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Impact of different seed rates with planting methods on withanolide content of ashwagandha

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Abstract
The present experiment was conducted at the Research Farm of Dr. KSG Akal College of Agriculture, Eternal University, Baru Sahib (H.P.) during the summer season of 2021. The trial consisted of two factors, namely seed rates and planting methods, resulting in eight treatment combinations. These combinations were arranged in a completely randomized design with three replications to ensure consistent and unbiased results. The eight treatments included two different seed rates (S1 - 4 kg ha^{-1}, S2 - 5.5 kg ha^{-1}) and four methods of planting (M1 - broadcasting, M2 - Line sowing, M3 - Transplanting randomly, M4 - Transplanting in lines). It was observed that treatment M3 (Transplanting randomly) resulted in significantly higher withanolide content. The different seed rates did not have a significant impact on the withanolide content. In terms of the interaction effect, the treatment M3S2 (Transplanting randomly + 5.5 kg per hectare) yielded the highest withanolide content.

Keywords: Ashwagandha, planting methods, seed rates, transplanting, *Withania*, withanolide content

Introduction
The Indian Himalayan region is a significant area for plant bio-diversity, housing a large number of medicinal plants that contribute to crop diversification in the Himachal Pradesh Agriculture model. However, the bio-diversity of Himachal Pradesh is currently facing substantial pressure due to the rising demand for natural health care substances for trade purposes. Ashwagandha (*Withania somnifera*) is particularly important as a medicinal plant due to its alkaloid and withanine content. Therefore, it is crucial for scientists to give special attention to the cultivation techniques of this herbal plant in order to enhance its productivity in the hill land agriculture of Sirmour district (H.P).

The experimental studies have also discovered that the ashwagandha plant contains active withanolides and alkaloids, which have medicinal properties that can be used in ayurvedic and yunani drugs to enhance brain function and improve memory. The alkaloid found in the plant roots has antibiotic, anti-tumour, and anti-cancer properties. The paste made from the plant's green leaves and roots is used to alleviate joint pain and inflammation. The leaves are also used to provide relief for eye diseases. Additionally, ashwagandha promotes reproductive functions and maintains balance in human beings.

With a demand of 7000 tonnes per year, the projected yearly production of ashwagandha in India is just about 1500 tonnes. The need to develop the appropriate technology to close the production gap has become crucial for agronomists working on this herbal crop (Anonymous, 2009). According to recent research studies, the main factors limiting this crop's poorer yields in the nation include its shy germination, inadequate planting techniques, and various environmental circumstances (Raghuveer, 2019) [7].

The Sirmour district is located in Himachal Pradesh's mid-Himalayan area, which has unusual weather features like erratic rainfall and chilly temperatures in the winter. The area's topography is sloppy, uneven, and dangerously prone to erosion. Another barrier to achieving the ideal plant population is the seed's poor intrinsic behavior for germination. It is necessary to determine the seed rate per unit area and adopt the proper plant geometries with the right to direct seedling v/s nursery transplanting randomly or in line sowing pattern to promote its aerial and root growth in order to overcome all of these limitations from the Mid-Himalayan Region of Himachal Pradesh. The farmer typically uses a seed rate of less than 4 kg/ha, however experimental results support using a higher seed rate. To reduce the shy germination effect of its seeds, its seeds must be sown in a covered nursery bed so that seedlings will be 30-35 days old before they are transplanted into the main field, either randomly or in line, depending on the soil conditions.

This technology may not only increase the plant's height, branching, and number of leaves, but it also has the potential to lengthen and improve the quality of the roots, which contain the majority of the active pharmacological ingredients like withanolides and alkaloids.

Material and Method

The experiment was carried out in the Dr. K.S. Gill Akal College of Agriculture's Research Farm in the summer of 2021 at Eternal University, Baru Sahib. About 985.8 mm of rain fell on the area each year, primarily from June to September. The direct seeding of ashwagandha was carried out by adopting two different sowing techniques (broadcasting and line sowing) with two different levels of seed rates (4 kgha-1 and 5.5 kgha-1), while maintaining the row distance at 25cm apart. This was done on July 25, 2021, simultaneously in the field and nursery bed. A modified spectrophotometer approach was used to estimate the withanolides content quantitatively.

Result and Discussion

The data in Table 4.8 revealed that the phytochemical property (withanolide) in plant root did not changed significantly with seed rates (S_1 and S_2) treatments. The withanolides contents under both seed rates S_1 (4kg ha^{-1}) and S_2 (5.5kg ha^{-1}) were read as 0.37 and 0.39 % respectively.

The different methods of sowing have shown significant difference for withanolide content in plant roots. Highest withanolide percentage was recorded in plant roots under M_3 (Transplanting randomly) with 0.51% value and rated significantly superior to the withanolide contents recorded under all other planting methods i.e M_4, M_1, M_2 with withanolide content of 0.45, 0.32 and 0.25 % respectively. M_2 (line sowing) methods of planting was rated inferior to all other method of planting in withanolide content of plant roots.

The data of interaction effect involving seed rates and methods of planting failed to enhance the phytochemical property (withanolides) of plant root significantly. Thought the highest value of withanolides content (0.52%) was recorded under S_2M_3 (5.5 kg ha^{-1} + Transplanting randomly) and lowest (0.24%) from S_1M_2 (4 kg ha^{-1} + line sowing) at 180 DAS (harvest).

There is no significant effect of seed rate on withanolide content in ashwagandha at harvested, through the higher numerical values of withanolide were obtained from high populated field plot in comparison to low populated crop. Similar observation have also been narrated by Desai *et al* (2017) [2] who reported the plant population have no impact on quality parameters like total withanolide yield.

The different methods of planting have shown significant differences on withanolide content of ashwagandha. Highest withanolide percent was recorded under M_3 (0.51%) at harvest, followed by M_4 (0.45%), M_1 (0.32%) and M_2 (0.25%). The broadcasting / random transplanting method is reported the best method of planting for ashwagandha which recorded highest value of alkaloid and withanolide content (Pandey and Shukla 2007) [8].

Table 1: Effect of seed rates and methods of sowing on phytochemical property (withanolides) at 180 DAS (harvest)

S. No	Treatments	Withanolides
A	**Seed rates**	
	S_1 - 4kg ha^{-1}	0.37
	S_2 – 5.5kg ha^{-1}	0.39
	S. Em±	0.01
	CD $_{0.05}$	NS
B	**Methods of planting**	
	M_1 – Broadcasting	0.32
	M_2 - Line sowing	0.25
	M_3 - Transplanting randomly	0.51
	M_4 - Transplanting in lines	0.45
	S. Em±	0.01
	CD $_{0.05}$	0.03

Table 2: Interaction effect of seed rates and methods of sowing on phytochemical property (withanolides) at 180 DAS (harvest)

	Harvest			
	M_1	**M_2**	**M_3**	**M_4**
S_1	0.31	0.24	0.50	0.44
S_2	0.33	0.26	0.52	0.46
S. Em ±	0.02			
$CD_{0.05}$	NS			

Conclusion

withanolide percentage in plant roots remained highest under M_3 (Transplanting randomly) with 0.51% value and rated significantly superior to the withanolide contents recorded under all the planting methods i.e M_4, M_1, M_2 with withanolide content of 0.45, 0.32 and 0.25 % respectively. M_2 (line sowing) methods of planting was rated inferior to all other method of planting in phytochemical (Withanolide) parameter of ashwagandha. The application of both seed rates S_1 and S_2 failed to affect its contents significantly

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