Journal of Pharmacognosy and Phytochemistry

Available online at www.phytojournal.com

J P P Journal of Pharmacognosy and Phytochemistry

E-ISSN: 2278-4136
P-ISSN: 2349-8234
JPP 2017; 6(5): 2563-2568
Received: 20-07-2017
Accepted: 22-08-2017

Deepak Chandra Sati
Department of Pharmaceutical Sciences, Kumaun University Nainital, Uttarakhand, India

Correspondence
Deepak Chandra Sati
Department of Pharmaceutical Sciences, Kumaun University Nainital, Uttarakhand, India

Pharmacognostical and phytochemical screening of leaf and fruit extract of *Pyracantha crenulata*

Deepak Chandra Sati

Abstract
Pyracantha crenulata is an evergreen shrub grows in the range between 1000 –2600m. In Uttrakhand it is wildly spread in the Nainital, Chamoli, Uttrakashi, Tahri, Almora, Bageshawr, Champawat and Pithoragarh district. Present study focused on the microscopical characteristic of Leaves and fruits of Pyracantha crenulata. Total Ash Value of leaves is observed 5.38%. Ethanolic extract 12.5%, water extractive value 15%, loss on drying 1.16%. Total Ash value of fruits is observed 5.966. Ethanolic and water extractive value s are 10 and 12.5% respectively. LOD is 1.33% observed. Phenolic content is 50% and Flavonoid content is 35%. In the T.S section of leave various cellular component are observed which are mention below in the pic. NO (A-E).

Keywords: *Pyracantha crenulata*, Ethanolic Extract, Ash Value, Extractive Value

1. Introduction
In India *Pyracantha crenulata* is grow in the range between 1000 –2600m [1]. In Uttrakhand it is wildly spread in the Nainital, Chamoli, Uttrakashi, Tahri, Almora, Bageshawr, Champawat and Pithoragarh district. *Pyracantha crenulata* commonly known as Ghingaru, *kingdom Plantae,Family* Rosaceae. *Pyracantha crenulat* contain leucocynidine flavanoids vitexin4 rhamnoside, vitexin glycoside and oligomeric leucoanthocyanidin.Fruits contain flavonol, kaemmpferol, quercetin and saponins, βsterol etc [2]. The fruits are full of medicinal properties [5]. It has some cardio tonic activity so it is useful in following cardiovascular disorders- Coronary vasodilator, Hypertension, Cardiac failure, myocardial weakness, Paroxysomal tachycardia, arteriosclerosis.
It is also useful in treatment of Burgors disease. Fruits contain anti-oxidant properties which reduced the free radical in our body Barks is useful in prevention of heavy bleeding during menstrual cycle [3].

2. Materials and Method:-
2.1 Powder Microscopic Examination
Glycerol reagent according to WHO rule was readied. Little measure of powder was gone up against the slide to this blend was included and secured with cover slip and analyzed under magnifying lens [4].

2.2 Microscopic Evaluation
Infinitesimal assessment is a stage towards confirmation of inward structure of the vegetable medication to build up legitimate distinguishing proof by uncovering tissue game plan. This is finished by Identifying inward structures, for example, epidermis, collenchyma, vascular groups, sorts and game plan of vascular packs, sclerenchyma, precious stones and whatever other particular highlights that lie there in. For this reason a transverse segment or longitudinal area by either free hand or utilizing microtome, might be readied. For the present work, free hand areas were set up as takes after [5].

2.3 Free Hand Sectioning
The midrib of the leaf was cut using a sharp razor including a small portion of lamina. The portion of midrib was put between the pith and fine sections were cut with the help of a sharp razor. The sections so obtained were cleared using chloral hydrate solution [6].

2.4 Staining
The cleared areas were exchanged to a watch glass containing recoloring arrangement (Safranin 1% arrangement). The areas were permitted to recolor for 2-3 minutes.

The segments were then exchanged to a watch glass containing plain refined water to wash away abundance of stain. The areas were then exchanged to a clean small scale slide and saw under magnifying instrument. In the event that any abundance of stain was watched the same was expelled by washing the segment with dil. HNO3 [7].

2.5 Microphotography
The photographs were taken with the help of a electric Microscope.

2.6 Physico-chemical Analysis
The parameter studied were total ash, acid insoluble ash, alcohol soluble extractives and water soluble extractives.

2.7 Recommended procedure
2.7.1 Loss on Drying:
The powdered leaves of *Pyracantha crenulata* was dried in the oven at 100- 105^{0}C to constant weight.

2.7.2 Total Ash:
Place about 2-4g of the ground air-dried material, accurately weighed, in a previously ignited and tared crucible (usually of platinum or silica). Spread the material in an even layer and ignite it by gradually increasing the heat to 450°C until it is white, indicating the absence of carbon. Cool in a desiccators and weigh. Ash value can be calculated by using [8].

Formula

$$\text{Ash value} = \frac{\text{Initial Weight} - \text{Final Weight}}{\text{Initial Weight}} \times 100$$

2.8 Determination of Extractive Value
This method determines the amount of active constituents extracted with solvents from a given
amount of medicinal plant material.

3. Method
3.1 Hot extraction
3.1.1 Water Soluble Extractive Value: Place around 15gm of coarsely powdered air-dried material, precisely weighed, in a glass-stoppered conelike jar. Include 300ml of water and weigh to acquire the aggregate weight including the carafe. Shake well and permit remaining for 60 minutes. Connect a reflux condenser to the cup and bubble tenderly for 6 hour; cool and weigh and channel quickly through a dry channel. Dry the removed powder in broiler till the weight is consistent. Ascertain the substance of extractable issue in mg per gm of air dried material [9].

3.1.2 Alcohol Soluble Extractive Value Place around 15gm of coarsely powdered air-dried material, precisely weighed, in a glass-stoppered conelike carafe. Include 300ml of water and weigh to get the aggregate weight including the jar. Shake well and permit remaining for 60 minutes. Append a reflux condenser to the jar and bubble delicately for 6 hour; cool and weigh and channel quickly through a dry channel. Dry the separated powder in broiler till the weight is steady. Ascertain the substance of extractable issue in mg per gm of air-dried material by utilizing this [10].

Formula

$$\text{Extractive value} = \frac{\text{Initial weight-Final weight}}{\text{Initial weight}} \times 100$$

3.1.3 Phytochemical Screening
Phytochemical screening of the extract gives general idea regarding the nature of chemical constituents present in the crude drug. Different test for Alkaloid, carbhohydrate, protein, flavinoids and Steroids has been carried out; there result has been mentioned in table 1 given below

Table 1: Table of Phytochemical screening of leaves

S. No. A	Tests for Alkaloids	Obserbation
1.	Dragandroff test	+
2.	Mayer's test	
3.	Hager's test	+
4.	Wagner's test	+
5.	Tanic acid test	+
B	Testsfor Carbohydrate	
1	Molish test	+
2	fehling test	+
3	Benedict test	+
4	Tollen's test	+
C	Tests for Proteins	
1	Biuret test	+
2	Millon's test	+
3	xanthoprotein test	+
D	Tests for Steroids	
1	Salkowski test	+
E	Tests for Flavanoids	
1	Shinoda test	+
2	Sulphuric acid test	+

4. Determination of total phenolic content
Add up to solvent phenols in the concentrates were resolved with Folin Ciocalteau reagent utilizing Gallic corrosive as a standard phenolic compound. 0.1 ml of concentrate arrangement taken in a test tube and 1ml of FC reagent was included and 3-5 min later, 2.5 ml of 20% sodium carbonate was included and the blend was permitted to remain for 30min with irregular shaking. The absorbance of the blue shading that created was perused at 765 nm. The convergence of aggregate phenols was communicated as mg/gm of dry concentrate. The grouping of aggregate phenolic mixes in the concentrate was dictated by utilizing the equation:
T=CV/M
Where, T= Total phenolic content mg/gm of plant extract in GAE, C= Concentration of Gallic acid from the calibration curve, V= volume of the extract in ml, M= wt of the pure plant methanol extract [11].

5. Determination of total flavonoids
The aggregate flavonoid substance of the concentrate was resolved utilizing a colourimeter examine created. 0.2ml of the concentrate was added to 0.3ml of 5% NaNO3 at zero time. After 5min, 0.6ml of 10% AlCl3 was included and after 6min, 2ml of NaOH was added to the blend took after by the option of 2.1ml of refined water. Absorbance was perused at 510nm against the reagent clear. The grouping of aggregate flavonoids was communicated as mg/gm of dry concentrate. The convergence of aggregate phenolic mixes in the concentrate was dictated by utilizing the equation [12].
T=CV/M
Where, T= Total flavonoid content mg/gm of plant extract, C= Concentration of quercetin from the calibration curve, V= volume of the extract in ml, M= wt of the pure plant methanol extract.

Fig 1: Powder microscopy of *P. crenulata* leaf

Fig 3: Glandular fiber

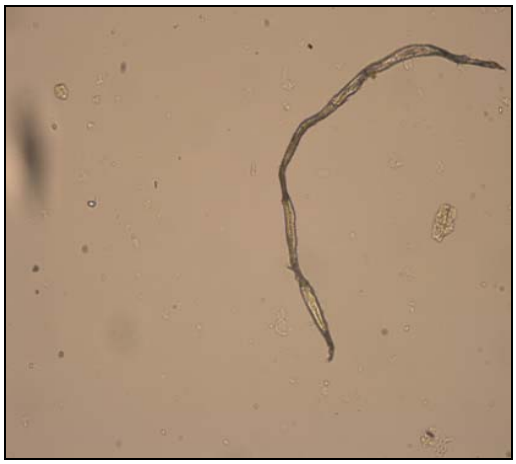

Fig 2: Starch grain

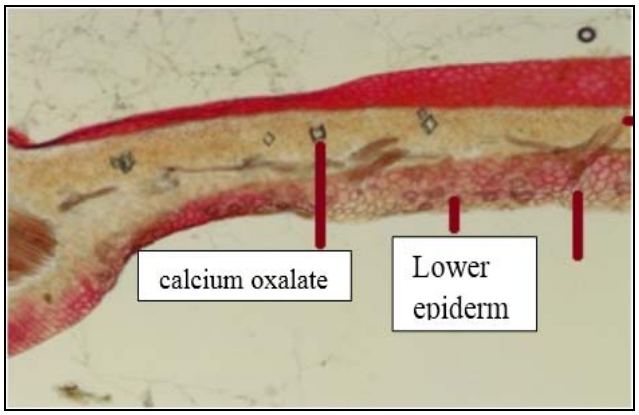

Fig 4: Prismatic calcium oxalate Cristal

Fig 5: T.S of Pyracantha Crenulata leaves

Table 2: Percentage loss in weight on drying

S. No	Weight of the powder taken (gm)	% Loss in weight (w/w)	Mean value	% Loss on drying
1	1.5	1.48	1.33	1.16
2	1.5	1.49	0.66	
3	1.5	1.49	0.66	
4	1.5	1.47	2	

Table 3: Extractive value

S. No	Solvent	Initial Weight of Sample	Amount of Solvent	Final Weight of Sample	Extractive Value
1	Ethanol	4	100ml	0.5	12.5%
2	Water	4	100ml	0.6	15%

Table 4: Total Ash value of leaves powder

S. No	Blank Crucible wt in gm	Crucible wt with 2 gm of powder in gm	Weight of ash obtained (gm)	% w/w total ash	Mean value
1	14.164	13.494	0.411	4.73	5.38
2	14.232	13.572	0.54	4.63	
3	13.87	12.931	0.94	6.77	

Table 5: Water soluble and acid soluble ash value

Water soluble ash value	Acid soluble ash value
3.72%	4.23%

Powder microscopy of *P. crenulata* fruit.

Table 6: Percentage loss in weight on drying of *P. crenulata* fruit.

S. No	Weight of the powder taken (gm)	% Loss in weight (w/w)	Mean value	% loss on drying
1	1.5	1.49	0.66	1.33
2	1.5	1.47	2	
3	1.5	1.48	1.33	
4	1.5	1.48	1.33	

Table 7: Extractive value of fruit

S. No	Solvent	Initial Weight of Sample(gm)	Amount of Solvent (ml)	Final Weight of Sample	Extractive Value
1	Ethanol	4	100	0.4	10 %
2	Water	4	100	0.5	12.5 %

Table 8: Total Ash value of fruits

S. No	Blank Crucible wt in gm	Crucible wt with 2 gm of powder in gm	Weight of ash obtained (gm)	% w/w total ash	Mean value
1	10.73	9.983	0.747	6.961	5.966
2	11.43	10.875	0.555	4.85	
3	11.45	10.753	0.697	6.087	

Table 9: Phenolic content in Galic acid.

S. NO	Sample solution(µg/ml)	Weight of dry extract (g/ml)	Volume of sample	Concentration of Gallic acid as mg/gm	Mean ±SEM
1	1000	0.001	1	81	82 ±0.8819
2	1000	0.001	1	82	
3	1000	0.001	1	82	

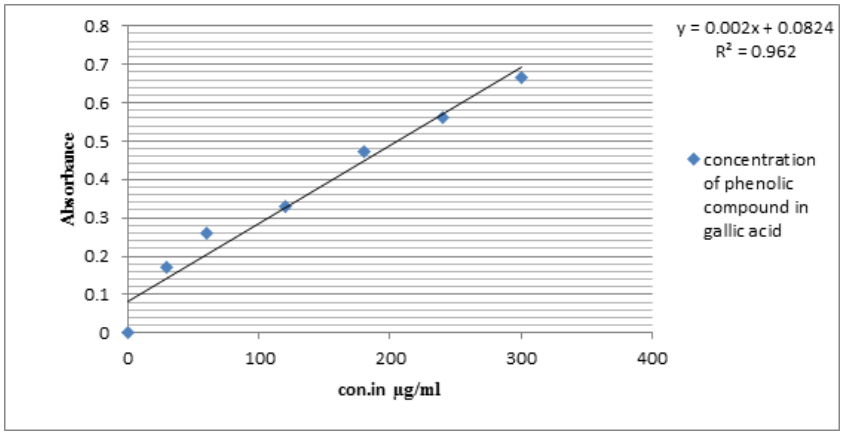

Fig 6: Calibration cruve of Galic acid at different concentration.Phenolic content was found to be 82 mg/g.

Table 10: Phenolic content in Extract

S. NO	Sample solution(µg/ml)	Weight of dry extract (g/ml)	Volume of sample	Concentration of extract as mg/gm	Mean ± SEM
1	1000	0.001	1	50	49.66 ± 0.3333
2	1000	0.001	1	50	
3	1000	0.001	1	49	

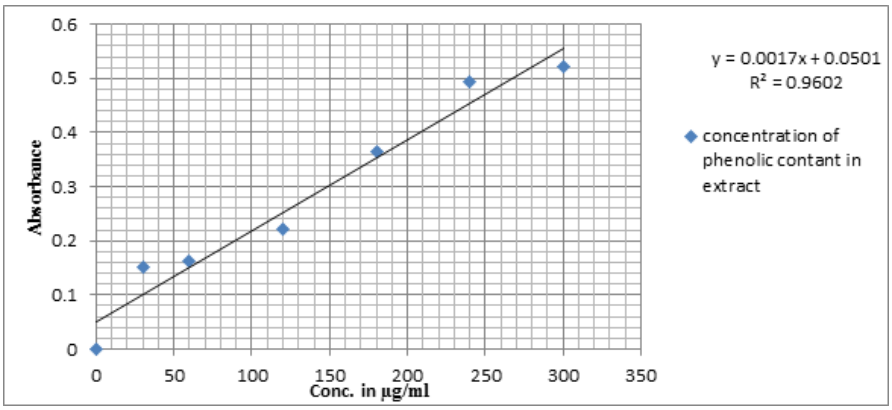

Fig 7: Calibration curve of different concentration of extract.Phenolic content was found to be 50mg/g

Total flavanoid content

Table 11: flavanoid content in Quercetin

S. No	Sample solution(µg/ml)	Weight of dry extract (g/ml)	Volume of sample	Concentration of QE as mg/gm	Mean ± SEM
1	1000	0.001	1	84	84 ± 0.6667
2	1000	0.001	1	86	

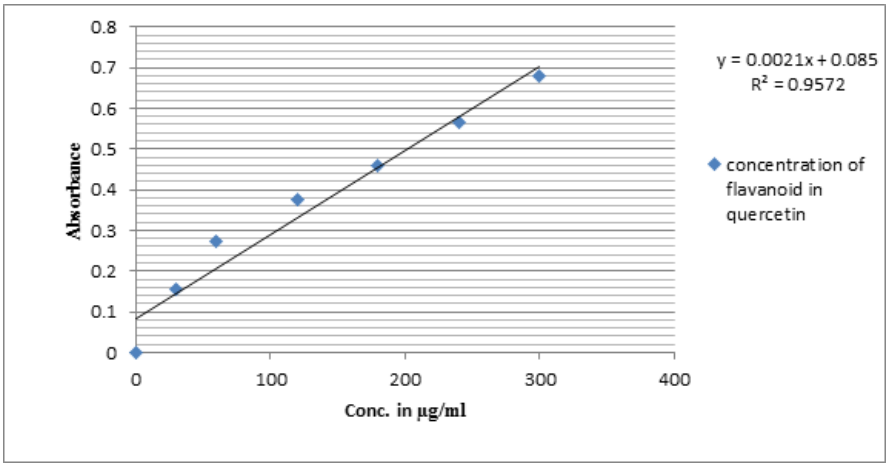

Fig 8: Calibration curve of extract at different concentration, flavanoid content was found to be 85mg/g

Table 12: Flavanoid content in extract

S. No	Sample solution(µg/ml)	Weight of dry extract (g/ml)	Volume of sample	Concentration of Extract as mg/gm	Mean ± SEM
1	1000	0.001	1	33	34.333 ± 0.8819
2	1000	0.001	1	36	
3	1000	0.001	1	35	

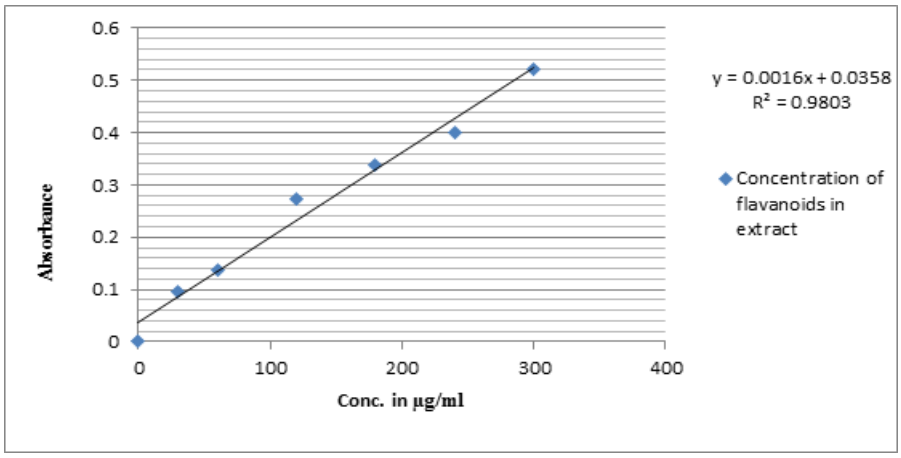

Fig 9: Calibration curve of extract at different concentration. Flavanoid content found to be 35mg/ml.

Conclusion

Pyracantha crenulata showed the presence of Glycoside, Carbohydrate, tennins, Amino Acids, Sterols and Terpenoids. The loss on drying of the powder of *P. crenulata* was found to be 1.16%. The ash value of powder of *P. crenulata* leaf was determine as total ash, water Soluble ash and acid insoluble ash was found to be 3.7%. The Extractive Value of *P. crenulata* was found to be 12.5% and 15% in Aqous and Ethanol respectivly and methanolic extract of fruit extract showed 50% Phenolic and 35% Flavanoid content respectivly. On powder microscopy following feature are observed like- Prismetic calcium oxalate, stomata, fiber, starch ggain,oil gland and Vascular bundle.These data can provide valuable information on standardization and evaluation of the leaves and fruits of *P. crenulata*.

Acknowledgement

I am thankful to Dr. Mahendra Rana Asst Prof. Kumaun University, Bhimtal campus for guiding me and providing laboratory facilities for my research work.

References

1. Saklani S, Chandra S. Preliminary phytochemical evaluation of Garhwal Himalaya wild edible fruit *Pyracantha crenulata*. Journal of Pharmacy Research. 2012; 5(6):15-21.
2. Saklani S. *In Vitro* Antimicrobial Activity, Nutritional Value, Anti Nutritional value and Phytochemical Screening of *Pyracantha Crenulata* Fruit. Int. J Pharm. Sci. Rev. 2014; 26(1):1-5.
3. Pal RS, Kumar A, Agrawal PK, Baatt JC. Antioxidant capacity and related phytochemicals analysis of methanolic extract of two wild edible fruits from north western indian Himalaya. *I*nternational journal of Pharma and Bioscience. 2013; 4(2):113-123.
4. Shah Shruti, Tiwari B, Bisht S, Tiwari A. Seed maturation indication in *Pyracantha crenulata* Roxab. in Kumaun Central Himalaya, Springer. 2005, 32(1).
5. Mehra A, Diversity. Utilization and sacred values of Ethno-medicinal plants of Kumaun Himalaya. Int. J Society for Tropical Plat Research. 2014; 1(3):80-86.
6. Bahuguna MY. Evaluation of *Pyracantha Crenulata* Roem for Antiurolithogenic Activity in Albino Rats. African Journal of Urology. 2009; 15(3):159-166.
7. Sharma R. Plants of India an Encyclopaedia. Daya Publishing house Delhi, 2003; 208-209.
8. Bahuguna MY. Phytochemical Examinatiin of Fruits of Pyracantha Crenulata (D.DON) M. Roemer. International Journal of Advances in Pharmaceuticl Research. 2014; 7(3):123-129.
9. Saklani S, Chandra S. Preliminary phytochemical evaluation of Garhwal Himalaya wild edible fruit *Pyracantha crenulata*. Journal of Pharmacy Research. 2012, 5(6).
10. Kamalesh Upreti, Amit Semwal, Kumud Upadhyaya1, Manoj Masiwal. Pharmacognostical and Phytochemical Screening of Leaf Extract of *Zanthoxylum armatum* DC, International Journal of Herbal Medicine. 2013; 1(1):6-11.
11. Rakhi Rawat, DP Vashistha. Common Herbal Plant in Uttarakhand, Used in The Popular Medicinal Preparation in Ayurveda, International Journal of Pharmacognosy and Phytochemical Research. 2011; 3(3):64-73.
12. Kamalesh Upreti, Amit Semwal, Kumud Upadhyaya, Manoj Masiwal. Pharmacognostical and Phytochemical Screening of Leaf Extract of *Zanthoxylum armatum* DC, International Journal of Herbal Medicine, Year. 2013; 1(1):1-6.