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INHOUSE CULTIVATION OF *Withania somnifera* (ASHAWANGHA) USING HYDROPONICS

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Abstract

There is an annual demand for medicinal plant based raw materials in the world. To fulfill the increasing demands, the medicinal plants are harvested excessively from their natural habitants causing loss of biodiversity. The hydroponics technologies have proven its benefits in increasing yields of many medicinal plants. Ashwagandha is widely used herb due to its anti-microbial, anti- inflammatory, anti-tumor, anti-stress, neuroprotective, cardioprotective, and anti-diabetic properties. The objective of the present study was to investigate effect of substrate moisture level and salicylic acid concentrations on the growth and yield of *W.somnifera*. The plants were grown in an open top coir dust grow bags inside a protected house. Four moisture stress levels 100% CC (container capacity)-M_1, 80% of CC-M_2, 60% of CC-M_3, 50% of (CC)-M_0 and four strengths (10^{-2}M-S_1,10^{-3}M-S_2,10^{-4}M-S_3 and control-S_0) of Salicylic acid as a foliar spray was applied to induce the root yield. The experiment was designed based on split plot design. There were 16 treatment combinations and 3 replicates for each treatment combination of moisture and salicylic acid. Plant height, number of leaves, and internodal length were taken as growth parameters and fresh weight and dry weight of the root was taken as the yield parameter. The data was analyzed by using one way ANOVA and means were separated by Duncan Multiple Range Test (DMRT) in SAS® software. The highest plant height (76.79cm) was recorded under treatment M_0 and the salicylic acid stress level S_3. The highest number of leaves were shown under the moisture stress level M_0 and the Salicylic acid stress level S_3 which were 117 and 86 leaves per plant respectively. There was no significant influence by treatments on internodal length. Root is the main plant part harvested from *W.somnifera* due to its rejuvenate properties. Highest fresh root weight was given under the interaction effect of treatments M_2 and S_3. Highest dry root weight was given under the interaction effect of treatments M_4 and S_3. According to the results of the experiment, it can be concluded that moisture stress level 50% of container capacity and a foliar spray concentration of 10^{-4}M Salicylic acid could be applied to increase root yield of *W.somnifera.*

Keywords: Coir dust, foliar spray, moisture stress, polytunnel, root yield, salicylic acid

Introduction

Demand for herbal products accounts for a significant portion of medicinal use in Europe, United States, Asian region, and other emerging regions and the growth of medicinal herb market is expected to expand with high profit margins in next decades (Mugundhan *et al*., 2011). Due to the excessive demand, most of the medicinal plants species and their population diminishing in alarming rate and in danger of extinction due to over exploitation of plant resources and deforestation (Xego *et al*., 2016). It is a timely requirement to develop of sustainable methodologies for the cultivation of medicinal plants in order to ensure consistent supply of quality raw materials for the wellbeing of present and future of medical care.

Withania somnifera (Ashawagandha) is a medicinal plant which has higher demand in both local and foreign market. The phytochemicals in *W.somnifera* contains many medicinal properties including anti-microbial, anti- inflammatory, anti-tumor, anti-stress,

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neuroprotective, cardioprotective, and anti-diabetic properties (Dar et al., 2015). The open field cultivation results in large year-to-year variability, as both biomass production and synthesis of secondary metabolites are affected by many factors such as genotype, climatic conditions, soil type, growing practices and the presence of pests and diseases. Hydroponic technology can be applied to produce high- standard plant material all year-round in consideration of the possibility to control growing conditions and to stimulate secondary metabolism by appropriate manipulation of mineral nutrition (Maggini *et al.*, 2012). Secondary metabolite production of plants depends on stress conditions, nutrient availability, drought stress, saline stress *etc.* (Teklic *et al*., 2020). Salicylic acid, well known for systemic acquired resistance, induces in the plant response to many pathogens, can also elicit the production of secondary metabolites in plants (Singh and Dwivedi, 2018).

LC1 and FR1 are the two local varieties present in Sri Lanka (Siriwardane *et al*., 2013). The usage of the local varieties of Ashwagandha in herbal medicine has been limited due to the lower content of Withanolides and higher fibre content compared to the Indian variety. The Indian varieties of Ashawagandha cultivated in Sri Lanka, does not show similar growth and yield performance levels compared to that of grown in India. The Indian variety of Ashwagandha is imported to Sri Lanka in large scale to fulfil the requirements in herbal medicine industries. In Sri Lanka, a proper agronomic practices have not been developed to grow Indian variety of *W.somnifera* to give maximum Withanolides content. The objective of the study was to investigate the growth and yield of *W.somnifera* as affected by different moisture stress levels and Salicylic acid concentrations.

Materials and Methods

Location of the Study

The experiment was carried out in a 1500ft^2 protected house in Faculty of Technology, University of Ruhuna, Sri Lanka. The experimental area is located in the WL2B climate zone, which is a low-lying wet zone.

Cultivation of Ashwagandha

W. somnifera was grown in an open top coir dust grow bags. The plants were established in the protected house after a nursery period of one month. Same fertilizer schedule was applied for all plants. A common fungicide, insecticide and other chemical application schedule were practiced for both crop species as an initial prevention method of pests and diseases. The treatments were started to apply one month after transplanting.

Application of Moisture stress

Four moisture stress levels as 100% container capacity (CC) (M_0), 80% of CC (M_1), 60 % of CC (M_2) and 50 % of CC (M_3) was maintained. The moisture stress was applied one month after transplanting. The specific moisture stress levels were maintained using XH-M214 humidity controller soil sensor modules. Emitters of drip irrigation line was buried in each grow bag.

Application of Salicylic acid stress

There were four Salicylic acid stress levels as without Salicylic acid (S_0) ,10^{-2} M (S_1),10^{-3}M (S_2), 10^{-4}M (S_3) under each moisture level. Salicylic acid was applied as foliar spray once in two weeks.

Experimental design

There were three replicates for each treatment combination and 16 treatment combinations for whole experiment. One grow bag was considered as one experimental unit and two plants were grown in each grow bag. The treatments were arranged in a split plot design.

Data collection

Growth parameters of the plant (plant height, internodal length, number of leaves in main stem/plant) were measured at two weeks interval after commencement of application of treatments. Fresh weight of root/plant and dry weight of root/plant were measured as yield parameters at harvesting.

Data analysis

Collected data was analyzed using one way ANOVA and the means were separated using Duncan Multiple Range test (DMRT). The test was carried out using SAS software.

Results and Discussion

The Salicylic acid levels and the moisture stress levels had a significant additive effect on the plant height of *W.somnifera* as given in Figure 1. The highest plant height (80.33cm) was recorded under treatment M_0 (substrate moisture equals to field capacity). The lowest average plant height of 62.79cm was recorded in the treatment of M_3 (substrate moisture equals 50% of field capacity). The highest plant height (76.79cm) was shown under the Salicylic acid stress level of S_3 while the lowest plant height (63.42cm) was given under the treatment S_0.

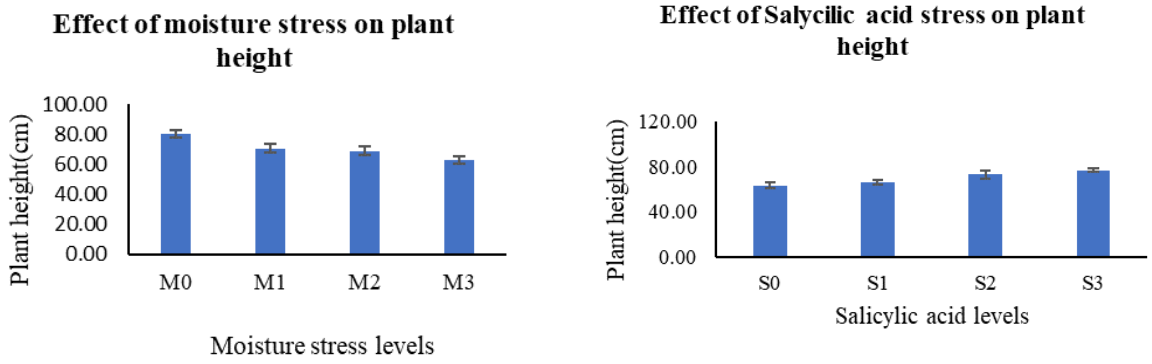

Figure 1: Plant height

There was a significant effect on number of leaves of *W.somnifera* from the applied moisture stress and Salicylic acid stress. The highest number of leaves was shown under the moisture stress level M_0 and the Salicylic acid stress level S_3 which were 117 and 86 leaves per plant respectively (Figure 2).

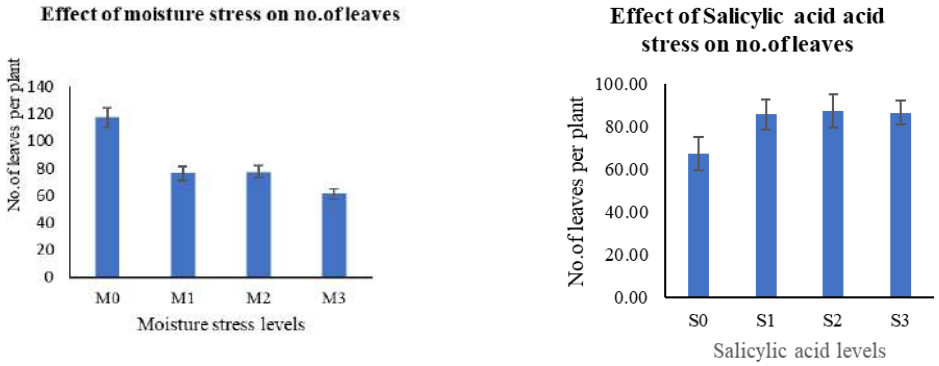

Figure 2: No. of leaves per plant

Root is the main plant part harvested from *W.somnifera* cause of its rejuvenative properties. Highest dry root weight was given under the interaction effect of treatments M_4 and S_3 as shown in Figure 3. The metabolic changes mediated by drought stress results in accumulation of higher natural products in medicinal plants (Kleinwachter and Selmar 2014). This can be the reason for the increment of dry weight observed in our study.

Similar results of decreased plant height with the application of drought stress have already been shown in *Satureja hortensis L* (Baher *et al*.,2002) and *Trachyspermum ammi l* (Azhar *et al*., 2011). Adequate water supply is one of the key factors which, determines the growth of a plant. The drought stress conditions created by the restricted water supply causes huge damage to physiology of the plant. The damages occurred to the plant physiology will be indicated by the degradation of morphological characteristics of plant including plant height, number of leaves and number of branches and number of fruits (Pradhan *et al*.,2017).

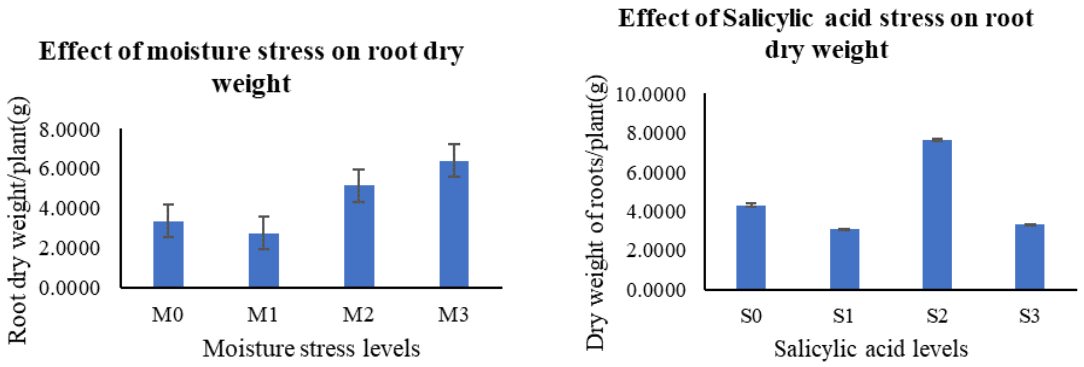

Figure 3: Dry weight

The Salicylic acid concentration 10^{-2}M (S_1) cannot be recommended for foliar spray since it was not shown any significant difference with that of control. Similar results were obtained during the study of "Salicylic acid-induced modulation of growth and metabolism of medicinal plant; *Mentha spicata*. That study has recommended 200 µg ml^{-1} concentration of Salicylic acid for the increment of shoot length, branch number per plant, leaf area, leaf number, and plant biomass, when compared with control plants (Kundu *et al*., 2018).

Conclusion

Results can be concluded that the *W.somnifera* plants have to be grown in moisture stress level -50% of container capacity with a foliar spray of Salicylic acid concentration 10^{-4}M. The recommended Salicylic acid level and the moisture stress level showed a root yield increment of 24.12% and 46.18% respectively related to the control.

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