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Morphological studies of medicinal plant of *Withania somnifera* (L.) Dunal grown in heavy metal treated (contaminated) soil.

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ABSTRACT
Plants were grown in pot culture experiments with three treatments in black soil, Treatment No I, a control without any addition to the soil, Treatment No II, Cadmium 10ppm, Chromium 20ppm, Nickel 16ppm were introduced into the soil, Treatment No III, one % of Calcium hydroxide was also added along with heavy metals to soil and was grown up to the productivity levels. To know the effect of the heavy metals on the growth and development of *Withania somnifera* plants, the study of macro morphology of external character was undertaken. Productivity was reduced in the plants grown in heavy metal treated plants compared to control plants. The experimental data revealed that the external morphology plants i.e., Plant height (cms), No of branches reduced in (Treatment No II) heavy metals treated plants and also was increased in No. of leaves compared to control plants and heavy metals + Calcium hydroxide treated plants. The No. of fruits per plants, leaf area reduced in the (Treatment No III) heavy metal+ Calcium hydroxide treated plant compared to control plants and heavy metals treated plants. This indicates that the heavy metal effects on the growth and development of the plants and ultimately the yields.

Keywords: *Withania somnifera*, yield, morphological features, leaf area.

1. Introduction

Withania somnifera is systematically placed into the division Magnoliophyta, class Magnoliopsida, order Solanales and family Solanaceae [7]. It is well known for its rejuvenating properties, and hence called "Indian Ginseng" [10]. *Withania somnifera* is an evergreen, erect, branching, tomentosa shrub, 30-150 cm in height. In Ayurvedic and Unani systems, the leaves of the plants are used for tumor and tubercular glands. Leaves are bitter in taste and used as an antihelminthic. Leaves are simple opposite, alternate, petiolate, elliptic ovate to broadly ovate, entire, exstipulate, the tip of the leaf is acute, cuneate or oblique and glabrous upto 8 to 12 cm in length. Flower is shortly pedicillate and 4-6 mm in dia. Flowers are greenish or lurid yellow, small about 1 cm long; few flowers (usually about 5) born together in axillary, umbellate cymes (short axillary clusters). Fruits are globose berries, 6 mm in diameter, orange red when mature, enclosed in the inflated and membranous persistent calyx. Seeds are smooth, discoid, 20-25 seed per fruit. Seeds are yellow, reniform and 2.5 mm in diameter [2, 3]. Roots are stout, fleshy, cylindrical and 1-2 cm thick, straight, unbranched, roots bear fiber like secondary roots, outer surface buff to grey-yellow with longitudinal wrinkles; crown consists of 2-6 remains of stem base; stem bases variously thicked; nodes prominent only on the side from where petiole arises, short and uneven; odour characteristic; taste bitter and acrid [2]. The roots are used as a nutrient and health restorative in pregnant women and old people. The decoction of the root boiled with milk and ghee is recommended for curing sterility in women. The roots are also used in constipation, senile debility, rheumatism, general debility, nervous exhaustion, loss of memory, loss of muscular energy and spermatorrhoea [11]. In Unani system of medicine, roots of *Withania somnifera* commonly known as Asgand are used for the medicinal properties. However, leaves of the plant are also reported to be used medicinally [1].

This shrub is common in Bombay and Western India, occasionally met within Bengal. It grows wildly throughout India particularly in hotter parts, on waste places and on road sides. It is also cultivated for medicinal purposes in fields and open grounds throughout India. This plant grows widely in all dried parts and subtropical India (widely cultivated in Bikaner and Pilani areas of Rajasthan, Rajputana, Punjab, Gujarat, Uttar Pradesh and Manasa (M.P.), Congo, South Africa,

Pakistan, Afghanistan, Egypt, Morocco, Jordan, Sri Lanka, Egypt, Morocco, Jordan, Iraq, Iran, Syria and Turkey [2-3-4-5-6-8-9].

2. Material and Methods

2.1 Plant material source

Withania somnifera seeds were procured from the CIMAP, Hyderabad. The plants were grown in earthen pots at Green house of Botanical Garden, Department of Botany, Osmania University and Hyderabad.

2.2 Experimental plant material cultivation

Sandy loam semi black soil was taken from clean area of Botanical Garden, Department of Botany, Osmania University, Hyderabad, A.P and India, and was passed through a 2mm sieve and air dried for one week, then filled into 20 pots so as to obtain 15 pots for individual exposure. Such prepared soil was taken and filled in 15 different pots divided into three treatments. Treatment No I control without any addition to the soil. Treatment No II Cadmium 10ppm, Chromium 20 ppm, Nickel 16ppm were introduced into the soil. Treatment No III one % of Calcium hydroxide was also added along with heavy metals to soil. Plants were grown in pot culture experiments with three treatments and were grown up to the productivity levels during years (2009 to 2010).

2.3 Plant height in Bikaner and Pilani areas of Rajasthan, Rajputana, Punjab, Gujarat, Uttar Pradesh and Manasa (M.P.), Congo, South Africa,

The plant height was measured with the help of scale and 100 plants. Where minimum and maximum readings were taken from ground to the apical part of the plants and values were expressed in centimeters.

2.4 Fresh Weight

The fresh weights of the plants recorded using a weighing balance. The fresh weights were expressed in grams.

2.5 Dry Weight

The plants were dried in the oven at 100 ^{0}C for 24 hours and their dry weights were recorded. The dry weighted were expressed in grams.

3. Result and Discussion

3.1 Plant height (cm)

The plant height in cms of *Withania somnifera* were 34.13±1.21 cms in plants grown in control soils, the plants grown in heavy metal treated soils were 29.38±1.44 cms and plants grown in heavy metal + $Ca(OH)_2$ treated soils were 34.20±1.08 cms. The difference between mean values of control plants and heavy metal treated plants differed significantly (t=0.004, df =144, p<0.05). The mean values of control plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=2.62, df=144, p<0.05). The mean values of heavy metal treated plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=2.66, df=144, p<0.05). The heavy metal treated (Treatment II) plants showed lowest plant height (cms) compared to control plants and heavy metal + $Ca(OH)_2$ treated plants.

3.2 No. of branches

The number of branches of *Withania somnifera* were 2.67±0.15 in plants grown in control soils, the plants grown in heavy metal treated soils were 2.35±0.19 and plants grown in heavy metal + $Ca(OH)_2$ treated soils were 2.49±0.16. The difference between mean values of control plants and heavy metal treated plants differed significantly (t=1.34, df =144, p<0.05). The mean values of control plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=0.70, df=144, p<0.05). The mean values of heavy metal treated plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=0.53, df=144, p<0.05). The heavy metal treated (Treatment II) plants showed lowest number of branches compared to the other two treatments.

3.3 No. of leaves

The number of leaves of *Withania somnifera* were 29.39±1.56 in plants grown in control soils, the plants grown in heavy metal treated soils were 25.57±1.61 and plants grown in heavy metal + $Ca(OH)_2$ treated soils were 26.92±2.11. The difference between mean values of control plants and heavy metal treated plants differed significantly (t=1.63, df =144, p<0.05). The mean values of control plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=0.97, df=144, p<0.05). The mean values of heavy metal treated plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=0.53, df=144, p<0.05). The heavy metal treated (Treatment II) plants showed lowest number of leaves compared to the other two treatments.

3.4 No. of fruits

The number of fruits of *Withania somnifera* were 15.50±1.73 in plants grown in control soils, the plants grown in heavy metal treated soils were 19.10±1.72 and plants grown in heavy metal + $Ca(OH)_2$ treated soils were 15.37±1.56. The difference between mean values of control plants and heavy metal treated plants differed significantly (t=1.51, df =144, p<0.05). The mean values of control plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=1.57, df=144, p<0.05). The mean values of heavy metal treated plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=0.05, df=144, p<0.05). The heavy metal + $Ca(OH)_2$ treated (Treatment III) plants showed lowest number of fruits compared to the other two treatments.

3.5 Fresh weight of leaves in grams

The fresh weight in leaves of *Withania somnifera* were 10.14±0.8 gm in plants grown in control soils, the plants grown in heavy metal treated soils were 7.58±0.52 gm and plants grown in heavy metal + $Ca(OH)_2$ treated soils were 10.78±0.62 gm. The difference between mean values of control plants and heavy metal treated plants differed significantly (t=2.73, df =144, p<0.05). The mean values of control plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=3.42, df=144, p<0.05). The mean values of heavy metal treated plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=0.69, df=144, p<0.05). The heavy metal + $Ca(OH)_2$ treated (Treatment III) plants showed highest fresh weight of leaves compared to the other two treatments.

3.6 Dry weight of leaves in grams

The dry weight in leaves of *Withania somnifera* were 1.48±0.94 gm in plants grown in control soils, the plants grown in heavy metal treated soils were 1.25±0.12 gm and plants grown in heavy metal + $Ca(OH)_2$ treated soils were 1.18±0.14 gm. The difference between mean values of control plants and heavy metal treated plants differed significantly (t=1.38, df =144, p<0.05). The mean values of control plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=1.70, df =144, p<0.05). The mean values of heavy metal treated plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=0.44,

df=144, p<0.05). The heavy metal + $Ca(OH)_2$ treated (Treatment III) plants showed lowest dry weight of leaves compared to the other two treatments.

3.7 Fresh weight of stem in grams

The fresh weight in stems of *Withania somnifera* were 12.93±1.56 gm in plants grown in control soils, the plants grown in heavy metal treated soils were 10.49±1.20 gm and plants grown in heavy metal + $Ca(OH)_2$ treated soils were 10.44±0.89 gm. The difference between mean values of control plants and heavy metal treated plants differed significantly (t=1.38, df =144, p<0.05). The mean values of control plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=0.028, df =144, p<0.05). The mean values of heavy metal treated plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=1.409, df=144, p<0.05). The control plants (Treatment I) showed highest fresh weight of stems compared to the other two treatments.

3.8 Dry weight of stem in grams

The dry weight in stems of *Withania somnifera* were 1.34±0.10 gm in plants grown in control soils, the plants grown in heavy metal treated soils were 1.01±0.09 gm and plants grown in heavy metal + $Ca(OH)_2$ treated soils were 0.82±0.11gm. The difference between mean values of control plants and heavy metal treated plants differed significantly (t=2.37, df =144, p<0.05). The mean values of control plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=3.41, df =144, p<0.05). The mean values of heavy metal treated plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=1.23, df=144, p<0.05). The heavy metal + $Ca(OH)_2$ treated (Treatment III) plants showed lowest dry weight of stems compared to the other two treatments.

3.9 Fresh weight of fruits in grams:-

The fresh weight in fruits of *Withania somnifera* were 3.57±0.35 gm in plants grown in control soils, the plants grown in heavy metal treated soils were 4.36±0.3 gm and plants grown in heavy metal + $Ca(OH)_2$ treated soils were 3.71±0.35 mg/gm/wt. The difference between mean values of control plants and heavy metal treated plants differed significantly (t=1.63, df =144, p<0.05). The mean values of control plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=1.33, df =144, p<0.05). The mean values of heavy metal treated plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=0.30, df=144, p<0.05). The heavy metal treated (Treatment II) plants showed highest fresh weight of fruits compared to the other two treatments.

3.10 Dry weight of fruits in grams

The dry weight in fruits of *Withania somnifera* were 2.23±0.62 gm in plants grown in control soils, the plants grown in heavy metal treated soils were 1.88±0.26 gm and plants grown in heavy metal + $Ca(OH)_2$ treated soils were 1.80±0.27gm. The difference between mean values of control plants and heavy metal treated plants differed significantly (t= 1.05, df =144, p<0.05). The mean values of control plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=1.17, df=144, p<0.05). The mean values of heavy metal treated plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=0.20, df=144, p<0.05). The control (Treatment I) plants showed highest dry weight of fruits compared to the other two treatments.

3.11 Fresh weight of roots in grams

The fresh weight in roots of *Withania somnifera* were 4.59±0.75 gm in plants grown in control soils, the plants grown in heavy metal treated soils were 3.14±0.30 gm and plants grown in heavy metal + $Ca(OH)_2$ treated soils were 2.63±0.44 gm. The difference between mean values of control plants and heavy metal treated plants differed significantly (t=1.91, df =144, p<0.05). The mean values of control plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=0.66, df=144, p<0.05). The mean values of heavy metal treated plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=2.58, df=144, p<0.05). The control (Treatment I) plants showed highest fresh weight of roots compared to the other two treatments.

3.12 Dry weight of roots in grams:-

The dry weight in roots of *Withania somnifera* were 0.429±0.06 gm in plants grown in control soils, the plants grown in heavy metal treated soils were 0.22±0.02 gm and plants grown in heavy metal + Ca $(OH)_2$ treated soils were 0.15±0.02gm. The difference between mean values of control plants and heavy metal treated plants differed significantly (t=3.12, df =144, p<0.05). The mean values of control plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=3.96, df=144, p<0.05). The mean values of heavy metal treated plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=1.10, df=144, p<0.05). The heavy metal + $Ca(OH)_2$ treated (Treatment III) plants showed lowest dry weight of roots compared to the other two treatments.

3.13 Fresh weight of whole plants in grams

The fresh weight in whole plants of *Withania somnifera* were 31.23±2.20 gm in plants grown in control soils, the plants grown in heavy metal treated soils were 25.59±1.29 gm and plants grown in heavy metal + Ca $(OH)_2$ treated soils were 27.58±1.41 gm. The difference between mean values of control plants and heavy metal treated plants differed significantly (t=2.23, df =144, p<0.05). The mean values of control plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=0.83, df =144, p<0.05). The mean values of heavy metal treated plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=1.39, df=144, p<0.05). The heavy metal treated (Treatment II) plants showed lowest fresh weight of whole plants compared to the other two treatments.

3.14 Dry weight of whole plants in grams

The dry weight in whole plants of *Withania somnifera* were 5.42±0.435 gm in plants grown in control soils, the plants grown in heavy metal treated soils were 4.35±0.37 gm and plants grown in heavy metal + $Ca(OH)_2$ treated soils were 3.88±0.52 gm. The difference between mean values of control plants and heavy metal treated plants differed significantly (t=1.79, df =144, p<0.05). The mean values of control plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=2.38, df=144, p<0.05). The mean values of heavy metal treated plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=0.38, df=144, p<0.05). The heavy metal + $Ca(OH)_2$ treated (Treatment III) plants showed lowest dry weight of whole plants compared to the other two treatments.

3.15 Leaf area (sq. cms)

The leaf area (sq. cms) of *Withania somnifera were* 10.27±0.99 gm in plants grown in control soils, the plants grown in heavy metal

treated soils were 7.27±0.95 gm and plants grown in heavy metal + $Ca(OH)_2$ treated soils were 6.61±1.30gm. The difference between mean values of control plants and heavy metal treated plants differed significantly (t=2.09, df =144, p<0.05). The mean values of control plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=2.34, df=144, p<0.05). The mean values of heavy metal treated plants and heavy metal + $Ca(OH)_2$ treated plants were found to be significantly (t=0.42, df=144, p<0.05). The heavy metal + $Ca(OH)_2$ treated (Treatent III) plants showed lowest leaf area (sq. cms) compared to the other two treatments

Table 1: Analysis of morphological parameters of plants growth:-

Parameters	Treatment No: I (Control soil)	Treatment No: II (Soil + Heavy metal)	Treatment No: III (Soil + Heavy metal + 1% $Ca(OH)_2$)
	Mean ± SE	Mean ± S.E	Mean ± S.E
Plant Height (cms)	34.13±1.21	29.38±1.44	34.20±1.08
No. of Branches	2.67±0.15	2.35±0.19	2.49±0.16
No. of Leaves	29.39±1.56	25..57±1.61	26.92±2.11
No. of Fruits	15.50±1.73	19.10±1.72	15.37±1.56
Fresh weight of leaves in grams	10.14±0.80	7.58±0.52	10.78±0.62
Dry weight of leaves in grams	1.48±0.94	1.25±0.12	1.18±0.14
Fresh weight of stems in grams	12.93±1.56	10.49±1.20	10.44±0.89
Dry weight of stems in grams	1.34±0.10	1.01±0.09	0.82±0.11
Fresh weight of roots in grams	4.59±0.75	3.14±0.30	2.63±0.44
Dry weight of roots in grams	0.429±0.06	0.22±0.02	0.15±0.02
Fresh weight of fruits in grams	3.57±0.35	4.36±0.30	3.71±0.35
Dry weight of fruits in grams	2.23±0.62	1.88±0.26	1.80±0.27
Fresh weight of Whole plants in grams	31.23±2.20	25.59±1.29	27.58±1.41
Dry weight of Whole plants in grams	5.42±0.435	4.35±0.37	3.88±0.52
Leaf Area (sq. cms)	10.27±0.99	7.27±0.95	6.61±1.30

4. Conclusion

Ashwagandha (*Withania somnifera*) is a dry land medicinal crop and roots are used as valuable drug in traditional systems of medicine. The present aims in a detailed of the plant regarding its morphological features, Plant height in cms, number of branches per plant, number of leaves per plant, number of fruits per plant, leaves, stem, roots, leaf area and whole plant yield in term of fresh weight and dry weight.

The experimental data revealed that the external morphology plants i.e., Plant height (cms), No of branches reduced in (Treatment No II) heavy metals treated plants and also was increased in No. of leaves compared to control plants (Treatment No. I) and heavy metals + Calcium hydroxide treated plants (Treatment No III). The No. of fruits per plants, leaf area reduced in the (Treatment No III) heavy metal+ Calcium hydroxide treated plant compared to control plants (Treatment No I) and heavy metals treated plants (Treatment No II). Productivity was reduced in the plants grown in heavy metal treated plants compared to control plants. This indicates that the heavy metal effects on the growth and development of the plants and ultimately the yields.

Ashwagandha germination in earthen pots

Measuring the plant parts

Ashwagandha cultivation in field

Ashwagandha mature plants

Flowering stage of Ashwagandha

Roots of Ashwagandha

Fig 1: Macro- morphological work of *Withania somnifera* in progress.

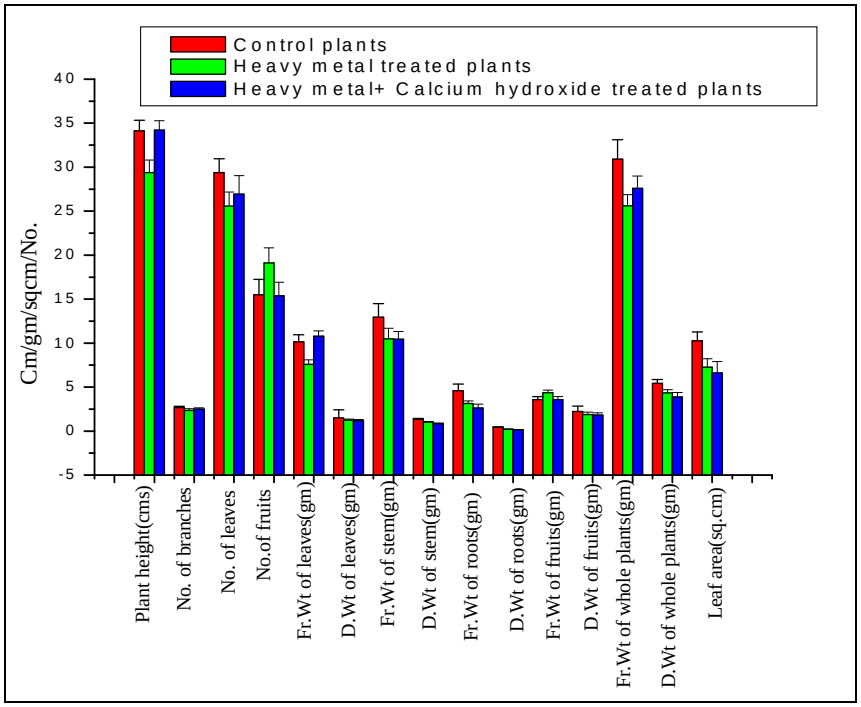

Fig 2: Graph showing the macro- morphological parameters of *Withania somnifera*

4.1 Statistical analysis
The data was analyzed using the statistical package for social science program (S.P.S.S.11). The results were expressed as Mean ± S.E.M. (standard error of mean) and % of change- Level of significance between groups were set at $P<0.05$. For comparison between different experimental groups, one way analysis of variation (ANOVA) was used followed by post hoc tests.

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6. References
1. Anonymous. The Wealth of India. Vol. 10 (Sp-W), Publications and Information Directorate, Council of Scientific and Industrial Research (CSIR), New Delhi 1982, 580-585.
2. Anonymous. The Unani Pharmacopoeia of India. Part I, Vol. I, Dept. of AYUSH, Ministry of Health & Family Welfare, Govt. of India, New Delhi 2007, 7-8.
3. Anonymous. Standardisation of Single Drugs of Unani Medicine. Part III, 1st ed. Central Council for Research in Unani Medicine (CCRUM), New Delhi, 2007, 9-14.
4. Chopra RN, Nayar SL, Chopra IC. Glossary of Indian Medicinal Plants. Council of Scientific & Industrial Research, New Delhi, 1980, 191, 258.
5. Dey KL, Bahadur R. Indigenous Drugs of India. Prime Lane, Chronica Botanica, New Delhi, 1973, 670.
6. Dymock W, Warden CJH, Hooper D. Pharmacographia Indica. Vol. 2, M/s Bishen Singh Mahendra Pal Sing, Dehradun & M/s Periodical Expert, New Delhi 1976, 566-572.
7. Heiser CB, Smith PC. The cultivated Capsicum. Econ Bot 1953; 1:214-227.
8. Kirtikar KR, Basu BD. Indian Medicinal Plants. Edn 2, Vol. 3, Lalit Mohan Basu, Allahabad, India, 1980, 1774-1777.
9. Nadkarni KM. Indian Materia Medica. Edn 3, Vol. 1, Popular Prakashan Pvt. Ltd., Bombay, 1982, 1292-1294.
10. Singh S, Kumar S. *Withania somnifera* – The Indian Ginseng Aswagandha. Central Institute of Medicinal and Aromatic Plants, 1998.
11. Scartezzini P, Speroni E. Review on some plants of Indian traditional medicine with antioxidant activity. J Ethnopharmacol 2000; 71(1-2):23-43.