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EFFECT OF 2, 4-DINITROPHENOL AND ATP ON UPTAKE, TRANSLOCATION AND DISTRIBUTION OF ^{32}P IN COTTON PLANTS UNDER DIFFERENT LIGHT CONDITIONS

N. I. ASHOUR, R. BARAKAT AND T. A. NOUR
National Research Centre, Dokki, Cairo-AREgypt

ABSTRACT

Cotton seedlings were subjected, in light and darkness, to DNP, ATP or both for 3 hours, then allowed to remain in contact with ^{32}P for further 6 hours. Darkness decreased phosphorus uptake and its translocation. In the light, DNP decreased the uptake of ^{32}P, but enhanced its translocation to the shoot. ATP did not affect ^{32}P uptake but enhanced its translocation. ATP could not overcome the negative effect of DNP on ^{32}P uptake. In the dark, neither DNP nor ATP exerted significant effect on ^{32}P uptake, yet accelerated its translocation. Under both light conditions, DNP accumulated most of the translocated ^{32}P in the stem, while ATP moved it further to the leaves. The enhancement of ^{32}P translocation was accompanied by the presence of higher percentage of ^{32}P in an organic form. It was concluded that high energy compound formed during photosynthesis may play a role in the metabolic active uptake of phosphorus.

INTRODUCTION

THE uptake of phosphorus and its translocation in plants were suggested to be metabolic active processes (Brower, 1965 and El-Fouly and Ashour, 1969). The energy required for such a process is supplied from the adenosine-triphosphate (ATP) deposited in the cells (Weigh, 1963). The high energy compound (ATP) is produced during the process of oxidative phosphorylation (Jackson *et al.*, 1962). Thus, 2, 4-dinitrophenol (DNP), an inhibitor of oxidative phosphorylation was found to decrease the uptake of phosphorus by plant roots (Stenlid, 1959).

The uptake of phosphorus by roots and its translocation to the shoot are higher in the light than in the dark (Linser, 1965), and increase with increasing light intensity (Ashour *et al.*, 1968). Raven (1969) suggested that the ATP required for regulation of ion pump can be produced in the light by cyclic photosynthetic phosphorylation.

The aim of this work was to investigate the effect of DNP and ATP on ^{32}P uptake, translocation and distribution in the cotton seedlings in light and in darkness.

MATERIALS AND METHODS

One month old, uniform cotton seedlings (*G. baradense*) cv. Ashmouni grown in water culture were selected. The plants were rinsed in large test tubes (2·5 cm dia and 20 cm long) filled with distilled water and left over-night. On the second day, the distilled water in each test tube was replaced by 50 ml of 1/4 strength complete Hoagland's nutrient solution containing 10^{-4} M 2, 4-dinitrophenol (DNP), 10^{-3} M ATP (the dipotassium salt of ATP) or both and left for 3 hours. Then 8μ Ci of ^{32}P as KH_2PO_4 supplied from the Egyptian Atomic Energy Establishment was injected in the nutrient solution in each tube, and the plants were allowed to remain in contact with the ^{32}P for 6 hours at 21° C. This experiment was conducted under conditions of both light (20·000 Lux) of fluorescent lamps, and darkness. Each treatment under both conditions had seven replicated tubes, thus each treatment included seven plants. At the end of the incubation period, the plants of four replicates were harvested, and the roots were washed carefully with running water for 2 min. then the plants were divided into roots,

stems and leaves and oven-dried separately at 105° C. Samples from different parts were prepared for counting on the same day. The shoots of the other three plants from each treatment were cut off 1 cm above the base. Filter paper (2 cm dia) was placed on the stems section for 15 min. to absorb all the translocated ^{32}P towards the stem of the plant. The radioactive circles were dried, counted, then washed with successive portions of 7% and 2% ice-cold trichloroacetic acid (TCA)– and finally with water to discard the inorganic phosphorus (Tobbert and Wiebe, 1955). The TCA-treated circles were counted once more. The percentage of inorganic– and organic–^{32}P was calculated.

Results

Under light conditions, DNP significantly decreased the uptake of ^{32}P, whereas ATP was without significant effect (Table I). The addition of ATP together with DNP did not change the uptake of ^{32}P more than that induced by DNP alone. In darkness, cotton plants absorbed much less amount of ^{32}P than in light. Under dark conditions, neither DNP nor ATP had any significant effect on the uptake of ^{32}P by plant roots as compared with the control plants. Combined treatment with DNP + ATP in the dark did not affect the uptake of ^{32}P.

Table I also shows that the darkness markedly retarded the translocation of absorbed ^{32}P from the root to the shoot of cotton plants. All treatments of metabolically active substances enhanced the translocation of the absorbed ^{32}P from the root to the shoot under both light conditions, the combined treatment of DNP + ATP was the most effective one.

In Table II it is clear that in the light DNP and ATP retained more or less an equal percentage of ^{32}P in the roots. However, much more of the ^{32}P that moved out of the roots in DNP-treated plants remained in the stems in comparison with the ATP-treated plants, where, further moving of ^{32}P towards the leaves was observed. In the dark, most of the absorbed ^{32}P was retained in the roots, while only traces were found in the stem and the leaves as compared with that in the light. However, the change in ^{32}P distribution among different plant organs in the dark due to DNP or ATP treatments was the same as observed in the light. When DNP and ATP were applied together most of the translocated ^{32}P under light conditions was accumulated in the leaves; whereas in darkness it was retained mainly in the stem.

Table III shows that in the light, all treatments increased the percentage of organic–^{32}P in the root

Table I

Effect of DNP and ATP on the uptake and the transport of ^{32}P in cotton plants in light and in darkness

Treatment	The uptake of ^{32}P				Transport index*	
	$\times 10^3$ counts/sec/plant		$\times 10^3$ counts/sec/g. root			
	Light	Dark	Light	Dark	Light	Dark
Control	326	213	1270	620	32·1	13·1
DNP	275	201	631	732	64·3	32·2
ATP	306	180	1062	599	65·4	28·5
DNP+ATP	237	208	787	702	86·6	68·6
P=0·05	37		123			

*Transport index = $\frac{^{32}P \text{ in shoot}}{^{32}P \text{ in whole plant}} \times 100$

Table II

Effect of DNP and ATP on the distribution of ^{32}P in cotton plant in light and in darkness

Treatment	Roots		Stem		Leaves	
	Light	Dark	Light	Dark	Light	Dark
	$\times 10^3$ counts/30 sec/organ					
Control	221	185	92	26	12·3	1·5
DNP	98	137	163	64	13·8	0·8
ATP	106	129	117	46	83·2	5·4
DNP+ATP	32	65	84	100	121·6	42·3
P=0·05	28		18		2·8	
	% of total radioactivity					
Control	67·9	86·9	28·3	12·4	3·8	0·7
DNP	35·7	67·8	59·3	31·8	5·0	0·4
ATP	34·6	71·5	38·2	25·5	27·2	3·0
DNP+ATP	13·4	31·4	35·4	48·2	51·2	20·4

Table III

Effect of DNP and ATP on the percentage of organic and inorganic ^{32}P in the root exudate of cotton plant in light and in darkness

Treatment	Organic-^{32}P		Inorganic-^{32}P	
	Light	Dark	Light	Dark
Control	49·7	68·6	50·3	31·4
DNP	69·2	84·3	30·8	15·7
ATP	79·3	66·0	20·7	34·0
DNP+ATP	79·6	91·3	20·4	8·7

exudate at the expense of the inorganic fraction. Darkness showed similar effect. Under such dark conditions, DNP also increased the percentage of organic–^{32}P in the root exudate, ATP was uneffective, while DNP + ATP appreciably increased it as compared with that in the control plants.

Discussion

The results indicate that light enhances the uptake of phosphorus by the roots of cotton plants and its translocation towards the stem thus confirming the results obtained by others (Linser, 1965 and Ashour *et al.*, 1968). Such effect was suggested by McEvoy (1967) to be due to the increased supply of the photosynthate under light conditions. In the light, the decrease in the uptake of ^{32}P after treatment with DNP, may be due to the inactivation of the phosphorylation processes in plant tissues. Under such conditions the formation of ATP was found to be partially blocked (Jachson *et al.*, 1962). However, in the dark, when only the oxidative phosphorylation was acting and not the photosynthetic phosphorylation, the DNP and the ATP were without effect on the uptake of ^{32}P. Thus, it seems that the high energy compounds formed during photosynthesis alongside with the downwards photosynthate may take part in the metabolic active uptake of phosphorus. On the other hand, when ATP is present in the root medium, the uptake of ^{32}P was not activated, but on the contrary, may be slightly retarded. A competitive effect between the molecule of ATP or its derivatives and the ion of phosphorus for a certain carrier was suggested for the explanation of such phenomenon (Vakhmistrov and Listova, 1967).

The translocation of ^{32}P from the root to the shoot was enhanced under both conditions of light due to DNP or ATP treatments, while DNP + ATP seemed to have an additive effect. It seems that high energy phosphorus compound is required for translocation. Randal and Vose (1963) found that DNP had a major possitive effect on the translocation of phosphorus to the shoots. In addition, it seems that when the translocation of phosphorus from the root was enhanced, the organic fraction of the translocated phosphorus was increased indicating a change in phosphorus metabolism. Further studies are needed to clarify the problem of translocation of phosphorus compounds in connection with the role of DNP, ATP and light.

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PROPAGATION OF *DIOSCOREA FLORIBUNDA* FROM *IN VITRO* CULTURE OF SINGLE-NODE STEM SEGMENTS

H. C. CHATURVEDI

Tissue Culture Laboratory, National Botanic Gardens, Lucknow

Abstract

Dioscorea floribunda plants were established in aseptic cultures from surface-sterilized single-node stem segments of field-grown vines. Axillary buds of nodal segments proliferated in presence of 6-benzylaminopurine (2 mg/l) unaccompanied by root formation. Whereas, shoot apices and single-node leaf cuttings rooted 100% in presence of NAA (0·5 mg/l), resulting into plantless, 100% of which were successfully grown in potted soil. It took about 40 days to obtain a 5–6 leaved plantlet in potted soil from single-node cutting taken from a plant grown in aseptic culture.

Introduction

DIOSCOREA FLORIBUNDA Mart. and Gal. is one of the three *Dioscorea* spp. (the other two are *D. composita* Hemsl. and *D deltoidea* Wall.) commercially yielding diosgenin, a main precursor, from plant source, for the synthesis of steroidal drugs, namely, cortisone, sex hormones, oral contraceptive pill, etc, which are so important in