxyl groups should react with ease. This idea has received experimental support from the work by Richzenhain and Alfredsson[17] who showed that diazomethane methylation drastically reduced the rate of hypochlorite oxidation from about 8 moles to less than 2 moles per phenyl propane unit.

Richzenhain and Alfredsson have been able to show that the oxidative degradation in dimeric model compounds can continue, under specific circumstances, from an unetherified unit to the next etherified unit.

Thus the available data suggest that the attack of HOCl on the aromatic moiety of the lignin molecule is essential for successful degradation in hypochlorite bleaching, and this, in turn, is dependent on the number of free phenolic units present. In this respect the action differs from that of

elemental chlorine which is capable of degrading etherified and unetherified units alike.

The reactivity ratio between lignin and carbohydrates in hypochlorite oxidation is of substantial interest, because it is known that the danger of cellulose degradation is more acute in hypochlorite oxidation than in acidic chlorination. It was pointed out earlier that the optimum pH for lignin is in the neighbourhood of 9, if it contains " normal " phenolic hydroxyl groups, while a maximum attack on cellulose occurs at about pH 7·0. Use of this fact is made in the pH adjustment of hypochlorite bleaching by which the degradation of cellulose can be kept at a minimum.

It appears, however, that the situation might actually be more complicated due to certain kinetic effects. At least the rapid substitution of unetherified guaiacyl propane units is first order with respect to hypochlorous acid concentration. The oxidation of cellulose which according to Herbst[18] has an intermediate in common with hypochlorous acid self-decomposition, is probably second order with respect to hypochlorous acid concentration. If this is true, the lignin to carbohydrate reactivity ratio will depend, even at constant pH, on the concentration of HOCl, low concentrations favouring increased reaction with lignin.

Very little information is available to evaluate how the degradation reaction of aromatic nuclei continues after the initial chlorine substitution stage. The hypochlorite consumption curves by Richzenhain are equivalent to an oxidant consumption corresponding to the conversion of the rings to oxalic acid and various maleic acid derivatives. This appears reasonable, since both oxalic and maleic acids are relatively stable towards oxidation by hyprochlorite.

BLEACHING WITH CHLORINE DIOXIDE AND CHLORITES

The inorganic system used in chlorine dioxide and chlorite oxidations is substantially more complicated than that used in aqueous chlorine solutions. It is very difficult to select proper conditions to study the specific actions of chlorine dioxide or that of chlorite alone, because the interconversion of these two oxidizing species can often not be avoided. A further complication arises from the fact that chlorine dioxide and chlorite may become reduced to elemental chlorine (or HOCl) during the oxidation. Consequently, the oxidation effects by the former species become masked by the relatively complicated reactions of the chlorine–water system.

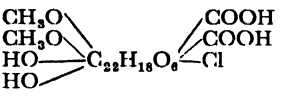
Only few degradation products have been characterized from the oxidation of lignin with chlorite and chlorine dioxide. Pearl[19] isolated 6-chlorovanillin from the filtrate of the chlorite holocellulose preparation of spruce wood. During the formation of this product the original side chain was oxidatively degraded and the aromatic ring remained intact; fumaric acid, isolated by Barton[20], probably derives its origin from aromatic rings. The monochloroacetic acid, isolated by the last-mentioned investigator, may again be a side-chain oxidation product.

It appears, therefore, that chlorine dioxide and/or chlorites may exert oxidative attack both on the side chains as well as on the aromatic nuclei. This complex action makes the interpretation of the analytical data by

Barton[20] and Purves and co-workers[21] on lignins oxidized by chlorine dioxide (*Table 3*) and chlorites somewhat difficult.

Table 3. Compositions of chlorine dioxide and chlorite lignins

Barton[20]:



Chlorite lignin *A*

Ether-solubles

$(COOH)_2$

trans—HOOCCH—CHCOOH

$CH_2ClCOOH$

Purves *et al.*[21]:

ClO_2—oxylignin

$C_{42}H_4O_6Cl_3(OCH_3)_2(COOH)_5$, (M.W. = 1193)

From the analytical data shown, it can be concluded that the following gross changes take place in the lignin molecule during the oxidation:

(i) reduction in methoxyl content;

(ii) increase in acidic groups which may be titrated, probably carboxyl groups. Purves has pointed out, however, that these groups may consist, in part, of hydroxyl groups associated with quinone nuclei, whose chemical behaviour cannot easily be distinguished from that of carboxyl groups;

(iii) introduction of chlorine in the molecule.

By alkaline saponification Purves *et al.* were able to almost double the number of acidic groups in their preparation; similar observations were made by Barton. This suggests that substantial amounts of lactone or ester groups might be present.

In all cases studied, the chlorine dioxide treatment results in the elimination of the 280 mμ maximum from the u.v. spectrum of lignin. This observation is generally interpreted to mean partial oxidative destruction of aromatic nuclei. Such reaction, if it occurs, would naturally account for the loss of methoxyl groups and contribute to the number of carboxyl and/or lactone groups.

This concept has received certain support from model compound studies. Already Schmidt and co-workers[22] were able to show that various phenols, phenol carboxylic acids and nitrophenols are degraded to maleic and oxalic acids by chlorine dioxide. Model compounds which possess closer structural relationship with lignin were later studied by Husband, Logan, and Purves[23]. It was shown by them that vanillin is converted to a crystalline compound $C_7H_5O_4(OCH_3)$. Subsequent studies[24] have shown that

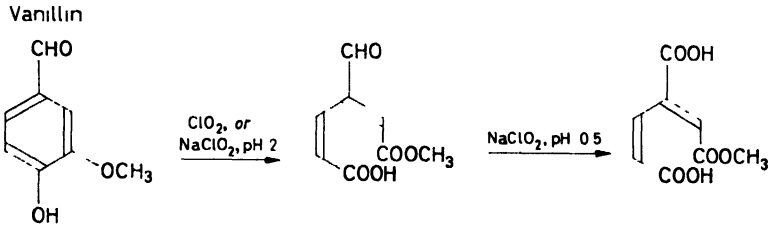


Figure 7. Oxidation of vanillin by chlorine dioxide and sodium chlorite

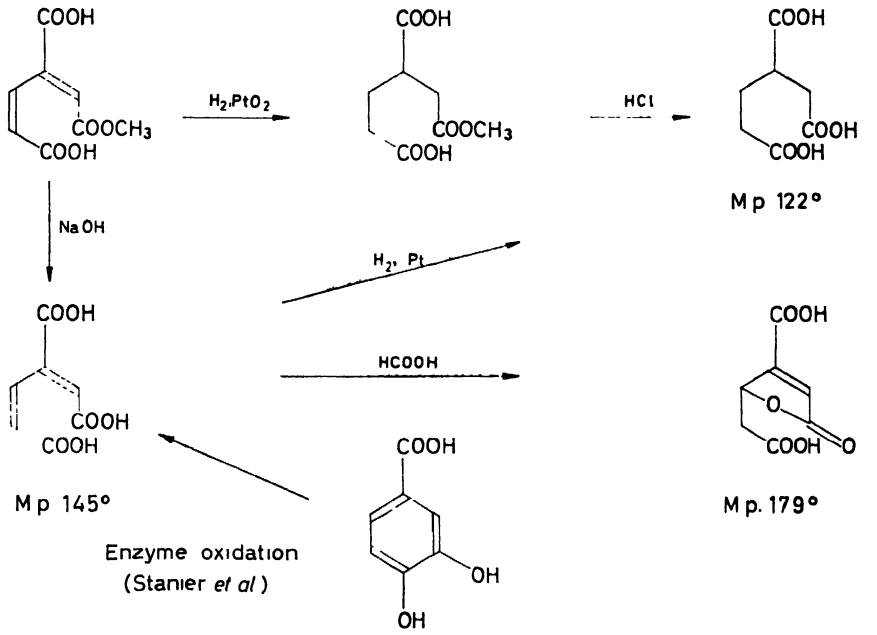


Figure 8. Reactions used for the structure assignment of β-formyl muconic acid monomethyl ester

the compound has the structure of β-formyl muconic acid monomethyl ester (*Figure 7*) which by chlorite oxidation is converted to corresponding muconic acid ester. *Figure 8* summarzies the reactions utilized for structure assignment.

Similar ring opening reaction occurs in the chlorine dioxide (or chlorite) oxidation of vanillyl alcohol[25], although the reaction product is isolated in the form of a δ-lactone (*Figure 9*). Consequently, an oxidative ring

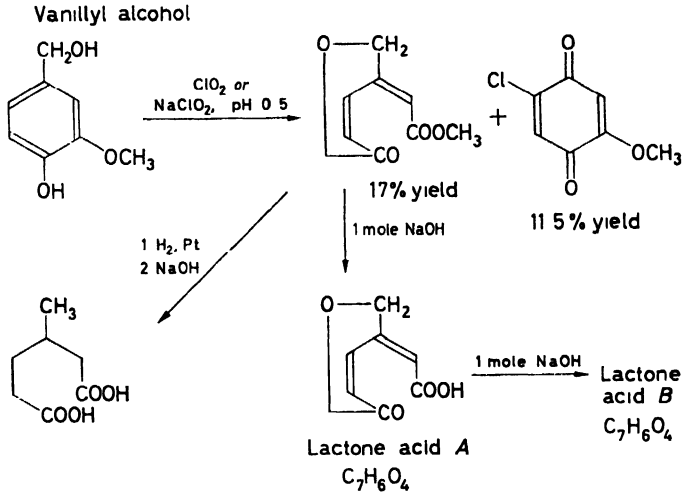


Figure 9. Oxidation of vanillyl alcohol by chlorine dioxide and sodium chlorite

opening between the carbons connected with the methoxyl and hydroxyl groups appears relatively general in guaiacyl derivatives.

2-Chloro-5-methoxybenzoquinone is an additional product from the oxidation of vanillyl alcohol. Quinones, with or without a chlorine substituent, are frequently obtained as products from the chlorine dioxide oxidation of various models[26].

Both in the ring opening and in the formation of methoxy quinones, the original aromatic methoxyl is converted to a less stable form which can be easily hydrolysed to methanol, while the methoxyl groups connected with unchanged aromatic nuclei are not hydrolysable under mild conditions. As a consequence, the amount of easily hydrolysable methoxyl groups can serve as a rough estimate on structures that have undergone either ring opening or conversion to methoxyquinones.

Table 4. Amount of hydrolysable methoxyl groups in model compounds and lignin preparations after oxidation with chlorine dioxide

Substance	*Methanol recovered (% of original* MeO)		
	Formed in oxidation	*Liberated in hydrolysis*	*Total*
Vanillyl alcohol	53	34	87
Veratryl alcohol	65	23	88
Lignosulphonic acid	58	7	65
Birch wood	27	7	34
Spruce wood	33	6	39

Table 4 presents some determinations of this sort carried out on model compounds and on various lignin preparations[25]. The chlorine dioxide and chlorite oxidations were performed under mild conditions to keep at minimum the hydrolysis of the ester and quinone-methoxyl groups, as well as the further oxidation of these structures to low molecular weight compounds. The results obtained in the oxidation of vanillyl alcohol are in good agreement with the combined yields of muconic acid and methoxyquinone derivatives which were actually isolated.

The values obtained for various lignins are, however, quite low, much lower actually than the free methanol formed directly during the oxidation reaction. These results unquestionably discourage the idea of regarding the formation of muconic acid esters and methoxyquinones as major degradative processes in the chlorine dioxide oxidation of lignin. Since, however, the amount of methanol directly liberated in the oxidation is relatively large and, furthermore, the u.v. absorption of oxidized lignins indicates extensive modification of the aromatic nuclei, other reaction pathways resulting in reduced aromaticity merit consideration.

One such possibility consists of the conversion of aromatic nuclei to furan derivatives. It was shown by Pearl and Barton[27] that vanillin, when oxidized with sodium chlorite in acetic acid solution, gives small amounts of furan-2, 4-dicarboxylic acid. Later studies[28] have shown that substantially higher yields of this compound (up to 15 per cent) are obtained when vanillic or

syringic acids are oxidized with aqueous chlorine dioxide or sodium chlorite. No furan-2,4-dicarboxylic acid is formed in the chlorite oxidation of β-carboxymuconic acid or its methyl ester, obviating the possibility of a common pathway.

It is obvious that data available on the chlorine dioxide and chlorite oxidations are insufficient to create more than a very preliminary picture of the chemistry of the process. The same is true, to a more pronounced degree, for the mechanism of peracetic acid delignification. Peracetic acid resembles chlorine dioxide in its capacity to oxidize phenols, catechols and their ethers to muconic acid derivatives. To what degree this similarity is related to the fact that both chlorine dioxide and peracetic acid are excellent delignifying agents, poses an intriguing problem for future research.

Anyone who visits a modern pulp bleaching plant cannot but be impressed by the scope of the operation and by the elaborate control devices. We are, unfortunately, only in the early stages of understanding the chemistry of bleaching processes, and it may be asked with good reason whether the basic knowledge of bleaching reactions lags behind the well-developed engineering of the bleaching operation.

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THE PHYSICAL CHEMISTRY OF LIGNIN

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INTRODUCTION

Perhaps we may begin with a rather naive view of the physical nature of lignin. Wood is an intimate mixture of polymeric components which interpenetrate each other to form a three-dimensional structure. It has been shown with a fair degree of certainty that the lignin moiety in wood is comprised of chains of phenylpropane units[1]. The chains are probably crosslinked to each other in a number of different ways to give an extensive network[2]. When the lignin is made soluble, fragments of the degraded network enter into solution as macromolecules of various sizes.

From this simplified picture it can be seen that there are two broad aspects to the physical behaviour of lignin. The first is concerned with the molecular properties of soluble lignins. Here, techniques such as viscometry, sedimentation, light-scattering and diffusion yield information on the size and configuration of the macromolecule in solution. The second type of investigation deals with the bulk properties of the solid or solvent-swollen polymer.

In recent years the behaviour of lignin derivatives in solution has been the subject of increasing study. Outstanding in this respect is the pioneering work of Gardon and Mason[3,4] and McCarthy *et al.*[5,6] on the lignin sulphonates. In contrast, scant reference can be found in the literature to measurements on isolated lignins of properties such as specific volumes or heats, accessibility, second-order transitions, softening or tack temperatures, viscoelasticity, adhesion and fracture energies. No doubt the dearth is due to the difficulty of isolating from wood a reasonably unchanged lignin in a form suitable for such studies.

The present paper is an outline of some of the work and ideas of my colleagues and myself on the physico-chemical behaviour of isolated lignins. Most of the results and conclusions are for softwood lignins and highlight the need for similar studies on lignins from deciduous trees. Many references to the results of others are included but no attempt has been made to review the entire field.

THE POLYDISPERSITY OF SOLUBLE LIGNINS

It is immediately apparent in reviewing the literature that the molecular weights reported for soluble lignins cover an immense range. McCarthy *et al.*[6] fractionated lignin sulphonates and found molecular weights as low as 260. In contrast, a weight average molecular weight of 50,000,000 was determined on a high molecular weight fraction of alkali-cooked periodate lignin[7]. Evidently, there can exist in solution, under the name of

lignin, molecules small enough to be well-defined chemical entities and others large enough to be regarded as colloidal particles.

A wide range of molecular size is also found when individual samples of soluble lignins are fractionated. This is illustrated in *Table 1*. Data for several fractionations of various types of soluble softwood lignins are given. Work on the sulphonates predominates but polydispersity is also apparent for organosolv lignins prepared by the dioxan-HCl extraction described by Pepper *et al.*[10]. Thus it seems likely that polydispersity may be a prominent feature of soluble softwood lignins regardless of the manner in which they are made soluble. In routine measurements of the molecular weights of soluble lignins, this property produces certain difficulties to be discussed in a later section.

Table 1. Molecular weight polydispersity of softwood lignins

Wood	*Method of isolation*	*Method of fractionation*	*Range of molecular weight*	M_w/M_n	*Ref.*
Balsam and fir	Sulphonation	Dialysis and ultrafiltration	3,700→58,000	2·2	3
Hemlock and fir	Sulphonation	Elution from column	400→150,000	7·0	6
Hemlock	Sulphonation	Precipitation with non-solvent	440→58,000	6·4	6
Hemlock	Sulphonation	Precipitation with non-solvent	1,400→96,000	3·3	6
Hemlock	Sulphonation	Precipitation with non-solvent	490→70,000	5·8	6
Spruce	Sulphonation	Successive cooking	1,700→24,000	3·3	8
Spruce	Alkali cook of periodate lignin	Precipitation with Ba^{2+}	35,000→17,000,000	5·7	7
Spruce	Dioxan and HCl	Successive extraction	2,900→50,000	2·7	9

It is interesting to remember that marked chemical differences have seldom been reported between fractions of any particular sample of a soluble lignin derivative isolated from the wood of gymosperms. Changes have been noted in the methoxyl content[3], ultra-violet light absorption[6] and C, H and O analysis[11], but in general such variations are relatively small[7,8,9,12,13]. Thus, soluble softwood lignins may be regarded as complex polymeric macromolecules, built up of the familiar lignin building blocks[14], differing only slightly in detailed chemical structure but showing large differences in molecular size.

Detailed results for the molecular weights of fractions of soluble hardwood lignins, analogous to the data for softwood lignins summarized in *Table 1*, have not been published. Therefore, it is not possible to draw conclusions on the polymolecularity of soluble lignins from deciduous wood. The few determinations available suggest that soluble hardwood lignins have lower molecular weights than softwood lignins. For example, McCarthy *et al.*[15] estimated that the molecular weights of lignin sulphonates from maplewood were between 800 and 2,100. The corresponding range for spruce lignin sulphonate was 2,800 to 14,000.

It is instructive to compare the molecular dispersity of soluble lignins with that of certain other macromolecules derived from biological tissue.

Many proteins, such as enzymes or seed globulins, are monodisperse or show an extremely narrow distribution of molecular weights[16]. On the other hand, linear polysaccharides like cellulose, amylose or certain hemicelluloses are invariably isolated in a polydisperse state. Usually the degree of polydispersity is not great. The ratio of the weight average molecular weight M_w to the number average molecular weight M_n is approximately 2. This is the ratio expected for molecular weight distribution produced by the random degradation of a linear polymer[17]. Some values of M_w/M_n derived from recently published data on cellulose nitrate, xylans and amylose are given in *Table 2.*

Table 2. M_w/M_n for several naturally-occurring macromolecules

Macromolecule	*Source*	$M_w \times 10^{-5}$	M_w/M_n	*Ref.*
Cellulose nitrate	Birch	27[a]	1·9[b]	18
Cellulose nitrate	Ramie	2·4[a]	1·7[b]	19
Amylose	Potato	8·8[c]	1·9[c]	20
Xylan	Birch	0·8[a]	2·3[d]	21
Xylan	Elm	0·7[a]	2·4[d]	21
Amylopectin	Waxy corn	1,700[a]	116[e]	22
Hydrolysed amylopectin	Waxy corn	15[a]	25[e]	22
Glycogen	Sweet corn	190[a]	15[e]	22
Hydrolysed glycogen	Sweet corn	20[a]	6·3[e]	22
Glycogen	Rabbit liver	390[a]	6·6[f]	23

[a] M_w by light scattering.
[b] M_n by viscometry from $[\eta] = 0.0091\ D\ P$.
[c] Calculated from fractionation data.
[d] M_n by osmometry.
[e] M_n from the alkali number.
[f] M_{ww} from sedimentation and diffusion used instead of M_n; usually $M_w > M_{ww} > M_n$.

There exists a third class of soluble macromolecule which is produced either by the synthesis or degradation of a three-dimensional network. Two examples which have been studied extensively in recent years are amylopectin and glycogen. These materials are made in plant and animal tissue by the enzymic coupling of glucose monomers. In this respect, glucose is polyfunctional and contains an aldehydic group and two alcohol groups (4 and 6) capable of bonding to a second glucose unit. In solution, amylopectin and glycogen are characterized by an extremely wide polydispersity. Some examples of M_w/M_n are given in *Table 2.* It is seen that in contrast to the linear polysaccharides, M_w/M_n can exceed 100. Erlander and French[24] have evolved a quantitative treatment of the degradation of such branched polymers and have related the polydispersity to the relative reactivities of the functional groups in the glucose monomer.

The wide polydispersity of the soluble softwood lignins suggests that they fall into the same category as the amylopectin or glycogen systems. The M_w/M_n ratios given in *Table 1* all exceed 2 and in one case the ratio is 7·0. These values were computed statistically from the weight percentage and molecular weights of the individual fractions obtained by fractionation of a particular source of lignin. Undoubtedly M_w/M_n would be even higher if weight and number average molecular weights were measured directly on the original unfractionated lignin.

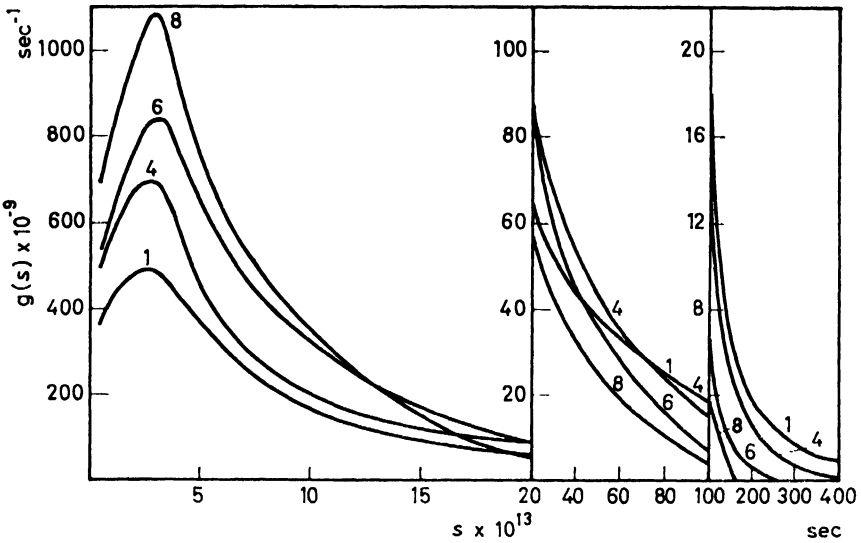


Figure 1. Distribution of sedimentation coefficients for alkali lignin fractions B-1, B-4, B-6 and B 8 described in Reference 25

The excessive polydispersity of soluble lignin is also demonstrated in the ultracentrifuge[25]. Distribution of sedimentation coefficients, s, for fractions of alkali periodate lignin is shown in *Figure 1.* Values of s from $0{\cdot}5 \times 10^{-13}$ to over 400×10^{-13} sec. were recorded. The low molecular weight material was distributed relatively closely about a sedimentation coefficient of 3×10^{-13} sec. The heavier molecules were present as a long, polydisperse " tail ". Fractions of successively lower molecular weight had smaller proportions of the heavy tail. Strikingly similar results have been obtained in degradation studies of glycogen[23,26,27] and amylopectin[27].

THE CONFIGURATION OF SOLUBLE LIGNINS

A characteristic feature of soluble lignin derivatives is the low viscosity which the macromolecule imparts to the solution. In *Table 3,* the intrinsic viscosity, $[\eta]$ which corresponds to a molecular weight of 50,000 is given for lignin sulphonate and dioxan-HCl lignin from spruce wood. Included for comparison are data for cellulose nitrate, xylan and two synthetic polymers. It is quite evident that the lignins give rather small intrinsic viscosities compared with other polymeric materials. The intrinsic viscosity is a measure of the effective hydrodynamic volume (per unit weight of dry solute) which the macromolecule presents to the shear field in the solvent. For a hard, non-swelling sphere $[\eta]$ in 100 ml/g is given by the Einstein equation

$$[\eta] = 0{\cdot}025\bar{V} \qquad (1)$$

where $\bar{V}$ is the specific volume of the material of the sphere. When the sphere is a macromolecule comprised of a porous, solvent-swollen network, $\bar{V}$ in equation (1) becomes the volume of the swollen macromolecules per

unit weight of solute. Thus $\bar{V}$, and therefore $[\eta]$, increases with increased swelling or extension of the macromolecule in the solvent. A low value of $[\eta]$ at a given molecular weight indicates that the molecule is compact and takes up relatively little volume in the solution. The data in *Table 3* show that, for a molecular weight of 50,000, soluble lignins occupy about 1/15 the volume of cellulose nitrate and about 1/3 the volume of poly-(α-methylstyrene).

Table 3. Intrinsic viscosities corresponding to a molecular weight of 50,000 for lignins and other macromolecular systems

Macromolecule	*Solvent*	$[\eta]$ 100 ml/g	*Ref*
Dioxan-HCl lignin	Pyridine	0·08	9
Sodium lignin sulphonate	0·1M NaCl	0·10	8
Poly(methyl methacrylate)	Benzene	0·23	28
Poly(α-methylstyrene)	Toluene	0·24	29
Xylan	Dimethyl sulphoxide	0·5	21
Cellulose nitrate	Ethyl acetate	1·4	30

Polymer configuration is also elucidated by the variation of parameters such as the intrinsic viscosity, diffusion constant or sedimentation coefficient with molecular weight. The exponent, a in the familiar relationship:

$$[\eta] = K_\eta M^a \qquad (2)$$

can vary between 0 and 1 depending on the configuration of the macromolecule[17]. In *Table* 4 exponents obtained for some soluble lignins are compared with theoretical values for model particles and also with results found for cellulose nitrate and other macromolecules. It is seen that

Table 4. Values of exponents a, b and c for lignins and other macromolecular systems

Macromolecule	*Solvent*	a	b	c	*Ref.*
Einstein sphere	- -	0*	0·33†	0·67†	—
Dioxan-HCl lignin	Pyridine	0·1	0·4		9
Alkali lignin	0·1M buffer	0·32	—	0·52	7
Lignin sulphonate	2M NaCl	0·47	-	—	4
Lignin sulphonate	0·5M NaCl	—	0·56	-	4
Lignin sulphonate	0·02M NaCl		0·57	—	5
Lignin sulphonate	0·1M NaCl	0·44	0·48	-	8
Non-free draining coil	-	0·5–0·8	0·5–0·6	0·4–0·5	17
Poly(α-methylstyrene)	Toluene	0·71	—		29
Poly(methyl methacrylate)	Benzene	0·73	0·57	0·43	28
Cellulose nitrate	Ethyl acetate	0·99	0·86	0·17	31
Free draining coils	-	1	1	0	32

* Equation (1).
† From Stokes' law.

a for soluble lignins lies between the expected value for an Einstein sphere and a non-freedraining random coil. This is in marked contrast to the behaviour of cellulose nitrate where a is 0·99, near the value expected for a completely freedraining coil.

Similar relationships may be established for D, the diffusion constant[17],

$$D = K_D M^{-b} \tag{3}$$

and for s, the sedimentation coefficient[17],

$$s = K_s M^c \tag{4}$$

The values of b and c for the lignins shown in *Table 4* lie between those expected for a non-freedraining random coil and an Einstein sphere.

The variation in hydrodynamic parameters with molecular weight suggests that soluble lignins are not hard, solvent-impermeable spheres. Yet they do not show fully the Gaussian behaviour expected of linear polymers. A suitable model for such a macromolecule would be a compact microgel (33–35) made up of crosslinked material, completely non-freedraining but capable of limited swelling. The compactness of such a macromolecule would produce the low values of $[\eta]$ shown in *Table 3*. Also the wide polydispersity in *Table 1* would be expected whether microgel was formed by random degradation of a network in the wood or by a polymerization reaction during the isolation of the lignin.

A diagrammatic representation of a water-soluble sulphonate microgel is shown in *Figure 2*. The bulk of the molecule is assumed to be made up

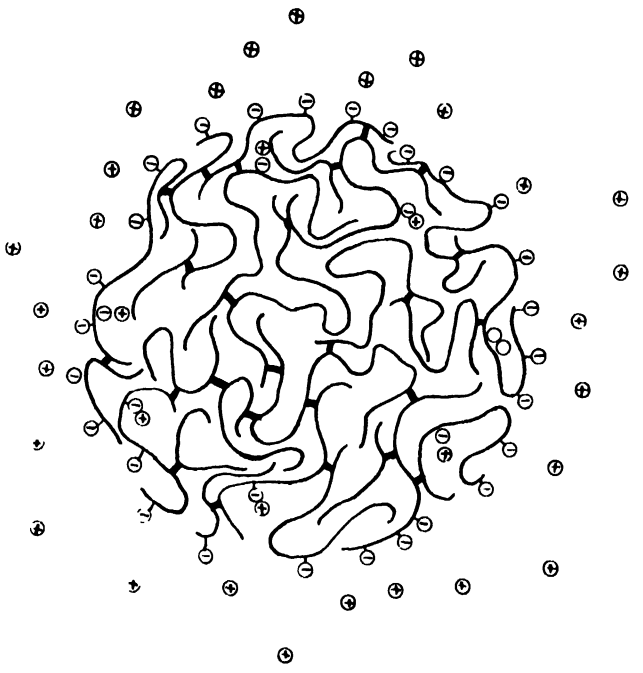


Figure 2. Idealized spherical model of the spruce lignin sulphonate macromolecule

of crosslinked, polyaromatic chains which are randomly coiled. The negatively charged sulphonate groups, near the surface of the particle, maintain its solubility in water. A double layer of counter ions is present in the solvent in the characteristic Boltzmann distribution.

The model shown in *Figure 2* was originally proposed to permit a quantitative theoretical treatment of the polyelectrolyte expansion of certain high molecular weight lignin sulphonates[36]. The data in *Tables 3* and *4* suggest that all high molecular weight, soluble lignins from softwoods may have a configuration approximated by the microgel model. One would expect differences in the degree of crosslinking, charge and swelling, depending on the method of isolation. Some such variations are indicated by the trends of the data for various lignins in *Tables 3* and *4*. In practice, the microgel particles would not be spheres but irregularly shaped, submicroscopic "hunks" of gell in a wide polydispersity of size. Also, for molecular weights lower than 5,000, it is unlikely that the model shown in *Figure 2* would be applicable. In spite of these reservations it seems probable that the microgel model is appropriate for high molecular weight, softwood lignins in organic or aqueous solvents.

MOLECULAR WEIGHT DETERMINATIONS

A common requirement in the routine characterization of polymers is a simple and reliable method of measuring the molecular weight. In this respect the soluble lignins present a special problem. Many references to the determination of number average molecular weights exist, and here the definitive work of Gross, Sarkanen and Schuerch[37] may be mentioned. In the present section a brief discussion of the determination of weight-type averages for lignins is given and the limitations of certain of the methods are discussed.

It may be said at once that the widely used method of measuring $[\eta]$ and then determining M from a relationship such as equation (2) is not to be recommended for the soluble lignins. The intrinsic viscosity of a particular lignin will be influenced not only by the molecular weight but also by the ability of the microgel to swell. Differences in the degree of crosslinking or charge density (in aqueous solutions) will affect $\bar{V}$ and thus $[\eta]$ in equation (1). Hence, it is not feasible to establish a unique relationship between M and $[\eta]$ for a particular lignin derivative in a given solvent. A further objection to such use of the intrinsic viscosity is its relative insensitivity to changes in molecular weight. This is particularly evident from the low value of a for the dioxan-HCl lignins in *Table 4*.

Similar reservations may be made for the use of the diffusion coefficient to measure the molecular weight by means of a relationship such as equation (3). McCarthy *et al.*[5,6,12,15] and Benko[38] have characterized lignin sulphonates in this manner and the latter author has made it clear that the molecular weights so obtained are only relative. The diffusion coefficient is given by[39]:

$$D = \frac{kT}{f} \tag{5}$$

where k is the Boltzmann constant, T the absolute temperature and f the

frictional constant of the macromolecule. If the molecule is approximately spherical in shape[39]:

$$f = 6\pi\eta_0 r \tag{6}$$

in which $[\eta_0]$ is the viscosity of the solvent and r the radius of the sphere. The volume of the sphere is related to its hydrodynamic specific volume, $\bar{V}$ by:

$$\frac{4}{3}\pi r^3 = \frac{\bar{V}MI}{N} \tag{7}$$

where N is the Avagadro number.

Thus, the relationship between D and M for a spherical molecule is obtained by combining equations (5), (6) and (7) to give:

$$D = \frac{kT}{\eta_0}\left(\frac{N}{162\pi^2\bar{V}MI}\right)^{1/3} \tag{8}$$

It is evident from equation (8) that variations in D will be linked not only to changes in M but also to the previously mentioned dependence of $\bar{V}$ on the swelling capacity of the microgel. A unique relationship between D and M may not be generally valid. However, it is interesting to note that the effect is small in the case of diffusion since $D \propto \left(\frac{1}{\bar{V}}\right)^{1/3}$ in equation (8) in contrast to the direct proportionality of $\bar{V}$ and $[\eta]$ in equation (1).

Elimination of $\bar{V}$ is possible by combining equations (1) and (8) to give:

$$M = \left(\frac{kT}{\eta_0}\right)^3 \frac{N}{6480\pi^2[\eta]D^3} \tag{9}$$

Equation (9) provides a method of determining M by measurement of $[\eta]$ and D in the same solution. Uncertainties due to the degree of swelling are eliminated. The assumptions involved are as follows:

(*a*) The macromolecule is non-freedraining and approximately spherical.

(*b*) The Einstein (equation 1) and Stokes (equation 6) relationships are valid.

From arguments advanced in the previous section it seems likely that assumption (*a*) is justified for soluble softwood lignins. Both the Einstein[39] and the Stokes[40] equations have been verified for macroscopic spherical particles and Polson[41,42] and Edward[43,44] have suggested, in recent work, that the approximate validity of equation (9) extends to proteins and even smaller molecules. Thus, assumption (*b*) is probably also valid in the range of precision usually required for characterization of soluble lignin derivatives.

By a measurement of the diffusion constant and sedimentation coefficient on any given sample the molecular weight can be computed from Svedberg's[45] well-known equation:

$$M = \frac{sNkT}{D(1 - \bar{v}\rho)} \tag{10}$$

in which ρ is the density of the solution. In equation (10), $\bar{v}$ is the partial specific volume, *i.e.*, the volume of solvent displaced per g of dry macromolecule; thus $\bar{v}$ does not include solvent immobilized by a swelling microgel in contrast to the term $\bar{V}$ in equations (1) and (8).

Although a mean diffusion constant for lignins is fairly easily obtained measurement of the sedimentation coefficient is complicated by the wide polydispersity of the macromolecule. An extreme example of this was found for the alkali lignins[25,46]. The maximum ordinate sedimentation coefficient was found to decrease by factors of up to 10 when the speed of rotation was increased from 12,000 to 60,000 rev/min. The explanation was that different fractions of the same sample were contributing to the peak at the two speeds. The anomaly could probably be resolved by use of a synthetic boundary cell and a suitable integration procedure for determining a weight average value of s. Such methods tend to be laborious compared with the simple procedure of measuring the velocity of the peak.

A more promising method for soluble lignins is the short column sedimentation equilibrium technique proposed by Van Holde and Baldwin[47] and by Yphantis[48]. The technique has all the advantages of the classical equilibrium method but precision is sacrificed for speed by working with liquid columns of only 1 mm in depth. Equilibrium is then reached in a few hours instead of days as with the older method[39]. Weight average molecular weights are obtained from

$$M_w = \frac{RT}{\bar{r}c_0\bar{\omega}^2(1 - \bar{v}\rho)}\left(\frac{dc}{dr}\right)_{r=\bar{r}} \tag{11}$$

in which c_0 is the initial concentration, $\bar{\omega}$ is the angular velocity and r is the distance from the centre of the rotor. The radius, $\bar{r}$ corresponds to the point in the column at which $c = c_0$. For a short column ($c = 1$ mm) this point is half-way between the meniscus and base of the column. Thus with the customary Schlieren optics, a determination is made simply by measurement of the gradient dc/dr at the mid-point of the column when equilibrium has been established. Results obtained with the equilibrium technique on soluble softwood lignins are currently being compared with molecular weights based on diffusion and viscometry, and calculated by means of equation (9).

The light scattering method has been extensively used for the determination of weight average molecular weights of macromolecules in solution[5,7,18,19,21]. The method is reproducible and can be reasonably precise when care is taken in preparation and handling of the solutions[49]. The problem of the removal of dust has been completely solved by the

elegant ultra-centrifugation technique of Dandliker and Kraut[21,49,50]. Even the presence of deep colour and strong fluorescence as found with soluble lignins can be accommodated with relatively minor refinements[7]. There remains, however, a basic characteristic of the technique which makes it unsuitable for routine molecular weight determinations of the soluble lignins.

From light scattering one obtains a " comprehensive " weight average molecular weight which includes all the particles in solution. With the type of distribution indicated by the sedimentation data in *Figure 1* for alkali lignin, a comprehensive weight average will be characteristic only of the high molecular weight part of the heavy tail. The molecular weight obtained will refer to a small proportion (perhaps < 5 per cent) of the sample and will be unaffected by changes in molecular weight of the major part of the material. Any clarification technique which is applied will tend to cause drastic changes in the light scattering molecular weight, M_{LS}, although only a small proportion of the sample is removed by the purification[21].

This objection to the light scattering technique is particularly apt for aqueous solutions in which aggregates of large particle size are readily produced and removed only with difficulty. In a recent paper on polyanhydroglucose addition polymers, Abe and Prins have noted ratios up to 15 for M_{LS}/M_W where M_W was determined by a sedimentation technique[51]. A similar discrepancy has also been obtained for aqueous solutions of carboxymethylcellulose[52]. Until some way around the problem is found, the comprehensive weight average molecular weight given by light scattering is likely to be unrepresentative of the molecular weight of the bulk of the sample of lignin.

The question now remains as to what method can be used to give a weight average which is representative of the greater part of the sample. The answer would be a technique which is not affected by the small proportion of very high molecular weight material. Short-column, sedimentation equilibrium falls into this category, because the very large particles are sedimented as a layer to the bottom of the cell before equilibrium is reached. The diffusion-viscosity method also would be expected not to be affected by a small proportion of very large particles. The type of average obtained for M will depend on the type of average for D substituted in equation 9. A weight average M will be given by a D corresponding to a weight average frictional constant. In a recent investigation[9], it has been shown empirically that if the so-called "area" diffusion constant D_A [53] is substituted in equation 5, an f value is obtained which approximates to a weight average f. Thus, substitution of D_A in equation (9) yields a value of M which approaches a weight average. At the same time the presence of a small proportion of very heavy particles will have a negligible effect on the shape of the diffusion patterns obtained.

It should be noted that the data from References 8 and 9 in *Tables 1 3* and *4* are based on molecular weights obtained by the diffusion viscosity method. On the other hand, the results in *Tables 1* and *4* from Reference *7* as well as the data in *Figure 6* are based on light scattering molecular weights.

POLYELECTROLYTE BEHAVIOUR

Gardon and Mason[4] first pointed out that lignin sulphonates behaved as swelling polyelectrolytes in aqueous solution. A characteristic feature of a polyelectrolyte is that the molecule expands when the concentration of counter-ion is decreased. When a solution of a polyelectrolyte in water is diluted with water (or 10^{-5} M NaCl) there is a marked increase in the reduced viscosity, due to the swelling of the molecule by the reduction in the ionic strength. In contrast, use of a high ionic strength aqueous

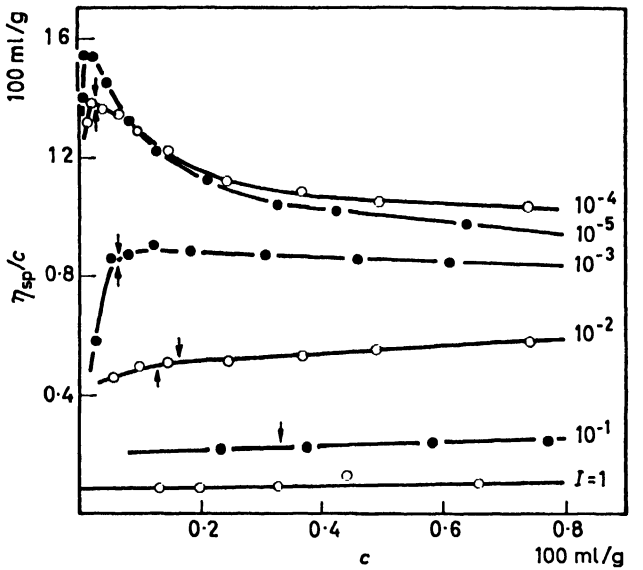


Figure 3. η_{sp}/c *v. c* for a sodium lignin sulphonate (fraction S–7, Reference 54) at ionic strengths from 1M to 10^{-5}M NaCl. The upward pointing arrows are theoretical predictions of the point of maximum bending while the downward pointing arrows correspond to the experimentally determined maxima

solvent such as 1M NaCl results in the linear graphs of $\eta_{sp/c}$ *v. c* usually found for synthetic polymers in non-polar solvents. This behaviour is shown in *Figure 3* for a high molecular sodium lignin sulphonate from spruce periodate lignin[36]. The maxima in the graphs have been shown to be due to intermolecular interaction which predominates at dilutions so low that the expansion has virtually ceased[54].

To obtain a linear plot of $\eta_{sp/c}$ *v. c* at low values of the ionic strength (I_E) it is necessary to dilute the solution so that the Na^+ counter-ion concentration remains constant[55,56]. The graphs of $\eta_{sp/c}$ *v. c* over the range of ionic strengths shown in *Figure 4* were determined by such an iso-ionic dilution[36]. From this data the variation of $[\eta]$ with ionic strength was measured for values I_E down to 0·00002 M. It was in order to explain certain unusual features of the dependence of $[\eta]$ on I_E that the idealized model of the lignin sulphonate macromolecule shown in *Figure 2* was proposed[36].

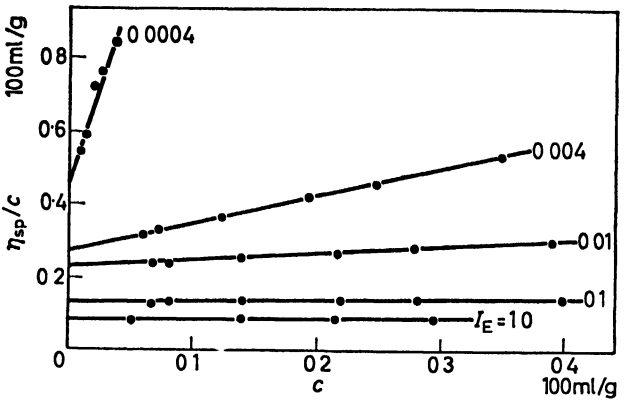


Figure 4. $\eta_{sp/c}$ *v.* c by iso-ionic dilution at various values of the ionic strength I_E for a sodium lignin sulphonate (fraction PS–5, Reference 36)

It is probable that the crosslinked, polyaromatic structure of lignin must acquire hydrophilic, charge-bearing groups in order to become water soluble. Thus any water-soluble lignin would be expected to show polyelectrolyte properties[57]. In this respect results similar to those in *Figure 3* have been obtained for alkali lignin prepared by cooking spruce periodate lignin in sodium hydroxide[58].

It is interesting to note that the lignin can acquire polyelectrolyte properties without becoming soluble. If the process of attaching charge groups to the lignin does not rend the network into soluble fragments, it will then become a polyelectrolyte gel showing the bulk properties of swelling or contracting in aqueous media, depending on the pH or ionic strength. It seems likely that the residual lignins, left on the fibres in the various semi-chemical or high yield pulping processes, would be in this category.

DISPERSING PROPERTIES

For some time water-soluble lignins have been used as cheap dispersing agents in colloidal systems. In a recent study of this effect[59] rather pronounced differences were found in the dispersing capacity of certain fractions of lignin sulphonate and alkali lignin.

Dispersion was measured by the decrease in viscosity produced in an aqueous suspension of titanium oxide (TiO_2) by the addition of 0·15 per cent lignin to the aqueous phase. Measurements were made with a Drage rheometer, and a standard dispersion number (SDN) was defined at a given rate of shear by

$$\text{SDN} = \frac{\text{shear stress of control suspension}}{\text{shear stress of dispersed suspension}} \qquad (12)$$

The results showed that some lignins were good dispersants whereas others were not dispersants at all for the conditions under which the standard dispersion numbers were measured. Several SDN values were greater

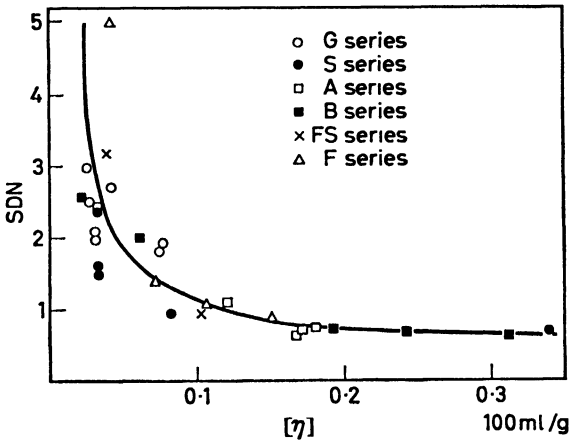


Figure 5. Standard dispersion number *v.* intrinsic viscosity for soluble lignin derivatives. The F-series are alkali lignins (see Reference 58) while all the others are lignin sulphonates described in Reference 59

than 5, while certain samples had SDN less than unity, *i.e.*, addition of the lignin increased the viscosity of the suspensions.

As shown in *Figure 5*, a well-defined, hyperbolic relationship was found between SDN and the intrinsic viscosities of the lignins. As expected from the correlation of $[\eta]$ and M shown in *Table 4*, a similar dependence of SDN on molecular weight was noted. The maximum in the graph of SDN *v.* M in *Figure 6* was caused by the abrupt drop in the dispersing power of the *D* series of fractions. These were separated by dialysis through a cellophane bag. All lignins were adsorbed on the titanium oxide to a mean value of 95 per cent, with the exception of the *D* series which were adsorbed to only 60 per cent. This smaller degree of sorption may have been the cause of the unexpectedly low SDN values found for these fractions.

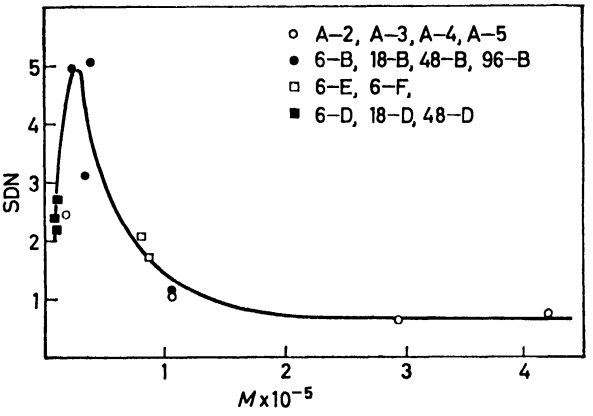


Figure 6. Standard dispersion number *v.* light scattering molecular weight, for some fractions of lignin sulphonate

The mechanism of dispersion was explained by the classical concept of a protective colloid, shown in *Figure 7*. The titanium oxide particles in water possessed a structure-forming tendency which produced aggregation in the slurry. Low molecular weight lignin microgels adsorb on to the particle, promote inter-particle repulsion, break down the structure and thus reduce the viscosity. With lignins of higher molecular weight, the coverage is not as great and thus the dispersion effect is less at a given lignin concentration. When the microgels are very large, inter-particle

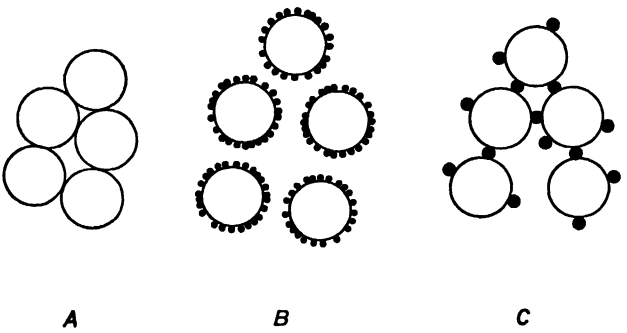


Figure 7. Pictorial illustration of the dispersion of titanium oxide by a lignin microgel. The large empty circles are the titanium oxide particles. The black circles are the lignin macromolecules

A—an aggregate in the undispersed suspension
B—dispersion on adsorption of a low molecular weight lignin
C—failure of a high molecular weight lignin to disperse the aggregate

adsorption occurs and the viscosity of the suspension is increased. Although certain detailed points remained unexplained, it seemed that the results in a general way fitted this simple colloidal theory.

ADHESIVE AND THERMOPLASTIC PROPERTIES

It is not known for certain whether lignin contributes to the strength of wood. Recent papers by Stone[60,61] and Kallmes[62] suggest that the lignin provides only a minor component of the shear strength of birch wood. On the other hand, one would expect that if lignin exists in wood as a three-dimensional network, comprising up to 30 per cent of the material in the cell wall, it must affect markedly the fibre-fibre adhesion in the wood. To answer such questions, more knowledge of bulk properties, such as softening or tack temperatures of solid or solvent-swollen lignins, is required. The results now to be discussed are taken from a recent study[63] of the conditions under which lignin will adhere to itself or to cellulose in the form of sheets of paper.

Several lignins were studied, their common characteristic being that they were insoluble in water. Samples included dioxan-HCl lignins, a Bjorkman lignin[64], periodate lignins and certain water-insoluble residues from soda and sulphite cooks.

The method consisted of sandwiching the powdered lignin between two small pieces of paper. Bond-strength was then measured as the force

required to pull the " sandwich " apart. Some sandwiches were made up dry. Some were made up with lignin and paper, soaking wet. In all cases, however, the sandwiches were dried before testing. Thus bond-strengths reported refer only to lignin and paper dried and equilibrated to ambient humidity at room temperature.

Normally, lignins did not bond to paper when the sandwiches were made up in the dry or wet state. However, a small but measurable bonding was found when spruce periodate lignin was soaked in a weak solution of NaOH, the sandwich made up wet, then dried and tested. As shown in

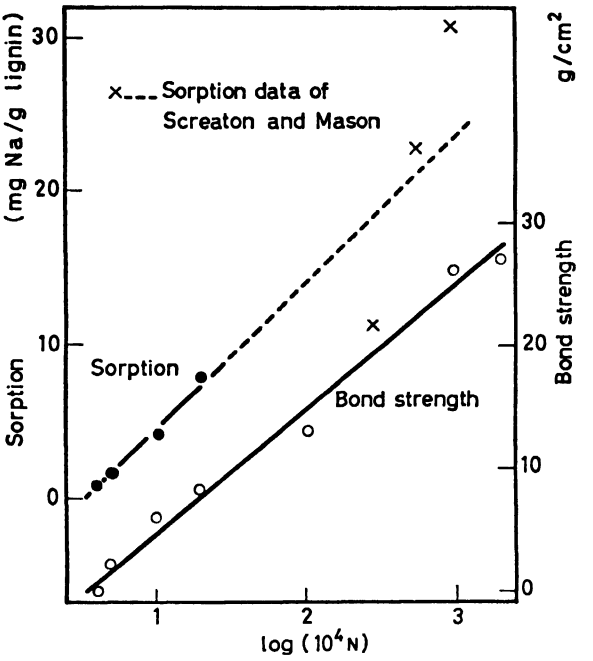


Figure 8. The lower line gives the increase in bond strength with increase in the concentration of alkali used for soaking. The upper line is the sorption of Na^+ on the periodate lignin. The points denoted by crosses are from the sorption data of Screaton and Mason[65]

Figure 8, the bond strength increased with the alkali concentration, as also did the sorption of sodium ion on the lignin.

The alkali induced bonding may probably be attributed to the mildly polyelectrolyte nature of the periodate lignin. As shown by Screaton and Mason[65], the sorbed alkali neutralizes the weakly acidic groups in the periodate lignin, thereby permitting their ionization. The free charges thus produced cause the lignin to swell like a neutralized polyelectrolyte gel. The lignin then develops adhesive properties and bonds when dried, in a manner analogous to the behaviour of other alkali-based adhesives[66]. When similar experiments were tried with insoluble residues from an alkali or sulphite cook, extremely strong bonds were formed, presumably because of the greater swelling tendency of such lignins.

As pointed out in an earlier section, the residual lignin left on the fibres in the various high yield or semi-chemical processes, will probably have acquired groups capable of ionization. This lignin, even though insoluble, will possess the swelling characteristics of a polyelectrolyte gel. It seems likely that under certain conditions, such lignin will contribute appreciably to the bonding characteristics of the fibres in the dry sheet of paper.

A study was then made of the effect of temperature on lignin bonding. The paper–lignin–paper sandwiches were pretreated by pressing at temperatures ranging from 25° to 250°. Pressing was done on dry sandwiches and also on sandwiches made up soaking wet. But, as before, all bond-strength tests were made on the dried sandwiches at room temperatures.

The way in which the temperature of pressing affected the bonding of dioxan lignins is shown in *Figure 9*. Pressing dry caused no bond to be formed until temperatures between 125° and 200° were reached. However, when the sandwich was pressed soaking wet and then dried, a bond was formed at a considerably lower temperature than when the pressing was made on a dry sandwich. Similar results were found for the spruce Björkman lignin, but a complete analysis over a range of temperature, as shown in *Figure 9*, was not possible because of the small size of the sample.

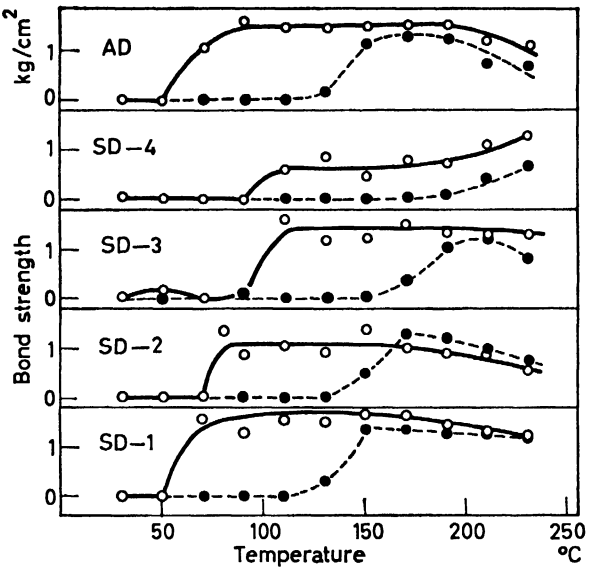


Figure 9. The effect of temperature of pressing on the bond strength of dioxan lignins. Dotted lines and filled circles represent dry pressing while full lines and empty circles correspond to wet pressing. The notation of the lignins is given in *Table 5*

The results for periodate lignins are shown in *Figure 10*. Here the bonding produced by dry pressing occurs at a higher temperature. Also there is a larger decrease of the bonding temperature with wet pressing. Measurements on Wiley milled wood gave results remarkably similar to those obtained for periodate lignin, except that in the case of spruce there

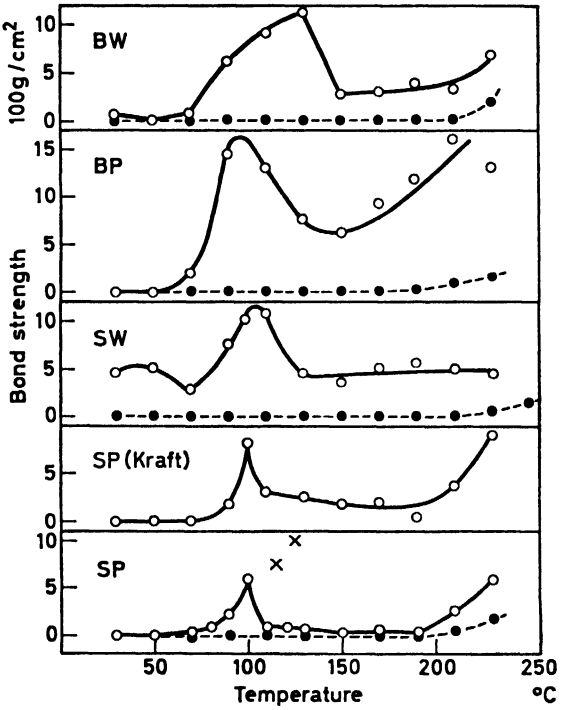


Figure 10. The effect of temperature of pressing on the bond strength for periodate lignins and for woods. Dotted lines and filled circles represent dry pressing while full lines and empty circles correspond to wet pressing. The notation of the lignins is given in *Table 5.* The two points indicated by crosses in the SP graph were for sandwiches pressed under steam pressure at 115° and 124°. The data labelled SP (Kraft) were for spruce periodate lignin sandwiched between 1/2 inch circles cut from a double weight kraft hand sheet

was a constant " base-line " of bonding at lower temperatures, which was probably due to the cellulose component of the milled wood. The maxima found in the bonding of wet-pressed periodate lignin could not be explained unambiguously. It is interesting to note that in this respect, the periodate lignins rather than the dioxan lignins parallel the behaviour of the milled woods.

It was desirable to ascertain the temperature at which bonding began to occur. This was arbitrarily taken to be the temperature at which a bond-strength of 200 g/cm^2 was first reached. Bonding temperatures for dry and wet pressing, T_1 and T_2 are given in *Table 5.* Also included is the sorption of water by the lignin from a saturated solution of sodium chloride at room temperature.

The bonding temperature, as measured in the present work, is analogous to the tack temperature found in polymeric materials. McLaren, Li, Rager and Mark[67] have pointed out that tack temperatures along with brittle points, softening temperatures and second order transition points belong to the category of non-equilibrium properties of high polymers. These effects all arise from the same basic molecular behaviour and are

Table 5. Bonding temperatures, water sorption and β for various lignins

Sample	*Notation*	(T_1) °C	(T_2) °C	(T_1-T_2) °C	*Sorption (mg/g)*	β
Spruce dioxan	SD–1	125	50	75	71	0·51
Spruce dioxan	SD–2	140	70	70	75	0·45
Spruce dioxan	SD–3	165	90	75	84	0·43
Spruce dioxan	SD–4	200	95	105	96	0·53
Aspen dioxan*	AD	130	50	80	86	0·45
Spruce periodate	SP	235	90	145	151	0·46
Spruce wood	SW	255	90	165	—	—
Birch periodate†	BP	235	70	165	119	0·67
Birch wood	BW	320	75	155	—	—

* Kindly supplied by Professor J. M. Pepper.
† Kindly supplied by Mrs P. A. Bradley and Professor C. B. Purves.

associated with increased freedom of molecular movement with increase in temperature. At temperatures below the softening temperature of an amorphous polymer the chains are " frozen " and the bulk polymer is a brittle solid. Above the softening temperature the chains acquire enough energy to move readily with respect to each other. The polymer then becomes soft, deformable and sometimes tacky. In the temperature range in which the change occurs there are usually variations in the rate of change of volume or heat content with temperature, giving rise to the so-called second order transitions[68].

If the polymeric solid absorbs a low molecular weight diluent, the chains are separated by the molecules of the absorbate and therefore require less energy to migrate with respect to each other. Second-order transitions or softening will then occur at a lower temperature. The diluent is said to " plasticize " the polymer[68].

If lignin in wood is made up of chains of phenyl propane units, cross-linked to give an amorphous, three-dimensional structure, it would be expected to undergo second-order transitions with increase in temperature, and thus to show a softening point. The postulate here is that the bonding temperature, T_1, can be regarded as the softening temperature of the lignin. The decrease of T_1 to T_2 on wet pressing is, therefore, a measure of the degree to which the lignin is plasticized by the presence of absorbed water.

An approximate quantitative treatment of the decrease in softening temperature with sorption of water is possible in terms of a relationship proposed by Fujita and Kishimoto[69]. If C is the concentration of diluent in g per g of polymer, the lowering of the transition temperature is given by

$$T_1 - T_2 = C\frac{\beta}{\alpha_2} \tag{13}$$

where α_2 is the difference between the thermal expansion coefficient above and below the transition temperature, and β is a parameter representing the contribution of the diluent to the free volume within the bulk polymer.

Williams, Landel and Ferry[70] have shown α_2 to have a constant "universal" value of $4{\cdot}8 \times 10^{-4}$ deg.$^{-1}$. Substituting for C the sorption of water shown in *Table 5*, the values of β were calculated by means of equation 13.

In spite of fairly marked changes in the water uptake and in $T_1 - T_2$, β in *Table 5* is relatively constant. If the unaccountably high figure for the birch periodate lignin is omitted, the average value of β is $0{\cdot}47 \pm 0{\cdot}03$. Under the conditions used for wet pressing, the uptake of water by the lignin corresponded to 100 per cent relative humidity. The sorption in *Table 5* was measured at 75 per cent relative humidity over saturated brine. Thus the values of β are too high by the ratio of the sorption at 100 per cent relative humidity to that at 75 per cent relative humidity. If an approximate correction is made based on the sorption isotherm for water on periodate lignin published by McKnight and Mason[71], the average β becomes 0·31. With water as diluent, Fujita and Kishimoto found β for poly(vinyl acetate) and poly(methyl acrylate) equal to 0·37 and 0·30, respectively.

A number of workers have previously discussed the thermoplastic behaviour of lignin. Kleinert and Tayenthal[72] have noted softening and melting of alcohol lignin in hot water. Lagergren, Rydholm and Stockman[73] have considered the effect of lignin softening in the defibreing of wood by different methods. The softening and steam plasticizing of lignin has been recognized for some time in the various processes for making board and dimensionally stabilized wood[74,75,76] and Higgins[77] has shown that the compressive strength of various types of veneer is markedly temperature dependent. The significance of lignin softening for mechanical pulping has been pointed out by Luhde[78] and for sulphite pulping by Bjorkman and Person[79]. It seems likely that both in the production of mechanical pulp[78] and in the drying of newsprint, temperatures and moisture conditions are reached which could cause the lignin to soften. Such softening of a major component must have an appreciable effect on the physical behaviour of the wood or the pulp. In a recent study of the fracture of wood, Atack *et al.*[80] have shown that at elevated temperatures the wood breaks as if the lignin has been softened. In chemical pulping, the wood is subjected to elevated temperatures in aqueous media. It is probable that the lignin will exist as a semi-molten phase before it is dissolved. Thus much of the loss of reactivity produced by the so-called " condensation " of lignin at high temperature may be due simply to physical coalescence of the molten lignin microstructure in the wood. Marraccini and Kleinert[81] are currently investigating the closely related phenomenon of the loss of lignin surface in the initial stages of the cooking process.

CONCLUDING REMARKS

This paper has been written around the concept of the lignin network proposed in the first paragraph. It must be admitted that none of the evidence gives an unambiguous answer to the important question of the molecular state of lignin in wood. With regard to the polydispersity of the softwood lignins, it is true that a considerable range of molecular weights would be produced by the degradation of a lignin network, in a fashion analogous to the degradation of a branched polymer such as amylopectin.

It is also true that appreciable polydispersity could arise from the synthesis of a branched network by polyfunctional radicals. such as are believed to be the precursors of lignin wood. In fact, the statistical polymer treatment of Erlander and French[24] was based on the condensation of polyfunctional glucose monomers. The concept was applicable to the acid hydrolysis of amylopectin and glycogen because the degradation reaction was found to be the reverse of a random condensation. Thus, the polydispersity of soluble softwood lignins may arise either from the degradation of a network already formed in the wood or from the induced syntheses of branched type macromolecules from reactive, low molecular weight molecules present in the wood[82]. Pronounced aggregation of low molecular weight units as suggested by Lindberg would also produce polydispersity[83].

The evidence supporting the network theory is more indirect. The universal insolubility of lignin is likely to be due to its extensive branched or crosslinked structure in wood. A network structure of protolignin would explain the well-known difficulty of separation from the carbohydrate moiety. Wardrop[81] has shown that lignification takes place at a comparatively late stage in the development of the cell wall. If the protolignin is laid down as a three-dimensional network, part of this material is likely to grow around individual carbohydrate molecules. The resulting entanglement of molecular chains could make separation difficult or impossible as in the case of the synthetic " snake cage " polymer mixture[85]. Thus it may not be necessary to postulate covalent linkages between lignin and carbohydrate molecules to explain their difficulty of separation[86].

If lignin is markedly polymerized by the usual methods and for delignification, one might expect an increase in the molecular weight of a lignin derivative made soluble by a relatively mild treatment and then re-cooked for a longer time. As yet there has been no demonstration of such an increase. In the case of sulphonation it has been shown that already soluble lignin sulphonates are degraded by further sulphonation[13,87].

Continuing definitive investigations on the physical chemistry of the lignins will eventually support definite conclusions concerning the molecular state of the lignin in the wood.

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