

## EFFECT OF SOME MICRONUTRIENTS ON THE YIELD AND ALKALOIDS CONTENT OF HARMAL PLANT (*PEGNAUM HARMALA* L.)

E.A. EL-GAMAL, A.A. ZAIED AND S.M. MAHMOUD

Horticultural Research Institute, Agricultural Research centre, Giza, Egypt.

(Manuscript received 22 September 1991)

### Abstract

In a study conducted on harmal plant (*Peganum harmala* L.) at Seds Experimental Station in Beni-Swaif Governorate, three powdered micronutrients formulations and two liquid ones named Wadi (3% Zn, 4.5 Mn and 1.5% Fe), Sahara (5% Zn, 3.5% Mn and 1.7% Fe), Giri (3.8% Zn, 3.8% Mn and 3.8% Fe), Shiring-Zn (4% Zn, 2.7% Mn and 1.3% Fe) and Shiring-Mn (2.8% Zn, 4.2% Mn and 1.4% Fe) respectively, were foliarly applied for three times in the rate of 1.5 g/L. or 1.5 ml/L. (i.e. for the powdered and liquid forms respectively) starting at flowering and after three week intervals. The results revealed that all the formulations applied increased the fresh and dry weight yields of herb, roots and the whole plant, as well as the alkaloid content in comparison with the control. The "Wadi formulation" produced the highest yield of fresh and dry herb and roots. However, it may be considered that the "Shiring-Mn" formulation is the favourable one to obtain the highest yield of alkaloids.

### INTRODUCTION

*Peganum harmala* L. (harmal) Zygophyllaceae, is a herbaceous plant growing wild in deep sandy places in Egypt, Mediterranean coast strip from El-Sallum to Rafah (Tackholm 1974). The plant was used medicinally as anthelmintic (tape worms), lactagogue and against malaria (Chopra and Chopra, 1958), antimicrobial (Ross *et al.*, 1980) and hypotensive (Siddiqui *et al.*, 1987). Also normal seeds have nematocidal effects (Sharma *et al.*, 1982) and allelopathic effects on seed germination

(Mohammad *et al.* 1982). In this connection most of the work done on *P. harmala* L. dealt with isolation and identification of alkaloids as well as their uses and applications in medicine or in other purposes (e.g. nematocide). Hilal *et al.* (1977) identified the alkaloids peganine, harmaline and harmine in the different organs of the plant, also they estimated the individual alkaloids by using T.L.C. and G.L.C. techniques; and reported the sum of total alkaloids calculated as harmine in the following values for the cultivated plants. 1.06%, 1.87% and 1.80% for leaves, stems and roots respectively. Jado *et al.* (1979) isolated the alkaloids of *Peganum harmala* L. growing in Saudi Arabia and they determined the total alkaloids in the plant organs by non-aqueous titration method.

Regarding the effect of micronutrients on the active ingredients in the medicinal plants, Ebrahim, 1981 reported that Zinc deficiency affected the total content of alkaloids of *Hyoscyamus muticus* L. Koreish and Helmy (1984) found that spraying of *Datura stramonium* L. plants with 0.25% Zinc sulfate singly or in combination with 0.1% ferrous sulfate increased fresh and dry weight of leaves as well as the total alkaloids. Zaied (1984) reported that spraying of *Saponaria officinalis* L. with Zn, Mn and Fe either separately or combined, resulted generally in high significant increases of saponin content in the different plant organs and the whole plant compared with the control. Haridi (1986) working on periwinkle plants (*Catharanthus roseus* G. Don.), reported that spraying plants with Zinc at the rate of 100 ppm stimulated significantly the herb and dry matter production as well as alkaloid percentage and yield (as perivine).

The aim of this investigation is to study the effect of three chelated microelements namely, zinc, manganese and iron, combined by different proportions, on the yield of herb and alkaloids of harmal. Such study was achieved by the foliar application of five commercial formulations containing the suggested three elements in order to find out the favourable one leading to obtain higher yield of harmal herb and alkaloids as well.

## MATERIALS AND METHODS

Harmal seeds were collected from the desert area of Borg El-Arab (North Coast of Egypt) and used in the present investigation. The experiment was conducted in Seds Experimental Station, Beni-Swaif Governorate for two successive seasons of 1987 and 1988 respectively. Such experiment was performed to study the effect of foliar spraying with five commercial formulations containing zinc, manganese and iron in a chelated form.

Three powdered micronutrient commercial formulations in addition to two liquid ones were used in the present investigation. The powdered formulations were produced by BASF Company, West Germany and having the following names, Wadi, Sahara and Giri, while the liquid ones were produced by Shiring Company West Germany and having the following names Shiring-Zn and Shiring-Mn. The chemical composition of the used formulation are shown in Table 1.

The experiment consisted of the five previously mentioned formulations (i.e. Wadi, Sahara, Giri, Shiring-Zn and Shiring-Mn respectively) in addition to the control treatment so, their was 6 treatments with three replicates. The plot area was 12 m<sup>2</sup> comprising 5 rows 60 cm apart and 4 m long. The plants were spaced by 40 cm each. The complete randomized blocks design was followed as an experimental

Table 1. Chemical composition of five micronutrient commercial formulations.

No.	Commercial Name	% Micronutrient			Form
		Zinc	Manganese	Iron	
1	Wadi	3.0	4.5	1.5	Powder (1)
2	Sahara	5.0	3.5	1.7	" "
3	Giri	3.8	3.8	3.8	" "
4	Shiring-Zn	4.0	2.7	1.3	Liquid (2)
5	Shiring-Mn	2.8	4.2	1.4	" "



design in this respect. Soil samples representing the experimental area were taken, well mixed and subjected to chemical analysis for macro and micro elements. The macro element contents were 6.32, 7.83, 180.10 and 58.95 mg/100 grams for nitrogen, phosphorous, potassium, magnesium and sodium respectively whereas the contents of microelements were 44.9, 24.9, 0.42 and 6.03 ppm for iron, manganese, zinc and copper respectively. Also, the conductivity and pH were found to be 0.36 m.mhos and 8.44 respectively.

The seeds were sown on February 1<sup>st</sup>, major element fertilization was done for all plants using the following quantities: 200 kg calcium super phosphate (15.5%  $P_2O_5$ ), 400 kg ammonium sulfate (20.6% N) and 100 kg potassium sulfate (48%  $K_2O$ ) per feddan (4200  $m^2$ ). Soil and plant management were done as usually followed for herbaceous plants. Regarding micronutrient application, the plants received 3 sprays in the concentration of 1.5 gram or 1.5 milliliters per liter for the powdered or liquid types respectively. The first spray was applied at the beginning of flowering, while the second and third sprays were applied later at 3 week intervals. The flowering started on April 9th and 5th of the 1987 and 1988 seasons respectively while fruit setting started on April 22nd and 16th of the two respective seasons. The statistical analysis was carried out according to Snedecor and Cochran (1956). At harvesting date on July, two representative plant samples were taken from each plot, then fresh weight of the whole plant and plant parts (herb and roots) followed by the determination of dry weight at 105 $^{\circ}$  (AOAC 1960) were recorded in order to calculate fresh and dry weight yields. Meanwhile similar samples were taken, dried at 50 $^{\circ}$ C, finely ground and kept for chemical estimation of alkaloids.

The estimated of alkaloids was carried out using non-aqueous titration method according to Jado *et al.* (1979) and Vogel (1975). The alkaloids previously extracted according to Hilal *et al.* (1977) were dissolved in 5 milliliters of chloroform and 5 ml of glacial acetic acid then the alkaloids were titrated with 0.05 N acetous perchloric acid in the presence of crystal violet indicator to the green violet color. The results were calculated in terms of harmine using the molecular weight of harmine ( $C_{13}H_{12}N_2O$ ) and the volume of perchloric acid consumed in the titration.

## RESULTS AND DISCUSSION

The results mentioned herein are dealing with the effect of foliar spraying with five commercial formulations (i.e. Wadi, Sahara, Giri, Shiring-Zn and Shiring-Mn). Containing Zinc, Manganese and Iron, on both plant and alkaloidal yields of *P. harmala* L. in 1987 and 1988 seasons.

### 1. Fresh and dry weight yields:

Data presented in Tables 2 and 3 show the effect of foliar spraying with the five used formulations on the fresh and dry weight per plot of the whole plant herb and roots of *P. harmala* L. in 1987 and 1988 seasons respectively. The results indicated that most of the applied treatments produced a high significant fresh and dry weight yield/plot in comparison with the control. The exception was found in formulation 3 (Giri) which did not exhibit significant effect. The highest significant value was recorded in treatment 1 (Wadi) followed by treatments 5, 2 and 4 corresponding to Shiring manganese, Sahara and Shiring zinc formulations respectively. The obtained result could be explained by referring to Table 1. It is obviously shown that formulation 1 (Wadi) contains the highest content of manganese (4.5%), also formulation 5 (Shiring manganese) has high content of manganese (4.2%) but lower than that of formulation 1. On the other hand, it is also shown from Table 1 that formulation 2 (Giri) contains the highest content of zinc (5%) followed by formulation 4 (4%) shiring zinc. Moreover formulation 3 which did not exhibit significant increase of yield is characterized by the highest value of Fe (3.8%), in balanced proportions with zinc and manganese consequently. It could be concluded that spraying with manganese in combination with Zn and Fe is more effective than the other used formulations to increase both fresh and dry yields since it is existed in a higher proportion than that of both zinc and iron. However, it may be mentioned that zinc element, as existed in the formulation used, is of moderate effect on the fresh and dry yields so that formulation 2 (Sahara) produced yields slightly higher than those resulted from both 1 and 5 ones. The obtained results were in agreement with those obtained by Haridi (1986) on periwinkle, Koreish and Helmy (1984) on *Datura* and Zaeid (1984) on *Saponaria officinalis* L.

### Alkaloid percentage:

Table 4 shows the alkaloid percentages in harmal plant parts (herb and roots) as affected by 5 micronutrient formulations in the 1987 and 1988 seasons. The data

Table 2. Effect of foliar spraying with some micronutrient formulations on the fresh weight yield (kg/plot) of harmful plants in 1987 and 1988 seasons.

No.	Formulation	1987			1988		
		Herb	Roots	Whole plant	Herb	Roots	Whole plant
1	Wadi	159.50	7.70	167.20	150.40	8.03	158.43
2	Sahara	140.1	6.67	146.82	134.78	6.25	141.03
3	Giri	94.50	3.70	98.20	103.78	4.75	108.53
4	Shiring-Zn	106.52	4.27	110.79	119.27	5.53	124.80
5	Shiring-Mn	149.23	6.88	156.11	144.28	7.52	151.80
6	Control	91.03	3.67	94.70	100.12	4.73	104.85
	L.S.D. 5%	15.70	2.71	13.40	20.77	1.38	7.14

Table 3. Effect of foliar spraying with some micronutrient formulations on dry weight yield (kg/Plot) of harmful plants in 1987 and 1988 seasons.

No.	Formulation	1987			1988		
		Herb	Roots	Whole plant	Herb	Roots	Whole plant
1	Wadi	88.38	5.57	93.95	84.65	5.83	90.48
2	Sahara	78.96	4.83	83.79	75.65	4.49	80.14
3	Giri	53.20	2.69	55.89	59.23	3.46	62.69
4	Shiring-Zn	61.23	3.10	64.3	67.11	4.00	71.11
5	Shiring-Mn	83.50	4.96	88.46	80.86	5.42	86.28
6	Control	51.64	2.63	54.27	57.25	3.40	60.65
	L.S.D. 5%	10.57	1.973	7.57	9.61	0.99	3.72



revealed that all the applied treatments increased the alkaloidal percentages in both herb and roots of the plant in comparison with the control. It could be observed that formulation 4 gave the highest alkaloidal percentage in the herb in both seasons but such increment was also produced in response to formulation 5 in the second season in comparison with the other treatments. In general it can be observed that formulations 2, 4 and 5 produced higher values in both seasons in comparison with the other treatments. However, the alkaloidal percentages in the roots exhibited a response with formulation 3 (3.8% Fe) which showed the highest value in this concern. It is shown from Table 1 that formulations 2 and 4 are of higher content of Zn and formulations 5 is characterized by a high content of Mn, moreover formulation 3 have the highest content of Fe, consequently. It may be concluded that zinc and manganese are required for the plant to synthesize the alkaloids in the herb. The obtained results agreed with those obtained by Haridi (1986), Koreish and Helmy (1984), Zaied (1984) and Ebrahim (1981).

#### Alkaloid yield:

Table 5 shows the effect of 5 micronutrient formulations on the total alkaloid yield in g/plot in the whole plant as well as in the plant parts (herb and roots) in 1987 and 1988 seasons. It could be observed from the results that all the applied treatment led to a significant increase in the alkaloid yield either in the whole plant or the plant parts in comparison with the control. Data of the first season revealed that formulation 5 recorded the highest value (680 g/plot) followed by formulation 2 (670 g/plot). Regarding the data of the second season, it may be noticed that a similar trend was shown in most cases. It can be concluded that, formulation 5 (Shiring manganese) was the highest treatment since it produced the highest yield of alkaloids in the herb.

Regarding the alkaloid yield produced by the roots, it was shown that the application of the five formulations has increased significantly the alkaloid yields especially in formulations 5 and 1 which recorded higher values. The previous results are in agreement with those obtained by Haridi (1986) and Zaied (1984).

It could be mentioned that although formulation 2 led to produce alkaloid yield slightly lower than that of formulation 5, treatment 2 was not the highest of all treatments in the second seasons 1988 as that produced from formulation 5. Consequently the later formulation (Shiring manganese) may be considered the favourable one to increase the alkaloid yield, since the results were confirmed for the two seasons 1987 and 1988.

Table 4. Effect of foliar spraying with some micronutrient formulations on the alkaloid percentages in harmful plants in 1987 and 1988 seasons.

No.	Formulation	1987		1988	
		Herb	Roots	Herb	Roots
1	Wadi	0.64	1.44	0.54	1.26
2	Sahara	0.77	1.31	0.77	0.95
3	Giri	0.60	1.51	0.52	1.71
4	Shiring-Zn	0.95	1.17	0.86	1.04
5	Shiring-Mn	0.73	1.44	0.85	1.45
6	Control	0.41	1.00	0.26	0.83
	L.S.D. 5%	0.10	0.10	0.06	0.16

Table 5. Effect of foliar spraying with some micronutrient formulations on the alkaloid yield of harmful plants (g/plot) in 1987 and 1988 seasons.

No.	Formulation	1987			1988		
		Herb	Roots	Whole plant	Herb	Roots	Whole plant
1	Wadi	570	80	650	460	70	530
2	Sahara	610	60	670	580	40	620
3	Giri	320	40	360	310	60	370
4	Shiring-Zn	580	40	620	550	40	590
5	Shiring-Mn	610	70	680	690	80	770
6	Control	210	30	240	150	30	180
	L.S.D. 5%	130	20	71	170	20	45.9



## REFERENCES

1. A.O.A.C. 1960. Methods of analysis of the Association of Official Agricultural Chemists Published by the A.O.A.C. Washington, D.C.
2. Chopra, R.N. and J.C. Chopra. 1958. "Indigenous Drugs of India" 2nd Ed., 368, Ohur and Sons Private, Ltd.
3. Ebrahim, D.M. 1981. Studies on the nutritional requirements of *Hyoscyamus muticus*. L.M.Sc. Thesis Agric. Fac. Zagazig Univ.
4. Haridi, I.M.A. 1986. Physiological studies on periwinkle plant, *Catharantus roeus* G. Don. Ph.D. Thesis Fac. Agric. Cairo Univ.
5. Hilal, S.H., M.Y. Haggag and S.A. El Kashoury. 1977. Chromatographic study of *P. harmala* L. Egypt. J. Pharm. Sci. 18(1): 9-18.
6. Joda, A.I., M.M.A. Hassan, S.T. Ezmirly and F.Y. Muhtadi. 1979. The chemical investigation of *P. harmala* grown in Saudi Arabia. Pharmazie 34(2): 108-109.
7. Koreish, S.M. and H.A. Helmy. 1984. Effect of spraying with some micronutrients on growth and alkaloidal content of *Datura stramonium* L. Second Conf. of Agric. Res. Center Giza April, 9-11.
8. Mohammad, H.U., S.I. Hussein and A.J. Al-Zarari. 1982. Effect of plant extracts of some poisonous plants of Iraq on mortality of citrus nematode. Tylemchulus - Semipentans: Acta Bot. Indica 9(2): 198-200.
9. Ross, S.A., S.E. Megralla, D.W. Bishary and A.H. Awad. 1980. Studies for determining antibiotic substances in some Egyptian plants part II Antimicrobial alkaloids from the seed of *P. harmala*. Fitoterpia 51: 309-312.
10. Sharma, K.D., K.L. Sidana and N.R. Singh V. 1982. Allelochemic effect of *Pegunum harmala* L. on pennisteum - Typhdeum. Indian J. Bot. 5(2): 115-119.
11. Siddiqui, S., O.Y. Khan and B.S. Siddiqui. 1987. Harmalidins, a beta carboline alkaloid from *Pegunum harmala* L. Phytochemistry, 26(5): 1548-1550.
12. Snedecor, G.W. and W.G. Cochran. 1956. Statistical methods, the Iowa State Univ. Press. P. 237, Ames Iowa U.S.A.
13. Tackholm, V. 1974. Student flora of Egypt. Cairo Univ. Printed by Cooperative Printing Company Beirut.
14. Vogel, A.I. 1975. Elementary practical organic Chemistry part III pp. 663-672. English Language Book Society and Language Group Limited. Pub. E.L.B.S.
15. Zaied, A.A. 1984. Studies on some factors affecting growth, yield and active principles of *Saponaria officinalis* L. Ph.D. Thesis Fac. Agric. El-Minia Univ.

## تأثير بعض العناصر الصغرى على المحصول والقلويدات فى نبات الحرمل فى مصر

السيد عبد الحميد الجمل ، عادل عبد العزيز زايد ، سعيد محمود محمد

معهد بحوث البساتين - مركز البحوث الزراعية

أجريت دراسة على نبات الحرمل الناتج من بذور جمعت من منطقة برج العرب وذلك فى محطة بحوث البساتين بسدس محافظة بنى سويف على خمسة أنواع من الاسمدة المحتوية على عناصر الزنك والمنجنيز والحديد بنسب مختلفة وهى: وادى (٢٪ زنك ، ٤,٥٪ منجنيز ، ١,٥٪ حديد) ، صحرا (٥٪ زنك ، ٣,٥٪ منجنيز ، ١,٧٪ حديد) ، جبرى (٢,٨٪ زنك ، ٢,٨٪ منجنيز ، ٣,٨٪ حديد) ، شيرنج زنك (٤٪ زنك ، ٢,٧٪ منجنيز ، ١,٣٪ حديد) ، شيرنج منجنيز (٢,٨٪ زنك ، ٤,٢٪ منجنيز ، ١,٤٪ حديد) ، على الترتيب استخدمت رشا على النباتات ثلاث مرات بتركيز ١,٥ جرام أو ملتر فى اللتر عند التزهير ثم كل ثلاث أسابيع. أوضحت النتائج أن كل أنواع أسمدة العناصر الصغرى الخمسة أدت الى زيادة كل من محصول الطازج والجاف من العشب والجذور والنبات الكلى وكذلك محصول القلويدات مقارنة بالكنترول. وقد أوضحت النتائج أن استخدام السماد المركب (وادى) أدى الى الحصول على أعلى محصول من الوزن الطازج والجاف للنبات الكلى والعشب والجذور ، ولكن يمكن أن نعتبر أن السماد المركب شيرنج منجنيز هو التركيبة المفضلة نظرا لأنها أدت للحصول على أعلى محصول للقلويدات خلال موسمى الزراعة.