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Ammonium Tolerance and Carbohydrate Status in Maize Cultivars

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Four maize (*Zea mays* L.) hybrids were grown hydroponically for 4 weeks with 20 mM ammonium or nitrate as the sole nitrogen source. Dry matter production was strongly depressed by ammonium nutrition in the hybrid Helga relative to plants grown on nitrate, and moderately decreased in the hybrid Melina. Ammonium had no inhibitory effect on total yield in the other two hybrids (Ramses and DK 261). The relative growth rate (RGR) of roots and shoots of the sensitive hybrid Helga decreased significantly under ammonium nutrition during the first 2 weeks of the experiment, while at the end of the experiment nitrogen form had no effect on the RGR in any of the four hybrids. The strong reduction in RGR of Helga in the early seedling stage was correlated with the accumulation of twice the concentration of free ammonium in the shoot tissue relative to the other hybrids. Helga was therefore unable to sufficiently detoxify ammonia in the roots. Root concentrations of water soluble carbohydrates (WSC) in Helga and Melina in the early seedling stage did not differ under ammonium and nitrate nutrition. In contrast, Ramses and DK 261 both had elevated WSC concentrations in ammonium-fed roots. It is hypothesized that a sufficient supply of carbon skeletons for ammonium assimilation in the roots is required for maximum growth under high ammonium concentrations, and that there is genotypic variability in this physiological trait.

Key words: Ammonium, carbohydrates, growth rate, maize, nitrate, roots, *Zea mays* L.

INTRODUCTION

In many higher plant species, ammonium as the sole nitrogen source leads to physiological disorders and reduced growth when compared with nitrate or mixed nitrogen nutrition (Goyal and Huffaker, 1984). Besides indirect toxic effects such as the acidification of the rhizosphere (Raven and Smith, 1976), ammonium *per se* can severely inhibit growth. Ammonium is generally not stored in cells and appears in only small amounts in root and shoot tissues of the plants. Ammonium toxicity is generally associated with the occurrence of large amounts of free ammonium and ammonia in plant tissues (Gill and Reisenauer, 1993). In photosynthesizing tissues, ammonium can uncouple electron transport from photophosphorylation (Peltier and Thibault, 1983). This has been considered a likely explanation for the reduced photosynthetic rates observed for wheat and maize if plants are supplied with ammonium instead of nitrate (Cramer and Lewis, 1993*a*).

Only a few species have the ability to store ammonium in shoot vacuoles without exhibiting ammonium toxicity symptoms (Smith, 1972). The satisfactory performance of most plants growing on high external ammonium concentrations may thus be due to their ability to detoxify ammonium via assimilation of ammonium in the root (Givan, 1979). Because the assimilation of ammonium into amides and amino acids requires carbon skeletons from the tricarboxylic acid cycle (Oaks, 1992), roots of ammonium-fed plants may form a stronger sink for carbohydrates than the roots of nitrate-fed plants, if nitrate is assimilated in the shoot (Barta, 1976). This may lead to larger net carbon fluxes from the shoot to the root for ammonium-fed plants (Lewis, Fulton and von Zelewski, 1987).

If carbohydrates transported to the roots are used for the assimilation of ammonium and then retranslocated to the shoot as amides or amino acids, this may be at the expense of root growth. The competition of ammonium assimilation and root growth for carbohydrates, was indicated by the split-root experiments of Schortemeyer, Feil and Stamp (1993), Schortemeyer and Feil (1996) and Cramer and Lewis (1993*b*). In these experiments one half of the root system of maize was supplied with ammonium, while the other half received nitrate as the nitrogen source. It was found that the ammonium-fed half of the root system generally exhibits restricted dry matter production. In a split-root experiment with soybeans, Chaillou *et al.* (1994) calculated that nitrate- and ammonium-fed halves of root systems received similar carbon import. Despite this, the ammonium-fed root halves produced less root dry matter (Chaillou *et al.*, 1994). Feil (1994) also showed for split-root-grown wheat that ammonium-fed roots produced less dry matter per unit nitrogen uptake than nitrate-fed roots. The enhanced need of carbon for the assimilation of ammonium was also demonstrated by Magalhães, Huber and Tsai (1992), who found that the supply of exogenous carbon sources such as α-ketoglutarate to ammonium-fed roots improved their growth.

Cultivar differences in ammonium tolerance or sensitivity may therefore be related to differences in production, transport, and shoot–root partitioning of soluble carbo-

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hydrates that are required for the assimilation of ammonium. Published research indicates that there is variation among maize genotypes in the response to ammonium nutrition only or to enhanced ammonium concentrations in mixed nitrogen substrates (Harvey, 1939; Handa *et al.*, 1984). Teyker (1991), working with soil-grown plants and the nitrification inhibitor nitrapyrin, found genotypic variation in the response (relative to nitrate) of inbred maize lines under enhanced ammonium supply (EAS). He related the positive response to EAS of some cultivars to the formation of longer roots in a fertilizer band containing anhydrous ammonia. Furthermore, the variability in the response of inbred lines of maize to ammonium nutrition may be related to their different glutamate dehydrogenase, glutamine synthetase, and glutamate synthase activities (Tsai *et al.*, 1991). Alfoldi, Pinter and Feil (1992) reported an interaction between genotype and nitrogen form in the accumulation of dry matter in roots and shoots of four maize hybrids grown under ammonium or nitrate, or a 1:1 mixture of both. Individual responses of the hybrids to different nitrogen forms were not related to differences in the concentrations of non-structural carbohydrates in roots or shoots.

The objective of this experiment was to determine if differences among commercially important maize cultivars in the response to ammonium as the sole nitrogen source could be attributed to differences in the concentrations of ammonium and of non-structural carbohydrates in shoot and root tissues. To impose stress conditions a large concentration of either nitrate or ammonium (20 mM) was supplied. The plants were sampled at three dates to investigate the stage at which the cultivars showed the largest response to nitrogen form treatments, and whether cultivar differences remained consistent over a longer period.

MATERIALS AND METHODS

Plant culture

Based on the performance of various maize genotypes under ammonium *vs.* nitrate nutrition in unpublished experiments, four cultivars were selected. Of these, growth of Melina and Helga (both from Pioneer Overseas Corporation, Johnston, IA, US. In US Helga corresponds to P3902) was significantly depressed when grown on ammonium instead of nitrate, while Ramses (Coop de Pau-Semences, Lescar, France) and DK 261 (Société RAGT, Rodez, France) grew slightly better under ammonium than under nitrate supply. Seedlings of these four maize hybrids were started in coarse vermiculite in a greenhouse under a 16 h photoperiod and day/night temperatures of 25/21 °C. The experiment was run throughout March 1993. Maximum light intensity at midday reached 1015 μmol m^{-2} s^{-1}. The plants were illuminated by natural daylight supplemented with artificial light from high pressure sodium lamps whenever the daylight intensity fell below 115 μmol m^{-2} s^{-1}. After 1 week, roots were washed free from vermiculite. Uniform plants were transferred to solution culture and grown for 4 weeks under the same environmental conditions. Concentrations of the macronutrients in the original nutrient solution were (mM): Ca, 5; Mg, 3; K, 3; P, 1; and S, 9, supplied as $CaSO_4 \cdot 2H_2O$, $MgSO_4 \cdot 7H_2O$, KH_2PO_4, and K_2SO_4, respectively. Concentrations of micronutrients were (μM): Fe, 89·0; Mn, 18·0; Cu, 0·9; Zn, 1·5; B, 12·0; Mo, 4·2; and Cl, 50·0, given as FeEDTA, $MnSO_4 \cdot H_2O$, $CuSO_4 \cdot 5H_2O$, $ZnSO_4 \cdot 7H_2O$, H_3BO_3, $(NH_4)_6Mo_7O_{24} \cdot 7H_2O$, and KCl, respectively.

TABLE 1. *Initial dry weights* (±*s.e.*), *WSC, and ammonium concentrations for the four hybrids before transfer to hydroponics. Values are based on the analysis of six pooled plants per hybrid*

Hybrid	Helga	Melina	DK 261	Ramses
Shoot dry matter (mg)	28±1	59±4	42±2	35±1
Root dry matter (mg)	67±4	78±13	84±7	61±4
Shoot WSC (%)	14·6	17·7	11·9	7·8
Root WSC (%)	15·4	21·8	10·7	9·5
Shoot NH_4 (μmol g^{-1})	33·9	27·5	16·7	26·0

The plants received an original N concentration of 20 mM supplied as either $(NH_4)_2SO_4$ or $Ca(NO_3)_2 \cdot 4H_2O$, respectively. The nutrient solution was changed weekly to restore nutrients to their original concentrations. The pH of the solution was restored to 5·5 by daily titration with KOH or H_2SO_4. FeEDTA was added every second day.

Twelve plants, three of each hybrid, were grown in each of 24 50-l-containers. In addition, six plants of each hybrid were retained for analysis at the time of transplanting when nitrogen treatments were imposed. The initial dry weight, as well as the concentrations of water-soluble carbohydrates, and of ammonium for the four hybrids are shown in Table 1. At 2, 3, and 4 weeks after transplanting, one plant of each hybrid was harvested per container. The data are based on 12 replications, i.e. 12 plants. Statistical differences displayed in Table 2 are based on analysis of variance.

Measurements

The plants were harvested 10 hours after onset of the light period. At each harvest shoots and roots of the plants were frozen and kept at −20 °C until they were freeze-dried. The shoot and root dry weights were then determined. RGR values for roots and shoots for each time interval were calculated using the running average method to reduce harvest to harvest variation (Poorter, 1989). Fifty to 60 mg of ground tissue for each sample were incubated at 60 °C with 0·5 ml of 80% ethanol for 10 min and then with 5 ml of water for 60 min. After centrifugation evaporation was measured by weighing, and soluble carbohydrates in the supernatant were determined with the anthrone method (Seifter *et al.*, 1950). An aliquot of the solution was hydrolysed in 4 ml anthrone solution in a boiling water bath for 15 min. After cooling, the glucose concentration was determined spectrophotometrically by absorption at 620 nm against a standard solution.

Nitrate and ammonium concentrations of the supernatant were determined on a continuous-flow autoanalyser as described by Schortemeyer *et al.* (1993). Starch in the residue of the shoot samples was digested in 3 ml boiling

TABLE 2. *Shoot and root dry matter, ammonium, and WSC concentrations of four maize hybrids exposed to 20 mM ammonium or nitrate. Values within each sampling date followed by the same letter do not differ significantly by l.s.d. at* $P \leqslant 0{\cdot}05$

	d 14		d 21		d 28	
	NH_4	NO_3	NH_4	NO_3	NH_4	NO_3
Shoot DM (g per plant)						
Helga	$0{\cdot}54^{a}$	$0{\cdot}93^{b}$	$1{\cdot}51^{a}$	$2{\cdot}62^{b}$	$4{\cdot}88^{a}$	$8{\cdot}69^{b}$
Melina	$1{\cdot}23^{c}$	$1{\cdot}41^{d}$	$3{\cdot}61^{c}$	$4{\cdot}15^{d}$	$9{\cdot}87^{b}$	$11{\cdot}96^{c}$
DK 261	$0{\cdot}98^{b}$	$1{\cdot}18^{c}$	$2{\cdot}73^{b}$	$2{\cdot}82^{b}$	$8{\cdot}16^{b}$	$8{\cdot}59^{b}$
Ramses	$1{\cdot}15^{c}$	$1{\cdot}18^{c}$	$2{\cdot}83^{b}$	$2{\cdot}66^{b}$	$8{\cdot}99^{b}$	$8{\cdot}64^{b}$
mean	0·97	1·18	2·67	3·10	7·89	9·47
Root DM (g per plant)						
Helga	$0{\cdot}22^{a}$	$0{\cdot}49^{c}$	$0{\cdot}48^{a}$	$0{\cdot}99^{cd}$	$1{\cdot}18^{a}$	$2{\cdot}46^{cd}$
Melina	$0{\cdot}50^{c}$	$0{\cdot}65^{d}$	$1{\cdot}08^{d}$	$1{\cdot}24^{e}$	$2{\cdot}23^{c}$	$2{\cdot}67^{d}$
DK 261	$0{\cdot}39^{b}$	$0{\cdot}48^{c}$	$0{\cdot}86^{bc}$	$0{\cdot}89^{bc}$	$1{\cdot}92^{bc}$	$1{\cdot}87^{bc}$
Ramses	$0{\cdot}45^{bc}$	$0{\cdot}46^{bc}$	$0{\cdot}88^{bc}$	$0{\cdot}79^{b}$	$1{\cdot}97^{bc}$	$1{\cdot}97^{b}$
mean	0·39	0·52	0·82	0·99	1·83	2·20
Shoot NH_4 (μmol g^{-1} DM)						
Helga	$62{\cdot}1^{d}$	$15{\cdot}2^{a}$	$37{\cdot}6^{c}$	$14{\cdot}3^{a}$	$25{\cdot}8^{d}$	$10{\cdot}3^{a}$
Melina	$34{\cdot}3^{b}$	$12{\cdot}8^{a}$	$21{\cdot}9^{b}$	$12{\cdot}7^{a}$	$18{\cdot}9^{bc}$	$8{\cdot}8^{a}$
DK 261	$37{\cdot}0^{bc}$	$13{\cdot}5^{a}$	$22{\cdot}7^{b}$	$12{\cdot}1^{a}$	$18{\cdot}1^{b}$	$9{\cdot}1^{a}$
Ramses	$41{\cdot}7^{c}$	$16{\cdot}1^{a}$	$27{\cdot}8^{b}$	$15{\cdot}0^{a}$	$21{\cdot}2^{c}$	$10{\cdot}3^{a}$
mean	43·8	14·4	27·6	13·6	21·0	9·6
Root NH_4 (μmol g^{-1} DM)						
Helga	162·1	48·4	143·1	52·8	126·8	68·9
Melina	148·7	44·7	122·3	47·3	110·7	79·5
DK 261	142·4	37·2	143·9	41·4	92·1	64·8
Ramses	173·4	54·2	146·2	54·8	95·4	55·5
mean	$156{\cdot}6^{b}$	$46{\cdot}1^{a}$	$138{\cdot}9^{b}$	$49{\cdot}1^{a}$	$106{\cdot}2^{b}$	$67{\cdot}2^{a}$
Shoot WSC (%)						
Helga	11·04	11·40	8·31	7·66	6·47	6·30
Melina	11·11	11·89	8·21	8·13	6·97	6·68
DK 261	12·21	11·79	8·57	7·83	6·96	6·06
Ramses	14·02	13·29	9·60	9·08	7·22	6·70
mean	12·10	12·09	$8{\cdot}66^{b}$	$8{\cdot}22^{a}$	$6{\cdot}90^{b}$	$6{\cdot}44^{a}$
Root WSC (%)						
Helga	$4{\cdot}90^{abc}$	$5{\cdot}12^{bc}$	$6{\cdot}66^{b}$	$5{\cdot}19^{a}$	3·33	3·01
Melina	$4{\cdot}35^{a}$	$4{\cdot}45^{ab}$	$5{\cdot}61^{a}$	$4{\cdot}78^{a}$	3·32	2·86
DK 261	$5{\cdot}93^{de}$	$4{\cdot}61^{abc}$	$6{\cdot}69^{b}$	$4{\cdot}90^{a}$	3·61	2·85
Ramses	$6{\cdot}28^{e}$	$5{\cdot}26^{cd}$	$7{\cdot}80^{c}$	$4{\cdot}58^{a}$	4·31	3·21
mean	5·36	4·86	6·67	4·85	$3{\cdot}64^{b}$	$2{\cdot}98^{a}$

water with termamyl (heat-stable α-amylase, Novo Nordisk Ferment Ltd., Dittingen, Switzerland). After cooling to room temperature, 2 ml NaOH-buffer (pH 4·6) was added. The starch was hydrolysed with amyloglucosidase in a water bath at 60 °C for 30 min. The glucose released was assayed with hexokinase/glucose-6-phosphate-dehydrogenase (Boehringer Mannheim GmbH, 1989). Some of the samples were also incubated twice for 60 min with 80 % ethanol and subsequently analysed for soluble carbohydrates and starch as described above. No significant differences in the detected concentrations of soluble carbohydrates and starch were found between the two extraction methods.

RESULTS

At all harvests, dry matter production of the hybrids Melina and Helga was restricted in both shoot and root if supplied with ammonium (Table 2). Helga suffered most when given ammonium only, as indicated by the dramatic restriction in growth rate of up to approx. 50 % compared with Helga receiving nitrate during the first 2 weeks of treatment. In contrast, ammonium nutrition did not significantly inhibit root or shoot dry matter production in the hybrids Ramses and DK 261. After 28 d of exposure to treatments, none of the hybrids differed in shoot RGR for ammonium- and nitrate-fed plants (Fig. 1). In the case of the most ammonium-susceptible hybrid, Helga, ammonium caused a greater restriction in root growth than in shoot growth. This resulted in a higher shoot/root ratio throughout the experiment (data not shown).

For ammonium-grown plants after 14 d of treatment, Helga had an ammonium concentration of 63 μmol g^{-1}, which was nearly double that of the other hybrids (Table 2). For all hybrids, ammonium concentrations declined to between 19 and 26 μmol g^{-1} at the end of the experiment (Table 2). Ammonium concentration in the roots was generally higher than in the shoots for all hybrids (Table 2). There was no cultivar effect on the ammonium concentration in the roots. The shoot and root nitrate concentrations of the hybrids ranged between 700 and 1300 μmol g^{-1} when

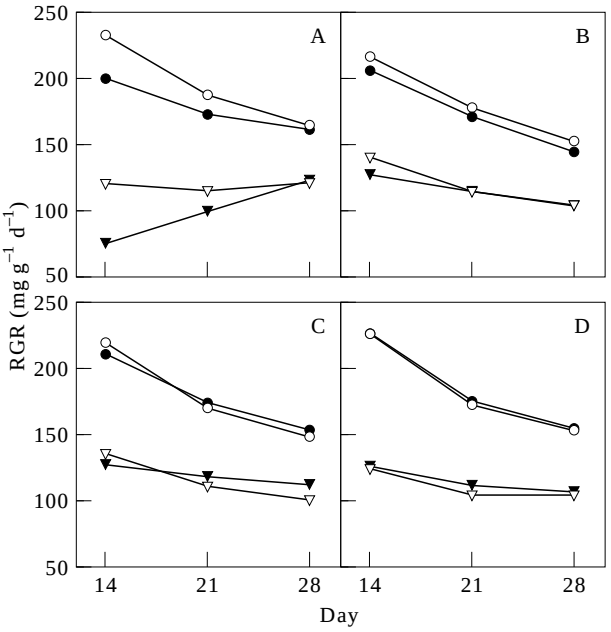

FIG. 1. Relative growth rates of shoots (○,●) and roots (▽,▼) of four maize hybrids exposed to either 20 mM ammonium (●,▼) or nitrate (○,▽). A, Helga; B, Melina; C, DK 261; D, Ramses.

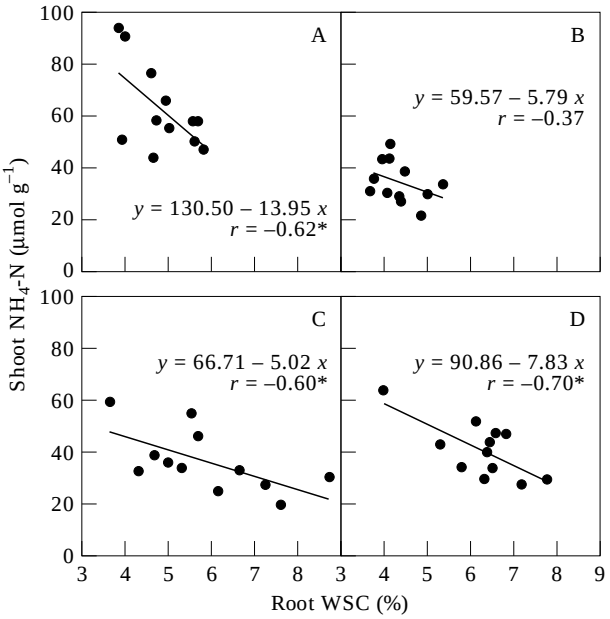

FIG. 2. Regression between root WSC concentration and shoot ammonium concentration for four maize hybrids after 14 d of exposure to 20 mM ammonium. Each point represents a single plant. * indicates significant correlation at $\alpha = 0{\cdot}05$. A, Helga; B, Melina; C, DK 261; D, Ramses.

supplied with nitrate (data not shown). Although no nitrate was applied in the ammonium treatment, and no nitrate contamination was detectable in the ammonium-based solutions, plants showed nitrate concentrations in roots and shoots of about 200 μmol g^{-1} at all harvests (data not shown).

The concentration of WSC in the shoot was slightly higher for Ramses than for the other genotypes at the first two harvests (Table 2). At the end of the experimental phase, the shoot WSC concentration had generally declined to the same value in all cultivars, between 6 and 7%. However, when the average value among hybrids was considered, no effect of the nitrogen form could be determined with certainty (Table 2). The starch concentrations of the shoot declined from 4% at the first harvest to 1% at the end of the experiment, but were little affected by either genotype or nitrogen form (data not shown).

Much greater differences among the hybrids occurred in the concentration of WSC in the root (Table 2). Under nitrate nutrition, the hybrids showed comparable WSC concentrations after two weeks of treatment, between 4 and 5%. For the ammonium-tolerant hybrids, Ramses and DK 261, the plants adapted to ammonium nutrition in the first 2 weeks of the experiment by substantially increasing the WSC concentration in the roots to 6%. In contrast, Melina and Helga showed a less pronounced response to ammonium feeding. After 14 d of treatment, the WSC concentration of the ammonium-fed and nitrate-fed roots was similar for the two ammonium-sensitive hybrids. In the third and fourth week of the experiment, the WSC concentration of ammonium-fed roots was still higher for the ammonium-tolerant hybrids Ramses and DK 261 than for the sensitive hybrids. In fact, the average WSC concentration was highest for Ramses and lowest for Melina, regardless of harvest date.

When ammonium only was supplied, the concentration of ammonium in the shoots could be related to the concentration of WSC in the roots. Figure 2 shows the relationship between the WSC concentration in the roots and the ammonium concentration in the shoots within each cultivar for the first harvest. The correlation between root WSC and shoot ammonium was significant at $P \leqslant 0{\cdot}05$ for the cultivars Helga, Ramses, and DK 261. No significant correlation between WSC concentration in the roots and ammonium concentration in the shoots was observed at the other harvest dates.

DISCUSSION

Our data show that there is genotypic variability in ammonium sensitivity or tolerance of maize, with the hybrid Helga being severely restricted in root and shoot growth if supplied only with ammonium (Table 2). Two hybrids, Ramses and DK 261, were completely insensitive to the nitrogen source, even at the high concentration of 20 mM in our experiment. Such concentrations are not unnaturally high when compared with fertilized soils, where ammonium concentrations higher than 20 mM may occur in the soil solution as far as 40 mm from a fertilizer band (Moody *et al.*, 1995).

Growth restrictions due to ammonium nutrition occurred in the early seedling stage, as shown by the RGR of shoots and roots during the first 3 weeks of the experiment.

Towards the end of the experiment, root and shoot RGR of Helga acclimatized to the ammonium supply, leaving little difference between ammonium- and nitrate-fed plants (Fig. 1). Thus, the low final biomass yield of Helga is explained mostly by reduced growth rates in the first 2 weeks of the experiment.

Although numerous studies reported plant growth inhibitions when only ammonium was supplied, little information is available about the developmental stages when these inhibitions occur. To our knowledge, there is no other study which investigates the effect of nitrogen source on RGR over time, except that of Atkin and Cummins (1994). In their study, the RGR of the arctic herb *Oxyria digyna* remained constant for 28 d under ammonium supply, whereas the RGR of nitrate-fed *Oxyria* exceeded that of ammonium during the first 3 weeks of their experiment and then decreased below the RGR of ammonium-fed plants. However, this pattern could not be observed for the slower-growing *Dryas integrifolia*, which did not respond to nitrogen form treatments (Atkin and Cummins, 1994). Nevertheless, it appears that for plants that are susceptible to ammonium stress, the early seedling stage is most responsive.

The growth reductions of Helga under ammonium nutrition were clearly correlated with the concentration of free ammonium found in the shoot (Table 2). One to 3-week old seedlings of Helga were apparently unable to detoxify all of the absorbed ammonium by assimilation (see Givan, 1974) or by ion release from the roots to the medium (see Morgan and Jackson, 1989). The suggestion that the concentrations of ammonium in Helga were in the toxic range is also supported by data from Gill and Reisenauer (1993), with growth of wheat inhibited if the ammonium concentration in the shoot tissue exceeded 43 μmol g^{-1}, a value which was clearly exceeded by Helga in the first phase of the present experiment.

High ammonium concentrations in the shoot tissue cause decreases in the net carbohydrate assimilation of plants (Cramer and Lewis, 1993*a*). As a consequence, less carbohydrate may be available for transfer to the roots. Therefore, less ammonium can be assimilated in the roots, and the excess of free ammonium moves to the shoots. This movement reduces, in turn, the production of carbohydrates and so forth. According to this, it is possible that reductions in growth due to excessive ammonium, as observed for Helga, have their origin in the fact that the adverse effects of insufficient assimilation of ammonium amplify themselves. We hypothesize that insufficient ammonium assimilation in the roots of Helga can be linked with insufficient supply of carbohydrates to the roots. The tolerant genotypes Ramses and DK 261 responded to ammonium nutrition by increasing the concentration of WSC in the roots by 29% and 19%, respectively, after 2 weeks of treatment. It appears that the ammonium-tolerant hybrids shifted their partitioning of carbohydrates towards the site where the bulk of nitrogen is assimilated, which was the shoot for nitrate-fed plants and the root for ammonium-fed plants.

The negative correlation between the WSC concentrations in the roots of ammonium-fed plants and the ammonium concentrations in the shoots within the hybrids (Fig. 2) further indicated that ammonium toxicity is probably a result of insufficient carbon supply to the roots. For the most susceptible hybrid, Helga, the slope of the regression was greatest, possibly indicating that this genotype reacts most sensitively to changes in the WSC concentration of the root. This action may be due to the feedback mechanism described above.

The consistently higher shoot:root dry matter ratios of Helga indicate that under ammonium nutrition, root growth suffered more than shoot growth. Restricted root growth under ammonium nutrition is a common observation: the assimilation of ammonium and root growth are competing for carbohydrates. The use of carbon skeletons for the assimilation of ammonium may be at the expense of root growth (Cramer and Lewis, 1993*b*; Schortemeyer *et al.*, 1993). For the present experiment, this scenario seems to have occurred only for the ammonium-sensitive hybrid, Helga.

It cannot, however, be generalized that hybrids characterized by high WSC concentrations in the root are ammonium-tolerant. The capability of a genotype to adapt to different nitrogen forms may not only depend on the absolute amount of carbohydrates in roots or shoots, but also on their flux between roots and shoots as a consequence of different sink demands. Concentrations of WSC in the tissue are only a rough measure for the availability of carbohydrates, since the distribution of WSC within the root may also be of importance. It is likely that factors other than WSC concentrations contribute to the cultivar variation in the sensitivity to ammonium feeding only. For example, the activity of ammonium assimilating enzymes may play a role (Tsai *et al.*, 1991). Furthermore, it is possible that there is genotype variation in the 'tolerance threshold' with regards to the concentrations of ammonium in the tissue.

It has been demonstrated that ammonium toxicity or growth reductions caused by ammonium can be alleviated by nitrate (Deignan and Lewis, 1988). This effect might also have happened in our experiment, since nitrate was detected in the plant tissue even when ammonium was the only nitrogen source applied. The occurrence of nitrate in ammonium-fed plants has been attributed to the contamination of the roots with nitrifying bacteria (Padgett and Leonard, 1993). It is therefore possible that in our experiment, ammonium toxicity symptoms would have been more severe without this steady supply of small portions of nitrate.

Even though it is difficult to translate results from hydroponic to field conditions, it is assumed that the reduced ammonium tolerance of some maize hybrids may retard development in the field. Due to the rapid conversion of ammonium to nitrate by soil micro-organisms, ammonium toxicity seldom appears to be a problem for crops, but seedlings may be subjected to localized high ammonium concentrations shortly after fertilization. This occurs predominantly in the phase from germination to the three-leaf stage of maize, which, according to our observations, seems to be most susceptible to ammonium toxicity. The use of ammonium-tolerant cultivars could prevent the expression of ammonium-induced growth reductions.

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