EFFECT OF N-SOURCE ON ACCUMULATION OF AMINO ACIDS IN SUGAR BEET

G. MÄCK

Institute of Sugar Beet Research, Göttingen, Germany

ABSTRACT

Nitrogen is taken up by the plant from the soil as nitrate (NO_3^-) and ammonium (NH_4^+) and reduced inside the plant to yield amino-N. Accumulation of amino-N in the beet is undesirable because it lowers the technical quality during the process of sugar crystallisation. The aim of the present study was to evaluate the effect of the N source of the plant on content and pattern of amino acids in the beet. The investigations were done on two genotypes which, in prior field trials, showed a different amino-N content of the beet (low- and high-N type). Seeds of these plants were germinated and the plants were grown hydroponically with either 5 mM NO_3^- or NH_4^+ as N source in the growth chamber for 40 days.

The results show that content and pattern of amino acids in the beet depend on the N source of the plant. Phenotypic differences between the two genotypes are reduced when NH_4^+ is supplied instead of NO_3^-. NH_4^+ as N source lowers the technical quality of the beet.

ABRÉGÉ - INFLUENCE DE LA SOURCE D'AZOTE SUR L'ACCUMULATION D'ACIDES AMINÉS DANS LA BETTERAVE SUCRIÈRE

L´azote que la plante absorbe du sol se présente sous la forme de nitrate (NO_3^-) ou d'ammonium (NH_4^-), deux substances qu'elle réduit en acides aminés. L'accumulation d'azote aminé dans la betterave est indésirable car elle détériore la qualité technique pendant la cristallisation du sucre. Cette étude avait pour but d'examiner les effets de la source d'azote de la plante sur la concentration et sur le schéma des acides aminés dans la betterave. Les analyses ont été accomplies sur deux génotypes qui, dans les essais du champ qui les avaient précédées, se distinguaient par la teneur en azote aminé de la betterave (type N haute et basse). Les plantes issues des graines de ces génotypes ont été cultivées pendant 40 jours de façon hydroponique en enceinte climatique aux paramètres contrôlés ; elles ont reçu, comme source d'azote, 5 mM soit de NO_3^- soit de NH_4^-.

Les résultats ont montré que la concentration et les schémas des acides aminés dans la betterave dépendent de la source d'azote offerte à la plante. Les différences phénotypiques entre ces deux génotypes diminuent lorsque la plante dispose de NH_4^- au lieu de NO_3^-. Le NH_4^- en tant que source d'azote préjudicie à la qualité technique de la betterave.

KURZFASSUNG - EINFLUSS DER N-QUELLE AUF DIE AKKUMULATION VON AMINOSÄUREN IN DER ZUCKERRÜBE

Stickstoff wird von der Pflanze aus dem Boden in Form von Nitrat (NO_3^-) oder Ammonium (NH_4^+) aufgenommen und in der Pflanze zu Aminosäuren reduziert. Die Akkumulation von Amino-N in der Rübe ist unerwünscht, denn sie verschlechtert die technische Qualität während der Zuckerkristallisation. Ziel der Studie war, den Effekt der N-Quelle der Pflanze auf Konzentration und Muster der Aminosäuren in der Rübe zu untersuchen. Die Untersuchungen wurden an zwei Genotypen durchgeführt, die sich in vorausgegangenen Feldstudien im Amino-N-Gehalt der Rübe unterschieden (hoch- und niedrig-N-Typ). Pflanzen aus den Samen dieser Genotypen wurden in der Klimakammer unter kontrollierten Bedingungen für 40 Tage hydroponisch angezogen mit entweder 5 mM NO_3^- oder NH_4^+ als N-Quelle.

Die Ergebnisse zeigen, dass Konzentration und Muster der Aminosäuren in der Rübe von der N-Quelle der Pflanze abhängen. Phänotypische Unterschiede zwischen den beiden Genotypen werden vermindert, wenn NH_4^+ anstelle von NO_3^- angeboten wird. NH_4^+ als N-Quelle verschlechtert die technische Qualität der Rübe.

INTRODUCTION

Amino-N compounds constitute 30 – 40 % of the so-called harmful nitrogen which considerably lowers the technical quality of the beet. A major aim in breeding is thus to minimize the concentration of N compounds in the beet. Amino-N is generated by the plant from nitrate (NO_3^-) and ammonium (NH_4^+). Nitrate is the predominant N source for plants in agricultural soils. Leaking of nitrate into the ground water as well as accumulation of nitrate in vegetables, however, bears risks for human health and natural ecosystems. Ammonium which is less mobile in the soil has thus received increasing interest as N fertilizer. Many herbaceous plant species, however, show deficiency symptoms when grown with ammonium as sole N source (CHAILLOU et al., 1986). The aim of the present study was therefore to investigate whether sugar beet tolerates ammonium as sole N source and whether ammonium affects the technical quality of the beet.

MATERIAL AND METHODS

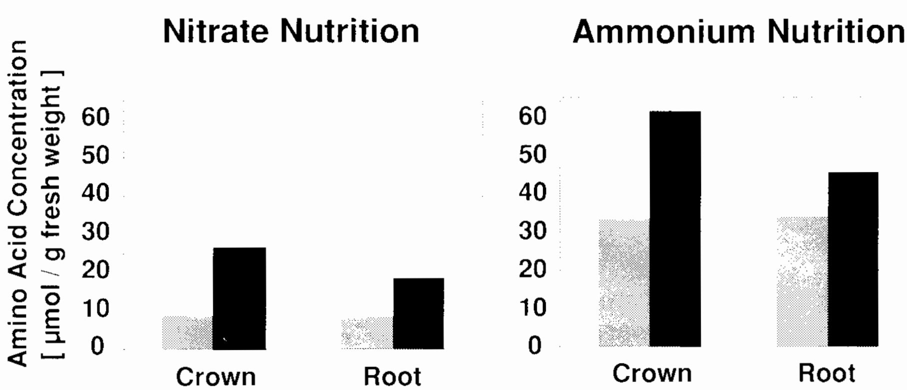
Two genotypes of sugar beet were selected from field trials; they were characterized by a different amino-N content of the beet (high- and low-N type). Seeds of these genotypes were germinated in vermiculite and thereafter the plants were grown hydroponically for 40 days in a growth chamber under controlled conditions. The plants were supplied with a full nutrient solution containing either 5 mM nitrate or ammonium as sole N source. The pH of the nutrient solution was adjusted to pH 6 and controlled by addition of solid $CaCO_3$ (GOYAL & HUFFAKER, 1986). At harvest, leaves and stems were removed from the thickened hypocotyl and the young beet was dissected into crown and root. Amino acids were extracted with methanol-chloroform and analysed by high pressure liquid chromatography (MÄCK, 1988).

RESULTS AND DISCUSSION

The 40-day-old plants had developed 5 pairs of foliage leaves, a thickened hypocotyl and a thickened root. No difference in plant growth with either nitrate or ammonium was observed at that stage as long as the pH of the nutrient solution was controlled carefully. Acidification as a result of ammonium uptake, however, resulted in reduced plant growth and symptoms of stress.

Ammonium, as compared to nitrate, elevated the concentration of total amino acids in crown and root. In the low-N genotype the level was increased 4-fold, but only 2-fold in the high-N type (Fig.1). Thus, although the concentration of amino acids was increased, the difference between the two genotypes became smaller after ammonium nutrition. When grown with nitrate, the genotypes differed by a factor of 4 and 2 in crown and root, respectively; when grown with ammonium the respective factors were 2 and 1.4.

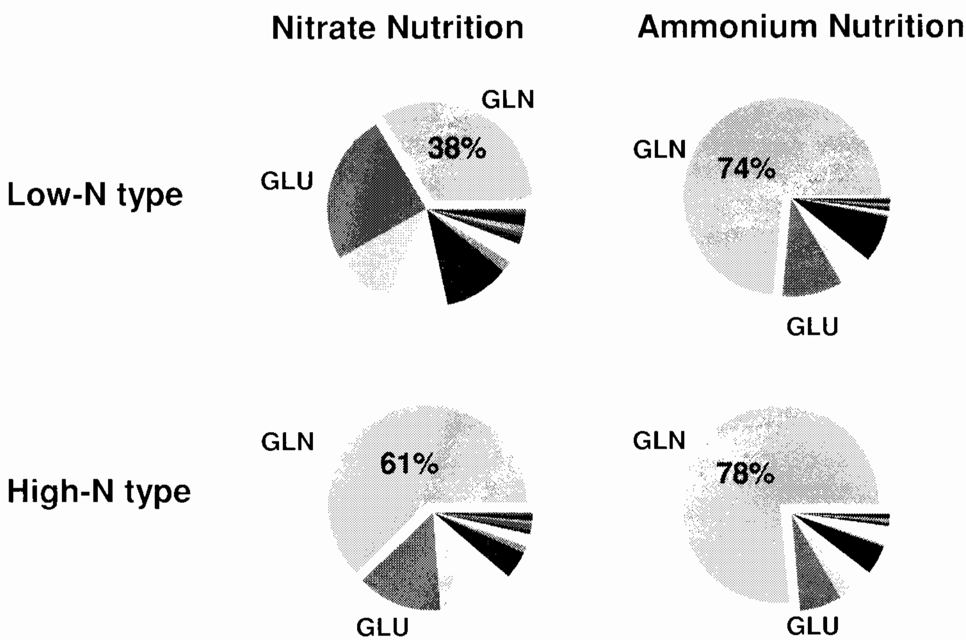
Figure 1: Concentration of total amino acids in crown and root of sugar beet. Two genotypes, a high-N type (black) and a low-N type (grey), were grown hydroponically with either 5 mM nitrate or ammonium as sole N source for 40 days.



The difference in the amino acid content of the two genotypes was more pronounced in the crown than in the root (Fig.1). This was probably due to the high density of vascular bundles in the crown; vessels of older leaves are connected to the inner cambial rings of the beet and are crossed by the vessels of young leaves which are connected to the outer rings (STIEBER & BEHRINGER, 1987).

It is generally reported that ammonium as N source increases the N content of plants (VAN BEUSICHEM et al., 1988). Amino acids like glutamine, asparagine, proline and γ-aminobutyric acid accumulate, most likely as storage compounds for excessive ammonium (SHELP et al., 1999). In sugar beet, α-amino-N increased not only in the beet but also in the leaves when grown with ammonium instead of nitrate (MÄCK, 1988). In crown and root the dominant amino acid was glutamine both when grown with nitrate or ammonium. When N was supplied as nitrate, 38 % and 61 % of total amino acids was glutamine in the root of the low-N and high-N genotype, respectively (Fig.2). With ammonium supply, the concentration of glutamine increased 7-fold and 3-fold in the root of the low-N and high-N genotype, respectively. This resulted in a proportion of glutamine of 74 % and 78 % of total amino acids in the low-N and high-N genotype, respectively (Fig.2).

Figure 2: Percentage of glutamine (GLN) relative to total amino acids in the root of sugar beet. Two genotypes, a low-N type (upper lane) and a high-N type (lower lane), were grown hydroponically with either 5 mM nitrate or ammonium as sole N source for 40 days.



Thus, when supplied with ammonium, the relative proportion of glutamine was similar in both genotypes whereas it differed by a factor of 2 when supplied with nitrate. This indicates that there is a limit for the increase of glutamine in response to ammonium supply. The limit seems to be reached when 70 to 80 % of all amino acids is glutamine. The capacity to respond to ammonium with an increase in glutamine was thus higher in the low-N genotype because its glutamine concentration under nitrate conditions was lower than that of the high-N genotype. Interestingly, when N was supplied as ammonium, arginine was measured in the roots of the high-N genotype but not of the low-N genotype (MÄCK, 1988). This indicates that excessive ammonium which could not be stored anymore as glutamine in the high-N genotype was stored as arginine. Arginine carries 4 N atoms per molecule and is thus a perfect storage compound for nitrogen.

CONCLUSION

To conclude, sugar beet tolerates ammonium as sole N source but the concentration of amino acids in crown and root increases considerably compared to plants supplied with nitrate. Thus, ammonium nutrition lowers the technical quality of the beet. Especially the concentration of glutamine increases and reaches a proportion of 70 % to 80 % of total amino acids which seems to be the upper limit for an accumulation of glutamine. Phenotypic differences between the genotypes become smaller when grown with ammonium instead of nitrate.

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