

SEPARATE PRINT

59
F48

Plant Research and Development

A Biannual Collection
of Recent German Contributions Concerning
Development through Plant Research

Volume 4

Edited in Conjunction with
Numerous Members of German Universities
and Research Institutions by the
Institute for Scientific Co-operation

HUMUS FORMATION FROM THE ROTTING DOWN OF MAIZE STRAW

by

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Besides containing relatively high amounts of minerals (plant nutrients), maize straw presents itself as a profitable source of humus for the soil. It is essential to promote the formation of humus as it is so important for the fertility and health of the soil. Ammonia has the property of reacting with rotting materials to form humic substances. In contrast to this, nitrate is hardly absorbed by the soil and is, in fact, either taken in by plants, washed away, or denitrified. The constant formation and breakdown of humus improves the physical properties of the soil and also provides plants with a steady source of nitrogen.

In the following model experiments, therefore, ammonia-producing nitrogenous fertilizers such as urea and calcium cyanamide were added to rotting maize straw. When plant material rots down, there is always a darkening of colour, which is indicative both of the more or less considerable change taking place in the existing fractions as well as of the formation of completely new organic substances. Humic substances are a heterogeneous mixture which is difficult to define, and the fractions into which they can be divided depend on the analytical procedure used. We used the following methods to determine and compare humic substances at particular stages of decomposition. We think this procedure adequately shows the extent of the humus formation according to the decomposition of the original material.

Test procedure

Maize straw was divided up into leaves, stalks and spindles. 0, 0.5, 1 or 1.5 % of nitrogen (based on the organic substance of the plant material)

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was added in the form of urea (U 1, U 2, U 3) or calcium cyanamide (C 1, C 2, C 3) and the material was then left to rot down. During the first five weeks, samples were taken every week for the various tests and then at five-weekly intervals for a total of 40 weeks [1, 2, 3].

Methodology

The degree of humification was determined by the following tests:

1. The *colour value* (CV) (= the humification index [HI]) according to Springer gives the extinction which the darkly coloured alkaline extract of 1 g of organic matter possesses after acid treatment (1 cm layer thickness, 100 ml extract, ELKO 57 filter). The colour value is 3.8 times smaller than the humification index [5].
2. The *humic acids* (HA) were obtained by extraction with an alkaline solution and then purified by means of precipitation, centrifugation, filtering and extraction with a benzol-alcohol mixture (1 : 1).
3. The *humic substances* (HS) were determined by sulphacetolysis according to Springer [5].
4. The lignin or lignin-humus complex was determined by extraction with 80 % sulphuric acid.

Results

1. *Colour value* (Table 1)

The colour value, which is a measure of the amount of humic substances dissolving in alkali, makes it possible to recognize the start of the humification process even before stable humus fractions are detectable. It is already fairly high in the leaves at the start of the rotting down process (0.46) because, due to the late harvesting of maize, a certain natural decomposition of its ingredients has doubtless already begun. In the stalks, and even more so in the spindles, yellow and reddy-coloured organic substances dissolve in alkali, these probably being oxidized lignin as well as derivatives of carotene, xantophyll, etc.

The colour values rise sharply in all test groups after only 5 weeks' decomposition and continue to increase up to the 40th week as the organic material breaks down. In the case of the *leaves*, the colour value rises up to the 5th week at practically the same rate in all test groups except the O group. At the end of 25 weeks, the values in all groups have to a large extent levelled out and are roughly the same at this point. Colour values of

Table 1 - Colour values, humic acids and humic substances resulting from the decomposition of maize straw
(Original quantity: 300 g organic matter/sample)

Rotting down period (weeks)	O			U ₁			U ₃			C ₁			C ₃		
	CV	HA	HS	CV	HA	HS	CV	HA	HS	CV	HA	HS	CV	HA	HS
<i>Leaves</i>															
0	0.46	-	9.0	0.46	-	9.0	0.46	-	9.0	0.46	-	9.0	0.46	-	9.0
5	1.05	0.19	8.3	1.53	11.9	18.6	1.53	0.4	19.7	1.50	5.4	16.9	1.54	16.1	21.5
10	1.51	0.45	11.5	2.42	12.7	26.0	2.00	7.0	25.0	1.85	5.8	24.0	1.92	22.2	30.6
15	2.64	12.5	26.3	3.08	12.4	34.0	2.63	11.1	32.0	2.90	10.9	34.7	2.05	22.4	32.5
20	3.06	13.2	30.3	3.20	11.7	36.6	2.78	10.8	34.4	3.12	10.7	38.4	2.53	22.2	31.1
25	3.21	13.1	30.8	3.28	13.0	36.7	3.17	12.0	37.1	3.35	10.8	38.0	3.14	23.6	34.4
40	3.9	15.0	35.0	3.5	16.0	40.0	3.8	16.0	39.0	3.8	16.0	39.0	3.9	27.0	40.0
<i>Stalks</i>															
0	0.12	-	3.3	0.12	-	3.3	0.12	-	3.3	0.12	-	3.3	0.12	-	3.3
5	0.79	0	5.3	0.61	0.4	8.3	0.55	0.2	4.7	0.50	3.2	3.7	0.52	5.8	4.7
10	1.29	0	4.6	1.19	0.4	9.0	0.79	0.3	11.9	0.90	2.9	5.7	0.92	15.6	9.5
15	1.60	0	5.4	1.41	3.8	12.7	1.25	13.0	12.8	1.23	3.4	9.7	1.23	17.6	11.4
20	1.82	0	7.0	1.53	8.0	13.0	1.55	12.8	12.8	1.58	12.5	12.0	1.27	17.2	11.9
25	1.94	0.3	8.3	1.84	12.5	15.8	1.55	15.7	12.4	1.69	17.6	15.9	1.68	17.1	13.2
40	1.8	1.2	11.0	1.9	11.4	13.0	1.8	12.0	13.0	1.7	14.0	18.0	1.9	19.0	18.0
<i>Spiraltes</i>															
0	0.33	-	17.4	0.33	-	17.4	0.33	-	17.4	0.33	-	17.4	0.33	-	17.4
5	0.53	2.3	14.6	0.44	1.2	17.0	0.44	2.2	29.3	0.45	1.5	25.6	0.55	2.8	35.7
10	0.63	2.3	13.5	0.60	1.9	16.0	0.50	1.4	28.1	0.63	1.8	22.1	0.66	2.5	39.7
15	0.72	1.4	12.1	0.84	1.4	16.0	0.58	1.5	25.0	0.92	1.8	25.0	0.75	2.3	36.5
20	0.82	1.5	13.5	0.99	1.4	20.4	0.69	1.8	26.5	1.04	1.3	26.4	0.92	2.9	32.1
25	0.89	1.7	16.5	1.20	1.9	22.2	0.74	1.7	27.0	1.15	0.8	25.4	1.16	3.7	30.0
40	1.40	1.8	18.0	1.40	1.7	21.0	0.90	1.7	26.0	1.40	1.2	29.0	1.80	4.1	37.0

CV = colour value; VA = humic acids; HS = humic substances; O = control group (no nitrogen added); U = urea added; C = calcium cyanamide added

3.9 correspond approximately to those which we obtained from previous tests on well rotted dung, compost, etc. In the *stalks*, the colour values behave in the same way as in the leaves except that they are barely half as high. As the colour values in the various test groups do not level out till later on (at the end of 40 weeks), it can be said that the stalks rot down much more slowly. The colour values of the *spindles* show the same tendency in the course of decomposition as the stalks and are also only half as high as in the case of the leaves. Even at the end of 40 weeks, there are considerable differences between test groups as a result of a very slow and irregular rotting down process. On the whole, the difference in colour values between leaves, stalks and spindles is greater than between test groups (use of nitrogenous fertilizers).

2. Humic acids (Table 1)

Humic acids occur in the soil both as an intermediary product of the formation of humus from fresh organic matter as well as a product of the breakdown of natural humus itself. They are therefore a measure of the change in humic substances. Compared with the "colour value", humic acids can be regarded as a much more stable humus fraction.

In the case of the *leaves* in the O and U₃ groups, considerable quantities of humic acid do not show up till the 15th and 10th week respectively, whereas in the other groups large amounts are present as early as the 5th week. The reason for the delay might be a certain lack of nitrogen in the case of the O group and the high NH₃ content and resulting pH change (pH 9–10) in the case of the U₃ group. The high humic acid values, however, in U₁ and C₃ (and to a lesser extent also in C₁) are due to the favourable NH₃ content arising from the breakdown of the urea and to the condensation and polymerization reactions caused by the cyanamide. Decomposition takes place much more slowly in the *stalks* than in the leaves, so that in groups U₁, U₃ and C₁ it is only at the end of 15–20 weeks and in the O group, only after 40 weeks, that considerable quantities of humic acid can be found. In the C₃ group, however, humic acid starts to form within a few weeks. At the end of 40 weeks, the humic acid content varies greatly in the different groups, in contrast to the colour values which have already levelled out. At this stage, the humic acid in the stalk material is roughly 3/4 of that in the leaf material. In comparison with the O group, the addition of nitrogen causes a more rapid decomposition and humification of the original material. The humic acid content in the rotting down spindles is very low. Even at the end of 40 weeks, when the

colour value is also low, only a few grams of humic acid are present. As in the case of the leaves and stalks, the highest values are to be found in the C₃ group. The main reason why the spindles do not rot down easily is their woodiness and low nitrogen content.

There is no connection between colour value and humic acid content. Thus, in the case of the leaves in the C₃ group, the colour values are no higher than in the other groups despite a much greater humic acid formation. Also in the control group, there are no measurable quantities of humic acid to compare with the colour values. This is because the colour value is not identical to the humic acid content but also includes the stages preceding the formation of humic acid such as lignic and humolignic acids, fulvic and hymatomelanic acids. These compounds are not condensed or polymerized to the point where they would be precipitated by acid, but they can already be darkly coloured. They are present in large quantities in the decomposing stalk material but are almost totally absent in the spindles.

3. Humic substances (Table 1)

The humic substances mainly include compounds which do not dissolve in acid or alkali and are hard to hydrolyze as well as higher molecular compounds, i. e. the both chemically and biologically very resistant humic complex. The latter also embraces those highly developed and very stable humic acids which occur almost only in the soil. Less stable humic acids, and particularly those in peat, compost and other decomposing products, are easily peptized in a sulphacetolysis mixture and can for the most part be precipitated out again by diluting the filtrate.

If fresh plant material is sulphacetolysed, there is generally a residue which arises from the condensation of lignin, protein and carbohydrates (artificial humification). In the leaves, it amounts to 3%, in the stalks to 1.1% and in the spindles to 5.8% of the dry matter. In the latter, a certain proportion of the lignin seems to be so strongly polymerized or woody that it remains insoluble in the reagent used.

Humic substances are formed at an earlier stage in the rotting down process and in larger quantities than humic acids. The most humic substances are formed in the *leaves*, where decomposition was by far the most advanced at the end of the experiment. After 25, and especially after 40 weeks, the same amount is present in all groups irrespective of the form and amount of nitrogen added. On average, barely half the quantity of humic substances are formed in the *stalks* compared with the leaves. At

the end of 40 weeks, there are in some cases still big differences between individual groups. The smallest amount is present in the O group and the largest amount in the C groups, despite equally strong decomposition in the U and C groups. In the stalks, therefore, which are low in nitrogen, calcium cyanamide has a much more favourable effect on the formation of humic substances as well as humic acids than in the case of the leaves. In the spindles, a residue of 5.8% or 17 g/sample is left after sulphacetolysis of the original material. This insoluble percentage, which is very difficult to break down, decreases in the O group up to the 15th week, i. e. this chemically and biologically very resistant material is broken down only very slowly. In the U 1 group, the decomposition corresponds roughly to the increase in newly formed humic substances. In the other N groups, the formation of humic substances is greater than the decomposition of the resistant, insoluble residue. The gradual breakdown is seen here, too, in an initial drop and subsequent increase in humic substance values. At the end of 40 weeks, the spindles have still decomposed relatively little despite considerable humic substance formation. The latter amounts to between 18 g (O group) and 37 g/sample (C₃). As the NH₃ losses are very great in the U groups at the start of the rotting down process (slow decomposition and nitrogen build-up, low water absorption capacity), decomposition and the formation of humic substances lag very much behind that of the calcium cyanamide groups. The slow formation of NH₃ from the cyanamide over a long period is better suited to the utilization of nitrogen by micro-organisms and to the hard job of breaking down the material than the sudden liberation of NH₃ from urea. Despite extremely low humic acid formation, therefore, almost as many humic substances occur in the spindles – at least in the C₃ group – as in the already much more decomposed leaves. Thus, under the same conditions, there are great differences in humic substance formation between the leaves, stalks and spindles. It depends on the structure (degree of woodiness) and on the nitrogen content or C/N ratio of the original material as well as on the form of nitrogen added.

Substance breakdown and humic substance formation

Table 2 compares substance breakdown (cellulose, pentosans, lignin) with humus formation at the end of 25 weeks' decomposition.

The cellulose and pentosan content in the *leaves* has already diminished to such an extent that it is roughly equivalent to that of a mineral soil. The organic matter which does not dissolve in 80% sulphuric acid, i. e. the lignin, decreases only slightly (from 42 to about 35 g) and has become

to a large extent insoluble in the sulphacetolysis mixture, i. e. has turned into humic substances. The high nitrogen content of these humic substances points to the fact that added nitrogen or nitrogenous metabolic products played a big part in their formation. A rough calculation shows that 20-25 % of the humic substances at most can have been derived from the protein. In reality, this percentage might be very much smaller, as lignin can also react directly with ammonia. At any rate, by far the greater proportion of lignin must have been converted into humic substances or

Table 2 - Cellulose, pentosans, "lignin-humus complex", humic substances and humic acids after rotting down for 25 weeks (g/sample)

	Original value	Test groups				
		O	U ₁	U ₃	C ₁	C ₃
<i>Leaves</i>						
Cellulose	94	6	7	8	8	13
Pentosans	93	10	10	12	10	12
"Lignin-humus complex"	42	36	35	35	35	41
Humic substances	(9)	31	37	37	38	34
Humic acids	-	13	13	12	11	24
% Nitrogen in humic substances	-	3.10	3.55	3.40	3.62	4.50
<i>Stalks</i>						
Cellulose	93	56	25	21	24	23
Pentosans	75	38	20	17	18	16
"Lignin-humus complex"	56	41	37	34	35	34
Humic substances	(3)	8	16	12	16	13
Humic acids	-	0	13	16	18	17
% Nitrogen in humic substances	-	3.77	3.97	4.28	4.08	4.60
<i>Spindles</i>						
Cellulose	107	72	42	64	52	55
Pentosans	107	71	46	63	46	39
"Lignin-humus complex"	57	45	43	45	48	48
Humic substances	(17)	17	22	27	25	30
Humic acids	-	2	2	2	1	4
% Nitrogen in humic substances	-	2.12	3.31	2.68	3.82	3.19

humic acids. Presumably, the formation of humic substances from lignin does not occur only out of its simple components (via polyphenol and quinone with further condensation), which in the first place produces humic acids, but also from larger molecular groups in the lignin. This could explain the early formation of insoluble humic substances which, for the most part, still contain lignin.

In the *stalks* which decomposed to a far lesser extent, the H_2SO_4 insoluble residue decreased much more (from 56 to about 35 g) during the 25-week rotting down period, producing a relatively low humic substance formation. The nitrogen content of the humic substances obtained from the stalks is higher than that from the leaves. This means that metabolic products played a bigger part in their formation. On the other hand, a considerable proportion of the lignin, in so far as it was still present in its original or little altered form, was released from the lignin-humin complex by sulphacetolysis. This applied especially in the case of the still only slightly decomposed O group. The lignin in the stalks appears to be easily soluble and very sensitive, as shown by the formation of large quantities of very darkly coloured, humic acid pre-stages which are not precipitated by acid (lignic acids, humolignic acids). This demethylated and oxydized, soluble lignin combines with protein or amino acids of the microbial metabolism to form lignin-protein complexes which are easily hydrolyzable from the chemical point of view but which are difficult to break down biologically, such as are found especially in cattle droppings and stable dung. In the O group in the case of the stalks, a considerable proportion was blocked in this way for a lengthy period by the very small amount of nitrogen present, thus greatly delaying microbial decomposition as well as the further development of humic acid pre-stages. In the N groups, however, the conversion of lignic and humolignic acids into lignohumic and humic acids, i. e. condensation and polymerization together with nitrogenous metabolic products, occurred very much earlier and faster than in the O group depending on the amount of nitrogen added.

The spindles, which are low in nitrogen, are broken down more slowly than the stalks. At the end of 25 weeks, they still contain 40-70% of their original cellulose content and 39-71% of their original pentosan content. The H_2SO_4 insoluble residue (lignin-humus complex) alters to a lesser extent in the spindles than in the stalks (decreases by 20-35%). The formation of humic substances, on the other hand, is almost twice as high in the spindles. A greater proportion of the lignin, therefore, has become soluble and been incorporated into the humic acids or their pre-stages. The lignin in the spindles, however, has become much less soluble as a result of the rotting down process (very low formation of humic acids or their

pre-stages) and a greater proportion (compared with the stalks) has been converted into humic substances. The lower nitrogen content of these humic substances also points to the fact that the lignin played a bigger part in their formation. In the H_2SO_4 insoluble lignin-humin complex of the spindles and stalks, practically the same amount of lignin is released by sulphacetolysis. The lignin in the spindles is presumably present in a higher polymer, very trapped form which is therefore not so easily soluble and immediately converted into humic substances when liberated with nitrogen or nitrogenous metabolic products. Lignin appears to be present in the stalks in a low molecular, mobile state and to become straightaway soluble and very sensitive after the breakdown of the carbohydrates. A lack of nitrogen or protein can inhibit further decomposition (protein blockage, trapping of the cellulose).

The two main rotting processes, i. e. the breakdown of organic matter and the simultaneous formation of humus, are shown in Figures 1 and 2. The leaves, especially in the C 3 group, represent an ideal case of decomposition. The rapid cellulose breakdown corresponds to a very early and rapid formation of humic substances or humic acids. In the control group, this does not start until the cellulose content has reached 20-30%, whereas in C 3, it starts when 30-40% of the original cellulose content is still present.

The low-nitrogen stalks decompose less satisfactorily when no nitrogen is added (O). Both curves do not move. Decomposition is seriously inhibited; only small quantities of humic substances and practically no humic acids are produced. Adding nitrogen (C 3) not only greatly accelerates the rotting down process and increases the formation of humic substances but, what is more, also produces a large quantity of humic acid.

The spindles, which are even lower in nitrogen and more difficult to rot down, show a similar curve picture in group O; humic substances, but no humic acids, are formed. The different curve development is due to the fact that the humic substances include lignin which is still polymerized and woody and which is broken down in the early stages of decomposition faster than humic substances are built up. The humic substance curve behaves differently in C 3: as a result of adding nitrogen, the build-up of humic substances initially outweighs the breakdown of the lignin content which decomposes with difficulty (rise of the curve). Then follows a more rapid breakdown or dissolving of the lignin and, after this change, again an increase in humic substance formation (Table 1). Humic acids occur only in very small quantities even when large amounts of nitrogen are added. The formation of humic substances in the spindles increases rapidly (especially in C 3) already when 60-70% of the cellulose and pentosans

humic acids. Presumably, the formation of humic substances from lignin does not occur only out of its simple components (via polyphenol and chinone with further condensation), which in the first place produces humic acids, but also from larger molecular groups in the lignin. This could explain the early formation of insoluble humic substances which, for the most part, still contain lignin.

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x—x = Cellulose, •—• = Humic substances, ◊—◊ = Humic acids
 Original quantity: 300 g org. matter/sample

a) Control group (no nitrogen added)

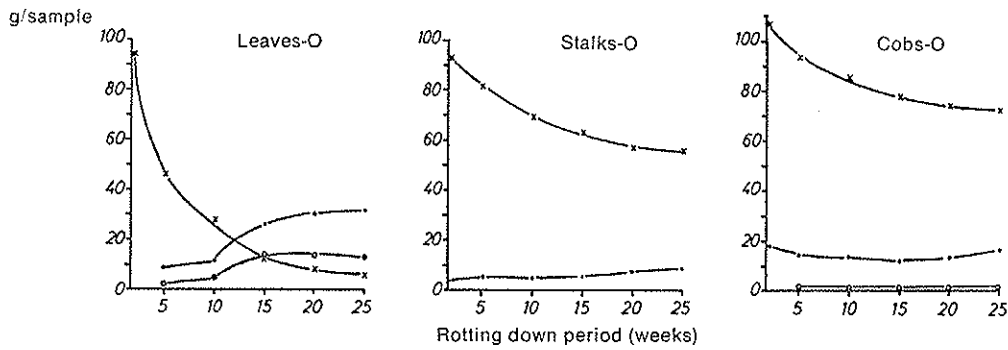


Figure 1 – Cellulose breakdown and humus formation during the rotting down of maize straw when no nitrogen is added

x—x = Cellulose, •—• = Humic substances, ◊—◊ = Humic acids
 Original quantity: 300 g org. matter/sample

b) 1.5 % Nitrogen added in the form of calcium cyanamide (C 3)

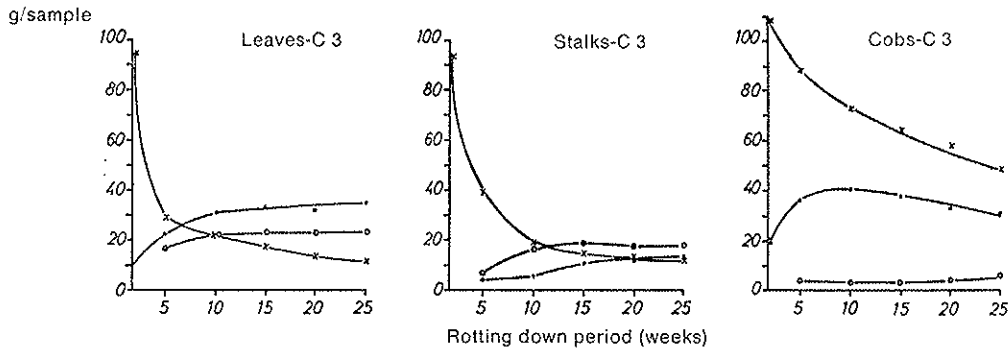


Figure 2 – Cellulose breakdown and humus formation during the rotting down of maize straw when nitrogen is added

and consequently less nitrogenous metabolic products, are still present. The lignin might here be the original material and basis of the humic substance formation more than in the leaves and stalks (lower nitrogen content [Table 2]).

The decomposition of maize straw depends on the following factors:

- the structure and texture (physical and mechanical properties) of the various plant parts;
- the percentage and nature of those substance groups which decompose easily or with difficulty;
- the nitrogen content.

The rotting down process is considerably accelerated by the use of nitrogen, depending on the form and amount added, and more humic substances with a richer nitrogen content are produced. The humic nitrogen increases considerably when large amounts of nitrogen are added, especially in the form of calcium cyanamide, as the breakdown of organic matter by microorganisms is slowed down by the presence of cyanamide. The cyanamide also seems even to promote condensation and polymerization and hence the formation of humic substances. The direct effect of the calcium is not immediately noticeable, as the reaction is always neutral to slightly alkaline as a result of the ammonia produced. In the open air or in the fields the alkaline reaction resulting from the application of large quantities of urea would, in all likelihood, produce scarcely any detrimental effect. The addition of 0.5 % nitrogen in the case of leaf material and about 1 % nitrogen in the case of stalks and spindles is sufficient to accelerate the decomposition of maize straw and produce a good quantity of high quality humus.

REFERENCES

- [1] AMBERGER, A. / A. WAGNER: Stoffliche Veränderungen bei der Rotte von Maisstroh (Substance Changes during the Decomposition of Maize Straw). In: Landwirtschaftliche Forschung 19. (1965) Sonderheft; pp. 3 to 11.
- [2] AMBERGER, A. / A. WAGNER / F. RASSADI: Über den Abbau der organischen Substanz bei der Verrottung von Maisstroh (The Breakdown of Organic Matter during the Decomposition of Maize Straw). In: Bayerisches Landw. Jb. 48 (1971); pp. 3-22.
- [3] AMBERGER, A. / A. WAGNER / F. RASSADI: Über die Festlegung des Stickstoffes bei der Verrottung von Maisstroh (Fixation of the Nitrogen during the Rotting Down of Maize Straw). In: Bayerisches Landw. Jb. 48 (1971); pp. 190-205.

- [4] WAGNER, A.: Untersuchungen über die nach mehrjähriger organischer Düngung gebildeten Huminsäuren (Tests on the Humic Acids Formed as a Result of Many Years of Organic Manuring). Dissertation Stuttgart-Hohenheim 1958.
- [5] Methodenbuch des Verbandes Deutscher Landw. Untersuchungs- und Forschungsanstalten; 1955, Bd. I, 46. Jg. Radebeul und Berlin: Verlag Neumann.