Biological Forum – An International Journal **16(8): 125-128(2024)**

ISSN No. (Print): 0975-1130
ISSN No. (Online): 2249-3239

Foliar Spray of Growth Regulators Modifies Phenology and Physiology of *Withania somnifera* (L.) Dunal

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(Received: 20 May 2024; Revised: 15 June 2024; Accepted: 11 July 2024; Published: 14 August 2024)
(Published by Research Trend)

ABSTRACT: Research experiment was conducted to investigate the effect of growth regulators spray on phenology and physiology of *Withania somnifera* during *Rabi* season 2022-2023 at Herbal Garden, Department of Plant Physiology, JNKVV Jabalpur (M.P.) India. Pot experiment comprises of eight treatments *viz*., water spray (T1), NAA@50ppm (T2), NAA@100ppm (T3), BA@50ppm (T4), BA@100ppm (T5), IBA@50ppm (T6), IBA@100ppm (T7), IBA@150ppm (T8) conducted in Completely Randomized Block Design replicated thrice. The result revealed the delayed occurrence of primary branches initiation with maximum delayed primary branching was found with the individual spray of IBA@50/100/150ppm and NAA@100ppm where as maximum delay in secondary branch initiation was noted with the spray of BA@50/100ppm and NAA@50ppm individually. The maximum days to floral initiation, first fruit maturity and 50% fruit maturity was found when no growth regulator sprayed. Fruit initiation was delayed to its maximum due to water, NAA@50ppm and BA@100ppm individually, while maximum delay in physiological maturity founded under the spray of BA@50ppm which is statistically similar with NAA@100ppm. The maximum RWC, WSD, MSI and Proline was achieved under the spray of NAA@50ppm, BA@50/100ppm and IBA@150ppm respectively. No definite sequence or pattern of CCI was recognized due to spraying of growth regulators.

Keywords: *Withania somnifera*, growth regulators, NAA, BA, IBA, RWC, MSI, WSD, Proline, phenology.

INTRODUCTION

Ashwagandha (winter cherry) is one of the most important medicinal crops commercially cultivated in India. Ashwagandha [*Withania somnifera* (L.) Dunal.] belongs to Solanaceae family. The roots and leaves of this plant contain many alkaloides and withanolides (Chadda, 2001). Root is the most important plant part used medicinally to cure many human ailments. It is used to increase vigour, vitality and to cure diseases like impotency, cough, rheumatism, skin diseases, fever, ulcers and leucoderma, dropsy, stomach and lung inflammation and several female disorders. Roots are also known as Indian Ginseng in International markets as it possesses rejuvenating, antistress and aphrodisiac properties.

It is grown mainly as late sown rainy season crop and is ready for harvest in 150-170 days after sowing (Bhaure *et al.,* 2012). It is cultivated over an area of 10,780 ha with a production of 8429 tonnes in India (Srivastava *et al*., 2018). The productivity of this important crop is low and the crop is not much responsive to fertilizers. Fertilizer application even affects the medicinal quality of ashwagandha root and other plant parts. In such situations, there is strong need to search possible inputs to improve the productivity and quality of this crop out of Plant growth regulators, biofertilizers, etc. Plant growth regulators play important role in seed germination, cell enlargement, cell division, root formation and thereby influencing growth, productivity and quality of many important horticultural crops. Spraying of various PGR with different doses modifies plant metabolism and thereby growth and productivity of many crops like chilli, tomato, coriander, fennel, fenugreek etc. Although some work has been done by scientist on *Withania somnifera* crop regarding optimization of protocols for plantlet regeneration (Jhankare *et al.,* 2011a; Jhankare *et al.,* 2011b), optimization of seed rate (Tiwari *et al*., 2002), development of resistant line against leaf blight disease (Jhankare *et al*., 2011b), effect of season of planting on productivity and alkaloid partitioning (Baraiya *et al*., 2005; Baraiya *et al*., 2012), but very limited work is reported so far on use of PGR to enhance growth, productivity and root quality of Withania somnifera (Mohare *et al*., 2023; Bhaure *et al*., 2012; Barathkumar and Manivannan 2018; Bhaure *et al*., 2014), comparative efficacy of chemical and plant based herbicides on Ashwagandha (Kulmi and Tiwari 2005; Kulmi and Tiwari 2006) to study the influence of growth regulators spray on phenology and physiology

of *Withania somnifera.* Hence the experiment was conducted.

MATERIALS AND METHODS

The experiment was laid out in polybags at the Herbal garden, Department of Plant Physiology, Jawaharlal Nehru Agriculture University, Jabalpur during *rabi* season 2022-2023 from 17th October 2022 to 18th May 2023 in complete randomized block design with 8 treatments (various concentrations of plant growth regulators) replicated thrice. The treatments were Control- water spray, NAA- 50 mg L^{-1}, NAA- 100 mg L^{-1}, BA- 50 mg L^{-1}, BA- 100 mg L^{-1}, IBA- 50 mg L^{-1}, IBA- 100 mg L^{-1} and IBA- 150 mg L^{-1}. The study site comes under the subtropical region. *Withania somnifera* (Variety: Jawahar Ashwagandha -20) was taken as experimental material and seed rate was 8 kg ha^{-1}. Soil was low in organic carbon with clay loam texture. The polybags were uniformly filled with the soil and FYM (2:1). Seeds were sown at the depth of 2-3 cm in polybags and covered with vermicompost (32.21 gm $polybags^{-1}$). Two thinning were done first at 30 DAS to remove excess plants for maintaining plant population (5 plant $polybag^{-1}$) and at 60 DAS. Four polybags were taken per treatment per replication. Eight plants were randomly selected in each treatment in the respective replication for morphological observations. RWC (Relative Water Content), WSD (Water Saturation Deficit), MSI (Membrane Stability Index) and proline content were estimated as per procedure used by Barrs and Weatherley (1962); Deshmukh *et al*. (1991); Bates *et al.* (1973) respectively. Statistical Analysis was done by the method given by Fisher (1967).

RESULTS AND DISCUSSION

A. Phenological characters

Phenology of *Withania somnifera* revealed varied results with reference to foliar sprays of various growth regulators at 25 and 55 days after transplanting (Table 1 and 2). Various growth regulators spray significantly affected days taken to primary branch initiation, days taken to secondary branch initiation, days to floral initiation, days to fruit initiation, days to first fruit maturity, days to 50% fruit maturity and days to physiological maturity (Table 1). IBA 50 mg l^{-1} delayed primary branch initiation as well as secondary branch initiation maximum upto 43.33 DAS and 53 DAS respectively, while IBA 150 mg l^{-1} promotes early initiation of primary as well as secondary branching.

Table 1: Effect of Plant Growth Regulators spray on Phenological characters of *Withania somnifera.*

Treatments	Days to first branch initiation	Days to secondary branch initiation	Days to tertiary branch initiation	Days to floral initiation	Days to 50% flowering	Days to fruit initiation
T_1 (water spray)	39.33	52.00	60.33	67.33	76.67	85.67
T_2 (NAA@50 ppm)	41.33	50.67	60.67	64.33	75.67	84.67
T_3 (NAA@100 ppm)	42.00	51.00	61.33	65.00	75.33	83.67
T_4 (BA@50 ppm)	39.67	49.67	62.67	65.33	75.67	83.00
T_5 (BA@100 ppm)	42.33	51.67	61.67	65.00	76.00	84.67
T_6 (IBA@50 ppm)	43.33	53.00	61.00	65.33	76.00	83.33
T_7 (IBA@100 ppm)	40.33	52.67	61.33	65.00	75.67	83.00
T_8 (IBA@150 ppm)	38.67	49.33	60.00	64.67	75.67	80.33
SEm±	**0.514**	**0.441**	**0.842**	**0.441**	**0.456**	**0.456**
CD (at 5%)	**1.540**	**1.322**	**NS**	**1.322**	**NS**	**1.368**

Table 2: Effect of Plant Growth Regulators spray on Phenological characters of *Withania somnifera.*

Treatments	Days to 50% fruiting	Days to first fruit maturity	Days to 50% fruit maturity	Days to physiological maturity	Days to physical maturity
T_1 (water spray)	109.33	138.67	148.33	164.00	173.67
T_2 (NAA@50 ppm)	108.67	137.33	147.67	162.67	173.33
T_3 (NAA@100 ppm)	108.00	136.67	146.67	164.67	172.67
T_4 (BA@50 ppm)	108.67	136.33	145.33	165.33	172.33
T_5 (BA@100 ppm)	108.67	137.00	145.67	163.67	174.67
T_6 (IBA@50 ppm)	107.00	136.33	145.67	164.00	174.00
T_7 (IBA@100 ppm)	107.00	135.33	146.33	163.67	173.67
T_8 (IBA@150 ppm)	108.00	135.00	146.00	163.33	173.00
SEm±	**0.745**	**0.408**	**0.471**	**0.408**	**0.540**
CD (at 5%)	**NS**	**1.224**	**1.413**	**1.224**	**NS**

Because of the spray of plant growth regulators of auxin nature promotes growth of plant and apical dominance, hence branching delayed (Prasad and Kumar 2005; Bhaure *et al*., 2012; Gare *et al*., 2017; Barathakumar *et al*., 2018). Water spray delayed floral initiation maximum upto 67.33 DAS as compared to NAA 50 mg L^{-1} (64.33 DAS) auxin application in lower dose promotes early flowering and fruiting. Days to fruit initiation delayed due to water spray (85.67 DAS), while IBA 150 mg l^{-1} (80.33 DAS) promotes early fruit initiation. Days to first fruit maturity and 50% fruit maturity also delayed due to water spray, while IBA 150 mg l^{-1} and BA 50 mg l^{-1} promotes early fruit maturity and 50% fruit maturity respectively. Physiological maturity is the development stage after which non significant increment in biomass occurs in

plants. Physiological maturity, if prolonged in *Withania somnifera*, the crop produces more biomass accumulation in plant and may increases the economic productivity of root. Maximum delay in physiological maturity is due to the spray of BA 50 mg l^{-1} (165.33 DAS), while earliest physiological maturity was noted in the spary of NAA 50 mg l^{-1} (162.67 DAS). Delayed maturity due to foliar spray of BA and IBA as individual application may possibly be due the reason that Cytokinin (BA) and Auxin (IBA) help to delay senescence, to promote biomass accumulation and to prolong vegetative growth of crops thus resulted in delayed occurrence of phenophases of Withania. These results are also in conformity with the findings of Bhaure *et al*. (2014).

B. Physiological Characters

Efficient use of captured solar energy is the key factor controlling the productivity of any crop. Plant growth comprises the conversion of substrate molecules into specific component and subsequent utilization and its storage in the plant. Higher yield per unit of substrate can be achieved only by the production of energetically cheaper storage organs. Maintenance of storage organs (sink) during their development also consumes 6 to 25% of total substrate for their growth (Penning Devries et al. 1983). Physiological efficacy of plant depends on accumulation and retention of water and nutrients within plants by maintaining good water status and stable membranes without any potential leakages. RWC, WSD, MSI as well as proline content in leaf of plants are important physiological parameters depicting suitability to survive and out performed under various environmental conditions. At the time of maturity, the maximum RWC (85.69%) was achieved in NAA@50 mgL^{-1}, while the minimum RWC (67.54%) was recorded in BA@100 mgL^{-1}. RWC is the parameters showing percentage amount of water retained by leaf of plant with reference amount of water retained in plant leaf at water saturation. This indicates that increase in RWC mean more suitability of plant to resist water stress. NAA is synthetic auxin in lower dose (50 mgL^{-1}) may trigger Ashwagandha plant to retain more water in their leaves in order to tolerate water stress and thereby biomass productivity. BA on the other hand delays senescence with less retention of water by decreasing RWC compromising their fitness against water stress of Withania commonly grown under water stress conditions.WSD is just opposite to RWC hence plant having maximum RWC will have least deficit against water saturation. The maximum WSD (31.51%) was registered with BA@100 mgL^{-1} and BA@50 mgL^{-1} individually, while the minimum WSD (19.95%) was observed in NAA@50 mgL^{-1} individually. The highest MSI (56.15%) was observed in BA@100 mgL^{-1}. It is an established fact that BA as synthetic cytokinin delays senescence by maintaining stay green character via preventing degradation of chlorophyll and other pigment along with anti oxidative activities for membrane proteins. This may increase the stability of membrane structure and functions. The proline content was maximum (80.91µmol g^{-1}fw) in IBA@150 mgL^{-1}, while the minimum (54.39 µmol g^{-1}fw) was noted with NAA@100 mgL^{-1}. Proline content always increases in response to water and any stress in order to increase compatible osmolyte in cell and their by improve cells fitness to accumulate more water in spite of stressfull conditions. IBA in higher dose of 150 mgL^{-1} mimic stress conditions and triggers proline synthesis to accumulate and retain more water for use in future commencement of stresses.

Table 3: Effect of Plant Growth Regulators spray on RWC (%), WSD (%), MSI (%) and Proline content (µmol g^{-1} fw) in *Withania somnifera*.

Treatment	RWC (%)	MSI (%)	WSD (%)	PROLINE (µmol g^{-1}fw)
T_1 (water spray)	67.85	47.35	31.18	58.56
T_2 (NAA@50 ppm)	85.69	52.94	19.95	76.81
T_3 (NAA@100 ppm)	75.85	38.58	20.84	54.39
T_4 (BA@50 ppm)	68.03	35.44	31.26	63.93
T_5 (BA@100 ppm)	67.54	56.15	31.51	59.60
T_6 (IBA@50 ppm)	77.84	51.64	21.43	70.52
T_7 (IBA@100 ppm)	76.57	39.69	23.76	75.42
T_8 (IBA@150 ppm)	74.9	51.63	21.36	80.91
SEm±	**0.151**	**0.156**	**0.105**	**0.341**
CD (at 5%)	**0.453**	**0.468**	**0.314**	**1.021**

CONCLUSIONS

Hence it is concluded that spray of IBA in lower dose (50 mgL^{-1}) delayed branching in Withania somnifera while higher dose (150 mgL^{-1}) promoted early flowering and fruiting. In addition, spray of NAA@50 mg L^{-1} found best for maximum Relative Water Content. Spray of BA@100/50 mg L^{-1} were equally effective in achieving maximum Water Saturation Deficit. The maximum Membrane Stability Index was achieved with BA@100 mg L^{-1}. Spray with IBA@150 mg L^{-1} recorded the highest Proline content.

Acknowledgement. Authors are highly thankful to the department of Plant Physiology, CoA, Jabalpur for providing field and lab facility to conduct this research work.
Conflict of Interest. None.

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How to cite this article: Pratibha Goud, Gyanendra Tiwari, Prabha Tigga, Abhilasha Tiwari, Subrata Sharma, Anubha Upadhyay, R. Shiv Rama Krishnan, Preeti Sagar Nayak and R.K. Samaiya (2024). Foliar Spray of Growth Regulators Modifies Phenology and Physiology of *Withania somnifera* (L.) Dunal. *Biological Forum – An International Journal, 16*(8): 125-128.