## Summary

1. It has been found that solid furoperacid decomposes spontaneously at 35°, yielding furoic acid, carbon dioxide, a resinous, alkali-soluble polymer and small amounts of 4,5-epoxyfuroic acid.

2. The decomposition of solid furoperacid is brought about, at room temperature, by certain types of inorganic and organic solids, and also by ultraviolet light in the region of 4500 to 3600 Å. An explanation has been offered to account for this effect.

3. It has also been found that furoperacid in chloroform solution decomposes mainly into furoic acid and oxygen.

4. The decomposition of furoperacid in chloroform has been measured at 35 and at 40°, and has been found to obey the monomolecular law. The temperature coefficient was found to be 2.30, and the heat of activation 15,800 calories per mole. The decomposition in chloroform can therefore be represented by the equation, $k = 1.035 \times 10^9 e^{-15.800/RT}$.

5. Ultraviolet light in the region of 4500 to 3600 Å. accelerates considerably the decomposition of furoperacid in glacial acetic acid solution.

CAMBRIDGE, MASS. RECEIVED FEBRUARY 13, 1934

---

[CONTRIBUTION FROM THE NEW JERSEY AGRICULTURAL EXPERIMENT STATION]

# Transformation of the Methoxyl Group in Lignin in the Process of Decomposition of Organic Residues by Microörganisms[1]

BY SELMAN A. WAKSMAN AND H. W. SMITH

It has been established definitely[2] that the lignin constituents of plant residues play a highly important role in the process of humus formation. Because of the resistance of lignin to decomposition by the great majority of microörganisms, it is allowed to accumulate in soils, peats and composts; however, lignin does not remain in an unmodified form in the humus, but it undergoes certain changes, both of a purely chemical and biological nature. Comparatively little is known concerning the modifications of the lignin in the process of humus formation, largely because of the limited knowledge of the chemical structure of its molecule.

Among the characteristic groups of the lignin molecule, the methoxyl ($OCH_3$) has been studied most extensively, and has been found to play a prominent part in the transformation of the lignin into humus. This group is present in lignin both in ester and in ether combinations. The determination of the methoxyl content in the ether form is frequently taken as a measure of the lignin content of the plant material. Powell and Whittaker[3] concluded that the lignin molecule (flax lignin prepared by alkali extraction) contains four methoxyl groups and five hydroxyl groups capable of acetylation, only three of the latter being phenolic in character. According to Brauns and Hibbert,[4] spruce lignin contains five methoxyl groups and five free hydroxyl groups, three of which are aliphatic in character, one phenolic or enolic, and one reacting with alkylating reagents. The methoxyl content of lignin has been shown to vary in concentration in lignins of different plants,[5] and even in the lignin of the same plant, at different stages of growth;[6] it may also depend on the position of the lignin in the plant tissues, the methoxyl content of the lignin in the middle lamella of wood having been found[7] to be 11–13%, while that of the lignin in the cell wall was only 4.3–4.8%.

According to Beckmann, Liesche and Lehmann,[6] there is not only an increase in the lignin content of plants with an increase in maturity, but the amount of methoxyl in the lignin also increases. Fuchs[5] concluded that formation and methylation of lignin do not go hand in hand, and that methylation is a secondary process. Assuming that the methoxyl present in wood is due entirely to lignin, he calculated for "native lignin"

---

[1] Journal Series Paper of the New Jersey Agricultural Experiment Station, Department of Soil Microbiology.

[2] S. A. Waksman, *Trans. Second Comm. Intern. Soc. Soil Sci. Budapest*, 3 (1929); *Cellulosechem.*, **11**, 209 (1930); J. Page, *J. Agr. Sci.*, **20**, 455 (1930).

[3] W. J. Powell and H. Whittaker, *J. Chem. Soc.*, **125**, 357 (1923); **127**, 132 (1925).

[4] F. Brauns and H. Hibbert, THIS JOURNAL, **55**, 4720 (1933).

[5] W. Fuchs, "Die Chemie des Lignins," Verlag Julius Springer, Berlin, 1926.

[6] E. Beckmann, O. Liesche, and F. Lehmann, *Biochem. Z.*, **139**, 491 (1923).

[7] G. J. Ritter, *Ind. Eng. Chem.*, **17**, 1194 (1925); **18**, 608 (1926).

or the unmodified lignin in wood, an average methoxyl content of 21.5%; according to Brauns and Hibbert,[4] "native lignin" contains only 17.5% methoxyl. Various lignin preparations obtained from wood by the acid treatment are usually reported to contain only 14.1 to 16.3% methoxyl. The methoxyl content of lignin in mosses is considerably lower: *Sphagnum medium* was shown[8] to contain only 0.3% methoxyl and *S. cuspidatum* 0.4%. The question is frequently raised, therefore, whether the lignin in mosses should be considered as "true lignin," or as a pectin-like substance containing a small amount of methoxyl.[5]

Assuming that the methoxyl content of the lignin molecule does not undergo any change during the decomposition of plant residues by microörganisms, König[9] concluded that lignin is the most resistant constituent. In the decomposition of stable manure in soil for a period of one and a half years, the pentosan disappeared completely, the cellulose was appreciably reduced, while a large part of the lignin, calculated on the basis of the methoxyl content, still remained. Fischer *et al.*[10] have shown that the methoxyl content of high moor peat increases with an increase in depth, from 0.49% at the surface to 1.67% at 1.8 meters; in the case of low moor peat, the methoxyl content decreases with depth, from 2.97% at the surface to 1.66% at a lower depth. They concluded[8] that during the decomposition of the plant materials, the lignin changes to the alkali-soluble "humic acid," which still contains some methoxyl; this group is gradually replaced by a hydroxyl group. In the case of a lake bottom formation, Ohle[11] obtained an increase in the methoxyl concentration with an increase in depth of sediment, up to a certain point; below that depth, the methoxyl content diminished rapidly. The methoxyl content of lignin in wood which has been decomposed by fungi was shown[12] to be lower than that of lignin in normal wood; the former is also more readily split off in the chemical preparation of lignin than the latter.

## Experimental

The following investigations deal primarily with the problem of transformation of lignin, especially its methoxyl groups, in the decomposition of plant residues; the nature of the plant materials, the conditions of decomposition and the method of lignin preparation are important factors; particular emphasis has been laid upon the relation of this process to humus formation in soils, peats and composts. The methoxyl content of the lignin fractions was determined by the Zeisel method, the pentosan content by the A. O. A. C. method, and the uronic acid by the method of Dickson, Otterson and Link.[13]

In Table I are given the changes in the methoxyl content of lignin, during the decomposition of horse manure in compost, as well as during the subsequent growth upon this compost of the cultivated mushroom, *Agaricus campestris*, a typical lignin-decomposing organism. The lignin content of the manure was found to increase in the process of composting, namely, from 32.9 in fresh manure to 42.1% in the compost. However, growth of the *Agaricus* resulted in a decrease in the lignin, both in fresh and in composted manures, since this organism feeds largely upon the lignin constituents of the manure.[14] The methoxyl content of the lignin changed very little, both during the process of composting of the manure and as a result of subsequent growth of the fungus. One may thus conclude that when plant residues are undergoing rapid decomposition, as in composting of stable manure and in the development of the mushroom fungus upon the compost, the lignin molecule is transformed as a whole, with little change in its methoxyl groups.

TABLE I

CHANGES IN LIGNIN CONCENTRATION AND IN METHOXYL CONTENT DURING THE DECOMPOSITION OF HORSE MANURE IN COMPOST AND DURING THE SUBSEQUENT GROWTH OF AGARICUS CAMPESTRIS

| Nature of manure | Growth of *Agaricus*[a] | Loss in wt. of organic matter as a result of *Agaricus* growth,[b] % | Total nitrogen in ash-free material, % | Lignin content of ash-free material,[c] % | Methoxyl content of ash-free material, % | Methoxyl content of "native lignin" calcd., % |
|---|---|---|---|---|---|---|
| Fresh | − | .. | 1.79 | 32.9 | 4.00 | 12.16 |
| Fresh | + | 29.5 | 2.82 | 19.2 | 2.26 | 11.77 |
| Composted | − | .. | 3.04 | 42.1 | 4.80 | 11.40 |
| Composted | + | 28.3 | 4.19 | 19.1 | 2.64 | 13.82 |

[a] − control material; + material (fresh or composted) upon which the *Agaricus* was grown. [b] For a period of 264 days. [c] Acid lignin.

Lignin preparations were obtained by the acid and alkali methods from the manure compost and the residue left after the growth of the fungus; the first were found to contain less methoxyl, as shown in Table II. As a result of the growth of the *Agaricus*, the lignin molecule is modified. The possibility is not excluded of course that some of the methoxyl in the compost may be due to non-lignin complexes, such as polyuronides;[15] this would affect considerably the calculation of the methoxyl in the

---

(8) Fr. Fischer and H. Schrader, "Entstehung und chemische Struktur der Kohle," Essen, 1922.

(9) J. König, *Biochem. Z.*, **171**, 261 (1926).

(10) F. Fischer, H. Schrader and Friedrich, *Abh. Kohle*, **5**, 530 (1922); *Brennstoff Chem.*, **3**, 341 (1923).

(11) W. Ohle, *Biochem. Z.*, **258**, 420 (1933).

(12) C. G. Schwalbe and A. Ekenstam, *Cellulosechem.*, **8**, 13 (1927).

(13) Dickson, Otterson and Link, THIS JOURNAL, **52**, 775 (1930).

(14) S. A. Waksman and W. Nissen, *Am. J. Bot.*, **19**, 514 (1932).

(15) G. J. Ritter and E. F. Kurth, *Ind. Eng. Chem.*, **25**, 1250 (1933).

TABLE II

CHANGES IN METHOXYL CONTENT OF LIGNIN, PREPARED BY ACID AND ALKALI TREATMENTS, AS A RESULT OF GROWTH OF AGARICUS CAMPESTRIS[a]

| Growth of *Agaricus* | Nature of lignin | Methoxyl in ash-free and protein-free lignin prepns., % |
|---|---|---|
| − | Acid[b] | 9.50 |
| + | Acid | 5.35 |
| − | Alkali | 12.08 |
| + | Alkali | 7.58 |

[a] Composted manure used. [b] Using 80% sulfuric acid for two and one-half hours in the cold, diluting with fifteen volumes of water and heating at 100° for five hours.

"native lignin." However, the fact that the carbohydrates are much more rapidly decomposed than the lignin, would tend to give more weight to the assumption that the methoxyl content of composts is due largely if not entirely to lignin.

TABLE III

CHANGE IN METHOXYL CONTENT OF LIGNIN IN DIFFERENT PLANT MATERIALS DECOMPOSING UNDER AEROBIC AND ANAEROBIC CONDITIONS. PER CENT. OF DRY MATERIAL

| Nature of material | Ash content[a] | Nitrogen content[a] | Total lignin, ash and N-free | Total methoxyl in ash-free material | Methoxyl in "native lignin," calcd. |
|---|---|---|---|---|---|
| Corn stalks, fresh | 7.5 | 0.66 | 16.8 | 2.33 | 13.93 |
| Corn stalks, decomposed aerobically | 29.7 | 2.34 | 33.6 | 3.10 | 9.24 |
| Corn stalks, decomposed anaerobically | 39.8 | 2.86 | 24.2 | 2.00 | 8.26 |
| Rye straw, fresh | 4.2 | 0.24 | 18.5 | 2.10 | 14.40 |
| Rye straw, decomposed aerobically | 19.3 | 2.37 | 35.0 | 4.58 | 13.09 |
| Rye straw, decomposed anaerobically | 12.0 | 1.74 | 34.1 | 3.36 | 9.86 |
| Oak leaves, fresh | 5.1 | 0.77 | 30.3 | 2.97 | 9.84 |
| Oak leaves, decomposed aerobically | 35.8 | 3.14 | 36.1 | 2.84 | 7.87 |
| Oak leaves, decomposed anaerobically | 34.9 | 2.03 | 41.9 | 2.49 | 5.93 |

[a] The increase in ash and nitrogen content as a result of decomposition is due both to the accumulation of these constituents from the original plant material and to the transformation of the minerals and inorganic nitrogen salts added to the compost into organic compounds.

A more detailed study of the effect of prolonged decomposition of plant residues upon the fate of the methoxyl group in the lignin molecule is given in Table III. Several plant materials, which have undergone decomposition for a period of nearly five years,[16] were used. Decomposition of the materials, both under aerobic and anaerobic conditions, led to an increase in the lignin content; this was accompanied by an increase in the relative amount of ash and of organic nitrogenous compounds. The methoxyl content of the residual lignin (acid) was considerably reduced as a result of decomposition, especially under anaerobic conditions. This tends to emphasize further the fact that the lignin molecule is modified considerably during the decomposition of plant residues by microorganisms, even if the lignin complex as a whole is not destroyed. This modification is particularly evident in the case of plant materials which decompose under anaerobic conditions. Similar, frequently even more striking, results were obtained in the decomposition of oat straw. The possibility is not excluded, of course, that some of the methoxyl destroyed by the microörganisms is not due to lignin, but to carbohydrate complexes.

A sample of cedar wood which has undergone a process of aerobic decomposition by fungi, whereby the cellulose was rapidly reduced and the lignin accumulated, was analyzed for lignin and methoxyl content. The total residual lignin (acid) in this decomposed wood was 71.14% and the methoxyl content of the wood was 10.90%; this amounted to 15.32% methoxyl in the lignin. The methoxyl content of "native lignin" in sound wood was found to be 18.37%, while the methoxyl content of acid lignin prepared from sound wood was 14.71% (Table IV). These results, as well, point to the fact that the aerobic decomposition of a plant material by microörganisms not able to attack the lignin molecule brings about only a limited reduction of its methoxyl content; even this limited reduction may be due not so much to the destruction of the methoxyl in the lignin as that of the carbohydrate complexes.

TABLE IV

INFLUENCE OF TREATMENT OF PLANT MATERIAL UPON THE METHOXYL CONTENT OF ITS LIGNIN

| Nature of material | Concn., % |
|---|---|
| Acid lignin in ether extracted wood | 35.38 |
| Methoxyl content of ether extracted wood | 6.50 |
| Methoxyl content of lignin in wood, calculated ("native lignin") | 18.37 |
| Methoxyl content of acid lignin, found directly by analysis | 14.71 |

The influence of anaerobic decomposition of plant materials upon the transformation of the lignin molecule is shown further in Table V. The peat samples were obtained from different depths of a low moor peat profile in the Everglades, Florida: this represents a *Cladium* formation with a distinct sedimentary layer. The results show that, although the lignin content increased considerably when the plant residues were changed into peat, the methoxyl content of the peat diminished rapidly; when the methoxyl content of the residual lignin is calculated, a still greater decrease is found with an increase in age of peat. These results tend to confirm the previous observations that, in the decomposition of plant residues under anaerobic conditions, the lignin complex loses its methoxyl groups more readily than under aerobic conditions of decomposition.

The methoxyl changes in the lignin of high moor or sphagnum peat were found to be similar (Table VI). The origin and chemical nature of this peat were described in detail elsewhere.[17] The methoxyl content of lignin in sphagnum plants is known to be considerably lower than that of lignin in grasses and in other high plants. In the decomposition of sphagnum plants under the anaerobic

---

(16) S. A. Waksman and F. G. Tenney, *Soil Sci.*, **26**, 155 (1928); **28**, 55 (1929); **30**, 143 (1930).

(17) S. A. Waksman and K. R. Stevens, *ibid.*, **27**, 271, 389 (1929).

TABLE V

METHOXYL CONTENT OF "ACID LIGNIN" IN DIFFERENT LAYERS OF A LOW MOOR PROFILE, AS COMPARED WITH THAT OF PEAT FORMING PLANTS

| Nature of material | Description of peat | Total lignin in ash-free material,[a] % | Total methoxyl in ash-free material,[a] % | Methoxyl content of lignin (calcd.),[b] % |
|---|---|---|---|---|
| *Eriophorum* plants | | 23.64 | 4.12 | 17.42 |
| *Cladium* roots | | 30.90 | 3.24 | 10.48 |
| Peat layer 51 | Upper fibrous layer | 51.24 | 4.44 | 8.66 |
| Peat layer 53 | Lower part of fibrous layer adjacent to sedimentary layer | 51.77 | 1.44 | 2.78 |
| Peat layer 54 | Sedimentary layer | 48.33 | 2.02 | 4.17 |
| Peat layer 55 | Lower part of sedimentary layer adjacent to lower fibrous layer | 49.08 | 2.26 | 4.60 |
| Peat layer 56 | Lower fibrous layer | 57.00 | 2.94 | 5.15 |

[a] In the case of the *Eriophorum* plants and the *Cladium* roots, the results are reported on a total dry material. [b] Assuming that all the methoxyl is due to lignin; in the case of the fresh plant materials, some of the methoxyl is no doubt due to the non-lignin constituents.

TABLE VI

METHOXYL CONTENT OF LIGNIN IN SPHAGNUM PLANTS AND IN SPHAGNUM PEATS[a]

| Nature of material | Lignin content in total dry material, % | Methoxyl content in total dry material, % | Methoxyl in lignin, % |
|---|---|---|---|
| Sphagnum plants | 12.50 | 0.881 | 7.05 |
| Young sphagnum peat | 41.45 | 1.518 | 3.66 |
| Older sphagnum peat | 61.92 | 1.816 | 2.93 |

[a] Assuming that all the methoxyl is due to lignin.

TABLE VII

CHEMICAL NATURE OF SPHAGNUM PLANTS AND SPHAGNUM PEAT. PERCENTAGE OF DRY MATERIAL

| Constituents | Sphagnum plants | Sphagnum peat |
|---|---|---|
| Total carbon | 41.95 | 46.36 |
| Total nitrogen | 0.72 | 0.78 |
| Total ash | 4.92 | 2.14 |
| Total "hemicellulose"[a] | 31.30 | 24.40 |
| Pentosan | 11.84 | 9.58 |
| Uronic acid anhydride | 16.50 | 11.70 |
| Cellulose, as glucose[b] | 14.50 | 16.00 |
| Acid lignin | 12.55 | 31.00 |
| Methoxyl content of original material | 0.91 | 0.81 |
| Methoxyl in "native lignin," calcd. | 7.25 | 2.61 |

[a] Hydrolyzed by 2% hydrochloric acid, at 100° for three hours, and reported as reducing sugar. [b] Residue left, after "hemicellulose" determination, was treated with 80% sulfuric acid solution in the cold, then diluted with fifteen volumes of water and heated, at 100° for five hours.

TABLE VIII

CHEMICAL NATURE OF "ACID LIGNIN" PREPARATIONS FROM SPHAGNUM PLANTS AND FROM SPHAGNUM PEAT. PERCENTAGE OF DRY MATERIAL

| Constituents | Acid lignin from sphagnum plants | Acid lignin from sphagnum peat |
|---|---|---|
| Total carbon | 49.86 | 53.85 |
| Total nitrogen | 0.79 | 0.84 |
| Total ash | 9.80 | 1.60 |
| Pentosan | 0.92 | 0.56 |
| Uronic acid anhydride | 5.97 | 5.08 |
| Methoxyl content | 1.06 | 3.04 |
| Fats and waxes (ether-soluble) | 6.30 | 5.34 |

conditions which prevail in a peat bog, the methoxyl concentration tends to diminish, while the relative "lignin" content increases. It is assumed again that all the methoxyl found in sphagnum plants and in peat is due to lignin. However, when lignin preparations were obtained by the acid and alkali treatments, they were found to contain much less methoxyl than the calculated "native lignin" in the plants; the methoxyl content of the lignin from sphagnum plants and from sphagnum peat were 1.5 to 2.3%, respectively. This points either to the fact that a large part of the methoxyl groups in the sphagnum is not due to true lignin, or that this methoxyl is loosely bound to the lignin molecule and is readily split off in the preparation of the lignin. The low methoxyl content of the lignin in sphagnum and in other mosses has been explained by Fuchs as due to the primitive nature of these plants.

The chemical composition of lignin preparations from sphagnum plants and peat, as compared with that of the original materials, is given in Tables VII, VIII and IX.

The acid lignin preparations were obtained by treating 200 g. quantities of dry sphagnum plants and sphagnum peat with 80% sulfuric acid for three hours in the cold, then diluting with fifteen volumes of water, heating for five hours, filtering through paper, washing and drying. The "alkali lignin" was prepared as follows: four 200-g. quantities of the two materials were extracted with hot water, followed by extraction with cold 2% ammonia solution, then by hot 4% sodium hydroxide solution; the last extraction was carried out first at 100°, then repeated at 120°. The hot water extracts of the materials were treated with two volumes of ethyl alcohol; the precipitate was collected, washed with acetone and dried. The alkali extracts were precipitated in the cold with hydrochloric acid solution. The residue after the second alkali extraction was washed with acetic acid and water and dried.

The "acid lignin" preparations can hardly be considered as true lignin: the carbon content is too low, even when calculated on an ash and nitrogen-free basis; the methoxyl content is too low, confirming the previous results that the methoxyl complex in mosses is easily split off on treatment with a mineral acid. The high uronic acid content of the preparations makes one question their true lignin nature. The same is true of the "alkali lignin" preparations. Whether the lignin from sphagnum has a constituent COOH group which is acted upon by hot 12% hydro-

TABLE IX

CHEMICAL NATURE OF HOT WATER EXTRACTS AND ALKALI SOLUBLE COMPLEXES FROM SPHAGNUM PLANTS AND SPHAGNUM PEAT. PERCENTAGE OF DRY MATERIAL

| Treatment | Chemical analysis | Sphagnum plants | Sphagnum peat |
|---|---|---|---|
| Hot water soluble, alcohol precipitated | Total yield[a] | 0.45 | 0.15 |
| | Uronic acid anhydride | 20.82 | 19.73 |
| | Reducing sugar on hydrolysis with 2% HCl | 73.03 | 77.72 |
| Fraction soluble in cold $NH_4OH$ and precipitated hy HCl | Total yield[a] | 0.72 | 2.51 |
| | Total carbon | 50.22 | 56.05 |
| | Total nitrogen | 4.59 | 4.20 |
| | Uronic acid anhydride | 10.00 | 9.20 |
| | Reducing sugar on hydrolysis with 2% HCl | ... | 8.00 |
| | Methoxyl | ... | 1.14 |
| Fraction soluble in 4% NaOH solution, precipitated by HCl | Total yield[a] | 5.65 | 24.85 |
| | Total carbon | 54.47 | 58.23 |
| | Total nitrogen | 2.06 | 2.17 |
| | Uronic acid anhydride | 8.92 | 6.17 |
| | Pentosan | ... | 4.18 |
| | Reducing sugar on hydrolysis with 2% HCl | 10.60 | 9.30 |
| | Methoxyl | ... | 1.13 |
| Residue, fraction insoluble in hot 4% NaOH | Total residue[a] | 28.5 | 23.6 |
| | Total carbon | 39.47 | 42.63 |
| | Total nitrogen | 0.24 | 0.46 |
| | Uronic acid anhydride | 10.7 | 7.8 |
| | Pentosan | 10.26 | 7.32 |
| | Reducing sugar on hydrolysis with 2% HCl | 17.3 | 11.3 |

[a] On the basis of 100 g. of original dry material.

chloric acid, giving carbon dioxide or whether the uronic acid anhydride is in some way admixed or attached to the lignin molecule in the sphagnum plant still remains to be determined. The presence of uronic acid no doubt accounts largely for the highly acid nature of the sphagnum plants and of sphagnum peats; it can serve to explain more readily the acid properties of this type of peat than the presence of hypothetical "humic acids." One may conclude from these results that "humic acid" of sphagnum peats consists of lignin derivatives with a low methoxyl content and of certain polyuronides, containing uronic acid complexes, and probably certain sugars.

It has been shown elsewhere[18] that synthetic humus complexes can be prepared by the interaction of alkali solutions of proteins and lignins. A series of such complexes were analyzed for their methoxyl content; the results (Table X) brought out the fact that there was no loss in methoxyl in the combination of the lignin with the protein. Humus or "humic acid" preparations ($\alpha$-humus) obtained from peat showed a much lower methoxyl content; this points to a chemical change in the lignin molecule in the process of its transformation into humus complexes, under natural conditions. This loss of methoxyl in the formation of humus from lignin is thus found to be not a matter of replacement by another group, such as a protein or a base, but is due to the direct activities of microorganisms bringing about the decomposition of the plant residues; this is especially true under anaerobic conditions which prevail in a peat bog.

TABLE X

METHOXYL CONTENT OF NATURAL AND ARTIFICIAL HUMUS COMPLEXES

| Nature of preparation | Nitrogen content, % | Methoxyl content, % |
|---|---|---|
| Wood lignin | 0.12 | 15.80 |
| H-ligno-proteinate | 3.62 | 11.85 |
| H-ligno-proteinate, insoluble in alcohol | 5.90 | 12.05 |
| H-ligno-proteinate, soluble in alcohol | 2.10 | 11.65 |
| Ca-ligno-proteinate[a] | 1.78 | 10.35 |
| $\alpha$-Humus from peat | 2.21 | 1.90 |

[a] Ash content 11.7%.

## Summary

Methoxyl is a characteristic group of the lignin molecule, but its relative concentration varies considerably with the nature of the plant and the degree of its maturity.

In the aerobic decomposition of plant residues in composts or in soils by microörganisms capable of utilizing the lignin molecule, the latter is attacked as a whole, while the methoxyl content of the residual lignin is not modified to any marked extent.

In the decomposition of plant residues in soils and in peat bogs, under conditions favorable to lignin accumulation, the methoxyl content is gradually reduced, especially under anaerobic conditions. Anaerobic bacteria and to a less extent aerobic organisms, which are unable to attack the lignin molecule as a whole, are still capable of modifying it by reducing its methoxyl content.

In the process of peat formation, which consists in the decomposition of plant residues under anaerobic conditions, there is an accumulation of lignin, because of its resistance to decomposition under these conditions; this is accompanied by a decrease in the methoxyl content. The gradual transformation of the lignin into the dark-colored humus substances is characterized by the reduction of the methoxyl content.

Sphagnum plants are characterized by a low methoxyl and a high uronic acid content; the uronic acid complexes in this material are also resistant to decomposition. Sphagnum plants give rise, as a result of anaerobic decomposition, to a type of peat which is rich in lignin-like complexes of a low methoxyl content and high in uronic acid.

NEW BRUNSWICK, N. J. RECEIVED FEBRUARY 14, 1934

---

(18) S. A. Waksman and K. R. N. Iyer, *Soil Sci.*, **34**, 43 (1932).