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EVALUATION OF THE NUTRIENT STATUS OF WHEAT PLANTS

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Evaluation of the nutrient status of wheat plants

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Summary

The balance of the principal ionic constituents in plants is reviewed in relation to the requirements for each of the elements, and for total accumulation as reflected in the total cation content (C), the inorganic anion content (A) and the organic anion content (C-A).

The balance in young wheat plants is investigated by means of adding various combinations of the principal ions to the soil.

A summary of tests on adequacy of supply with the principal nutrient ions and of accumulation of cations, inorganic anions and organic anions, based on critical values for contents in the plant material, is presented for diagnosing the nutrient status in relation to capacity for maximum growth.

Introduction

Evidence was obtained that the grain yield is often increased when the early growth of wheat plants is retarded by a limited supply with nitrogen. An example is summarized in Fig. 1, where straw and grain yields have been plotted against the amount of nitrogen supplied in the fertilizer. The solid lines show the relation for single applications at sowing of 0, 60, 90 or 120 kg N/ha. The broken lines refer to an application at sowing of 30 kg N/ha, and supplements of 30, 60 or 90 kg N/ha applied at the beginning of shooting.

The split applications of nitrogen led to lower straw and higher grain yields compared with the single dressings at sowing. Since the same was observed on CCC-treated plots without lodging suppression of lodging by split applications could not have been the only cause of the improved grain yields. Apparently, moderate dressing with nitrogen at the onset of growth, followed by supplementary dressings at shooting, may shift dry matter distribution in favour of the grain yield.

However, regulation of the time course of growth by split applications of nitrogen can be satisfactory only if at the time of the second application the other conditions are adequate to support maximum growth required after shooting.

One of these other conditions concerns the supply with major elements from the soil, as reflected in leaf composition with respect to K, Na, Mg, Ca and NO₃, Cl, H₂PO₄, SO₄. A balance-sheet of these ions also yields the organic salt content (C-A) as the difference between the equivalent sum of inorganic cations (C) and the inorganic anions (A) in the tissues. Previous research showed that maximum growth may be

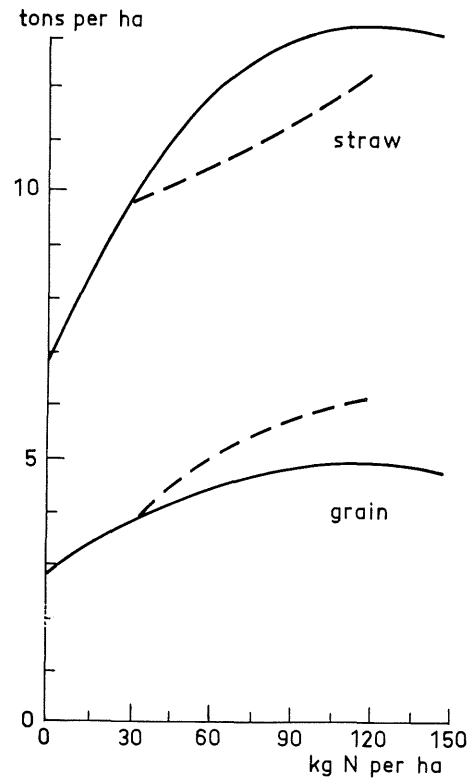


Fig. 1 Straw and grain yields of wheat, grown in the field with varying rates of N applications (abscissae). Solid lines: all N applied at sowing. Broken lines: 30 kg N/ha at sowing, the remainder applied on July 6 at the beginning of shooting. Data of Jonker and de Jong (1966).

obtained only if the organic salt content is at the normal level (de Wit et al., 1963). In view of the perspectives of nitrogen-controlled growth it was considered useful to examine the ionic balance of wheat in the pre-shooting stage, to evaluate the normal organic salt content and to attempt to develop a key for diagnosing the nutrient status.

It seems useful first to review the main characteristics of the ionic balance and its response to different fertilizer formulas.

Among the anions phosphate is present as inorganic phosphate and in the form of phosphorylated organic compounds, but the ionic form of all phosphates may in good approximation be considered as H_2PO_4^- . Phosphate absorbed distributes over the nucleoproteins and phospholipids, the TCA-soluble organic phosphates and free orthophosphate. In ryegrass the nucleoproteins (plus phospholipid) phosphorus was 0.025 gram atoms P per gram atom of protein nitrogen, the TCA-soluble organic phosphate 0.03 gram atoms P per kg of dry plant material, and the remainder was free orthophosphate (Dijkshoorn and Lampe, 1961). According to these figures the organic-phosphate content of plants with 3 gram atoms of organic nitrogen (mainly protein-nitrogen) will be $3 \times 0.025 + 0.03 = 0.1$ gram atoms P per kg dry matter.

From data on the relation between plant growth and phosphate fractions (Sokolov, 1945) it can be inferred that the phosphate requirement for maximum growth is about twice the amount transformed into organic phosphates. The plant need will then be

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0.2 gram atoms. Expressed as the more convenient meq. units, this requirement for growth will be 200 meq. H_2PO_4 per kg dry plant material. Therefore, values for phosphate greater than 200 meq./kg dry matter indicate adequate phosphate supply for plants with nitrogen below 3 gram atoms of organic N per kg dry matter.

Sulphate absorbed in excess of its metabolic conversion to non-ionic organic sulphur is retained by the tissues as inorganic sulphate. With shortage in the supply, the sulphate content drops by dilution and metabolic consumption, and plants deficient in sulphur have very low sulphate contents of about 12 meq./kg dry matter (Dijkshoorn and van Wijk, 1967). Since some reserve in the tissues seems desirable, inorganic-sulphate values of less than 20 meq./kg dry matter should be considered critical for growth.

The chloride requirement for growth is very low (Ulrich and Ohki, 1956) and is always met in soil-grown plants. When supplied in the fertilizer, chloride may accumulate in high concentrations in the tissues.

When nitrogen is in adequate supply, the organic-nitrogen content of young wheat plants is in the range of 3000 to 4000 milligram atoms of N per kg dry matter. It originates from absorbed nitrate or ammonium and is conveniently expressed as the meq. of these ions which give the same numerical value as milligram atoms of N. Ammonium absorbed is nearly completely transformed into organic N, but if nitrate acts as the source of nitrogen some of it is retained in the ionic form by the tissues. If during growth nitrate becomes exhausted in the supply there is a subsequent fall in nitrate concentration in the tissues as a result of metabolic consumption and dilution (Dijkshoorn, 1958). In grass, a nitrate content of 100 meq./kg dry matter is indicative of sufficient supply (van Burg, 1966), but depending on plant species and ion supply the nitrate content is often much higher.

Of these absorbed anions, some of the nitrate and sulphate, and all of the chloride and phosphate is retained in the ionic form. The quantities are obtained by analyses on nitrate, chloride, total phosphate and (inorganic) sulphate in the plant material. The sum of meq. $NO_3^- + Cl^- + H_2PO_4^- + SO_4^{2-}$ is called the inorganic anion content A. For practical reasons, this value includes total phosphorus. The organic phosphates are thus grouped under the anions A, since their ionic form is similar to orthophosphate in the range of tissue pH. The organic anions of carboxylate character (see below) are then placed in one group of carboxylates only.

Of the cations absorbed, potassium, sodium, magnesium and calcium contribute in significant amounts to the final cation content of the tissues. Irrespective of their distribution at the cellular level, their dissolved or undissolved state or linkage with other constituents, they contribute as metallic cations to the formation of the various salts present in the tissues. The sum of meq. $K^+ + Na^+ + Mg^{++} + Ca^{++}$ represents the total cation content C of the plant material.

In plant tissues there is always an excess of the cations (C) over the inorganic anions (A) and this excess is balanced by organic anions. It has been found that the organic salt content (C-A), inferred from the balance of inorganic ions, consists of carboxylates only, and originates mainly from the common organic plant acids, being recovered as malate, citrate, oxalate etc. (van Tuil, 1965).

Previous research on the relation between growth and the ionic balance showed that gramineous plants tend to maintain an organic salt content (C-A) of a certain value which, depending on the species, lies in the range of 800-1000 meq./kg dry matter. This organic salt content has been named the normal.

With adequate nutrient supply the normal organic salt content is maintained during

the vegetative growth of grass plants. It can be reduced to a subnormal level by a change in the nutrient supply. It appeared that with such fertilizer formulas maximum growth was never obtained. Apparently, maximum-growth capacity requires a nutritional status which, among other characteristics, will enable the plant to maintain its normal organic salt content. A fall in (C-A) to values less than the normal is a sign of nutritional stress caused by shortage or excess of nutrients which affect the (C-A) (de Wit et al., 1963).

One of the conditions required for maintaining the normal (C-A) content is a sufficient uptake of cations (C). According to data of Lundegårdh (1951), Broeshart and van Schouwenburg (1961), Garner et al. (1930), Key et al. (1962), and others, the minimum contents of magnesium and calcium are in the order of 50 and 100 meq./kg dry matter, respectively, and values above 100 meq./kg dry matter may be considered adequate for the specific requirements. For potassium, the minimum content is about 200 meq./kg dry matter (van Tuil, 1965).

Compared with the other cations potassium is most readily absorbed by gramineous plants as the main constituent of the cation content C. Since potassium in the tissues can be made to vary considerably, the total cation content C is often much interfered with by the presence or absence of sufficient potassium.

The plant need for cations is conditioned by its efforts to maintain the normal organic salt content (C-A) and depends on the anion content A. If the latter is raised by an increased accumulation of inorganic anions as e.g. chloride, nitrate, more of the cations are required to keep (C-A) at the normal level. Since potassium aids in the uptake of cations to a considerable extent, the potassium requirement will depend to a large extent on the inorganic anion content, and there will be no distinct potassium concentration of adequacy in the tissues valid under different conditions of ionic supply.

It is therefore common experience that growth can often be improved by an increase in potassium from any initial value in the range of, say, 200 to 1000 meq. K/kg dry matter, provided that (C-A) is less than the normal, and is raised with the increase in potassium. The plant need for potassium is therefore dependent on its inorganic anion content, and is conditioned by its particular ability to readily supply extra cation equivalents by uptake, if required for the further maintenance of the normal (C-A) content.

Some gramineous species are able to accumulate sodium in large amounts but they do so only when they are deficient of potassium. In this case, potassium in the tissues may fall below the minimum content of 200 meq./kg dry matter, as a result of further dilution by extra growth initiated by sodium (Lehr and Wybenga, 1958; de Wit et al., 1963).

At sufficient supply a large proportion of ion absorption is occupied by the nitrogenous ions nitrate or ammonium.

With nitrate the uptake of anions proceeds in large quantities and the salt cation uptake is enhanced to balance the anion uptake. The nitrate absorbed is for the greater part metabolized. Only some of it is retained in the ionic form as unchanged nitrate which contributes to the final inorganic anion content in the tissues. The result is that (C-A) accumulation is enhanced. For this reason nitrate nutrition will effectively support maintenance of the normal organic salt content.

With ammonium a large quantity of salt cations, including ammonium, is absorbed and this will promote the uptake of anions to balance the increased cation uptake. The ammonium absorbed is metabolized and since the uptake of potassium, sodium,

cerns maize plants grown with varying supplies of micronutrients. These factors may also be related to the supply with major elements included in the ionic balance, as e.g. sulphate and phosphate shortage, which interfere with growth but not with the (C-A) content. Their status can be inferred from the results of chemical analysis.

The final make-up of the balance gives a systematic review of the contents of each of the elements which can be compared with the critical values. The additive values for C, A and (C-A) can be used for defining the status of (C-A) with respect to the normal value. Sufficient knowledge on the effect of added fertilizers on composition of the plant species concerned may render it possible to suggest changes in nutrition which may improve the ionic balance.

For young wheat plants the present experience has been summarized in Table 8, to be used as a key for the nutritional status. The reader should bear in mind that for values close to the limits the alternative course should also be followed, and an intermediate conclusion should be drawn. For instance, treatment 9 of experiment 1 shows a too high anion content A caused by excess chloride. But (C-A) is close to the limit and the alternative course without excess chloride is also applicable, which shows that the excess was not serious. A similar conclusion applies to the data of treatment 7.

When the supply changes from ammonium to nitrate, a transition stage may be encountered where the (C-A) content is low and the nitrate content high. Table 8 reveals this condition when applied to the data of e.g. treatment 15 of exp. 4, referring to plants grown in the early season with ammonium nitrate as the fertilizer.

In many cases there may be other evidence that the soil used supplies sufficient calcium, magnesium, phosphate and sulphate for maximum growth. If so, the procedure may be simplified by confining plant analysis to nitrate, potassium and ash-alkalinity, and calculating (C-A) by subtracting nitrate from ash-alkalinity (van Tuil et al., 1964). When total nitrogen is also determined the nutrient status can be examined for potassium, nitrogen and (C-A). Test 5 cannot be made, but when potassium values are greater than 1100 meq./kg dry matter it is likely that the anion contents A are too high and correspondingly have increased the plant need for cation uptake.

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Table 7 Plant composition in experiment 4. The fields were sampled by removing 100 wheat plants from each treatment in the pre-shooting stage.

No.	K	Na	Mg	Ca	C	NO ₃	Cl	H ₂ PO ₄	SO ₄	A	N _{org}	(C-A)
1	913	34	208	275	1490	157	180	92	280	709	3257	781
2	907	31	148	214	1300	186	152	96	257	691	3335	609
3	849	28	148	225	1250	105	144	114	261	624	3509	626
4	1072	27	131	230	1460	98	307	133	256	794	3945	666
5	1150	23	107	230	1510	181	99	161	267	708	4205	802
6	1176	23	140	214	1553	152	228	120	222	722	3526	831
7	1021	31	164	180	1396	53	310	133	267	763	3618	633
8	1163	29	171	252	1615	176	262	143	296	877	3781	738
9	1166	33	67	239	1505	170	180	140	319	809	3573	696
10	1100	33	156	264	1553	264	214	173	292	943	3622	610
11	1019	41	164	259	1483	281	178	133	271	863	3676	620
12	960	40	173	249	1422	184	240	134	298	856	3737	566
13	945	46	173	262	1426	222	240	143	264	869	3671	557
14	989	38	—	—	—	166	234	143	175	718	3591	—
15	930	36	173	264	1403	194	194	123	210	721	3520	682
16	926	28	173	252	1379	220	254	182	242	898	3737	481
17	881	30	173	234	1318	167	254	95	251	767	3447	551
18	860	32	181	264	1337	236	234	119	276	865	3585	472

Table 8 Test table for the nutritional status of wheat

Test	meq./kg d.m.	Comments	go to:	
1	NO ₃	above 100	sufficient N for maximum growth, but undesirably high in the pre-shooting stage	2
		below 100		3
2	(C-A)	above 800		4
		below 800		5
3	(C-A)	above 800	moderate N shortage, but desirable in the pre-shooting stage	4
		below 800		8
4	K	above 800		13
		500-800	K shortage if N is applied after shooting	13
5	A	below 500	K shortage	13
		above 1500	too high, the anions NO ₃ , Cl, H ₂ PO ₄ , SO ₄ are unnecessarily high if above 500, 200, 300 and 100	6
		1500-800		6
6	K	below 800		7
		above 1100		13
		1100-800	if A cannot be reduced, (C-A) may be increased by raising the K level	13
		below 800	K shortage	13
7	K	above 800		13
		below 800	K shortage	13
8	N _{org}	above 3000		9
		3000-2000	N shortage, but desirable in the pre-shooting stage	10
		below 2000	N shortage	10
9	(C-A)	above 500	history of ammonium uptake	11
		below 500	too much ammonium absorption	11
10	K	above 900		12
		below 900	K shortage	12
11	K	above 600		12
		below 600	K shortage	12
12	Cl	above 500	too much Cl	13
		below 500		13
13			H ₂ PO ₄ , SO ₄ , Mg, Ca shortage if below 200, 20, 100, 100 meq./kg	13

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magnesium and calcium, which compose the final cation content C in the tissues, is depressed by the large uptake of ammonium, the accumulation of organic salts (C-A) becomes strongly reduced compared with nitrate nutrition. For this reason ammonium fed plants have (C-A) contents much less than the normal (van Tuil, 1965). In this way the balance-sheet reflects the form of nitrogen absorbed. When organic N is high, nitrate is present and the (C-A) content is normal, nitrate must have been in adequate supply. With high organic N and nitrate, a subnormal (C-A) content may have been caused by too low potassium or by too high chloride contents. When organic N is high, nitrate is absent and (C-A) is less than the normal even at moderate chloride contents, nitrogen must have been absorbed as ammonium. If in this case, nitrate is present, but the (C-A) content is still below the normal, plant nutrition may have changed recently from ammonium to nitrate as a result of nitrification or refertilization.

Many fertilizer formulas will be able to supply sufficient quantities of each of the elements for maximum growth, but will result in a subnormal (C-A) content and, in this way, create an unsatisfactory nutrient status for maximum growth. The balance-sheet of ions and some knowledge of the characteristics of the plant species with respect to ion uptake and accumulation will often help to indicate in what respects the formulas should be changed to raise (C-A) in the plants to the normal value. Of course, this improvement of the ionic balance does not necessarily bring about maximum growth. Other factors, some related to ion supply as e.g. phosphorus, sulphate, minor elements, others to water, temperature, light and so on, may restrict growth, and thus reduce the size of the yield increment expected from the improvement of (C-A) or even make it zero.

From this it follows that plants with submaximum growth may have varying (C-A) contents, including the normal content. But maximum growth will invariably be associated with the normal (C-A) content.

For each plant species the evaluation of the normal organic salt content and the critical values for ion contents interfering with this normal value can only be made by means of experiments on yield and plant composition with systematic variations in fertilizer formulas. In the present investigation this was done for wheat. Of course, some caution is necessary when using definite critical values. The values given have been inferred from the present results and, in addition, from those of earlier research on related species. They represent the best estimates available at present.

Experimental

Experiment 1

To sandy soil, poor in N and K, sufficient calcium carbonate, magnesium oxide, magnesium sulphate and calcium phosphate was added to bring these constituents to an adequate level. The exchange capacity was 140 meq./kg.

The pots were filled with 7 kg of soil, and solutions of various salts were added in meq. quantities, as listed under the various treatments recorded in Table 1. In each pot, 24 seeds of the variety San Pastore were sown on April 9, 1964, and the pots placed in the greenhouse at about 20 °C. The plants were harvested on May 6 when in the fifth to sixth leaf stage.

The experiment was made in 4 replicates for dry weights. For plant analysis the material of the replicates was combined to form one sample for each treatment.

Table 1 The salt ions, in meq. per pot, supplied to the pots of experiments 1 and 2. The yields are given as the mean of replicates with standard error of the mean

No.	K	Na	Ca	NH ₄	NO ₃	Cl	CO ₃	SO ₄	Yield (g d.m. per pot)	
									exp. 1	exp. 2
1	40				40				7.5 ± 0.2	14.8 ± 0.6
2	20		20		40				7.1 ± 0.1	14.8 ± 0.6
3	0		40		40				4.3 ± 0.2	11.8 ± 0.5
4	40		40		40	40			6.6 ± 0.1	15.7 ± 0.7
5	20		60		40	40			6.8 ± 0.4	14.1 ± 1.4
6	0		80		40	40			3.5 ± 0.2	11.8 ± 0.2
7	80		0		40	40			6.5 ± 0.2	16.9 ± 0.9
8	60		20		40	40			6.1 ± 0.3	15.1 ± 0.6
9	40		40		40	40			6.5 ± 0.4	15.1 ± 1.1
10	80		0		40		40		6.6 ± 0.3	13.6 ± 0.5
11	60		20		40		40		7.6 ± 0.3	15.4 ± 0.5
12	40		40		40		40		7.3 ± 0.2	14.9 ± 0.4
13	20	20			40				7.1 ± 0.1	14.7 ± 0.6
14	10	30			40				7.2 ± 0.3	14.8 ± 0.3
15	0	40			40				4.3 ± 0.2	12.3 ± 0.5
16									1.6 ± 0.1	3.7 ± 0.2
17	20				20				6.6 ± 0.2	11.9 ± 0.5
18	0	20			20				4.0 ± 0.3	10.0 ± 0.4
19	40			40		40	40		6.6 ± 0.3	—
20	40			40		40	40		7.1 ± 0.2	14.2 ± 0.4
21	40			20	20		40		8.3 ± 0.2	—
22	40			20	20		40		8.4 ± 0.1	14.9 ± 0.6
23	40				40				7.6 ± 0.3	—

The results are listed in Table 1 and 2. The ammonium treatments were also made in the presence of 2-chloro-6(trichloromethyl) pyridine (N-serve). Although complete inhibition of nitrification of added ammonium by N-serve was observed earlier (van Tuil and Lampe, 1964), it appeared ineffective in the present experiment.

Experiment 2

A soil-sand mixture, low in N and K, and with an exchange capacity of 250 meq./kg was supplied with the basic dressing and with the additional salt treatments as described under experiment 1. The pots were filled with 7 kg of the soil-sand mixture with basic dressing, salts were added according to the treatments, and 30 seeds were sown of the variety Mara on March 13, 1965. The pots were kept in the open at Kragujevac and harvested on May 18 at the fifth to sixth leaf stage. There were 5 replicates for dry weight, the combined material of each treatment was analysed. The treatments are given in Table 1, together with the dry weights, and the results of plant analysis in Table 3.

Experiment 3

The pots were filled with the soil-sand mixture with basic dressing made as under experiment 2, and salt solutions were added according to the treatments listed in Table 4 and 5. The pots were sown with 30 seeds of the variety San Pastore on October 14, 1965 and kept in a greenhouse. The plants were cut on November 15 when in the 4th-leaf stage and another group on December 7 when in the 6th-leaf

In some of the treatments of experiments 1 and 2 nitrogen was added as ammonium, but the results were comparable to the nitrate treatments. Previous research had shown that replacing nitrate by ammonium reduced both yield and (C-A) content although sufficient supply with nutrients did, in fact, exist. The similarity between nitrate and ammonium in the present experiment should be related to a more rapid conversion of the added ammonium to nitrate by nitrification in the soil.

Another example of treatments affecting markedly the (C-A) content but not the yield is provided by the results of experiment 4. The data, summarized by the closed circles in Fig. 3, show that there was no systematic change in yield when passing from treatments with subnormal (C-A) contents to those where the normal (C-A) content was reached. As pointed out before, this is indicative of some other factor unfavourable for growth which restricts dry matter production to a submaximum level. The data of Table 7 show that phosphate was lower than 200 meq. per kg dry matter with organic nitrogen contents greater than 3000 meq./kg for all treatments. This indicates phosphate shortage. That phosphate was restricting growth became likely when the total amount of phosphate in the plants was plotted against the dry plant weights. Fig. 4 shows that there was a systematic increase of plant weight with the increase of the absolute amount of phosphate in the plants. Thus it seems that phosphate shortage had caused a consistent depression of the yield which prevented growth increments otherwise associated with increments of (C-A) towards the normal value.

Of course there are factors influencing the yield but not the (C-A) content. Treatments related to these factors may reduce the plant weight considerably without lowering the (C-A) content. An example is shown by the open circles in Fig. 3. It con-

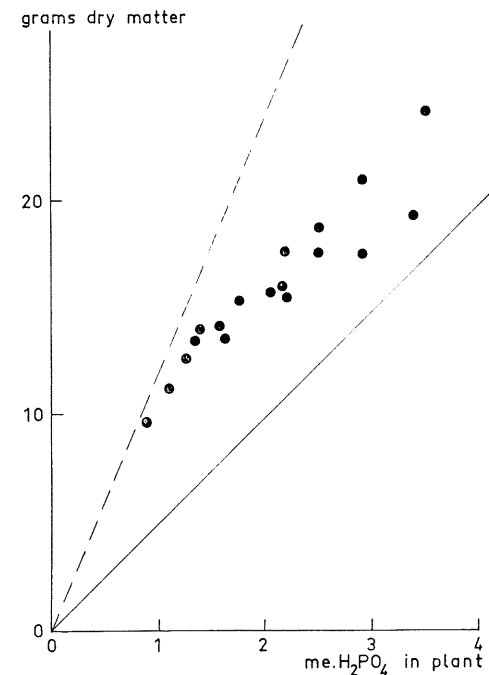


Fig. 4 Relation between the dry weights per plant and the total amounts of phosphate in the plants of experiment 4. The broken line shows the organic phosphate, calculated for the average level of 3500 meq. organic N/kg dry matter, the solid line the phosphate requirement of 200 meq. H₂PO₄/kg dry matter for maximum growth.

This effect of chloride was small compared with earlier results on barley and oats where the yield and (C-A) content were reduced by more than 25% by similar applications of chloride (de Wit et al., 1963).

In experiment 2 there was no effect of chloride on the yield. The low nitrate and organic nitrogen contents show that nitrogen had been short in the supply. These nitrogen deficient plants would not be expected to make additional growth when chloride was removed from the supply. The (C-A) content was raised but the yield remained unchanged at the submaximum level since it was restricted by shortage of nitrogen and not by (C-A).

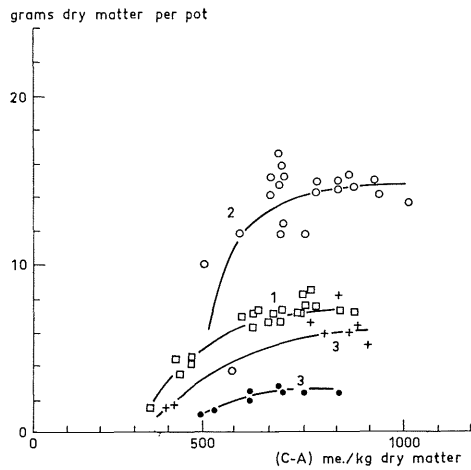


Fig. 2 Yields and organic salt content (C-A) in the three pot experiments. The numbers correspond to those of the experiments mentioned in the text, with two growth stages for experiment 3.

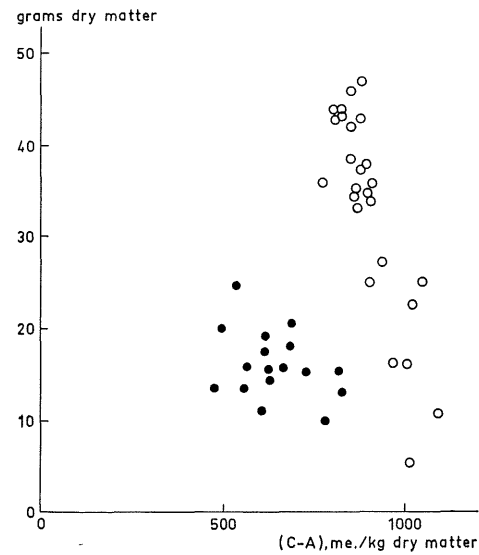


Fig. 3 Open circles: yields and (C-A) contents in corn, grown with varying amounts of micro-nutrients in the supply which caused the plant weights to vary considerably without much change in (C-A) contents. Data of Scharrer and Jung (1956). Closed circles: results of experiment 4, with differences in (C-A) contents due to treatment, and variations in yield due to varying low phosphate levels.

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Table 2 Inorganic cations, their sum (C), inorganic anions, including total phosphorus, their sum (A), organic nitrogen (N_{org}) and the organic salt content (C-A) in the plants of pot experiment 1, made at Wageningen. All the data in meq./kg dry matter, N_{org} in mg atoms/kg dry matter. The first column shows the treatment numbers specified in Table 1.

No.	K	Na	Mg	Ca	C	NO_3	Cl	H_2PO_4	SO_4	A	N_{org}	(C-A)
1	1872	8	213	179	2274	856	160	280	95	1394	3304	880
2	1713	8	246	254	2223	880	152	303	104	1440	3382	782
3	434	52	666	518	1672	358	183	581	81	1203	3802	468
4	1877	4	213	274	2369	664	620	300	60	1645	3495	724
5	1596	4	246	319	2167	313	541	313	78	1548	3521	618
6	506	34	592	613	1747	329	234	610	104	1278	3717	469
7	2010	4	213	239	2467	700	580	306	59	1647	3517	820
8	1982	4	213	254	2454	871	510	316	68	1765	3472	688
9	1867	8	213	284	2373	661	597	309	68	1637	3464	736
10	1897	8	197	179	2283	977	174	296	130	1579	3365	704
11	1833	8	197	174	2214	835	157	300	103	1397	3279	817
12	1838	8	213	204	2265	941	152	296	101	1493	3309	772
13	1971	17	246	184	2420	879	157	296	130	1464	3350	955
14	1263	65	328	239	1897	685	152	319	81	1239	3452	658
15	455	347	534	319	1657	524	194	429	86	1234	3716	422
16	746	8	320	319	1395	12	380	548	159	1101	1781	293
17	1383	8	164	164	1721	95	166	335	202	799	2602	921
18	404	56	641	459	1561	262	174	558	102	1089	3531	462
19	1578	4	123	149	1855	64	186	464	401	1116	4004	738
20	1567	4	106	109	1788	111	183	438	364	1098	3752	690
21	1647	4	156	139	1947	519	152	326	132	1130	3321	817
22	1716	8	156	129	2010	616	141	313	134	1204	3281	803
23	1744	4	213	204	2167	717	152	300	79	1249	3259	917

Table 3 Plant composition in experiment 2, made at Kragujevac. The treatment numbers refer to Table 1. For plant dry weights see Table 1.

No.	K	Na	Mg	Ca	C	NO_3	Cl	H_2PO_4	SO_4	A	N_{org}	(C-A)
1	1009	11	128	235	1383	6	150	186	117	459	2208	924
2	886	13	157	261	1317	8	138	182	109	437	2085	880
3	421	118	274	417	1230	10	153	205	101	469	2668	761
4	1076	12	128	297	1513	5	458	179	81	723	1980	790
5	948	14	154	325	1441	10	443	192	79	724	2190	717
6	594	75	266	456	1391	6	337	176	71	590	2714	801
7	1044	14	123	270	1451	6	463	146	81	696	2101	755
8	1038	11	120	283	1452	8	456	119	89	672	1863	780
9	1068	11	130	304	1513	8	499	182	89	778	2056	735
10	1209	12	127	253	1601	11	159	186	123	479	2332	1122
11	1041	10	123	253	1427	8	137	169	124	438	2099	989
12	1007	15	144	301	1467	8	143	192	126	469	2178	998
13	890	18	157	262	1327	6	125	192	122	445	2172	882
14	694	48	177	283	1202	6	138	182	112	438	2080	764
15	425	228	270	349	1272	6	159	211	108	484	2594	788
16	573	61	109	243	986	8	182	119	68	377	1292	609
17	773	12	122	221	1128	5	176	215	92	488	1545	640
18	528	71	196	303	1098	5	180	189	210	584	1795	514
20	1076	10	196	234	1516	8	127	189	145	469	1870	1047
22	1013	10	197	250	1470	10	124	197	123	436	1947	1034

stage. The experiment was made at Kragujevac. The first harvest comprised 6, the other 5 replicates. For analysis the material of the replicates was combined to one sample for each treatment. The results are listed in Table 5.

Experiment 4

This was made in the field at Kragujevac with the variety San Pastore. The plots were sown on October 24, 1964. On March 31, 1965 100 plants were removed from each plot for the determination of dry weight and composition. The experiment was continued to ripeness and records were made of grain and straw yields.

Ammonium nitrate was applied; one-fourth before sowing, one-half at the onset of growth, and the remainder after sampling on March 31. Prior to sowing superphosphate and muriate of potash were applied according to the treatments (Table 6).

Table 4 Treatments and yields of experiment 3, made at Kragujevac

No.	Treatments (meq. per pot)		Yield (g d.m. per pot)	
	KNO ₃	KHCO ₃	Yield	
			4 leaves	6 leaves
1	9	0	1.0 ± 0.05	1.7 ± 0.04
2	10	20	2.3 ± 0.10	5.2 ± 0.14
3	11	40	2.3 ± 0.12	6.1 ± 0.26
4	12	60	2.2 ± 0.14	6.2 ± 0.05
5	13	0	1.3 ± 0.06	1.8 ± 0.05
6	14	20	2.3 ± 0.17	5.7 ± 0.12
7	15	40	2.5 ± 0.13	6.3 ± 0.27
8	16	60	2.4 ± 0.11	6.1 ± 0.20

Table 5 Plant composition in experiment 3, made at Kragujevac. For treatments see Table 4.

No.	K	Na	Mg	Ca	C	NO ₃	Cl	H ₂ PO ₄	SO ₄	A	N _{org}	(C-A)
1	1205	42	273	304	1824	14	664	485	148	1311	2341	513
2	1545	49	308	304	2206	777	383	328	46	1534	3642	672
3	1654	35	289	335	2313	809	319	319	58	1505	3593	808
4	1789	41	263	334	2427	858	300	312	36	1506	3642	921
5	1271	28	290	302	1891	11	690	453	153	1307	2362	584
6	1658	30	279	307	2274	857	359	325	52	1593	3408	681
7	1742	32	263	340	2377	928	327	323	44	1622	3501	755
8	1735	37	296	307	2375	856	306	325	128	1615	3541	760
9	995	32	237	346	1610	11	626	393	181	1211	1589	399
10	1240	32	220	315	1807	164	299	280	68	811	3045	996
11	1498	32	238	315	2083	604	259	238	40	1141	3226	942
12	1615	35	344	298	2222	741	259	241	97	1338	3233	954
13	1089	32	229	261	1611	15	628	380	178	1201	1727	410
14	1345	24	176	305	1850	159	310	394	113	976	3058	874
15	1544	24	237	298	2103	639	259	257	128	1283	2387	820
16	1613	28	246	325	2212	742	254	252	57	1305	2486	907

Table 6 Treatments, plant weights in shooting stage, and final total dry weights and grain weights (14% moisture basis) in experiment 4, made in the field at Kragujevac. For split application of N compare text. K was given as 40% muriate of potash, N as ammonium nitrate limestone. The degree of lodging was estimated at two dates.

No.	Treatments (kg/ha)			% lodging		Grams of dry weight per plant 31/3	Yield at ripeness (ton/ha)	
	N	P	K	31/5	22/6		total	grains
						1		
2	100	—	—			11.3	11.6	5.1
3	—	100	—			15.4	8.7	4.2
4	—	—	100			15.7	8.3	4.1
5	100	100	—			15.0	11.9	5.2
6	100	—	100			12.9	10.4	5.2
7	—	100	100			14.1	9.1	4.2
8	50	100	100			15.4	10.9	5.0
9	100	100	100		+	21.2	12.0	5.1
10	150	100	100	50	100	17.3	11.9	5.0
11	200	100	100	70	100	18.9	11.5	4.6
12	100	50	100		+	15.9	12.1	5.1
13	100	200	100		+	24.3	11.9	5.2
14	100	300	100		100	17.5	12.7	5.1
15	100	100	40	30	100	17.9	11.9	4.9
16	100	100	160	50	100	19.1	11.9	5.3
17	100	100	200		100	13.7	12.1	5.1
18	200	300	200	90	100	13.4	11.9	4.7

Discussion of the results

The data on yields and (C-A) contents have been summarized in Fig. 2 for the experiments 1, 2 and 3. Most of the treatments in experiments 1 and 3 resulted in organic nitrogen contents of about 3500 meq./kg of dry matter. The (C-A) contents varied according to the treatments but did not exceed the value of 1000 meq./kg dry matter which represents the normal organic salt content of gramineous plants.

The yield dropped when (C-A) fell below 750 meq. per kg dry matter owing to nitrate shortage (e.g. treatments 5 and 13 of exp. 3), or potassium shortage (e.g. treatments 3, 6 and 18 of exp. 1 and 2) or both (treatment 16 of exp. 1 and 2).

In treatments 1 and 10 of exp. 2, the low nitrate content indicates shortage, but nitrate had not come to exhaustion but shortly before harvesting, since the (C-A) content was still high. Treatment 17 of exp. 1 gave similar results. But in treatment 17 of exp. 2, nitrate exhaustion had advanced much further since both the nitrate and the organic nitrogen contents were low, and the (C-A) content had dropped to a value significantly lower than the normal.

Some of the treatments had chloride in the fertilizer, others not. Since chloride is readily absorbed but not metabolized, its presence in the fertilizer is often associated with increased inorganic anion contents A. With chloride there may be some increase in the cation content C but this increase is less than the increment in A and the (C-A) content falls to a subnormal value. For instance, treatments 1, 2, 10 and 11 without chloride gave average yields, (C-A) and chloride contents of 7.2 g, 795 meq./kg and 160 meq./kg, respectively, in experiment 1, whereas treatments 4, 5, 7 and 8 with chloride yielded 6.5 g, 711 meq./kg and 563 meq./kg.