



Study of *Withania somnifera* with the spatial reference of phytochemical, FTIR and flavonoids quantification

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Abstract

Plant is oldest existing complete medical significance in the world. Its origins go back nearly 5000 years. In this research article make known on *Withania somnifera* (Ashwagandha) as Medicinal plants have therapeutic potential due to the presence of natural antioxidants functioning as reducing agents, free radical scavengers and quenchers of singlet oxygen. Majority of different medicinal plant and their antioxidant activity is due to bioactive compounds viz. flavones, isoflavones, flavonoids, anthocyanins, coumarins, lignans, catechins and isocatechins. Extraction of the bioactive plant constituents has always been a challenging task for the researchers. And this article also provides an overview of extraction, photochemical analysis, Flavonoide quantification and FTIR analysis of *Withania somnifera* (Ashwagandha).

Key-Words: Medicinal Plants, *Withania somnifera*, Flavonoids

Introduction

It is evident that anti-oxidants play an important role in the prevention of cancer, cardiovascular and neurogenerative diseases. There has been a surge of research in its effect in animal models of atherosclerosis, hyperlipidemia, myocardial infarction, myocardial ischemia reperfusion injury, cerebral ischemia, cardiomyopathy, cardiac hypertrophy, cardiotoxicity and congestive heart failure. 13 different constituents of WS for their anti proliferative capabilities on lung, colon, central nervous system (CNS), and breast tumor lines (Jayaprakasam B et al 2003). Many pharmacological studies have been conducted to investigate the properties of ashwagandha and to authenticate its use as a multi-purpose medicinal agent. Studies on *Withania somnifera* suggests that it reduces tumor cell proliferation and enhances the effectiveness of radiation therapy while potentially mitigating undesirable side effects (Kaur et al., 2004). *Withania somnifera* Dunal (WS), commonly known as ashwagandha, has been used for centuries in Ayurvedic medicine to increase longevity and vitality (Altern Med Rev 2004). WS acts as an anti-inflammatory agent through inhibition of complement, lymphocyte proliferation, and delayed-type hypersensitivity (Rasool M et al 2006).

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Scientific classification

Kingdom:	Plantae
Order:	Solanales
Family:	Solanaceae
Genus:	<i>Withania</i>
Species:	<i>somnifera</i>



Fig.1: *Withania somnifera*

Material and methods

Plant material collection

Plant *withania somnifera* were collected from sanjivani ayurvedic nursery Bhopal during October month. Then dried up under the shed dry for three week furthermore crush it.

Soxhlet extraction: acetone water (1:1)

Soxhlet extraction is only required where the desired compound has a limited solubility in a solvent, and the impurity is insoluble in that solvent. If the desired compound has a high solubility in a solvent then a simple filtration can be used to separate the compound from the insoluble substance. The advantage of this system is that instead of many portions of warm solvent being passed through the sample, just one batch of solvent is recycled. This method cannot be used for thermo labile compounds as prolonged heating may lead to degradation of compounds (Nikhil SB, et al 2010).

Phytochemical analysis

Phytochemical analysis is a major procedure for estimation of particular chemical compound so that following soxhlet extraction crude drug evaporated under steam bath tank. And Phytochemical analysis was proceeded by the help of different specific method for each test. All phytochemical analysis (test) has been done by the using Kokate (Practical Pharmacognosy) as a reference book.

Total flavonid estimation

Total flavonoid contents were measured with the aluminum chloride colorimetric assay. Hydro-alcoholic extracts that has been adjusted to come under the linearity range and different dilution of standard solution of Quercetin (10-100 μ g/ml) were added to 3ml of water. To the above mixture, 0.1ml of 5% C₄H₄O₆KNa₄H₂O was added. After 5 minutes, 0.1ml of 10% AlCl₃ was added and the total volume was made up to 3 ml with distill water. It was left at room temperature for 30 min after which the absorbance of the reaction mixture was measured at 430 nm with a single beam Visible spectrophotometer, Systronic (106).

Results and Discussion

The FT-IR analysis of the samples was done and the functional groups associated were determined (Fig.2). The FT-IR spectrum of the sample was obtained effective peaks. the FT-IR spectrum of the *Withania somnifera* samples recorded the number of peaks lying between 3320.29 cm⁻¹, 2945.67 cm⁻¹, 2834.64 cm⁻¹, 1652.83 cm⁻¹, 1449.39cm⁻¹, 1417.14 cm⁻¹, 1113.62cm⁻¹, 1016.45cm⁻¹, 755.15 cm⁻¹, 575.61 cm⁻¹, 546.14 cm⁻¹, 534.78 cm⁻¹, 510.12 cm⁻¹ respectively. This finding helps in further research in the investigation of other medicinal plant with different solvent fraction for their antioxidant activity and it also useful to utilize of *Withania somnifera* plant as a source medicine.

Current study revealed the flavonoid scontents of the leaves, of *withania somnifera*. (Quercetin standard

plot: $y = 0.0966x$, $R^2=0.9878$) (Elija Khatiwora et al 2010). On the basis that calibration curve was plotted by preparing the Quercetin solutions at concentrations 12.5 mg/ml-1. Total flavonoid content of the extracts was expressed as percentage of flavonoid in plant extract 4.78 equivalents per dry weight of sample and take notice of (Fig.3) $y=0.002x+0.004$, $R^2=0.999$.

Current era herbal products are measured to be symbols of safety in comparison to the synthetic product that are regarded as hazardous to human life and environment. Although herbs had been priced for their therapeutic importance. But now everyday phytochemical and pharmacological studies are conducted on different parts of these plants. The present literature supports the potential of certain medicinal plants. More research can be done to investigate the unknown and unexplored potential of these plants. Further structural analysis of *Withania somnifera* plant (flavonoid compounds) can be carried out by way of making use of different analytical methods such as NMR and Mass spectrophotometer analysis.

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References

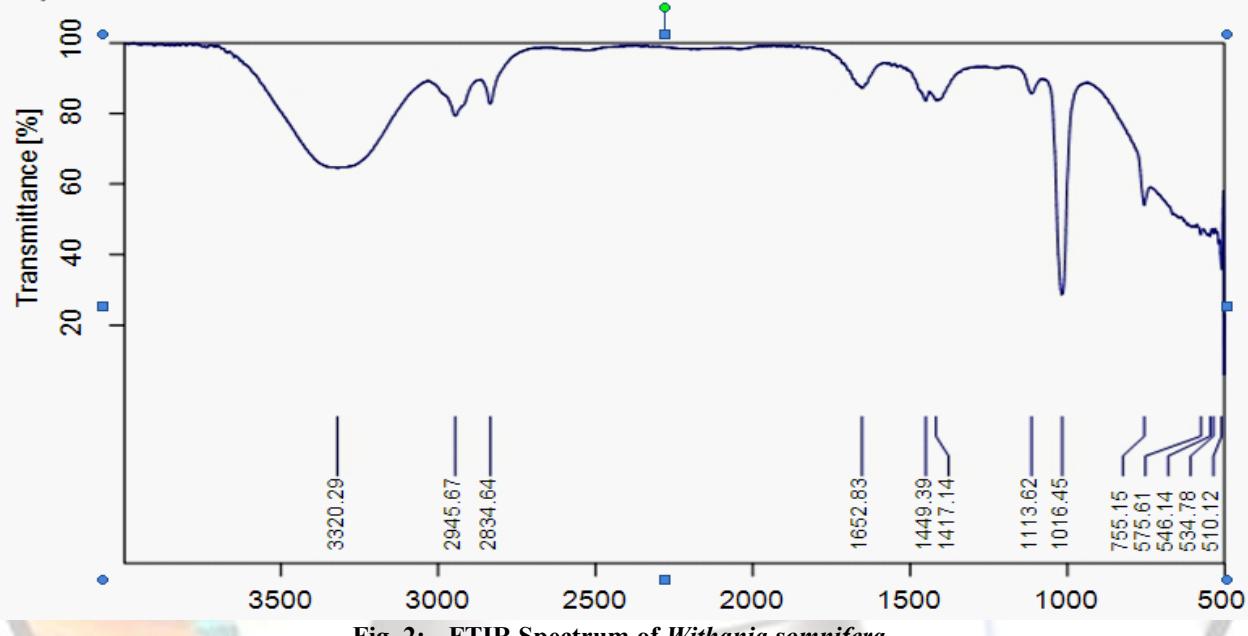
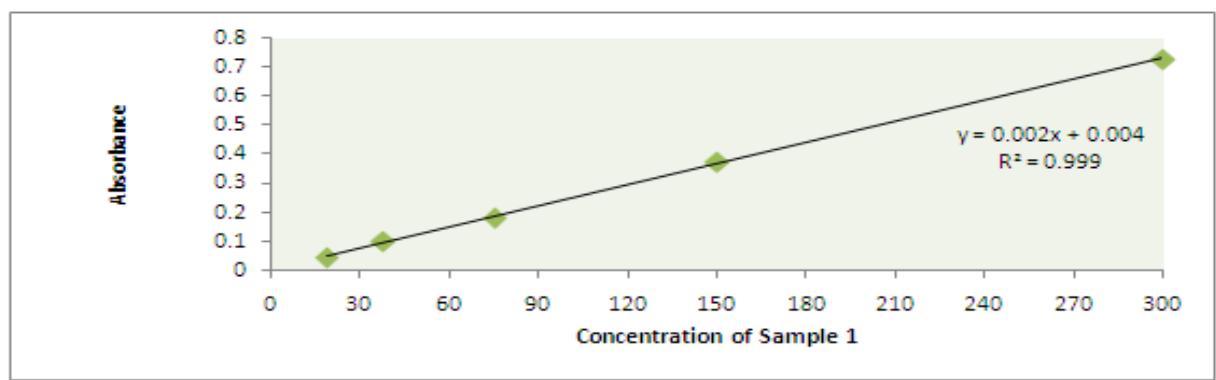
1. Elija Khatiwora, Vaishali B. Adsul, Manik M. Kulkarni, N.R. Deshpande and R.V Kashalkar., (2010). Spectroscopic determination of total phenol and flavonoid contents of Ipomoea carnea; International Journal of Chem Tech Research Vol.2, No.3, pp 1698-1701.
2. Jayaprakasam B, Zhang Y, Seeram NP, Nair MG., (2003). Growth inhibition of human tumor cell lines by withanolides from *Withania somnifera* leaves. *Life Sci* ;74: 125-132.
3. Kaur K, Rani G, Widodo N, et al., (2004).Evaluation of the anti-proliferative and anti-oxidative activities of leaf extract from *in vivo* and *in vitro* raised ashwagandha. *Food Chem Toxicol* ;42:2015- 2020.
4. Nikhal SB, Dambe PA, Ghongade DB, Goupale DC., (2010) Hydroalcoholic extraction of *Mangifera indica* (leaves) by Soxhletion. International Journal of Pharmaceutical Sciences ; 2 (1): 30-32.
5. No authors listed., (2004). *Withania somnifera* monograph. *Altern Med Rev* ;9:211-214.
6. Rasool M, Varalakshmi P., (2006). Immunomodulatory role of *Withania somnifera* root powder on experimental induced

inflammation: an *in vivo* and *in vitro* study. *Vascul Pharmacol*, 44:406-410.

7. C.K.. Kokate., (2008). Pharmacognosy; Published Nirali Prakashan ISBN 8185790094 (ISBN13: 9788185790091).

Table 1: Phytochemical nature *W. somnifera*

S/No.	Natural product	Test performed	Result
1	Carbohydrate	Molish test	+ve
2	Starch	Iodine	-ve
4	Protein	Millions	+ve
5	Amino acid	Cysteine test	+ve
6	Steroid	Salkowski test	+ve
7	Flavonoids	Ferric chloride test	+ve
8	Alkaloid	Mayer's test	+ve
9	Tannic andphenolic compound	%5FeCl ₃ test	+ve
10	Oxalic acid	Test	+ve
11.	Inorganic acid	Sulphate test	+ve

Fig. 2: FTIR Spectrum of *Withania somnifera*Fig. 3: Flavonoid quantification curve of *Withania somnifera*