



## Physiological and Morphological Variation in *Withania somnifera* under Contrasting Agro-Climatic Conditions

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**Abstract:** *Withania somnifera* (Ashwagandha) is a medicinal plant from the Solanaceae family that is highly regarded for its root-derived withanolides. In order to identify the primary traits that influence productivity, this investigation evaluated physiological and morphological variation in two starkly different agro-climatic environments: Chennai (Tamil Nadu) and Jabalpur (Madhya Pradesh). Uniform agronomic practices were implemented during field trials in rainfed kharif conditions. The average plant height was 80.30 cm in Chennai and 88.53 cm in Jabalpur, while the root yield was 8.58 g and 10.57 g per plant, respectively. Regression analysis revealed that plant height and root area significantly influenced root yield in Chennai ( $p < 0.05$ ), whereas leaf length was the only trait showing a significant association with root yield in Jabalpur ( $p < 0.05$ ). Differences in shoot–root ratio between locations were also statistically significant, indicating environment-specific biomass allocation patterns. In Chennai, root area and plant height were identified as significant predictors of root yield, while leaf length was the sole significant factor in Jabalpur. The increased allocation of biomass to roots was indicated by lower shoot–root ratios. The results emphasize the genetic and phenotypic diversity of *Withania somnifera* and identify traits—particularly shoot–root ratio, root area, and leaf length—as potential selection criteria for breeding initiatives that are designed to enhance yield and stress tolerance. This study contributes to plant science by demonstrating how agro-climatic variability alters trait–yield relationships in a medicinal crop of high economic importance. It advances understanding of phenotypic plasticity and biomass allocation strategies in *Withania somnifera*, and establishes measurable traits—such as shoot–root ratio, root area, and leaf length—as reliable indicators of adaptive performance and yield potential.

**Keywords:** *Withania somnifera*, Agro-climatic adaptation, Physiological variation, Root yield, Breeding selection.

### 1. INTRODUCTION

Commonly referred to as Ashwagandha, Indian ginseng, or winter cherry, *Withania somnifera* (L.) Dunal is a perennial medicinal shrub from the Solanaceae family that has been utilized in Ayurveda for over 4,000 years as a Rasayana (rejuvenate tonic) to enhance resilience, vitality, and longevity (Ashwagandha extraction and its health benefits (Bokelmann, 2022)). It is cultivated in Africa, the Middle East, and the Mediterranean regions, and it is native to the Indian subcontinent. It thrives in arid, subtropical climates (Akbar, 2020). The plant is highly regarded for its adaptogenic, neuroprotective, immunomodulatory, and anti-inflammatory properties, which are primarily attributed to steroidal lactones (withaferin A, withanolide A, withanone), as well as alkaloids and flavonoids (Jamalludin et al., 2022; Kumar et al., 2023). Pharmacological investigations have illustrated numerous mechanisms of action, such as the regulation of neurotransmitters, the hypothalamic-pituitary-

adrenal axis, antioxidant activity, and cytokine balance (Singha et al., 2024; Shivananjappa et al., 2022). Traditionally, root and leaf preparations have been employed to alleviate tension, anxiety, reproductive disorders, arthritis, respiratory ailments, and metabolic dysfunctions (Pandya et al., 2022; Tuli et al., 2023). It is renowned for its adaptogenic, anti-inflammatory, and neuroprotective properties (Mir et al., 2012). The physiology of this plant involves the biosynthesis of bioactive secondary metabolites, including withanolides, alkaloids, and flavonoids, which are responsible for its pharmacological potential (Kumar et al., 2007). Its advantages in augmenting cognitive performance, reducing cortisol levels, supporting physical endurance, and improving sleep quality are further substantiated by contemporary clinical trials (Jędruszczak et al., 2023; Raj et al., 2022). In order to optimize its therapeutic potential, the necessity for standardized cultivation, phytochemical

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characterization, and region-specific selection is underscored by the increasing global interest in *Withania somnifera*.

Genetic diversity, environmental conditions, and agronomic inputs influence physiological and morphological characteristics, including the number of seeds per berry, the length of the leaf, the height of the plant, and the biomass yield. Amphimixis, open pollination, and facultative autogamy are the defining characteristics of reproductive biology, which guarantees reproductive resilience. The number of seeds per berry varies between 10 and 30, contingent upon the genotype and environmental conditions (Mir et al., 2012; Kumar et al., 2007). Nitrogen application at 120 kg/ha has been reported to optimize leaf area (Thakur et al., 2014), while optimal tillage depth and plant density can increase shoot biomass by over 20% (Kothari et al., 2003). Das et al. (2010) have also disclosed stable phenotypic variants, such as thick-leaf mutants with enhanced leaf dimensions, through induced mutagenesis studies. It is essential to comprehend the interplay between physiological processes, morphological variability, and environmental adaptability in *Withania somnifera* in order to optimize cultivation strategies, enhance phytochemical yield, and create stress-tolerant, high-yielding varieties that guarantee sustainable medicinal production.

## 2. MATERIALS AND METHODS

### 2.1. Site Location

Chennai (Tamil Nadu, India). Chennai is characterized by a tropical savanna climate with minimal seasonal temperature variation. In May–June, temperature maxima frequently exceed 38 °C, while winter minimums are typically around 22 °C. The northeast monsoon (Sept–Dec) is the primary source of precipitation, with an average annual rainfall of approximately 1,400 mm. Southwest rainfall (June–Sept) is also present. Jabalpur (Madhya Pradesh, India) is located at an elevation of approximately 412 meters. The city is characterized by a subtropical climate, with summer temperatures reaching as high as approximately 46 °C and winter temperatures as low as 2 °C. The majority of the annual rainfall, approximately 1,315 mm, is received between mid-June and September, with intermittent winter showers occurring in December and January.

Predominantly sandy loam to light red soil with excellent drainage and neutral to alkaline pH (7.5–8.0) are the soil characteristics of Chennai. The soil was well-aerated and had minimal moisture retention, which facilitated root growth. However, it is typically low in organic carbon and

nitrogen. The soil at the experimental site in Jabalpur is sandy loam with a pH of approximately 7.5. The nutrients were moderate, with P and K at approximately 16 and 236 kg/ha, respectively. Nitrogen was low at approximately 206 kg/ha, and organic carbon was approximately 0.3%.

### 2.2. Crop Management and Cultivation

Uniform agronomic practices were implemented in the planting of *Withania somnifera* varieties in both Chennai and Jabalpur locations (Kumar et al., 2020; Sharma et al., 2017). An excellent tillage was established by pre-monsoon tillage. In accordance with the cultivation of the kharif season, direct sowing took place in July–early August following the initial rainfall. Irrigation was infrequent and was implemented solely during periods of drought, as both regions depend on rainfed conditions (Sharma et al., 2017). Observations: Vegetative characteristics include the number of branches, the duration of flowering and maturity, and the height of the plant. Plant reproductive characteristics include the number of fruit and the quantity of seeds produced per plant (Kulkarni et al., 2017; Nikam et al., 2013). Root characteristics include the number of primary and secondary roots, the length and diameter of the main root, and the weight of the fresh and dried roots. At the appropriate growth phases, all measurements were conducted in accordance with established protocols (Kumar et al., 2020), (Figure 1 a,b,c). Data on morphological and physiological traits and root yield were analyzed to determine the significance of differences between the two agro-climatic locations, Chennai (Tamil Nadu) and Jabalpur (Madhya Pradesh). Mean values were compared using Student's *t*-test to assess location-wise differences at a significance level of  $p < 0.05$ . Prior to analysis, data were tested for normality and homogeneity of variance to ensure compliance with test assumptions.

In addition, multiple regression analysis was performed separately for each location to identify traits significantly influencing root yield. Statistical analyses were carried out using standard statistical software, and results were considered significant at  $p < 0.05$ .

## 3. RESULTS AND DISCUSSION

### 3.1. Physiological Variation and Trait–Yield Relationships in Chennai

*Withania somnifera* reached 50% flowering at 105.63 days and exhibited a mean plant height of 80.30 cm under Chennai conditions (Table 1). The plants reached maturity in 216.85 and yielded 43.15 fruit per plant. The mean seed yield was 13.54 g, with 250.75 seeds. The root length averaged 16.72 cm, while the root area exhibited exceptionally high

variability (159.03 cm<sup>2</sup>). The fresh root yield was 8.58 g. Root yield was significantly influenced by root area. The accumulation of root biomass is directly supported by the larger root systems, which likely enhance nutrient and water absorption. Additionally, root yield was substantially influenced by plant height (which may be attributed to its correlation with the overall photosynthetic capacity and assimilate availability for root growth). The number of days to maturity approached significance (indicating that certain genotypes may be able to develop roots more effectively as a result of extended vegetative periods). Root yield was not significantly influenced by other variables, such as root length, leaf length, leaf area ( $F = 0.67$ ;  $p = 0.750$ ), biomass yield, and flowering time. The shoot-to-root ratio of 5.041, is lower than approximately 10.3 (Sangwan et al., 2013), suggests that the biomass is more effectively allocated to the roots, which is a beneficial characteristic for the production of medicinal roots. The significance of root architectural traits in Chennai's sandy loam and humid conditions is emphasized by this allocation pattern, in conjunction with significant root area effects.

### 3.2. Physiological Variation and Trait-yield Relationships in Jabalpur

The average height of the plants in Jabalpur was 88.53 cm, with 50% of the plants flowering at 105.58 days and reaching maturity at 213.35 days. The seed count averaged 277.70, and the seed yield exhibited considerable variability (135.74 g). The mean berry production per plant was 43. The

fresh root yield was 10.573 g, and the root area averaged 35.47 cm<sup>2</sup>. The root length was 18.62 cm. The sole significant predictor of root yield was leaf length. This is likely due to the fact that larger leaves facilitate photosynthetic assimilation, which in turn allows for a more concentrated allocation of carbon to root development. Although not statistically significant, the F-values of root length and days to maturity were high, indicating that they may be of significance with a larger sample size. No significant impact was observed on root yield by other characteristics, including plant height, root area, or biomass yield.

The shoot-to-root ratio (4.761) was once again lower than the literature reports (~10.3), which corroborates the observation of a favourable resource allocation toward root biomass in both locations. Unlike Chennai, where root architectural characteristics were the most influential, Jabalpur's results indicate that leaf morphology is the primary factor influencing root productivity. This may be a reflection of the subtropical climate and soil nutrient profile, which encourage photosynthetically driven growth.

Breeding implications and comparative interpretation - A combination of potentially superior genotypes and favourable environmental conditions was suggested by the fact that both locations recorded greater plant height (80.30–88.53 cm) and fresh root yields (8.58–10.57 g) than previous studies (Sangwan et al., 2013: 34.08 cm height, 4.64 g root yield). Nevertheless, the determinants of root

**Table 1.** Physiological variation observed at Chennai (Tamil Nadu) and Jabalpur (Madhya Pradesh) conditions

Physiological character	Mean $\pm$ Std. deviation observed at Chennai	Mean $\pm$ Std. deviation observed at Jabalpur (Madhya Pradesh)
Plant height (cm)	80.30 $\pm$ 10.47	88.534 $\pm$ 11.71
50% flowering	105.63 $\pm$ 4.64	105.58 $\pm$ 7.097
Days of maturity	216.85 $\pm$ 4.66	213.35 $\pm$ 5.575
No. of berries/plant	43.15 $\pm$ 10.04	43.00 $\pm$ 11.16
No. of seeds / plant	250.75 $\pm$ 113.75	277.70 $\pm$ 71.82
Seeds yield per plant (in gm)	13.54 $\pm$ 4.078	135.74 $\pm$ 522.15
Root area	159.03 $\pm$ 557.44	35.47 $\pm$ 8.45
root length	16.72 $\pm$ 3.87	18.62 $\pm$ 5.26
Fresh root yield/ plant	8.58 $\pm$ 1.70	10.573 $\pm$ 2.13
Leaves length	7.78 $\pm$ 0.83	42.889 $\pm$ 153.964
Leaves area	12.45 $\pm$ 3.18	13.135 $\pm$ 2.530
Biomass yield at maturity	67.72 $\pm$ 16.38	81.980 $\pm$ 22.96
Shoot root ratio	5.041 $\pm$ 1.29	4.761 $\pm$ 1.269
Root shoot ratio	0.210 $\pm$ 0.0511	.223 $\pm$ 0.55509

productivity varied significantly between Chennai and Jabalpur. In Chennai, root area and plant height were significant factors, while in Jabalpur, leaf length was a more significant factor. The significance of location-specific reproductive targets is underscored by these distinctions. To optimize root-yield enhancement programs, Chennai genotypes should be chosen for their expanded root systems and optimal shoot growth, while Jabalpur genotypes should be optimized for their larger, more photosynthetically efficient leaves. Stable flowering periods across sites suggest genetic uniformity, which facilitates synchronized seed production and harvesting.

#### 4. CONCLUSION

This study demonstrates that the determinants of root yield in *Withania somnifera* vary distinctly across environments. Root architectural traits were the primary contributors to yield performance under Chennai conditions, whereas leaf morphological traits were more influential in Jabalpur. Both locations supported favourable biomass allocation patterns, indicating strong genetic potential for improvement. These findings highlight the importance of site-specific selection strategies in breeding programmes, emphasizing root-related traits for Chennai and leaf-related traits for Jabalpur to enhance productivity and stress resilience.

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#### Credit authorship Contribution Statement

Neha Singh: Conceptualization, Methodology, Investigation, Data curation, Formal analysis, Writing – original draft. Nisha Singh: Data curation, review & editing. Anita R. J. Singh: Supervision, Validation, Writing – review & editing.

#### Conflict of Interest

The authors declare no conflict of interest.

#### Funding Declaration

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