



PHYTOCHEMICAL, PHARMACOGNOSTIC AND ANTIMICROBIAL EVALUATION IN SELECTED PLANT SPECIES

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ABSTRACT

Plants are potent biochemist and have components of phytomedicine, since time immemorial. Man is able to obtain from them a wondrous assortment of industrial chemicals. The beneficial medicinal effects of plants such as healing and curing of disease is because of the presence of phytochemicals present in them. Phytochemical screening was done in selected five plant species for alkaloids, glycosides, flavonoids, phenols, steroids, tannins, etc.; histochemical study was conducted to locate the areas where these economically and biologically valuable phytochemicals were deposited, and the antimicrobial study helps to reveal the effect of these phytochemicals on selected microbes. However, depending on their solubility the action of these chemicals may vary, though some chemicals are common in all the species tested. The study revealed the presence of these biocompounds in these common plants and the reason for utilizing them in the home remedies, ethno and ayurvedic therapies.

KEY WORDS: *Mangifera indica, Artocarpus heterophyllus, Asparagus racemosus, Dioscorea oppositifolia, Plectranthus rotundifolius, secondary metabolites, antimicrobial activity.*



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INTRODUCTION

Plant based natural constituents can be derived from any part of the plants like bark, leaves, flowers, roots, fruits, seeds, etc.¹, i.e. any part of the plant may contain active components. The beneficial medicinal effects of plants such as healing and curing of disease is because of the presence of phytochemicals present in them. The medicinal plants constitute an effective source of both traditional and modern medicines, herbal medicine has been shown to have genuine utility and about 80% of rural population depends on it as a primary health care². Medicinal plants are the richest bio-resource of drugs of traditional systems of medicine, modern medicine, nutraceuticals, food supplements, folk medicines, pharmaceutical intermediates, chemical entities for synthetic drugs. Mango is one of the most popular of all tropical fruits. Mangiferin, being a polyphenolic antioxidant and a glucosyl xanthone³, has strong antioxidant^{4,5,6}, antidiabetic^{7,8}, anti-viral⁹, anthelmintic and anti allergic¹⁰, anti parasitic¹¹, antibone resorption¹², anti-tumor, anti HIV¹³, antispasmodic¹⁴, immunomodulation¹⁵, antidiarrhoeal¹⁶, anti-inflammatory¹⁷, antibacterial, antifungal¹⁸, anti microbial¹⁹, hepatoprotective²⁰ and gastroprotective²¹ activities. *Asparagus racemosus* (Shatavari) is one of the well known drugs in Ayurveda, effective in treating madhurasam, madhurvipakam, seet-veeryam, somrogam, chronic fever and internal heat²². Shatavari means, 'who possesses a hundred husbands or acceptable to many'. It is considered both a general tonic and a female reproductive tonic. The plant is known as the 'Queen of herbs', because it promotes love and devotion. Shatavari is the main ayurvedic rejuvenative tonic for the female, whereas *Withania* is for the male²³. This herb is highly effective in problems related to female reproductive system. *Charak Samhita* and *Ashtang Hridayam*, are the two main texts on

ayurvedic medicines, Shatavari as part of the formulas to treat women's health disorders²⁴. The tubers of several species of *Dioscorea* are edible and are counted just after potato for their food value. Diosgenin is a pharmacologically active component of *Dioscorea*, obtained from root and rhizome which is one of the most costly and important steroidal drugs used worldwide²⁵. Dietary plant estrogens of *Dioscorea* can provide wide range of health benefits including protection against development of some cancers, osteoporosis, cardiovascular disease, asthma, diabetes, etc., and for the production of contraceptives; and in the treatment of various genetic disorders. Jackfruit, its pulp and seeds are rich sources of several high-value compounds with potential beneficial physiological activities. The rich bioactive profile of jackfruit makes it a highly nutritious and desirable fruit crop. There are ample evidences that routine dietary supplementation with jackfruit may give protection against stomach ulcer, cardiovascular disease, development of certain cancers; in addition to protecting the health of the mouth and skin²⁶. Phytochemical studies have shown that jackfruit contains useful compounds like the flavonoids, sterols and prenylflavones which may have been responsible for the various pharmacological properties²⁷. *Plectranthus rotundifolius* tuber contains nutritional and large amount of secondary metabolites which help in improving body immune system and defense against diseases²⁸.

MATERIALS AND METHODS

Five plant species of high economic interest (Table-1) were collected from Palakkad district, Kerala. The species were identified by Prof. P. V. Madusoodanan, Department of Botany, University of Calicut. The voucher specimens were deposited in the herbarium of Govt. Victoria College, Palakkad as Coll. L. 1546-1555.

Table 1
Plant species selected for the present study

Sl.	Common name	Botanical name	Family	Part used
1	Mango	<i>Mangifera indica</i> , L.	Anacardiaceae	Stem Bark
2	Jack fruit	<i>Artocarpus heterophyllus</i> , Lam.	Moraceae	Stem Bark
3	Shathavari	<i>Asparagus racemosus</i> , Willd.	Liliaceae	Tuber
4	Dioscorea	<i>Dioscorea oppositifolia</i> , L.	Dioscoreaceae	Tuber
5	Coleus	<i>Plectranthus rotundifolius</i> , (Poir.) Spreng.	Lamiaceae	Tuber

The cleaned and shade dried materials were powdered and extraction was done as cold aqueous percolation, hot aqueous extraction (decoction), and solvent extraction in water (cold and hot), petroleum ether, ethyl acetate and methanol. After 2 hours of wetting, the conical flasks were placed at Remi rotary shaker adjusted to 100 rpm for 6 hours and filtered through Whatman No.1 filter paper. The filtrates were stored in darkness.

1. The filtrates were tested for different phytochemicals. The following tests have been done to find the presence of active chemical constituents such as alkaloids, flavonoids, steroids²⁹ and tannin³⁰ by the following methods.

- Alkaloids: Wagner's test
- Glycosides (cardiac): Keller Killiani's test
- Flavonoids: Alkaline reagent test.
- Phenolics: i). Ferric chloride test.
ii). Quinones: Acid test.
- Saponins: Foam test
- Tannins: Braymer's test
- Terpenoids: i). Salkowski test:
ii). Lieberman Burchardt test.
- Anthraquinones.
- Steroids: i). Salkowski Test.
ii). Sterol: Lieberman Burchardt test.
- Localization of phytochemicals was done in hand sections of fresh and healthy twigs treated with different stains. Depending on the phytochemicals

Histochemical studies for localization of phytochemicals (Fig.1-5, Table-3).

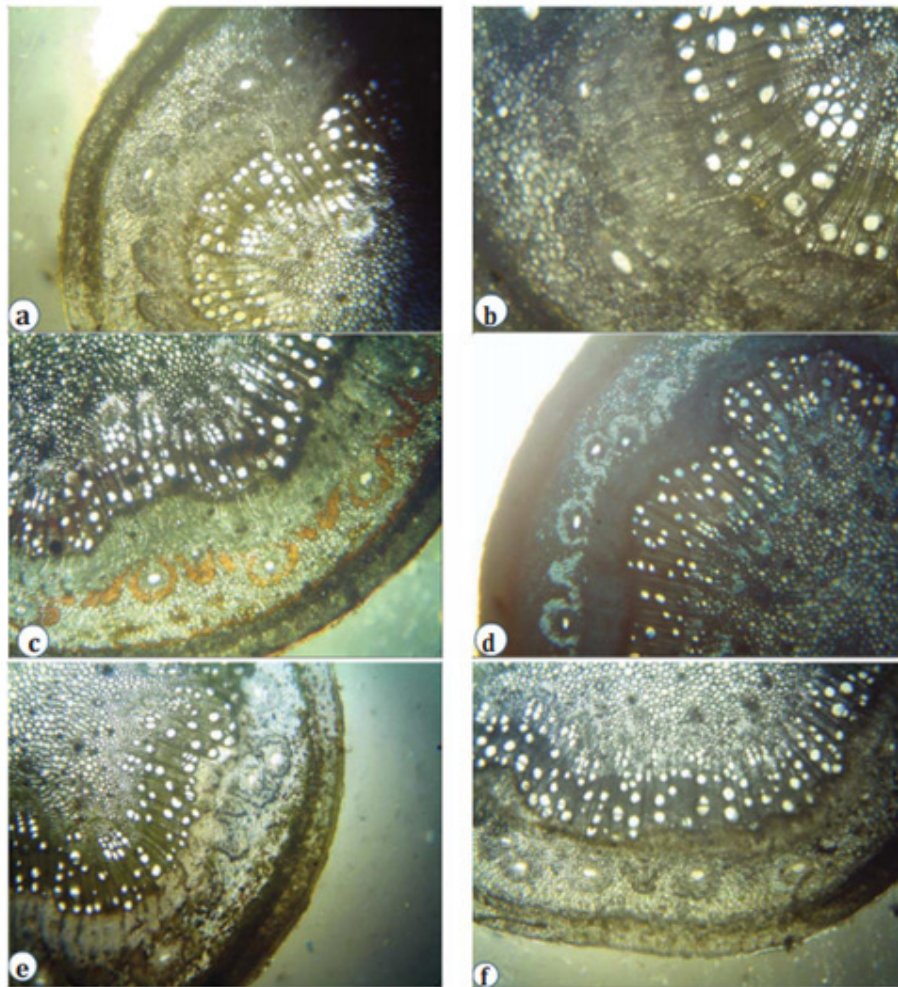
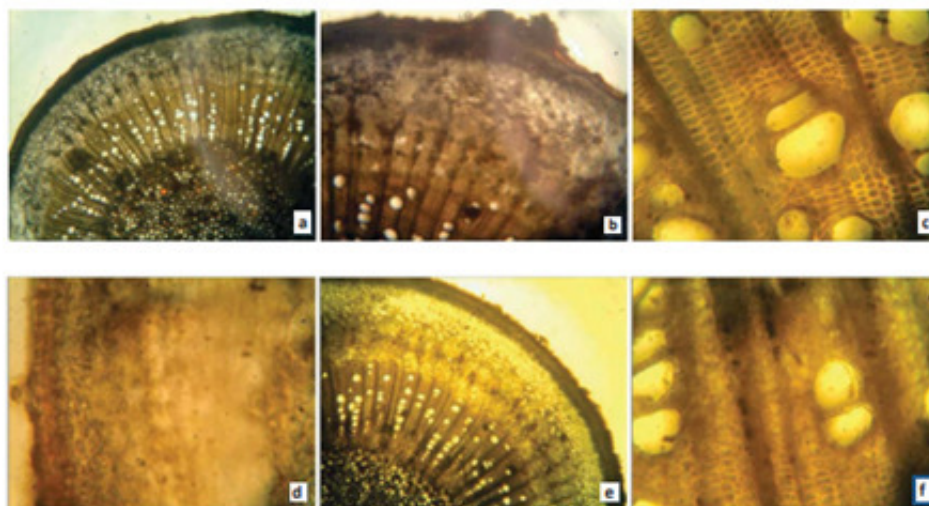


Figure 1

Histochemistry of *Mangifera indica* L. stem
a.,b.alkaloid;c.starch;d.protein



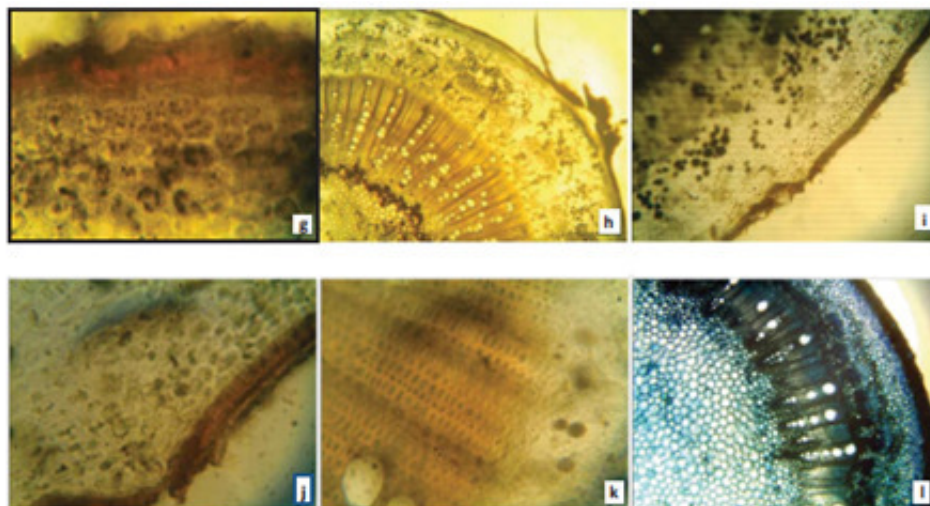


Figure 2

Histochemistry of Artocarpus heterophyllus, Lam. stem. a., b., c., d. starch; e., f., g. alkaloid; h., i., j., k saponin; i. protein

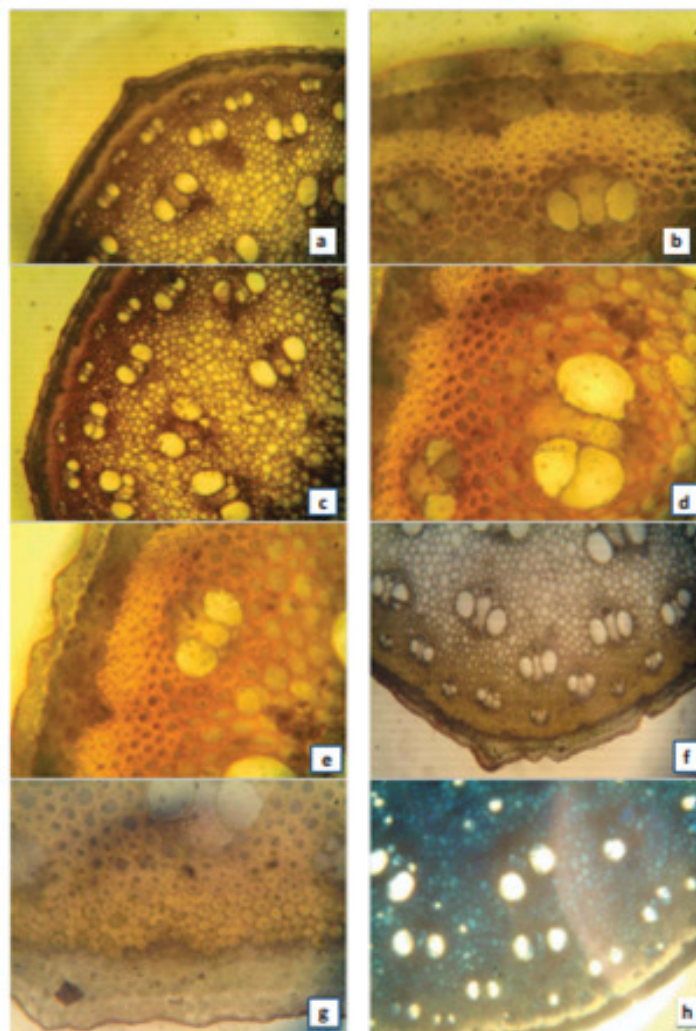


Figure 3

Histochemistry of Asparagus racemosus Wild. stem. a., b. starch; c., d., e. alkaloid; f., g. saponin; h. protein

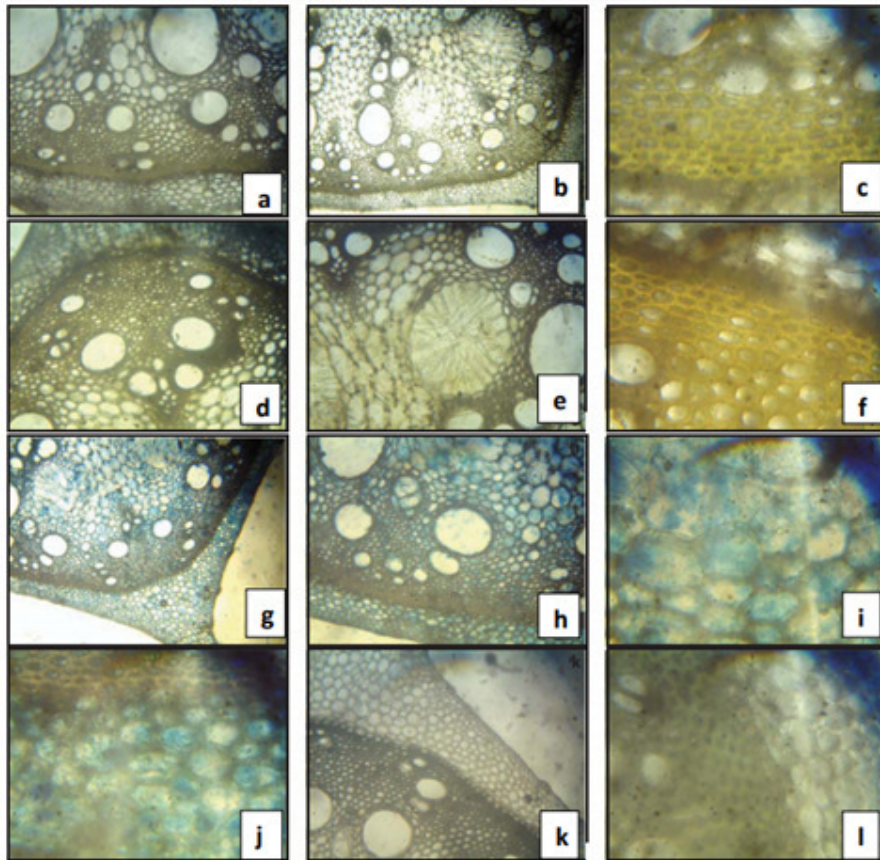
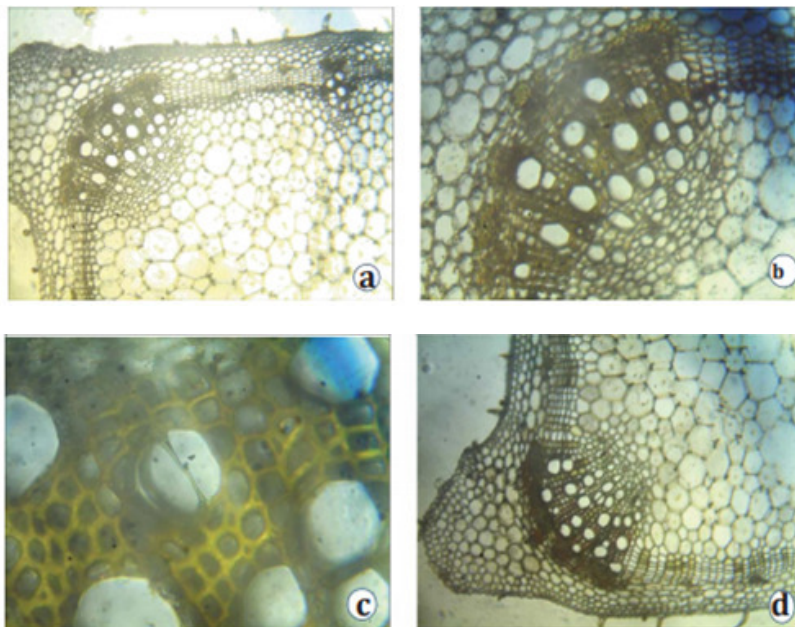


Figure 4

Histochemistry of Dioscorea oppositifolia, L. stem. a., b., c. starch; d., e., f. alkaloid; g., h., i., j. protein; k., l. saponin



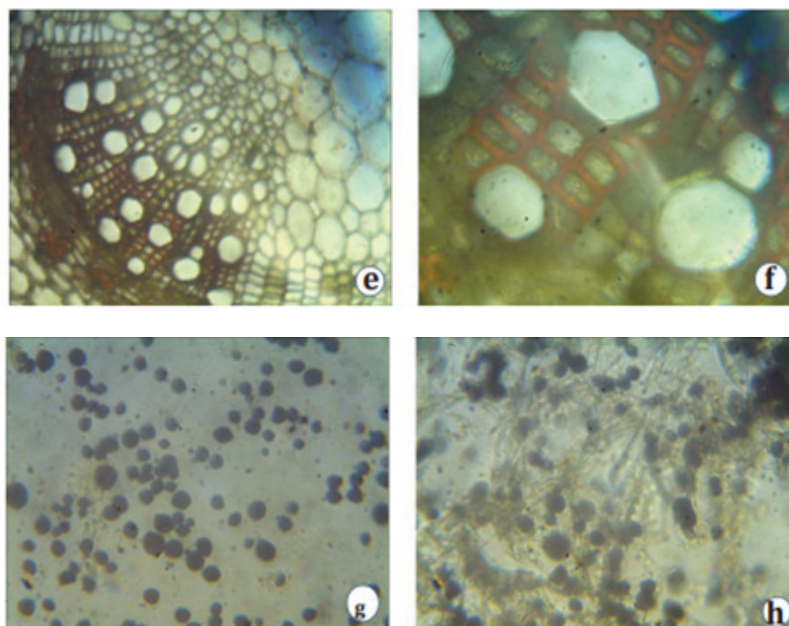


Figure 5
Histochemistry of *Plectranthus rotundifolius* Spreng, stem. a., b. alkaloid; c., d., e., f. starch; g. starch grains; h. starch grains with in the cells

Table 3
Deposition of phytochemicals in different parts of the plant body.

Biochemical	epidermis/ phellum	Phellogen	Phelloderm	cortex	endodermis/ bundle sheath	Medullary rays	phloem	Xylem	Pith
<i>Mangifera indica</i>									
Alkaloids	+	+	+	+	-	-	-	+	-
Glycosides	-	-	-	-	-	-	-	-	-
Saponins	+	+	+	+	-	-	-	+	+
Starch	+	-	-	+	-	-	-	-	-
Tannins	-	-	+	+	-	-	+	-	+
<i>Artocarpus heterophyllus</i>									
Alkaloids	+	+	+	-	-	-	-	+	-
Glycosides	-	-	-	-	-	-	-	-	-
Saponins	+	+	+	-	-	-	-	+	-
Starch	+	+	+	+	-	-	-	+	-
Tannins	-	-	-	-	-	-	-	-	-
<i>Asparagus racemosus</i>									
Alkaloids	-	-	-	+	+	-	-	-	-
Glycosides	-	-	-	-	-	-	-	-	-
Saponins	-	-	-	+	-	-	-	-	-
Starch	-	-	-	+	-	-	-	-	-
Tannins	-	-	-	-	-	-	-	-	-
<i>Dioscoreaoppositifolia</i>									
Alkaloids	-	-	-	-	-	-	+	+	+
Glycosides	-	-	-	-	-	-	-	-	-
Saponins	-	-	-	-	+	+	+	+	+
Starch	-	-	-	-	+	+	+	+	+
Tannins	-	-	-	-	-	-	-	-	-
<i>Plectranthusrotundifolius</i>									
Alkaloids	-	-	-	-	-	-	+	+	-
Glycosides	-	-	-	-	-	-	-	-	-
Saponins	-	-	-	-	-	-	-	-	-
Starch	-	-	-	+	-	-	+	+	-
Tannins	-	-	-	-	-	-	-	-	-

+ present, - absent

Antimicrobial activity**Against *Escherichia coli***

None of the aqueous extract was inhibitory to *E. coli*. Other three solvent extracts of *Mangifera indica*, L. was inhibitory to the bacteria tested. All the solvent extracts except petroleum ether in *Artocarpus heterophyllus*,

Lam. and ethyl acetate in *Dioscorea oppositifolia*, L. inhibited the growth of *E. coli*. Petroleum ether extract of *Asparagus racemosus*, Willd has a positive inhibition whereas *Plectranthus rotundifolius*, (Poir.) Spreng. did not have any inhibitory effect (Table-4).

Table 4
Antibacterial effect of different plant extracts on *Escherichia coli*.

No.	Extracts Plants	Aqueous	Ethyl acetate	Methanol	Petroleum ether
1	<i>Mangifera indica</i> , L.	-	+	+	+
2	<i>Artocarpus heterophyllus</i> , Lam.	-	+	+	-
3	<i>Asparagus racemosus</i> , Willd.	-	-	-	+
4	<i>Dioscorea oppositifolia</i> , L.	-	-	+	+
5	<i>Plectranthus rotundifolius</i> , (Poir.) Spreng.	-	-	-	-

+ present, - absent

Antifungal action against *Pyricularia oryzae*

The mycelia of *Pyricularia oryzae* showed a profuse growth in PDA medium. The modified PDA media with the plant extracts showed a differential growth pattern. *Mangifera indica*, L. extract inhibited the growth of *Pyricularia* in culture. Whereas the culture with *Asparagus racemosus*, Willd. and *Plectranthus rotundifolius*, (Poir.) Spreng. showed a profuse initial growth of mycelia; then showing secondary inhibition. *Artocarpus heterophyllus*, Lam. failed to inhibit the growth of mycelium.

DISCUSSION

Phytochemical processing is an area of engineering that is critical to the growing multimillion dollar global business of healthcare in pharmaceutical, nutraceutical, and herbal based industries. There is a growing trend of people moving from synthetic allopathic drugs to herbal cures. A key driving force is the growing knowledge of consumers about traditional medicines. The alkaloids function in the defence of plants against herbivores and pathogens, and are widely exploited as pharmaceuticals, stimulants, narcotics, and poisons due to their potent biological activities. Alkaloids have pharmacological applications as anesthetics and CNS stimulants³². Glycosides are usually administered in order to promote appetite and aid digestion. These are purely bitter principles which act on gustatory nerves, which results in increased flow of saliva and gastric juices. Some foodstuff containing cyanogenic glycosides can cause poisoning³³. Over four thousand flavonoids are known to exist and some of them are pigments in higher plants. Quercetin, kaempferol and quercitrin are common flavonoids present in nearly 70% of plants³⁴. Phenolics, phenols or polyphenolics are chemical components that occur ubiquitously as natural colour pigments responsible for the colour of fruits of plants. Their most important roles are in plant defence against pathogens and herbivore predators, and thus are applied in the control of human pathogenic infections³⁵. Phenolics essentially represent a host of natural antioxidants, used as nutraceuticals. Saponins are extremely poisonous, as they cause hemolysis of

blood and are known to cause cattle poisoning³⁶. They possess a bitter and acid taste, besides causing irritation to mucous membranes. Saponins are also important therapeutically as they are shown to have hypolipidemic and anticancer activity. Saponins are also necessary for activity of cardiac glycosides³³. Steroidal saponins are used in the commercial production of sex hormones for clinical use. The most abundant starting material for the synthesis of progesterone is diosgenin isolated from *Dioscorea* species³³. Tannins are used as antiseptic and this activity is due to the presence of phenolic group. In Ayurveda, formulations based on tannin-rich plants have been used for the treatment of diseases like leucorrhoea, rhinorrhoea and diarrhea³⁶. Terpenes are the most widespread and chemically diverse group of natural products. They are flammable, unsaturated hydrocarbons, existing in liquid form commonly found in essential oils, resins or oleoresins³⁷. Plant Steroids have an inherent ability to afford a very specific and powerful action mainly on the cardiac muscle when administered through injection into man or animal^{36,37}. Table-3 and Fig.1-5 illustrates the areas where these biochemicals get concentrated. In the case of *Asparagus racemosus*, Willd., *Dioscorea oppositifolia*, L. and *Plectranthus rotundifolius*, (Poir.) Spreng. stem sections were made instead of tubers. Epidermis/phellum, phellogen, phelloderm, cortex, endodermis/bundle sheath, medullary rays, phloem, xylem and pith stained differentially with suitable stains. Aqueous and organic solvent (ethyl acetate, methanol and petroleum ether) extract of *Mangifera indica*, L., *Artocarpus heterophyllus*, Lam., *Asparagus racemosus*, Willd., *Dioscorea oppositifolia*, L., and *Plectranthus rotundifolius*, (Poir.) Spreng. when tested on *Escherichia coli* showed that aqueous extract of all the species was not inhibitory to *E. coli*. Other three solvent extracts of *Mangifera indica* were inhibitory to the bacteria tested. All the solvent extracts except petroleum ether in *Artocarpus heterophyllus* and ethyl acetate in *Dioscorea oppositifolia* inhibited the growth of *E. coli*. Petroleum ether extracts of *Asparagus racemosus* had a positive inhibition whereas *Plectranthus rotundifolius* did not have any inhibitory effect (Table-4). All these results show that these plants can control the growth of bacteria and that may be the

reason for incorporating them in all traditional medicines. *Mangifera indica* extracts showed an extensive antifungal activity against *Pyricularia oryzae*. Extract of *Asparagus racemosus* and *Plectranthus rotundifolius* significantly reduced the mycelial growth of the tested fungi; but there was no significant difference between inhibitory effects. All these results agree that plant secondary metabolites have great potential as a source of effective antifungal agents.

CONCLUSION

The phytochemical and antimicrobial studies in the

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CONFLICT OF INTEREST

Conflict of interest declared none.

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