Calcium uptake by cowpea as influenced by mycorrhizal colonization and water stress

G. Pai, D. J. Bagyaraj, T. Padmavathi Ravindra and T. G. Prasad*

Department of Agricultural Microbiology, *Department of Crop Physiology, University of Agricultural Sciences, GKVK, Bangalore 560 065, India

The role of vesicular-arbuscular mycorrhizal (VAM) colonization on calcium uptake was studied under different levels of moisture stress. Pots maintained at different moisture levels were given water containing known amount of radioactive calcium. The radioactivity in different parts of the plant was assessed 60 h after giving ^{45}Ca to the soil. High ^{45}Ca activity was present in all parts of VAM plants compared to non-mycorrhizal plants at all levels of moisture stress.

IT is estimated that about 90% of vascular plants harbour vesicular-arbuscular mycorrhizal (VAM) fungi in their root system[1]. It is also well established that these fungi improve plant growth, mainly through phosphorus nutrition[1,2]. The other beneficial effects are their role in the biological control of root pathogens, biological nitrogen fixation and hormone production. These VAM fungi can also cause changes in plant water relations and improve drought resistance[3-5]. Changes caused in water relations were attributed to improved phosphorus nutrition[6]. The improved nutrient uptake by VAM fungi is not confined to phosphorus alone, as these fungi also help in the uptake of other nutrients such as Zn, Cu, Ca etc.[7,8]. Calcium, an essential element which confers integrity to membranes[9], is also involved in phosphorus transfer to the host from the fungus because of its ability to stimulate alkaline phosphatase activity[10]. Enhanced Ca uptake in VAM plants improving host-water relations and plant hormone levels was reported by Rhodes and Gerdemann[8]. Calcium is now recognized as a second messenger which influences a number of physiological and biochemical processess in plants[11]. The present study attempts to understand the role of VAM fungi on growth, calcium uptake and water relations of cowpea. A pot culture experiment was conducted to study the influence of VAM inoculation at different levels of moisture stress on Ca uptake by cowpea.

Plastic pots of 17.5 × 20.0 cm holding 6 kg sterilized soil with 5 ppm applied phosphorus were used. Six cowpea seeds of a relatively drought-resistant variety C-152 were sown per pot at 3 spots (with 2 seeds at each spot). Treatments with VAM inoculation received *Glomus fasciculatum* (Thaxt) Gerd and Trappe inoculum at the rate of 0.21×10^4 infective propagules per pot. The inoculum was placed 2 cm below the seeding spot during sowing. The pots were maintained in a glass house. After 15 days of sowing, three seedlings were maintained per pot and the following treatments were imposed.

T_1 = Non-mycorrhizal (NM) cowpea maintained at field capacity (FC), T_2 = Mycorrhizal (VAM) cowpea maintained at FC, T_3 = NM cowpea maintained at 75% FC, T_4 = VAM cowpea maintained at 75% FC, T_5 = NM cowpea maintained at 50% FC, T_6 = VAM cowpea maintained at 50% FC.

Table 1. Influence of VAM fungi, moisture levels and their interaction on ^{45}Ca activity per leaves, stem and root, ^{45}Ca activity/plant, DPM/g plant sample and DPM/g root weight

Treatment		^{45}Ca activity/plant (DPM) Leaves ($\times 10^4$)	Stem ($\times 10^4$)	Root ($\times 10^4$)	Total ^{45}Ca/plant ($\times 10^4$)	DPM/g plant sample ($\times 10^4$)	DPM/g Root wt ($\times 10^4$)
NM		6 7	5 4	5 0	17.2	18 2	64 5
	M	21 2	24 8	11.5	61.4	17.8	120 1
100%	FC	16 7	11 0	4 6	32 0	17.9	94 3
75%	FC	18 4	18 9	11.0	48 3	17.4	85 0
50%	FC	6 8	15 4	9 2	37 6	18 6	97 7
Moisture NM/M level							
100%	NM	5 7	5 4	4 0	15.0	18.4	57.7
FC	M	27 6	16 7	5 2	48 9	17.3	130 8
75%	NM	12.7	9 5	8 0	30.1	18 6	69 9
FC	M	21.1	28 4	14 0	66.5	16 3	100 0
50%	NM	1.83	1.48	3 3	6 6	17 5	65 9
FC	M	24.2	29 3	15 5	68 6	19.7	129.5

Six replications per treatment were maintained. The pots were weighed every morning to estimate the water lost and the required amount of water was added to bring back the water status of the soil to the desired level. Twenty days after imposing moisture stress treatments, 3 soil core spots, 2 cm deep were made uniformly in all the 6 pots. One millicurie of ^{45}Ca was diluted to 300 ml. Fifty ml of the diluted ^{45}Ca solution was added per pot through the soil cores. Sixty hours after adding ^{45}Ca plants were harvested and dried in an oven at 60°C for 5 days. Dry weight of the root, stem and leaves was recorded. The dried samples were powdered separately and 10 mg of each were taken and hydrolysed using 0.1 N NaOH. Radioactivity was measured using liquid scintillation counter (model ECIL, India) and radioactivity was expressed in disintegrations per minute (DPM) per gram plant samples.

The radioactivity in different parts of the plant assessed 60 h after giving ^{45}Ca to the soil is given in Table 1. Mycorrhizal plants showed high ^{45}Ca activity in all parts compared to NM plants at all the levels of moisture stress. With increase in moisture stress, the ^{45}Ca activity in the root of VAM plants increased while the activity in the leaf showed marked reduction. No definite trend in ^{45}Ca was observed in stem, although the activity in VAM plants was always high compared to NM plants. Increased root volume and surface area of absorption occurring in mycorrhizal plants[12] might have influenced ^{45}Ca uptake. Calcium is known to move from soil to plant by mass flow[13]. This indirectly indicates that high amount of calcium entered the plant system via increased water uptake. Rhodes and Gerdemann[8] have shown that hyphal translocation can account for the movement of some Ca in VAM roots.

Total ^{45}Ca activity expressed as DPM/plant was also markedly high in VAM plants at all levels of moisture stress. With increase in moisture stress the VAM plants showed significant increase in ^{45}Ca activity. Non-mycorrhizal plants at 75% FC showed higher ^{45}Ca activity compared to NM plants maintained at 100% FC. In plants grown at low soil moisture regimes high ^{45}Ca content was seen in root. At low moisture regimes, hyphae in VAM plants might have actively absorbed and translocated ^{45}Ca to roots and availability of water may be limiting to these plants for further translocation of ^{45}Ca from root to shoot.

When the total radioactivity in the plant was expressed per unit weight of root an interesting trend was observed. A marked increase in ^{45}Ca per g weight of root was observed at all levels of stress. Ca is known to be an essential element which confirms integrity to membrane[9] and differential movement of nutrients[14]. Ca may also be involved in P transfer to host because of its ability to stimulate alkaline phosphatase activity[10]. This suggests the possibility of nutritional aspect in maintaining positive water balance in plant under stress due to VAM association. Thus, the present results clearly bring out that VAM fungi help in the uptake of Ca, which in turn plays a role in phosphorus and water uptake by plants.

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Morphological variations, qualitative and quantitative changes in alkaloid pool in the protoclone progenies of *Hyoscyamus muticus* (Egyptian henbane)

C. C. Giri and P. S. Ahuja*

Department of Genetics, Osmania University, Hyderabad 500 007, India

* Plant Tissue Culture Division, Central Institute of Medicinal and Aromatic Plants (CIMAP), Lucknow 226 016, India

Protoplast-derived plants of *Hyoscyamus muticus* L. ($2n = 28$) from mesophyll source when transferred to glasshouse and field exhibited a wide range of morphological variations with respect to growth habit, biomass yield and differences in total alkaloid content and relative concentrations of hyoscyamine and scopolamine or hyoscine. Total alkaloid content in the protoclones varied from 0.2% to 1.7% on dry weight basis compared to 0.4% to 1.01% in control (hyoscyamine and scopolamine content was 68% and 8.6%), respectively. Scopolamine content in the