Current Pharma Research
ISSN-2230-7842
CODEN-CPRUE6
www.jcpronline.in/

Research Article

Quantitative Estimation of Guggulsterone E & Z in Polyherbal Tablet Formulation by HPLC.

Pratap B. Pawar*[1], Sunil A. Nirmal[2]
[1]Department of Pharmacognosy, SGSS'S Loknete Dr. J. D. Pawar College of Pharmacy, Manur, Kalwan (423501), Maharashtra, India,
[2]Department of Pharmacognosy, Pravara Rural College of Pharmacy, Pravaranagar, Loni (413736), Maharashtra, India.

Received Apr 27, 2016; received in revised form June 18, 2016; accepted June 20, 2016
***Corresponding author E-mail address:** pawar1pratap@rediffmail.com*

ABSTRACT
Guggulsterone is the active constituent of *Commiphora mukul* which is the main ingredients of formulation. The formulation was found to be potent for Triton X-100 induced Hyperlipidemic model of anti-hyperlipidemic activity. In the present study an attempt has been made to develop a HPLC method for quantitative estimation of Guggulsterone in anti-hyperlipidemic polyherbal tablet formulation. The HPLC separation was performed on a C18 column (250 x 4.6 mm, 5 µ) using solvent system acetonitrile: water (45: 55) at flow rate of 2 ml/min. Detection was carried out at 242 nm. The content of guggulsterone E & Z in polyherbal formulation was found to be 0.561 mg/tablet.

KEYWORDS
Guggulsterone, *Commiphora mukul*, HPLC, polyherbal formulation, anti-hyperlipidemic.

1. INTRODUCTION

Naturally occurring drugs are widely used in the traditional medicinal system for the treatment of various diseases. Standardization of natural products is a complex task due to their heterogeneous composition. To ensure reproducible quality of herbal products, proper control of starting material is essential.[1,2] Natural product quality control generally necessitates a multidisciplinary approach which requires resources and sophisticated equipments.[3,4] For identification of the crude drug authentic reference standard of that particular crude drug are required.[5,6] Guggul recognized as traditional medication for the treatment of different diseases and ailments of human beings.[8,9] Anti-hyperlipidemic polyherbal tablet formulation containing Arjunsal Ghana100 mg, Shuddha Guggul 100 mg, Lashuna Kalka 50 mg, Brahmi Ghana 50 mg, Triphala Kwatha, Draksha Swarasa, and Jatamamsi Phant.[7,8] Guggulsterone is one of the active constituent of *Commiphora mukul* known as 'Guggul'. HPLC is a valuable tool for the investigation of herbal products with respect to different aspects of their quality. HPLC analysis is comparatively fast and multiple samples can conveniently be analyzed at a time on the same column.[1,2] This is particularly important for quantitative and qualitative screening of raw materials for process control during manufacturing.[10]The objective of the present study is to develop a method for quantitative analysis of guggulsterone from a polyherbal tablet formulation using HPLC.

2. MATERIALS & METHODS

2.1. Chemicals

Guggulsterone standards were procured from Natural Remedies Pvt. Ltd., Bangalore. HPLC grade Acetonitrile was supplied by Finar chemicals Ltd., India and HPLC grade Water supplied by Merck specialties Pvt. Ltd. Prepared formulation was used for analysis.

2.2. Instrumentation

HPLC separation was carried out on Waters multi solvent delivery system using water W600 E gradient model. The mobile phase was degassed using water in line degasser AF. The detection was carried out on water 486 absorbance detector. The data was accessed using Data Ace chromatographic work station.

2.3. Experiment Formula

2.3.1. Determined % w/w and mg/tablet by given formula,

Estimation of Guggulsterone E & Z in (% w/w) and (mg/tablet):

$$(\%\ w/w) = \frac{\text{Sample Area}}{\text{Standard Area}} \times \frac{\text{Standard wt. (mg)}}{\text{Dilution (ml)}} \times \frac{\text{Sample Dilution (ml)}}{\text{Standard wt. (mg)}} \times \text{Purity} \quad \text{--- (1)}$$

$$\text{(Mg/tablet)} = \frac{\text{\% Guggulsterone X Average weight of Tablet (mg)}}{100} \quad \text{--- (2)}$$

2.4. Chromatographic Condition

Chromatographic separation was performed on Guard Column C18 (250 x 4.6 mm, 5 µ), using injection volume 20 µl, solvent system acetonitrile: water (45: 55) at flow rate of 2 ml/min at an ambient column temperature. Detection of a guggulsterone was performed at UV detector 242 nm.[1,3] Sample solution was prepared using Sample weight 406.95 mg to 50ml in Acetonitrile. Preparation of standard solution using Reference Standard weight: (Guggulsterone E & Z > 99%) 10.68 mg diluted to 100 ml in Acetonitrile.

3. RESULTS AND DISCUSSION

3.1. Optimization of HPLC chromatographic conditions

Optimum chromatographic conditions were obtained after running different mobile phase with a C18 column. Acetonitrile was preferred over methanol as mobile phase because it results into improved separation. Many different gradient systems of mobile phase were tried for the best separation of peaks. Selecting 242 nm as the detection wavelength resulted in an acceptable responses and enable the detection of compounds used in this study.[1,3] An HPLC fingerprint for the formulation was developed. Elution was carried out at a flow rate of 2 ml/min with acetonitrile as solvent. Under the chromatographic condition employed, standard compound guggulsterone and the formulation have shown sharp peaks and good separation (Figure 1 and 2).

3.2. Analysis of the Prepared Formulation

Guggulsterone extracted from polyherbal tablet formulation showed good resolution with a gradient elution (Figure 2). The content of guggulsterone E & Z was found to be 0.561mg/tablet and was within the limits. Chromatogram of sample solution showed other peaks than those of standards which might be due to the presence of other phytoconstituents present in the formulation in various concentrations.

4. REFERENCES

1. Agrawal Himani et al. (2004) HPTLC method for guggulsterone: I. Quantitative determination of E- and Z-guggulsterone in herbal extract and pharmaceutical dosage form. Journal of Pharma and Biomed Ana. 36(1): 33-41.1
2. Bhattacharya Kaviraj Niwargchandra, Bharat Bhaishajya Ratnakar Motilal Banarasidas (1988) Delhi. 1:5-7.
3. Deng Ruitang (2007) Therapeutic Effects of Guggul and Its Constituent Guggulsterone: Cardiovascular Benefits. Cardiovascular Drug Reviews. 25(4): 375.

4. Indian Herbal Pharmacopoeia, A Joint Publication of Regional Research Laboratory Council of Scientific and Industrial Research Jammu Tawi and Indian Drug Manufacturers Association, Mumbai (1999) 1 (30): 99-105.
5. Indian Herbal Pharmacopoeia, A Joint Publication of Regional Research Laboratory Council of Scientific and Industrial Research Jammu Tawi and Indian Drug Manufacturers Association, Mumbai (1999) 2: 50.
6. Kirtikar KR, Basu BD (1991) Indian Medicinal Plants, International Book Distibutors, Dehradun. 1(3): 526-528, 607-609.
7. Kokate C.K., Purohit A.P., Gokhale S.B. (2008) Pharmacognosy, Nirali Prakashan, Pune. 41: 8.57, 9.4-9.10, 11.49, 11.52, 11.134.
8. Mukherjee P.K. (2002) Quality Control of Herbal Drugs, Business Horizone, Pharmaceutical Publishers, New Delhi. 1: 545, 707, 725.
9. Nadkarni K.M. (1976) Indian Materia Medica, Bombay Popular Prakashan, Bombay. 1(3): 65, 374, 480, 840, 1198, 1202, 1205, 1287.
10. The Useful Plants of India, Council of Scientific and Industrial Research, New Delhi (1994): 28, 65, 138, 195, 392, 627, 628, 684-685.

Table 1: HPLC Chromatogram data Guggulsterone E & Z Pure & Reference Standard.

Peak	Ret. Time (min)	Area	Name	Area %	Height %
1	21.195	739800	Guggulsterone E	11.970	15.778
2	29.969	5440816	Guggulsterone Z	88.030	84.222
Total		6180616		100.00	100.00

Table 2: HPLC Chromatogram Reading of Formulation.

Peak	Ret. Time (min)	Area	Name	Area %	Height %
1	21.325	383728	Guggulsterone E	37.818	43.964
2	28.212	84091		8.288	8.788
3	30.244	355916	Guggulsterone Z	35.077	32.087
4	34.934	122630		12.086	10.100
5	36.295	68299		6.731	5.061
Total		1014663		100.00	100.00

Table 3: Estimation of Guggulsterone in Formulation by HPLC.

Sr. No.	Compound	% (w/w)	mg/ Tablet
1.	Guggulsterone E	0.084	0.294
2.	Guggulsterone Z	0.076	0.267
Total	Guggulsterone E & Z	0.160	0.561

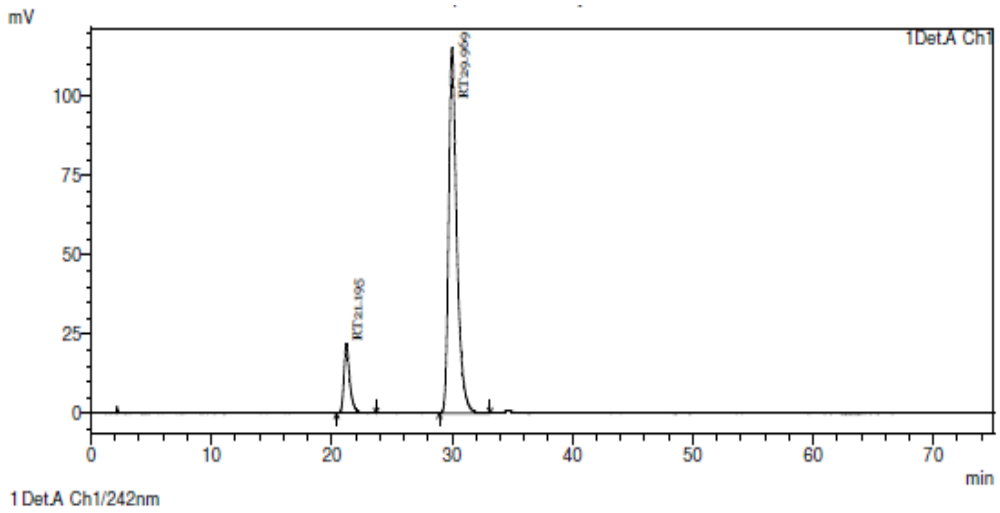

Figure 1: HPLC Chromatogram of Guggulsterone E & Z Pure & Reference Standard

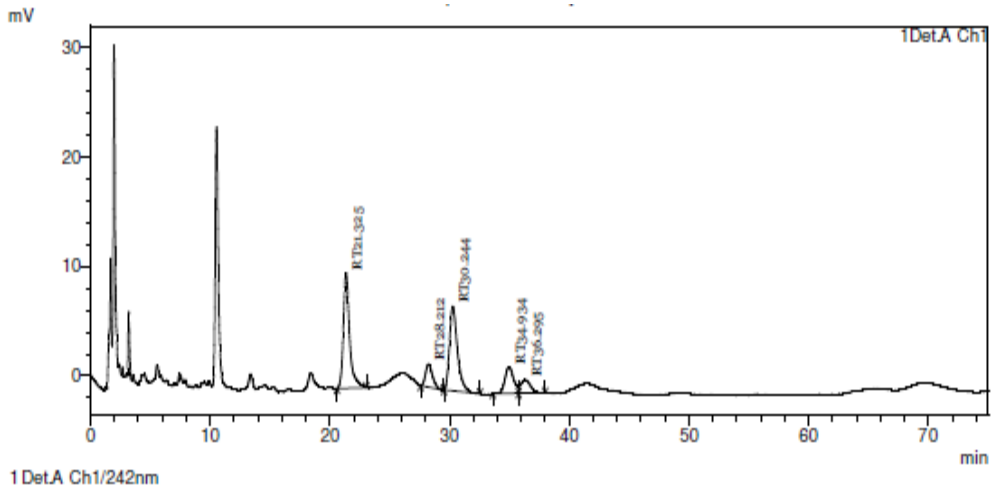

Figure 2: HPLC Chromatogram of Formulation.