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## Effect of Different Seed Treatments and Media on Growth and Biomass of Indian Cheese Maker - *Withania coagulans* (Stocks) Dunal.

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### Abstract

The present investigation was conducted to assess the effect of different seed soaking treatments and media on growth and alkaloid content of Indian cheese maker—*Withania coagulans* (Stocks) Dunal. to develop effective nursery production technology. The experiment was conducted in completely randomized design with factorial concept, including twenty treatment combinations comprising of four levels of seed treatments with GA<sub>3</sub> (0, 50, 100, 150 ppm) and five levels of different growing media. Among various concentrations of GA<sub>3</sub> as pre-soaking treatment, 100 ppm GA<sub>3</sub> (S<sub>3</sub>) showed significantly better results for germination, growth, biomass and alkaloid content in *W. coagulans*. Similarly, among growing media, red soil (M<sub>1</sub>) recorded maximum germination percentage, collar diameter, number of branches per plant, number of leaves per plant, total leaf area, length of the main root, thickness of main root, root dry biomass, shoot dry biomass, fresh weight of plant, dry weight of plant, survival percentage of the plant and total alkaloid content of plant. Overall, the result indicated that that pre-sowing *W. coagulans* seeds with GA<sub>3</sub> -150 ppm for 24 hrs and subsequently sowing treated seeds in M<sub>1</sub> media comprising of red soil (control) can enhance growth and alkaloid content.

**Keywords:** Alkaloid content, Indian cheese maker, seed treatment

### 1. Introduction

Drastic increase in sales of herbal products in global markets underscores the growing popularity of herbal therapies (Singh, 2009). In last few decades, there is a shift within many developing countries from subsistence to commercial usage of medicinal plants. In India, MAPs play an important role in the country's agricultural profile due to quantitative and qualitative advantages. MAPs cultivation can help small-scale farmers to strengthen their livelihoods and as a result, greater access to a wider range of assets can be achieved, and a capacity to build these into successful and sustainable activities.

The genus *Withania* (Family: Solanaceae) is a highly acclaimed genus of medicinal plants in the Indian Ayurvedic system of medicine because of its valuable pharmaceutical and nutraceutical properties. Among the twenty-six known species of this genus, only two (*W. somnifera* and *W. coagulans*) are economically significant (Negi et al., 2006). *W. coagulans* is commonly known as Indian cheese maker or vegetable rennet has been used for preparing vegetables rennet ferment for making cheese in different parts of India. Different parts of this plants have been reported to possess a variety of biological activities (Gupta and Keshari, 2013). The fruits are sedative, emetic alterative and diuretic possession. The leaves are used

as vegetables in Pakistan and as camel and sheep fodder. The seeds are helpful, and minimize pile inflammation (Hemalatha et al., 2008). It is distributed in the East of the Mediterranean region extending to South Asia i.e., Iran, Afghanistan, Pakistan (Sind and Baluchistan), Nepal and India, up to 1700 m. In India, it is found in Himachal Pradesh, Punjab, Uttarakhand and Rajasthan. In Rajasthan, it is sporadically distributed in Barmer, Jaisalmer and Jodhpur districts of Western Rajasthan desert and it is not common, categorized as "vulnerable species" (Pandey et al., 2012).

In last few decades, ruthless collection of *W. coagulans* for medicinal purposes, habitat destruction and climate changes has made the species to become endangered in their natural habitats. Further, low rates of natural regeneration from seeds resulted into decrease in its populations and has become scattered in the wild only (Edalatifard et al., 2014; Vakeswaran and Krishnasamy, 2003). Propagation technique needed to develop for profuse plant production of *W. coagulans*. Hence, present investigation was conducted to establish effective nursery production technique for *W. coagulans*.

### 2. Materials and Methods

The present investigation was carried out during the year 2019–20 at the Model Nursery on Medicinal and Aromatic



Plants, ASPEE College of Horticulture and Forestry, Navsari Agricultural University, Navsari. It is geographically located at 20°57' North latitude and 72°54' E longitude at an altitude of about 11.83 meters above the MSL. It is about 13 km away from the historical place "The Dandi" on the Arabian seashore. According to agro-climatic conditions, Navsari is classified under 'South Gujarat Heavy Rainfall Zone and Agricultural Ecological Situations-III'. The climate of this region is characterized by fairly hot summer, moderately cold winter and humid warm monsoon with heavy rainfall.

The experiment was laid out in Completely Randomized Design with factorial concept (FCRD). Two factors, growing media and pre-sowing treatments constituted 20 treatment combinations, three replication each and each replication consisted 15 plants per treatment. The first factor, pre sowing seed soaking in Gibberellic acid ( $GA_3$ ) had four levels  $S_1$  (Control i.e., 0 ppm  $GA_3$ ),  $S_2$  (seed soaking in 50 ppm  $GA_3$  for 24 hours),  $S_3$  (seed soaking in 100 ppm  $GA_3$  for 24 hours) and  $S_4$  (seed soaking in 150 ppm  $GA_3$  for 24 hours). Later the treated seed were sown 2-3 cm deep in perforated polythene bags (dimension: 15 x 20 cm) filled with various types of soil media under net house condition with 50% shade net. The second factor i.e., soil media had five levels namely  $M_1$  (Control),  $M_2$  (Red soil + Sand in the ratio 2:1),  $M_3$  (Red soil+Vermicompost in the ratio 2:1),  $M_4$  (Red soil+Sand+Vermicompost in the ratio 1:1:1) and  $M_5$  (Red soil + Sand + Vermicompost+Castor cake in the ratio 1:1:1:1).

The shoot characters like germination (%) up to one month, plant height (cm), collar diameter (mm), number of branches/plant, number of leaves plant<sup>-1</sup> at interval of 30 days, shoot dry biomass (g), plant fresh weight (g), plant dry weight (g) at

final stage and root characters like length of main root (cm), thickness of main root (cm), root dry biomass (g), survival percentage (%) at final stage and total alkaloid content (%) were regularly recorded. The data pertaining to all the characters studied were subjected to the statistical analysis of variance technique as described by Panse and Sukhatme (1985).

### 3. Results and Discussion

The findings revealed that application of seed treatment with  $GA_3$  and media significantly differed for growth and biomass parameters in *W. coagulans*. Among different seed treatments,  $S_4$  (seed soaked in 150 ppm  $GA_3$ ) recorded highest plant height (8.46 cm, 25.75 cm, 42.50 cm, 59.05 cm), collar diameter (0.95 mm, 1.11 mm, 1.24 mm, 1.47 mm), number of branches plant (0.22, 1.62, 2.71, 3.70), number of leaves plant<sup>-1</sup> (7.68, 33.34, 53.71, 74.79) at 30, 60, 90 and 120 DAS, respectively (Figure 1). Further, length of main root (21.24 cm), thickness of main root (0.89 cm), root dry biomass (3.20 g), total alkaloid content (4.71%), shoot dry biomass (37.39 g), plant fresh weight (61.41 g) and dry weight of plant (18.93 g) was also reported maximum in treatment  $S_4$  (seed soaking with 150 ppm) (Table 1).

Gibberellic acid is known to induce the expression of genes encoding enzymes that mobilize the reserved food material stored in the endosperm which includes starch, protein and lipids. This helps in increasing the biological yield of the plant. The overall increase in the growth of the plant is due to gibberellic acid which leads to the increase in rate of photosynthesis.

Table 1: Effect of different seed treatments and media on germination, biomass, survival percentage and total alkaloid content (%) in *Withania coagulans* (Stocks) Dunal

Treatments	Germination (%)	Fresh weight of plant (g)	Dry weight of plant (g)	Shoot dry biomass (g)	Root dry biomass (g)	Survival percentage (%)	Total alkaloid content (%)
$S_1$	54.39	54.48	16.24	31.50	2.38	78.32	2.15
$S_2$	62.76	58.38	18.11	35.59	2.95	87.81	3.93
$S_3$	64.60	59.89	18.52	36.49	3.07	89.90	4.32
$S_4$	66.43	61.41	18.93	37.39	3.20	91.98	4.71
SEm±	1.47	1.34	0.47	0.47	0.03	2.36	0.04
CD ( $p=0.05$ )	4.19	3.82	1.33	1.34	0.010	6.76	0.12
$M_1$	66.38	61.36	18.92	37.36	3.19	91.92	4.70
$M_2$	65.36	60.52	18.69	36.86	3.12	90.76	4.48
$M_3$	61.28	57.16	17.78	34.87	2.85	86.13	3.61
$M_4$	60.26	56.32	17.55	34.37	2.78	84.97	3.40
$M_5$	56.94	53.59	16.81	32.75	2.55	81.21	2.69
SEm±	1.64	1.49	0.52	0.52	0.04	2.64	0.05
CD ( $p=0.05$ )	4.69	4.27	1.49	1.49	0.11	7.55	0.27
CV %	9.16	8.96	10.04	5.14	4.56	10.52	4.30



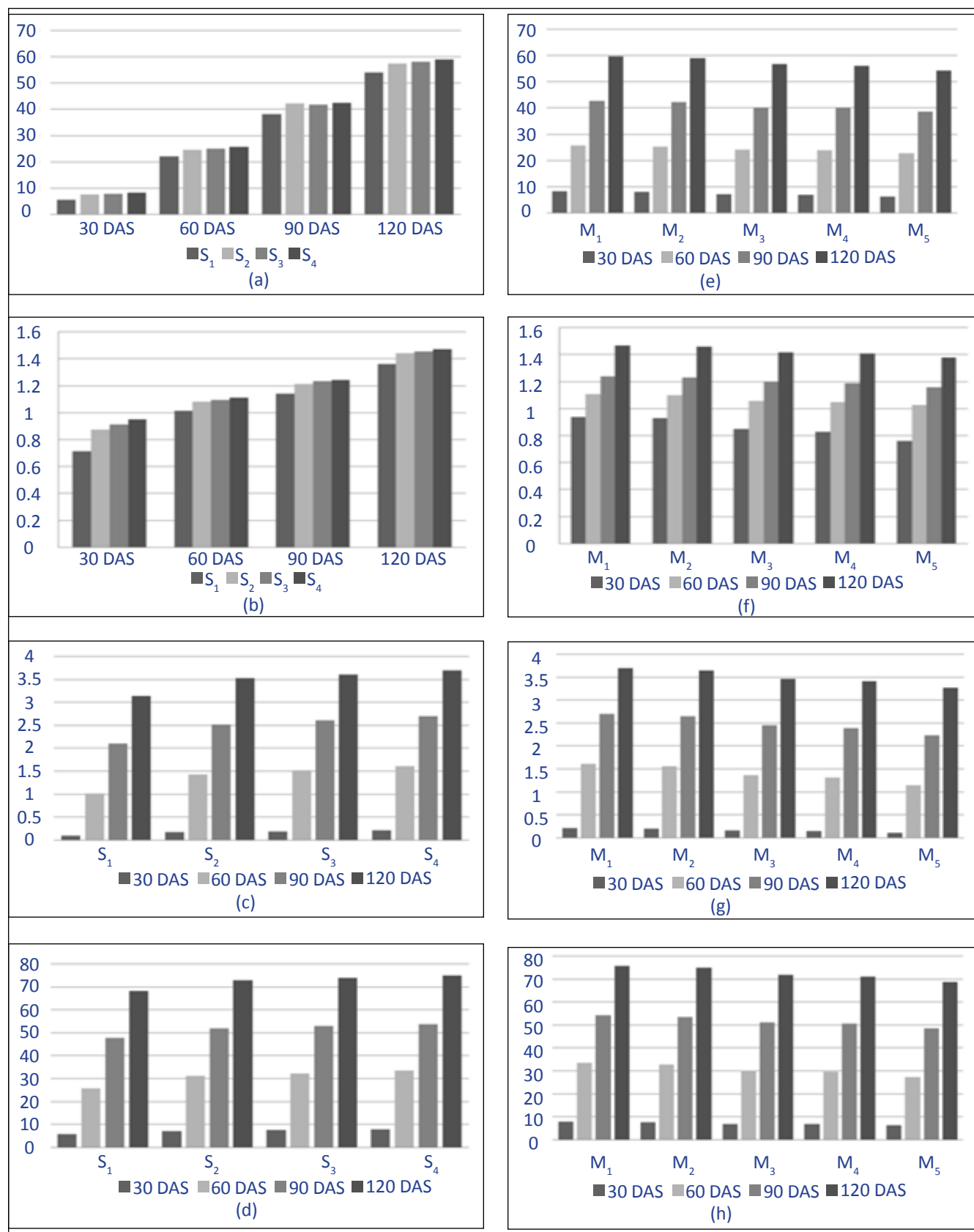


Figure 1: (a-h) Effect of pre-sowing treatment (a-d) and growing media (e-h) on (a & e) plant height, (b & f) collar diameter, (c & g) number of branches/plant and (d & h) number of leaves plant<sup>-1</sup>

The increased rate of photosynthesis helps in the higher accumulation of photosynthates within the cell and the plant and resulted in higher fresh weight and dry weight of root, shoot and plant Polaiah et al. (2020). Similar positive effect of GA<sub>3</sub> on the growth and biomass in has been reported by Velmurugan et al. (2003) in *Withania somnifera* and Swain and Malik (2018) in *Rauvolfia serpentina*. All the growth and biomass parameters were recorded lowest in treatment S<sub>1</sub> (control).

Among different media treatments, M<sub>1</sub> recorded highest plant height (8.45 cm, 25.74 cm, 42.88 cm, 59.73 cm), collar diameter (0.94 mm, 1.11 mm, 1.24 mm, 1.47 mm), number of branches plant<sup>-1</sup> (0.22, 1.62, 2.71, 3.70), number of leaves plant<sup>-1</sup> (7.67, 33.31, 54.01, 75.67) at 30, 60, 90 and 120 DAS, respectively (Figure 1). Further, M<sub>1</sub> reported maximum length of the main root (21.21 cm), thickness of main root (0.89 cm), total alkaloid content (4.71%), root dry biomass (3.19 g), shoot dry biomass (37.36 g.), fresh weight of plant (61.36 g) and dry weight of plant (18.92 g) (Table 1).

Media plays crucial role in growth and biomass under nursery condition. Vishnoi et al. (2010) recorded higher whole plant length, stem length, root length, whole plant biomass and root biomass of *Terminalia arjuna*, when seed grown in sand: soil (80:20). Dharmveer et al. (2016) found in the good results in the Sand+Soil + Cocopeat+Vermicompost (1:1:1:1) which showed significant positive effect on seed germination and seedling development in *Angelica glauca*. Abirami et al. (2010) found greater seedling height, number of leaves, shoot length, root length and plant biomass of *Myristica fragrans* when grown in soil: coir dust: sand: vermicompost. Mathowa et al. (2014) reported higher plant, leaf area, stem diameter, number of leaves, branches and pod of *Corchorus olitorius* in clay, sandy and some extent loam soil. Panchal et al. (2014) reported maximum plant height, root length, number of leaves per plant, number of branches per plant, leaf area per plant, root dry weight, root/shoot ratio, stem dry weight, total dry weight and minimum mortality in *Manilkara hexandra* when grown in Soil+Coco peat+FYM (1:1:1) along pre-treatment with IBA (1000 ppm).

#### 4. Conclusion

Germination and growth of Indian cheese maker—*Withania coagulans* can be enhanced by pre-soaking seed in the 150 ppm GA<sub>3</sub> for 24 hours and then seeding it in the red soil. Positive effect of pre-treatment on alkaloid content suggests the potential of enhancing productivity of *W. coagulans* in net-house.

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