form gives readings of 60–80. The mild form of rusty mottle gives an average colorimeter reading of around 200, while the severe form averages nearly 400. On peaches, the mild form of Western X-disease gives an average reading of around 60; the moderate form, 175; and the severe form, 250.

As shown in Table 1, there seems to be a group of "mild" virus diseases of peaches, the leaves of which do not differ significantly in color reaction from that of "normal" peach foliage. There is a possibility that some of the trees from which the "normal" samples were taken may have contained a "latent" virus. Truly virus-free material might have given a consistently lower reading. On sweet cherries, it is also difficult to find a bearing tree that is completely free from all viruses, but with the spur type of growth there seems to be a certain amount of isolation for some spurs. Thus, it was possible to find "normal" spurs even on trees affected with a "mild" virus such as ring spot.

At the present time it is possible to characterize a given virus disease only by a certain colorimeter reading (or percentage transmission) because the exact chemical compound or compounds that give the color have not yet been determined. Tests on pure chemicals indicate that the color reaction is probably due to polyhydroxy phenols, possibly of the tannin group. Other workers (*1*) have shown that some virus diseases cause an increase in tannin content in the affected plants. Since tannins are recognized as protein precipitants, it seems possible that a virus infection may initiate a defense mechanism within the host plant, leading to the production of tannins.

The only factor known at present to interfere with the test is girdling. Leaves from a girdled branch of a virus-free tree give a red coloration similar to that obtained from some virus-infected leaves. Thus, the mechanism of the test may depend upon the virus causing some disturbance in the phloem of the host plant.

Preliminary studies indicate that the test will probably work on virus diseases of other trees such as apples and apricots, as well as on some virus diseases of berries, including raspberries, strawberries, and blueberries. Time has not permitted a study of the virus diseases of annual plants. Some plants may not be suitable for the particular test described here. Leaf samples of quick decline and psorosis of oranges, for example, do not give a color test. Whether tissue other than the leaf could be used in such cases is not known.

A color test for plant virus diseases would seem to have many potential uses. It should be of great aid in establishing sources of virus-free plant material for propagation purposes, and it should aid materially in the diagnosis of cases of some virus diseases where symptom expression is meager or atypical. It might also serve as a tool in physiological studies of the interaction between virus and host.

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Inhibition of Gastric Ulceration in the Rat by *o*-Hydroxybenzoic (Salicylic) Acid

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It has been shown (*2*) that extensive ulceration develops in the rumen of the stomach of the rat following ligation of the pylorus if the animals have been previously fasted for a length of time which depends upon their age. This ulceration may be inhibited and in some cases entirely prevented by the administration of certain substances. In examining the activity of monohydroxybenzoic acids, one of them was found to have a striking anti-

TABLE 1

Influence of Monohydroxybenzoic Acids on Gastric Ulceration in the Rat*

Exp. No.	Acid administered	Dose/rat† (mg)	Avg. body weight‡ (gm)	Gastric juice (ml)	Ulceration: % clear	Ulceration: Average ulceration	Ulceration: Index
1	Saline (controls)		130–109	7.6	0	3.0	3.0
	p-hydroxybenzoic	27.4	129–107	5.3	17	2.2	1.8
	m- " "	27.4	128–108	5.8	0	2.8	2.8
	o- " " (salicylic)	27.4	127–108	3.0	100	0	0
2	Saline (controls)		139–117	5.5	0	3.7	3.7
	Salicylic (*o*-hydroxybenzoic)	9.6	140–115	4.6	0	2.5	2.5
	Salicylic (*o*-hydroxybenzoic)	27.4	141–119	6.5	100	0	0
3	Sodium chloride (controls)	25	136–116	6.1	17	2.2	1.9
	Salicylic	41.4	134–112	2.8	87	0.2	0
	Acetyl salicylic (aspirin)	54.0	133–112	5.6	67	0.3	0.1

* Exp. 1 and 2 female and Exp. 3 male rats fasted 48 hr. before pylorus ligation when the trial substance was administered, and fasting continued an additional 9 hr. The acids were given as their sodium salts at pH 7.2.

† The test doses were administered intraperitoneally in Exp. 1, intravenously in Exp. 2, and *per os* in Exp. 3.

‡ There were 6 rats in each group. The first body-weight average was at the beginning of fasting, and the second, preoperative.

ulcer effect (Table 1). The sodium salts were used and the therapeutic effect obtained whether the compound was administered intraperitoneally, subcutaneously or intravenously. The latter route is usually the best for purposes of comparison. Administration of the active compound by mouth some little time preceding the ligation of the pylorus showed that it is effective when given in this manner.

Data on the antiulcer actvitiy of *o*-hydroxybenzoic (salicylic), *m*-hydroxybenzoic, and *p*-hydroxybenzoic acids

given as their sodium salts are presented in Table 1. The latter two acids have some activity, but it is very small in comparison with *o*-hydroxybenzoic (salicylic) acid. The antiulcer and antigastric secretory activity of various substances may not always go hand in hand; however, salicylic acid is not only a very potent antiulcer agent—it also reduces secretion of gastric juice. Long ago it was reported that sodium salicylate inhibits gastric acid secretion in man (*1*). It is interesting to note that of the three acids only the *o*-hydroxybenzoic acid (salicylic) gives relief in rheumatic fever. Stockman (*3*) showed that both the *m*-hydroxybenzoic acid and the *p*-hydroxybenzoic acid are practically inert as antiseptic and antirheumatic agents.

Acetyl salicylic acid (aspirin) is almost as active as salicylic acid in the prevention of gastric ulceration. The activity of other derivatives of salicylic acid, various dihydroxybenzoic acids, and related compounds is now under study.

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Constitution of Gymnosperm Lignin[1]

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Freudenberg's deduction (*5*) of a benzpyrane ring constitution for gymnosperm lignin has recently been extended by Russell's (*19*) proposal of a polyflavanone structure as its specific form. However, the alleged synthesis offered as evidence is open to doubt (*2*). In fact, for substances such as lignin and its derivatives, which are unresponsive to most criteria for identity save ultimate and functional group analysis, even the best evidence of synthesis is contributory but scarcely conclusive. This is certainly the case when the reaction used is one so little suited to give predictable results as is the aluminum chloride-catalyzed Fries rearrangement, by means of which vanillin monoacetate is claimed to rearrange and condense to a polymer corresponding to gymnosperm lignin.

Evidence of a different and more reliable character has been accumulating in this laboratory, and we are prompted to report, perhaps somewhat prematurely, a summary of our investigations on the structure of lignin derivatives and the tentative conclusions drawn from our observations. The hypothesis that lignin from Western hemlock (*Tsuga heterophyla*) is a polyflavanone and that lignin sulfonic acid may be the polyflavanone 3-sulfonic acid is a concept upon which our experimental program was based as early as May 1947.

The evidence rests upon the use of periodic acid to determine the arrangement of oxygen substituted in the nonbenzenoid portion of lignin and its derivatives; upon the behavior of lignin sulfonic acid and other lignins in methylation and acetylation reactions, with particular attention to the influence of alkali; and upon the chemical and physical demonstration of the presence of carbonyl groups in the nonbenzenoid portion of the lignin molecule.

The use of periodic acid for structural determination of lignin rests upon the fact that lignin sulfonic acid is attacked by that reagent (*14*). It was soon understood that reaction with free phenols can scarcely be extensive, considering the small quantity present (*15*), but the large amount of demethylation with the formation of methanol does demonstrate that the reaction involves the aromatic ring (*14, 15, 20*). That phenols contribute to only a minor portion of the reactivity is shown also by the failure to block the oxidation by a single treatment with diazomethane, which should quantitatively methylate most phenolic substituents. Successive methylations with diazomethane fail to eliminate the oxidation until a composition corresponding to $C_9H_{7.75}O_{1.8}(SO_3NH_4)_{0.5}(OCH_3)_{1.52}$ (A) is reached. Reaction with periodate then ceases. Phenolic groups become available for methylation with diazomethane through an equilibrium promoted by alkali, in the presence of which successive methylations give the product (A), containing 19.6% methoxyl. The same number of methylations (five) carried out in neutral solution gives a product with only 14.5% methoxyl. More methoxyl can be introduced by dimethyl sulfate in cold aqueous alkaline solution, as found by Hibbert and his co-workers (*11*). This derivative, which has the composition $C_9H_{7.6}O_{2.2}(SO_3NH_4)_{0.42}(OCH_3)_{2.17}$, is not oxidized by periodic acid. A water-soluble acetyl derivative resistant to periodic acid oxidation has the composition $C_9H_6O_{2.6}(SO_3NH_4)_{0.5}(OCH_3)_{0.9}Ac_{1.2}$. During these reactions no alteration in molecular weight occurs.

Comparisons of the composition of ammonium lignin sulfonate, $C_9H_6O_{2.2}(SO_3NH_4)_{0.5}(OCH_3)$, and its methyl and acetyl derivatives show the introduction of a new oxygen atom for each methylene group.[2] Thus, during the methylation reactions there are added also the elements of water.

Since neither diazomethane nor cold dilute alkaline dimethyl sulfate will react with any but phenolic groups, phenols must be liberated during the methylations. For this to occur without alteration in molecular weight or change in the generic composition (*i.e.* no fragments lost) can mean only that the phenols are liberated from an intramolecular linkage. This means a ring opening. Assuming the benzpyrane structure, the phenolic libera-

[1] Contribution from Pulp Mills Research Project, University of Washington, Seattle 5, Washington.

[2] Methylation of a phenol adds not a methoxyl, but a methylene, group. Thus,

$$C_6H_5OH + CH_3OSO_2OCH_3 \longrightarrow C_6H_5O\overset{H}{\underset{|}{C}}H_2,$$