The relationship of proline accumulation to phosphorus content in oilseed rape

JANINA ROGOZIŃSKA, STANISŁAW FLASIŃSKI

Laboratory of Plant Physiology, Technical-Agricultural University, Bernardyńska 6, 85-029 Bydgoszcz, Poland

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Abstract

Phosphorus (P) deficiency in the medium caused proline accumulation in the leaves and stems of oilseed rape. This accumulation took place when the P level had decreased in the plant to ca 0.2% d.w. and proceeded very fast when the P content dropped below 0.1% d.w. The magnitude of proline accumulation depended strictly on the P level in the plant.

Key words: proline accumulation, P content, oilseed rape

INTRODUCTION

Amino acids play an essential role in plant metabolism, being the primary products of inorganic nitrogen assimilation and precursors of proteins and nucleic acids (Stewart and Larher 1980). As a result of nutrient deficiencies the amino acid metabolism is changed in many plants (Machicado and Boynton 1961, Stewart 1962, Labanauskas and Handy 1970). One of the most pronounced events is the accumulation of free proline associated not only with a number of mineral element deficiencies but also with various other stress conditions (Savickaya 1976, Stewart and Larher 1980, Flasiński et al. 1986).

This paper is a continuation of our previous studies regarding the effect of various stress conditions (nutrient, salt and osmotic stresses) on proline accumulation in oilseed rape (Rogozińska and Flasiński 1987). It was shown that one of the factors causing accumulation of this amino acid could be P deficiency in the nutrient solution. The aim of the present study is to demonstrate the relationship between proline accumulation and phosphorus content in oilseed rape.

MATERIAL AND METHODS

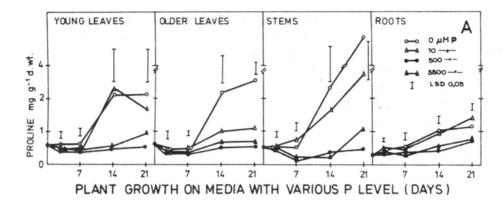
Oilseed rape (Brassica napus var. oleifera cv. Start) was cultured in a phytotrone (prototype OBRUCiG, Bydgoszcz) under 12/12 photoperiod and 70% humidity, 36 W m⁻² ligh intensity, temp $24\pm2^{\circ}$ C. The seeds were germinated on perlite, and 7 day-old seedlings were transferred to water culture on Hoagland's solution diluted 1:1. Next, three week-old plants were transferred to media with different phosphorus (P) levels: 0, 10, 500 (control), 3500 μ M. After 3, 7, 14 and 21 days of growth, the proline content according to the method of Bates et al. (1973) and phosphorus content by the method of Fiske and Subbarow (1925) were determined. Total P was estimated in the various organs after digestion of dried samples in 2.5 M $_{2}$ SO₄ with 70% $_{2}$ HClO₄ (5:1). The results from three replicates were subjected to variance analysis and the least significant differences (LSD) were calculated.

RESULTS

It is seen that proline accumulation depended on the P level in the medium (Fig. 1A). In oilseed rape plants grown on control medium (500 μ M P), the proline content amounted to ca 0.5 mg g⁻¹ d.w. On P deficient media (0, 10 μ M P) the proline content increased. Proline accumulation differed depending on the organ. The most significant accumulation took place in the stems, next in the leaves. Proline accumulation in the roots was low.

Proline started to accumulate after 7 days of stress and continued for the duration of the experiment. After 3 weeks the proline content in the leaves and stems increased ca 10 times and in the roots 4 times only. Distinctly higher proline accumulation in the plants growing on medium lacking P than in plants growing on P deficient medium occurred only in older leaves. In the case when the medium contained an excess of P, the proline content was only insignificantly higher than in the control, but the differences were statistically insignificant.

The phosphorus level in the media strongly affected the P content in the plant. A similar relationship as between proline accumulation and P level in the medium was found in relation to P content in the plant (Fig. 1B). When the P level decreased from 0.5 to 0.2% d.w. the increase in proline content was negligible. When, however, the P level in the plant dropped from 0.2 to 0.1% d.w. the proline content increased considerably. Thus, intensive proline accumulation occurred after the P level had decreased below 0.1% d.w. Proline accumulation in the plant could thus be some kind of an indicator, though not specific, of the P deficiency, both in the medium and in the plant.



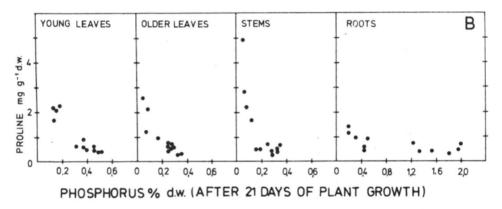


Fig. 1. Proline accumulation depending on P supply in the medium (A) and P content in the plant (B)

DISCUSSION

Some early responses of plants to P deficiency are ammonia accumulation and its removal by amino acid synthesis (Rabe and Lovatt 1986). In this case considerable changes in the free amino acid pool and amides content occur (Stewart and Larher 1980). As shown, P deficiency may affect the individual amino compounds differently in various plants. For example in P-deficient ryegrass the predominant accumulated compounds were asparagine and glutamine (Nowakowski et al. 1977) while in linseed plants, arginine and glutamic acid (Ranjan and Malaviya 1962). Proline accumulation and particularly of arginine and lysine in response to P deficiency was found also in several citrus plants (Rabe and Lovatt 1984). From all the amino acids accumulated under stress conditions, the most common in various plant species, also in oilseed rape, is proline. The literature concerning this amino acid is the most abundant.

It seems that the most specific linkage between amino acid and phosphate metabolism in plants occurs probably through pyridoxal phosphate (Achituv and Bar-Akiva 1976). In P deficiency the transamination of α-carboxylic acid would be depressed and production of amino acids shifted to the urea cycle. Under these conditions an increased proline and arginine content would be important in nitrogen transport and storage (Miflin and Lea 1977).

Strict relationship was shown between the P content in the plant and the amount of accumulated proline. Proline accumulation, however, is caused not only by P deficiency but also by other kinds of stresses.

REFERENCES

- Achituv M., Bar-Akiva A., 1976. Glutamic-oxaloacetic transaminase in leaves of phosphorus-deficient citrus plants. Physiol. Plant. 36: 368-373.
- Bates L. S., Waldren R. P., Teare I. D., 1973. Rapid determination of free proline for water-stress studies. Plant and Soil 39: 205-207.
- Fiske C. H., Subbarow Y., 1925. The colorimetric determination of phosphorus. J. Biol. Chem. 66: 375-400.
- Flasiński S., Rogozińska J., Drozdowska L., 1986. The effect of phosphorus and water deficit on phosphatase activity and proline accumulation in seedling cotyledons and roots of oilseed rape as compared to that of excised cotyledons and roots. Acta Soc. Bot. Pol. 55: 83-96.
- Labanauskas C. K., Handy M. F., 1970. The effects of iron and manganese deficiencies on protein amino acids in macadamia leaves. J. Amer. Soc. Hort. Sci. 95: 218-223.
- Machicado M., Boynton D., 1961. Effects of potassium, magnesium and calcium deficiencies of the intermediary nitrogenous constitutents of cocoa (*Theobroma cacao* L.) leaves. Proc. Amer. Soc. Hortic. Sci. 77: 231-235.
- Miflin B. J., Lea P. J., 1977. Amino acid metabolism. Ann. Rev. Plant Physiol. 28: 299-329.
 Nowakowski T. Z., Mattingly G. E. G., Lazarus W., 1977. Effect of nitrogen and phosphorus fertilizers on yield and on inorganic and organic composition of Italian ryegrass grown on phosphorus-deficient soil. J. Sci. Food Agric. 28: 491-500.
- Rabe E., Lovatt C. J., 1984. De novo arginine biosynthesis in leaves of phosphorus-deficient Citrus and Poncirus species. Plant Physiol. 76: 747-752.
- Rabe E., Lovatt C. J., 1986. Increased arginine biosynthesis during phosphorus deficiency.
 A response to the increased ammonia content of leaves. Plant Physiol. 81: 774-779.
- Ranjan S., Malaviya B., 1962. Effect of phosphorus deficiency on the free and protein bound amino acids of the linseed plant. Flora 152: 399-408.
- Rogozińska J., Flasiński S., 1987. The effect of nutrient, salt and osmotic stresses on proline accumulation in oilseed rape plants. Acta Physiol. Plant. 9: 61-68.
- Savickaya N. N., 1976. O fizjologicheskoj roli prolina w rastieniakh. Nauch. Dokl. Wyzhsh. Shkoly. Biol. Nauki 2: 49-61.
- Stewart I., 1962. The effect of minor element deficiencies of free amino acids in *Citrus* leaves. Proc. Amer. Soc. Hort. Sci. 81: 244-249.
- Stewart G. R., Larher F., 1980. Accumulation of amino acids and related compounds in relation to environmental stress. In: The biochemistry of plants. Vol. 5. Miflin B.J. (ed.), Acad. Press, New York. pp. 609-635.

Akumulacja proliny u rzepaku ozimego w zależności od zawartości fosforu

Streszczenie

Niedobór fosforu (P) w pożywce powoduje akumulację proliny w liściach i łodygach rzepaku. Proces ten rozpoczyna się, kiedy poziom P obniży się w roślinie do okoła 0.2% s.m. i pogłębia się w miarę dalszego zmniejszania zawartości P. Wielkość akumulacji zależy wyraźnie od poziomu P w roślinie.