RESEARCH ARTICLE

Evaluation of phytochemical, radical scavenging and antimicrobial profile of *Pittosporum eriocarpum* royal (Agni) from Uttarakhand Region, India

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Abstract

Aim: The current study was designed to investigate total phenolic content (TPC), total flavonoid content (TFC), radical scavenging activity and antimicrobial profile of the aqueous, methanolic, ethanolic, and acetone leaf extract of *Pittosporum eriocarpum*. **Materials and Methods:** TPC was performed by the Folin–Ciocalteu method and TFC was measured by aluminum chloride assay. The free radical scavenging activity of leaf extracts of *P. eriocarpum* was ascertained by 1, 1-diphenyl-2-picrylhydrazyl assay and antimicrobial activity were studied by agar well diffusion method and were measured on the basis of the zone of inhibition (ZI) in millimeters. **Results and Discussion:** Screening of phytochemicals is a basic step in the investigation of the new potent bioactive component before huge extraction. *P. eriocarpum* leaf extracts possess significant results in terms of TPC and TFC, maximum value of TPC (544.60 ± 28.70 gallic acid equivalent [GAE] mg/g) was recorded in acetone extract while the minimum value of TPC (352.01 ± 32.4 GAE mg/g) was recorded in aqueous extract. The maximum (786.00 ± 13.75 QE mg/g) and minimum (493.30 ± 33.02 QE mg/g) values for TFC were recorded in methanolic and aqueous extracts, respectively. Radical scavenging activity was recorded to be maximum in the methanolic extract (66.91 ± 0.60%), whereas, lowest scavenging activity was recorded for aqueous extract (45.99 ± 0.7%), respectively. *P. eriocarpum* leaves extracts showed significant antimicrobial potential against three yeast strains (*Candida albicans*, *Candida glabrata*, and *Candida tropicalis*) and three bacterial strains (*Staphylococcus aureus*, *Escherichia coli*, and *Pseudomonas aeruginosa*). The maximum ZI was recorded in the methanolic extract (19.03 ± 0.47 mm) against *Candida glabrata* while minimum ZI 11.53 ± 0.41 mm for *C. tropicalis* was recorded. Methanolic extract showed maximum ZI of 14.67 ± 00.30 mm against *P. aeruginosa* while aqueous extract showed minimum ZI of 11.60 ± 00.65 mm for *P. aeruginosa,* respectively. Fluconazole (for fungal strains), ampicillin (for bacterial strains) and DMSO were used as a positive and negative control. **Conclusion:** These results may serve as the basic footsteps in the field of biomedical research, and further studies can be directed toward the investigation of novel molecules associated with its antioxidants, phytochemical properties, and antimicrobial properties.

Key words: Bacterial strains, phytochemicals, *Pittosporum eriocarpum*, radical scavenging, yeast strains

INTRODUCTION

The genus *Pittosporum* comprises 200 species of trees or shrubs, belonging to the family Pittosporaceae. They are distributed in the Western Pacific region, Southern Asia, Africa, Hawaiian Islands, and Australia. In India, nine species of *Pittosporum* were reported, among them *Pittosporum eriocarpum* is an important member and is commonly known as Agni. *P. eriocarpum* is an endemic tree species of Uttarakhand Himalaya[1,2] and is cited as vulnerable in "Red Data Book" of "Botanical Survey of India.[3]" International Union for Conservation of Nature and Natural Resources World Conservation Monitoring Centre[4] have been categorized *P. eriocarpum* as an endangered species. In Uttarakhand, it is

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reported from Sahastradhara (Dehradun), Mussoorie, and Tehri district.[5-7] It is used in Uttarakhand for fodder, fuel, wood, and for the treatments of narcotic, expectorant, bronchitis, etc.[8,9] To the best of our knowledge, it is the first report on radical scavenging and antimicrobial properties of leaf extract of *P. eriocarpum*, while preliminary investigation of phytochemicals was previously reported by Semwal *et al*.[10]

MATERIALS AND METHODS

Sample Collection

P. eriocarpum leaves were collected from Dehradun district, Uttarakhand (India), and the plant sample was identified by Dr. R.M. Painuli (Curator) and submitted to the Departmental Herbarium of Department of Botany and Microbiology at Hemwati Nandan Bahuguna Garhwal University, Srinagar, Uttarakhand, with voucher I.D. of GUH8376.

Extraction

Fresh leaves of *P. eriocarpum* were shade dried and coarsely powdered. Absolute ethanol, methanol, acetone, and water were used as solvents. The ethanolic, methanolic and acetone extracts were prepared by Soxhlet extraction technique, whereas the aqueous extract was prepared by maceration (48 h) technique. For each extraction 1:10 ratio (10 g of plant sample was added in 100 mL of each solvent) of powdered plant and solvent was used. After extraction, the extracts were filtered and centrifuged (15 min, 4000 rpm). The supernatant was collected, dried and stored at 4°C for further use.

Total Phenolic Content (TPC)

Folin–Ciocalteu reagent (FCR) assay was performed for the estimation of TPC of the extracts.[11,12] Gallic acid was used as a standard at different concentrations (0.2 mg/mL, 0.4 mg/mL, 0.6 mg/mL, 0.8 mg/mL, and 1 mg/mL) and quantification of TPC was based on gallic acid calibration curve. The extract (1 mg/mL)/standard having 150 µL of volume was mixed with 240 µL of distilled water and 150 µL of 0.25 N FCR. The solution was incubated for 3 min at room temperature in the dark, and afterward, 300 µL of 1N Na_2CO_3 was added. The solution was further incubated for 30 min. Absorbance was measured at 760 nm VS blank and the results were calculated in terms of mg of gallic acid equivalent/g (GAE mg/g) of extract.

Total Flavonoid Contents (TFC)

The TFC of the extracts was determined using aluminum chloride calorimetric method.[13] Quercetin was taken as standard at different concentrations (0.2–1 mg/mL) and quantification of TFC was based on quercetin calibration curve. 100 µL of extract (1 mg/mL)/standard was mixed with 400 µL of distilled water, 30 µL of 5% $NaNO_2$ and 10% aluminum chloride and it is then allowed to stand for 15 min. Absorbance was measured at 510 nm and TFC was expressed as mg quercetin equivalent (QE mg)/g of extract.

Radical Scavenging Activity using DPPH Assay (1, 1-Diphenyl-2-Picrylhydrazyl)

Free radical scavenging activity of *P. eriocarpum* was determined as described by Bibi *et al*.[14] with few modifications. 10 µL of extracts was mixed with 990 µL of DPPH solution and was incubated for 2 h in the dark. Ascorbic acid was used as a standard (0.2–1.0 mg/mL). Absorbance was measured at 515 nm using a spectrophotometer. The percentage scavenging effect was calculated using the equation:

$$\text{Scavenging rate (\%)} = [(A_{control} - A_{sample})/A_{control}] \times 100 \quad (1)$$

Where $A_{control}$ was the absorbance of the control (without extract) and A_{sample} was the absorbance in the presence of the extract.

Antimicrobial Activity

Microorganism strains and growth media

MTCC culture of three yeast strains naming *Candida albicans* (MTCC 3017), *Candida glabrata* (MTCC 3019), and *Candida tropicalis (MTCC 1406)* and three bacterial strains naming *Staphylococcus aureus* (MTCC 6908), *Escherichia coli* (MTCC 1698), and *Pseudomonas aeruginosa* (MTCC 4306) were, chosen and, obtained from Institute of Microbial Technology, Chandigarh, India. Yeast extract peptone dextrose (YPD) broth and YPD agar were used for maintenance of yeast strains, whereas Luria bertani (LB) broth and Mueller-Hinton agar (MHA) were used for maintaining the growth of bacterial strains. The yeast and bacterial stock culture were prepared on YPD and MHA plates and were incubated for 24 h at 28°C and 37°C, respectively. All strains were regularly sub cultured and stored at 4°C for further used.

Agar Well Diffusion Assay

In vitro antifungal and antibacterial activities were determined for the aqueous, ethanolic, methanolic, and acetone extracts of *P. eriocarpum* by agar well diffusion method as suggested by Alzoreky *et al*.[15] with some modifications. The ethanolic, methanolic, and acetone extracts were prepared in 10% of dimethyl sulfide (DMSO) whereas, the aqueous extract was prepared in water. Fluconazole (1 mg/mL) was used as a positive control against all selected fungal strains, whereas for bacterial strains ampicillin (1 mg/mL) was used as positive control. For estimation of antifungal activity YPD broth was inoculated with fungal strains *(C. albicans*, *C. glabrata*, and

C. tropicalis) and was incubated at 28°C for 24 h, whereas for antibacterial activity LB broth was inoculated with bacterial strains *(S. aureus*, *E. coli*, and *P. aeruginosa*) and was incubated at 37°C for overnight, respectively. When the fungal and bacterial cultures (OD_{600}–0.8) achieve log phase, 0.1 mL of each diluted culture (10^5 CFU/ml) was spread on respective agar (YPD or MHA) plates. Wells of 6 mm diameter were punched into plates with the help of sterilized borer. The wells were filled with either 20 µL of plant extract (5 mg/mL) or positive control or vehicle control (10% DMSO). The plates were incubated at 28°C for fungal strains and 37°C for bacterial strains or 24 h. The antimicrobial activity was evaluated by measuring the zone of inhibition (ZI) (mm) and all the experiments were performed in triplicates.

Statistical Analysis

The data was expressed as mean ± standard deviation from three individual tests performed at 3 different times.

RESULTS AND DISCUSSION

TPC and TFC

TPC and TFC contents of different extracts (ethanolic, methanolic, acetone, and aqueous) of *P. eriocarpum* were represented in terms of gallic acid and QE, respectively. TPC was observed to be highest for acetone extract (544.60 ± 28.7 GAE mg/g of extract) followed by a methanolic extract (530.20 ± 30.2 GAE mg/g of extract), ethanolic extract (416.67 ± 21.7 GAE mg/g of extract), and aqueous extract (352.01 ± 32.4 GAE mg/g of extract). Whereas, total flavonoids content was observed to be maximum for methanolic extract (786.0 ± 13.75 QE mg/g of extract) followed by acetone extract (689.3 ± 27.00 QE mg/g of extract), ethanolic extract (624.0 ± 40.30 QE mg/g of extract), and aqueous extract (493.3 ± 33.02 QE mg/g of extract), respectively. Most of the studies reported that these phytochemicals play an important role in different biological activities such as antioxidant, antimicrobial, and anticancer activities. Quantitative analysis of TPC and TFC is represented in Table 1.

Radical Scavenging Activity using the DPPH Assay

Radical scavenging activity of *P. eriocarpum* extract was determined by the DPPH assay. In this assay, maximum free radical scavenging activity was observed in the methanolic extract (66.91 ± 0.60%) followed by an ethanolic extract (64.53 ± 0.7%), acetone extract (52.21 ± 1.15%), and aqueous extract (45.99 ± 0.7%), respectively (represented in Table 1).

Antimicrobial Activity

Antibacterial and antifungal activities of *P. eriocarpum* extract were measured by calculating ZI (mm). The aqueous extract of *P. eriocarpum* showed maximum ZI against *C. glabrata* (15.97 ± 1.23 mm), followed by *C. albicans* (13.77 ± 1.57 mm) and *C. tropicalis* (13.40 ± 0.90 mm). The methanolic extract of *P. eriocarpum* showed maximum ZI against *C. glabrata* (19.03 ± 0.47 mm), followed by *C. tropicalis* (13.16 ± 0.32 mm) and *C. albicans* (12.30 ± 0.70 mm). The ethanolic extract *of P. eriocarpum* showed maximum ZI for *C. glabrata* (16.70 ± 1.21 mm), *followed* by *C. albicans* (12.70 ± 0.97 mm) *and C. tropicalis* (11.53 ± 0.41 mm). Acetone extract of *P. eriocarpum* showed maximum ZI against *C. glabrata* (16.33 ± 0.94 mm), while similar ZI was seen against *C. albicans* (12.30 ± 0.70 mm) and *C. tropicalis* (12.23 ± 0.32 mm), respectively. *P. eriocarpum* showed good results against *C. glabrata* while *C. tropicalis* showed lower ZI against the extract. Known drug (fluconazole) showed maximum ZI of 25.63 ± 0.35 mm against *C. tropicalis* and minimum ZI of 24.23 ± 1.17 mm for *C. glabrata* and 25.43 ± 0.85 mm ZI was seen against *C. albicans*. Negative control (DMSO) does not show ZI for any of the yeast strain.

For antibacterial activity, aqueous extract of *P. eriocarpum* showed maximum ZI of 14.06 ± 0.42 mm for *S. aureus*, followed by *E. coli* (12.47 ± 0.80 mm) and *P. aeruginosa* (11.60 ± 0.65 mm). Methanolic extract of *P. eriocarpum* showed maximum ZI of 14.67 ± 0.30 mm for *P. aeruginosa*, followed by *S. aureus* (13.93 ± 0.81 mm) and *E. coli* (12.40 ± 0.82 mm). Ethanolic extract of *P. eriocarpum* showed maximum ZI of 13.4 ± 0.80 mm for *E. coli* followed by *P. aeruginosa* (13.03 ± 0.37 mm) and *S. aureus* (12.07 ± 0.61 mm). Acetone extract of *P. eriocarpum* showed maximum ZI of 13.73 ± 0.32 mm for *S. aureus*, followed by *P. aeruginosa* (13.50 ± 0.55 mm) and *E. coli* (12.73 ± 0.41 mm). Ampicillin, a positive control for bacterial cultures showed maximum ZI of 29.9 ± 1.57 mm against *S. aureus* and minimum ZI of 25.66 ± 0.57 mm against *E. coli* while ZI of 26.50 ± 0.37 mm was showed against *P. aeruginosa.* Negative control (DMSO) does not show any kind of ZI for any of the bacterial strain. Antimicrobial activity of *P. eriocarpum* extracts is shown in Figures 1 and 2, respectively.

Table 1: Quantitative analysis of TPC, TFC, and DPPH assay of *P. eriocarpum* extracts

Test	Ethanol	Methanol	Acetone	Aqueous
TPC (GAE mg/g)	416.67±21.7	530.20±30.2	544.60±28.7	352.01±32.4
TFC (QuE mg/g)	624.0±40.30	786.0±13.75	689.3±27.00	493.3±33.02
DPPH (%)	64.53±0.7	66.91±0.60	52.21±1.15	45.99±0.7

TPC: Total phenolic content, TFC: Total flavonoid content, DPPH: 1, 1-diphenyl-2-picrylhydrazyl, GAE: Gallic acid equivalent, *P. eriocarpum: Pittosporum eriocarpum*

P. eriocarpum contains different types of phytochemicals, i.e., phenolics, tannins, terpenoids, flavonoids, and alkaloids.[10] These phytochemicals play an important role in the treatment of different diseases or disorders. Different reactive oxidative species are natural products and are mostly produced by living organisms.[16] These free radicals can damage essential proteins, DNA, lipids, and shows harmful effects in cell structure and cause different kind of diseases/ disorders, i.e., cancer, stroke, and diabetes. Antioxidants play an important role against free radicals and provide a defense mechanism of living cells against oxidative damage.[17,18] Phytochemical screening is significant for the isolation of natural products from medicinal plants. The Chinese Culture used several species of Pittosporaceae family for their sedative and cough-relieving effects. Several studies on *Pittosporum*, from different regions, reported the presence of phytochemicals such as saponins, triterpenoids, carotenoids, and essential oils.[19-22] The biological properties of *Pittosporum* have been mainly attributed to the presence of a number of mono and sesquiterpenes in leaves.[20]

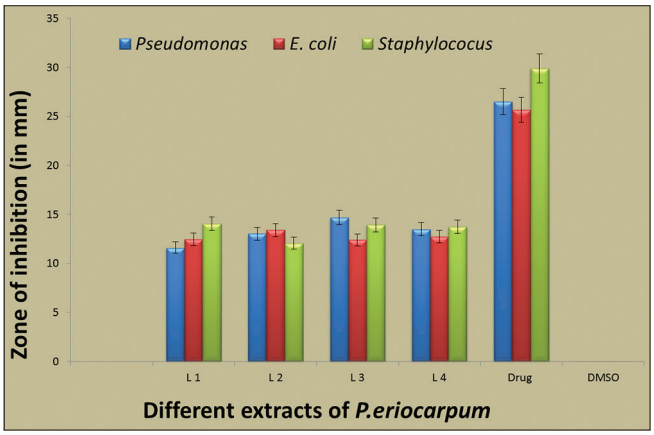

Figure 1: Antibacterial activity of different extracts of *Pittosporum eriocarpum* leaves in terms of zone of inhibition (ZI). A zone of inhibition (in mm diameter) including the diameter of well (6 mm) in agar well diffusion assay and assay was performed in triplicate, and the results are the mean of three values. L1: Indicates aqueous extract, L2: Ethanolic extract, L3: Methanolic extract, L4: Acetone extract, and drug: Ampicillin for antifungal test, respectively

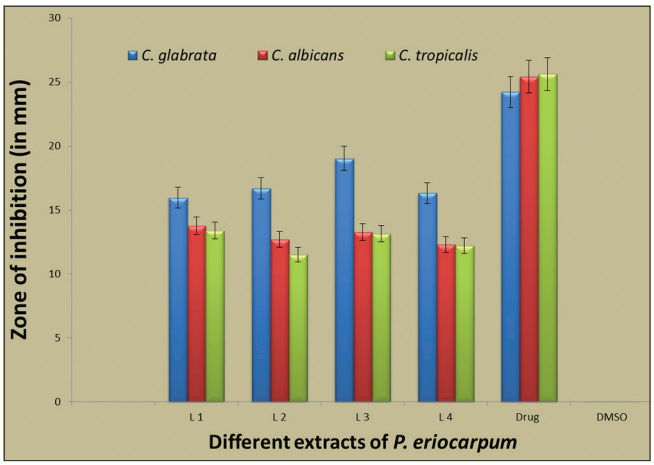

Figure 2: Antifungal activity of different extracts of *Pittosporum eriocarpum* leaves in terms of zone of inhibition (ZI). A zone of inhibition (in mm diameter) including the diameter of well (6 mm) in agar well diffusion assay and assay was performed in triplicate, and the results are the mean of three values. L1: Indicates aqueous extract, L2: Ethanolic extract, L3: Methanolic extract, L4: Acetone extract, and drug: Fluconazole for antifungal test, respectively

P. eriocarpum leaves extracts showed antimicrobial potential against selected yeast (*C. albicans, C. glabrata,* and *C. tropicalis)* and bacterial strains (*S. aureus, E. coli,* and *P. aeruginosa*). Previously, the antibacterial activity of leaves extract of *Pittosporum viridiflorum* against *Enterobacter aerogenes*, *E. coli*, *Proteus vulgaris*, *Bacillus cereus,* and *Salmonella typhi* was recorded by Swamy *et al*.[23] The extract was active against all the other organisms except *S. typhi,* and zones of inhibition were observed to be much similar to the present study. The same results of antioxidant activity of *Pittosporum mannii* Hook f. were reported by Momeni *et al*.[24] TPC, TFC, and antioxidant potential of *Pittosporum dasycaulon* Miq. were reported by Mani and Dennis.[25] They reported maximum scavenging activity in aqueous extracts than methanolic extract against the DPPH free radicals. In the present study, maximum radical scavenging activity value was recorded in the methanolic leaf extract, and minimum value was observed in aqueous leaf extracts of *P. eriocarpum*. These types of activities may depend on the quantity and composition of phytocompounds. Al-Dhab *et al*.[26] reported the detailed investigation of medicinal profile of *Pittosporum tetraspermum* and the results of the study also supports the present study.

P. eriocarpum is an endemic tree of the Himalayan region and is not been studied for radical scavenging and antimicrobial potential. Therefore, the present study highlights the first report on *P. eriocarpum* leaf extract with different solvents against antimicrobial and radical scavenging profile. Our findings showed that there is no uniform response against microbial strains in all the crude extract of *P. eriocarpum*. A small amount of extract (5 mg/mL) showed inhibitory effects against different microbial strains. Our observation also supports the ethnobotany and medicinal properties of *P. eriocarpum* and provides an opportunity to researchers to target new components and drug development and along with its conservation program.

CONCLUSION

Current findings of this research suggest that *P. eriocarpum* extract possesses radical scavenging activity and antimicrobial activity. Bioactive components of this plant species are responsible for these kinds of biological activities. Further studies are necessary to elucidate the mechanism lying behind these effects. This study may serve as a footstep in understanding the biological and pharmacological activities of leaf extract of *P. eriocarpum*.

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REFERENCES

1. Osmaston AE. Forest Flora of Kumaun. Dehradun: International Book Distributers (Reprinted 1987); 1927.
2. Kanjilal UN. Forest Flora of the Chakrata, Dehradun and Saharanpur Forest Divisions. Vol. 3. Uttar Pradesh: Managers of Publications; 1928. p. 593.
3. Nayar MP, Sastry AR. Red Data Book on Indian Plants. Calcutta: Botanical Survey of India; 1987.
4. Walter KS, Gillett HJ. IUCN Red List of Threatened Plants; 1998.
5. Singh D, Goel R. *Pittosporum eriocarpum* - An endangered species with its new distribution recorded from Tehri district. Ann For 1999;7:185-97.
6. Pundir YP, Basera H. Conservation and Propagation of Three Endemic, Endangered and Rare Plant Species of Saharanpur-Shiwaliks, Doon Valley and LOWER MAUSSOORIE FORESTS. Almora: A Project Report Submitted to G.B. Pant Institute of Himalayan Environment and Development; 2001.
7. Padalia A, Bharti RR, Pundir YP, Sharma KP. Geospatial multiple logestic regression approach for habitat characterization of scare plant population: A case study of *Pittosporum eriocarpum* Royal (An endemic species of Uttarakhand). J Indian Soc Remote Sen 2010;38:513-21.
8. Rawat VS, Rawat YS. Indigenous knowledge and sustainable development in the Tones valley of Garhwal Himalaya. J Med Plant Res 2010;4:2043-7.
9. Semwal P, Kapoor T, Anthwal P, Thapliyal A. *Pittosporum eriocarpum* (agni) endangered medicinal plant species of Uttarakhand and its conservation. Biotechnol Int 2013;6:25-30.
10. Semwal P, Anthwal P, Kapoor T, Thapliyal A. Preliminary investigation of phytochemicals of *Saussurea obvallata* (Brahm kamal) and *Pittosporum eriocarpum* (Agni) endangered medicinal plant species of Uttarakhand. Int J Pharmacogn 2014;1:266-9.
11. Fakruddin M, Mannan KS, Mazumdar RM, Afroz H. Antibacterial, antifungal and antioxidant activities of the ethanol extract of the stem bark of *Clausena heptaphylla*. BMC Complement Altern Med 2012;12:232.
12. Khan FA, Hussain I, Farroq S, Ahmad M, Arif M, Rehman I. Phytochemical screening of some Pakistanian medicinal plants. Middle East J Sci Res 2011;8:575-8.
13. Meda A, Lamien CE, Romito M, Millogo J, Nacoulma OG. Determination of the total phenolic, flavonoid and proline contents in burkina fasan honey, as well as their radical scavenging activity. Food Chem 2005;91:571-7.
14. Bibi Y, Nisa S, Chaudhary FM, Zia M. Antibacterial activity of some selected medicinal plants of Pakistan. BMC Complement Altern Med 2011;11:52.
15. Alzoreky NS, Nakahara K. Antibacterial activity of extracts from some edible plants commonly consumed in Asia. Int J Food Microbiol 2003;80:223-30.
16. Khan H, Saeed M, Muhammad N, Rauf A, Khan AZ, Ullah R. Antioxidant profile of constituents isolated from polygonatum verticillatum rhizomes. Toxicol Ind Health 2013;32:138-42.
17. Rauf A, Khan R, Muhammad N. Antioxidant studies of various solvent fractions and chemical constituents of *Potentilla evestita* Th. Wolf. Afr J Pharm Pharmacol 2013;39:2707-10.
18. Rauf A, Uddin G, Arfan M, Muhammad N. Chemical composition and biological screening of essential oils from *Pistacia integerrima*. Afr J Pharm Pharmacol 2013;7:1220-4.
19. Seo Y, Berger JM, Hoch J, Neddermann KM, Bursuker I, Mamber SW, *et al*. A new triterpene saponin from *Pittosporum viridiflorum* from the madagascar rainforest**.** J Nat Prod 2002;65:65-8.
20. Chou CH, Chen IS, Hwang TL, Wang TC, Lee TH, Cheng LY, *et al*. Phthalides from *Pittosporum illicioides* var. Illicioides with inhibitory activity on superoxide generation and Elastase release by neutrophils. J Nat Prod 2008;71:1692-5.
21. Maoka T, Akimoto N, Kuroda Y, Hashimoto K, Fujiwara Y. Pittosporumxanthins, cycloaddition products of carotenoids with α-tocopherol from seeds of *Pittosporum tobira***.** J Nat Prod 2008;71:622-7.
22. Feng C, Li BG, Gao XP, Qi HY, Zhang GL. A new triterpene and an antiarrhythmic liriodendrin from *Pittosporum brevicalyx***.** Arch Pharm Res 2010;33:1927-32.
23. Swamy TA, Obey JK, Kipngetich TE, Edwin M. *In vitro* antibacterial activity of ethanolic extract of *Pittosporum viridiflorum* leaves extract against laboratory strains of selected microorganisms. Int J Bioassays 2014;3:3388-94.
24. Momeni J, Ntchatchoua DW, Akam FM, Ngassoum MB. Antioxidant activities of some cameroonian plants extracts used in the treatment of intestinal and infectious diseases. Indian J Pharm Sci 2010;72:140-4.
25. Mani B, Dennis T. Evaluation of the antioxidant potential of *Pittosporum dasycaulon* Miq. stem bark. Food Sci Biotechnol 2014;23:539.
26. Al-Dhabi NA, Arasu MV, Rejiniemon TS. *In-vitro* antibacterial, antifungal, antibiofilm, antioxidant, and anticancer properties of isosteviol isolated from endangered medicinal plant *Pittosporum tetraspermum*. Evid Based Complement Alternat Med 2015;164261. DOI: 10.1155/2015/164261.

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