Estimation of rutin and quercetin Terminalia chebula by HPLC

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Citation

Abstract
Tannins and Flavonoids present in the Terminalia chebula. Flavonoids like Rutin and quercetin possess many biochemical effects like inhibition of enzymes, regulatory role on different hormones and pharmacological activities like antimicrobial, antioxidant, anticancer, antihepatotoxic, protection of cardiovascular system. An HPLC method was developed for the estimation of rutin and quercetin from methanol methanolic extract of Terminalia chebula.

INTRODUCTION
Terminalia chebula is one of the ingredients present in the many Ayurvedic and other traditional medicine system. Terminalia chebula is traditionally used in formulation for anti-diabetic, anti-inflammatory, laxative, antibacterial, antifungal, cardiotonic, diuretic, hyperlipidemic activity, jaundice (Anonymous. 1999, Kirtikar KR and Basu BD, 1987, Inamdar et al., 1959, Sabu, M.C. 2002 Miglani, et al., 1971, Khanna, et al., 1993). Flavonoids are a group of polyphenolic compounds, which are widely distributed throughout the plant kingdom. To date about 300 varieties of flavonoids are known (Anonymous, 1996). Many have low toxicity in mammals and some of them are widely used in medicine for maintenance of capillary integrity (Kuhnau, J. 1976). Rutin, 5,7,3’, 4’, tetrahydroxy flavonol-3-rhamnoglucoside and quercetin 5,7,3’,4’,- tetrahydroxy flavonol are exhibits anti-inflammatory, antihypertotoxic (Cesarone, 1992), antiulcer (Clack et al., 1950), antiallergic, antiviral actions and some of them provides protection against cardiovascular mortality (Colergie Smith et al., 1980, Hertog et al., 1993). Both possess antioxidant activity and reduce low density lipoproteins (LDL) oxidation (De-whalley et al., 1990), quercetin in combination with other flavonoids, are inhibiting a number of enzymes like bradykinin (Bamard et al., 1993), tyrosine kinase (Hur, et al., 1994), and 5’-nucleotidase activity (Beladi, et al., 1987). Rutin and quercetin have shown regulatory activity of hormones like affect the transport, metabolism and action of thyroid hormones. High performance layer chromatography (HPLC) (Harbone, J.B. 1984) method is the suitable method for estimation of chemical constituents present in plant materials. Hence Terminalia chebula contains rutin and quercetin are important active constituents and is estimated by HPLC method.

MATERIALS AND METHODS

INSTRUMENTATION
The Shimadzu class LC-10AT HPLC, Hichrom C18 and a Rheodyne 7725i injector fitted with a 20µl loop, column oven, and a photodiode array detector. The output signal was monitored and processed using chromquest version 3.0 software on Pentium computer (Hewlett Packard).

SOLVENT AND CHEMICALS
Rutin and quercetin obtained from natural remedies (Bangalore) chromatographic grade methanol, formic acid and acetoniitrile (AR), were obtained from Merck (Mumbai, India).

PLANT MATERIAL
Terminalia chebula fruits were extracted with distilled water by Soxhlet apparatus. The pooled aqueous extract was evaporated under vacuum to dryness, yielding was noted. Aqueous extract was subjected for estimation of rutin and quercetin.

PREPARATION OF STANDARD AND SAMPLE SOLUTIONS
Rutin and quercetin 10 mg were accurately weighed into a 10 mL volumetric flask, dissolved in 5 mL methanol and the solution was made up to 10 mL with the same solvent (1
mg/mL). T. Chebula fruit extract was accurately weighed (10 mg) into a 10 mL of volumetric flask and dissolved in methanol the solution was filtered through Whatman filter paper No. 42 and the filtrate was made up to the mark with methanol.

RESULTS AND DISCUSSION

The retention time of standards rutin and quercetin were found to be 4.072 and 19.104 (Graphs1). The retention time of rutin and quercetin in Terminalia chebula were found to be 4.802 and 19.040 (Graphs2), which are matching with standard R values respectively. Then the amount of rutin and quercetin in Terminalia chebula was found to be 59.52 and 9.06 % w/v respectively graphs

ACKNOWLEDGEMENT

The authors are thankful to K.V.Naveen Kiran, Chairman, Sri K.V.College of Pharmacy, Chickballapur, for providing required facilities.

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