ISSN : 0974 - 7508 Volume 7 Issue 4

Trade Science Inc.

Natural Products

An Indian Journal

Short Communication

NPAIJ, 7(4), 2011 [171-173]

Determination of total phenol & flavonoid content in seeds of *Psoralea corylifolia*

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Received: 20th June, 2011 ; Accepted: 20th July, 2011

ABSTRACT

The aim of this work is to estimate the total phenolic and flavonoid content of alcoholic extract of seeds of Psoralea *corylifolia*. The total phenolic content was determined using the Folin- Ciocalteau assay. The total flavanoid content was also measured spectrophotometrically by using aluminum chloride colorimetric assy. The Total Phenolic Content &Total Flavanoid Content in the seeds of Psoralea corylifolia was found to be 149.2981µg/mg of extract & 132.3479 µg/mg of extract respectively.

KEYWORDS

Psoralea corylifolia;
Phenolic content;
Flavonoid content;
Gallic acid;
Quercetin.

INTRODUCTION

Psoralea corylifolia is a medicinally importantplant, belongs to family Fabaceae. The plant is wellrecognized in Chinese and Indian folkloric medicine[1]. The seeds have been used for over many decades as traditional medicine. The plant is of immense biological importance and it has been widely exploited since ages for its magical effect against several skin diseases like psoriasis, leucoderma and leprosy[2]. It is reported to contain essential oil, coumarins, alkaloids, flavonoids and terpenoids[3]. It shows antitumor[4], anti-allergic[5], antioxidant[6], insecticidal[7] and antimicrobial activity[8]. They have beenspecially recommended in the treatment of leucoderma, leprosy, psoriasis and inflammatory diseases of the skinand are prescribed both for oral administration and for local external application in the form of a paste or ointment[9].

MATERIALS AND METHODS

Instruments

A Shimadzu UV-Visible Spectrophotometer (UV-1700) with a matched pair of 10 mm quartz cells were used for experimental purpose. Shimadzu AUX-220 balance was used for weighing the samples.

Materials

The seeds of Psoralea corylifolia were procured from local market and authenticated for their correct botanical identity N.B.R.I., Lucknow, India for identification & Taxonomic authentification.(Specification No.: NBRI-SOP-202)

The seeds were dried, powdered and kept in an air tight container. All chemicals were AR grade were procured from S.D. Fine chemicals, Mumbai.

Preparation of the methanolic extract of Psoralea

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corylifolia: – The seeds of Psoralea corylifolia should not be crushed for extraction, as some of the active constituents lie in seed coat also. The seeds of plant were dried at 31ºC. 20 gm of seeds were extracted with 250 ml of methanol by hot extraction process for about 4-6 hours. The extract was filtered through Whatman filter paper no. 1 and the residue was dried and used for further analysis.

Determination of total phenolic content

The total phenolic content was determined using Folin Ciocalteau assay[10].

Preparation of standard Gallic acid solution

Standard stock solution (primary) was prepared by dissolving 100 mg of gallic acid in 10 ml of 80% methanol and then making the volume to 100ml with distilled water in a 100 ml volumetric flask to get concentration of 1mg/ml (1000µg/ml).

Preparation of calibration standard solutions

The calibration standard solutions were prepared by diluting primary stock solution with distilled water to get calibration standard solutions of 50, 100, 150, 250 and 500µg/ml of gallic acid solution, the absorbance was measured at λmax 725 nm, against reagent blank (TABLE 1) to construct standard plot (Figure 2).

Preparation of sample solution

100 mg of dry extract was dissolved in 10ml of 80% methanol and diluted to 100 ml with the solvent to get the conc. of 1mg/ml.

Procedure for estimation of total phenolic content

1 ml of sample (methanolic solution of extract) was added to 25 ml volumetric flask, containing 9 ml of distilled water. 1 ml of Folin-Ciocalteu phenol reagent was added to the mixture and shaken. After 5 min, 10 ml of 7% sodium carbonate solution was added to the mixture. The solution was diluted to volume (25 ml) with

TABLE 1 : Linearity table of Gallic acid solution.

Conc.(µg/ml)	Absorbance
50	0.0591
100	0.1126
150	0.1693
250	0.2791
400	0.4287
500	0.5362

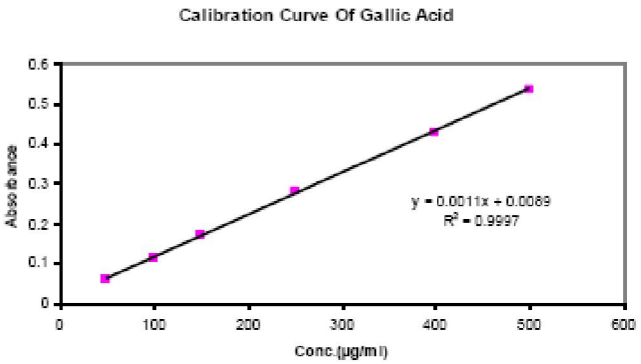

Figure 2 : Linearity curve of Gallic acid solution

distilled water and mixed. After incubation for 90 min at room temperature, the absorbance against prepared reagent blank was determined at 765 nm. Test samples were analyzed in triplicate and the conc. of the sample was determined using the calibration curve. The mean value is reported in gallic acid equivalents (GAE) using units of µg/mg of extract (TABLE 2)

TABLE 2 : Results for total phenolic content.

Sample Absorbance	Calculated Conc.(µg/ml)	Mean	SD	%RSD
0.1697	152.2330			
0.1652	147.9727	149.2981	2.5456	1.705
0.1649	147.6887			

Determination of total flavanoid content

Aluminum chloride colorimetric method was used for flavanoid determination[11].

Preparation of standard Quercetin solution

Standard stock solution (primary) was prepared by dissolving 10 mg of quercetin in 2 ml of 80% methanol and making the volume to 10ml with 80%methanol in a 10 ml volumetric flask to obtain concentration of 1mg/ml (1000µg/ml).

Preparation of calibration standard solutions

The calibration standard solutions were prepared by diluting primary stock solution with 80% methanol to obtain calibration standard solutions of 12.5, 25, 50, 75, 100,125 and 150µg/ml of gallic acid solution, the absorbance was measured at λmax 415 nm, against reagent blank (TABLE 3) to construct standard plot (Figure 3).

Preparation of sample solution

100 mg of dry extract was dissolved in 10ml of 80% ethanol and diluted to 100 ml with the solvent to

TABLE 3 : Linearity table of quercetin solution.

Conc.(µg/ml)	Absorbance
12.5	0.08
25	0.158
50	0.3152
75	0.4476
100	0.6182
125	0.7653
150	0.9061

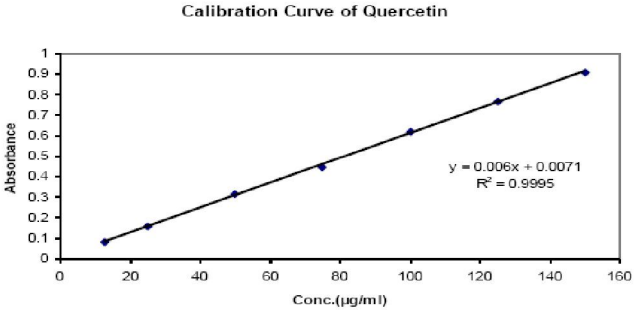

Figure 3 : Linearity curve of Quercetin solution

get the conc. of 1mg/ml.

Procedure for estimation of total flavanoid content

1 ml of sample (ethanolic solution of extract) was added to 10 ml volumetric flask containing 3 ml of 95% ethanol (v/v). To the flask was added 0.2 ml 10% aluminum chloride. After 5 min 0.2 ml of 1M potassium acetate was added and total volume was made upto 10 ml with distill water. The solution was mixed well and incubated at room temperature for 30 min. The absorbance of reaction mixture was measured at 415 nm. A volume of 10% aluminum chloride was substituted by same volume of distilled water in blank. Test samples were analyzed in triplicate and the conc. of the sample was determined using the calibration curve. The mean value is reported in quercetin equivalent using units of µg/mg of extract (TABLE 4).

TABLE 4 : Results for total flavanoid content

Sample Absorbance	Calculated Conc.(µg/ml)	Mean	SD	%RSD
0.8107	133.3394			
0.8013	131.7761	132.3479	0.8626	0.6518
0.8022	131.9258			

CONCLUSION

The Total Phenolic Content &Total Flavanoid Content in the seeds of Psoralea corylifolia was found to be 149.2981µg/mg of extract & 132.3479 µg/mg of extract respectively. The present data for total phenolic content & total flavanoid content are a basis for assessment of the preventive role of Psoralea corylifolia against free radical effects.

ACKNOWLEDGEMENTS

Authors are thankful to Prof. S.W.Akhtar Honourable Vice Chancellor Integral University, Lucknow for providing necessary facilities for the work.

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