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Variation in Total Phenolics Content in Elite Germplasms of Indian Ginseng *Withania somnifera* (Ashwagandha)

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Abstract: Total phenolics content in leaves, stem and root for 64 pure lines of *Withania somnifera* were estimated. The phenol content in its root ranged from 0.09 - 0.69 percent with a mean value of 0.43±0.10, while in stem it ranged from 0.36 - 1.02 percent with mean value of 0.62±0.14. In leaves, the phenolic content ranged from 0.75 - 2.42 percent with a mean value of 1.72±0.41. Order of TPC was leaf > stem > root. Outcome of the present study could be useful for selection of germplasms for development of cultivar with high phenolics content.
Keywords: Solanaceae, Withania, Phenolics, Germplasm.

1 Introduction

Withania somnifera (Ashwagandha) belongs to Solanaceae plant family and finds a reputed position in many traditional systems of medicine. The Solanaceae plant family is important in economic, agricultural and pharmaceutical terms. It includes about 96 genera and 300 to 400 species [1]. *Withania* (Solanaceae), is a small shrub distributed in east of the Mediterranean region, extending to South Asia [2]. The twenty three known *Withania* species are widely distributed in drier parts of tropical and subtropical zones [3]. Among them *W. somnifera* (L.) Dunal and *W. coagulans* (L.) Dunal are economically significant and widely cultivated [4].

W. somnifera (commonly known as Ashwagandha), is an important medicinal plant of India. It is being used in Indian Systems of Medicine for over 3,000 years. It is used as herbal medicine in various forms (decoction, infusions, ointment, powder and syrup) in different parts of the world [5,7]. Roots, leaves and preparations thereof are traditionally used in more than 100 formulations of Ayurveda, Unani and Siddha. The ethno-pharmacological properties of this plant include adaptogenic, hypnotic, sedative and diuretic [8,9]. Singh et al. [10] compared therapeutic value of roots of *W. somnifera* with *Panax ginseng* and found them with similar in many respects. The effects of different dietary phenols have been confirmed in decreasing the risk of cardiovascular diseases in human intervention studies. Also, other effects of phenolics in improving gut inflammation, obesity, diabetes, hypertension and metabolic syndrome were also established [11].

Hypogycaemic and hypolipidaemic effects of *W. somnifera* root and leaf extracts on alloxan –induced diabetic rats was reported [12]. Also, determination of phenolic compounds in the extracts of *W. somnifera* root and leaf was reported. These extracts were orally administered daily to diabetic rats. Diabetic rats showed a significant elevation in glucose and thiobarbituric acid reactive substances and a significant reduction in glycogen, vitamin C and E, superoxide dismutase, catlase, glutathione peroxidase, glutathione-S-transferase levels as compared to normal control rats. Administration of above mentioned two extracts of *W. somnifera* to diabetic rats restored the levels to normal. The author suggested that the presence of phenolic compounds in extracts and their antioxidant activity could have played a vital role in reduction of blood glucose level in alloxan-induced diabetic rats [13].

A literature search revealed that studies pertaining to concentration of total phenolics in *W. somnifera* tissue is lacking. Keeping in view of medicinal importance of phenolics, the present study was carried out for estimation of total phenolics content (TPC) in leaves, stems and roots of *W. somnifera* elite germplasms.

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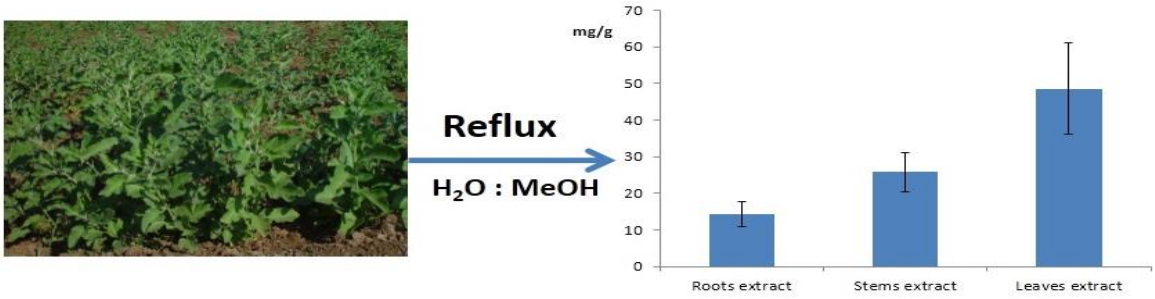

Figure 1.total phenolics content (mean) in different plant parts of *W. somnifera* elite germplasms

2 Experimental

2.1 Plant Material

W. Somnifera crop was grown in the experimental fields of the ICAR-Directorate of Medicinal and Aromatic Plants Research (DMAPR), Boriavi, Anand, Gujarat, India under open conditions. The cultivation was done by following standard package of practices. The experimental plot represented the subtropical area of India (22.556°N, 72.951°E) at an altitude of 40.63 m above mean sea level with average rainfall of 800 mm. The minimum and maximum temperature ranged between 12.7 -42ºC. Elite lines of *W. somnifera* (DWS-55 to DWS-123 excluding DWS-60, 69, 109, 112, 122) (64×5 Plants) were collected from the research farm (*W. Somnifera* breeding block) of the Directorate of Medicinal and Aromatic Plants Research, Boriavi, Anand, Gujarat, India at flowering stage of the crop in month of January 2015. The roots, stems and leaves were separated from each plant and kept for air drying in shade and in a hot air oven.

2.2 Preparation of Extracts

The air dried plant materials (root, stem and leaves) were finely powdered using an electric grinder. For conventional extraction (cold percolation), different weight (in gm.) of powdered plant material was mixed with 1:15 ratio of 10% methanol in a flat bottom flask and kept about 24 hours at room temperature. Liquid extracts obtained were separated from the solid residue by vacuum filtration and concentrated using a rotary evaporator. The same process was repeated for three times.

2.3 Determination of Total Phenolics in Extracts

Total phenolics content in the extracts were determined spectrophotometrically by Folin–Ciocalteau method. Dried extracts were reconstituted in distilled water (1 mg/mL). Folin–Ciocalteau reagent (0.5 mL) was added to the extract solution (0.5 mL) and the total volume was adjusted to 8.5 mL with distilled water. The tubes were kept at room temperature for 10 min and thereafter 1.5 mL of sodium carbonate (20%) was added. The tubes were incubated in a water bath at 40º C for 20 min. The intensity of the blue colour developed was measured by recording the absorbance at 755 nm using a UV–visible spectrophotometer. The reagent blank was also prepared using distilled water (without plant extract). For quantification of the total phenolics in the extract, a standard calibration curve was prepared using gallic acid. Total phenolics content of the extract samples was expressed as gallic acid equivalent (GAE) milligrams per gram of the extract [14].

3 Results and Discussion

W. somnifera is widely used as a traditional remedy for several illnesses in many countries and finds use in more than 200 formulations in Ayurveda, Siddha and Unani systems of medicine. Also, *W. somnifera* is becoming a popular adaptogenic in the western world. All parts of this plant are used medicinally. Practically, phenolic compounds are synthesized and accumulated in all plant tissues. There are very few studies about the TPC of *W. somnifera*. The extract yield (%) and total phenolics content (%) in the roots, stems and leaves of *W. somnifera* germplasms are described in table 1. The extract yield for roots, stems and leaves was in the range of 10.00 – 41.50, 16.50 – 32.50 and 26.00 – 50.00 percent, respectively. TPC in extracts of roots, stems and leaves was in the range of 9.03-21.83, 16.29-39.16 and 19.83-73.89 mg/g, respectively. TPC in plant parts of roots, stems and leaves was in the range of 0.09-0.69, 0.36-1.02 and 0.75-2.42 percent, respectively.

Table 1. Extract yield and total phenolics content in different plant parts of *W. somnifera* elite germplasms

Plant parts	Extract yield (%)		TPC in extract (mg/g)		TPC in plant (%)	
	Range	Mean±SD	Range	Mean±SD	Range	Mean±SD
Root	10.00 – 41.50	30.61±5.53	9.03-21.83	14.37±3.32	0.09-0.69	0.43±0.10
Stem	16.50 – 32.50	24.22±3.20	16.29-39.16	25.79±5.43	0.36-1.02	0.62±0.14
leaves	26.00 – 50.00	35.83±4.12	19.83-73.89	48.60±12.51	0.75-2.42	1.72±0.41

Udayakumar et al. (2010) reported TPC in *W. somnifera* root and leaves extract as 28.26±1.2 and 17.32 ± 0.9 mg/g, respectively. Fernanando et al. [15] reported phenolic contents of different parts of *W. somnifera* from three different growth stages. Order of TPC was leaf>flower>fruit>stem>root. Karakoti et al. [16] reported TPC in callus culture of two varieties of *W. somnifera*. namely Poshita (325 mg/kg) and Jawahar 20 (340 mg/kg).

4 Conclusions

TPC showed wide variation in roots, stems and leaves of *W. somnifera*. Based on the present study selected elite germplasms could be utilized in breeding programme for high phenolic content genotype. Further, as these parts are renewable in nature, more advance profiling of individual phenolicsare under progress.

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