

TABLE 1: ANALYSIS OF LINEZOLID IN TABLET FORMULATION

Method	Batch	Label Claim (mg/tab)	% of Label Claim Estimated*	S.D	% Recovery**
I (BCG)	A	600	98.71	0.682	101.29
	B	600	98.94	0.564	
	C	600	99.02	0.714	

BCG stands for bromocresol Green. \*Denotes average of three determinations. \*\*Denotes average of recovery studies at three different concentration levels. The tablets used were Linox, (600 mg) Unichem Ltd.

The optical characters such as Beer's law limit 0-70 µg/ml, molar absorptivity  $1.3 \times 10^4$  l/mol/cm, Sandell's sensitivity  $0.0227 \mu\text{g}/\text{cm}^2/0.001$  absorbance unit, slope 0.0084, intercept 9.9991 correlation coefficient is 1.02 and relative standard deviation is 0.3059. The colour of the complex formed has been found to be stable for 8 h. The drug dye ratio is 1:1.

#### ACKNOWLEDGEMENTS

The authors are grateful to Dr. Nalla. G. Palanisamy (Chairman), Dr. Thavamani. D. Palanisamy (Trustee), Prof.

Abhay Dharamsi (Principal) K.M.C.H. College of Pharmacy, Coimbatore for providing the necessary facilities.

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## HPLC Analysis of Withaferin A in *Withania somnifera* (L.) Dunal

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Accepted 8 December 2003

Revised 25 September 2003

Received 7 February 2003

**A reversed phase liquid chromatographic method for analysis and quantitative estimation of withaferin A in roots of *Withania somnifera* (L.) Dunal collected from different geographical zones, has been developed using a symmetry C18 column and a binary gradient profile. The various aspects of analysis such as extraction, efficiency, detection limits, reproducibility and peak purity were validated using photodiode array detector.**

The roots of *Withania somnifera* (L.) Dunal (Family: Solanaceae) is commonly known as *Ashwagandha* have been employed in Ayurveda and Unani system of medicine as nervine tonic. This plant is reported as an antistress, antiinflammatory, antitumor and CNS depressant agent.

Withaferin A was isolated from species of *Withania*<sup>1</sup> and from *Acnistus arborescens*<sup>2-3</sup>. It is reported to be an antibiotic<sup>4</sup>, anticancer agent<sup>5</sup> and a radiosensitizer<sup>6</sup>. In present study attempts were made to develop a reliable, simple and reproducible method for extraction of withanolides and their quantitative estimation in roots of *Withania somnifera*.

Withaferin A was purchased from Natural Remedies

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Pvt. Ltd. Bangalore, acetonitrile (Spectrochem., Mumbai) and water (Mallinckrodt Baker, Paris) used were of HPLC grade. The solvents were filtered through 0.45 µm filter before use.

The roots of *Withania somnifera* were procured from market of Kochi, Bangalore and Kanyakumari and collected from Lucknow. A small amount of dried, powdered roots of *Withania somnifera* (10 g) was extracted with 50 ml (x3) of methanol for 24 h at room temperature (25°). The extracts were combined together and concentrated under reduced pressure. The concentrated extract was fractionated with n-hexane (25 mlx3) and subsequently with Diethyl ether (25 mlx3). Residue was then treated with 0.5% H<sub>2</sub>SO<sub>4</sub> (10 ml). After filtering, residue was washed free of acid with distilled water. An aliquot of this residue (10 mg) was redissolved in 1 ml of methanol and 20 µl subjected to HPLC.

HPLC was performed on a Waters modular system consisting of two 515 pumps, a Rheodyne injector, a pump control module, a 2996 photodiode array detector and Millennium 32 chromatography manager. The injector, pump control module and Millennium 32 chromatography manager were integrated to give reproducible results. Symmetry C18 (5 µm, 150x3.9 mm, Waters) column was used for the analysis.

Solvents A water:acetonitrile (80:20 v/v), B water:acetonitrile (20:80 v/v). The linear gradient profile was initially 100% A, changing to 70% A at 12 min, 50% A at 20 min, 20% A at 35 min and then 100% A at 40 min and allowing 10 min for equilibration of column. Initial flow rate was 0.6 ml/min for 20 min and increased 1.0 ml/min till 40 min. The spectral acquisitions were done at 228 nm. Injection size for standard and sample was 20 µl each. Calibration curves for standard withaferin A was found to be linear in the range of 0.2–20 µg/ml.

The main objective was to devise a more simple, fast and reproducible method. The analytical parameters were selected after screening a number of solvent systems, gradient profile and adsorbent such as Lichrosorb SI-100, µ Porasil A, R-Sil C18 HL Lichrosorb C18 and ODS-supelcosil C18<sup>7-11</sup>. The analysis was done on symmetry C18 column with acetonitrile-water and reported gradient profile. It provides best baseline resolution for crude withanolides mixtures with reference to withaferin A (retention time t<sub>r</sub>=15.68 min. fig. 1A and B). This method can also be used for determination of other withanolides present in *W. somnifera*. All the samples were analyzed under identical conditions and it provides good baseline resolution. The

validation was further confirmed by comparing the UV spectra of peak with reference compound using PDA detector. The percentage of withaferin A present in different sample collected/ procured from different geographical zones (viz. Lucknow, Kochi, Kanyakumari and Bangalore) are 13.85, 11.64, 9.30 and 4.87, respectively (Table 1).

#### ACKNOWLEDGEMENTS

We are grateful to Dr. P. Pushpangadan, Director, NBRI, Lucknow, for providing facilities and Dr. G. C. Uniyal, Sci-III, CIMAP, Lucknow, for their valuable suggestions. Authors are also thankful to Dr. S. Khaton and Mr. S. K. Srivastava for identifying the plant material and Mr. Manoj

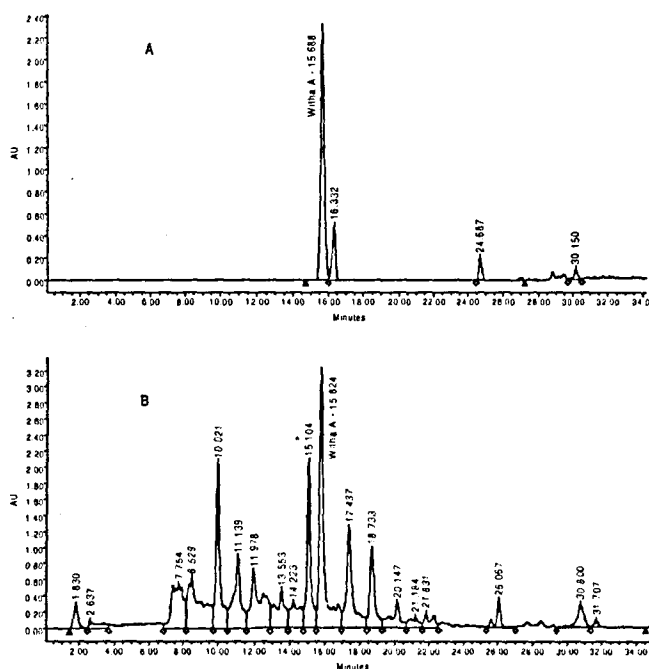


Fig. 1: (A) – HPLC profile of withaferin A; (B) – HPLC profile of crude withanolides present in roots of *Withania somnifera* under identical conditions.

TABLE 1: PERCENTAGE OF WITHAFERIN A IN DIFFERENT SAMPLES IN CRUDE WITHANOLIDES

Place	Percentage
Lucknow	13.858
Bangalore	4.874
Kanyakumari	9.302
Kochi	11.640

Dhoundiyal for typing the manuscript. One of us (SB) is grateful to CSIR, New Delhi, for financial support.

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## Mouth Dissolve Tablets of Sumatriptan Succinate

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Accepted 6 February 2004

Revised 1 October 2003

Received 23 January 2003

Mouth dissolve tablets of sumatriptan succinate were prepared using disintegrants, sodium starch glycolate, carboxy methylcellulose sodium and treated agar by direct compression method. The prepared tablets were evaluated for thickness, uniformity of weight, content uniformity, hardness, tensile strength, porosity, friability, wetting time, water absorption ratio, *in vitro* and *in vivo* disintegration time and *in vitro* drug release. The tablets disintegrate *in vitro* and *in vivo* within 10 to 16 s and 12 to 18 s, respectively. Almost 90% of drug were released from all formulations within 10 min. The formulations containing combination of sodium starch glycolate and carboxy methyl cellulose was found to give the best results. The tablets apart from fulfilling all official and other specifications, exhibited higher rate of release.

Sumatriptan succinate is a potent and selective 5-hydroxytryptamine agonist. Chemically it is 3-[2-(dimethylamino) ethyl]-N-methyl-1H-indol-5-methanesulfonamide butane-1,4-dioate<sup>1</sup>. It is an effective agent in the treatment of acute migraine attack. It provides rapid symptoms relief up to 85-90% of migraine patients within 2 h of treatment<sup>2</sup>. However, oral bioavailability is poor with only 14% of the dose reaching systemic circulation<sup>3</sup>. This is likely due to extensive pre-systemic clearance on first pass. As migraine sufferers have markedly reduced functional ability, they would be benefited from acute treatment that helps them to resume their functional

activities as quickly as possible. Rapid onset of action was demonstrated an important attribute for an acute migraine treatment.

Mouth dissolving tablets which disintegrate or dissolve in saliva and swallowed without the water<sup>4</sup>. As tablet disintegrates in mouth this could enhance the clinical effect of the drug through pre-gastric absorption from the mouth, pharynx and esophagus this leads to an increase in bioavailability by avoiding first pass liver metabolism<sup>5</sup>. Sumatriptan succinate was obtained as a gift sample from Natco Pharma Ltd. Hyderabad. Avicel PH-102 was a gift sample from Reliance cellulose, Secunderabad. Aerosil and flavor was obtained from Epic Pharmaceuticals, Satara. Sodium starch glycolate (SSG), carboxy methylcellulose

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