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Evaluation of Phytochemical Profile and *In Vitro* Antioxidant, Anti-bacterial and Anti-inflammatory activity of *Piper schmidtii* Hook. fil. A Wild Edible Fruit

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Abstract

Aim: The present study aims at screening the phytochemical components and evaluates the antioxidant, anti-bacterial, and anti-inflammatory activity of fruit of *Piper schmidtii,* an endemic plant species from The Nilgiris, Tamil Nadu. **Materials and Method:** The different polar solvents such as petroleum ether, chloroform, ethyl acetate, methanol, and water were used and extraction was carried out using the soxhlet apparatus. The extracts were screened for qualitative and quantitative phytochemical analysis. The extracts of *P. schmidtii* were also subjected to *in vitro*-antioxidant activity by DPPH assay, Phosphomolybdenum assay, Ferric reducing antioxidant power (FRAP), superoxide radical scavenging activity, and reducing power assay. **Results:** Among all the extracts, methanol extract exhibited the maximum amount of phenolics (731.91 mg GAE/g extract), tannin (726.6 milligrams of Gallic acid equivalent/g extract), and ethyl acetate extract depicted the maximum quantity of flavonoids (698.17 mg QE/g extract). Methanol extract of *P. schmidtii* revealed the higher antioxidant activity in all the assays with IC_{50} values of 15.19 µg/ml (DPPH), 135.67 mg AAE/g (Phosphomolybdenum assay), 380.98 mM Fe/mg (FRAP), 60.94% (Superoxide) and higher reducing power was depicted in the ethyl acetate extract, respectively. Further anti-bacterial activity revealed that the methanol extract shows highest inhibitory activity against the tested bacterial pathogens. The methanol extract showed high degree of inhibition (71.24%) in anti-inflammatory assay. **Conclusion:** Thus, the result support that *P. schmidtii* is a potential source of natural antioxidant that can inhibit bacterial growth and subside inflammation.

Keywords: Anti-bacterial, Anti-inflammatory, Antioxidant, *In vitro* assays, *Piper schmidtii*

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Introduction

Plants are a resourceful preference with diverse assortment of organic compounds which are used to enhance human health, ever since the early civilization.[1] Most of the world's population relies on herbal medicine, where the modern medicines are exorbitant and represent the only accessible treatment. In the recent years, scientific attention toward the plant kingdom has increased considerably to bring about the experimental evidence to prove that plants are more effective and less toxic than that of synthetic drugs.[2] The plants are rich sources of secondary metabolites which not only protect the plants but they also play a pivotal role in scavenging free radicals formed in the human body.

Free radicals are unstable compounds synthesized during normal metabolism of aerobic organism, excessive free radicals are formed due to abnormal metabolism, external factors such as pollutants and radiation which results in oxidative stress. At low levels reactive species has in dispensable role in cell signaling and defense, but when the level outstrips than the normal level, it results in oxidative stress which deregulates the normal metabolism and elevates the risk of several diseases.[3,4] The etiology of several chronic disorders and other diseases are highly co-related with the damage caused by free radicals.[5] To cope up with excessive free radicals and to counteract the detrimental effects, exogenous anti-oxidants are required to prevent or delay the oxidation of biomolecules.[6] The natural antioxidants have gained more interest in the recent years since the synthetic antioxidants (BHA and BHT) are suspected and reported with high toxicity, side effects and high manufacturing cost.[7]

Piper schmidtii is a climbing shrub that belongs to the family Piperaceae. It is endemic to Southern Western Ghats. The tropical plant family Piperaceae has been used as a source of diverse medicine in many traditional medicine systems and also holds commercial and economic importance.[8] A large number of reports provide evidences in the favor of therapeutic uses of the Piperaceae family that holds the bioactive compound piperamide piperine which is used to cure various ailment.[9] The *P. schmidtii* fruits are used to cure tooth aches, head ache, cough, rheumatism, cold, bronchitis, dyspepsia, insect, or scorpion bites.[10]

Considering the medicinal activity of *P. schmidtii* based on the aforesaid traditional information, the present study was focused on the phytochemical, antioxidant, anti-bacterial, and anti-inflammatory activity of the selected plant sample to add scientific conclusion to the traditional claims.

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Materials and Methods

Collection and Identification of Plant Material

The fresh fruits of *P. schmidtii* were collected from Shola Forest of Coonoor, Tamil Nadu, India, in the month of July 2019. The plant taxonomic identification was confirmed by Botanical Survey of India, Southern Circle Coimbatore, Tamil Nadu. The surface pollutant was removed with washing under running tap water. The shade dried fruit samples were powdered for further use.

Preparation of *P. schmidtii* Fruit Extract

The *P. schmidtii* fruit powder was packed in thimbles using Whatman no.1 filter paper and extracted progressively with Petroleum ether, Chloroform, Ethyl acetate, Methanol using Soxhlet apparatus. Each time before extracting with next solvent the thimbles were air dried. Finally, the sample was macerated using hot water with constant stirring for 24 h and water extract was filtered. The different extracts were concentrated by rotary vacuum evaporator (Yamato BO410, Japan), and then air-dried extracts were weighed and collected in storage containers for further use.

The percentage of extract yield was calculated for each solvent using the formula given below,

$$\text{Extract recovery percentage} = \frac{\text{Amount of extract recoverd(g)}}{\text{Amount of fruit sample(g)}} \times 100$$

Qualitative Phytochemical Analysis

All extracts were subjected to preliminary qualitative phytochemical screening to detect the presence of carbohydrates, proteins, amino acids, alkaloids, saponins, phenols, flavanols, flavones glycosides, and phytosterols using standard methods.[11]

Quantitative Analysis of Secondary Metabolites

Determination of total phenolics and tannins

The total phenolic content of *P. schmidtii* extracts were determined using Folin–ciocalteu method.[12] Different concentration of fruit extracts (100–250 µl) was made up to 1 ml with distilled water. Distilled water served as blank. 500 µl of Folin–ciocalteu phenol reagent was added to all test tubes; 2.5 ml sodium carbonate was added and incubated in dark for 40 min. Absorbance spectra were recorded at 725 nm. To minimize standard error, thereaction was performed in triplicate and results were expressed in milligrams of Gallic acid equivalent (mg GAE).

Using the same extract tannin was also estimated, 100mg of polyvinylpolypyrrolidine was added to Eppendroff tube with 1ml distilled water and 1ml of sample and kept for incubation of 15 min at 4°C at freezer. Then, it is centrifuged at 4000 rpm for 10 min. The non-tannin phenolics containing supernatant was used to determine the tannin which calculated by

Tannin (%) = Total phenolics (%) – Non-tannin phenolics (%)

Determination of total flavonoids

The flavonoid content of different extract of fruit *P. schmidtii* was determined by Zhishen *et al*. (1999)[13] with Rutin as reference compound. Different concentration of extracts was added with 2 ml of distilled water followed by 150 µl of 5% $NaNO_2$, 150 µl of 10% $AlCl_3$ were added and the mixture was incubated for 6 min. 2 ml of 4% NaOH was added, and the volumes of the test tube were made up to 5 ml with distilled water. After 15 min of incubation, the sample with pink shadings was perused at 510 nm against the blank.

In Vitro Antioxidant Activity

DPPH radical scavenging activity

The DPPH assay was performed as described by Braca *et al*. (1985).[14] Aliquots of the samples and standard of various concentrations (20–100 µL) were made up to 100 µL with methanol and 3 ml of 0.004% methanolic solution of DPPH was added and incubated for 30 min. The solvent alone is considered as blank. The decrease in absorbance of test-mixture was determined at 517 nm; BHT and Rutin (8.8 µg/ml and 7.43µg/ml) were used as standard.

Phosphomolybdenum assay

The antioxidant activity of all the extracts of fruit of *P. schmidtii* was determined by the method of Prieto *et al.* (1999).[15] Triplicates of fruit sample are made up to 300 µl with methanol. All the triplicates were added with 3ml of reagent solution (0.6 M sulfuric acid, 28 mm sodium phosphate and 4 mM ammonium molybdate). The mixture was vortexed and incubated at 95°C for 90 min and cooled to room temperature. Absorbance was measured at 695 nm; antioxidant activity of each sample was expressed as ascorbic acid equivalent.

Ferric reducing antioxidant power (FRAP) assay

The antioxidant capacities of different extracts of fruit of *P. schmidtii* were estimated according to the procedure described by Pulido *et al.* (2000).[16] FRAP reagent (900 µL) was mixed with 90 µL of distilled water and 30 µL of test sample or methanol (blank). BHT and Rutin were used as the standards. All the test tubes were incubated at 37°C for 30 min in a water bath. At the end of incubation, the absorbance of the blue color developed was read immediately at 593 nm against the reagent blank.

Superoxide radical scavenging activity

The assay was based on the capacity of various extracts to inhibit formazan formation by scavenging the superoxide radicals generated in riboflavin light nitro blue tetrazolium (NBT) system (Beauchamo and Fridovich, 1971).[17] The scavenging activity is calculated by

$$\text{Scavenging activity}\,(\%) = \frac{\text{controlOD} - \text{SampleOD}}{\text{ControlOD}} \times 100$$

Reducing power assay

The method reported by Pulido *et al*. (2000)[16] was used to determine the reducing power of different *P. schmidtii* extracts. The aliquots of different concentration were made up to 1 ml with methanol. All the samples are added with phosphate buffer (2.5 ml, 0.2 M, ph-6.6), potassium ferric cyanide (2.5 ml 1%) and incubated for 20 min at 50°C.

then trichloroacetic acid (2.5 ml 10%) was added. After centrifugation for 10 min at 3000 rpm, 2.5 ml distilled water and 0.5 ml ferric chloride (0.1%) were mixed with 2.5 ml of aliquot supernatant and allowed to stand for 10 min and the absorbance at 700 nm was measured.

Antibacterial Activity

Agar-well diffusion method and minimum inhibitory concentration (MIC)

The various extracts of *P. schmidtii* were checked for its antibacterial activity using agar well diffusion methods. Bacterial cultures of *Bacillus subtilis, Bacillus coagulans*, *Citrobacter, Esherichia coli,* and *Pseudomonas aeruginosa* were used.

Petriplates containing 20 ml Muller Hinton agar medium were inoculated with bacterial strains enriched for 24 h. Wells were cut and 20 µl of the methanol extract of fruit *P. schmidtii.* Various concentrations of fruit (100–1000 µg/ml) were added to each well to check the MIC. The plates were then incubated overnight at 37°C. The antibacterial activity was assayed by measuring the diameter of the inhibition zone formed around the well.

In vitro anti-inflammatory activity by membrane stabilization method

The blood collected from retina of Wistar albino rats was mixed with equal volume of sterilized Alsever's solution (2% dextrose, 0.8% sodium citrate, 0.05% citric acid, 0.42% sodium chloride in distilled water). The blood was centrifuged for 10 min at 3000rpm and washed with isosaline, pensionin. The reaction mixture (4.5 ml) contained 1ml of phosphate buffer, 2 ml of hyposaline, 1 ml extract, 0.5 ml RBC). The mixture is incubated at 37°C for 30 min and centrifuged. The absorbance of the supernatant was measured at 560 nm, with phosphate buffer as blank and reaction mixture without sample as negative control.

$$\text{Percentage inhibition} = \frac{\text{absorbance of control} - \text{absorbance of treated sample}}{\text{absorbance of control}} \times 100$$

Results and Discussion

Extract Recovery Percentage

The percentage yield of *P. schmidtii* fruit extracts from different polar solvents is presented. The maximum percentage of extract was recovered from aqueous extract (10.41%) followed by the extracts of methanol (7.65%), petroleum ether (5.18%), chloroform (4.69%), and ethyl acetate (3.48%).

Polarity of solvents has a significant impact on the extraction efficacy since the solubility range of the natural components increases with increasing order of polarity.[18,19] Thus, the results reveal that high polar solvent attributes high dissolution of active compound present in *P. schmidtii* fruit might be contributed to the phenolic compounds in the extracts which have high effectiveness as antioxidants.[20,21]

Qualitative Phytochemical Analysis

Medicinal plants are intrinsic to human kind since the dawn of civilization. The phytochemicals present in the plants and plant derived products are generally non-toxic and contains many medicinal properties.[22] Plant phytochemicals have been reported to have various biological activities including antioxidant, antimicrobial, antidiabetic, antifungal, and anti-inflammatory.[23] In the same context plant extracts facilitates development of pharmaceutical drugs for the management of complex diseases.

The qualitative phytochemical screening was carried out for the various extracts of *P. schmidtii* fruit to identify the primary and secondary metabolites. The results are depicted in the Table 1. The results revealed that the *P. schmidtii* fruits are mostly affluent with the primary metabolites (carbohydrates, proteins, amino acids) and secondary metabolites (alkaloid, flavanol glycosides, phytoserol, flavonoid, cardiac glycoside, phenolic compound, and saponins). The "+" sign indicates high concentration of particular secondary metabolites which was indicated by the high intensity of the color developed and "–" sign indicates absence of chemical compound.

Quantitative Analysis of Secondary Metabolites

Determination of total phenolics, tannins and flavonoid contents of P. schmidtii fruit

The content of total phenolics was estimated based on the absorbance values of various extracts of *P. schmidtii* fruit, reacted with Folin–Ciocalteu reagent and compared with the standard of Gallic acid equivalent per g of extract (mg GAE/g extract). The total phenolic, tannin and flavonoid content of the different extracts of *P. schmidtii* are presented in Table 2. Among all the extracts studied methanol extract depicted the highest amount of phenolics (731.91 mg GAE/g extract) followed by aqueous extract (420 mg GAE/g extract). The tannins were found to be higher in the methanol extract (726.6 mg GAE/g extract). The highest level of flavonoid content was observed in the ethyl acetate extracts (698.17 mg QE/g extract) followed by methanol extract.

Phenolic compound exhibit strong antioxidant potential by chelating redox- active metal ions, inactivating lipid free radical chains and inhibit hydrogen peroxide conversion into reactive oxygen radical.[24] Flavonoids are one of the paramount group of secondary metabolites which acquires an array of pharmacological and therapeutic values.[25,26] Interestingly, the results on total phenolics, tannins and flavonoid contents of *P. schmidtii* fruit were higher in most cases than that of previously reported *Piper* species namely *Piper mullesua*,[23] *Piper wallichi,*[27] *Piper auritum*.[28] Thus, the high phenolic content in the *Piper* species might be the reason for its novel therapeutic uses.

In Vitro Antioxidant Assay

DPPH radical scavenging activity

DPPH is an easy, rapid, stable sensitive way to determine antioxidant activity. 2,2, Diphenyl-1-picrylhydrazine (DPPH) is a stable free radical, which becomes unstable in the presence of methanol (deep violet color), when it reacts with antioxidant it gets converted into 2,2, Diphenyl-1-picrylhydrazyl with discoloration.[29] DPPH radical scavenging activity of *P. schmidtii* fruit extract was examined and depicted in Figure 1. Among all the extracts, methanol extract exhibited superior IC_{50} value

Table 1: Preliminary phytochemical screening of various extracts of *Piper schmidtii* fruit

Fruit Extracts	*Petroleum ether*	*Chloroform*	*Ethyl acetate*	*Methanol*	*Water*
Carbohydrate	+	+	+	+	+
Protein	+	+	+++	+	+
Amino acid	–	–	++	++	++
Alkaloid	+	+	+++	++	++
Flavanol glycosides	+	+	++	+	+
Phytosterol	+	+	++	++	++
Flavonoid	+	+	+++	++	++
Cardiac glycoside	-	++	++	+++	+
Phenolic compound	+	+	+++	+	++
Saponins	-	++	++	+	++

(+): Presence of chemical compound, (–): Absence of chemical compound

Table 2: Total phenolic, tannins and flavonoid contents of *Piper schmidtii* fruit extracts

Extracts	*Total Phenolics (mg GAE/g extract)*	*Tannins (mg GAE/g extract)*	*Flavonoids (mg QE/g extract)*
Petroleum ether	94.29±1.57	93.22±1.02	117.12±3.03
Chloroform	89.49±2.31	88.12±3.49	103.78±4.82
Ethyl acetate	352.12±0.75	347.21±1.7	698.17±2.19
Methanol	731.91±1.57	726.6±1.53	405.89±1.82
Water	420.56±3.58	417.67±2.53	110.10±2.1

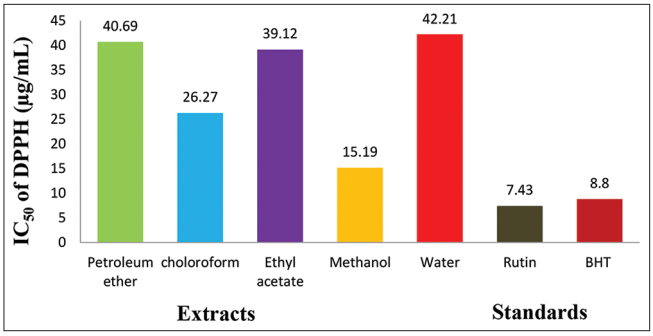

Figure 1: DPPH radical scavenging activity of *Piper schmidtii* fruit. Values are mean of triplicate determination ($n = 3$) ± standard deviation

15.19 µg/mL, the results were compared with that of Rutin and BHT. The result discloses that *P. schmidtii* fruit retains high electron donating capacity to act as an antioxidant. As for as the literatures reviewed, the radical scavenging activity of *P. schmidtii* was found to depict higher activity when compared with results disclosed by the earlier findings. The work carried out by Satyanshu *et al*., 2021[30] revealed the radical scavenging activity of four *Piper* species namely *Piper longum, Piper peepuloides, P. longum,* and *P. mullesua* were the IC_{50}values ranged between 54.52 µg/mLto 85.35µg/mL which is comparatively lower than that of the selected plant sample. The appraised antioxidant activity of *Piper cubeba*[31] exhibited similar radical scavenging activity as of *P. schmidtii*.

Phosphomolybdenum assay

The antioxidant capacity of *P. schmidtii* fruit was measured through phosphomolybdenum reduction method based on the reduction of Mo (VI) to Mo (V) and the subsequent formation of green phosphate Mo (V) with maximum absorbance at 695 nm.[32] The maximum reduction was found in methanol extract (135.67 mg AAE/g extract) followed by aqueous extract (72.53 mg AAE/g extract). The antioxidant capacity of different extract of *P. schmidtii* can be ranked in the order of methanol> water > ethyl acetate > chloroform > petroleum ether.

FRAP assay

FRAP is a relatively simple method frequently used in the evaluation of total antioxidants. This assay is based on a redox reaction in which antioxidant acts as reductants and ferric ions acts as oxidants. The principle of this method is based on the reduction of ferric-tripridyltridyltriazine to the ferrous, colored form in the presence of antioxidants.[33] The result revealed that the maximum ferric reducing capacity was found to be higher in the methanolic fruit extract 380.98 mM Fe (II)/mg extract). This reducing capacity is found to be comparable with that of standard BHT (566. 91 mM Fe (II)/mg extract) and rutin (492.83 mM Fe (II)/mg). Thus the ferric reducing power of different extracts of *P. schmidtii* reveals that there are compounds which have high affinity to the ferrous ions and thereby quench/scavenge them through redox reactions.

Superoxide radical scavenging activity

Superoxide radicals are formed as a result of cellular oxidation which results in the pathogenesis of various diseases.[34] The superoxide radical scavenging activity of different extracts of *P. schmidtii* was measured by reduction of NBT. The results of super oxide radical activities of *P. schmidtii* are shown in the Figure 2. The methanolic extract (60.94%) exhibited higher radical scavenging activity than ethyl acetate (59.06%). Thus, the result exhibit that *P. schmidtii* holds a high antioxidant properties which could be a promising remedy for radical mediated diseases by scavenging the superoxide radical.

Reducing power assay

The reducing capacity of a compound (ferricyanide complex to ferrous) by donating electron indicates its potential antioxidant activity. Figure 3 depicts the results of reducing power assay. Higher absorbance indicates the higher reducing power. Thus, the results imply that higher reducing power was evident in ethyl acetate extract of *P. schmidtii* which could act as primary and secondary antioxidants by inhibiting the lipid peroxidation.

Phenolic and flavonoids play an essential role in scavenging free radicals. Moreover, the hydroxyl group positions in phenolic groups enhance the antioxidant and antiradical activity. Besides

phenolics, flavonoids exerts direct role in scavenging ROS and counteract lipid peroxidation by donating hydrogen atom.[35,36] Thus, the result revealed that antioxidant composition in plant is strongly influenced by total amount of phenolics.

Antibacterial Activity

Agar well diffusion assay

The antibacterial screening of *P. schmidtii* fruit against *B. subtilis*, *B. coagulans, Citrobacter*, *E. coli* and *P. aeruginosa* are shown in Table 3. The methanol extract *P. schmidtii* fruit samples showed significant zone of inhibition than the other solvents against *B. coagulans* around 24.56 mm at a concentration of 100 µg/ml. The activity of solvents were in the order of methanol > ethyl acetate >water >chloroform > petroleum ether.

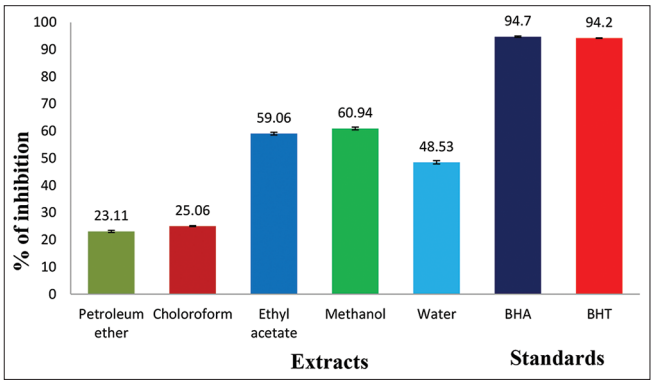

Figure 2: Superoxide radical scavenging activity of *Piper schmidtii* fruit.

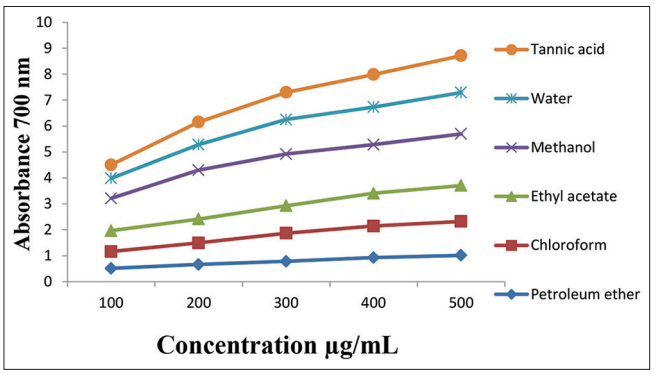

Figure 3: Reducing power assay of Piper *schmidtii fruit* extracts. Values are mean of triplicate determination (n = 3) ± standard deviation

Several studies reported that phenolic compounds play vital role in inhibitory effect against bacteria by binding with its cell wall. Thus, the antibacterial finding suggests that the *P. schmidtii* can be used effectively to treat various disease caused by bacterial pathogens.

MIC

The antimicrobial activity of the samples was determined using the MIC. MIC values of the extract against the tested bacteria were shown in Table 4. The methanol extract of the fruit sample of *P. schmidtii* showed significant activity against all the selected microorganisms. The MIC values of fruit extracts against the tested bacteria were ranging from 400 µg/mL to 1000 µg/mL (*B. coagulans, B. subtilis, E. coli*, *P. aeruginosa* and *Citrobacter*).

Among the various fruit extracts used for the antibacterial assay, methanol extracts of *P. schmidtii* showed high degree of antibacterial activity against the selected pathogens with varied rate of inhibition than the other solvent extracts. Most of the *Piper* plants are renowned for their anti-bacterial properties but the major antibacterial activity has been examined only leaves and stems of various *Piper* species. Only scanty research works are available on antibacterial activity of *Piper* fruit.[37,38]

Thus, the antibacterial findings suggest that the plant extract can be effectively used to treat the infectious disease caused by these bacteria species. However, it has to be considered that the results of antibacterial activity in this work might be used as a lead to continue the search of active substances in the extracts.

In vitro Anti-inflammatory Activity

Membrane stabilizing assay

Inflammation is a normal protective response to tissue injury alongside inflammation, it initiates the healing process.[39] During inflammation, lysosomal enzymes are released into the cytosol which affects the surrounding tissues and results in cause of several diseases. The results are expressed as percentage inhibition of extract. The results of anti-inflammatory activities of fruit extracts of *P. schmidtii* are shown in Figure 4. The investigation disclosed that the methanolic extract of *P. schmidtii* shows significant inhibition of inflammation (71.24%). The methanol extract was found to be comparable with that of standard diclofenac (92.02%).

Several studies have reported that flavonoids play important role to avert and reduce the inflammatory responses thus the results put forth that there is a correlation between the flavonoids and anti-inflammatory activity. As indicated by the previous results, the methanol extracts of *P. schmidtii* exhibited good antioxidant and anti-inflammatory property. More than a spice, *Piper* acquires desperate

Table 3: Antibacterial activity of *Piper schmidtii* fruit extracts

Extract	*Zone of inhibition (mm)*				
	Gram positive		*Gram negative*		
	Bacillus coagulans	*Bacillus subtilis*	*Citrobacter*	*Escherichia coli*	*Pseudomonas aeruginosa*
Petroleum ether	8.0±1.0^{f}	6.7±0.5^{f}	8.7±1.5^{f}	7.3±1.7^{e}	5.0±1.5^{f}
Chloroform	11.0±1.0^{e}	10.3±1.5^{e}	12.6±1.5^{e}	12.6±1.5^{d}	11.6±2.5^{e}
Ethyl acetate	18.3±1.5^{c}	18.6±1.5^{c}	19.0±2.0^{c}	20.0±2.0^{c}	21.0±0.5^{c}
Methanol	24.56±1.5^{b}	21.0±1.3^{b}	23±0.55^{b}	22.5±1.8^{a}	23.±53^{b}
Water	17.6±1.5^{d}	17.0±1.0^{d}	18.3±1.5^{d}	16.4±2.0^{c}	18.7±2.0^{d}
Control	24.1±1.0^{a}	23±1.5^{a}	24.2±1.0^{a}	22.4±2^{a}	23.3±1.7^{a}

Values are mean of triplicate determination (n=3) ± standard deviation, statistically significant at P<0.05 where $^{a}>^{b}>^{c}>^{d}$ in each column

Table 4: The minimum inhibitory concentration of m fruit extracts of *Piper schmidtii* on certain pathogenic bacteria

Organism	*Piper schmidtii fruit extract (µg/ml)*									
	100	*200*	*300*	*400*	*500*	*600*	*700*	*800*	*900*	*1000*
*Standard	+	+	+	+	+	+	+	+	+	+
Bacillus coagulans	_	_	_	_	+	+	+	+	+	+
Citrobacter	_	+	+	+	+	+	+	+	+	+
Bacillus subtilis	_	_	+	+	+	+	+	+	+	+
Escherichia coli	_	_	_	+	+	+	+	+	+	+
Pseudomonas aeruginosa	_	_	_	+	+	+	+	+	+	+
**Negativecontrol	_	_	_	_	_	_	_	_	_	_

(+) - Activity, (–) - no activity. *Tetracycline. **DMSO: Dimethyl sulfoxide

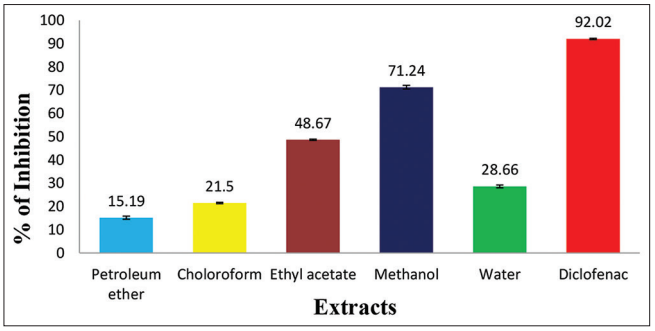

Figure 4: Membrane stabilizing assay of *Piper schmidtii* fruit extracts

therapeutic values which are used as conventional source of medicine ever since the ancient times. Several authors have congregated various pharmacological activities of *Piper* species and envisaged that the *Piper* plants play pivotal role against inflammations.[38] Thus, the result of the present study elucidated the anti-inflammatory property of *P. schmidtii* which could serve as an excellent anti-inflammatory agent analogously reducing the oxidative stress.

Conclusion

The present study onantioxidant, anti-bacterial, and anti-inflammatory activities of *P. schmidtii*, which is an endemic plant in The Nilgiris are reported for the 1st time. Accordingly, the *P. schmidtii* could offer a significant contribution for the prevention and treatment of several diseases. The present study has revealed the important pharmaceutical properties of *P. schmidtii*, and further studies are required to identify the bioactives and elucidate their pharmacological perspectives to use in therapeutics.

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