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IMPROVEMENT OF THE TANNING POTENTIAL OF LIGNIN DERIVATIVES BY CHEMICAL MODIFICATIONS

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I INTRODUCTION

The tanning industry of the United States is largely dependent upon imports from foreign markets for its supply of natural tanning materials. Because of the rising costs of these imports and with the possibility of a future critical shortage due to the destruction of domestic chestnut trees by the blight, or, due to import failures, the need for some program to alleviate these conditions is obvious. This research is part of a program directed toward the development of a replacement supply of tanning materials to meet this contingency.

In making a source of tanning materials available one is led to several possible choices. One direction toward this goal lies in the cultivation of sources of natural vegetable tannins in this country and the U. S. Department of Agriculture is, at present, taking an active interest in this development by cultivating canaigre.

The natural tannins are known to contain phenolic groups and this knowledge suggests the use of compounds with phenolic groups as replacement tanning materials. Some such synthetics have been prepared. Turley (52) has described a phenol aldehyde condensate which has received commercial acceptance. However, the greater adaptation of synthetic tanning materials awaits the time when suitable materials become more competitive with natural tanning materials.

Economically, one logically turns to the utilization of

low cost commercial products, which suggests lignin wastes, and these materials are not only abundant in this country, but their tanning potential has been widely studied and definitely demonstrated (28). The utilization of this material will not only prevent any critical shortage of tanning materials, but will also partially reduce the stream pollution problem faced by those firms who must now dispose of such wastes. Lignin wastes, therefore, appear to be a justified raw material for this program from a practical as well as an economic point of view.

II DEVELOPMENT OF THE RESEARCH

A. The Mechanism of Vegetable Tannage

The theories concerning the mechanism of vegetable tannage are many and varied. In the past, there has been a general tendency to classify these theories as "chemical" and "physical". The former necessarily implies a stoichiometric relationship between the tannin and skin substance, while the physical theory requires tannage to be a surface type of phenomenon in which the adsorption isotherms of Freundlich and/or Langmuir are obeyed. As we shall see this distinction in the tanning theory is no longer valid.

An excellent review of the literature concerning the theories of vegetable tannage prior to the last war may be found in the monograph by McLaughlin and Theis (29). A more recent review is that of Shuttleworth and Cunningham (48) who

emphasize the modern Hydrogen Bond Theory of tannage. Tu and Lollar also favor this modern concept and in their recent publication (53) present rather conclusive evidence toward its confirmation. Since that theory is fundamental to the development of this thesis it will be considered in some detail.

The hydrogen bond was first postulated by Moore and Winmill (33). In this type of bond the hydrogen atom serves as the bridging atom between two other atoms. There is a considerable amount of evidence that such a bond exists, but its nature is still not completely understood. It is generally recognized (38,58) that resonance is not a major factor in the hydrogen bond, but that its particular character can be explained by a consideration of the electrostatic forces of the atoms involved. Thus the only atoms that can be bridged by the hydrogen atom are the most electronegative ones: F, O, and N.

Although there is little valid evidence for the contribution of resonance to the hydrogen bond per se, this does not imply that resonating structures do not influence hydrogen bonding. In fact, quite the converse is true. Jenkins (22) has shown a relationship between resonating structures involving hydrogen bonding and the dissociation constants of several carboxylic acids. Gill and Stonehill (15) have shown that many dyes, the color of which has been attributed to resonance, can form hydrogen bonds. Tu and Lollar (53) have

shown that only those hydroxy derivatives of benzhydrol, benzophenone, di- and tri-phenyl-methane, and xanthene which can partake of quinoid resonance are tanning compounds. From a consideration of the resonance structure involved it follows that this quinoid resonance enhances hydrogen bonding and thus the significance of hydrogen bonding and the tanning mechanism is demonstrated.

In the heterogeneous system existing during tannage (i.e. protein, tannins, non-tannins, and solvents) there are many active centers at which it is possible to speculate that reaction between tannin and collagen occurs. The free amino and carboxyl groups, quanidyl groups, hydroxy groups, and the peptide link itself are all possibilities for reaction loci. As Tu and Lollar (53) point out, such heterogeneity does not indicate that vegetable tannage occurs by a single mechanism, but their data clearly indicate that in the case of the phenolic tanning materials studied, hydrogen bonding of the phenolic groups of the tannin to the peptide linkage of the collagen plays a very significant role.

B. <u>The Tannins</u>

The chemical classification of the tannins is complicated by several factors. First, it is not correct to assume that the structure of any of the vegetable tannins is explicitly known. Secondly, the most common method of assaying the tannins (that is by their uptake on hide powder) is a very unsatisfactory method. Nevertheless, several classifications

have evolved.

Early classification of the tannins was based upon the color reactions obtained by treating tannin solutions with iron salts, or based upon the end products obtained by heating the tannins at about 200 degrees C. Since the impurities in the tannins had the same nuclei and thus gave the same end products, and also since the color reactions indicated only free phenolic groups, these classifications were not satisfactory. In 1918 Perkin and Everest (41) classified tannins into three groups:

a. Depsides of gallotannins

b. Diphenylmethylolid or ellagitannins

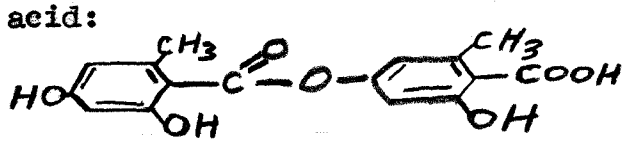
c. Phlobatannin or catechol tannin

However, the most widely accepted classification is that of Freudenberg (13) and has been used by Nierenstein (35) in his book concerning the organic tannins. Under this classification the tannins consist of:

1. Hydrolyzable Tannins

These are composed of benzene nuclei united in a larger complex by ester linkages and are degraded into simple components by the hydrolyzing action of acids or enzymes.

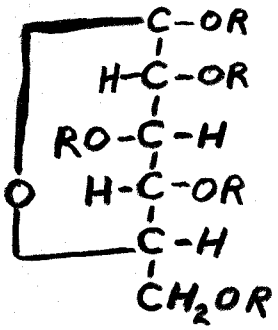
a. Mutual esters of phenolcarboxylic acids or with hydroxy acids (depsides). As an example, consider lecanoic acid:



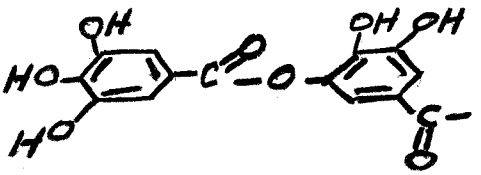
b. Esters of phenolcarboxylic acids with poly-

atomic alcohols and sugars.

c. Glucosides of the gallotannin and ellagitannin types. Fisher (9) established that gallotannin, the natural tannin developed from the depsides, was penta-meta-digalloyl-a-glucose:

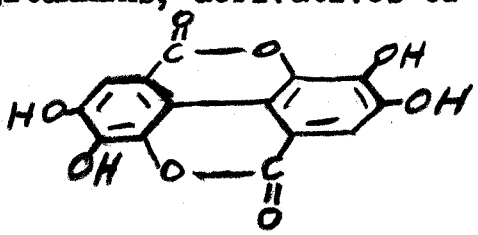


where R is

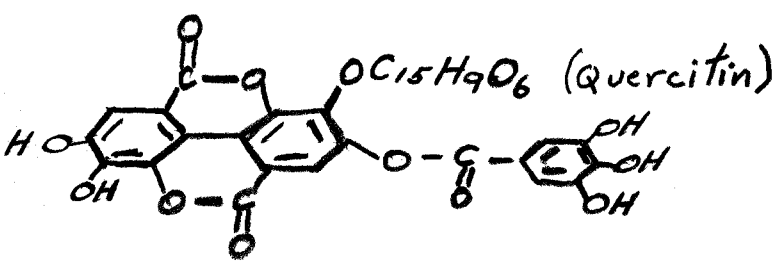


Nierenstein, however, takes exception to this structure. He believes that some of the R groups may be larger than dimers and consequently, not all of the OH groups need be involved in the esterification.

Ellagitannins, derivatives of ellagic acid



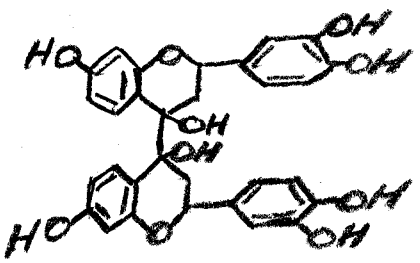
also occur as glucosides and are characterized by the tannin in myrobalan nuts and by valonia. Chestnut tannin, on the other hand, yields ellagic acid as well as gallic acid and quercitin on hydrolysis, but is generally not considered to be a glucoside. It has been indicated by Flinn (10) to be



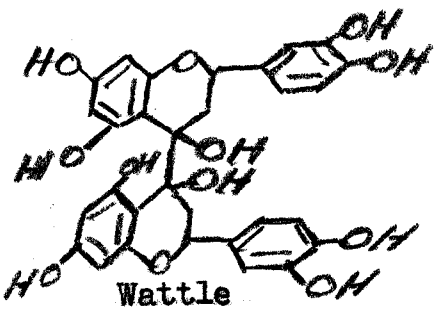
but it is possible that during the purification of the tannin for examination, the glucose portion may have been cleaved and thus lost for identification.

2. Condensed Tannins

These tannins have their nuclei held together by carbon linkages and thus cannot be hydrolyzed by acids or enzymes. This group (commonly called catechol tannins or phlobotannins) makes up the majority of the commercial tannins. Freudenberg (13) believed that the tannins in this class were polymers of 3-hydroxy-polyhydroxy flavan. Russell (46) however, used the chateehin model to develop his theory that the tannins in this class are bis-hydroxyflavopinacols and his synthesis of these materials (46) do in many respects closely approximate the natural tannins. Russell actually claims the synthesis of quebracho, wattle and other tannins.



Quebracho



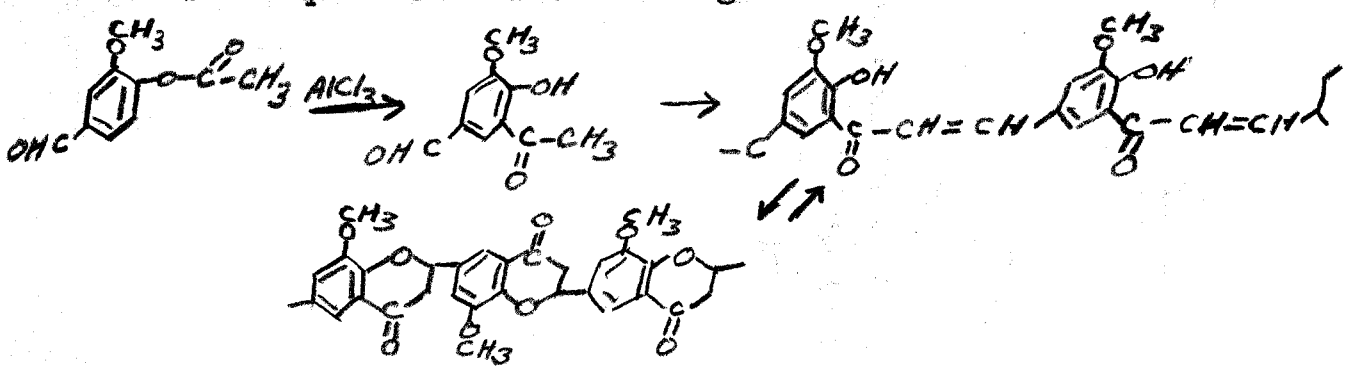
Wattle

There are however, certain diserepancies such as molecular size and the phenomenon of phlobaphene condensation that leaves his synthesis open to question. Further, the recent work of White (57) has shown natural quebracho tannin to contain at least 14 entities distinguishable by paper chromatography and solubility studies. Flinn (10) thinks it

is likely that the condensed tannin molecule is made up of long chain polymers with the flavopinacol or an open ring form as the basic unit.

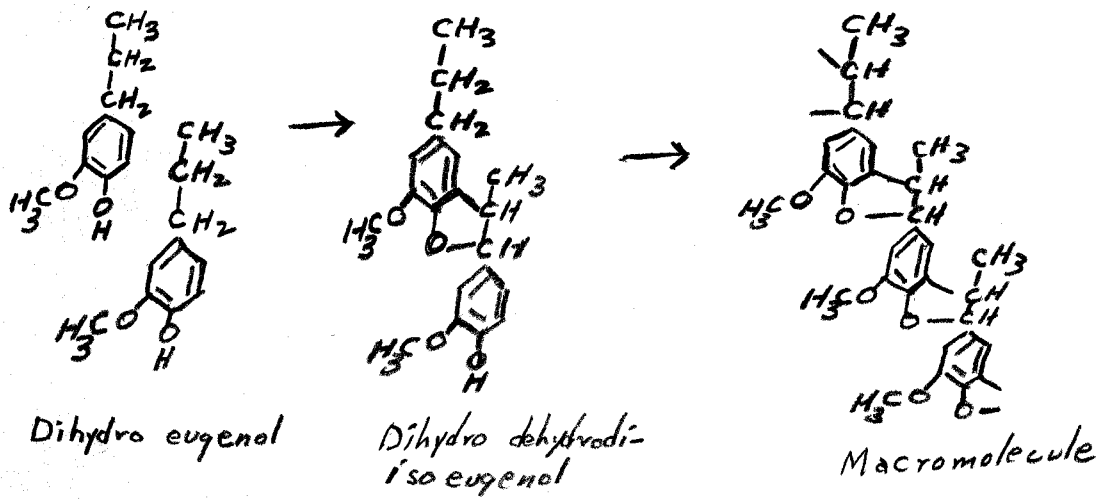
C. The Nature of Lignin Wastes

Despite the fact that the synthesis of gymnosperm lignin has been claimed by Russell (45), his work has been criticized by such lignin experts as Erdtman (7), Brauns (2) and others and the true structure of lignin remains unknown. Russell's synthesis consists of first preparing 2-hydroxy-3-methoxy-5-formylacetophenone from vanillin monoacetate by the Fries rearrangement. This then undergoes repeated intermolecular condensation polymerization (aldolization, loss of water, Claisen condensation) to give the open chain structure which is in equilibrium with the lignin structure:

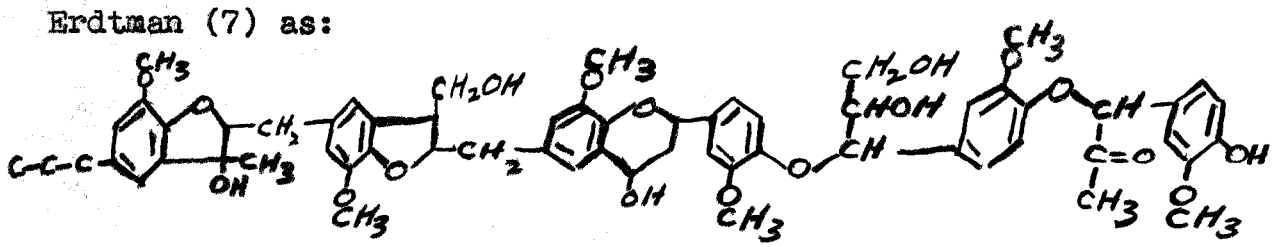


Russell claims that this product is indistinguishable from natural spruce lignin, but Brauns (2) points out that the calculated carbon, hydrogen and methoxyl do not agree well with the values obtained by actual analysis of spruce lignin. Although Freudenberg has not specifically claimed a lignin

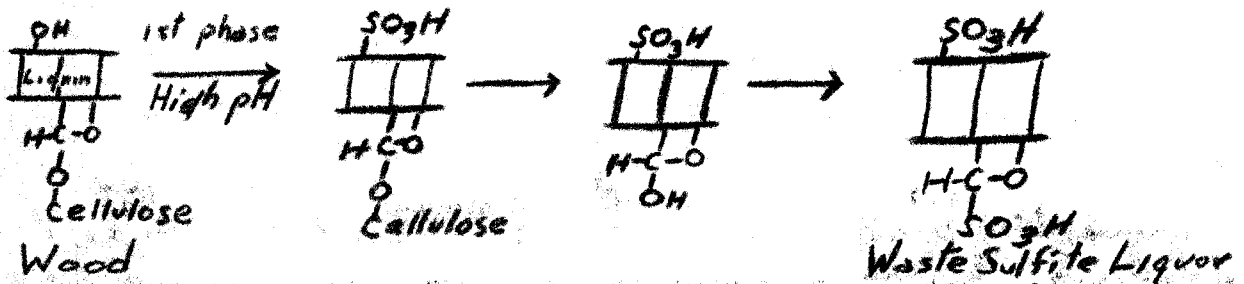
synthesis, he has recently reported (11) the formation of lignin-like products by the action of a dehydrase on coniferyl alcohol. Aulin-Erdtman (1) has also suggested that lignin formation is probably due to such a reaction. This would lend support to Freudenberg's conception of the lignin macromolecule as being formed from dihydro eugenol as follows:



This structure has been further supported by the ultraviolet absorption studies of Aulin-Erdtman (1), but she assigns the Freudenberg formula to only a part of the total structure. A structure for lignin that would appear the explain most of what is known about its chemical properties is given by Holger Erdtman (7) as:



Not only is the structure of lignin in doubt, but so also is its reaction with bisulfite. Erdtman (8) has recently discussed the chemistry of the sulfite pulping process in relation to the sulfonation of lignin. He points out that as sulfonation increased the hydroxyl content decreases and the conclusion is therefore drawn that the sulfonation process is essentially a substitution of hydroxyl groups by sulfonic acid groups. In the first stage of the sulfonation process a solid lignosulfonic acid of very large molecular weight is formed. Upon additional sulfonation under the acid conditions employed in the commercial sulfiting process, the acetal linkages supposedly connecting lignin units to each other or to carbohydrate units are cleaved. This, of course, liberates hydroxy groups which impart solubility to the sulfonated lignin (Kullgren's lignosulfonic acid). Additional sulfonation now occurs with the replacement of the hydroxyl groups by sulfonic acid groups. A summary of these reactions is given by Erdtman as:



Although this mechanism of the sulfiting process has its opponents, it is more or less generally accepted as the best representation of what actually occurs.

D. The Utilization of Lignin as Tannin

Lignin derivatives have long been studied in regard to their tanning properties. One of the earliest applications of lignin to the leather industry is the patent of Mitscherlich (31) in 1878 who pointed out that the waste sulfite liquor from the cooking of oak wood could be used as a tanning agent. Since that time much has been published and the interested reader is referred to the recent article by Johnson and Marshall (23) for a comprehensive review of the subject up to 1948. It is of interest, however, to consider the salient points concerning the theories of tannage as related to the application of lignin derivatives to the tanning industry.

In 1914 Moeller (32) observed that the combination of lignosulfonic acids and hide substance is very stable and that the lignin derivative cannot readily be removed by leaching with water. Although this stable fixation cannot be attributed to the reaction between the sulfonic acid groups of the lignin and the free amino groups of the collagen because a salt linkage of this type would be expected to be ruptured by water, the inactivation of these polar groups, nevertheless, is well established. Evidence for this ionic type of reaction can be found in the work of Gustavson and Larsson (19) who showed the greatly decreased uptake of lignosulfonic acid when the free amino groups of the collagen were inactivated by synthetic tannins of the sulfonic acid type. Gustavson also found (17) that deaminated collagen

fixed less lignosulfonic acid than the normal material. Supporting evidence for this ionic reaction can be found in the recent publication of Lipsitz, Kremen, and Lollar (28) who used the ninhydrin reaction on leather made with a lignosulfonate. Since in the case of the natural tannins the reaction was positive, indicating that free amino groups were present, the inactivation of these groups is inferred by the negative test obtained in the case of the lignosulfonate tanned leather. Thus the reaction between the sulfonic acid groups of the lignin and the hide substance is generally accepted.

This ionic reaction, however, is not the only combination that occurs between lignosulfonic acid and hide substance. It will be recalled from the above that the tanning mechanism is attributed to hydrogen bonding between the phenolic hydroxyl groups of the tannin and the peptide link of the protein. Lipsitz et al. (28) have produced leather from lignosulfonates that have the "feel" and appearance of vegetable tanned leather. The conclusion is drawn that this leathering quality of the lignin derivatives is due not to amino specificity, but "to some other mechanism". If this conclusion and the theory of the tanning mechanism is correct, it follows that the "other mechanism" is, to a great extent, hydrogen bonding at the peptide link. Opposition to this view has very recently been expressed by Gustavson (18). Gustavson refers to the work of

Kremen and Lollar (25) who produced a material with leather characteristics from deaminated calfskin and he argues that "the lignosulfonate fixed by deamino-collagen is adequately accounted for by ionic interaction with the remaining e-amino groups of lysine residues and the quanidyl groups of the arginine residues." Such an arguement, however, does not invalidate the theory maintained by Lollar and his associates that leathering properties (not simply fixation) of lignosulfonates is due to hydrogen bonding at the peptide link. Thus the fact that ✗ leather made from a lignosulfonate does not have an increased shrinkage temperature is attributed to the few available loci for hydrogen bonding to occur.

This low degree of hydrogen bonding potential is evident from a comparison of the natural tannins with the various lignin derivatives. In the former case, an abundance of phenolic hydroxyl groups is evident, while the lignin derivatives have a scarcity of these groups or other hydrogen bonding functions.

In view of this interpretation, it is felt that proper chemical modification of lignin derivatives will lead to an improved product that will overcome the objection of the unmodified material and will meet the minimum requirements necessary for the production of a commercial leather. Obviously, it will be necessary to modify these materials in such a manner that their ability to hydrogen bond at the peptide link will be heightened and their natural affinity for combination with the free amino groups decreased.

D. The Predicted Modifications

As has just been pointed out, the theory of tannage suggests to the chemist a modification of lignin derivatives in such a manner as to increase their hydrogen bonding ability at the peptide link of the skin protein. The various methods of modification will, of course, depend upon the type of lignin derivatives available. Since the use of any such modified material by the leather industry will depend upon the availablility of the raw materials, a program of such modifications must limit the substances to be investigated to commercially available and preferably low cost lignins. In addition, these commercial lignins must also meet the requirement of having a fairly constant composition so that reproducibility of any chemical process will be obtained. Lignin products are now being produced in this country that very nearly meet the stated requirements: for example, a magnesium lignosulfonate which is a by-product of the sulfite process of wood pulping (21) and two alkali lignins, obtained from the soda pulping process (42) are available.

Having established a satisfactory source of raw material we can turn our attention to the specific chemical reactions that will convert lignin wastes to satisfactory tanning materials. On the basis of the specificity of the lignosulfonates for free amino groups of the hide protein and the fact that leathering properties are imparted to the skin by hydrogen bonding at the peptide link, it is not difficult to predict the type of chemical modification best suited to this purpose. There is another factor that must be considered, however, when chemically modifying these materials. The tanning extracts

that are used today are water soluble materials and all tanning procedures and equipment are geared to an aqueous tannage. The lignosulfonic acids are water soluble by virtue of their sulfonic acid groups and so let us first consider these materials with regard to their possible chemical modifications.

A chemical modification that would reduce the amino specificity of the lignosulfonates and subsequently enhance hydrogen bonding ability is that of minimum desulfonation. This would involve only partial removal of the sulfonic acid groups so that water solubility would not be too greatly reduced. Another method of decreasing amino specificity and creating additional hydrogen bonding possibilities would be the condensation of the sulfonic acid groups with phenolic substances to form sulfones. Recent German work (36) on the sulfone bridge type of synthetic tannin supports the view that this type of modification will increase the tanning potential of lignin materials. In addition, the particle size of the lignin units will be increased which is presumed to be advisable. Such a modification, however, will probably result in a water insoluble material and thus it will be necessary to subject the product to a minimum sulfonation to regain water solubility.

Another possible modification of the sulfonic acid group of the lignosulfonates is the conversion to the

sulfonamide through the sulfonyl chloride. Aliphatic sulfonyl chlorides have very recently assumed considerable importance as tanning materials.[3] A lignosulfonyl chloride, however, by virtue of its affinity for amino groups would not be expected to produce a "full" leather of the sole leather type, but it, too, may be useful in the production of other leathers. The lignosulfonamide, on the other hand, has greater possibility for combination at the peptide link and might possibly be of value in heavy leather tanning.

The modifications of the lignosulfonates just discussed are unique in that they concern the sulfonic acid portion of the molecule. Other types of modification are, of course, possible and should be included in any program designed to improve the tanning potential of the lignin wastes. The removal of methoxyl groups in the lignin material, for example, would increase the phenolic hydroxyl content. Costwise, this operation might be difficult to justify and other effects on the lignin molecule may be too drastic. However, it would seem wise to consider this possibility. The lignosulfonic acids as well as the alkali lignins contain several reactive alcoholic groups whose chemical reactivity permit combination with carboxylic and sulfonic acid chlorides to form esters. By proper selection of the acid chloride the phenolic hydroxyl groups of the lignin can readily be increased. Lignin ethers can be prepared to give similar results. Lignin derivatives can be readily nitrated and here, too, the degree of hydroxylation might be increased through diazotization of the reduced nitro

derivative.

A disadvantage inherent to the alkali lignins is their water insolubility. Because of the comparatively large size of the lignin molecule it would appear reasonable to assume that the discussed derivatives of the alkali lignins are also water insoluble. Even though solvent tannage has been successfully accomplished in the laboratory (44), the tanner is not yet prepared for such a revolutionary change from his present methods. Water solubility could no doubt be imparted to these derivatives by a minimum sulfonation procedure, and although this process would involve an additional cost factor, the product may still meet the economic requirements of a replacement tannin. From this viewpoint the lignosulfonic acids may offer a distinct advantage.

F. Plan of the Research

The chemical modifications just discussed are many and varied and will require considerable time for their complete evaluation. The plan and purpose of this program is fourfold, viz:

1. To obtain a preliminary evaluation of some of these predicted modifications of the lignin derivatives. This was accomplished by preparing the derivatives and subjecting them to the evaluation by tanning calfskin squares with them. The tanned squares were then examined with regard to "feel", shrinkage temperature, and general appearance.

2. To select those derivatives with the greatest promise and further study them by means of a calfskin piece

tannage. This permitted a more complete analysis of their tanning potential by means of leather analysis, microscopic examination and resistance to proteolytic enzymes.

3. To obtain fundamental chemical information concerning the most promising derivatives. This included elemental analysis where required (i.e. sulfur, magnesium, etc.) and functional group analysis (i.e. methoxy and hydroxy).

4. To correlate the information in 2 and 3 with the postulated theory of the tanning mechanism.

III EXPERIMENTAL

A. Preparation of the Derivatives

Three types of commercial lignin materials were used in the following synthesis. These consist of:

1. Meadol--- a purified hardwood lignin
2. Indulin-- a purified alkali softwood lignin
3. Marathon Tan A-- a magnesium lignosulfonate resulting from the sulfite process of wood cooking.

These names will be used as an aid in classifying the derivatives prepared.

All of the derivatives to be described were evaluated in regard to their tanning potential. The data for this evaluation will be found in Tables I-A, I-B, and I-C.

1. Meadol Derivatives

a. Meadol-poly-2,4 dihydroxybenzoate

Lignin esters have been described by Lewis, Brauns et al. (26). Their method of preparation was to react an aliphatic acid chloride with the hydroxyl groups of the Meadol in the presence of pyridine. To employ this method for the synthesis of a polyhydroxy aromatic acid ester of a lignin material would be most impractical because it is doubtful that the acid chloride of 2,4 dihydroxy benzoic acid (beta-resorcylic acid) could successfully be prepared. Therefore a method of direct esterification was used. This consisted of refluxing the lignin product with the acid in dioxane solution in the

bulb portion of a Soxhlet extractor and shifting the equilibrium by allowing the condensed dioxane vapors containing the water of reaction (B. P. dioxane 101° C.) to pass through anhydrous sodium sulfate placed in the extractor cup.

20 g. of Meadol were dissolved in 200 ml. of dioxane, 14.7 g. of beta-resorcylic acid and 5 ml. of concentrated hydrochloric acid added. The mixture was refluxed in the bulb portion of a Soxhlet extractor for 16 hours allowing the condensate to pass through the cup which was packed with anhydrous sodium sulfate. After cooling, the solution was slowly poured into ether while stirring vigorously and the precipitate filtered, washed with ether, and dried in vacuum over sulfuric acid. Yield of product: 20 g.

b. Demethylated Meadol

Moore, Wright and Hibbert (33) describe a procedure for the demethylation of a spruce lignin. The method was applied to Meadol and its details follow:

20 g. of air dry Meadol was dissolved in 1600 ml. of glacial acetic acid, 200 ml. of 57 per cent hydriodic acid (Sp. G. 1.7) added and the mixture refluxed for two hours in an atmosphere of carbon dioxide. After concentration of the solution to about 200 ml. it was poured into 6 liters of 0.1 N sodium bisulfite solution. This decomposed the excess hydriodic acid and caused the lignin material to be precipitated. The

precipitated material was separated by centrifugation, washed with water, and dried in air.

The dried material was then extracted with ether to remove any tar-like products and the tar free derivative dissolved in glacial acetic acid and refluxed in the presence of zinc dust to remove any iodine in the lignin material. After filtering the residual zinc from the solution, it was poured (after first concentrating it to 200 ml.) into 2 liters of water. The precipitate was centrifuged, washed with water and dried over sulfuric acid in a vacuum desiccator. 6 g. of a dark brown powder was obtained.

c. Hydrolysis Product of Chlorinated Meadol

Aromatic chlorine compounds can be hydrolyzed to phenols and thus a hydrolysis product of a chlorinated lignin might have an increased number of phenolic groups. To prepare such a product, Meadol was suspended in water at 80° C. and chlorine passed through. The chlorinated product was then subjected to hydrolysis by refluxing with alkali. After cooling, the modified lignin was recovered by pouring the solution into acid, filtering the precipitate, washing and drying in air.

d. Alkaline Hydrolysis Product of Meadol

Peniston and McCarthy (40) have claimed the conversion of methoxyl groups in spruce lignin to phenolic hydroxyl groups by a mild alkaline hydrolysis. Meadol was therefore subjected to a 10 per cent sodium hydroxide solution and refluxed

for about four hours. The solution was dropped into a hydrochloric acid solution, the precipitate filtered off, washed with water and dried in air.

e. Condensation Product with Ethanolamine

Reid, Dryden, and Aronovsky (43) show that monoethanolamine condenses with corncob lignin and the condensate can be remethylated. They conclude therefore, that the condensate has a greater number of hydroxyl groups present. Although it is believed that the interpretation of their experiment is open to some question, this reaction was employed in this research with alkali lignin by adding monoethanolamine to a dioxane solution of Meadol and the condensed product filtered off, washed with water and dried in air.

f. Resorcinol-Meadol Condensate

100 g. of Meadol (dried in vacuum over sulfuric acid) and 300 g. of Resorcinol were intimately mixed and placed in a two necked flask fitted with a water cooled reflux condenser and a thermometer. 100 ml. of ethyl ether was added and then 10 ml. of a concentrated hydrochloric acid. After refluxing for three hours, the hot resinous mass was poured into vigorously stirred water.

The brown precipitate was allowed to settle, washed with water several times by decantation, filtered, washed repeatedly with water and dried in vacuum. 10 g. of vacuum dried material was obtained.

2. Indulin Derivatives

a. Indulin-poly-2,4 dihydroxy benzoate

20 g. of Indulin was treated exactly as was Meadol. A yield of 24 g. of air dried product was obtained.

b. Demethylated Indulin

The procedure for demethylation was essentially that described for Meadol. A sodium fusion of the material before deiodination and a subsequent test for iodide with silver nitrate was negative. There was no need therefore for the deiodination procedure. 19 g. of demethylated Indulin was obtained from 20 g. of the original lignin.

c. Aluminum Chloride treated Indulin

It is well known that anhydrous aluminum chloride is a good demethylating agent (20). It was hoped that it could be made to convert the aromatic methoxy groups of lignin to phenolic hydroxyl groups. Indulin was suspended in nitrobenzene, aluminum chloride added and the mixture refluxed for an hour. The modified lignin was then filtered off, washed with ether and air dried.

d. Phosphoric Acid treated Indulin

100 per cent phosphoric acid is reported to convert aromatic ethers to phenols (51). Approximately pure phosphoric

acid was prepared by the addition of phosphorous pentoxide to 85 per cent phosphoric acid until the specific gravity of the pure acid was obtained. A check by titration indicated a purity of 98.6 per cent. When Indulin was suspended in this acid and heated, considerable frothing occurred. The lignin material was recovered by pouring the mixture into water, filtering off the product, washing with water and drying in air.

e. Rearrangement of Ether structure of Lignin to Phenolic Hydroxyl

Alkyl-phenyl ethers undergo rearrangement in the presence of certain reagents to form substituted phenols (6). Since lignin is said to consist of a phenyl-propyl-ether type structure (12) it is conceivable that certain reagents might produce a lignin derivative with a greater phenolic content. A mixture of glacial acetic and sulfuric acids is an effective agent for this rearrangement and it was therefore employed with Indulin by adding the lignin to the acid mixture and heating. The gummy precipitate formed was separated by pouring into water, filtering, washing with water and drying.

f. Hydrolyzed Chlorinated Indulin

Indulin was chlorinated and then hydrolyzed exactly as was Meadol. The product obtained was recovered and dried in

the manner previously employed.

g. Condensation with Phenol

5 g. of Indulin was dissolved in 60 g. of Phenol and 12 ml. ethyl ether and 1 ml. concentrated hydrochloric acid added. After refluxing for one hour, part of the excess phenol was removed by vacuum distillation and the rest separated by dissolving the reaction mixture in 150 ml. of dioxane and adding that solution dropwise into 1500 ml. of water. The precipitate was filtered off, washed with ethyl ether, petroleum ether, and dried in air to yield 7 g. of material.

h. Condensation with Resorcinol

Indulin was condensed with resorcinol in exactly the same manner described for Meadol. The resulting product had characteristics similar to the other resorcinol-lignins.

i. Alkaline Hydrolysis Product of Indulin

Indulin was hydrolyzed with 10 per cent sodium hydroxide exactly as was Meadol. The resulting solution was adjusted to proper pH (5.0) and used for the preliminary tannage.

3. Derivatives of Magnesium Lignosulfonate

a. Alkaline Hydrolysis Product

As already noted, Peniston and McCarthy (40) have shown

that mild alkaline hydrolysis of a spruce lignosulfonate results in demethylation with an increase in phenolic groups. Magnesium lignosulfonate was therefore subjected to alkaline hydrolysis in the manner previously described for the alkali lignins.

b. Hydriodic Acid treated Product

An attempt was made to demethylate magnesium lignosulfonate in the manner used for the alkali lignins. It was observed that although the lignosulfonate was soluble in 57 per cent hydriodic acid, coagulation occurred upon heating and the product became very difficult to handle. For this reason an aqueous medium was used. The precipitate formed after refluxing the lignosulfonate with hydriodic acid was separated by centrifugation, treated with a solution of sodium bisulfite to remove iodine and then washed with water, filtered, washed again and air dried. When an aqueous suspension of the material was tested with silver nitrate no precipitate was obtained.

c. Magnesium Lignosulfonate-Resorcinol Condensate

100 g. of dry magnesium lignosulfonate was intimately mixed with 100 g. of Resorcinol, 100 ml. of ethyl ether and 10 ml. of concentrated hydrochloric acid were added and the mixture refluxed at 120-160° C. for two and one-half hours.

While still molten, the plastic mass was dropped into water, the precipitate filtered off, washed thoroughly with water and dried in air to yield 80 g. of material.

B. Preliminary Evaluation of Tanning Potential

All of the lignin derivatives prepared were subjected to a preliminary evaluation of their tanning ability by means of tannage of calfskin squares. Since most of the products obtained were insoluble in water, it was necessary, in these instances to employ a solvent system of tannage. Tu and Lollar (53) have shown that methanol is the most satisfactory solvent and it was therefore selected for use. The procedure for tannage was essentially that used by Tu and Lollar (53) and consisted of immersing several acetone-dehydrated calfskin squares (prepared as described by Lipsitz, et al. (27)) in a 20-40 per cent methanol solution of the lignin derivative. After penetration had occurred (16-24 hours), the squares were placed in water for several hours to permit fixation to occur and then the pieces allowed to air dry in the dark overnight or longer.

Where a water soluble material was evaluated, a single bath tannage was conducted in an aqueous system, the pH being gradually reduced from 5.0 to 3.0 before removal from the tanning liquors.

The dried squares were then examined visually for "feel" and appearance and a shrinkage temperature determination also

made on the Theis Shrinkage Meter. The results will be found in Table 1-A, I-B, and I-C.

C. The Promising Derivatives and the Problem of Water Solubility

It is readily observed from Table I that five of the lignin derivatives produce leather with desired characteristics and deserve further study. These are:

(1) demethylated Meadol

(2) demethylated Indulin

(3) the resorcinol-Meadol condensate

(4) the resorcinol-Indulin condensate

(5) the condensate of resorcinol with magnesium lignosulfonate

All of these products were, therefore, prepared on a larger scale, but under essentially the same conditions as previously described.

None of these materials, however, is soluble in water. Under the present commercial tanning conditions it is essential to the successful development of any commercial tannin that the substance be soluble or at least dispersable in aqueous system. Since the cheapest available source of lignin is the magnesium lignosulfonate, its derivative was chosen as the most logical material for solubilization studies.

A common method of imparting water solubility is by sulfonation. One method of sulfonation is by reaction with bisulfite and when successful, is simply carried out. A preliminary experiment in which the magnesium lignosulfonate condensate was refluxed with a sodium meta-bisulfite solution indicated that a water soluble product resulted.

In order to determine the optimum concentration of bisulfite required a series of reactions at varying bisulfite concentrations was allowed to proceed, and after cooling, calfskin squares immersed in the resulting solution and the shrinkage temperature of the tanned squares determined. The experimental data will be found in Table II.

It is evident from Table II that bisulfiting at the 5 per cent level is most effective. Subsequent bisulfiting of the insoluble condensate was carried out at this concentr - ation and the tanning potential of the soluble derivative evaluated.

D. Complete Evaluation of Tanning Potential

In addition to the five lignin derivatives tabulated above, the water soluble product was included in this more complete evaluation. For this evaluation of these promising derivatives, pieces of acetone-dehydrated calfskin (about 3" by 5") were subjected to tannage. The water insoluble materials were employed in a manner identical to that used for the calfskin square tannage, the skin to tannin ratio being 2:1

and a volume to skin ratio of 10:1. In the case of the water soluble derivative, a single bath tannage at pH 5.0 was used. The skin to tannin ratio was also 1:1, but the solution to skin ratio was 20:1.

After tannage, the pieces were bleached, fatliquored and dried in the manner described in a previous publication (28).

The leathers thus produced were then examined with regard to the usual criteria of leather quality. This consisted of:

1. Visual observation for color and general leather character

2. A modified A.L.C.A. leather analysis. This consisted of determining hide substance, grease, and ash in the usual manner and determining water solubles by means of Wilson-Kern washing with water.

3. Microscopic examination. Histological examination of cross-sections was made for tannin and fatliquor penetration. In addition, the cross-sections were subjected to the ninhydrin test as described by Lipsitz et al. (28).

4. A shrinkage temperature determination on the Theis shrinkage meter.

5. A determination of the leathers' resistance to proteolytic attack and a leather made from magnesium lignosulfonate alone was also evaluated for comparison. The method employed is that recently developed by Undeutsch (54). Essentially it is conducted in the following manner:

About 0.5 g. of finely ground leather sample is weighed into two bottles. Into one bottle 25 ml. of a 0.2 per cent solution of trypsin in 0.1 N sodium bicarbonate is added and the same amount of a similar tryptic enzyme solution which has been inactivated by boiling introduced into the second bottle. Both bottles are then agitated at 104° F. for 18 hours. After the digestion has occurred the solutions are made up to 250 ml. in volumetric flasks, filtered with suction, the filtration being aided with "Celite". 50 ml. of the filtrate are then pipetted into tared dishes for solids determination. The difference in solids between the control and experimental sample divided by the dry sample weight represents the degree of tryptic hydrolysis.

The results of this evaluation together with the data obtained from the other indicated studies will be found in Table III which also lists the properties of leather made from an unmodified magnesium lignosulfonate.

E. Fundamental Studies of the Modified Lignins

1. Determination of Homo-/or Heterogeneity

Accurate interpretation of analytical results depend upon the use of samples representative of a homogeneous entity. It was therefore necessary to determine the homo- or heterogeneity of the lignin derivatives and since the resorcinol-condensates appeared to be the most promising derivatives, those materials were subjected to ascending,

one dimension paper chromatography. Two solvent mixtures for development of the chromatograms were used: (1) the butyl alcohol-acetic acid-water mixture (4:1:5) used by Partridge (39) and also by White (57) who separated quebracho tannin into many components with this mixture and (2) a 3:1 mixture of ethyl acetate and methyl alcohol used by Grassman (16) in his work on vegetable and synthetic tannins. Strips (2.5 by 30 cm.) were cut from Whatman #2 paper which was extracted with methanol to remove a fluorescent impurity and spotted 2.5 cm. from one end with 0.5 ml. of solution containing 10-15 micrograms of solids. The strips were then suspended in the solvent mixtures in a closed container (a covered battery jar was used) and the developing liquid allowed to rise. When the advancing front had travelled two-thirds of the length of the strip, the strips were removed, air dried and examined under ultraviolet light.

Table IV indicates the results obtained and it is evident from the two bands obtained in the case of the magnesium-lignosulfonate-resorcinol condensate, that it is composed of at least two components.

2. Separation of the Components in the Magnesium Lignosulfonate-Resorcinol Condensate

Preliminary experiments with a chromatographic column of calcium carbonate indicated also that two substances were present. Both materials were methanol soluble, but one

fluoresced with a yellow color when its methanol solution was placed under ultraviolet light while the other substance gave a blue-white fluorescence. Neither calcium carbonate, alumina, nor hide powder produced a completely satisfactory chromatographic column, but the hide powder column indicated that the use of hide powder in a batch treatment might preferentially adsorb one of the components. It should then be possible to leach off the fraction adsorbed on the hide powder with a 50-50 water-acetone mixture which Merrill (30) has shown to be very effective in removing many tannins from hide powder. Details of the method employed follow:

312 g. of a grease extracted, methanol washed hide powder was wetted with water and allowed to stand for 30 minutes. After filtering with suction the hide powder was added to 5 liters of methanol containing 18.7 g. of the magnesium-lignosulfonate-resorcinol condensate and the mixture was stirred vigorously for one hour. The tanned hide powder was washed once with methanol and allowed to air dry overnight. The filtrates were combined for further treatment.

The air dry tanned hide powder was then extracted by repeated shakings with four 2 liter portions of 50-50 acetone-water mixture. Although all of the material was not removed, the fourth extraction removed very little material. These extracts were then concentrated to yield 6.4 g. of material

(designated 106-T) which represents 34.2 per cent of the amount taken (18.7 g.) When dissolved in methanol the material exhibits a blue-white fluorescence under ultraviolet light.

The filtrate and washing which contain the material not adsorbed by the hide powder were combined and concentrated to yield 3.9 g. of solids which is 20.9 per cent of the total amount taken (18.7 g.). This material when dissolved in methanol exhibited a yellow fluorescence under ultraviolet light. Each of these fractions were again chromatographed and in each case a single band was obtained. Thus it appears that a separation was achieved.

3. Chemical Analysis of the Derivatives and Fractions

In order to have a better knowledge of the composition of the modified lignins it was necessary to subject them to elemental and functional group analysis. Since the structure of the lignins employed is unknown and the fundamental unit quite large (about 840) little value could be obtained from the usual carbon-hydrogen analysis. Of much greater interest are such functional groups as methoxyl and hydroxyl and the elements of sulfur and magnesium in the case of the magnesium lignosulfonate derivatives. The methods employed for these analyses follow:

a. Methoxyl:

The standard method (T2M-43) of the Technical Association of the Pulp and Paper Industry was used (50). Essentially it is a modified method of Viebock and Schwappach (55).

b. Hydroxyl:

The method used was that described by Siggia (49) and taken from the publication of Ogg, Porter and Willits (37). It consists of acetylating the material with a known amount of acetic anhydride and determining the excess by titration.

c. Sulfur

The method of Salvesan and Hogan (47) was used for sulfur analysis. It is a wet oxidation method (perchloric-nitric acids) with subsequent precipitation of barium sulfate.

d. Magnesium:

Magnesium was determined in the solution obtained by the perchloric-nitric acid oxidation. 8-hydroxyquinoline was used as described by Kolthoff and Sandell (24).
The results of these analysis will be found in Table V.

IV DISSCUSSION

It is interesting to note in Table I-A and I-B that the lignin-poly-2,4 dihydroxy benzoates produced leather samples that had good appearance, but no increased shrinkage temperature. At first this might appear contradictory to the hydrogen bonding theory of the tannin mechanism since phenolic hydroxyl groups have been added to the lignins. Tu and Lollar (53) have shown that tanning properties cannot be expected from meta substituted phenolic hydroxyl groups nor can they be expected from aromatic hydroxy carboxylic esters. The carbonyl group of the ester linkage limits to a large degree the quinoid resonance in the resorcinol ring and therefore the hydrogen atoms of the hydroxyl groups have little tendency to hydrogen bond. This interpretation might be questioned on the grounds that hydrolyzable tannins are aromatic hydroxy carboxylic esters and that they do convert hide protein into leather. But the additional fact remains that Lipsitz et al. (27) have recently shown that chestnut tannin, a representative of the hydrolyzable tannins, does not make a leather that resists proteolytic attack. This raises the interesting question as to whether or not other tannins in this class exhibit the same characteristics. It is true that this class of tannins produces leather with an elevated shrinkage temperature (although chestnut tanned

leather has, in general, a lower shrinkage temperature than leather made from the condensed tannins type; viz. quebracho and wattle) and explanation for the tanning ability of chestnut can be found in the quercitin part of the chestnut tannin, which structure is postulated by Flinn (10). This poly-hydroxy-flavanone structure contains phenolic hydroxy groups which can take part in quinoid resonance and are therefore loci of hydrogen bonding. Thus the hydrogen bond theory adequately accounts for the facts observed.

A mild alkaline hydrolysis of the lignins might be expected to produce suitable tanning materials since, as already noted, Peniston and McCarthy (40) have shown that such treatment increases the phenolic hydroxyl content and decreases the sulfonic acid character of the lignin. Unfortunately, this treatment also results in considerable breakdown of the lignin material. It is well known that particle size of the tanning material is an important factor in affecting a tannage. Very probably, the failure to obtain a satisfactory tanning material from an alkaline hydrolysis modification is the result of such extreme lignin breakdown.

The fact that some of the other modified lignins failed to yield the desired improvement in tanning ability must be attributed to the possibility that reaction occurred other than was expected. Since the structure of the lignin materials

is not known and since its chemical properties are not completely understood, unexpected and unexplainable reactions are quite possible. Nevertheless, the lignin derivatives which were promising in the preliminary evaluation and which were selected for complete evaluation did fulfill the predictions made in regard to the improvement of tanning potential.

Table III indicates the data obtained from the complete evaluation of the modified lignins. It is evident that the demethylated lignins produced leather with a poor general appearance, but which did have an elevated shrinkage temperature and was considerably more resistant to tryptic attack than the leather made from the unmodified lignosulfonate. The leathers made from the resorcinol condensate derivatives are, in general, excellent in regard to visual observation and "feel" characteristics. They, too, have an elevated shrinkage temperature and are quite resistant to tryptic attack, having the same range of hydrolysis as vegetable tanned leathers (54).

The yield obtained and the degree of tannage was, in all cases somewhat low which is a reflection of the high hide substance value. Unfortunately the results of the ninhydrin reaction are not as definite as one would like. Only in the case of the bisulfited-resorcinol-condensate was there a decided blue coloration. In the other instances, the leather

cross-sections were so dark that no blue coloration could be seen. There was, however, a decided further darkening of these cross-sections when treated with the ninhydrin reagent which would indicate a positive reaction. The interpretation of the positive reaction is that inactivation of the free amino groups of the hide protein has not occurred and therefore the tanning mechanism does not involve reaction at these loci. The significant data obtained in the test is that the bisulfited-resorcinol condensate produced a positive reaction in contrast to the unmodified magnesium lignosulfonate tannage. Since the bisulfited-resorcinol condensate is the result of increasing the hydrogen bonding loci of the magnesium lignosulfonate this is additional evidence that the basic contention of the tanning mechanism (i.e. hydrogen bonding) is of considerable significance.

Table V indicates the analytical data obtained from the modified lignin derivatives. The analysis of the demethylated lignin derivatives (Table V-A) indicates that the hydriodic acid treatment reduced the methoxy content about 12 per cent. Although only a small increase in hydroxy content is evident it must be remembered that treatment with hydriodic acid destroys the aliphatic hydroxyl groups and thus the per cent hydroxyl represents phenolic hydroxyl. In addition to destruction of the aliphatic hydroxyl groups, considerable degradation of the lignin probably occurred since the lignin

structure is reported to contain ether linkages in its fundamental unit (21). This breakdown is reflected in the rather poor leather quality obtained from tannage with these materials. Thus, the only conclusion warranted is that the phenolic hydroxyl content has been considerably increased with a subsequent increase in shrinkage temperature and resistance to tryptic attack of leather made from such modified lignins.

The analysis of the Meadol-resorcinol condensate is compared with that of Meadol in Table V-B. If one assumes that the literature is correct and Meadol has a unit weight of 860 and that this unit contains 6 methoxy groups (26), some interesting calculations can be made. It follows from the nature of the modification that the number of methoxy groups was not affected and therefore, the unit weight of the Meadol-resorcinol condensate can be calculated thus:

$$\frac{6 \times 31 \times 100}{\text{Unit Wt.}} = \text{per cent methoxy} = 14.5$$

$$\text{Unit wt.} = 1285$$

From this unit weight, the number of hydroxyl groups present in the Meadol derivative can be obtained.

$$\frac{\text{No. of OH groups} \times 17 \times 100}{1285} = \text{per cent OH} = 17.0$$

$$\text{No. of OH groups} = 12.9 \text{ or } 13$$

Therefore, since five hydroxyl groups are present in the unmodified Meadol, eight hydroxyl groups must have been added

by the reaction.

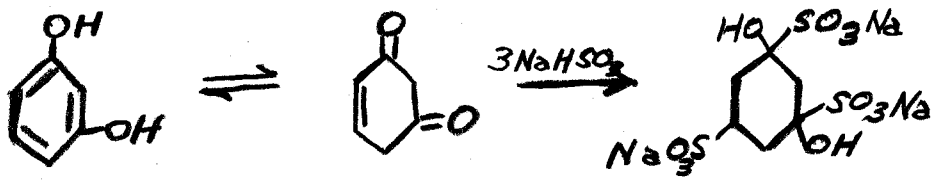
As a further check on this conclusion it is found that the unit weight of the Meadol derivative is 1300 if four resorcinol molecules have been added per 860 unit: [860+(4 x 110)] = 1300. This is a good check on the value calculated from the analytical data. Thus it appears that the reaction between Meadol and resorcinol involves the addition of four resorcinol molecules per lignin unit of 860 and the data further suggests that the reaction does not involve hydroxyl groups.

Similar reasoning from the analytical data for the Indulin resorcinol condensate (Table V-C) indicates that six hydroxy groups have been added per Indulin unit of 840. The unit weight calculated from the analytical data is 1410 which is in agreement with the value obtained (1390) on the assumption that five resorcinol molecules combine with Indulin. Thus it would appear that the reaction between Indulin and resorcinol involves the addition of five resorcinol molecules per lignin unit of 840 and that four hydroxyl groups are involved.

The interpretation of the analytical data obtained in the case of the magnesium lignosulfonate resorcinol condensate (Table V-D) is limited because the material is not homogeneous. The two fractions that have been obtained are decidedly different, but no other conclusions about them can be made.

Of considerable interest however, are the hydroxyl to methoxyl ratios of the water insoluble and bisulfited derivatives. Again making the logical assumption that the

methoxyl content should not change with bisulfiting it is at once apparent from the ratios that the hydroxyl groups were not affected by the sulfiting reaction since, if they had been, a decrease in the ratio would have been noted. Bucherer (4) and Fuchs and Elsner (14) found that resorcinol can be sulfonated with bisulfite and postulated that the reaction would involve the tautomeric enol form as follows:



The resulting product has one sulfonic acid group firmly attached to the ring and the other two are bisulfite addition compounds of carbonyl groups. Wise (58) points out the fact that the ratio of the firmly bound to the loosely bound sulfonic acids is not comparable to that indicated by the above equation and concludes that this is strong evidence against this concept of the reaction between bisulfite and lignin. But, even if this reaction were to occur at the condensed resorcinol groups it is true that the hydroxyl to methoxyl ratio would not change, but the resulting compound would not be a tanning material because the aromatic character of the hydroxyl groups no longer exists. Since the bisulfited derivative has good tanning ability, the

original deduction that the hydroxyl groups are not involved in the bisulfiting reaction would appear to hold.

As a general conclusion, it is evident that this program for the improvement of the tanning potential of lignin derivatives is well founded. It is obvious, however, that additional research must be carried out to make any process toward this goal economically sound. It is felt that the next logical step should be directed toward this end.

V SUMMARY

The hydrogen bond theory of tannage suggests that modified lignin derivatives are a potential source of tanning materials. Three types of commercially available lignins have been subjected to chemical modifications designed to increase their hydrogen bonding ability. The derivatives thus obtained were given a preliminary evaluation by means of a calfskin square, solvent tannage and the most promising derivatives selected for further study.

In addition to the water insoluble promising derivatives, viz:

a. a demethylated alkali hardwood lignin

b. a demethylated alkali softwood lignin

c. a condensate of resorcinol with an alkali hardwood lignin

d. a condensate of resorcinol with an alkali softwood lignin

e. a condensate of resorcinol with a magnesium lignosulfonate,

a water soluble derivative was also prepared by bisulfiting the resorcinol-magnesium lignosulfonate condensate. These six lignin derivatives were used to tan pieces of calfskin and the resulting leathers given a complete evaluation in

regard to shrinkage temperature, general leather "feel" and appearance, chemical and microscopic analysis, and resistance to proteolytic enzymes.

In addition to the above evaluation, fundamental information concerning the derivatives themselves was obtained. This consisted of elemental analysis (Mg and S where necessary) and functional group analysis (OCH_3 and OH). A study was also made of the homo- or heterogeneity of the compounds by paper chromatography and where a separation of the components was made, each fraction was also subjected to chemical analysis.

The data obtained in this investigation permit the following conclusions to be made:

1. As predicted, increasing the phenolic hydroxyl content of lignin derivatives converts them to tanning materials. This is additional evidence for the significance of hydrogen bonding in the tanning mechanism.

2. Leather produced from such modified lignins have leather character not obtained from the unmodified lignosulfonate tannage, i.e. increased shrinkage teperature and resistance to enzymatic hydrolysis.

3. Lignin-resorcinol condensates are better tanning materials than demethylated lignins.

4. A bisulfited magnesium lignosulfonate-resorcinol condensate is an excellent water soluble tanning material.

5. The reaction between sodium bisulfite and the magnesium lignosulfonate condensate does not involve hydroxy groups.

6. The reaction between an alkali hardwood lignin and resorcinol results in the introduction of four resorcinol molecules per lignin unit and the data suggests that the hydroxyl groups are not involved.

7. The reaction between an alkali softwood lignin and resorcinol results in the addition of five resorcinol molecules and the data suggests that four hydroxyl groups are involved.

8. The magnesium lignosulfonate-resorcinol condensate is a mixture of two components. Preferential adsorption of one fraction permits a separation yielding two distinct fractions.

BIBLIOGRAPHY

1. Aulin-Erdtman, G., TAPPI 32, No. 4, p. 160 (1949)
2. Brauns, F.E., "Nature of the Chemical Components of Wood", TAPPI, Monograph No. 6, Tech. Assoc. of the Pulp and Paper Ind., (1948)
3. Brown, J.B., White, M.F., Roddy, W.T. and O'Flaherty, F. J.A.L.C.A. 42, 625 (1949)
4. Bucherer, H., Angew. Chem. 17, 1068 (1904)
5. Bulletin L-5 of the Devel. Dept. West Virginia Pulp and Paper Company (1948)
6. Claisen, Ber. 48, 3157 (1912)
7. Erdtman, H., TAPPI 32, No. 2, p. 72 (1949)
8. Erdtman, H., TAPPI 32, No. 2, p. 75 (1949)
9. Fisher, E. and Anger, G., Ber. 52, 854 (1919)
10. Flinn, E.S., "Nature of the Chemical Components of Wood", Chapt. VII, TAPPI Monograph No. 6, Tech. Assoc. of the Pulp and Paper Ind. (1948)
11. Freudenberg, E., Sitzber, heidelberger Akad. Wiss., Mathe.-naturw. Klasse 1949 No. 5, 151 C.A. 44, 2235 (1950)
12. Freudenberg, K., J. Chem. Ed. 9, 1171 (1932)
13. Freudenberg, K.J., "Die Chemie der Naturlichen Gerbstoffe" Berlin, Julius Springer (1920)
14. Fuchs and Elsner, Ber., 52B, 2281 (1919)

15. Gill, R. and Stonehill, H.J., J. Soc. Dyers Col. 60, 183 (1944)

16. Grassman, W., Collegium p. 401 (1935)

17. Gustavson, K.H., "Ingenior Vetenskaps Akademien" (Stockholm) Proceedings No. 177 (1944)

18. Gustavson, K.H., J.A.L.C.A. 45, 163 (1950)

19. Gustavson, K.H., and Larsson, A., Papperstidning No. 11B (1947)

20. Hartmann and Gatterman, Ber. 25, 3531 (1892)

21. Howard, G.C., Ind. and Eng. Chem. 26, 614 (1934)

22. Jenkins, H.O., Nature 151, 561 (1943)

23. Johnson, A.M., and Marshall, H.B., Pulp and Paper Mag. of Canada 50, 98 (1949)

24. Kolthoff, I.M. and Sandell, E.B., "Textbook of Quantitative Inorganic Analysis", The Macmillan Co. (1946)

25. Kremen, S.S. and Lollar, R.M., J.A.L.C.A. 43, 543 (1948)

26. Lewis, H.F., Brauns, F.E., Buchanan, M.A., and Brookbank, E.B., Ind. and Eng. Chem. 35 1113 (1943)

27. Lipsitz, P., Kremen, S.S., Lollar, R.M., J.A.L.C.A. 44, 371 (1949)

28. Lipsitz, P., Kremen, S.S., and Lollar, R.M., J.A.L.C.A. 44, 194 (1949)

29. McLaughlin, G.D. and Theis, E.R., "The Chemistry of Leather Manufacture", Monograph No. 101 Reinhold Publ. Corp. (1945)

30. Merrill, H.B. et al., J.A.L.C.A. 42, 536 (1947)

31. Mitscherlich, A., German Patent 4178 (1878)

32. Moeller, W., Ledertech. Runschau 6, 81 (1914) C.A. 8, 2082 (1914)

33. Moore, R.G., Wright, G.F. and Hibbert, H., Can. J. Research 15B, 532 (1937)

34. Moore, T.S. and Winmill, T.J., J. Chem. Soc. 101, 1635 (1912)

35. Nierenstein, M., "The Natural Organic Tannins", The Sherwood Press (1935)

36. Office of Technical Service Report, Leather Series Report No. 2, German Synthetic Tanning Agents

37. Ogg, Porter, and Willits, Ind. Eng. Chem., Anal. Ed. 17, 394 (1945)

38. Pauling, L., "The Nature of the Chemical Bond", Cornell Univ. Press (1945)

39. Partridge, S.M., Biochem. J., 42, 238 (1948)

40. Peniston and McCarthy, J. Am. Chem. Soc. 70, 1329 (1948)

41. Perkin and Everest, "The Natural Organic Coloring Matters", London (1918)

42. Plunguian, M., Ind. and Eng. Chem. 32, 1399 (1940)

43. Reid, Dryden, and Aronovsky, Paper Trade Journal 113, 27 (1941)

44. Roddy, W.T., J.A.L.C.A. 38, 184 (1943)

45. Russell, A., Science 106, 372 (1947)

46. Russell, A., Chem. Rev. 17, 155 (1935)

47. Salvesan, I.R. and Hogan, D., Anal. Chem. 20, 909 (1948)

48. Shuttleworth, S.G., and Cunningham, G.E., J. Soc. Leather Trades Chemists 32, 183 (1948)

49. Siggia, S., "Quantitative Organic Analysis via Functional Groups", J. Wiley and Sons (1949)

50. Testing Methods, Recommended Practices, Specifications of the Technical Association of the Pulp and Paper Industry

51. Theilheimer, W., "Synthetic Methods of Organic Chemistry" Vol. I Interscience Publ. Inc. (1948)

52. Turley, H.G., J.A.L.C.A. 40, 58 (1945)

53. Tu, Shu-Tung and Lollar, R.M., J.A.L.C.A. In print

54. Undeutsch, W.C., Enzymatic Stability of Leather Univ. of Cincinnati Masters Thesis (1950)

55. Viebach, F. and Schwappach, A., Ber. 63, 2818 (1930)

56. Wheland, G.W., "The Theory of Resonance", J. Wiley and Sons (1944)

57. White, T., J. Soc. Leather Trades Chemists 33, 39 (1949)

58. Wise, L.E., "Wood Chemistry", Reinhold Publ. Corp. (1944)

Table I-A

Preliminary Evaluation of the Tanning

Potential of Modified Lignin Derivatives

Lignin Source - Meadol

Lignin Derivative	Type of Tannage	"Feel"	Color	Shrinkage Temperature
Meadol-poly-2,4 Dihydroxy-benzoate	Solvent	Excellent	Tan	67°C.
Demethylated Meadol	Solvent	Excellent	Dark brown	81
Hydrolysis product of Chlorinated Meadol	Aqueous	Fair	Very dark brown	62
Alkaline hydrolysis product	Solvent	Good	Brown	66
Condensation product with ethanolamine	Solvent	Poor	Dark brown	-
Resorcinol-Meadol Condensate	Solvent	Excellent	Tan	80

Table I-B

Preliminary Evaluation of the Tanning Potential of

Modified Lignin Derivatives

Lignin Source - Indulin

Lignin Derivative	Type of Tannage	"Feel"	Color	Shrinkage Temperature
Indulin-poly-2,4 dihydroxy benzoate	Solvent	Very good	Brown	65°C.
Demethylated Indulin	Solvent	Excellent	Dark brown	85
Aluminum Chloride treated Indulin	Solvent	Fair	Black	62
Phosphoric Acid treated Indulin	Solvent	Poor	Black	-
Rearrangement of Ether Structure to Phenolic hydroxyl	Solvent	Good	Brown	64
Hydrolyzed Chlorinated Indulin	Solvent	Fair	Black	57
Phenol-Indulin Condensate	Solvent	Good	Brown	70
Resorcinol Indulin Condensate	Solvent	Excellent	Tan	79
Alkaline hydrolysis product	Solvent	Fair	Black	61

Table I-C

Preliminary Evaluation of the Tanning Potential of

Modified Lignin Derivatives

Lignin Source - Magnesium Lignosulfonate

Lignin Derivative	Type of Tannage	"Feel"	Color	Shrinkage Temperature
Alkaline hydrolysis product	Aqueous	Fair	Tan	68
Hydriodic acid treated product	Solvent	Very poor	Black	--
Resorcinol-Condensate	Solvent	Excellent	Tan	81

Table II

Concentration Studies for Optimum Bisulfiting Conditions

Magnesium Lignosulfonate-Resorcinol Condensate taken----------3.0 g.

Aqueous solution volume----------30.0 ml.

Reflux time----------24.0 hrs.

Concentration of Sodium Bisulfite*	Shrinkage Temperature
1 %	74 °C.
2	75
3	77
5	80
10	77
15	76
20	75

* Based on lignin derivative

Table III

Complete Evaluation of Leathers Made from Modified Lignins

Tanning Material	Demethylated Meadol	Demethylated Indulin	Resorcinol Meadol Condensate	Resorcinol Indulin Condensate	Resorcinol Magnesium Lignosulfonate Condensate	Bisulfited Resorcinol Magnesium Lignosulfonate	Magnesium Lignosulfonate
Yield % (hide substance basis)	160	152	164	169	157	164	192
Fullness	Poor	Poor	Good	Good	Good	Excellent	Excellent
Firmness	Sl. stiff	Sl. stiff	Firm	Firm	Firm	Firm	Firm
Color	Brown Black	Brown Black	Tan	Brown	Brown	Tan	Tan
Shrinkage T. °C.	77	77	83	85	88	79	68
Moisture %	10.1	10.4	9.6	8.8	10.2	10.0	11.1
Total Ash++%	0.01	0.02	0.04	0.05	0.11	0.82	0.7
Hide Substance++%	62.4	65.7	61.0	59.2	63.8	61.1	52.1
Chloroform Extract++%	4.7	4.6	7.8	8.3	4.3	3.4	4.4
Water Solubles++%	3.0	3.7	9.9	7.3	8.0	12.1	2.3
Combined Tannin++%	29.9	26.0	21.3	25.2	23.8	22.7	40.1
Degree of Tannage++%	48.0	39.5	35.0	42.5	37.4	37.1	76.8
Tryptic Hydrolysis	22.5	11.1	2.3	8.7	5.4	3.0	43.2
Tannin Penetration	Excellent	Excellent	Excellent	Excellent	Excellent	Light through center	----
Ninhydrin Reaction	+?	+?	+?	+?	+?	+	—

+ Analysis on dry basis
? Leather too dark for absolute detection of blue coloration, but darkening occurred

Table IV

Paper Chromatography of Modified Lignins

	Developing Mixture					
	Ethyl Acetate-Methanol (3:1)			N-Butyl Alcohol-Acetic Acid-Water (4:1:5)		
Lignin Derivative	Rf.	Visible Color	Color under Ultraviolet	Rf.	Visible Color	Color under Ultraviolet
Resorcinol-Indulin	1.0	Orange	Yellow	1.0	Orange	Yellow
Resorcinol-Meadol	1.0	Orange	Blue-white	1.0	Orange	Yellow
Resorcinol-Magnesium Lignosulfonate	1.0	Orange	Blue-white	1.0	Colorless	Blue-white
				0.98	Orange	Yellow
Fraction 106-T	1.0	Orange	Blue-white	1.0	Orange	Yellow
Fraction 106-N-T	1.0	Orange	Blue-white	1.0	Orange	Yellow

Table V-A

Analysis of Demethylated Lignin Derivatives

(All data on dry basis)

	Indulin	Demethylated Indulin
% Methoxy	13.9*	1.1
% Hydroxy	14.5	15.8

	Meadol	Demethylated Meadol
% Methoxy	21.6	9.2
% Hydroxy	9.9**	14.7

*Identical to value found in the manufacturers bulletin (5)

**Calculated from literature statement of 5 OH groups per lignin unit of 860 (26)

Table V-B

Analysis of Meadol and the Meadol Resorcinol Condensate

(All data on dry basis)

	Meadol	Resorcinol Meadol
% Methoxy	21.6*	14.5
% Hydroxy	9.9*	17.0

Table V-C

Analysis of Indulin and the Indulin Resorcinol Condensate

(All data on dry basis)

	Indulin	Resorcinol Indulin
% Methoxy	13.9	8.8
% Hydroxy	14.5	16.8

* Taken from literature (26)

Table V-D

Analysis of Magnesium Lignosulfonate Derivatives

(All data on dry basis)

Derivative	% Mg	% S	% OCH_3	% OH	OH : OCH_3
Magnesium Lignosulfonate	2.5	6.5	10.0	11.0	-----
Magnesium Lignosulfonate Resorcinol Condensate	0.2	2.0	7.9	16.6	3.8 : 1
Bisulfited Magnesium Lignosulfonate Resorcinol Condensate	---	4.2	7.2	16.2	4.3 : 1
Fraction 106-T*	---	1.3	7.4	16.1	-------
Fraction 106-N-T**	---	0.94	8.1	18.8	-------

* A methanol solution of this fraction gave a blue-white fluorescence when placed under ultraviolet light.

** A methanol solution of this fraction gave a yellow fluorescence when placed under ultraviolet light.