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MACROSCOPIC, ETHNOMEDICINAL STUDY, PHOTOLUMINESCENCE AND PHYTOCHEMICAL SCREENING OF BARK OF PADMAK (*Prunus cerasoides)*

[1]Kanika*, [1]Chinu Kumari, [1]Dev Prakash Dahiya, [1]Rahul Sharma, [1]Nikhil Rana, [1]Vishakha Patial

Research Scholar, Associate professor, Professor, Research Scholar, Research Scholar, Research Scholar

Department of Pharmaceutics, School of Pharmacy

Abhilashi University, Chail-Chowk, Mandi, Himachal Pradesh, India

ABSTRACT

The Himalayan cherry tree, or Padmak (*Prunus cerasoides* D. Don), is an herbal drug with important ethno-botanical and therapeutic properties. Its range of distribution is limited to the sub-montane and montane Himalaya, which stretches from North Central India's Himachal Pradesh to Sikkim, Nepal, Bhutan, Myanmar, West China and Thailand. It is widely distributed in Uttarakhand state's temperate Garhwal Hills. Plants have a variety of chemical compounds that give them therapeutic qualities. This is probably the reason that different regions of the world have utilized plants in traditional medicine. Bark of *Prunus cerasoides* may be utilized to mend broken bones. The bark is brownish-grey, smooth, and peels off in thin, gleaming horizontal stripes, revealing a copper-colored surface beneath. The kernel can be used to treat kidney stones and bladder gravel. The smaller branches are crushed and soaked in water before being ingested internally to prevent abortion. Its stem has refrigerant and antipyretic qualities and can be used to treat leukaemia, leprosy and vomiting. Among its many benefits are diuretic, depurative, carminative, expectorant, analgesic, febrifuge, and tonic properties.

Index Terms-*Prunus cerasoides* D. Don, Macroscopy, Photoluminescence, Phytochemical screening, Extraction, Maceration, Soxhlet Extraction.

INTRODUCTION

The Padmak or paja, *Prunus cerasoides* belongs to Rosaceae family. *Prunus cerasoides* is deciduous tree present in temperate Himalayan regions. P. cerasoides is found in temperate forests all over the Himalayas, from Nepal, Sikkim, Bhutan, Myanmar, West China, and Thailand to Himachal Pradesh in north-central India.(1) In India, the plant is restricted to the 1500–2400 m (3,900–7,900 ft) Himalayan sub-montane and montane ranges.(2) It is grown in large quantities in the Dhanolti region as well as the temperate zones of Uttarakhand's Garhwal Hills, which include the districts of Pauri, Tehri, Chamoli, and Uttarkashi.(3) A medium to large tree with smooth, brown bark that peels off in horizontal sections to reveal a gleaming copper colored surface.(4) The bark is brownish-grey, smooth, and peels off in thin, gleaming horizontal stripes, revealing a copper-colored surface beneath. Heart wood is reddish brown, closely grained,

moderately hard and strong, durable, and seasons well. Sap wood is whitish and lustrous. It is resistant to fungus and insect attack. Stipules are long, glandular, and fringed.(5) The stipules are delicate. Fruits have insufficient pulp and are rarely eaten, are famous for producing the well-known cherry brandy. Leaves are membranous, ovate-lanceolate or elliptic-lanceolate, 7.5-12.5 cm long, glossy, nearly glabrous, with one or more conspicuous glands on the petiole. At the base of the petiole, there are 2-4 glands. Flowering occurs between October and November. Flowers are bisexual and appear in fascicles of rose-red color that fade to virtually white. Insects are the primary pollinators.(6)

PLANT PROFILE

Scientific Name: *Prunus cerasoides*

Common Name: Himalayan wild cherry, bird cheery and paja

Ayurveda: Padmak

Hindi: Padam (6)

Taxonomical Classification:

Kingdom: plantae

Subkingdom: Tracheobionta (vascular plant)

Infrakingdom: Streptophyta (Land plant)

Super division: Spermatophyta (Seed plant)

Division: Magnoliophyta (Flowering plants)

Subdivision: Spermatophytina (Spermatophyes)

Class: Magnoliopsida (Dicotyledons)

Subclass: Rosidae

Superorder: Rosanae

Order: Rosales

Family: Rosaceae (Rose family)

Subfamily: Amygdaloideae

Genus: *Prunus*

Subgenus: Cerasus

Species: *cerasoides* (7)

Phytogeography:

In the temperate Himalayan regions, *Prunus cerasoides* is a deciduous tree. In the Himalayas, P. cerasoides can be found in temperate forests in Nepal, Sikkim, Bhutan, Myanmar, West China, and Thailand, as well as in Himachal Pradesh in north central India.(8)

Only the Himalayan sub-montane and montane ranges, spanning 1500-2400 meters (3,900-7,900 feet), in India are home to the plant. It grows widely in the districts of Pauri, Tehri, Chamoli, and Uttarkashi as well as in the temperate zones of Uttarakhand's Garhwal Hills.(9)

Pharmacognosy:

The Himalayan wild cherry, or *Prunus cerasoides*, is a medium-sized deciduous tree. This tree has smooth, brownish-grey bark that removes in thin, glossy horizontal stripes to reveal a copper-colored surface below. Its leaves are glabrous, oblong, acuminate, and doubly serrate.(10)

Medicinal Uses:

- Cures Leprosy
- Hallucinations
- Burning of the Body
- Leukoderma(11)

MATERIAL AND METHODS

ETHNOMEDICINAL STUDIES

For collection of data regarding the medicinal uses of *Prunus cerasoides* a questionnaire was prepared. Without regard to their occupation, sex, or level of education, respondents over 30 were chosen at random.

The interviews were performed ideally at a time and in a language that the respondents could understand in order for them to communicate more effectively.

PLANT MATERIAL

Collection, Authentication and Preservation of Sample

The plant specimen was gathered at Urla in Mandi, Himachal Pradesh in its natural setting. The plant specimen was authenticated by Dr. Pankaj Sharma, Sr. Scientific Professional Himachal Pradesh State Biodiversity Board, Shimla, Himachal Pradesh Letter no.- HPSBB/274. (12)

MACROSCOPIC STUDY

Morphological characteristics:

Morphological characteristics of bark of *Prunus cerasoides* such as shape, texture, colour and peeling sections were studied as per visual observations and verified with standard taxonomical books.(13)

Organoleptic study:

Organoleptic properties such as odour, taste and touch were visually and sensory observed. The collected data was precisely recorded and documented.(14)

PREPARATION OF PLANT EXTRACT

In order to compare effectiveness of extraction methods, the Phyto-constituents of the plant's stem bark were extracted using both maceration and Soxhlet extraction techniques.(15)

Extraction:

Maceration

Six grams of the powdered stem bark was added to a separate 250 mL conical flask, to which 90 milliliters of ethanol and distilled water were added. After completely wrapping the conical flask with aluminium sheet and sealing it with a rubber stopper, the macerated samples were shaken continuously for 72 hours at 180 revolutions per minute using an electrical shaker.

Using Whatman No. 1 filer paper, the marc of each crude extract was isolated from the solvent containing the extract. The phytochemical screening of both extracts was carried out separately using various methods.(16,17)

Soxhlet Extraction

The Soxhlet extraction method was used to extract the powdered plant material with ethanol. 130 milliliters of ethanol were used to extract 10 grams of plant powder at 60 degrees in a round-bottom flask. The mixture was extracted for a full day, or longer, if necessary, until it lost its entire color.

The extract was left to evaporate at room temperature until one-third of its initial volume was reached. The extracts were refrigerated at 40 degrees Celsius and stored in vials.(18)

PHOTOLUMINESCENCE STUDIES

The fluorescence of plant powdered substance with various chemical reagents and extracts was observed under visible and ultraviolet radiations. The purpose of photoluminescence study is to provide an idea regarding chemical nature of the herbal extract and determine its chemical constituents.(19)

PHYTOCHEMICAL SCREENING

Phytochemical screening for both aqueous and ethanolic extract were studied for various phytochemicals such as alkaloids, flavonoids,(20) carbohydrates, tannins, terpenoids, steroids, reducing sugars, saponins, glycosides using coloration and precipitation reaction methods. (21)

Various Qualitative tests for different Phytochemicals:

1. Tests for alkaloids:

Mayer’s test: In a test tube, 1 mL of extract will be added. After that, 1 mL of Mayer's reagent, potassium mercuric iodide solution, will be shaken into the mixture. Alkaloids can be detected by the appearance of a whitish or cream precipitate.(22)

Dragendorff’s test: In a test tube, 1 mL of extract will be added. The mixture is going to be shaken with one millilitre of the Dragendorff's reagent (potassium bismuth iodide solution). A bright red precipitate that forms indicate the presence of alkaloids.(23)

Wagner’s test: In a test tube, 1 mL of extract will be added. Then, 1 millilitre of potassium iodide (Wagner's reagent) will be added and shaken with the mixture. Reddish-brown precipitate is a sign that alkaloids are present.(24)

2. Tests for flavonoids:

Lead acetate test: A test tube containing 1 millilitre of extract will be used to look for the presence of flavonoids. After that, shake in a few drops of lead acetate. When a yellow precipitate forms, it indicates the presence of flavonoids.(25)

Alkaline reagent test: In a test tube, 1 mL of extract will be added. A few drops of sodium hydroxide will then be added and the mixture will be shaken.

Flavonoids can be identified by the appearance of a bright yellow color that eventually becomes colorless when diluted acid is added.(26,27)

3. Tests for carbohydrates:

Molisch's test: Molisch reagent test, also called the purple ring test. Molisch reagent is a solution of naphthol in 95% ethanol.

To perform Molisch's test, take two milliliters of each of the test sugar and distilled water solutions in four separate test tubes. Then, add two drops of Molisch reagent to each tube. Place the test tube on an incline and slowly pour 1 millilitre of concentrated H_2SO_4 along the test tube wall.

Avoid combining the solution and the acid. If concentrated acid is not added gradually, the heat produced by the reaction may char the carbohydrates, resulting in the formation of a black ring.

Keep an eye out for the development of a purple ring at the interface between the acid and solution in the test tube.(28)

4. Tests for tannins:

Ferric chloride test: Around 0.5 mg of dried powdered plant extract should be added to a test tube along with 20 mL of water, boiled, and filtered. A few drops of a solution containing 0.1% ferric chloride will be added. If it's blue-black or brownish-green, there will be tannins.

Gelatin test: In a test tube, 1 mL of extract will be added. Shake well after adding the 1% gelatin solution containing sodium chloride. If white precipitate is present, it means that tannins are present.(29)

5. Tests for saponins:

Froth test: In a test tube, three milliliters (3 mL) of the extract's aqueous solution were combined with ten milliliters (mL) of distilled water. The test tube was then sealed and shaken vigorously for approximately five minutes, after which it was left to stand for thirty minutes and checked for the presence of honeycomb froth, which is a sign of saponins.(30)

6. Tests for steroids:

Salkowski reaction: After shaking the test extract with chloroform and adding concentrated H_2SO_4 along the test tube walls, a red color emerged, signifying the presence of steroids.(31)

7. Tests for reducing sugars:

Fehling's test: Transfer 1 ml of the sample into a dry test tube. As a control, place 1 millilitre of distilled water in another tube.

Fill each tube with 1 millilitre of Fehling's reagent (A and B). Place in a bath of boiling water. Watch for the formation of red precipitation.

Reddish brown ppt indicates a positive Fehling's test result (glucose, fructose, lactose).(32)

Benedict's test: A test tube is filled with about 1 millilitre of the sample. Benedict's reagent ($CuSO_4$), 2 ml (10 drops), is added to the test tube. Next, the solution is heated for three to five minutes in a bath of boiling water.

Keep an eye out for any precipitate formation or color changes in the test tube solution. The formation of green, orange, or red colors indicates the presence of reducing sugars.(33)

8. Tests for glycosides:

Legal's test: After taking one millilitre of the extract, adding the same volume of sodium nitroprusside, a tiny amount of sodium hydroxide solution, and shaking the mixture.

The formation of a pink to blood-red precipitate will indicate the presence of cardiac glycoside.(34)

9. Tests for terpenoids:

Salkowski's test: Concentrated sulfuric acid (3 ml) was cautiously added to the mixture of extract (5 ml) and chloroform (2 ml) to create a layer. The interface developed a reddish-brown coloration, indicating the presence of terpenoids.(35)

THIN LAYER CHROMATOGRAPHY

Preparation of Stationary Phase

TLC-Precoated Plates with silica gel G F254 (Fluorescent indicator) were taken on which the sample was applied 1cm above from the baseline.(36)

Activation of Pre-coated TLC plates

The TLC-Precoated placed were placed in hot air oven at temperature of 105^0C for 30 minutes.

Development of Chromatographic Chamber

The chromatographic chamber was developed using solvent system of water and methanol (1:1) in a glass container. This chromatographic chamber was pre-saturated for removal of edge effect.

Development of Chromatogram

The sample- loaded stationary plate was placed in pre-saturated chromatographic chamber such that the mobile phase or solvent system remains below the sample application spot.

The capillary action mechanism was responsible for the sample elution through the TLC-plate.(37)

Scanning and detection of the spots

The TLC-plates were air-dried. The spots were scanned and detected using UV chamber under UV-Visible radiations including both Near and Far UV radiations.

The Rf value of spots were calculated using following formula:

$$R_f = \frac{Distance\ \ travelled\ by\ spot\ from\ origin}{Distance\ travelled\ by\ solvent\ front}$$

All values were recorded precisely and documented.(38)

RESULTS AND DISCUSSION

ETHNOMEDICINAL STUDIES

Broken bones can be fixed with the bark. The kidney stone and gravel in the bladder can be treated with the kernel.

In order to prevent abortion, the smaller branches are crushed and soaked in water before being consumed internally.

Leprosy, leukoderma, and vomiting can all be treated with its stem, which also has antipyretic and refrigerant properties.

Diuretic, depurative, carminative, analgesic, expectorant, febrifuge, analgesic, and tonic qualities are among its many qualities. It can alleviate seminal weakness and stomach issues. It relieves leprosy, hallucinations, burns, erysipelas, leukoderma, vomiting, coughing, and thirst.

Treatment for sprains, wounds, ulcers, skin discoloration, diarrhoea, and cardiac debility can also benefit from it.

MACROSCOPY STUDY

Morphological characteristics:

The bark of *Prunus cerasoides* is round, blood red to brownish-grey color, smooth, and peels off in thin, gleaming horizontal stripes, revealing a copper-colored surface beneath.

Organoleptic study:

Table1: Organoleptic properties of *Prunus cerasoides* bark

Parameter	Observations
Touch/Texture	Smooth
Colour	Blood Red(outside) Dark Brown-Light Brown (inside)
Taste	Bitter/Astringent
Odour	Woody

PHOTOLUMINESCENCE STUDY

The fluorescence of plant powdered substance with various chemical reagents and extracts was observed under visible and ultraviolet radiations.

The UV radiations are further classified into two types on the basis of wavelength i.e., Short Wavelength (254nm) and Long Wavelength (365nm). The obtained results are mentioned below:

Table 2: Fluorescence characteristics of *Prunus cerasoides* bark

Sr. No.	Treatment	Visible light	UV Light 254nm (Shorter Wavelength)	UV Light 365nm (Long Wavelength)
1.	Bark Powder	Brown	Greenish-Brown	Dark Brown
2.	Bark Powder + 1N NaOH	Dark Brown-Black	Blood Red	Greenish-Black
3.	Bark Powder + 1N HCl	Pale Yellow	Green	Yellowish-Green

4.	Bark Powder + 1N H2SO4	Pale Yellow	Green	Dark Green
5.	Ethanolic extract	Yellow	Yellowish-Green	Dark Purple
6.	Aqueous extract	Pale Yellow	Green	Purple

PHYTOCHEMICAL SCREENING

Various phytochemical tests were performed by using coloration and precipitation reactions.

The obtained results are as follow:

Table3: Qualitative analysis of *Prunus cerasoides* bark in ethanolic and aqueous extract

Phytochemicals	Phytochemical Test	Ethanolic Extract	Aqueous Extract
Alkaloids			
	Mayer's test	+	+
	Dragendorff's test	+	+
	Wagner's test	+	+
Flavonoids			
	Lead acetate test	+	+
	Alkaline reagent test	+	-
Terpenoids			
	Salkowski's test	-	+
Saponins			
	Froth test	+	-
Tannins			
	Ferric chloride test	+	+
	Gelatin test	+	+
Reducing Sugars			
	Fehling's test	+	+
	Benedict's test	+	+
Steroids			
	Salkowski reaction	+	-
Glycosides			
	Legal's test	+	+
Carbohydrates			
	Molisch's test	+	+

+ = Present

- = Absent

THIN LAYER CHROMATOGRAPHY

Qualitative chromatography of ethanolic and distilled water extract was carried out using Methanol: Distilled Water (1:1) (MeOH: DW) as mobile phase.

Total 5 spots were found in chromatogram of ethanolic extract and 3 spots were observed in distilled water extract under 254nm Ultra-Violet radiation.

Table4: Rf value calculation in thin layer chromatography of *Prunus cerasoides* bark under 254nm.

Extract	Mobile Phase	Total run(cm)	Number of spots	Distance of Solvent front from origin	Distance of spot from origin (Sample application site)	Rf value
Ethanolic extract	MeOH + DW (1:1)	3	5	8	1.6 3.5 4.3 6.7 7.9	0.2 0.4375 0.5375 0.8375 0.9875
Distilled water extract	MeOH + DW (1:1)	3	3	8	1.8 4.5 6.5	0.225 0.5625 0.8125

CONCLUSION

Prunus cerasoides is a medium to large tree with smooth, brown bark that peels off in horizontal sections to reveal a gleaming copper colored surface. The bark is brownish-grey, smooth, and peels off in thin, gleaming horizontal stripes, revealing a copper-colored surface beneath. Heart wood is reddish brown, closely grained, moderately hard and strong, durable, and seasons well. Sap wood is whitish and lustrous. It is resistant to fungus and insect attack. The bark of *Prunus cerasoides* is bitter/astringent in taste with woody odour.

The photoluminescence study is carried out to determine chemical constituents or chemical nature of herbal extract. The photoluminescence study of *Prunus cerasoides* determined presence of flavones and terpenoids.

The phytochemical screening showed the presence of alkaloids, flavonoids, carbohydrates, tannin, glycosides and reducing sugars in both aqueous and ethanolic plant extracts. The aqueous plant extract showed slight presence of terpenoids whereas ethanolic plant extract showed slight presence of steroid and saponin in addition to other chemical constituents.

The thin layer chromatography of aqueous plant extract shoed 3 spots with 0.225, 0.5625, 0.8125 whereas ethanolic plant extract showed 5 spots with 0.2, 0.4375, 0.5375, 0.8375, 0.9875 with methanol and distilled water (1:1) as mobile phase under 254nm i.e., short UV radiation.

ABBREVIATIONS

TLC Thin Layer Chromatography

UV Ultraviolet

MeOH Methanol

DW Distilled water

REFERENCES

1. Cerasoides P, Don D. Agro-techniques of Selected Medicinal Plants: Volume-III.
2. Negi GCS, Dhyani PP, Environmental Information System on Himalayan Ecology. Glimpses of forestry research in the Indian Himalayan region: special issue in the international year of forest-2011. 184
3. Virupaksha Gupta K, Rishi R. Ethnopharmaco-Botanical Review of Padmaka-Prunus puddum Roxb. Vol. 1, International Journal of Ayurvedic and Herbal Medicine. 2011.
4. Kim SD, Kim M, Wu HH, Jin BK, Jeon MS, Song YS. *Prunus cerasoides* extract and its component compounds upregulate neuronal neuroglobin levels, mediate antioxidant effects, and ameliorate functional losses in the mouse model of cerebral ischemia. Antioxidants. 2022 Jan 1;11(1).
5. Tiwari C, Chubey S, Kurele R, Nautiyal R, Scholar MD. A REVIEW ON PADMAKA (*PRUNUS CERASOIDES* D. DON): DIFFERENT SPECIES AND THEIR MEDICINAL USES. An International Journal of Research in AYUSH and Allied Systems AYUSHDHARA.
6. Nino Joseph NAYCT. *Prunus cerasoides* D. Don: A Review on Its Ethnomedicinal Uses, Phytochemistry and Pharmacology. Int J Pharm Sci Rev Res, 48(1). 2018;48(1):62–69.
7. Sharma I, Tyagi M, Sharma R. A REVIEW ON PADMAKA (*PRUNUS CERASOIDES* D. DON): AN EMERGING TRADITIONAL DRUG FOR THERAPEUTIC BENEFITS. 2022
8. Dhrubajyoti S, Hiranmoy S, Jyoti TA, Zahidul M, Neelakshi S, Nipan S, et al. A detail microscopic studies and pharmacognostical evaluation of *Prunus cerasoides* D. Don (Rosaceae) leaf found in North East India (Meghalaya). Journal of Medicinal Plants. 2016;4(4):215–218.
9. Tewari B, Tewari A, Shah S, Pande N, Singh RP. Physical attributes as indicator of seed maturity and germination enhancement in Himalayan Wild Cherry (*Prunus cerasoides* D. Don.). New For (Dordr). 2011; 41:139–146.
10. Tiwari C, Chubey S, Kurele R, Nautiyal R. A review on padmaka (Prunus cerasiodes D. Don): Different species and and their medicial uses. Ayushdhara. 2017;4(1):1051–1055.
11. Sharma A, Joshi R, Kumar S, Sharma R, Rajneesh, Padwad Y, et al. *Prunus cerasoides* fruit extract ameliorates inflammatory stress by modulation of iNOS pathway and Th1/Th2 immune homeostasis in activated murine macrophages and lymphocytes. Inflammopharmacology. 2018; 26:1483–1495.
12. Techen N, Crockett SL, Khan IA, Scheffler BE. Authentication of Medicinal Plants Using Molecular Biology Techniques to Compliment Conventional Methods. Vol. 11, Current Medicinal Chemistry. 2004.
13. Tabasum S, Khare S, Jain K. MACROSCOPIC AND MICROSCOPIC EVALUATION OF ABRUS PRECATORIUS L. (GUNJA), SEEDS. Int J Pharm Sci Res [Internet]. 2017;8(6):2631–2635.
14. Shukla K, Saraf S, Saraf S. Study of macroscopic and microscopic plant characters: Assure the presence of plant constituents in traditional ayurvedic formulation. Vol. 4, Biotechnology Research Asia. 2007.
15. Abubakar A, Haque M. Preparation of medicinal plants: Basic extraction and fractionation procedures for experimental purposes. J Pharm Bioallied Sci. 2020;12(1):1.
16. Poole CF. Solvent Selection for Liquid-Phase Extraction. In: Liquid-Phase Extraction. Elsevier; 2020. 45–89.
17. Kim SD, Kim M, Wu HH, Jin BK, Jeon MS, Song YS. *Prunus cerasoides* extract and its component compounds upregulate neuronal neuroglobin levels, mediate antioxidant effects, and ameliorate functional losses in the mouse model of cerebral ischemia. Antioxidants. 2021;11(1):99.
18. Redfern J, Kinninmonth M, Burdass D, Verran J. Using Soxhlet Ethanol Extraction to Produce and Test Plant Material (Essential Oils) for Their Antimicrobial Properties. J Microbiol Biol Educ. 2014 May;15(1):45–46.
19. Kumar M, Mondal P, Borah S, Mahato K. Physico-chemical evaluation, preliminary phytochemical investigation, fluorescence and TLC analysis of leaves of the plant Lasia spinosa (Lour) Thwaites. Int J Pharm Pharm Sci. 2013;5(2):306–310.

20. Poonam V, Kumar G, Reddy CL, Jain R, Sharma S, Prasad A, et al. Chemical Constituents of the Genus Prunus and their Medicinal Properties ¥. Vol. 18, Current Medicinal Chemistry. 2011.

21. Jayaprakash A, Sangeetha R. Phytochemical screening of Punica granatum Linn. peel extracts. Journal of academia and industrial research. 2015;4(5):160–162.

22. Brazdovlcova B, Kostalova D, Tomko J, Jin HY. Isolation and identification of alkaloids from fruits of Berberis thunbergii DC. Chem zvesti. 1980; 34:259–262.

23. Craker LE. Plant alkaloids: a guide to their discovery and distribution. Routledge; 2018.

24. Bhandary SK, Bhat VS, Sharmila KP, Bekal MP. Preliminary phytochemical screening of various extracts of Punica granatum peel, whole fruit and seeds. Journal of Health and Allied Sciences NU. 2012;2(04):34–38.

25. Sharma V, Janmeda P. Extraction, isolation and identification of flavonoid from Euphorbia neriifolia leaves. Arabian Journal of chemistry. 2017;10(4):509–514.

26. Sharma V, Agarwal A, Chaudhary U, Singh M. Phytochemical investigation of various extracts of leaves and stems of Achyranthes aspera Linn. Int J Pharm Pharm Sci. 2013;5(1):317–320.

27. Joseph N, Anjum N, Tripathi YC. Phytochemical screening and evaluation of polyphenols, flavonoids and antioxidant activity of *Prunus cerasoides* D. Don leaves. J Pharm Res. 2016;10(7):502–508.

28. Elzagheid MI. Laboratory activities to introduce carbohydrates qualitative analysis to college students. World J Chem Educ. 2018;6(2):82–86.

29. Fecka I. Qualitative and quantitative determination of hydrolysable tannins and other polyphenols in herbal products from meadowsweet and dog rose. Phytochemical Analysis. 2009;20(3):177–190.

30. Oyekunle MA, Aiyelaagbe OO, Fafunso MA. Evaluation of the antimicrobial activity of saponins extract of Sorghum bicolor L. Moench. Afr J Biotechnol. 2006;5(23).

31. Kancherla N, Dhakshinamoothi A, Chitra K, Komaram RB. Preliminary Analysis of Phytoconstituents and Evaluation of Anthelminthic Property of Cayratia auriculata (In Vitro). Maedica (Bucur). 2019 Dec;14(4):350–356.

32. Al Jamal A, Al Yousef M. Phytochemical analysis of some herbal medicines. Medbiotech Journal. 2018;2(02):82–84.

33. Shubham S, Mishra R, Gautam N, Nepal M, Kashyap N, Dutta K. Phytochemical analysis of papaya leaf extract: screening test. EC Dent Sci. 2019;18(3):485–490.

34. Shaikh JR, Patil M. Qualitative tests for preliminary phytochemical screening: An overview. Int J Chem Stud. 2020;8(2):603–608.

35. Hemaia M, Motawe L, Ibrahim FM, Ibrahim ME, Mahmoud EA, Aly HF. Isolation and identification of terpenoids and sterols of Nepeta cataria L. Int J Pharmtech Res. 2015;8(10):10–17.

36. Matysik E, Woźniak A, Paduch R, Rejdak R, Polak B, Donica H. The New TLC Method for Separation and Determination of Multicomponent Mixtures of Plant Extracts. J Anal Methods Chem. 2016.

37. Santiago M, Strobel S. Thin layer chromatography. In: Methods in Enzymology. Academic Press Inc.; 2013,303–324.

38. Talukdar A Das, Choudhury MD, Chakraborty M, Dutta BK. Phytochemical screening and TLC profiling of plant extracts of Cyathea gigantea (Wall. Ex. Hook.) Haltt. and Cyathea brunoniana. Wall. ex. Hook (Cl. & Bak.). Assam University Journal of Science and Technology. 2010;5(1):70–74.