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Quality Assessment of *Withania somnifera* (L.) Dunal by Analyzing Its Functionalities and Elemental Compositions

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ABSTRACT

Withania somnifera (L.) Dunal is one of the indigenous medicinal plant in India as well as worldwide. Since it contains number of health-promoting chemical compounds includes withaferin A and variety of steroids, it is commonly used in the ayurvedic system for the treatment of various disorders. Therefore several mono- and poly-herbal products of *W. somnifera* are commercially available in the Indian market. Hence there is an emergent necessity to analyze the quality retain of commercial products from its genuine real samples. In this present work the commercial sample *W. somnifera* available from southern part of India and genuine real samples available in the Western-Ghats South-East region are tested for its chemical constituents qualitatively as well as quantitatively. The analysis FT-IR, UV-Vis and SEM-EDAX has been adopted to study the functional presence and elemental presence of *W. somnifera*. The results revealed minor variations in the content of both *W. somnifera* samples.

1. Introduction

Ashwagandha, "*Withania somnifera*" (L.) Dunal (Solanaceae) commonly known as winter cherry, is one of the most valued medicinal plants in Ayurveda and other traditional systems of medicine. In Ayurveda, *W. somnifera* is regarded as one of the most useful herbs having 'Vata' pacifying properties [1–3]. Plant is widely found in India, Sri Lanka, Iraq, Iran, Syria, Turkey and Africa, as well as in North America [4]. It is act as an herbal tonic, health food in *Vedas* and commonly used in Indian traditional health care systems. The leaves are bitter and used in painful swallowing, anti-inflammatory medicines and opthalmitis [5]. Some people also used paste of roots and leaves to cure ulcer [6].The roots of the plant are categorized as rasayanas, which are reputed to promote health and longevity, arresting the ageing process, increasing the capability of the individual to resist adverse environmental factors [7], promote vitality during recovery from chronic diseases [8] and also used to control the pain during arthritic conditions [9]. The leaves are applied locally to tumors and to tuberculous glands. A fomentation of the leaves is used to cure sore eyes, ulcers, and swellings. They are also used as a hypnotic and an anthelmintic [10]. The plant has been reportedto have adaptogenic, anticancer, anti-convulsant, immunomodulatory, antioxidative and neurological effects [11, 12]. It is also considered efficacious in the treatment of arthritis, geriatric, behavioural and stress-related problems.

Withaferin A, chemically characterized as 4*b*, 27-dihydroxy-5*b*-6*b*-epoxy-1-oxowitha-2, 24-dienolide, is one of the main withanolidal active principles isolated from the plant (Fig. 1). Withaferin A inhibits cyclooxygenase-2(COX-2) but not cyclooxygenase-1 (COX-1) [13], desired flora non-ulcerating anti-inflammatory/chemotherapeutic drug. Withaferin A has also been reported to have immune suppressive action on B-lymphocyte proliferation [14]. Other withanolides, including glycosylated ones present in medicinalplants are reported to have antioxidant, immunomodulatory and other activities [15–17]. Some withanolides are known to have quinine reductase induction-mediated protective activity against chemical carcinogenesis [18]. A variety of mono- and poly-herbal preparations are commercially sold in India.

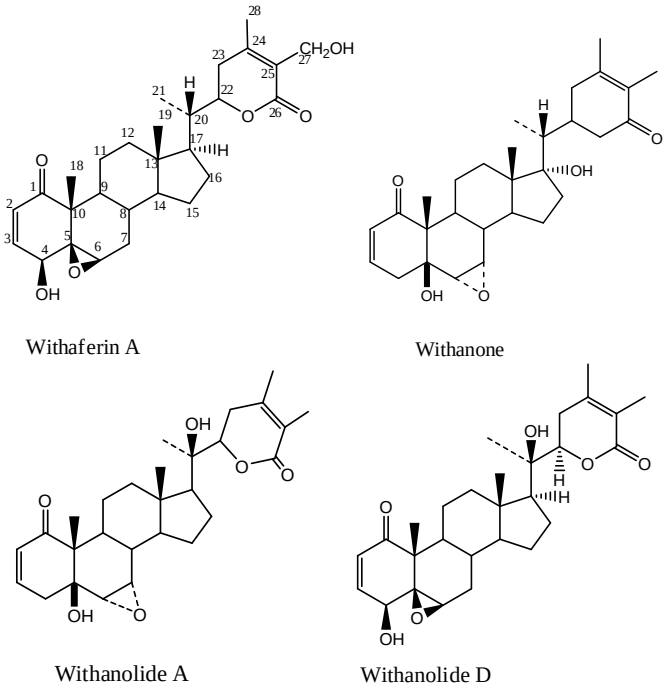

Fig. 1 Some withanolides of Ashwagandha (*Withania somnifera*)

The present study was, therefore, undertaken to make an assessment of the quality as well as the element quantity of genuine and commercial samples of *W. somnifera* using the spectral investigations.

2. Experimental Methods

2.1 Collection of Plant Materials

The plant chosen for this present study namely *W. somnifera* (Solanaceae) was collected from Salem district, Tamil Nadu, India in July 2010 and authenticated by Dr. V. Chelladurai, Research Officer (Botany),

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Retired, Survey of Medicinal and Aromatic plants Unit- Siddha, CCRAS, Palayamkottai, Tirunelveli District, TN, India. The root was shade dried for obtaining a constant dry weight. Then the dried root was powdered and noted as genuine sample (GS). The dried roots of *W. somnifera* (Solanaceae) were procured from the local market, powdered and noted as market sample (MS). Both samples were passed through sieve of size 53 µm, stored in a closed vessel individually.

2.2 Extraction

About 5 grams of the root powder of the plant samples were extracted with 95% methanol (1:10 w/v) for 48 hours with constant stirring individually. Suspensions were filtered through Whatman No. 1 filter paper to retain the clear solution. The pooled extracts were vacuum evaporated below 50 °C, the residues were dissolved in the same solvent and were stored at 4 °C separately.

2.3 Instrumentation

The IR measurements were recorded for methanolic extract of the plant samples in the transmittance mode Range: 4000 to 400 cm^{-1} by using Perkin-Elmer-Spectrum RXI FT-IR (RXI FT-IR) instrument individually. Ultra Violet-visible spectroscopy analyses were carried out by UV–visible spectrophotometer JASCO V-530 in the range of 200 nm – 1100 nm, with the scanning speed of 400 nm/min for both samples. The morphology examination of dried powder samples were analyzed with Scanning Electron Microscope (SEM) HITACHI-S-3400N model fitted with an energy dispersive X-ray analyzer (EDAX) allows a qualitative detection and localization of elements in the samples. The SEM enables a direct observation of the surface microstructures of the plant samples.

3. Results and Discussion

The obtained FT-IR spectra for both GS and MS *W. somnifera* are shown in Fig. 2 and Fig. 3. From Fig. 2 the FT-IR spectrum of GS *W. somnifera* is having very strong peaks at 3328 cm^{-1} and 1022 cm^{-1}. Also it shows the strong peaks at the frequencies 2944 cm^{-1}, 2832 cm^{-1}, 1448 cm^{-1}, 1115 cm^{-1} and 623 cm^{-1}. The peaks at 3328 cm^{-1} and 623 cm^{-1} that the peaks cover the entire region with a very broad peak. The peak at 3328 cm^{-1} is corresponds to hydrogen bonded O-H stretching frequency. The peak at 2944 cm^{-1} is assigned to H-C-H stretch. The strong peak at 1022 cm^{-1} responsible for C-CO-C stretching frequency and 1115 cm^{-1} responsible for C=O functional groups. The stretching frequency for C=O has obtained at 1737 cm^{-1} and the C-H stretching frequency obtained at 1448 cm^{-1} response the plane asymmetric stretch. The parallel and perpendicular stretching frequencies are obtained at 2944 cm^{-1} and 2832 cm^{-1}. The similar observations were obtained for MS *W. somnifera* (Fig. 3), i.e., the O-H stretching frequencies at the broaden area at 3330 cm^{-1}; the C-H stretching frequencies at 2943 cm^{-1}, 2831 cm^{-1} and 1448 cm^{-1}; the C-CO-C and C=O stretching frequencies at 1022 cm^{-1} and 1112 cm^{-1} respectively. The obtained results from both figures ensured the presence of withanolides in studied sample.

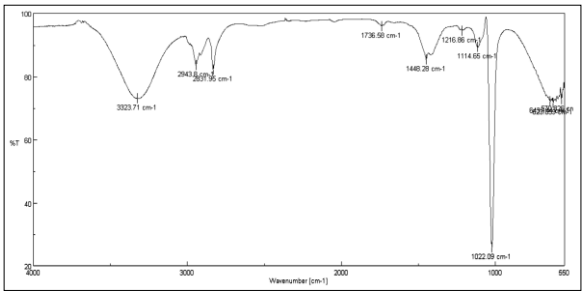


Fig. 2 FT-IR spectrum of GS *W. somnifera* genuine sample

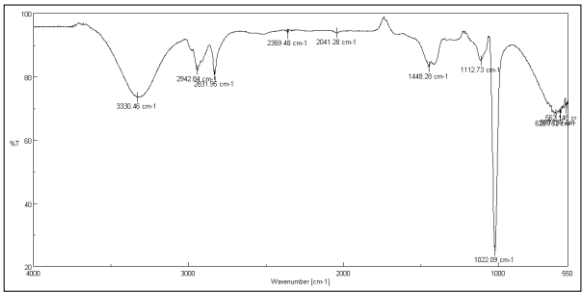


Fig. 3 FT-IR spectrum of MS *W. somnifera* commercial sample

The UV-Vis spectra of both GS and MS *W. somnifera* are shown in Fig. 4. The maximum absorptions obtained at the λ_{max} value of 218 nm (n-π*, π-π*), 283 nm (n-π*, π-π*) and 322 nm (n-π*) for GS and the λ_{max} value of 219 nm (n-π*, π-π*) and 279 nm (n-π*, π-π*) for MS of *W. sominfera*. The results indicates that the presence of C=C, C=O and C-O functional grouped compounds in the samples. These results are correlated with our FT-IR spectral reports. The quality of the *W. somnifera* herbal is retaining its own medicinal properties in both samples.

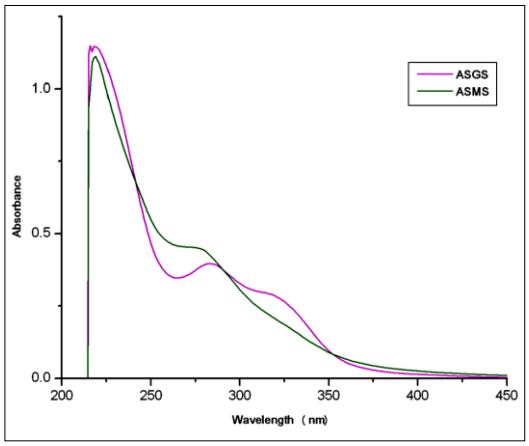
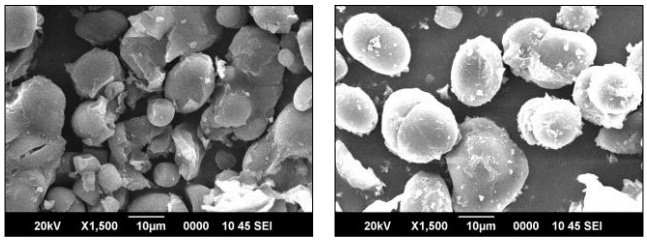
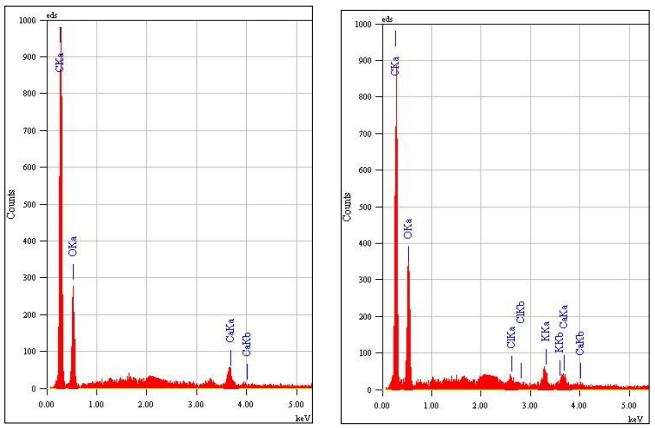


Fig. 4 UV spectrum of *W. somnifera* samples



(a) (b)

Fig. 5 SEM images of *W. somnifera* samples (a) GS and (b) MS



(a) (b)

Fig. 6 EDAX spectrum of *W. somnifera* samples (a) GS and (b) MS

The SEM-EDAX spectra obtained for the study samples are given in Fig. 5 and Fig. 6. From SEM figures Fig. 5a and Fig. 5b, by morphologically the both samples look similar to other while the particle size showing very minor variations. As it can be seen from Fig. 6a, the GS shows the presence of various elements such as C, O and Ca in which C is the highest percentage 90.41% followed by O as 8.1% and with small quantities of Ca (1.48%). But in the Fig. 6b for the MS, in addition to C (83.5%), O (12.2%) and Ca (1.32%) it contains K (1.32%) and Cl (0.83%) also. As a report, the SEM-EDAX analysis shows minor variations between the both.

4. Conclusion

The GS and MS of *W. somnifera* have been employed for their quality and elemental quantity analysis. The results clearly imply that the both samples are retaining its organic compositional values and correspondingly its medicinal values obviously. There is a small variations in their elemental compositions. These variations can originate from several factors that include: (i) diversified bioresources of heterogeneous nature from the wild and/or under cultivation, (ii) physiological and ecological variations in plantations, (iii) harvest and post-harvest operations, (iv)processing of biomass, (v) manufacture process for product, (vi) unregulated and often non-descript supplementations, etc.

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