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# BIO-ORGANIC PRODUCTION OF MEDICINAL CONSTITUENTS FROM *DATURA STRAMONIUM*, L.

BY

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## ABSTRACT

Physiological studies in relation to development and growth of medicinal *D. stramonium* L. plants are rather restricted. This study illustrated seed bacterization affecting plant growth and alkaloids content as well as total count of total microbes in rhizosphere of *D. stramonium* grown under organic fertilizer. Quantitative data on the growth of *D. stramonium* plant were positively influenced by seed bacterization with *Azotobacter chroococcum* and organic amendment in absence of N-fertilization. The highest increase in growth rate (branching, stem diameter, leaf area and number; plant height. fresh and dry weight) of plants grown in clay loamy soil and fertilized with bio-organic were recorded is such case. This was followed descendingly by plants inoculated with *Az. chroococum* in the presence of N-fertilizer. Non-bio-organic treatments gave significantly lower growth rates of plants when compared with bio-organic ones.

The highest alkaloids content in plants was recorded for those cultivated in amended clay loamy soil inoculated with *Az. Chroococcum* being 1.45, 1.15, 1.05, 0.63 and 0.20% in leaves, stem, flowers, seeds and roots respectively of dry matter after 6 months post cultivation. In the absence of organic amendments the alkaloids content was significantly decreased.

The rhizosphere of *D. stramonium* roots, the highest increased in total microbes found in the amended soil (125.0 x $10^4$ cells $g^{-1}$ dry soil). While decreased in non-amended soil ones (115.2 x $10^4$ cells $g^{-1}$ dry soil).

**Key words:** *D. stramonium*, Organic fertilizer, *Az. Chroococcum* seed bacterization, Alkaloids.

## INTRODUCTION

Many higher plants accumulate extractable organic substances in sufficient quantities to be economically useful as chemical feed stocks or raw materials for various scientific, technological and commercial applications. Natural substances are employed, either directly, by a large number of industries and natural plant products (phytochemicals) figure prominently in several of these (Porde and Doty, 1981). Economically important plants serve as sources of industrial oils, resins, tannins, saponins, natural rubber, gums, waxes, dyes,

pharmaceuticals and many special products (Tayler *et al.*, 1981). In higher plants such compounds are often concentrated in seeds and vegetagive storage organs and are needed for physiological development because of their role in basic cell metabolism.

*Datura ceratocauia* ort (Solanaceae) is an aquatic, hollow-stemmed, prostrate, cruping plant know by the Mexicans as the maredic lornaloca or maddening plants. This species is the connecting link between herbaceus daturas and brugmansias (Griffin and Lin, 2000 and Nari *et al.*, 2005). Only a few alkaloids have been reported for *D. stramonium* (Lounasmaa and Taminnea, 1993). Knowledge of the complete alkaloid pattern is of interest not only phytochemically, but also in relation to aspects of alkaloid biogenesis metabolism and application in the plant biotechnology.

The present work was designed to study the effect of seed bacterization with an active strain of *Az. Chroococcum* and organic fertilizer on plant growth and production of active constituent (mainly hyoscyamine) of *Datura stramonium* plants, as well as density of *Az. chroococcum*.

## MATERIAL AND METHODS

The present study aimed to investigate the effect of seed bacterization with *Az. chroococcum* on the plant growth and the production of active constituent of the selected medicinal *D. stramonium* plants grown under organic amendment.

The seeds *D. stramonium* were kindly supplied from the National Organization of Drug Control and Research, Giza, Egypt.

Seed bacterization: Heavy cell suspension of *Az. chroococcum* was obtained from (Unit of Biofertilizers, Fac. of Agic., Ain Shams Univ.) growing on Ashby medium for 5 days at 29 ± 1°C. Suspension of cells containing about $10^8$ $mL^{-1}$, was used as a standard inoculum.

**Determination of the total microbial count:**

**Bunt and Rovira medium (Bunt and Rovira, 1955):**

| | | | |
|---|---|---|---|
| Peptone | 1.000 gm | $(NH_4)_2SO_4$ | 0.50 gm |
| Yeast extract | 1.000 gm | $FeCl_3.6H_2O$ | 0.01 gm |
| Glucose | 5.00 gm | $Na_2MoO_{43}$ $2H_2O$ | Traces |
| $K_2HPO_4$ | 0.40 gm | $CaCl_2$ | 0.10 |
| $MgSO_4$ $7H_2O$ | 0.50 gm | Soil extract | 250.00 ml |
| NaCl | 0.20 gm | Dis. water | 750.00 ml |
| pH | 6.8 | Agar agar | 20.00 gm |

This medium was used for determination of the total microbial counts. The inoculated plates were incubated at 30°C for 7 days. A greenhouse pot experiment was carried out using a fertile clay loamy soil. The soil was mixed with calcium super phosphate (15.5% $P_2O_2$) at the rate of 100 kg $feddan^{-1}$. The

amended soil was distributed in earthenware pots (30 cm diameter) at the rate of 7.5 kg $pot^{-1}$ clay loamy soil. Nitrogen fertilizer of ammonium nitrate (33.5% N) was added to the amended pots. The nitrogen fertilizer was added 33.33% of total amount after cultivation and 66.67% after two months of cultivation. Three seedlings of *Datura* were planted in each pot. Five replicates for each treatment.

In all treatments, seeds were surface sterilized by immersion in 75% alcohol for 5 min followed by transferring in 0.01% $HgCl_2$ solution for 2 min. Seeds were then washed repeatedly with a sterile water and kept to dry at room temperature. After that, seeds were immersed in a bacterial cell suspension for 30 min. then dried at room temperature before being cultivated in pots.

**Determination of morphological parameters:**

Plant samples were taken after 4 and 6 months of cultivation. Morphological parameters recorded were: plant height (cm), number of leaves, rate of branching, leaf diameter (cm), the stem diameter (cm), fresh and dry weight of shoot and roots (g).

**Determination of alkaloids:**

The active constituent was determined in leaves, stem, flowers, seeds and roots after 4 and 6 months from cultivation. It was determinated using the modified method as described by the Hashim *et al.* (1983). Plant samples were dried at 50°C and macerated in 3% $H_2SO_4$ for 2 h at room temperature. The supernatants were made alkaline with 2.5% $NH_4OH$ (pH 9-10) and applied to Extrelut columns (Marck) and the alkaloids were eluted by $CH_2CL_2$ (6 ml/1 g Extract) and the extracts were evaporated to dryness. Thus obtained residues were resolved in $CH_3OH$ for the further determination. The active constituent was expressed as gram hyoscyamine per 100 g dry matter. Each ml of $H_2SO_4$ is equivalent to 0.01447 g of alkaloids calculated as hyoscyamine.

The experiment was carried out inpots under greenhouse condition in complete randomized design, (Snedecor and Cochran, 1969).

## RESULTS

A green house pot experiment was carried out to evaluate the effect of seed bacterization with *Az. chroococcum* strain on the growth of *D. stramonium* plants in the presence of organic amended and or full inorganic N-fertilizer. The plant is known to contain medium constituents of a high pharmaceutical value hyoscyamine.

Total count of total microbes in the rhizosphere of plant was presented in Table (1). Data show that the highest densities of total microbes in the cultivated soil (125 x $10^4$ cells/g soil) were obtained in the rhizosphere soil of organic amended pots inoculated with *Az. chroococcum* without inorganic N-fertilizer, 3 month spost-cultivation. The count figure was found decrease reaching 90 x $10^4$ $cell/g^{-1}$ dry soil 4 month spost-cultivation. It was also noticed that when full dose of N-fertilizer was applied, the density of total microbes reached 110 x $10^4$ and

75.5 x $10^4$ cells $g^{-1}$ dry soil after 3 and 4 months respectively post-cultivation. Unamended treatments generally showed lower densities of total microbes in rhizosphere soil of the tested plants when compared with amended ones. The count figures of unamended treatments after 3 months post-cultivation were only 62.5, 55.0; 75.5 and 65.5 x $10^4$ cells/$g^{-1}$ dry soil in the absence or presence of inorganic N-fertilizer respectively.

**Table (1): Densities of total microbes in rhizosphere soil of *D. stramonium* seed bacterization.**

| Treatments | Without organic fertilizer | | | |
|---|---|---|---|---|
| | Without N-fertilizer | | N-fertilizer | |
| | 3 month | 4 month | 3 month | 4 month |
| Seed without bacteria | 60.5 | 54.5 | 73.9 | 60.5 |
| Seedbacterization | 62.5 | 55.0 | 75.5 | 65.5 |
| | Organic fertilizer | | | |
| Seed without bacteria | 115.2 | 72.92 | 98.5 | 65.3 |
| Seedbacterization | 125.0 | 90.0 | 110.0 | 75.5 |

Total microbial count x$10^5$ $g^{-1}$ dry soil.

The growth and quantity of hyoscyamine content of *D. stramonium* plant showed positively significant increase as influenced by seed bacterization and organic amendment without N-fertilizer. On the other hand, in the presence N-fertilizer the increase in growth of plants and hyoscyamine was non-significant compared with non seed bacterization and non-organic amendment in presence full dose N-fertilizer (Table 2).

The application of organic amendments and seed bacterization resulted in increasing hyoscyamine in the plant organs (leaves, stem, flowers, seeds and roots) in without N-fertilizer comparing with non-amended and non seed bacterization. However the increasing in hybocyamine quality with presence N-fertilizer was non significant (Table 2).

The fresh weight of shoots was non significantly influenced by inoculation with *Az. chroococcum* in organic amended treatments in the presence N-fertilizer as was 90.498 and in without N-fertilizer was 88.5 g per plant after 6 months of cultivation. This was followed in a descending order by non inoculaiton with Azotobacter and organic amended in the presence and without inorganic fertilizer (Table, 3).

With respect to dry weight of shoots, the same trend was generally noticed when data of the fresh weight were recorded in Table (3).

Data recorded in Table (3) indicate that fresh weight of roots was influenced by inoculation with *Az. chroococcum* and organic amendment. The highest weight of fresh root was obtained by inoculated in *Azotobacter* and organic matter in the presence of N-fertilizer, being 2.85 per plant and 3.10, in without N-fertilizer ones after 6 months of cultivation. The stimulatory effect of

organic amendment on increasing the weight of roots was not significant when cultivation of such plant was carried out in non-seed bacterization in the presence of N-fertilizer and followed without N-fertilizer.

The results in Table (3), illustrated that, the rate of branching of *D. stramonium* was highest in organic amended and seed bacterization in the presence of N-fertilizer being 7 branches plant$^{-1}$ after 6 months of cultivation. This was followed in a descending order by the organic amended with seed bacterization and without N-fertilizer, being 6 branches plant$^{-1}$ after the same period. On the other hand, without organic amended and seed bacterization gave the lowest number of the branches plant$^{-1}$ as 2 and 3 in without and presence N-fertilizer respectively.

**Table (2): Effect of seed bacterization and organic fertilizer on Hyoscyamine content of *D. stramonium* L. plants.**

| Plant organs | Treatments | Organic amended | | | |
|---|---|---|---|---|---|
| | | Non N-fertilizer | | N-fertilizer | |
| | | 4 mon | 6 mon. | 4 mon | 6 mon. |
| Leaves | Non seed bacterization | 1.12 | 1.25 | 1.21 | 1.40 |
| | Seed bacterization | 1.25 | 1.37 | 1.40 | 1.45 |
| Stem | Non seed bacterization | 0.85 | 0.90 | 0.93 | 0.98 |
| | Seed bacterization | 0.95 | 1.02 | 1.07 | 1.15 |
| Flowers | Non seed bacterization | - | 0.75 | - | 0.91 |
| | Seed bacterization | - | 0.85 | - | 1.05 |
| Seeds | Non seed bacterization | - | 0.35 | - | 0.51 |
| | Seed bacterization | - | 0.47 | - | 0.63 |
| Roots | Non seed bacterization | - | 0.11 | - | 0.17 |
| | Seed bacterization | - | 0.11 | - | 0.20 |
| | | No organic amended | | | |
| Leaves | Non seed bacterization | 0.69 | 0.92 | 0.95 | 1.05 |
| | Seed bacterization | 0.75 | 0.78 | 0.90 | 1.05 |
| Stem | Non seed bacterization | 0.45 | 0.59 | 0.62 | 0.74 |
| | Seed bacterization | 0.50 | 0.62 | 0.72 | 0.95 |
| Flowers | Non seed bacterization | - | 0.43 | - | 0.52 |
| | Seed bacterization | - | 0.50 | - | 0.78 |
| Seeds | Non seed bacterization | - | 0.35 | - | 0.43 |
| | Seed bacterization | - | 0.42 | - | 0.52 |
| Roots | Non seed bacterization | - | 0.08 | - | 0.10 |
| | Seed bacterization | - | 0.10 | - | 0.15 |

- No determination.
* The results were calculated from three replicates.
* The active constituent was expressed as gram hyoscyamine per 100 g dry weight %. Alkaloids were calculated as hyoscyamine.
S.D. 1.0%, organ·plant = 0.25 and period = 0.07

**Table (3): Effect of seed bacterization with *Az. chroococcum* and organic amendment on the growth rates of *D. stramonium* L.**

| Growth rates | Treatments | Organic amended | | | |
|---|---|---|---|---|---|
| | | Non N-fertilizer | | N-fertilizer | |
| | | 4 mon | 6 mon. | 4 mon | 6 mon. |
| (1) Plant height (cm) | Non-bacterization | 19.6 | 58.6 | 21.2 | 60.7 |
| | Seed bacterization | 31.5 | 69.8 | 33.5 | 72.0 |
| (2) Rae of Branching plant$^{-1}$ | Non bacterization | 1 | 3 | 2 | 4 |
| | Seed bacterization | 2 | 6 | 3 | 7 |
| (3) Number of Leave plant$^{-1}$ | None bacterization | 14 | 35 | 15 | 37 |
| | Seed bacterization | 24 | 57 | 21 | 60 |
| (4) Leaf length (cm) | Non bacterization | 6.9 | 8.5 | 9.7 | 10.0 |
| | Seed bacterization | 9.2 | 11.5 | 10.5 | 12.0 |
| (5) Leaf width (cm) | Non bacterization | 4.5 | 4.6 | 4.5 | 4.9 |
| | Seed bacterization | 5.2 | 5.6 | 4.9 | 5.7 |
| (6) Stalk diameter (cm) | Non bacterization | 0.7 | 0.9 | 0.7 | 1.0 |
| | Seed bacterization | 0.8 | 0.9 | 0.8 | 1.2 |
| (7) Shoot fresh weight Plant$^{-1}$ (g) | Non bacterization | 14.5 | 49.7 | 13.9 | 50.9 |
| | Seed bacterization | 24.5 | 88.5 | 25.1 | 90.4 |
| (8) Shoot dry weight Plant$^{-1}$ (g) | Non bacterization | 2.5 | 9.7 | 2.9 | 10.2 |
| | Seed bacterization | 3.9 | 12.8 | 4.9 | 13.9 |
| (9) Root fresh weight Plant$^{-1}$ | Non bacterization | 0.50 | 1.80 | 0.44 | 1.95 |
| | Seed bacterization | 1.20 | 2.85 | 0.95 | 3.10 |
| (10) Root dry weight Plant$^{-1}$ | Non bacterization | 0.20 | 0.62 | 0.25 | 0.66 |
| | Seed bacterization | 0.31 | 0.95 | 0.45 | 1.25 |
| | | No organic amended | | | |
| (1) Plant height (cm) | Non-bacterization | 16.5 | 40.9 | 25.5 | 52.7 |
| | Seed bacterization | 25.7 | 57.5 | 27.2 | 64.5 |
| (2) Rae of Branching plant$^{-1}$ | Non bacterization | 1 | 1 | 1 | 2 |
| | Seed bacterization | 2 | 2 | 2 | 3 |
| (3) Number of Leave plant$^{-1}$ | None bacterization | 11 | 22 | 18 | 18 |
| | Seed bacterization | 16 | 27 | 18 | 45 |
| (4) Leaf length (cm) | Non bacterization | 5.2 | 6.5 | 6.9 | 7.5 |
| | Seed bacterization | 7.5 | 7.5 | 7.5 | 8.1 |
| (5) Leaf width (cm) | Non bacterization | 4.2 | 4.4 | 4.3 | 4.5 |
| | Seed bacterization | 5.1 | 4.5 | 4.6 | 4.7 |
| (6) Stalk diameter (cm) | Non bacterization | 0.5 | 0.4 | 0.6 | 0.7 |
| | Seed bacterization | 0.5 | 0.7 | 0.0 | 0.2 |
| (7) Shoot fresh weight Plant$^{-1}$ (g) | Non bacterization | 8.5 | 22.4 | 14.5 | 34.5 |
| | Seed bacterization | 12.5 | 27.5 | 19.7 | 72.5 |
| (8) Shoot dry weight Plant$^{-1}$ (g) | Non bacterization | 1.3 | 3.5 | 3.0 | 5.1 |
| | Seed bacterization | 2.4 | 5.2 | 4.0 | 10.1 |
| (9) Root fresh weight Plant$^{-1}$ | Non bacterization | 0.20 | 1.30 | 0.20 | 1.50 |
| | Seed bacterization | 0.40 | 1.59 | 0.45 | 1.75 |
| (10) Root dry weight Plant$^{-1}$ | Non bacterization | 0.10 | 0.36 | 0.08 | 0.42 |
| | Seed bacterization | 0.15 | 0.52 | 0.21 | 0.65 |

The results were calculated from three replicates.

L.S.D. 0.1%

| (1) | (2) | (3) | (4) | (5) | (6) | (7) | (8) | (9) | (10) |
|---|---|---|---|---|---|---|---|---|---|
| 2.5 | 1.5 | 4.5 | 2.1 | 0.5 | 0.4 | 0. | 0.25 | 0.42 | 0.10 |

It was also found that organic amendments and seed bacterization gave significant effect on branching figures when compared with un-organic amended plants.

The number of leaves per plant was non-significantly influenced by organic amended with seed bacterization in the presence of N-fertilizer, where gave 60 leaves plant$^{-1}$ after 6 month post-cultivation. This was followed in a descending order by the organic amended with seed bacterization and without N-fertilizer being 57 leaves plant$^{-1}$ after the same period. On the other hand, without organic amended and seed baccterization (bio-organic) gave the lowest number of the leaves per plant as 27 and 45 in without and presence of N-fertilizer, respectively.

Non-significant increase in leaf length and width were obtained in amended soil treatment inoculated with *Az. chroococcum* in the presence of N-fertilizer being 12.0 and 5.7 cm, respectively after 6 months from sowing, while in without N-fertilizer were found 11.6 and 5.6 cm in length and width respectively

Inoculation with *Az. chroococcum* in soil resulted in a significant increase of the stalk diameter of plants from 1.0 cm in inoculated organic amended to 1.2 cm in without N-fertilizer.

## DISCUSSION

This work aim aimed to bio-organic production of medicinal constituent from *D. stramonium* without N-fertilization as well as density of total microbes in greater absorption of nutrients from soil.

In addition a pot experiment was designed to study the effect of inoculating *Az. chroococcum* in the presence of organic matter on the growth of *D. stramonium* L. and its contents of active substances. It is clear from the obtained results that, inoculation with *Az. chroococcum* non significantly increased total microbial count in rhizosphere of *D. stramonium*, the plant growth, branching rate, number of leaves plant$^{-1}$, length and width of leaves; plant height, stalk diameters, fresh and dry weight of shoot and roots and hyoscyamine content of the plants. The benificial effect of *Az. chroococcum* on plant development could be attributed not only to the $N_2$-fixation process but also to the production of growth promoting substances. Harper and Lynch (1979) reported that *Az. chroococcum* produces thiamine, nicotinic acid, pantothenic acid, biotin, pyridoxine, gibberellins and other compounds of the auxin type which gave a positive effect on plant growth root biomass, and thus indirectly help in greater absorption of nutrients from soil. In addition it may be due also to the successful competition of the bacteria with antagonists of plant growth (Lynch and White, 1977), therefore a series of comperhensive experiments for inoculation with asymbiotic $N_2$fixers with different plants were carried out by other investigators and clearly showed that inoculation with *Azotobacter* (Lavahmann, 1982; Ishac *et al.*, 1984 and Sharaf, 1985) led to considerable improvements in the plant growth and yield as well as the reducing the costs of agricultural production by reducing the amount of inorganic nitrogen fertilizers through the enhancement of asymbiotic $N_2$-fixation.

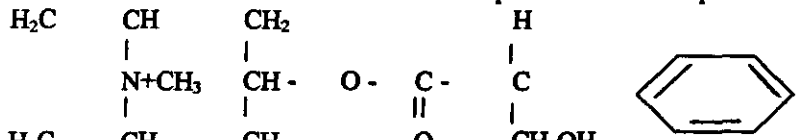
It was also found that application of organic amendments (compost of plant residues) resulted in increasing the plant growth, dry matter and hyoscyamine. This

may be due to that organic matter amendment has a fundamental effect on biological, physical and chemical properties of soil (Baver, 1963 and Bear, 1965). Supplementing the soil with different forms of carbonic materials having a wide C/N ratio results in a marked increase in densities of *Azotobacter* spp. accompained by appreciable gains in nitrogen. These results are in line with those obtained by Eweda, (1983); Ishac *et al.* (1985) and Sharaf (1985). Moreover, a significant correlation between densities of N2-fixers and amount of organic matter added was also due to the preference of these N2-fixers to organic acids than other energy sources, thus accumulation of these compounds in soil due to organic matter degradation, will encourage the prevalence of such group of bacteria in soil and rhizosphere (Kuto and Mann, 1968; Dobereiner and Day, 1976, Mostafa, 1983 and Sharaf, 1985).

The results showed that, the presence of full dose of N-fertilizer inhibited $N_2$-fixation (Neyra and Dobereiner, 1977 and Pederson *et al.*, 1978). On the contrary without or low concentration of N-fertilizers promote the response of plants to inoculation and the establishment of well functional $N_2$-fixing association (Reynders and Vlassak, 1979).

The present study showed that, quantitative investigation of hyoscyamine revealed that the *D. stramonium* accumulates relatively high hyoscyamine as compared with other plant species. This result is in agreement with a previous report (Brachet *et al.* (1997). The hyoscyamine content was differed among organs of plants. The highest hyoscyamine content was found in leaves followed by stem, flowers, seed and roots (Berkov and Philipov, 2003 and Nari *et al.*, 2005, Philipor and Berkov, 2003 and El-Dougdoug *et al.*, 2007).

The drug occurs generally in a dry condition in matter masses or broken loose pieces of shrivelled leaves intermixed with stems and flowering tops and also few fruits. The characters of the powdered of Egyptian Hyoscyamus is light greenish yellow with a slightly foetid and necrotic and a bitter acrid test (Hashim *et al.*, 1983). The chemical nature of active constituent is tropine base and di-tropic acid




Atropine (di-Hyoscyamine)

## REFERENCES

Baver, L.D. (1963): Soil physics. 3rd Ed. John Wiley and Sons, Inc., New York.

Bear, F.E. (1965): Chemistry of the soil. Painhold publishing Comperation, New York.

Berkov, S. (2003): Alkaloids of *Datura ceratocoula*. Z. Nature Forsch 85: 455-458.

Berkov, S. and Philipov, S. (2003): Alkaloid production in diploid and tetraploid plants of *Datura stramonium*. Phermocent, Biol., 40: (In Press).

Brachet, A.; Munoz, D.; Gupta, M.; Veuthey, J.L. and Christen, P. (1997): Alkaloids of *Erytroylum luudum* stem-bark. Phyochemistry, 46: 1439-1442.

Bunt, J.S. and Rovira, A.O. (1955): Microbiological studies of some sub-antaretic soil. J. Soil Sci., 6: 119-128.

Dobereiner, J. and Day, J.M. (1976): Associative symbioses in tropical grasses. Characterization of microorganisms and dinitrogen fixing sites, p. 518-538. In Newton, W.E. and Nymon, C.J. (Eds), Proceedings of the First International Symposium on Nitrogen fixation. Washington State Univ. Press Pullman, USA.

El-Dougdoug, Kh.A.; Hala, Mohamed and Amal Abou Senna (2007): Effect of PVY viral infection on alkaloid content of cultivated medicinal plants. Journal of Applied Sciences, 2007 (Under press).

Eweda, Wedad E. (1983): Effect of soil inoculation with *Azotobacter* on some economical plants. Ph.D. Thesis, Fac. of Agric., Ain Shams Univ., Cairo, Egypt.

Griffin, W. and Lin, D. (2000): Chemotaxonomy and geographical distribution of tropane alkaloids. Phytochemistry, 55, 623-637.

Harper, S.H.T. and Lynch, J.M. (1979): Effects of *Azotobacter chroococcum* on barley seed germination and seedling development. J. Gen. Microbiol., 112: 45-51.

Hashim, F.M.; Shabana, M.M. and El-Shamy, A.M. (1983): Pharmacognosy for second year students. Second Ed., 2: 24-31. Fac. of Pharmacy, Cairo Univ. Saad Sammak Publishing House, Hadaik El-Koba, Egypt.

Ishac, Y.Z.; El-Haddad, M.E.; Eid, M.; Saleh, E.A.; El-Borolosy, M.A. and El-Demerdash, M. (1984): Effect of seed bacterization and organic amendment on the growth of some economical crops in Egypt. I. Maize. International Symposium on $N_2$-fixation by non-leguminous plants, Finland.

Ishac, Y.Z.; El-Haddad, M.E.; El-Borollosy, M.A.; Rahal, A.G. and Mostafa, M.L. (1985): Effect of organic acids and carbon dioxide on asympbiotic $N_2$-fixation. Egypt. J. Microbiol., Special Issue

Kuto, S.B. and Mann, H.S. (1968): Effect of green manuring on physical, chemical and biological properties of soil. Indian J. Agron., 13: 20-25.

Lavahmann, M. (1982): Response of crop plants to *Azotobacter* treatment. Second International Symposium on $N_2$-fixation with non-legumes. Banff, Canada, p. 32.

Lounasmaa, H. and Taminnea, T. (1993): The tropane alkaloids In: The alkaloids (Brassi. A. Ed.). Academic Press, New York, pp. 1-114.

Lynch, J.M. and White, N. (1977): Effect of some non-pathogenic microorganisms on growth of antibiotic barley plants. Plant and Soil. 47 (1): 61-170.

Mostafa, M.I.M. (1983): Some factors affecting $N_2$-fixation by non-symbiotic microorganisms in the rhizosphere. M.Sc. Thesis, Fac. Agric., Ain Shams Univ., Cairo, Egypt.

Nari, T.; Takata, Z. and Graham Cooks, R.G. (2005): Rapid *in situ* detection of alkaloids in plant tissue under ambient conditions using desorption Electro spray ionization. J. Analyst, 130: 1624-1633.

Neyra, C.A. and Dobereiner, J. (1977): Nitrogen fixation in grasses, Adv. Agron., 29: 1-38.

Pederson, W.L.; Chakrabarty, K.; Klueas, R.V. and Vidaver, A.K. (1978): Nitrogen fixation (acetylene reduction) associated with roots of winter wheat and sorghum in Nebraska, Appl. Environ. Microbiol., 35: 129-135.

Philipov, S. and Berkov, S. (2003): GC-NS investigation of tropone alkaloids in *Datura stramonium*. Z. Nature Forsch, 57c. 559-561.

Porde, E.H. and Doty, H.O. (1981): In New Sources of Foots and Oils, E.H. Drincon; K.D. Mukherjee Eds. (American Oil Chemical Society, Champaign 111) 3-14.

Reyriders, L. and Vlassak, K. (1979): Conversion of tryptophan to indole acetic acid by *Azospirillum brasilense*. Soil Biol., Biochemistry 11: 547-548.

Sharaf, M.S. (1985): Studies on nitrogen fixers in some non-legumionous plants. M.Sc. Thesis, Fac. of Agric., Ain Shams Univ., 163 pp.

Snedecor, G.W. and Cochran, W.G. (1969): Statistical methods. 8th Ed., Iowa State Univ. Press. Ames, Iowa, U.S.A.

Tayler, V.E.; Brady, L.R. and Robbers, J.E. (1981): Pharmacognoxy (Lea and Febiger, Philadelphia, ed. 8.

## الإنتاج الحيوى – العضوى للمادة الطبية الفعالة من نبات الداتوره

**حسن محمود قرقار**
المعهد العالى للتعاون الزراعى – القاهرة – مصر

الدراسات الفسيولوجية على المادة الفعالة وارتباطها بتكشف نمو نبات الداتوره الطبى محدودة ، وهذه الدراسة توضح تأثير تلقيح بذور الداتوره ببكتريا الأزوتوباكتر على نمو النبات ومحتواه من المادة الطبية الفعالة والمسمد عضوياً وكذلك أعداد والميكروبات فى تربة الريزوسفير . أوضحت النتائج التأثير الموجب للتلقيح بالأزوتوباكتروالتسميد العضوى فى عدم وجود النتروجين المعدنى . وسجلت الزيادة المرتفعة فى معدل نمو النبات (التفريع وطول النبات وعدد الأوراق وعرض وطول الورقة وسمك الساق والوزن الرطب والمادة الجافة للمجموع الخضرى والجذرى) للنباتات المسمدة حيوياً عضوياً يليها النباتات المسمدة حيوياً فى وجود التسميد النتروجينى المعدنى . أما النباتات الغير مسمدة حيوياً وعضوياً فقد أظهرت إنخفاضاً معنوياً فى معدلات نمو النبات بمقارنتها بالنباتات فى المعاملات الأخرى .

كما سجلت النباتات المسمدة حيوياً – عضوياً والمنزرعة فى أرض طينية طمية والملقحة بالأزوتوباكتر والمسمدة عضوياً محتوى عالى من المادة الفعالة للقلويدات . وأظهرت الأوراق أعلى محتوى من المادة الفعالة عن مكونات النبات الأخرى والنسبة المئوية للمادة الفعالة هى على التوالى ١,٤٥ و ١,١٥ و ١,٠٥ و ٠,٦٣ و ٠,٢% فى الأوراق والسيقان والأزهار والبذور والجذور فى المادة الجافة . بعد ٦ أشهر من الزراعة انخفضت مكونات النبات من المادة الفعالة فى النباتات الغير مسمدة حيوياً – عضوياً .

كما أدى التسميد الحيوى – العضوى الى زيادة فى أعداد الميكروبات الكلى فى تربة لريزوسفير النبات (التربة الملاصقة للجذور) وكانت ١٢٥,٠ × ١٠^٦ خلية لكل جرام تربة جافة ، ولكن إنخفضت أعداد الميكروبات فى تربة الريزوسفير للنباتات الغير مسمدة حيوياً – عضوياً وكانت ١١٥,٢ × ١٠^٦ خلية لكل جرام تربة جافة.