Changes in nitrogen fractions and carbohydrates by foliar application of phosphorus under salt stress in peanut plants (Arachis hypogaea L.)[01/]

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COMPENDIO

Se cultivó maní (Arachis hypogaea *L. 'TMV-2') en macetas con arena salinizada con Na Cl a 0,4%. Se asperjó fosfato como Na H_2 PO_4 a 0,1% (p/v) con 0,01% (v/v) Tween-80, hasta el punto de goteo, una vez al día desde el 20º al 25º día y desde el 30º al 35º día, después del sembrío. Se cosecharon raíces, tallos y hojas al 30º y al 40º día para análisis. El rendimiento de materia seca fue acentuadamente reducido a causa de la salinidad y fue parcialmente restituido por la aplicación foliar de fósforo. La salinidad provocó una reducción del nitrógeno proteico y un aumento en el nitrógeno soluble. La aplicación de fósforo al follaje de plantas tratadas con sal condujo a una acentuada reducción del nitrógeno soluble y a un aumento en el nitrógeno proteico. El tratamiento de sal disminuyó el nivel de azúcares y aumentó el contenido de almidón. Sin embargo, la aspersión de fósforo aumentó los azúcares totales y bajó los niveles de almidón.*

Introduction

THE plant responses to salts are of agricultural as well as academic interest, since yields are reduced when the salt content of the soil exceeds the optimum value (1, 17, 20). Growth depression under saline and alkaline conditions also results from a sharp decrease in the absorption of nutrient elements such as phosphorus (12, 13, 16). The impairment of growth by any factor will lead to a change in the metabolite concentration (3). Consequently, all the metabolic processes are altered by salinity (23). Disturbances in carbohydrate and nitrogen metabolism due to salinity are reported by several investigators (7, 10, 18, 22). Much work has not been carried out in supplying the nutrients through foliage when they are not absorbed by the root system under saline and alkaline conditions. Since phosphate in particular, is reduced in its uptake by the root system under conditions of soil salinity (13), the study of growth, nitrogen, and carbohydrate metabolism in salt stressed peanut plants by foliar application of phosphorus was undertaken.

Materials and Methods

Peanut (*Arachis hypogaea*, L., Var. 'TMV-2') seeds of uniform size were surface sterilized, washed and sown in glazed pots containing acid washed sand. The temperatures ranged between 60° to 90° F. The experimental station was located in South India with 13°N latitude, 80°E longitude and 250 ft. above mean sea level. The seedlings were grown under natural photoperiod and deionized water was added to maintain the moisture content around 60 per cent of the water holding capacity of the sand. Salt treatment (0.4% of NaCl) was given 10 days after sowing. Hoagland and Arnon's (11) nutrient solution was supplied 15 days after sowing to all the plants. Phosphate as NaH_2PO_4 at 0.4% (w/v) with 0.01% (v/v) Tween-80 was sprayed to the plants to the drip point, once daily for 5 days from 20th day. On the 30th day, a set of plants were harvested for analyses. Five more sprays were given from 30th day after sowing and plants that received 10 sprays were harvested on 40th day for analyses. Thus the treatments were:

a) CC - non-salinized and sprayed with distilled water

b) CP - non-salinized and sprayed with phosphate solution

c) ClC - salinized and sprayed with distilled water

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d) CIP - salinized and sprayed with phosphate solution

The stages were:

0 - without P-treatment (20 days old plants)

1 - at the end of first period of treatment (30 days old plants)

2 - at the end of second period of treatment (40 days old plants)

The pots were covered by polyethylene hoods to avoid rainfall which might leach the salts from the pots

Roots, stems and leaves were separated, washed, oven dried and dry weights were taken. Total and protein nitrogen was estimated by microKjeldahl method using bromocresol green as an indicator (14, 21). Soluble nitrogen was obtained by subtracting protein nitrogen values from total nitrogen. Sugars were extracted in alcohol to estimate colorimetrically using phenol reagent (5). The residue left behind after alcohol extraction was taken for starch extraction and it was estimated following the method of McCready *et al* (15). The results presented are means of three replications

Results and discussion

The yield of all plant parts in control plants increased rapidly from 20 to 30 days and slightly at 40 days, whereas in salinized plants the increase during the experimental period was not as steep as in control (Table 1). The results are in agreement with the earlier studies (1, 13). Reduction in the uptake of phosphorus is one of the several factors that cause reduction in growth (12, 13, 16). When phosphorus was given through foliar sprays, dry weights of control as well as salinized plants increased more markedly when the plants received phosphate for two periods (Table 1). Thus the partial restoration of growth in salinized plants occurred with foliar supply of phosphorus when it is not taken up by the root system, is of considerable importance (13)

A rapid decrease in protein nitrogen with age was observed in all plant parts of both control and salinized plants. Foliar P increased the protein nitrogen content in control plants at 30 days without much change at 40 days (Table 2). However, with foliar spray of phosphate to salinized plants, the increase in protein nitrogen was consistent both at 30 and 40 days in all plant parts studied. In general, total nitrogen decreased with age in control plants whereas in salinized plants phosphate spray did not show any clear trend on total nitrogen (Table 2). Soluble nitrogen was high in salinized plants compared to control plants and in general, it increased with age. However with

Table 1.—Effect of foliar application of phosphorus under saline conditions on changes in dry weight (g/plant).

P-Treatment	Days After Sowing	CONTROL (CC)				CONTROL+P (CP)				SALINIZED (CIC)				SALINIZED+P (CIP)			
		Leaves	Stems	Roots	Total Plant	Leaves	Stems	Roots	Total Plant	Leaves	Stems	Roots	Total Plant	Leaves	Stems	Roots	Total Plant
0	20	0.21 ±0.02	0.11 ±0.12	0.07 ±0.01	0.39					0.16 ±0.08	0.10 ±0.09	0.04 ±0.08	0.31				
1	30	0.60 ±0.09	0.30 ±0.09	0.28 ±0.09	1.19	0.71 ±0.14	0.30	0.32 ±0.08	1.33	0.45 ±0.10	0.15 ±0.09	0.14 ±0.07	0.75	0.48 ±0.07	0.16 ±0.16	0.15 ±0.05	0.79
2	40	0.70 ±0.11	±0.08	0.30 ±0.13	1.32	0.75 ±0.07	0.35 ±0.09	0.34 ±0.07	1.44	0.53 ±0.09	0.18 ±0.09	0.17 ±0.05	0.88	0.60 ±0.06	0.20 ±0.07	0.18 ±0.07	0.99

Table 2.—Effect of foliar application of phosphorus under saline conditions on changes in total, protein, and soluble nitrogen contents (mg/g dry wt.).

P-Treatment	Days After Sowing	CONTROL (CC)				CONTROL+P (CP)				SALINIZED (CIC)				SALINIZED+P (CIP)			
		Leaves	Stems	Roots	Total Plant	Leaves	Stems	Roots	Total Plant	Leaves	Stems	Roots	Total Plant	Leaves	Stems	Roots	Total Plant
		Total Nitrogen															
0	20	7.07 ±0.12	5.79 ±0.09	4.45 ±0.17	17.31					5.96 ±0.31	4.31 ±0.40	3.92 ±0.26	14.19				
1	30	6.27 ±0.23	4.84 ±0.05	3.69 ±0.23	14.80	7.46 ±0.16	5.68 ±0.28	4.33 ±0.33	17.47	5.87 ±0.05	4.20 ±0.18	3.83 ±0.23	13.90	5.74 ±0.17	4.30 ±0.17	3.31 ±0.14	13.35
2	40	5.66 ±0.38	4.51 ±0.26	3.57 ±0.18	13.74	5.78 ±0.27	4.23 ±0.20	3.71 ±0.34	13.72	7.54 ±0.73	4.83 ±0.22	3.86 ±0.20	16.23	6.12 ±0.19	4.20 ±0.34	2.96 ±0.23	13.28
		Protein Nitrogen															
0	20	6.85 ±0.45	5.55 ±0.19	4.27 ±0.09	16.67 ±0.					4.95 ±0.09	3.70 ±0.33	3.83 ±0.26	12.48				
1	30	5.70 ±0.28	4.40 ±0.05	3.54 ±0.23	13.65 ±0.	7.01 ±0.36	5.33 ±0.20	4.16 ±0.40	16.50	4.10 ±0.26	3.75 ±0.21	3.00 ±0.11	10.85	4.55 ±0.17	4.25 ±0.05	3.27 ±0.16	12.07
2	40	5.19 ±0.28	4.33 ±0.05	3.29 ±0.11	12.81 ±0.	5.32 ±0.12	3.86 ±0.24	3.28 ±0.14	12.46	3.95 ±0.23	2.82 ±0.11	2.42 ±0.11	9.19	4.46 ±0.14	4.25 ±0.23	2.73 ±0.16	10.44
		Soluble Nitrogen															
0	20	0.22	0.24	0.18	0.64						1.01	6.61	0.09	1.17			
1	30	0.56	0.44	0.15	1.15	0.45	0.35	0.17	0.97	1.77	0.55	0.83	3.05	1.19	0.05	0.04	1.28
2	40	0.47	0.18	0.28	0.93	0.46	0.37	0.43	1.26	3.59	2.01	1.44	7.04	1.66	0.95	0.23	2.84

Table 3.—Effect of foliar application of phosphorus under saline conditions on changes in total sugars, and starch content (mg/g dry wt.).

P-Treatment	Days After Sowing	CONTROL (CC)				CONTROL+P (CP)				SALINIZED (CIC)				SALINIZED+P (CIP)			
		Leaves	Stems	Roots	Total Plant	Leaves	Stems	Roots	Total Plant	Leaves	Stems	Roots	Total Plant	Leaves	Stems	Roots	Total Plant
		Total Sugars															
0	20	16.7 ±0.26	10.3 ±0.35	11.5 ±0.0	38.5					10.4 ±0.38	8.5 ±0.43	9.0 ±0.10	27.9				
1	30	22.0 ±0.05	16.0 ±0.43	18.3 ±0.52	56.3	18.1 ±0.57	12.2 ±0.57	13.5 ±0.50	43.8	12.3 ±0.24	6.2 ±0.15	7.1 ±0.47	25.6	12.3 ±0.45	8.7 ±1.01	9.5 ±0.50	30.5
2	40	24.0 ±0.50	20.5 ±0.50	18.5 ±0.50	63.0	21.8 ±0.55	15.0 ±0.0	13.3 ±0.66	50.1	13.8 ±0.47	8.3 ±0.43	6.3 ±0.24	28.4	9.3 ±0.42	6.5 ±0.50	7.5 ±0.43	23.3
		Starch															
0	20	34.7 ±0.99	25.2 ±1.20	21.2 ±1.01	81.1					46.3 ±1.25	41.0 ±0.50	38.3 ±0.99	126.6				
1	30	28.0 ±0.43	20.2 ±1.32	11.8 ±0.68	60.0	19.0 ±0.50	13.5 ±0.0	16.3 ±0.69	46.8	39.5 ±0.50	35.5 ±0.87	26.8 ±0.59	101.8	38.8 ±0.69	27.2 ±0.57	14.2 ±0.94	80.2
2	40	23.3 ±1.14	17.7 ±1.50	16.2 ±1.45	57.2	14.5 ±0.0	14.0 ±0.5	10.2 ±0.5	33.7	31.0 ±0.87	20.8 ±0.73	23.0 ±1.32	74.8	30.5 ±2.29	23.8 ±1.25	19.5 ±1.01	73.8

foliar application of phosphate, a decrease in the leaves, stems and roots of control and salinized plants was observed (Table 2). Thus the reduction in protein and total nitrogen in salinized plants lends support to the findings of earlier investigations (4, 6, 18). Ergle and Eaton (8) noticed in cotton plants that at low P concentration, nitrate and protein nitrogen were lowered associated with an increase in soluble nitrogen which indicates disruption in protein synthesis. The P deficiency effects appear to be similar to the effect of salinity with regard to nitrogen fractions.

Carbohydrates are regarded as the starting point for both respiration and growth of the plant (2). In control, the total plant showed an increase in total sugar content with age whereas in chloride plants it decreased from 20 to 30 days and increased from 30 to 40 days. Carbohydrates accumulated more in leaves, followed by stems and roots (Table 3).

Starch content increased with salt treatment in all the plant parts studied (Table 3) as had also been observed by other investigators (9, 10). Bernstein (3) has emphasized that impairment of growth by any factor results in a change in the metabolite concentrations in view of the reduced consumption and that considerable accumulation of starch in tissues is often regarded by various authors as concerned in maintaining concentration gradient.

With foliar application of phosphate on control plants the total sugar content of the entire plant decreased by 22.4 per cent compared to the control, but the same treatment to salinized plants resulted in an increase of 19.6 per cent over chloride plants at 30 days. Considering the effect of phosphate spray to the control plants, it is interesting to note that the total sugars of different plant parts were considerably lowered while the starch content was also lowered in the leaves and stems but not in the roots; it may be noted that the control plants were growing vigorously and phosphate spray increased the phosphate content of these plants perhaps to an excess extent. Ergle and Eaton (8) observed that with high P content the carbohydrates are decreased.

Phosphate spray to salinized plants resulted in decreased starch content in general; the total sugar content however, showed fluctuations, it being lowered in the sprayed plants in leaves and stems at 40 days, while it was higher in the roots at 30 to 40 days. Ergle and Eaton (8) in their studies on deficient supply of phosphorus to the rooting medium, observed increased levels of reducing sugars and starch. Similarly Singh and Singh (19) noticed in mint plants, with P-deficiency, an increase in all sugar fractions in roots and leaves and reduction in stems. Although the results in the present study partially support the above observations, it should be remembered that the technique leading to P-deficiency in studies of the above mentioned authors (actual reduced supply of P in the rooting medium) is altogether different from the deficiency of P content in the salinized plants which is due to its reduced uptake by the roots. Thus the discrepancies can be attributed to some extent to salinization also in the present study. Moreover, fluctuations of carbohydrates in different plants under deficient P conditions are explained by Ergle and Eaton (8) on the basis that the curtailment of growth associated with P-deficiency is greatly due to photosynthetic inhibition.

Summary

Peanuts (*Arachis hypogaea* L., 'TMV-2') were raised in sand cultures salinized with NaCl at 0.4%. Phosphate as NaH_2PO_4 at 0.1% (w/v) with 0.01% (v/v) Tween-80 was sprayed to the drip point, once daily from 20th day to 25th day and from 30th day to 35th day after sowing. Roots, stems and leaves were harvested at 30th day and 40th day for analyses. The yield of dry matter was markedly reduced due to salinity which was partially restored by foliar application of phosphorus. Salinity resulted in reduction of protein nitrogen and increase in soluble nitrogen. Phosphorus supply to the foliage of salt-treated plants showed a marked reduction in soluble nitrogen with an increase in protein nitrogen. Salt treatment decreased the level of total sugars and increased starch content. However, phosphorus spray enhanced the total sugars and lowered the starch levels.

Literature cited

1. ASANA, R. D. and KALE, V. R. A study of salt tolerance of four varieties of wheat. Indian Journal of Plant Physiology 13: 5-22. 1965.

2. BEEVERS, H. Respiratory metabolism in plants. Row-Peterson Biological Monograph, New York, 1961.

3. BERNSTEIN, L. Salt affected soils and plants. The problem of arid zones. Proceedings of the Paris Symposium (Arid Zone Research XVIII) UNESCO 139-174. 1962.

4. BHARADWAJ, S. N. and RAO, I. M. Physiological studies on salt tolerance in crop plants. IX. Effect of NaCl and Na_2CO_3 on seedling respiration and growth of wheat and grain. Indian Journal of Plant Physiology 5:56-71. 1962.

5. DUBOIS, M., GILLES, K. A., HAMILTON, J. K., REBERS, P. A. and SMITH, F. Colorimetric method for determination of sugar and related substances. Analytical Chemistry 28: 350-356. 1956.

6. DYER, H. J. Influence of K and Na on the metabolism of peanut cotyledons during germination. Botanical Gazette 108: 570-589. 1947.

7. EATON, F. M. Toxicity and accumulation of chloride and sulphate salts on the growth of tomato. Journal of Agricultural Research 64: 357-399. 1942.

8. ERGLE, D. R. and EATON, F. M. Aspects of phosphorus metabolism in the cotton plant. Plant Physiology 32: 106-113. 1957.

9. GAUCH, H G and EATON, F M Effect of saline substrate on hourly levels of carbohydrates and inorganic constituents of barley plants Plant Physiology 17: 347-365 1942.

10 HAYWARD, H E and LONG, E M Anatomical and physiological responses of the tomato to varying concentrations of sodium chloride, sodium sulphate and nutrient solutions Botanical Gazette 102: 437-462 1941.

11. HOAGLAND, D R and ARNON, D. I The water culture method for growing plants without soil Circular 347 California Agricultural Experiment Station 1950.

12 KOVDA, U A Genesis and regime of saline soils. Part 2 Moskva-Leningrad Izdatel' Stvo Akademii NAUK USSR 1947

13. MALAKANDAIAH, N and RAJESWARARAO, G Effect of foliar application of phosphorus on growth and mineral composition in peanut plants *(Arachis hypogaea*, L) under salt-stress Plant and Soil 52: 41-48. 1979

14. MARKHAM, R A steam distillation apparatus for micro-Kjeldahl analysis Biochemistry Journal 36: 790-791 1942.

15. McCREADY, R M., GOGGOLZ, J., SILVIERA, U and OWENS, H S. Determination of starch and amylase in vegetables Application to peas Analytical Chemistry 29: 1156-1158. 1950

16. NIEMAN, R H and WILLIS, C Correlation between the suppression of glucose and phosphate uptake and release of protein from viable carrot root cells treated with monovalent cations Plant Physiology 48: 287-293 1971

17 OSAWA, T. Effect of various concentrations of sodium chloride on the growth, flowering, fruit bearing and chemical composition of egg plants in sand culture. Journal of Horticultural Association, Japan 26: 9-14 1957.

18 RAMANA, K V R. Salt induced and natural changes in metabolism during seedling growth of *Raphanus sativus*, L. Ph D Thesis, S V University, Tirupati, India 1968

19 SINGH, J N and SINGH, D P. Effect of phosphorus deficiency on carbohydrate metabolism of *Mentha arvensis* Plant Physiology 21: 1341-1347 1968

20 SUTCLIFFE, J F Mineral salt absorption in plants Pergamon, Press, New York, 1962

21 THIMMAN, M V and LOOS, G M Protein synthesis during water uptake by tuber tissues Plant Physiology 32: 275-279 1957

22 WADLEIGH, G H and AYERS, A D Growth and biochemical composition of bean plants as conditioned by soil moisture tension and salt concentration. Plant Physiology 20: 106-132 1945

23 ZHUKOVSKAYA N V Structure and function of plant cells under salinization - a review Soviet Plant Physiology 18: 551-553. 1971.

Notas y Comentarios

Y ahora, melocotoneros enanos

Los tractoristas podrían pronto estar cosechando melocotones casi como ahora cosechan trigo, con una cosechadora que economiza mano de obra al cortar toda la planta por la base y al peinar después la fruta de las ramas Para obtener este grado de mecanización, los fruticultores tendrán que emplear el método de cultivo llamodo "prado-huerto", en el que se plantan árboles pequeños juntos unos de otros, y que rebrotan todos los años, después de cada cosecha La idea se originó en la Estación Experimental de Long Ashton, de Inglaterra, para los productores de manzanas. Todavía no es económico para manzanas, pero los investigadores en los Estados Unidos están optimistas en su aplicación a los melocotoneros

La mano de obra temporal para la cosecha de frutas es escasa en muchas partes de Europa y de América del Norte. Las máquinas cosechadoras existentes no pueden llenar la brecha del todo Estas máquinas, que funcionan sacudiendo el árbol y cogiendo la fruta al caer, son apropiados para proveer fruta para los fabricantes de conservas pero no para las fruterías; muy poca fruta sobrevive la caída a la red colectora sin sufrir daños y no está entonces lista para el ojo crítico del comprador. El cortar pequeños árboles por la base hace relativamente fácil para una máquina cosechadora recoger la fruta sin daños peinándola de las ramas cortadas con "dedos" de caucho. El secreto es cultivar los pequeños árboles económicamente y uniformemente

La mayor valla económica está en los costos iniciales. Para mantener rendimientos altos por hectárea con sus árboles pequeños, el sistema prado-huerto requiere unas 300 veces el número de árboles que un huerto tradicional, o sea, unos 65 000 por hectárea. Al precio usual de alrededor de US$ 1,40 por una plantita de melocotonero norteamericano o de $ 5,50 por un arbolito de manzano británico, esto es prohibitivamente caro Pero un nuevo método de obtener plantitas de melocotonero provenientes de estacas, desarrollado por investigadores de la Universidad de Georgia, ha bajado el costo por árbol a unos 24 centavos, al evadir la necesidad de hacer injertos

Las plantas que se están probando ahora parecen promisoras En Georgia, el promedio de rendimiento por hectárea es de unos 8 000 dólares de melocotoneros, más que suficiente para recuperar la inversión inicial de 2 000 dólares en árboles y una enorme mejora en los normales cinco a ocho años necesario para que los árboles maduren en un huerto convencional. Aún mejor, algunas áreas están todavía rindiendo bien, aunque a niveles ligeramente menores, después de ocho años de cosechas

Todavía quedan algunas cuestiones pendientes sobre cuánto tiempo se pueden mantener estos rendimientos Nuevos brotes emergen naturalmente de los tocones cosechados, pero la competencia por los nutrimentos, entre los árboles densamente plantados, es intensa Algunos investigadores se preguntan si esta competencia no incidirá eventualmente en los rendimientos, particularmente al depender una buena cosecha de que todos los frutos maduren simultáneamente para la labor eficiente de la cosechadora Es esencial una cuidadosa vigilancia de los nutrimentos del suelo y la aplicación de productos químicos para controlar el crecimiento y la fructificación.