



## **Evaluation for Total Phenolic, Total Flavonoid and Antioxidant activity of leaves and roots of *Pyrus pashia***

Preeti Rawat<sup>1\*</sup>, Neetishwar Saroj<sup>2</sup>, Pinki Rawat<sup>3</sup>, Piyush Kumar<sup>4</sup>, Tryambak D Singh<sup>5</sup>, Mahesh Pal<sup>1</sup>

<sup>1</sup>Phytochemistry Division, CSIR- National Botanical Research Institute, Lucknow, India.

<sup>2</sup>DJ College of Pharmacy, Modinagar, Gaziabad, India

<sup>3</sup>Dept. of Pharmaceutical Sciences and Technology, Birla institute of Technology, Ranchi, Jharkhand, India

<sup>4</sup>Dept. of Chemistry, Indian Institute of Technology, Kanpur, India

<sup>5</sup>Dept. of Medicinal Chemistry, Institute of Medical Sciences, Banaras Hindu University, Varanasi, India

**Abstract:** The antioxidant activity and total phenolic content of methanolic extracts of leaves and fruits of *Pyrus pashia* were evaluated by using a model system containing DPPH free radical and Folin-Ciocalteu method. The total phenolic content of the extracts was determined spectrophotometrically according to the Folin-Ciocalteu procedure and found to be 325±10 mg/g and 98±5 mg/g in leaves and fruits respectively on fresh weight basis. The total flavonoid content of extracts of leaves and fruits were determined by Aluminium chloride colorimetric assay and ranged from 150±20 mg/g to 10.30±10 mg/g respectively.

**Key words:** *Pyrus pashia*, Antioxidant, Phenolic, Flavonoid, DPPH.

### **Introduction**

Medicinal plants are best remedies used as alternative tools for the prevention and treatment of many ailments. The recent studies have investigated that the antioxidant effect of plant products is mainly attributed to phenolic compounds such as flavonoids, phenolic acids, tannins etc<sup>1,2</sup>.

Rosaceae (the rose family) is a medium-sized family of flowering plants, including about 2830 species in 95 genera<sup>3</sup>. Rosaceae includes herbs, shrubs and trees. Most species are deciduous, but some are evergreen<sup>4</sup>.

*Pyrus pashia* is commonly known as wild Himalayan pear, Punjabi pear, Indian pear belongs to family Rosaceae. It is a small to medium size deciduous tree of the small and oval shaped crown with ovate, finely toothed leaves, attractive white flowers with red anthers and small pear-like fruits. *Pyrus pashia* is a tolerant tree that grows on sandy loamy soil that is well drained. It is adapted to a precipitation zone that ranges from 750 to 1500mm/yr or more, and a temperature that ranges from -10 to 35 C<sup>5</sup>. Its fruit is edible and characterized as being pome<sup>6</sup>. It looks like the russet apple and has an astringent but sweet taste when ripe. The astringent juice is used to stop diarrhoea.

It is native to southern Asia. Locally, it is known by many names such as batangi<sup>4</sup> (Urdu), tangi (Kashmiri), mahal mol (Hindi) and passi (Nepal)<sup>5</sup>. Leaves are used as fodder, leaf extract is tonic for hair fall and wood of the tree are used as a source of fuel in Himalayan region<sup>7</sup>. The leaves are consumed as tea beverages by the Monpa community of Tawang, Arunchal Pradesh.



**Figure 1: *Pirus pashia* Plant**



**Figure 2: *Pirus pashia* fruit**

## Material and method

### Extraction

Samples were shade dried at ambient temperature ( $\pm 24^{\circ}\text{C}$ ) and powdered. Powdered leaves and fruits were extracted with methanol. The methanolic extract was evaporated in a rotatory evaporator and dried by vacuum pump. The methanolic extract was suspended on water and extracted successively with hexane, ethyl acetate and butanol respectively. Extract was filtered with Whatman no.1 filter paper and concentrated under reduced paper to dryness below  $40^{\circ}\text{C}$ .

### Determination of Total Phenolic Content

Total phenolic compound contents were determined by the Folin-Ciocalteu method<sup>8,9</sup>. The extract samples (0.5 ml) were mixed with 2.5 ml of 0.2 N Folin-Ciocalteu reagent for 5 min and 2.0 ml of 75 g C 1 sodium carbonate were then added. The absorbance of reaction was measured at 760 nm after 2 h of incubation at room temperature. Results were expressed as gallic acid equivalents.

### Determination of Flavonoid Content

Total flavonoids were estimated using the method of Ordonez et al.<sup>8,9</sup>. 0.5 mL solution of each plant extracts in methanol were separately mixed with 1.5 mL of methanol, 0.1 mL of 10% aluminum chloride, 0.1 mL of 1 M potassium acetate, and 2.8 mL of distilled water and left at room temperature for 30 minutes. The absorbance of the reaction mixture was measured at 415 nm with a double beam spectrophotometer (Perkin Elmer). Total flavonoid contents were calculated as quercetin from a calibration curve.

### DPPH Radical Scavenging Activity

The stable 1,1-diphenyl-2-picryl hydrazyl radical (DPPH) was used for determination of free radical scavenging activity of the extracts<sup>9,10</sup>. Different concentrations of each extracts were added, at an equal volume, to methanolic solution of DPPH (100  $\mu\text{M}$ ). After 15 min at room temperature, the absorbance was recorded at 517 nm. The experiment was repeated for three times. Quercetin were used as standard controls.

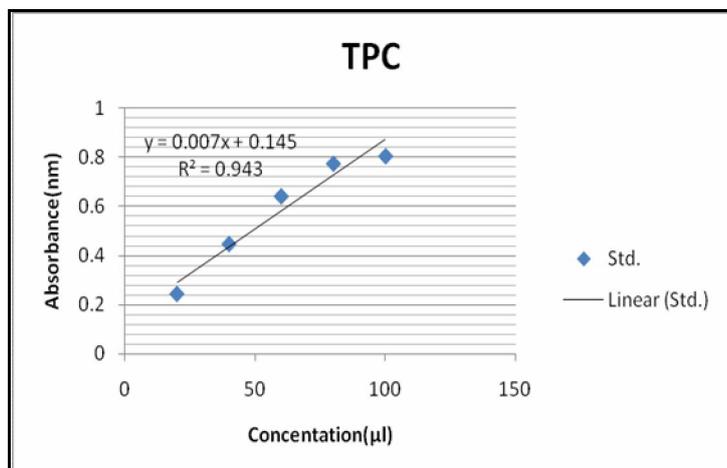
### Statistical analysis

Experimental results are expressed as means  $\pm$  SD. All measurements were replicated three times. The data were analyzed by an analysis of variance ( $p < 0.05$ ) and the means separated by Duncan's multiple range test. The  $\text{EC}_{50}$  values were calculated from linear regression analysis.

## Result and Discussion

### Total Phenol Content

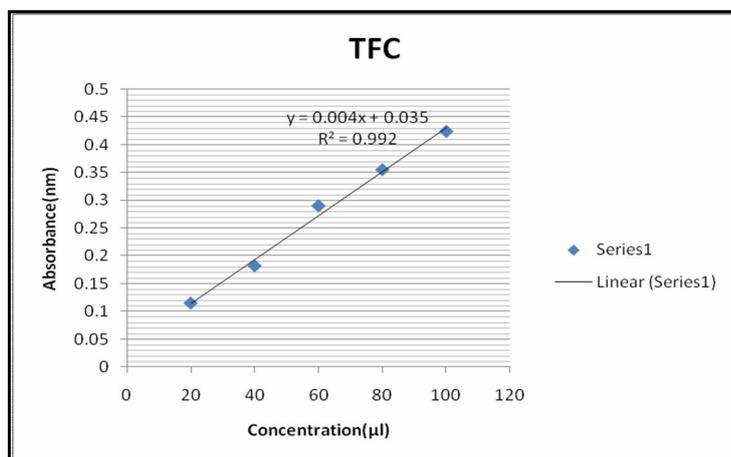
The Total phenolic content were reported as gallic acid equivalents by reference to standard curve . The total phenolic contents of leaves and fruits of *Pyrus pashia* were found to be  $325 \pm 10$  and  $98 \pm 5$  mg gallic acid equivalent/g of extract powder, respectively.



**Graph 1. Standard curve of Gallic acid**

#### Total Flavonoid Content

The Total flavonoid content of leaves and fruits of *Pyrus pashia* were found to be  $150 \pm 20$  and  $10.30 \pm 10$  mg quercetin equivalent  $g^{-1}$  of extract powder, respectively, by reference to standard curve .



**Graph 2. Standard curve of quercetin**

#### DPPH Radical Scavenging Activity

The radical-scavenging activity of leaves and fruits of *Pyrus pashia* for DPPH radical-scavenging activity was found to be  $15.12 \pm 10$  and  $8.5 \pm 5$  mg/ml respectively.

**Table 2 : Extractive yield , TPC, TFC and % Inhibition of extracts of *Pyrus pashia***

	Leaves	Fruits
Extraction yield ( % )	20	22
Total phenolic content	$325 \pm 10$ mg / g	$98 \pm 5$ mg / g
Total flavonoid content	$150 \pm 20$ mg / g	$10.30 \pm 10$ mg / g
% Inhibition at 200 mg / ml	$15.12 \pm 10$	$8.5 \pm 5$

## Conclusion

The present study evaluated the antioxidant activity, total phenolic and flavonoid contents in leaves and fruits of *Pyrus pashia*. It is important to measure the antioxidant activity using various radicals and oxidation systems in order to realize the health benefits from potential plant sources. *Pyrus pashia* possess significant antioxidant property.

## Acknowledgement

The authors are thankful to the Director, CSIR-National Botanical Research Institute, Lucknow, India for facilities and encouragements. The financial support received from University Grant Commission, New Delhi, India to carry out the research work is duly acknowledged.

## References

1. Nagavani V, Rao TR. Evaluation of antioxidant potential and qualitative analysis of major polyphenols by RP-HPLC in *Nymphaea nouchali* Burn flowers. International Journal of Pharmacy and Pharmaceutical Sciences 2 (Suppl 4) 2010, 98-104.
2. Cartea ME, Francisco M, Lema M, Soengas P, Velasco P. Resistance of cabbage (*Brassica oleracea* capitata group) crops to *Mamestra brassicae*. Journal of Economic Entomology 103(5), 2010, 1866-1874.
3. "Angiosperm Phylogeny Website". *mobot.org*.
4. Watson, L., and Dallwitz, M.J. (1992 onwards). The families of flowering plants: descriptions, illustrations, identification, and information retrieval. Version: 21 March 2010. <http://delta-intkey.com/angio/www/rosaceae.htm>
5. Zamani, A. et al. "*Pyrus pashia* (rosaceae), a new record for the flora of Iran. Iran.Journ. Bot. 2007, 15 (1), 72–75. Tehran.
6. Parmar, C. and Kaushal MK. *Pyrus pashia* Buch. & Ham, In: Wild Fruits. Kalyani Publishers, New Delhi, India. 1982,78–80.
7. Sheikh, MI. "*Pyrus Pashia*". Trees of Pakistan 1992, 5–142.
8. M.A. Ebrahimzadeh, S.J. Hosseinimehr, A. Hamidinia and M. Jafari. Antioxidant and free radical scavenging activity of *Feijoa sallowiana* fruits peel and leaves. Pharmacology online 2008, 1: 7-14.
9. Ebrahimzadeh MA, Pourmorad F and Hafezi S. Antioxidant Activities of Iranian Corn Silk. Turkish Journal of Biology 2008, 32: 43-49.
10. Nabavi SM, Ebrahimzadeh MA, Nabavi SF, Hamidinia A and Bekhradnia AR. Determination of antioxidant activity, phenol and flavonoids content of *Parrotia persica* Mey. Pharmacologyonline 2008, 2, 560-567.

\*\*\*\*\*