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QUANTITATIVE ANALYSIS OF TOTAL PHENOLIC CONTENT IN BANAFSHAH FLOWER (VIOLA ODORATA. LINN) EXTRACTS

Abdul Wahab[1]*, Aisha Siddiqui[2], Umar Jahngir[3], Ayman Kirmani[4], Anwar Jamal[5] and Rasikh Javaid[6]

[1,4]Research Scholar, Department of Pharmacology (Ilmul Advia) School of Unani Medical Education and Research Jamia Hamdard New Delhi-62.
[2]Associate Professor, Department of Pharmacology (Ilmul Advia) School of Unani Medical Education and Research Jamia Hamdard New Delhi-62.
[3]Dept. of Amraze-Jild wa Tahziniyat School of Unani Medical Education and Research Jamia Hamdard New Delhi-62.
[6]Assistant Professor, Department of Pharmacology (Ilmul Advia) School of Unani Medical Education and Research Jamia Hamdard New Delhi-62.
[5]Research Officer (U)/SL-IV RRIUM(CCRUM), D-11 Abul Fazal Enclave, Jamia Nagar, Okhla, New Delhi-25.

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***Corresponding Author**
Dr. Abdul Wahab
Research Scholar, Department of Pharmacology (Ilmul Advia) School of Unani Medical Education and Research Jamia Hamdard New Delhi-62.

ABSTRACT

Viola odorata flower were analyzed for total phenolic contents. This phenolic component is responsible for antioxidant activity. The amount of total phenols were analyzed with the Folin-Ciocalteu reagent. Gallic acid was used as a standard compound and the total phenols were expressed as mg/g gallic acid equivalents (Standard curve equation: y = 0.0507 + 0.0619, R2= 0.9689). The hydroalcoholic extract had a lower TPC value of 11.496±0.207 mg/g, indicating that the aqueous extract contains a higher concentration of phenolic compounds. The total phenolic content of the aqueous extract was found to be 22.163 ±0.5356 mg/g.

KEYWORDS: *Viola odorata, total phenols, Folin-Ciocalteu reagent, Gallic acid.*

The terms "Banafshah" and "sweet violet" refer to the medicinal plant Viola odorata Linn. (Violaceae). It is found in high altitudes in North America, Europe, and the Himalayas. This plant has a long trailing stalk and is less than 6 inches tall. The broad subterranean stem of the plant is covered in scaly rooting runners. Its

heart-shaped leaves are smooth, dark green, or rarely downy, and they are grouped in a rosette at the base of the plant. The edges of the leaves are serrated, or scalloped. Bright yellow-white flowers range in color from deep purple or blue to pink.[1-2] Viola odorata L., often known as the sweet violet, grows wild in natural environments next to riverbanks, hedgerows, the boundaries of deciduous forests, and forest glades. It is believed that the Mediterranean, which now encompasses portions of North America, South Africa, Tierra del Fuego, Australia, and Antarctica, was the sweet violet's original home. Violet flowers were traditionally used to treat a wide range of ailments, especially eye, fever, and chills.[3]

Sweet violet is used to treat pneumonia, diabetes, postoperative tumor metastases, cancer, and common stomach problems. Numerous chemical classes have been identified phytochemically from different species of this genus, including cyclotides, flavonoids, alkaloids, and triterpenoids.

MATERIAL AND METHODS

Authentification of plant material

The *Banafshah* (*Viola odorata* Linn.) flower was identified by the taxonomist at the National Institute of Science Communication and Information Resources (NISCAIR), New Delhi, which were bought from Khari Baoli, Old Delhi (Authentication No. NIScPR/RHMD/Conult/2023/4318-19).

Sample preparation

Hydro-alcohol extract

Banafshah flower coarse powder was extracted over a 6-hour period using a Soxhlet device and solvents were used hydro-alcoholic solvent (Ethanol and Distilled water in ratio of 50:50).

Aqueous extract

The plant material (20 gm of powdered Viola odorata flower) was extracted using 250 ml of water and refluxed at 70^0c for 18 hours. The crude extracts were obtained after filtering and evaporation to dryness.[4]

Chemicals and Instruments

Methanol, gallic acid, anhydrous sodium carbonate, and Folin-Ciocalteu's phenol reagent. Shimadzu UV spectrophotometer**.**

Preparation of Folin-Ciocalteu's Phenol reagent

A 1500 ml flask was filled with 800 ml of water, 100 g of sodium tungstate and 25 g of sodium molibedate, 50 ml of phosphoric acid, and 100 ml of HCl. The flask was then left to reflux for ten hours. After the mixture cooled, mix with 50 milliliters of water, 150 grams of lithium sulfate, and four to six drops of bromine water. Let the mixture stand for two hours. After a 15-minute boil, the mixture was allowed to cool before being filtered. There should be no trace of green in the reagent.[5]

Procedure for determination of total phenolic contents

Using the Folin-Ciocalteu reagent, the total phenolics in extracts were measured as milligrams per gram of gallic acid equivalents (GAE), using gallic acid as the standard.[6] the Concentration of standard solution (gallic acid) 0.5,1.0,2.5,5.0, and 10.0 mg/ml were prepared in methanol. Concentration of 0.5 ml of 10 mg/ml of plant extract was also produced in methanol. 0.5 ml of each sample was added to test tubes along with 4 ml of 7.5% sodium carbonate and five tablespoons of a 10-fold diluted Folin-Ciocalteu reagent. Following a half-hour of room temperature storage in parafilm-wrapped tubes, the absorbance was determined spectrophotometrically at 716 nm.[7]

Quantitative analysis of total phenolic content

Tab. 1: Absorbance of standard compound (Gallic acid).

S. No	Concentration (µg/ml)	Absorbance (Mean) λmax=716 nm
1	0.5	0.118
2	1.0	0.158
3	2.5	0.199
4	5.0	0.286
5	10.0	0.307

Tab. 2: Total phenolic content of viola odorata in different plant extracts.

S. No	Extract sample	Concentration (µg/ml)	Mean±SD
1	Aqueous extract	100	22.163 ±0.535
2	Hydro-alcoholic extract	100	11.496±0.207

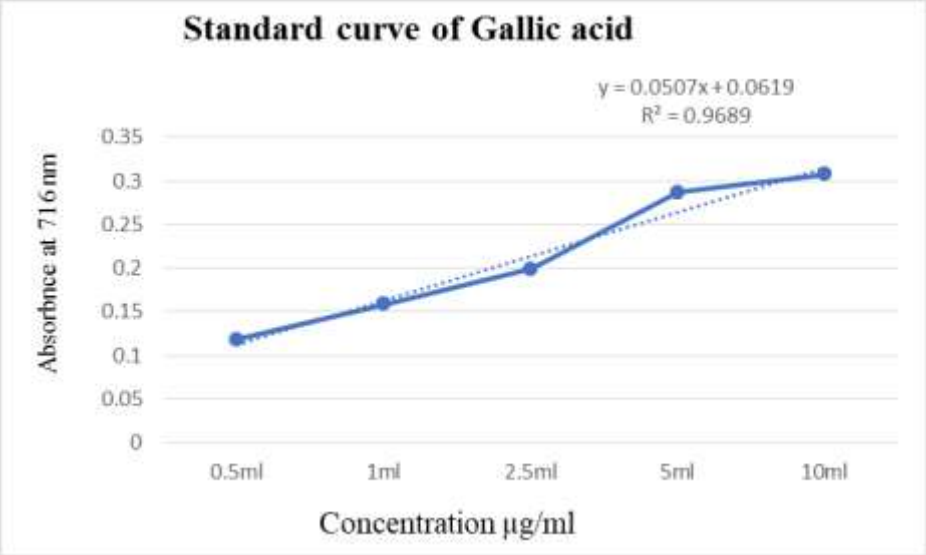

Figure 1: Standard curve of gallic acid.

RESULTS AND DISCUSSION

The Folin-Ciocalteu reagent was used to calculate the total phenol content. Using gallic acid as a reference chemical, the total phenols were reported as milligrams per milligram of gallic acid equivalent using the standard curve equation: y = 0.0507x + 0.0619, R2=0.9689, where x is the total phenolic content, expressed in mg/gm, of the various Viola odorata preparations and y is the absorbance at 716 nm. A group of antioxidant drugs that functions as a free radical terminator is phenolic compounds.[8] Table.1 shows the variation of mean absorbance with concentration of Gallic acid. Table.2 shows the contents of total phenols that were measured by Folin Ciocalteu reagent in terms of gallic acid equivalent. The total phenolic content of the aqueous extract was found to be 22.163 ±0.5356 mg/g**.** the aqueous extract contains a higher concentration of phenolic compounds.

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