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Nitrogen nutrition of rice plants measured by growth and nutrient content in pot experiments. 2. Uptake of ammonium and nitrate from a waterlogged soil

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Summary

Rice plants were cultivated in a flooded soil in pots provided with varying proportions of the nutrient salts of ammonium, potassium, nitrate, chloride and sulphate.

With nitrate alone denitrification caused shortage of nitrogen and restricted uptake. When replaced by the stable ammonium form of nitrogen, growth of the plants and their capacity to incorporate nitrogen determined the quantity absorbed.

When potassium nitrate was replaced by ammonium chloride the plants grew less than when ammonium sulphate was used to replace potassium nitrate. In view of the relatively small difference in the ionic strength of the medium, this difference in growth was related to the greater accumulation of chloride within the tissues which caused a marked fall in the concentration of carboxylates to a very low level indicative of nutritional stress.

Introduction

We (Ismunadji and Dijkshoorn, 1971) studied the effect of ammonium and nitrate on growth and composition of rice plants in sand culture. When the increase in osmotic pressure of the medium caused by substitution of salts of ammonium and potassium for potassium nitrate was restricted by the use of sulphates instead of chlorides or phosphates, the ammonium plants yielded more dry matter than the nitrate plants.

Variance in nitrogen uptake due to form of nitrogen was not significantly different from the variance due to error. Since all the available nitrogen was absorbed, both forms must have been equally available. Therefore, it seemed safe to conclude that no denitrification had occurred in the waterlogged sand which contained no organic matter. A loss of one-fourth of both forms of the added nitrogen was attributed to consumption by algae growing in the water layer above the sand.

In the present experiment use was made of a humic sandy soil with 5 % organic matter, to bring to light the effect of loss of nitrate by denitrification and the stability of ammonium. Furthermore, the increase in osmotic pressure of the added salts, which went together with the substitution of a salt of ammonium and a salt of potassium for

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potassium nitrate alone, was avoided by merely replacing potassium nitrate by ammonium chloride, or ammonium sulphate without restoring the potassium supply. In this way the nitrogen was in constant supply and varied in form, whereas the supply of potassium was reduced with replacement by ammonium. It appeared that the soil alone was unable to supply the plants with sufficient potassium, so that the ammonium plants showed reduced growth from shortage of potassium.

Experimental

The pots (diameter 20 cm) were filled with 6.5 kg of a sandy soil with 5 % organic matter, pH 5.5. Measured quantities of normal solutions of the various nutrient salts were added to the soil. Enough water was added to form a water layer of 0.5 cm above the soil, and this was mixed to distribute the salts and to remove trapped air.

Another 1 kg of the unfertilized soil was added as a top layer and water was again added to restore a water layer of 0.5 cm above the soil surface.

Each pot received 4 healthy seedlings of the rice vartiety IR-5. With the growth of the plants, the water level was raised to 3 cm above the soil and kept there by frequent watering.

Sixty days after transplanting the tops of the plants were harvested by cutting at a height of 7 cm above the soil surface. The material was dried, weighed, powdered and stored for analysis.

The quantities of salts are listed in Table 1. The supply of potassium and form of nitrogen was varied step-by-step by substituting for x: 0, 10, 20, 30 or 40, thus varying the amounts from 0 to 40 meq per pot.

In Series a the supply of nitrate was increased by replacing potassium sulphate by its equivalent of calcium nitrate. At more complete substitution potassium shortage was induced because its supply was reduced and the growth was improved by the increase in nitrate.

Table 1. The treatments in ion milliequivalents added per pot. Only the first (x = 0) and the last (x = 40) members of the replacement series a, b and c are recorded.

Series	Substitution of salts	Milliequivalents per pot								
		NH_4	K	Na	Mg	Ca	NO_3	Cl	H_2PO_4	SO_4
a	x Ca $(KN0_3)_2$		40		10	10	0		10	50
	(40-x) K_2S0_4		0		10	50	40		10	10
b_1	x $KN0_3$		0	40	10	10	40		10	10
	(40-x) $NaN0_3$		40	0	10	10	40		10	10
b_2	x $KN0_3$		0		10	50	40		10	10
	(40-x) Ca $(N0_3)_2$		40		10	10	40		10	10
b_3	x $KN0_3$		0		50	10	40		10	10
	(40-x) Mg $(N0_3)_2$		40		10	10	40		10	10
c_1	x NH_4CL	0	40		10	10	40	0	10	10
	(40-x) $KN0_3$	40	0		10	10	0	40	10	10
c_2	x $(NH_4)_2$ $S0_4$	0	40		10	10	40		10	10
	(40-x) $KN0_3$	40	0		10	10	0		10	50

In Series b nitrate was kept at the highest level of 40 meq per pot, and the potassium supply was varied form shortage (x = 0) to the highest level (x = 40 meq per pot) by stepwise substitution of potassium nitrate for sodium nitrate (b_1), calcium nitrate (b_2) or magnesium nitrate (b_3).

In Series c ammonium was introduced by a stepwise substitution of ammonium chloride (c_1) or ammonium sulphate (c_2) for potassium nitrate. In these series ammonium substituted for potassium, and the anion for nitrate, so that with a constant nitrogen supply of 40 meq per pot, the introduction of ammonium went together with a reduction in the potassium supply.

This experiment was made with vegetative plants in a growth room under 400 W mercury vapour lamps with a 14-hour light period per 24 hours and a light intensity of 700 cal m^{-2} min^{-1} at plant height.

Table 2. Concentration of ionic constituents and of organic nitrogen in the tops of the rice plants. The code refers to the treatment series of Table 1.

Series	meq per pot	Yield (g DM)	Concentration (meq/kg DM)								
			K	Na	Mg	Ca	NO_3	Cl	H_2PO_4	SO_4	org. N
a	0	2.61	868	6	200	354	8				1352
	10	3.11	847	5	235	279	8				1364
	20	3.47	847	5	292	344	8				1558
	30	3.39	791	7	372	319	8				1626
	40	3.15	394	35	629	319	21				2196
b_1	0	5.10	404	311	553	324	5	265	161	298	1870
	10	4.57	765	54	472	324	10	262	168	348	1990
	20	5.04	862	36	421	324	8	240	158	358	1912
	30	5.15	906	17	378	339	8	245	155	350	1718
	40	4.59	847	7	355	334	10	245	145	356	1842
b_2	0	3.67	473	34	595	369	15	321	119	273	2111
	10	3.80	791	9	367	299	10	245	113	327	2036
	20	4.84	883	10	390	314	6	223	135	353	1777
	30	4.53	901	15	369	344	8	237	145	345	1695
	40	4.33	908	14	360	294	8	243	139	378	1901
b_3	0	3.18	461	25	828	294	32	409	119	276	2288
	10	5.28	783	9	527	329	8	268	152	325	1604
	20	4.49	875	7	491	304	15	262	145	359	1791
	30	5.36	901	7	447	309	6	240	139	349	1617
	40	5.60	924	7	389	319	6	248	139	356	1617
c_1	0	5.85	855	7	415	309	6	237	139	332	1708
	10	5.74	842	7	368	260	15	587	122	247	2123
	20	4.97	837	7	406	289	18	711	119	189	2645
	30	4.55	747	10	441	324	21	708	113	150	2654
	40	2.17	345	36	444	354	16	677	103	74	3024
c_2	0	5.03	850	7	370	284	10	290	129	297	1785
	10	6.80	868	7	352	299	6	203	129	366	1891
	20	7.07	798	10	321	284	8	178	113	409	2484
	30	6.54	737	19	405	299	10	209	106	345	2619
	40	4.18	350	31	517	260	24	336	97	135	2925

Results and discussion

The results of plant analysis are listed for all treatments in Table 2. Organic nitrogen is expressed as milliequivalents of nitrate or ammonium ions consumed for its production, which equals millimoles of organic N. This makes the data fit in well with the electroneutrality conditions in the ionic balance (Dijkshoorn, 1962).

Uptake of cations by the rice plants

The change in concentration of the metal cations within the plants with substitution of one for the other in the added salts may give some idea of the selective and competitive uptake by rice plants. These data are summarized in Fig. 1.

Where potassium substitutes for sodium (Series b_1) or for magnesium (Series b_3), the line for potassium curves upward whilst the curveture for sodium and magnesium is downward. From this it would seem that potassium competes effectively with sodium and magnesium. That potassium competes more effectively than sodium with magnesium is evident from the marked increase in magnesium when potassium was replaced by sodium at constant supply of magnesium (Series b_1).

It will be noticed that magnesium concentrated to a high level when supplied in the absence of potassium (Series b_3) and showed distinct competitive interaction with the monovalent cations. In this detail the rice plant seemed to differ from the uptake peculiarities observed in other gramineous species (de Wit et al., 1963).

The point about which there might be a doubt in the above interpretation is that concentrations in the plant were related to proportions in the added salts, and not to their relative availability in the treated soil. Work on solution culture is in progress.

Soil moisture level and availability of nitrate to ryegrass

In the present context the evaluaton of availability of nitrate nitrogen from plant analysis can hardly be better exemplified than by a representation of some data obtained with ryegrass.

To judge the availability it is instructive to express the results in terms of the absolute amounts absorbed per pot, in order to see whether uptake could have been restricted by supply. These figures can be obtained by multiplying the grams dry weight found in the the pot experiment by the concentration per gram dry matter found in the laboratory.

Ryegrass was grown in pots kept at various, constant soil moisture levels by daily watering to predetermined pot weights. In Fig. 2 the total nitrogen absorbed by the herbage is plotted against the dry weight of the herbage.

In Experiment II the points were found to lie on a straight line through the orgin of the graph. This shows that when the plants yielded more dry matter at a better soil moisture level, the amount of nitrogen absorbed increased in proportion.

In this way, the concentration of nitrogen in the dried herbage remained constant at the value indicated by the slope of the line of 2.7 meq/g DM. Here, available nitrogen remained undetermined, but was more than 43 meq/pot absorbed at the soil moisture level with the best growth. Consequently, the smaller plants in the drier pots had nitrogen available in excess, so that the concentration of 2.7 meq/g DM must have been the maximum the plants could incorporate. Here, the amount of absorbed nitrogen was entirely determined by the growth and the data supply no means of testing the availability.

Each series of moisture levels was concluded with pots containing excess of water.

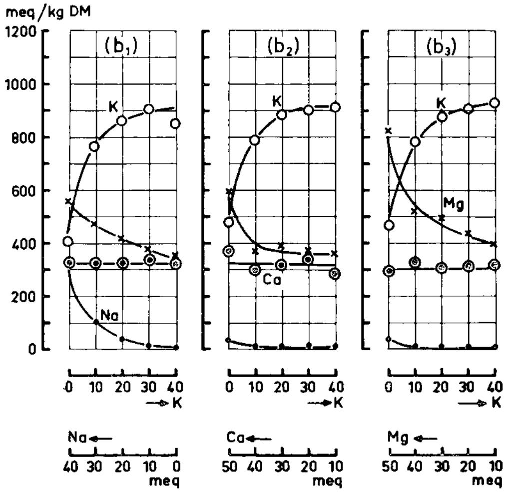

Fig. 1. Concentration of cations in the dried tops of the rice plants grown in the series with substitution of potassium for sodium (b_1), for calcium (b_2) and for magnesium (b_3) in the nutrient salts added to the soil.

The points referring to this waterlogged soil are located at the bottom of the broken line drawn for orientation. Both the uptake of nitrogen and the dry weight of the herbage were markedly reduced. But the observations do not fit in with the relationship for the aerated soils as they would have if the growth had been the regulating factor for uptake of nitrogen. The location of the points shows that the plants absorbed only 1.5 meq/g DM. Since this concentration was much lower than the maximum, it could be stated with safety that waterlogging had reduced available nitrogen from more than 43 meq to about 15 meq per pot and that, for this treatment, absorption was a measure of availability.

In Experiment I growth determined uptake of nitrogen only in the drier pots with the lower yields. At the more favourable moisture levels, producing more dry matter, the uptake of nitrogen lagged disproportionately behind and the concentration in the dried herbage fell correspondingly to submaximum values. From then the uptake of nitrogen was mainly determined by its availability. The highest value of 32 meq per pot was at-

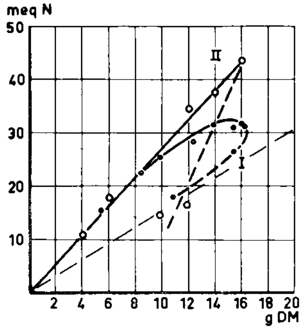

Fig. 2. Nitrogen uptake and dry matter production by ryegrass grown at soil moisture levels ranging from very dry to waterlogged. With excess water (dashed sections of the curves) nitrogen uptake was reduced to 1.5 meq/g DM. Experiment I showed shortage of nitrogen at the soil moisture levels with better growth. Experiment II had sufficient nitrogen to permit uptake to be raised in proportion to the dry weight so as to maintain the maximum level within the foliage.

tained as a gradual approximation to the available amount, the plants containing lower and lower concentrations the further dry weight increased. In the waterlogged soil, indicated by the dashed section of the curve, there was again a marked fall in nitrogen available to the plants.

Thus available nitrogen can only be estimated from weight and composition of plants if they show evidence of shortage for establishing their maximum nitrogen concentration. In terms of Fig. 2: the observations should lie below the proportionality line of maximum concentration, irrespective of the values for dry weight and absorption.

It would naturally be concluded that waterlogging made the nitrate added as a fertilizer to disappear by denitrification.

But since ryegrass is poorly adapted to waterlogged soil, the reduction in availability might also have originated from a disfunction of the root system caused by anaerobiosis.

The former concluson will grow more definite when the evidence obtained with rice plants is outlined.

Uptake of nitrate by rice from waterlogged soil

The rice plant is marked out from other Gramineae that it is well adapted to waterlogged soils. It seems interesting to see whether the uptake by this species brings to light any parallelism, if nitrate is chosen as the one source of nitrogen in which losses by denitrification are likely to occur and ammonium as the other one in which changes are unlikely to occur in waterlogged soil.

In Fig. 3, Graph 1 refers to Series a of Table 1, and begins with the addition of 40 meq per pot of potassium sulphate and no nitrate. Since the concentration of nitrogen in the dry matter was low, growth was retarded and the 3.6 meq of N absorbed per pot was all the unfertilized soil could provide.

With the substitution of calcium nitrate for potassium sulphate absorption of nitrogen increased to 7 meq per pot. At first, the uptake of potassium increased by better growth but, at more complete substitution, shortage of potassium made its appearance and the uptake fell to a lower level with low concentrations of potassium in the plant material.

Plant analysis (Table 2) showed that the plants of all treatments contained rather high levels of unconsumed sulphate anion, indicating that sulphate was invariably absorbed in excess of the metabolic requirements (Dijkshoorn and van Wijk, 1967). Therefore, no weight should be assigned to variations in sulphate supply. What matters in the present experiment is the anion chosen to replace the sulphate in the medium.

Graph 2 of Fig. 3 shows the uptake of potassium and nitrogen for Series b_2 where potassium nitrate substituted for calcium nitrate. With the increase in uptake of potassium the yield was raised, but the uptake of nitrogen remained unchanged at the level of 8 meq per pot. The data of Table 2 show that the higher the yield, the lower was the concentration of nitrogen in the plant material. On the face of this it would seem that shortage of nitrogen restricted uptake to 8 meq per pot in the tops. Since the whole plant contains 1.5 times as much, available nitrogen must have been 12 meq per pot which is astonishingly little compared with the 40 meq applied.

A loss of nitrate from the medium is not the only possible conclusion which can be drawn from this small recovery of nitrogen. Perhaps, the rate of uptake was slow, and uptake would have continued if only further growth had been permitted by cutting at a later date. The conclusion will grow more definite when the effect of replacing nitrate by ammonium is considered.

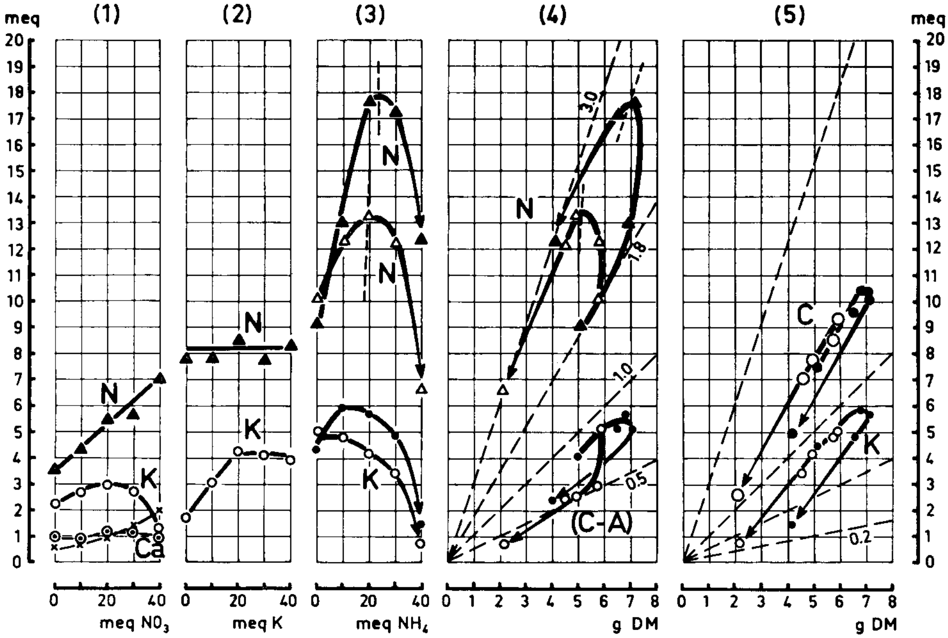

Fig. 3. Accumulation of nitrogen, potassium, total cations (C) and carboxylates (C-A) in the tops of rice plants grown at varying levels of nitrate potassium (Graphs 1 and 2), and with progressive substitution of ammonium chloride (open symbols) or ammonium sulphate (solid symbols) for potassium nitrate (Graph 3). Graphs 4 and 5 are uptake-yield diagrams for the data of Graph 3. For further explanation see text.

The recognition of denitrification

Graph 3 of Fig. 3 records the uptake of nitrogen for Series c_1 with ammonium choride, and Series c_2 with ammonium sulphate. With 40 meq potassium nitrate alone (zero ammonium) the uptake was somewhat higher than in the identic treatment of the preceding series, but what really matters here is that substitution of ammonium at the first two levels, viz 10 ammonium plus 30 nitrate, and 20 ammonium plus 20 nitrate, raised the uptake of nitrogen considerably.

For Series c_2 with ammonium sulphate plants grown with 10 meq of ammonium application absorbed 13 meq nitrogen per pot. If the uptake for 30 meq nitrate alone is taken from Graph 1 we arrive at 6 meq. The difference of 7 meq came from the added ammonium. Since the whole plant contains about 1.5 times the amount of nitrogen in the tops, it would follow that the whole plants absorbed $1.5 \times 7 = 10$ meq of nitrogen per pot as ammonium, when 10 meq ammonium was applied in addition to the nitrate.

A similar reasoning can be applied to the 20 meq ammonium plus 20 meq nitrate treatment of Graph 3. Uptake by the tops was 18 meq of which, according to Graph 1 at the 20 meq nitrate level, 5 meq came from the soil supplied with the nitrate only. Consequently, the 20 meq ammonium added in addition to the nitrate, accounts for the uptake of 13 meq by the tops, and $1.5 \times 13 = 20$ meq by the whole plants.

Of course such conformity is not realistic. In view of the uptake of 8 meq per pot in

Graph 2 one might prefer to draw a steeper line in Graph 1 which ends at this value. But then the value arrived at above will not differ by more than 1 meq.

The evidence seems to support what has already been said about the complete availability of added ammonium and the incomplete availability of added nitrate. It also suggests that at least with respect to ammonium, the uptake system worked adequately.

If this were true, the uptake of nitrogen should increase further, because at completion of the replacement, 40 meq ammonium would be completely available. Instead, the curve for uptake of nitrogen declines, and at the 40 meq ammonium level the uptake by the tops is only 12 meq, and by the whole plants 18 meq per pot for Series c_2 with ammonium sulphate. Consequently, available nitrogen was in excess of the uptake, and Table 2 shows that, with the reduction in dry weight of the plants, nitrogen concentrated markedly in the plant material.

The nature of the optimum curve for the uptake of nitrogen is now beyond doubt. The ascending branch results from the increase in the proportion of stable ammonium which increased the amount remaining available to the plants in the waterlogged soil. The descending branch originates form the reduction in pant weight caused by less and less available potassium, with uptake of potassium below the requirements for better growth.

The uptake-yield diagram for the rice plants

The state of affairs can be established more directly by an arrangement of the data in an uptake-yield diagram as in Graph 4 of Fig. 3. The corresponding treatments can be identified by reading the corresponding points in Graph 3. It was found convenient to symbolize the change in treatment by drawing the lines thinner, and providing them with arrows in the direction of more complete substitution of ammonium for nitrate as the source of nitrogen.

The graph will bring to light the milliequivalents absorbed, the dry matter produced, and the concentration in meq/g DM at any level of yield and uptake. It gives a definite outline of the way the element concentrates in the dry matter, since the representation brings out that concentration is the integrated result of accumulation and dry matter production, a fact which is apt to be obscured when concentrations are directly related to treatment.

For orientation, straight dashed lines through the origin were drawn at different indicated levels of concentration. Inspection of the graph will make it obvious that if the element concentrates in the dry material, the next point lies on a steeper concentration line.

In the curves for nitrogen in Graph 4, the right-hand branch begins with the treatment 40 meq nitrate alone. For the sulphate series c_2, with the solid triangles, the first step of 10 meq ammonium plus 30 meq nitrate moved the point nearly along the concentration line of 1.8 meq/g DM. Here, the increase in available nitrogen due to the substitution of stable ammonium for labile nitrate, produced more dry matter of nearly unchanged, low nitrogen concentration. In this range, available nitrogen apparently restricted the growth.

The subsequent step, from 10 meq ammonium plus 30 meq nitrate to 20 meq ammonium plus 20 meq nitrate, resulted in a marked increase in nitrogen uptake, but without further increase in dry matter. This proves that some other factor was limiting growth.

It must be borne in mind that in this experiment we are dealing with one fixed growing period. The possibility of further expansion of the dry mattter did not come into view but with a longer duration of growth it can be predicted that the point would lie somewhere nearer the concentration line of 1.8 meq/g DM.

The further decrease in weight of dry matter, when 30 or 40 meq per pot of ammonium replaced the nitrate, proved that the faculty of growth was definitely reduced. The curve for the uptake of potassium in Graph 4 shows that where ammonium completely replaced potassium, the uptake of potassium fell to only 0.3 meq/g DM. At this low level of potassium within the plants, the slow uptake restricted growth.

With the decrease in dry weight due to shortage of potassium, the uptake of nitrogen was diminished (Graph 4), but the latter element concentrated to the high level of 3 meq/g DM.

The curve for the other series (c_1), where ammonium chloride was used to replace potassium nitrate, which is marked by the open triangles, fits in a similar way between the concentration lines. Both curves point alike to the view that in the right hand branch, emerging from the 1.8 meq/g DM concentration line, the availability of nitrogen was the more important part, whilst in the left-hand branch the yield of dry matter was the essential variable for nitrogen uptake.

Between the treatments with 40 meq ammonium alone, the dry matter yield differed by a factor two, but both points are found on the same concentration line of 3 meq/g DM. Here, the expansion of the dry matter determined the uptake of nitrogen, and this concentration was the highest the plants could establish. This implies that soil nitrogen was in excess of the actual growth requirements of the plants.

The obvious difference between the two sources of nitrogen would be that ammonium was stable in the waterlogged soil, and nitrate was lost by denitrification.

The nitrogen balance of uptake and availability

With regard to the availability of added nitrogen it may be recalled that about one-fourth was consumed by algae which grew in the water layer on the top of the soil. Furthermore, about two-thirds of all the nitrogen absorbed by the whole plant appeared to accumulate in the tops (Ismunadji and Dijkshoorn, 1971). This would mean that half of the nitrogen applied to the soil was potentially available to the tops.

Since in the present experiment only the tops were collected and analysed, we have to account for both the consumption by algae, and the distribution ratio of nitrogen within the plants. To arrive at a grouping of the figures in the form of a balance of available and absorbed nitrogen, we assumed that half of the ammonium form of nitrogen applied to the soil remained available to the tops of the plants.

The proportion of nitrate available to the tops can be found directly from Graph 1. If the uptake of 3.6 meq per pot from the unfertilized soil is considered to be constant the increment indicates the uptake from the added nitrate at its various levels of application.

But the uptake observed at 40 meq potassium nitrate per pot did not fit in well with the higher uptakes for the identic treatments of Graphs 2 and 3. This uptake was roughly averaged to 8.6 meq at the 40 meq potassium nitrate level of application. With 3.6 meq from the unfertilized soil, 5 meq was absorbed by the tops per 40 meq of nitrate added to the soil. At the lower levels of application the uptake of nitrate decreased in proportion. Since in Graph 4 the points for the 40 meq nitrate treatment were located well below the highest concentration line of 3 meq/g DM, the plants had absorbed all the available nitrate.

Table 3 shows the manner in which the figures were grouped. The figures marked

Table 3. Levels of application of ammonium and nitrate, and of the various forms of nitrogen available to the tops of the plants. The last two columns show the amount of total nitrogen found in the tops. Derived from treatment series c_1 and c_2.

Applied. (meq/pot)		Available to tops (meq/pot)				Total nitrogen found in tops (meq/pot)	
NH_4	NO_3	NH_4	NO_3	soil-N	sum	(c_1)	(c_2)
0	40	0	5	3.6	8.6	10.1	9.0
10	30	5	3.8	3.6	12.4	12.2	12.9
20	20	10	2.5	3.6	16.1	13.4	17.6
30	10	15	1.4	3.6	20.0	12.1	17.2
40	0	20	0	3.6	23.6	7.6	12.3

'sum' give the total nitrogen available to the tops of the plants from the various sources of nitrogen. In the last two columns the findings for total nitrogen in the tops are listed for Series c_1 and c_2.

For the first two members of each series the uptakes lie very close to the sum of available nitrogen. All of it was absorbed at some earlier stage of growth, and with the further depleted growth the concentration in the dried material fell to a low level.

For the last member available nitrogen remained in excess of uptake and the plants maintained their maximum concentration until harvest.

Owing to the poorer growth in Series c_1, the demand of nitrogen was smaller and, for this series, nitrogen remained available in excess for the last three members. As a consequence, the area enclosed by the uptake-yield curve in Fig. 3 is centred more to the left, the points shifting to higher concentrations lines, compared with Series c_2 with better growth and a higher demand for nitrogen.

The chloride effect and its ionic parallel

The origin of the poorer growth in Series c_1 with ammonium chloride instead of ammonium sulphate is now considered.

On the one hand, the introduction of the monovalent chloride in place of the divalent sulphate included the risk of a rise in osmotic pressure not well tolerated by the plants (Ismunadji and Dijkshoorn, 1971).

On the other hand, chloride may concentrate to a much higher level than sulphate within the plant. Since in the tissues the anions accumulate as salts of the metal cations, either more of these cations will be required to balance the increase in inorganic anions, or fewer will be available to balance organic anions such as malate, citrate, oxalate, succinate, etc.

The combination of neutral inorganic salts absorbed, and the metabolic conversion of the greater part of the nitrate, all of the ammonium, and some of the sulphate into organic nitrogen and sulphur, results in the final accumulation of inorganic and organic salts of the metal cations, with a sum of milliequivalents C.

The inorganic anions left negative by metabolism are some nitrate (low or absent with shortage of nitrate), some sulphate (very low with shortage of sulphur), all the phosphate (including the organic phosphate which retains the monovalent ionic form of inorganic phosphate) and all the chloride. Their sum of milliequivalents is called A. The excess of metal cations C over het inorganic anions A is balanced by anions of the carboxylic plant acids generated by metabolism. The total amount of these carboxylates is numerically fixed by the difference (C-A) obtained from analysis for inorganic ions in the plant material (Dijkshoorn, 1962).

By this scheme of analysis for the major inorganic ions it is possible to fit together into a connected whole the ionic balance within the plant.

Changes in nutrition such as substitution of calcium for potassium, and ammonium for nitrate, may bring about a reduction in the uptake of metal cations and in the value of (C-A).

If chloride is supplied, the increment in inorganic anions A may exceed the increment in metal cations C so that the value of (C-A) is reduced.

Attempts to force the effect of chloride on the growth of plants into line with that of the other ions known to reduce (C-A) have the decided advantage of bringing a unifying conception to bear upon the problem of relating the pattern of the ionic balance to the faculty of maximum growth.

The paper of de Wit et al. (1963) contains evidence on some of the grass species and cereals, but it should be developed on a wider scale where other plant species are concerned.

It is based upon assumption that, in addition to the specific requirements for the essential elements themselves, there is a limited freedom to substitute cations for other cations, and anions for other anions, without losing the plant's faculty of maximum growth, if only the concentration of carboxylates is kept at the level of (C-A) which is characteristic of the plant species.

It is worth mentioning that no speculations were made as to requirements of carboxylates. Their additive, numerical value merely supplied a means of testing the adequacy of ion accumulation as a whole. With values higher or lower than the normal the plants were found to have lost the power for maximum growth, a rule to which no exceptions were found. This situation, then, was related to some sort of nutritional stress. The data on concentration of the different ion species can throw more light upon the nature of this stress, and the measures to be taken to releave it.

There remains the problem of identifying the normal value of (C-A). By its very nature, we are forced to rely upon our judgement of experimental results, and to select a value which appears to be associated with the best growth. But if some other factor interfered, maximum growth may not be manifested, and the best growth may occur within a certain range of values for (C-A). Then, selection of a more discrete value may become a mere guess which is most congenial to our taste or experience.

Turning to the present line of evidence it is seen that the highest dry weight of 7.2 per pot was attained in the series with ammonium sulphate. If a straight line is drawn through this point and the origin of Graph 4, Fig. 3, this line slopes at 0.75 meq/g DM. Let this value be identifiable with the normal value for (C-A).

The loop-shaped curve for (C-A) in the series with ammonium sulphate (solid circles) is centred along this line, with higher concentratons on the nitrate side, and lower concentrations on the ammonium side. But the curve for ammonium chloride (open circles) drops at the very first introduction of 10 meq ammonium chloride to the subnormal level of 0.5 meq/g DM, with no significant change in dry weight of the plants.

Examination of Graph 5 in Fig. 3, of Fig. 5 and of Table 2 will show that there was little difference between the series in the concentration of potassium and total cations at the same levels of application of ammonium.

All this goes to show that we are dealing with the effect of accumulation of chloride

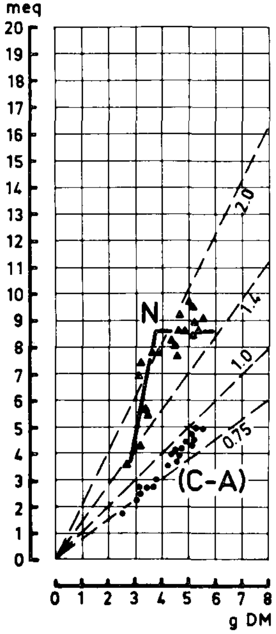

Fig. 4 The uptake-yield diagram for the series with varying potassium and nitrate levels of application and no ammonium in the fertilizer (Series a and b of Table 1).

in the rice plants which made subnormal levels of (C-A) of earlier appearance merely by an increase in the level of the inorganic anions A.

For the relation between (C-A) and the yield the results for the treatments with nitrate alone remains to be considered.

Fig. 4 shows the same manner of plotting for Series a and Series b_1, b_2 and b_3 where nitrate and the metal cations were varied in the added salts. Nitrogen uptake was raised from 3.6 meq to a level which was eralier averaged to 8.6 meq per pot, with a dispersion of the points due to variations in potassium and experimental errors. Depending on the supply of nitrate, the concentration of nitrogen varied between 1.4 and 2.0 meq/g DM, and these low values indicate that denitrification was fast enough in the waterlogged soil to cause shortage of nitrogen.

At the lower levels of nitrate application, accumulation of (C-A) occurred at 0.75 meq/g DM, but the dry weight was very low due to severe shortage of nitrogen. With the increase in nitrate supply, the accumulation of (C-A) increased disproportionately with the dry weight, and the concentration attained at the highest nitrogen level was 0.9 meq/g DM.

In the previous communication the higher concentration of (C-A) with nitrate was qualified as above normal. Since in that experiment the nitrate plants and the ammonium plants absorbed the same amount of nitrogen without any interference of denitrification, the conclusion that the nitrate plants grew less because they contained a surplus of (C-A) would seem an easy exercise.

In the present experiment the nitrate plants had more shortage of nitrogen than the ammonium plants. But it would be naturally concluded that plants, continuously grown at an adequate supply of nitrate and, consequently, containing a surplus of (C-A), might be improved in growth by permitting them to absorb chloride in sufficient amount to reduce (C-A) to the normal level. At this point it is not possible to discuss the somewhat thorny question of defining the meaning of the normal concentration of (C-A), because no material is available to permit a further qualification of possibilities.

Composition of the rice plants

In the preceding sections enough has been said about the nature of the change in plant composition with the replacement of potassium nitrate by the salts of ammonium. For the sake of clearness it may be well to review some details by plotting the concentrations in the dried material against the treatment. In Fig. 5, the left-hand graph refers to Series c_1 where, from the right to the left, ammonium chloride replaced potassium nitrate. The other graph refers to Series c_2 with ammonium sulphate as the substituting salt.

The graphs show more directly the marked increase in concentration of nitrogen when the nitrate in the fertilizer was replaced by the stable ammonium form of nitrogen. It may be recalled that in the previous experiment the ammonium plants were lower in concentration of nitrogen than the nitrate plants, because there was some shortage in the nitrogen supply, no loss of nitrate by denitrification, equal uptake of both forms, but more dilution by better growth of the ammonium plants (Ismunadji and Dijkshoorn, 1971).

With the increase in the level of application of ammonium chloride (Series c_1) there

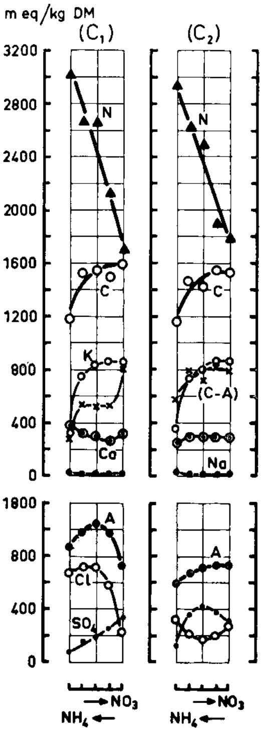

Fig. 5. Concentrations of ionic constituents and of organic nitrogen in the tops of the rice plants for Series c_1 where ammonium chloride replaced potassium nitrate, and Series c_2 where ammonium sulphate replaced potassium nitrate in the fertilizer. Nitrogen concentration increased when nitrate was replaced by the stable ammonium in the waterlogged soil. The concentration of carboxylates (C-A) is indicated by crosses.

was a substantial increase in the concentration of chloride. The concentration of the anions A increased but the increment was reduced by a fall in the concentration of sulphate anion.

The increase by 1200 meq of organic nitrogen would mean that synthesis of organic sulphur increased by about 6 % of this value. If the increased demand for organic sulphur had been satisfied by a further metabolization of the sulphate pool within the tissues, sulphate would have declined by not more than 50 meq/g DM. Since the sulphate concentration dropped by 250 meq, the greater part of the decline in sulphate anion concentration should be attributed to the antagonistic depression of sulphate uptake by chloride.

The very low concentration of nitrate in the plants was negligible throughout.

Both series were comparable in concentration of the metal cations, and the sharp drop in (C-A) in the chloride series was completely mirrored by the increase in concentration of the inorganic anions A.

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