Indian Journal of Natural Sciences www.tnsroindia.org.in ©IJONS

Vol.13 / Issue 76 /February/ 2023 *International Bimonthly (Print)* *ISSN: 0976 – 0997*

RESEARCH ARTICLE

Screening of Phytochemicals and Antimicrobial Activity of Lithophytic Fern *Pyrrosia porosa* (C.Presl) Hovenkamp

D.Sowmiya[1]*, H. Rehana Banu[2], R. Divya[3], R.Rakkimuthu[4], P.Sathishkumar[5] and A.M Anandakumar[5]

[1]Ph.D Research Scholar, Department of Botany, PSGR Krishnammal College for Women, Coimbatore, Tamil Nadu, India.
[2]Assistant Professor, Department of Botany, PSGR Krishnammal College for Women, Coimbatore, Tamil Nadu, India.
[3]Msc Student, PG and Research Department of Botany, Nallamuthu Gounder Mahalingam College, Pollachi, Coimbatore, Tamil Nadu, India.
[4]Assistant Professor and Head, PG and Research, Department of Botany, Nallamuthu Gounder Mahalingam College, Pollachi, Coimbatore, Tamil Nadu, India.
[5]Assistant Professor, PG and Research, Department of Botany, Nallamuthu Gounder Mahalingam College, Pollachi, Coimbatore, Tamil Nadu, India.

Received: 23 Sep 2022 Revised: 20 Nov 2022 Accepted: 23 Dec 2022

*Address for Correspondence
D.Sowmiya,
Ph.D Research Scholar,
Department of Botany,
PSGR Krishnammal College for Women,
Coimbatore, Tamil Nadu, India.
Email: d.sowmiya22@gmail.com

ABSTRACT

The objective of the present study was to carry out phytochemical, inorganic constituents, GC-MS analysis, and antimicrobial activity of the lithophytic fern *Pyrrosia porosa* (*C.Presl*) Hovenkamp. The leaves of *Pyrrosia porosa* were subjected to preliminary phytochemical screening using four different solvents. Phytochemicals such as volatile oils, saponins, and steroids were present in petroleum ether extract. Glycosides, saponins, terpenoids, and steroids were present in chloroform extract. The ethyl acteate extract showed the presence of alkaloids, volatile oils, glycosides, resin, saponin, steroids, phenols, and flavonoids. Methanolic extract showed the presence of alkaloids, saponins, phenols, and tannin. Inorganic constituents from the chloroform leaf extract showed the presence of phosphate, nitrate and calcium. The GC-MS analysis of the chloroform extract showed 16 different compounds. The antimicrobial activity of *Pyrrosia porosa* against four bacterial and two fungal species was studied. Among four studied bacterial species, *Staphylococuus aureus* showed 09 mm zone of inhibition and a fungus, *Candida* species, showed 07 mm zone of inhibition.

Keywords: *Pyrrosia porosa*, Phytochemicals, inorganic constituents, GC-MS analysis, antimicrobial activity.

INTRODUCTION

The survival of the human race without plants on the earth is inconceivable. Since the beginning of the human race, humans have been dependent on plants. Herbal use was originally intended to produce a favourable interaction with body chemistry [1]. Traditional medicinal information is important not only for its potential contribution to drug development but also for people's healthcare. According to the World Health Organization, 80% of the world's population, mostly individuals in developing countries, depends on plant-derived medicines for their healthcare needs. Most of the aboriginal people are not well identified with the uses of pteridophytes ever since they're not simply available like flowering plants. Pteridophytes have a vital role in the earth's biodiversity [2]. The word "pteridophyta" has Greek origins. 'Ptero' means fern, 'phyta' means plant. Ferns and their ailles are one of the oldest major divisions of the Pteridophyta, the second largest group of vascular plants. They have a long fossil history on our planet in the early carboniferous period. They have been known since 380 million years ago. There are about 45,000 plant species, in which pteridophytes are represented by 144 genera and about 1200 species distributed in India [3]. Several plants or their parts have been exploited for this purpose. The value of these plants is attributed to the presence of some chemical substances that produce a definite physiological action. These substances are called phytochemicals. These phytochemicals have multiple biological functions such as antioxidant, anticancer, antimicrobial activities, *etc.* [4]. It is a well-known fact that plants produce these chemicals to protect them, but recent research demonstrates that they can also protect humans against diseases [5]. The GC-MS method for analysing plant extracts can be a useful tool for determining the amount of active principles in ferns used in cosmetics, drugs, pharmaceutical or food industry, environmental and forensic applications [6]. It combines two analytical techniques into a single method of analysing mixtures of chemical compounds. Gas chromatography separates the components of the mixture and mass spectroscopy analyses each of the components separately [7]. Since bioactive compounds occurring in plant material consist of multicomponent mixtures, their separation and determination still creates problems. Almost all of them have to be purified by the combination of several chromatographic techniques and various other purification methods to isolate bioactive compounds. Many ferns and fern allies have antimicrobial properties that could be used as alternative medicine for the treatment of various human illnesses instead of allopathic medicines. Therefore, the present study has been carried out to investigate the phytochemicals and antimicrobial properties of *Pyrrosia porosa*, a small lithophytic fern belonging to the family Polypodiaceae and distributed in abundance in the study area.

MATERIALS AND METHODS

Collection of plant materials

The plant material was collected from the Valparai Hills, Western Ghats. The collected specimen was identified and authenticated by Dr.M. Johnson, Director, Center for Biotechnology, St. Xavier's College, Palayamkottai, Trichy with reference number (CPB2097). The collected specimen was identified as *Pyrrosia porosa* (*C.Presl*) Hovenkamp. Leaves were shade dried for 30 days to remove all the moisture content and to preserve the maximum bioactive compounds. The organic solvents in the increasing order of polarity (Petroleum ether, Chloroform, Ethyl acetate, Methanol) were used to extract the powder sample of *Pyrrosia porosa* according to the method described by Harborne (1998). The sample was sequentially extracted using a soxhlet apparatus at a temperature (40–50°C) and was subjected to a test to detect the presence of different phytoconstituents.

PHYTOCHEMICAL ANALYSIS

Qualitative estimation of phytoconstituents

The petroleum ether, chloroform, ethyl acetate, and methanol extracts of *P. porosa were* tested using the standard procedures for the presence/absence of phytochemical constituents viz., alkaloids, glycosides, saponins, volatile oils, terpenoids, and resins [8]; phenolic compounds and flavonoids [9]; tannins, steroids, and anthraquinones [10].

Tests for inorganic constituents

The chloroform extract was tested for the presence and absence of the inorganic constituents *viz.*, chloride, sulphate, nitrate, carbonate, iron and calcium [11].

GC-MS Analysis

The Vellore Institute of Technology (VIT) in Chennai conducted a gas chromatography (GC) analysis. It is one of the key techniques generally used for the screening and identification of many groups of plant phytochemicals. The high attainable separation power in combination with the wide range of the detectors employing various detection principles to which it can be coupled makes GC an important, often irreplaceable tool in the analysis at trace level of plant phytochemical compounds. Gas Chromatographical study includes the important optimization processes such as introduction of the sample extract onto the GC column, separation of its components on an analytical column and detection of target analysis by using a Mass Spectrometric (MS) detector. The Clarus 680 GC was used in the analysis. It employed a fused silica column, packed with Elite-5MS (5% biphenyl, 95% dimethylpolysiloxane, 30m × 0.25mm ID × 250µm df), and the components were separated using helium as carrier gas at a constant flow of 1 ml/min. The injector temperature was set at 260°C during the chromatographic run. The 1µL of extracted sample injected into the instrument at the oven temperature was as follows: 60°C (2min); followed by 300°C at the rate of 10°C min-1; and 300°C, where it was held for 6m in. The mass detector conditions were: transfer line temperature of 240°C; ion source temperature of 240°C; and ionisation mode electron impact at 70 eV, a scan time of 0.2 sec, and a scan interval of 0.1 sec. The fragment size ranges from 40 to 600 Da. The spectra of the components were compared with the database of spectra of known components stored in the GC-MS NIST (2008) library.

Media preparation for antimicrobial activity

The materials which are required for bacterial and fungal media preparation were sterilized in an autoclave at 121°C for 15 min at 15lb pressure, and all procedures were done under Aseptic conditions.

Preparation of Agar plates

Muller Hinton agar (Hi Media) and Potato Dextrose Agar (PDA) was used for antibacterial and antifungal activity. The media was sterilized in the autoclave and was poured onto sterile petriplates and was allowed to solidify. The test organisms used for antibacterial activity were *Staphylococcus aureus*, *Bacillus spp.*, *Pseudomonas aeruginosa* and *Escherichia coli* and that for antifungal activity were *Aspergillus niger* and *Candida spp.*

Disc-diffusion method

Disc-diffusion method was adopted to study antibacterial and antifungal activity [12]. Circular disc of 5mm diameter were prepared and then sterilized in an autoclave. With a pair of sterile forceps (dipped into ethyl alcohol and flamed), a disc was picked up and placed into one sector of the plate. To the impregnated disc, leaf extract was added. The bacterial and fungal plates were then sealed and incubated in an incubator at 37°C for 24 hours and 48 hours respectively. They were then examined for the appearance of clear zone around the disc, *i.e.*, inhibition of growth of the test organisms.

RESULTS AND DISCUSSION

The results of the preliminary phytochemical analysis using different solvents such as petroleum ether, chloroform, ethyl acetate and methanol extracts of *Pyrrosia porosa* are given in Table 1. The petroleum ether extract of *P. porosa* showed the presence of three phytochemicals such as saponins, steroids, and volatile oils. The chloroform extract showed the presence of five phytochemicals such as glycosides, saponins, steroids, tannins, and terpenoids. The ethyl acetate showed positive results for alkaloids, volatile oils, glycosides, resins, saponins, steroids, phenols, and flavonoids. The methanol extract reported the presence of four phytochemicals, viz., alkaloids, tannins, saponins, and resins. Similar work was carried out by Pandian prabhakaran et *al.* [13] in six aqueous extracts of ferns, *viz.*, *Adiantum raddianum, Aspleniuma ethiopicum, Cyclosorus interruptus, Dicranopteris linearis, Diplazium polypodioides, and*

Pteridium aquilinum. A high amount of saponin was found in *C. interruptus, D. linearis,* and *P. aquilinum,* and the minimum amount of saponin was found in *A. raddianum, A. aethiopicum,* and *Diplazium polypodioides*. A minimum amount of tannins and phenols was found in all the fern extracts. Alkaloids were present in all the aqueous extracts except *A. raddianum* and *A. aethiopicum*. Shakoor *et al*. [8] evaluated the preliminary phytochemical screening of some pteridophytes from district Shopian (Jammu and Kashmir). 34 pteridophyte species were screened for phytochemical constituents. Screening was performed with acetone, ethanol, methanol and aqueous extracts of the plants that made up a total of 136 extracts. Out of 34 pteridophyte species, the aqueous extract of the ferns was found to contain flavonoids, terpenoids, saponins, and phenolic compounds.

Inorganic constituents

The chloroform extract of *Pyrrosia porosa* was subjected to the analysis of inorganic constituents, which revealed the presence of three inorganic constituents. The results are given in Table 2. The chloroform extract of *Pyrrosia porosa* was subjected to the analysis of inorganic constituents, which resulted in the presence of phosphate, nitrate and calcium, whereas chloride, sulphate, iron and carbonate were absent. The results confirm the presence of constituents which are known to exhibit medicinal as well as physiological activities. Bharti [11] reported the inorganic constituents present in the methanol leaf extract of *Lygodium flexuosum* and *Amylopteris prolifera*. The result revealed the presence of six inorganic constituents (sulphate, phosphate, potassium, iron, chloride, and calcium). In the present study, chloroform extract of *Pyrossia porosa* expressed positive results for three of the above mentioned inorganic constituents.

GC-MS Analysis

Chloroform extract of *Pyrossia porosa was* subjected to the identification of a number of compounds which were identified using mass spectrometry coupled with GC (Fig-2). Using the recorded retention time, molecular weight, molecular formula, peak area percentages, CAS number, and different compounds were identified (Table-3). The compounds identified in the chloroform extract of *Pyrrosia porosa* have important biological activities, which were illustrated in (Table -3). It showed 16 peaks majorly which indicates the presence of 16 compounds, with the retention time ranging from 2.593 to 30.519 minutes. The prevailing compounds identified with high or major peaks were observed with 25.063 area % at 2.593 retention time (RT), which mainly possessed butane, 2-ethoxy-2-methyl). The next major peak was observed with 13.631 area % at 18.620 retention time, which has 3,7,11,15-Tetramethyl-2-hexadecen-1-OL. Followed by Docosane, 2,4- dimethyl9.483 area % at 28.444(RT), Heptadecane,2,6,10,15-Tetramethyl- 6.993 area % at 17.074 (RT), Nonadecane – 6.337 area % at 19.315 (RT), Eicosane,9-octyl- 4.985 area % at 26.289 (RT), Phytol – 4.812 area % at 18.685 (RT),Nonadecane – 4.594 area % at 20.370 (RT), Hexadecanolc acid,2-hydroxy-1- (hydroxyme-methyl)ethylester- 3.938 area % at 25.127 (RT), Heptadecane,2,6,10,15-Tetramethyl- 3.774 area % at 18.205 (RT), 3,7,11,15- Tetramethyl-2-hexadecen-1-OL-3.362 area % at 19.100 (RT), Docosane, 2,4-dimethyl- 3.111 area % at 28.559 (RT), Docosane, 2,4-dimethyl- 3.073 area % at 28.289 (RT), Eicosane,9-octyl- 2.478 area % at 29.324 (RT), 3,7,11,15-Tetramethyl-2-hexadecen-1-OL2.291 area % at 18.885 (RT), Henelcosane,3-methyl- 2.074 area % at 30.519 (RT).

Similar work was done by Arockia Badhsheeba and Vadivel [14] in methanol extracts of leaves and rachis of *Acrostichu maureum*. The GC-MS chromatogram of the leaf methanol extract of *A. aureum* shows 19 peaks, indicating the presence of 19 compounds, with the retention time ranging from 3.94 to 35.77 minutes. The prevailing compound identified with a high peak was DL-phenylalanine, N-Chlorodiflouro acetyl-ethyl ester (15.75%). In all, fifteen compounds with a retention time ranging from 3.92 to 27.12 minutes were identified in the rachis of methanol extract of *A. aureum*. The component that has the highest peak is Denotonium benzoate (31.14%). GC-MS studies have been increasingly applied for the analysis of medicinal plants. This technique has proved to be a valuablemethod for the analysis. GC-MS analysis of *A. aureum*, *A. trapeziformae*, *B. orientale*, *D. linearis* and *L. flexuosum* in ethanol extract evaluated the existence of the GC-MS Chromatogram of the 11, 14, 5, 3 and 7 peaks respectively. The majorcompounds like Tetradecanoic acid, ethyl ester, n-Hexadecanoic acid, Hexadecanoic acid, ethyl ester and Octadecanoic acid, ethyl ester shows important medicinal properties [15].

Antibacterial Activity

The chloroform extract of *Pyrrosia porosa* was subjected to the evaluation of antibacterial activity using Muller-Hinton agar and PDA medium by following disc diffusion method. The result of the study was reported in Table-4. The chloroform extract of *Pyrrosia porosa* showed no zone of inhibition against *Pseudomonas aeruginosa*and minimum zone of inhibition was observed against *Staphylococcus aureus* (09 mm), *Bacillus spp* (08mm) and *Escherichia coli* (05mm). This result reveals that chloroform extract of *Pyrossia porosa* do have some antibacterial activity against three test organisms out of four studied organisms. Similar work was carried out by Manhas *et al.* [16] in the fern *Christella dentata*. Antimicrobial activities of methanolic extracts at different fractions were evaluated by the agar well diffusion method according to their zone of inhibition against *Rhodococcus pyridinivorans* NIT-36 (gram positive) and *Geobacillus stearothermophilus* MAC 1. The zone of inhibition against *Rhodococcus pyridinivorans* NIT-36 and at a concentration of 260 mg was seen to be maximum at 25 mm with respect to the standard taken as antibiotic streptomycin at 26 mm. As in the case of *Geobacillus stearothermophilus* MAC 1, it did not show any zone of inhibition at any concentration, as this strain of bacteria can tolerate extreme conditions as its habitat. Being a thermophile, it is resistant to any amount of phytochemical action.

Antifungal Activity

The chloroform extract of *Pyrrosia porosa* were subjected for the evaluation of antifungal activity in Tryptone Soya Agar medium using disk diffusion method. The result of the study is reported in Table -5. The Chloroform extract of *Pyrrosia porosa* showed no zone of inhibition against *Aspergillus niger* whereas minimum zone of inhibition was observed against *Candida spp* (07mm). This reveals that Chloroform extract of *Pyrrosia porosa* possess antifungal activity against *Candida spp*.

CONCLUSION

The present study was proposed to investigate the secondary metabolites of *Pyrrosia porosa,* a fern that belongs to the family Polypodiaceae. The leaves of *Pyrrosia porosa* were analysed for their phytochemical and inorganic constituents. Preliminary phytochemical screening reported the presence of glycosides, saponins, terpenoids, and steroids in chloroform extract. The presence of alkaloids, volatile oil, glycosides, saponins, resin, flavonoids, steroids, phenol in ethyl acetate extract. The methanol extract reported the presence of four phytochemicals, viz., alkaloids, tannins, saponins, and resins. The petroleum ether showed the extract presence of three phytochemicals, viz., volatile oil, saponins, and steroids. The analysis of inorganic constituents in the chloroform extract of *Pyrrosia porosa* contains phosphate, nitrate, and calcium. The GC-MS analysis of chloroform extract showed 16 peaks corresponding to 16 different compounds. The antibacterial and antifungal activities of *Pyrrosia porosa* chloroform extract exhibited a minimum zone of inhibition. The compound phytol identified in the GC-MS analysis possesses anti-inflammatory activity. So, this studied fern may possess anti-inflammatory activity.

CONFLICT OF INTEREST

Anthors have no conflict of interest.

ACKNOWLEDGEMENT

The authors expresses their sincere thanks to the Principal Dr. R. Muthukumaran, M.A.,M.Phil., B.Ed., Ph.D, Nallamuthu Gounder Mahalingam Collage, Pollachi for necessary facilities and infrastructure at the institution and moral support.

REFERENCES

1. Muhammad ShahzadAslam, Muhammad Syarhabil Ahmad (2016). Worldwide Importance of Medicinal plants: Current and Historical Perspectives. *Recent Advances in Biology and Medicine* 2, 88-96.

2. Pradnya N.G., Thakar S.B., Dongare M.M., Manisha V.K. (2015). Phytochemical analysis of four *Cheilanthes*species from Northern Western Ghats of India. *Research Journal of Life Sciences, Bioinformatics, Pharmaceutical and Chemical Sciences* 1(2) 92-100.
3. Chandra S., Fraser- Jenkins C.R., AlkaKumari, ArchanaSrivatava (2008). A summary of the status of threatened pteridophtes of India, Taiwania. 53(2).170.
4. Tawheed Amin, Monika Thakur (2014). A Comparitive study on Proximate Composition, phytochemical screening, antioxidant and antimicrobial activities of *Linumusitatisimum* L. (flaxseeds). *International Journal of Current Microbiology and Applied sciences* 3(4)465-481.
5. Smitha V., Vadivel V. (2019). Phytochemical screening for active compounds in*Ceratopteristhalictroides* (L.) Brogen. *Journal of Pharmacognosy and Phytochemistry* 8(3): 3556- 3559.
6. Uma B., Prabhakar K., Rajendran S., Lakshmi sarayu (2009). Studies on GC-MS of Spectroscopic analysis of some bioactive antimicrobial compounds from *Cinnamomumzeylanicum.' Journal of Medicinal Plants* 8(31) 125-131.
7. DuraisamyGomathi, Manokarankalaiselvi, GanesanRavikumar, KanakasabapathiDevaki and Chandrasekar Uma (2015). GC-MS analysis of bioactive compounds from the whole plant ethanolic extract of *Evolvulusalsinoides* (L.). *Journal of Food Sciences and Technology,* 52(2):1212-1217.
8. Shakoor A. mir, Anand K. Mishra, Zafar A.Reshi, Maheswar P. Sharma (2013). Preliminary phytochemical screening of some pteridophytes from district Shopian (J&K). *International journal of Pharmacy and Pharmaceutical sciences,* 5(14): 975-1491.
9. Kalpana Devi Rajesh, Subramani Vasantha, Nakulan Valsala Rajesh, Annamali Pannerselvam (2016). Qualitative and Quantitative Phytochemical analysis in four Pteridophytes' *International Journal of Pharmaceutical Sciences* 27(2) 408-412.
10. ShindeMahavidyalaya M.H., Tisangi Tal., Mengane S.K. (2016). Phytochemical analysis of *Adiantumlunulatum. International journal of current moicrobiology and applied sciences,* 5(11): 351-356.
11. Bharti M (2018). Studies on phytochemical analysis and Screening for cctive Compounds in some ferns of Ranchi and Latehar Districts' *International Journal of Academic Research and Development* 3(1): 33-41.
12. Azoro C. (2002). Antibacterial activity of crude extract of *Azadiracta indica* on *Salmonella typhi. World J of Biotechnol,* 3, 347-357.
13. Pandian Prabhakaran, Balakrishnan Radhakrishnan, Kodakkadal Kotian Srikumar and Bastian Suresh Kumar (2017). Efficacy of certain common ferns against red spider mite *Oligonychus coffeae* and Tea Mosquito bug *Helopeltis theivora* infesting tea. Plant Protect. Sci. 53(4): 232-242.
14. Arockia Badhsheeba, M. and Vadivel, V. (2019). Physicochemical and phytochemical constitutions of rachis of *Acrostichum aureum* L. International Journal of Current Advanced Research, 8(9): 19863-19868.
15. Manisha V Kale (2015). GC-MS analysis of phytocomponents on whole plant extract *Adiantum capillus-veneris* L.- A potential folklore medicinal plant. *Life Science informatics Publications,* 1(2): 116.
16. Shubhangi Manhas, Chandrika Attri, M.K. Seth and Amit Seth (2018). Determination of phytochemical constituents and evaluation of Antimicrobial activity of medicinal fern *Christella dentata. Indian Fern J.* 35:169-178.

Table.1: Phytochemical analysis of *Pyrrosia porosa* (C. Presl) Hovenkamp

Phytoconstituents	Petroleum ether	Chloroform	Ethyl acetate	Methanol
Alkaloids	-	-	+	+
Volatile oils	+	-	+	-
Glycosides	-	+	+	-
Tannin	-	-	-	+
Saponins	+	+	+	+
Terpenoids	-	+	-	-
Resin	-	-	+	-
Flavonoids	-	-	+	-
Steroids	+	+	+	-
Phenols	-	-	+	+
Anthraquinones	-	-	-	-

+ Present - Absent

Table 2: Inorganic constituents of *Pyrrosia porosa*

Elements	Observation
Chloride	-
Sulphate	-
Phosphate	+
Nitrate	+
Carbonate	-
Iron	-
Calcium	+

+ Present - Absent

Table 3: Compounds identified in GC-MS analysis of chloroform extract of *P.porosa*

RT	Compound name	Molecular weight	Molecular formula	Area %	CAS No.	Biological role
2.593	Butane,2-ethoxy-2- methyl	116	$C_7H_{16}O$	25.063	919-94-8	-
17.074	Heptadecane 2,6,10,15-tetramethyl-	296	$C_{21}H_{44}$	6.993	54833-48-6	-
18.205	Heptadecane 2,6,10,15-tetramethyl-	296	$C_{21}H_{44}$	3.774	54833-48-6	-
18.620	3,7,11,15- Tetramethyl-2-hexadecen-1-OL	296	$C_{20}H_{40}O$	13.631	102608-53-7	-
18.685	Phytol	296	$C_{20}H_{40}O$	4.812	150-86-7	Anti-inflammatory activity Activitantiprolife rative activity
18.885	3,7,11,15- Tetramethyl-2-hexadecen-1-OL	296	$C_{20}H_{40}O$	2.291	102608-53-7	-
19.100	3,7,11,15-Tetramethyl-2-hexadecen-1-OL	296	$C_{20}H_{40}O$	3.362	102608-53-7	-
19.315	Nonadecane	298	$C_{19}H_{40}$	6.337	629-92-5	NCL yeast anti-cancer drug screen.
20.370	Nonadecane	268	$C_{19}H_{40}$	4.594	629-92-5	NCL yeast anti-cancer drug screen
25.127	Hexadecanolc acid,2-hydroxy-1-(hydroxyme-methyl)ethylester	330	$C_{19}H_{38}O_4$	3.938	23470-00-0	-
26.568	Elcosane,9octyl	394	$C_{28}H_{58}$	4.984	13475779	NCL yeast anti-cancer drug screen
28.289	Docosane,2,4- dimethyl	338	$C_{24}H_{50}$	3.073	77536-30-2	-
28.444	Docosane,2,4- dimethyl	338	$C_{24}H_{50}$	9.483	77536-30-2	-
28.559	Docosane,2,4- dimethyl	338	$C_{24}H_{50}$	3.111	77536-30-2	-
29.324	Eicosane,9octyl		$C_{28}H_{58}$	2.478	23475-77-9	NCLyeast

		394				anticancer drug screen
30.519	Henelcosane,3- methyl	310	$C_{22}H_{46}$	2.074	6418-47-9	-
9.081	Pentanoic acid, 2(Aminooxy)	133	$C_5H_{11}O_3N$		5699-55-8	-
20.370	Octacosane	394	$C_{28}H_{58}$	4.594	630-02-4	Antimirobial activity
20.370	Heneicosane	296	$C_{21}H_{44}$	4.594	629-94-7	Inhibits aflatoxin production, Antineoplastic
25.127	Hexadecanoyl hydrazide	270	$C_{16}H_{34}ON_2$	3.938	2619-88-7	-
28.289	2-Methyldocosane	324	$C_{23}H_{48}$	3.073	1560-81-2	-
28.289	1-Heptadecanamine	255	$C_{17}H_{37}N$	3.073	4200-95-7	NCI yeast Anticancer drug screen
28.289	1H-Tetrazol-5-amine	85	CH_3N_5	3.073	4418-61-5	NCI yeast Anticancer drug screen
25.127	1-Tridecene	182	$C_{13}H_{26}$	3.938	2437-56-1	Cell viability counter screen
25.127	3-Heptadecenal	252	$C_{17}H_{32}O$	3.938	900143-48-7	-
25.127	Octadocanoic acid,Octadecyl ester	536	$C_{36}H_{72}O_2$	3.938	2778-96-3	-
25.127	16-Hexadecanoly hydroxide	270	$C_{16}H_{34}ON_2$	3.938	2619-88-7	-
18.620	Hexadecanal	240	$C_{16}H_{32}O$	13.631	629-80-1	Antiviral activity
18.620	1-Octadecanol	250	$C_{18}H_{34}$	13.631	629-89-0	Antiviral activity, Anticancer
18.620	16-Heptadecanol	252	$C_{17}H_{32}O$	13.631	900144-57-9	-
18.620	16-Heptadecanol	252	$C_{17}H_{32}O$	13.631	900144-57-9	-
18.620	Cyclododecanol	184	$C_{12}H_{24}O$	13.631	1724-39-6	NCI yeast anticancer drug sreen
18.620	Tridecanal	198	$C_{13}H_{26}O$	13.631	10486-19-8	Nematicidal activity
18.620	Hexadecyne	222	$C_{16}H_{30}$	13.631	629-74-3	-
19.100	1-Octadecyne	250	$C_{18}H_{34}$	3.362	629-89-0	Antiviral activity, Anticancer
19.100	1-Hexadecyne	222	$C_{16}H_{30}$	3.362	629-74-3	-
19.100	1-Pentadecyne	208	$C_{15}H_{28}$	3.362	765-13-9	-
19.100	1-Tetradecyne	194	$C_{14}H_{26}$	3.362	765-10-6	-
19.100	1-Heptadecyne	236	$C_{17}H_{32}$	3.362	26186-00-5	-
19.100	1-Eicosyne	278	$C_{20}H_{38}$	3.362	765-27-5	Colorimetric assay for SAR study

18.685	3,4-dimethylcyclohexanol	128	$C_8H_{16}O$	4.812	5715-23-1	NCI yeast anticancer drug screen
9.081	2,6-Pyrazinediamine	110	$C_4H_6N_4$		41536-80-5	-
9.081	4-Nonanol	144	$C_9H_{20}O$		5932-79-6	-
29.324	2-methldocosane	324	$C_{23}H_{48}$	2.478	1560-81-2	-
29.324	Nona Hexacontanoic acid	998	$C_{69}H_{138}O_2$	2.478	40710-32-5	-
26.568	Di-N-Decylsolfone	345	$C_{20}H_{42}O_2S$	4.985	111530-37-1	Antifungal, Antimicrobial
26.568	15-Methyltriacontane	478	$C_{34}H_{70}$	4.985	900131-20-3	-
17.074	Octadecane	254	$C_{18}H_{38}$	6.993	593-45-3	NCI yeast anticancer drug screen
17.074	Heptadecane	240	$C_{17}H_{36}$	6.993	629-78-7	Antibacterial

Table 4: Antibacterial activity of *Pyrrosia porosa*

Test Organisms	Zone of inhibition in mm
Staphylococuus aureus	09mm
Bacillus spp	08mm
Pseudomonas aeruginosa	No Zone
Escherichia coli	05mm

Table. 5: Antifungal activity of *Pyrrosia porosa*

Test Organisms	Zone of inhibition in mm
Aspergillus niger	No zone
Candida spp	07mm

Figure 1: Habit of *Pyrrosia porosa*

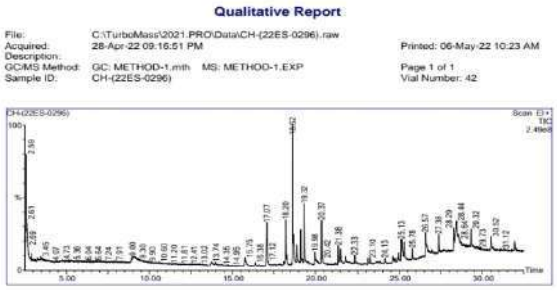

Fig. 2: Total ion chromatogram of Chloroform extract of *P.porosa*