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CHLOROPHYLL-TYPE COMPOUNDS IN SOIL

III. THEIR SIGNIFICANCE IN ARABLE SOILS

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INTRODUCTION

Chlorophyll-type compounds in soil come mainly from fresh plant material and from faeces of grazing animals [4]. They decompose in soil at rates that depend on the history of the material, on the activity of the plant tissue enzymes, and on soil conditions, particularly those affecting the microflora [5]. Consequently, their amounts in arable soils should differ according to their properties and recent histories. The ability of soils to supply nitrogen to plants is often associated with recent additions of plant materials which cannot be readily identified, soils containing plant residues that may release nitrogen are not readily distinguished from those without such residues.

The work described in this paper was done to find whether chlorophyll-type compounds in arable soils might be used to indicate the content of undecomposed plant material and, whether this was relative to their ability to supply nitrogen to crops.

ANALYTICAL METHODS

The methods to measure chlorophyll-type compounds were as already described [4]; 25 g of each soil were extracted with 75 ml of 90 per cent aqueous acetone. Absorption readings were made on extracts in 4-cm glass cells at 665 mμ (used as the absorption peak); background absorption was allowed for by subtracting the mean of the readings at 630 and 750 mμ.

Absorption readings for the 24 soils in the second experiment were examined to find how well the analytical methods developed earlier [4] measured

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chlorophyll-type compounds. Readings were taken on extracts of the 24 soils from 350 to 760 mμ at 10-mμ intervals with an additional reading at 665 mμ. Absorption by extracts from 3 soils was greater at 660 mμ than at 665 mμ and from one soil, greater at 670 mμ; however, the reading at 665 mμ was used as the peak.

The minimum absorption reading preceding 665 mμ was always at 630 mμ. The mean of the readings at 630 and 750 mμ would not correctly represent the background absorption at 665 mμ if it decreased linearly over this range. Background absorption values close to 665 mμ were therefore calculated from readings at 630 and 740 mμ, assuming absorption decreased linearly; the chlorophyll units (CU) calculated using these values were correlated ($r = 0.99$) with those using the means of the absorption readings at 630 and 750 mμ. Consequently, the mean of the readings at 630 and 750 mμ was considered an adequate measure of background absorption.

EXPERIMENTAL AND RESULTS

Quantities of chlorophyll-type compounds found in field-rotations

Soil samples were taken 0–9 inches deep from the Ley–Arable rotation experiments on Highfield and Fosters Field at Rothamsted and on Stackyard Field at Woburn; samples were taken from a rotation that included a ley and from an arable rotation.

The ley–arable rotation consists of three years of grazed grass–clover ley followed by three arable crops. The leys are ploughed in autumn. Soil samples were taken in May, 1962, from plots in the three stages of arable cropping at 0–1, 1–2 and 2–3 years after ley.

The arable rotation consists of a one-year grass-clover ley cut for hay, followed at Rothamsted by 5 arable crops and at Woburn by 4 arable crops. The plots were sampled the year before 1 year-ley was to be grown. Soil samples were taken in May, 1962 from sub-plots given farmyard manure (FYM) at 12–15 tons per acre once in the course of each of the rotations (1 year before sampling at Rothamsted and 3 years before at Woburn) and from sub-plots not given FYM.

TABLE 1

Quantity of chlorophyll-type compounds in some plots of field experiments testing different crop sequences, expressed as CU/100 g soil

Location	Field	Ley–arable: Years from ploughing the ley			Continuous arable	
		0–1	1–2	2–3	With FYM	Without FYM
Rothamsted	Highfield	6.4	5.6	3.8	4.9	5.0
	Fosters				7.0	3.4
Woburn	Stackyard	2.7	3.0	2.3	2.7	1.6

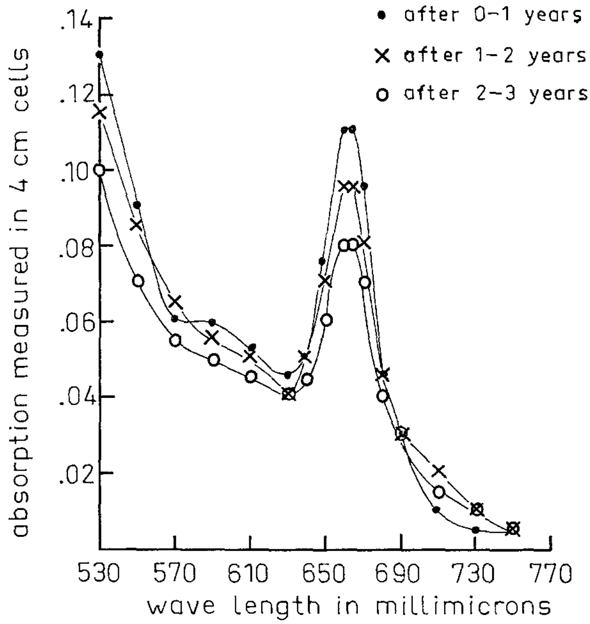

Fig. 1. Absorption spectra of acetone extracts of soil after ploughing ley. Highfield.

Chlorophyll units (the measure of chlorophyll-type compounds as used here) decreased slowly under arable cropping after ley in Highfield soil at Rothamsted, but not in Woburn soil (Table 1). Figure 1 shows absorption spectra of acetone extracts of the Rothamsted soil.

Plots with and without FYM on Highfield had the same chlorophyll units (CU), whereas on Fosters Field at Rothamsted, and Stackyard Field at Woburn, soil with FYM had the more CU.

The relationship between chlorophyll units in soil and the nitrogen available to plants

The soils used in this experiment were those used by Gasser [1 3] to study "available" soil nitrogen and results of pot experiments and other measurements are quoted from him.

The soils were sampled 0–9 inches deep in 1958 and 1959 from farms within 25 miles of Rothamsted; previous cropping was usually arable but some soils were from ley-arable rotations. Organic C was determined by wet oxidation, total N by the Kjeldahl method and pH was measured in a 1 : 2.5 soil : water suspension. The increases in mineral-N content (abbreviated to Δ mineral-N) were measured after incubating re-wetted air-dry soils at 25°C for 21 days. The chlorophyll-type compounds (expressed as chlorophyll units) were extracted in 1963; the soil samples were stored air-dry from 1959 to 1963.

Measurements of CU were compared with dry matter and N-uptake of ryegrass measured in glasshouse pot experiments; the soils received a basal PK fertilizer but no N. Both the dry matter and N-uptake results are totals of 3 cuts.

Table 2 shows measurements used in all correlations and regression analyses. Table 3 shows that in the 1958 samples of the four soil measurements chlorophyll units (CU) were best related to N-uptake

TABLE 2

Soil characteristics and data from the pot experiment on ryegrass, used in correlations and regressions

Soil	Soil characteristics					Ryegrass data	
	CU/ 100 g soil	Δ Mineral N air dry ppm	Total N %	Organic C %	pH	N-uptake mg/pot	D.M. g/pot
1958							
1	1.88	13	0.156	1.90	7.9	28	2.67
2	2.40	28	0.178	1.66	7.9	38	3.58
3	2.48	35	0.215	2.30	8.0	42	3.56
4	2.48	11	0.094	0.92	6.0	26	2.72
5	2.62	27	0.168	1.90	7.9	39	3.46
6	2.78	18	0.195	1.96	7.9	41	3.57
7	2.85	40	0.236	2.33	7.9	42	4.02
8	4.12	26	0.143	1.62	6.4	45	4.22
9	5.48	35	0.242	2.32	8.0	59	4.76
10	9.60	110	0.336	4.61	6.8	74	6.13
1959							
11	1.88	26	0.177	1.90	8.0	63	4.40
12	2.40	23	0.191	1.80	8.0	54	4.30
13	2.78	41	0.210	2.01	8.0	70	4.94
14	3.15	39	0.205	2.10	7.6	56	4.36
15	3.38	45	0.199	2.27	7.6	76	5.64
16	3.38	39	0.229	1.91	8.0	71	4.95
17	3.52	30	0.174	1.65	6.6	52	4.03
18	3.60	26	0.139	1.98	7.9	45	3.53
19	3.60	30	0.177	1.95	7.6	71	4.97
20	3.75	38	0.157	1.62	7.4	66	4.88
21	3.75	31	0.173	1.55	8.1	62	4.66
22	4.65	31	0.162	1.80	7.0	65	4.87
23	5.85	43	0.264	2.36	7.8	82	6.03
24	7.35	83	0.494	4.15	6.3	119	8.18

(r = 0.93) and yield (r = 0.94) of ryegrass, whereas in the samples taken in 1959 CU had the smallest correlation coefficients (r being 0.77 and 0.81). On combining results for the samples taken in 1958 and 1959 (and adjusting the differences between the mean yields and mean N-uptake in the two years), the correlation of N-uptake and CU (r = 0.79) was little less than with Δ mineral-N (r = 0.82). Figure 2 shows the relationship between CU and N-uptake. The correlation coefficient for organic C with N-uptake was 0.83, and for total N with N-uptake 0.90.

TABLE 3

Total correlation coefficients between nitrogen uptake and yield of ryegrass and the soil measurements

Soil measurements	Nitrogen uptake by ryegrass			Yield of ryegrass		
	1958	1959	1958 and 1959	1958	1959	1958 and 1959
CU/100 g soil . . .	0.93**	0.77**	0.79**	0.94**	0.81**	0.84**
Δ Mineral-N. . . .	0.88**	0.92**	0.82**	0.90**	0.93**	0.85**
Total N	0.89**	0.91**	0.90**	0.86**	0.91**	0.89**
Organic C	0.87**	0.87**	0.83**	0.86**	0.88**	0.84**

Significant at ** P = 0.01

Although Table 3 shows that each of the four soil measurements was closely associated with N-uptake, this does not necessarily imply cause and effect relationships. Table 4 shows that the soil measurements, CU, Δ mineral-N, total N and organic C were all correlated at the one per cent probability level with one another in all possible comparisons. However, chlorophyll units correlated best with Δ mineral-N. Table 5 shows partial correlations between pairs of soil measurements, holding the other two constant. Of the partial corre-

TABLE 4

Total correlation coefficients between soil measurements

	CU	Δ Mineral-N	Total N	Organic C	pH
CU	—	0.86**	0.71**	0.81**	−0.43*
Δ Mineral-N	0.86**	—	0.82**	0.93**	−0.28
Total N . .	0.71**	0.82**	—	0.89**	−0.17
Organic C .	0.81**	0.93**	0.89**	—	−0.19
pH	−0.43*	−0.28	−0.17	−0.19	—

Significant at * P = 0.05; ** P = 0.01.

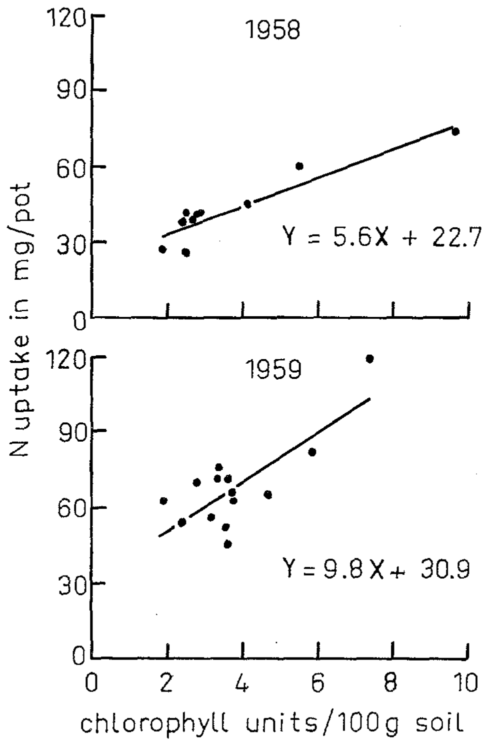

Fig. 2. Regression of nitrogen uptake by ryegrass on chlorophyll units in the soil.

lations involving CU, only that with Δ mineral-N was significant. The significant partial correlations among the other three soil measurements were between Δ mineral-N and organic C and between

TABLE 5

Partial correlation coefficients between two soil measurements holding the other two constant		
Measurements compared	Measurements held constant	Partial coefficients of correlation
CU and total N	Δ Mineral-N, organic C	−0.03
CU and organic C	Δ Mineral-N, total N	0.06
CU and Δ mineral-N	Organic C, total N	0.50*
Δ Mineral-N and total N	CU, organic C	−0.01
Δ Mineral-N and organic C	CU, total N	0.63**
Organic C and total N	Δ Mineral-N, CU	0.59**

Significant at * $P = 0.05$; ** $P = 0.01$.

organic C and total N. These partial correlation coefficients suggest that there was a 'real' relationship between CU and Δ mineral-N; close association between Δ mineral-N and organic C, and in turn, between organic C and total N, was responsible for the correlations between CU and organic C and between CU and total N (Table 4).

The partial correlation coefficients of Δ mineral-N with both CU and organic C were significant, so Δ mineral-N probably varied independently with both the CU and the organic C of the soils. The regression equation of Δ mineral-N (Y) on CU (X_1) and organic C (X_2) was $Y = 3.7X_1 + 18.3X_2 - 16.1$. The t-values for the partial regression coefficients of Δ mineral-N on CU and on organic C were significant at the five and one per cent levels of probability respectively (the same as for the partial correlation coefficients).

DISCUSSION

Chlorophyll-type compounds in soil were significantly correlated ($P = 0.01$) with nitrogen uptake by ryegrass grown in pots. However, the chlorophyll-type compounds themselves could have made only an infinitesimal contribution to the total nitrogen needed by the ryegrass plants, because they amounted to only from 0.5 to 3.0 pounds per acre. Therefore, the chlorophyll units (CU) indicate quantitatively the presence of other compounds that decomposed to supply nitrogen to the plants.

It was found earlier [5] that 75–80 per cent of the chlorophyll-type compounds in ground grass added to clay soil (in a field experiment at Rothamsted) were lost after 72 days. When green materials like these decay, they release nitrogen and CU remaining in soil may be roughly proportional to the amounts of nitrogen yet to be released from them. As changes in mineral nitrogen on incubating re-wetted air-dry soils were related to both CU and organic C, it seems that CU do indeed represent a fraction of plant remains containing easily mineralizable nitrogen.

Earlier laboratory incubation experiments showed that the residual chlorophyll-type compounds in soil decomposed slowly [5]. This work shows a slow loss of chlorophyll-type compounds in the field over 3-years after ploughing a ley at Rothamsted; the soils used in the pot experiment contained few CU and their cropping histories (mostly arable) suggests that the chlorophyll-type compounds were mainly in a slowly decomposing residue. The residual CU found is thought to represent either a resistant unidentified chlorophyll-type compound, or pheophytin that is protected in some way against microbial attack (unprotected pheophytin is easily attacked [5]).

The group of soils used were those readily obtainable and were not altogether suitable for relating CU and nitrogen supply. The soils were from arable rotations and the samples had been stored for several years which obscures [3] relationships between laboratory measurements of nitrogen fractions and the power of soil to supply nitrogen to crops. If a series of soils containing recently ploughed ley or green manure or added organic manures had been available for the pot experiments that Gasser [1] did relationships between CU and available nitrogen might have been better. Despite this nitrogen uptake by ryegrass was nearly as well correlated with CU as it was with Δ mineral-N. Using these and other soils, Gasser and Williams in 1963 [2] reported that Δ mineral-N was the most consistently correlated of several soil measurements with the portions of the soil nitrogen that is available to plants. This paper suggests that CU might be used to predict nitrogen fertilizer requirements of crops. Advantages are that CU can be measured rapidly, the measurement is free from interferences and the soil samples can be handled in any convenient way.

SUMMARY

The quantities of chlorophyll-type compounds in a group of 24 arable soils were significantly correlated with nitrogen uptake ($r = 0.79$ and yield ($r = 0.84$) of ryegrass grown in pots in the glasshouse. Partial correlation coefficients showed the amounts of mineral N released on incubating the re-wetted air-dry soils were related both to chlorophyll units (measure of chlorophyll-type compounds) and organic C. The chlorophyll units (CU) apparently represented some fraction of organic matter that liberated inorganic nitrogen, because the quantities of chlorophyll-type compounds in the soil were too small (only 0.5–3.0 pounds per acre) to supply this nitrogen. It is likely that the compounds were a residue that resisted decomposition. The CU in soil that had been ploughed from ley decreased very slowly during 3 years of arable cropping.

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