African Journal of Pharmacy and Pharmacology Vol. 5(9), pp. 1190-1195, 8 September, 2011
Available online http://www.academicjournals.org/AJPP
ISSN 1996-0816 ©2011 Academic Journals

Full Length Research Paper

Phytochemical analysis and antimicrobial evaluation of *Andrographis lineata* Nees leaves and stem extracts

Chinnappan Alagesaboopathi

Department of Botany, Government Arts College (Autonomous), Salem – 636 007, Tamilnadu, India.
E-mail: alagesaboopathi@gmail.com.

Accepted 8 August, 2011

The present investigation was designed to evaluate the antibacterial, antifungal properties and physiochemical screening of the various leaves and stem extracts of *Andrographis lineata*. Acetone, ethanol, methanol, petroleum ether and chloroform extracts of shade dried plant leaf and stem of *A. lineata* were tested for antibacterial, antifungal and phytochemical analysis. The antibacterial properties of various extracts of leaves and stem of *A. lineata* were assayed using the standard disc diffusion method, against five strains of bacterial species, namely, *Proteus vulgaris, Escherichia coli, Klebsiela pneumoniae, Staphylococcus aureus and Pseudomonas aeruginosa.* Among different solvent extracts studied, the methanol leaf extract showed highest antibacterial activity against *staphylococcus aureus* (19.41 mm); followed by petroleum ether extract (18.30 mm). The highest inhibition zone observed for acetone extract of *A. lineata* leaves against *P. vulgaris* was 17.40 mm followed by ethanol extract (16.10 mm). The highest activity of chloroform extract against *K. pneumoniae* was 14.07 mm. Phytochemical investigation confirmed the presence of flavonoids, saponins, gums and mucilages, triterpenoids, steroids, glycosides, phenolic compounds and tannins. The acetone leaf extracts of *A. lineata* showed highest antifungal activity against *Penicillium pinophilum* (13.40 mm). The methanol leaf extracts showed maximum activity against *Aspergillus niger* (12.15 mm). The petroleum ether leaf extracts showed highest activity against *P. pinophilum* (12.11 mm). The ethanol leaf extracts showed significant activity against *Aspergillus flavus* (11.24 mm). The chloroform leaf extracts showed higher activity against *A. niger* (9.75 mm). The leaf extract showed more inhibitory effect than the stem extracts. The present research justifies the claimed uses of this herb in the traditional system of medicine to treat different diseases.

Key words: *Andrographis lineata,* antibacterial activity, antifungal activity, phytochemicals, agar well diffusion method.

INTRODUCTION

Medicinal plants are eminent natural sources for the treatment of various ailments since primitive times. Infectious ailments are the second leading effect of death worldwide. Treatment of infections continues to be problematic in recent time because of the severe side effects of some drugs and the growing resistance to antimicrobial properties. Hence, investigation for newer, safer and more potent antimicrobial is in pressing need. Herbal medications have received much attention as a source of new antibacterial drugs since they are considered as time-tested and comparatively safe both for human use and the environment. (Faziy-Bazzaz et al., 2005) World Health Organization (WHO) recognized that medicinal plants play an important role in the health care of about 80% of the world population in developing countries and depend largely on traditional medicine (Ikegami et al., 2003). The increase in prevalence of multiple drug resistance has slowed down the progress of new synthetic antimicrobial medicines and has necessitated the investigation for new antimicrobial sources for replacement. Herbs are supposed to be safe but numerous unsafe and fatal side effects have recently been reported (Fabricant and Fansworth, 2001). Hence, there is an critical need to study the evaluation of antimicrobial activity of herbs, which will be useful in the treatment of various diseases caused by microorganisms.

Phytoconstitiuents from medicinal plants showing antimicrobial properties have the potential of filling this demand, because their structures varies from those of the more studied microbial reasons, and therefore their

mode of achievement may too very likely differ (Al-Bayati and Al-Mola, 2008). There is growing attention in correlating the phytochemical of a medicinal plant with its pharmacological activity (Chen et al., 2008; Costa et al., 2008; Kumar et al., 2004). Screening active compounds from plants regulate the discovery of modern drugs, which have efficient protection and treatment roles against many diseases including cancer (Sheeja and Kultan, 2007). The useful medicinal effects of plant materials typically effect from the combinations of secondary products present in the plant. In plants, these compounds are mostly secondary metabolites such as flavonoids, tannins, alkaloids, steroids, phenolic compound and resins fatty acids gums which are efficient of producing physiological action on body (Bishnu et al., 2009).

Species of *Andrographis* Wallich ex Nees (Acanthaceae) are used in the Indian systems of medicines such as Ayurveda, Homeopathy, Naturopathy, Amchi , Modern, Siddha and Unani and exhibit antipyretic properties (Kirtikar and Basu, 1975). This genus consists of 40 species distributed in Tropical Asia. Among them 24 species have been found mainly in the hill areas of Tamilnadu, India (Gamble, 1924), of which 18 species are reported to be endemic in India (Ahmedullah and Nayar, 1986). *Andrographis lineata* Nees is a medicinal herb (Alagesaboopathi, 1993) found wild in Shevaroy Hills of Salem District , Tamilnadu (11°45'N and 11 55' and 78°11'to 78° 20'E) with elevation up to 1600 m. Various medicinal properties such as antipyretic (Balu et al., 1993) anti-inflammatory (Balu and Alagesaboopathi, 1993) antivenom (Balu and Alagesaboopathi, 1995) anti- diabetic, jaundice , diabetes, snake bite, skin diseases and also as veterinary medicine have been attributed to this plant in the traditional system of Indian medicine (Ayyanar et al., 2008; Karuppusamy, 2007; Kadhirvel et al., 2010; Sivaperumal et al., 2010; Sangameswaran and Ilango, 2010). It is used as hepatoprotective (Sangameswaran et al., 2008; Sharma Bhawna and Sharma Upendra Kumar, 2009) antibacterial (Perumalsamy and Ignacimuthu, 2000) and diuretic (Sangameswaran et al., 2007). Three flavonoids were isolated from the leaf extract (Hari Kishore et al., 2003), therefore, it is necessary to establish the scientific basis for therapeutic action of this plant. The present research is an attempt to evaluate the phytochemical, antibacterial and antifungal activities of the leaves and stem of *A. lineata.*

MATERIALS AND METHODS

Collection and identification of plant materials

A. lineata leaves and stem were collected in July 2010 from Shevaroy Hills of Salem district of Tamilnadu, India and dried at 31°C for 15 days. The plants specimens were identified and confirmed with the Flora of Tamilnadu and voucher specimen (No.21/10.07.2010 CA) deposited in the department of Botany, Government Arts College (Autonomous), Salem for the future reference.

Preparation of plant extracts

The solvents used were acetone, ethanol, methanol, petroleum ether and chlororform. The powdered leaves and stem (25 g) were taken and extracts were prepared with Soxhlet using 150 ml of solvent. The extract was filtered through membrane filter (0.45 µm size) with the aid of a suction pump. The obtained filtrate was evaporated to dryness at 37°C. The extract was then weighed dissolved in the minimal volume of dimethyl sulphoxide (Silva et al., 1997) and used for phytochemical, antibacterical and antifungal activity.

Microorganisms used

In the experiment, we have used five bacteria (*Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus, Klebsiella pneumoniae* and *Proteus vulgaris*) and five fungi (*Penicillium pinophilum, Fusarium solani, Asoergillus flavus, A.niger* and *Alternaria alternata*). All the cultures were procured in pure form from the Biomedical Engineering Research Foundation, Salem, Tamilnadu, India.

Antibacterial assay

The agar well diffusion method was employed for the determination of antimicrobial activity of the extracts. The Petriplates containing 20 ml of Muller Hinton Agar medium were seeded with 24 h culture of the microorganisms. The wells (6 mm in diameter) were cut from the agar and the extract solution (5 mg/ml) were then added into it. The plates were incubated at 37°C for 24 h. The diameter of the inhibition zone were measured on millimeters (mm).Each experiment was performed in triplicates, repeated twice and were tabulated

Antifungal activities

The antifungal activities of *A. lineata* were proved in a radical growth inhibition properties. A fungal plug was placed in the centre of the Potato Dextrose Agar Plate. Extract (50 mg/ml) was applied into the wells .The Petriplates were incublated in the dark at 23°C. Antifungal activities were observed as a cresent shaped zone of inhibition at the mycelial form. The effect on fungal growth was expressed qualitatively. Ciprofloxacin and fluconazole were used as positive controls for bacteria and fungi, respectively.

Phytochemical screening

Phytochemical investigation was carried out on the acetone, ethanol, methanol, petroleum ether, chloroform extracts and on the powdered specimens using standard procedures to identify the phytoconstituents as described by Sofowara (1993) and Kokate et al. (2003). The leaf and stem extracts was assayed for the presence of tannins, flavonoids, triterpenoids, saponins, steroids and sterols, glycosides, phenolic compounds, gums and mucilages.

RESULTS AND DISCUSSION

The phytochemical screening of *A. lineata* investigated

Table 1. Phytochemical analysis of the leaf and stem of *Andrographis lineata.*

Phytoconstituents	Acetone extract		Ethanol extract		Methanol extract		Petroleum ether extract		Chloroform extract	
	Leaf	Stem	Leaf	Stem	Leaf	Stem	Leaf	Stem	Leaf	Stem
Tannins	+	+	+	+	+	+	+	+	+	+
Triterpenoids	+	+	+	+	+	+	+	+	+	+
Flavonoids	+	+	+	+	+	+	+	+	+	+
Saponins	+	+	+	+	+	+	+	+	+	+
Carbohydrates	-	-	-	-	-	-	-	-	-	-
Phenolic compounds	+	+	+	+	+	+	+	+	+	+
Glycosides	+	+	+	-	-	+	-	+	+	-
Gums and mucilages	+	+	+	+	+	+	+	+	+	+
Protein and Amino acids	-	-	-	-	-	-	-	-	-	-
Alkaloids	-	-	-	-	-	-	-	-	-	-
Steriods and Sterols		+	+	+	-	-	+	-	-	-

Key: +, present; -, absent

Table 2. Phytochemicals composition of *Andrographis lineata.*

Phytoconstituents	Acetone extract		Ethanol extract		Methanol extract		Petroleum ether extract		Chloroform extract	
	Leaf	Stem	Leaf	Stem	Leaf	Stem	Leaf	Stem	Leaf	Stem
Tannins (%)	6.42± 0.21	2.70± 0.15	6.10± 0.05	2.10± 0.14	6.73± 0.33	2.68± 0.07	6.27± 0.65	2.03± 0.28	7.80± 0.23	2.90± 0.50
Triterpenoids (%)	0.35± 0.13	0.20± 0.10	0.27± 0.20	0.19± 0.31	0.40± 0.08	0.15± 0.18	0.54± 0.43	0.23± 0.18	0.41± 0.29	0.27± 0.15
Flavonoids (%)	15.36± 0.43	8.65± 0.11	16.28± 0.20	7.40± 0.15	17.65± 0.28	8.86± 0.49	18.20± 0.10	7.40± 0.36	19.60± 0.55	9.20± 0.38
Saponins(%)	3.40± 0.21	1.95± 0.17	4.10± 0.41	2.10± 0.15	3.94± 0.60	1.95± 0.38	4.35± 0.70	2.05± 0.14	3.01± 0.15	1.83± 0.21
Carbohydrates (%)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Phenolic compounds (%)	0.48± 0.50	0.27± 0.17	0.53± 0.20	0.26± 0.15	0.95± 0.20	0.15± 0.08	0.98± 0.41	0.47± 0.60	0.71± 0.36	0.30± 0.11
Glycosides (%)	0.36± 0.24	0.00	0.70± 0.15	0.00	0.61± 0.20	0.42± 0.15	0.00	0.32± 0.27	0.29± 0.15	0.00
Gums and mucilages (%)	13.80± 0.53	7.78± 0.15	11.40± 0.27	9.11± 0.10	15.77± 0.60	7.20± 0.33	12.37± 0.20	8.38± 0.25	11.60± 0.31	8.27± 0.23
Protein and Amino acids (%)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Alkaloids (%)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Steriods (%)	0.61± 0.13	0.00	0.75± 0.48	0.38± 0.17	0.45± 0.15	0.00	0.00	0.00	0.29± 0.10	0.00

are presented in Tables 1 and 2. Preliminary phytochemical test for acetone, ethanol, methanol, petroleum ether and chloroform extracts of the drug were carried out. The phytochemical constituents analysis of the various extracts from the leaf and stem sample of *A. lineata* revealed the presence of phytoconstituents such as tannins, triterpenoids, flavonoids, steroids, glycosides, gums and mucilages, saponins and phenolic compounds and absence of protein and amino acids, carbohydrates and alkaloids (Table 1). The presence of these

Table 3. Antibacterial activity of the various leaf and stem extracts of *Andrographis lineata* by agar well diffusion method.

Plant part	Plant extracts	Zone of inhibition (in mm)				
		Proteus vulgaris	***Escherichia coli***	***Klebsiela pneumoniae***	***Staphylococcus aureus***	***Pseudomonas aeruginosa***
Leaves	Acetone	17.40±0.35	14.07±0.71	12.34±0.69	15.65±0.40	14.63±0.26
	Ethanol	16.10±0.28	13.20±0.60	14.11±0.11	14.36±0.13	15.30±0.25
	Methanol	-	-	16.40±0.38	19.41±0.20	16.45±0.01
	Petroleum ether	14.16±0.80	15.10±0.31	12.01±0.30	18.30±0.15	16.50±0.65
	Chloroform	13.50±0.17	13.11±0.30	14.07±0.03	12.30±0.05	13.14±0.10
Stem	Acetone	15.11±0.20	12.40±0.71	10.41±0.18	13.17±0.02	12.11±0.30
	Ethanol	12.17±0.11	13.71±0.15	8.70±0.11	13.07±0.11	-
	Methanol	-	-	13.58±0.47	15.30±0. 28	10.30±0.11
	Petroleum ether	13.20±0.18	8.40±0.21	6.40±0.17	11.47±0.18	-
	Chloroform	12.20±0.40	9.36±0.17	10.30±0.27	10.35±0.21	11.20±0.41
	Ciprofloxacin 25 µg/ml	21.0±0.19	23.0±0.40	26.0±0.63	27.0±0.11	25.0±0.27

Data given are mean of triplicates ±standard error. - indicates no activity. Concentration used 50 µg/ml.

phytochemicals suggests that the plant might be important in medical and pharmaceutical industry. Table 2 show qualitative evaluation of the percentage phytoconstituents of *A. lineata. A. lineata* has the maximum percentage yield of flavonoids (19.60%) in chloroform leaf extract. The content of gums and mucilages were found highest (15.77%) in methanol leaf extract. *A. lineata* contained the least yield of phenolic compounds (0.15%) but the maximum percentage yield of tannins (7.80%). Triterpenoids obtained from the plant were found to be minimal (0.54 to 0.15%). Saponin recorded high yield of 4.35% and least yield was found in 1.83%. Steroids were obtained in the plant but the yields recorded were 0.75 to 0.29%. The content of glycosides that was found in *A. lineata* was 0.70 to 0.29%.

The results of antibacterial properties of acetone, ethanol, methanol, petroleum ether and chloroform extracts of the leaves and stem of *A. lineata* are given in Table 3. All the extracts exhibited broad spectrum of activity. When the five extracts were compared with each other and with that of standard antibiotic Ciprofloxacin, the methanol leaf extract showed the highest activity compared to that of the ethanol, acetone, chloroform and petroleum ether extracts. The extract obtained using methanol showed highest activity against *S. aureus* (19.41 mm), *P. aeruginosa* (16.45 mm) and *K. pneumoniae* (16.40 mm) and minimal inhibition zone was observed against *P. aeruginosa* (10.30 mm). No activity was observed against *P. vulgaris* and *E. coli*. The study made on acetone extract recorded highest activity against *P. vulgaris* (17.40 mm), *S. aureus* (15.65 mm) and *P. aeruginosa* (14.63 mm). Further, it showed least activity against *K. pneumoniae (*10.41 mm) in stem extract.

Ethanol extract showed maximum activity against *P. vulgaris* (16.10 mm), *P. aeruginosa* (15.30 mm) and *S. aureus* (14.36 mm) and the minimal activity was against *K. pneumoniae* (8.70 mm). Whereas it has no activity against *P. aeruginosa* in stem extract. The extracts using petroleum ether showed highest inhibition zone observed against *S. aureus* (18.30 mm), *P. aeruginosa* (16.50 mm), *E.coli* (15.10 mm) and *P. vulgaris* (14.16 mm). Least inhibition zone was observed against *K. pneumoniae* (6.40 mm) and *E. coli* (8.40 mm). It has no activity against pathogen like *P. aeruginosa* in stem extract. The chloroform extract antimicrobial activity results showed diameter of inhibition zones ranging from 14.07 to 9.36 mm, with the highest zone was inhibition shown towards *K. pneumoniae* (14.07 mm). Minimum inhibition zone was observed against *E. coli* (9.36 mm).

The phytochemical investigation and quantitative estimation of the percentage yields of chemical constituents of the plant studied was that the leaf and stem were rich in flavonoids, gums and mucilages, saponins and tannins. The plant was known to show medicinal activity as well as

Table 4. Antifungal activity of the various leaf and stem extracts of *Andrographis lineata*.

Test organisms	Acetone extract		Ethanol extract		Methanol extract		Petroleum ether extract		Chloroform extract		Fluconazole 25 µg
	Leaf	Stem	Leaf	Stem	Leaf	Stem	Leaf	Stem	Leaf	Stem	
Aspergillus flavus	9.16± 0.42	7.30± 0.11	11.24± 0.35	6.47± 0.66	8.35± 0.80	6.70± 0.12	9.11± 0.43	5.80± 0.05	8.38± 0.55	6.80± 0.65	16±0.32
Aspergillus niger	10.15± 0.18	0.00	9.40± 0.30	0.00	12.15± 0.03	7.40± 0.15	10.70± 0.15	6.30± 0.25	9.75± 0.20	6.80± 0.12	15±0.17
Penicillium pinophilum	13.40± 0.03	8.40± 0.64	10.48± 0.37	7.31± 0.17	10.70± 0.61	6.80± 0.14	12.11± 0.39	0.00	9.36± 0.19	6.13± 0.71	18±0.20
Fusarium solani	8.10± 0.07	7.20± 0.10	10.18± 0.29	6.71± 0.13	9.14± 0.20	6.11± 0.08	8.70± 0.25	5.14± 0.17	8.13± 0.10	4.70± 0.50	19±0.39
Atternaria alternata	9.35± 0.20	8.11± 0.13	10.31± 0.40	6.03± 0.07	10.70± 0.20	5.90± 0.32	8.30± 0.04	5.31± 0.10	8.20± 0.71	5.10± 0.13	15±0.50

exhibiting physiological properties (Sofowara, 1993). Tannin has been found to have antibacterial, antiulcer and antiviral activities (Moreman, 1998). Flavonoids on the other hand, are potent water soluble antioxidants and free radical scavengers, which prevent oxidative cell damage and have strong anti-cancer activity (Okwu, 2004; Okwu and Josaiah, 2006). Saponin possesses specific chemical, physical and biological properties that make them useful as medicines. Some of the biological activities include antimicrobial (Margineanu et al., 1976) and hemolytic effects (Abe et al., 1981).

The antibacterial activity of another species of this family (Acanthaceae) has been reported (Santhi et al., 2006; Mishra et al., 2009). Currently the attention has been directed toward extracts and biologically active compounds isolated from familiar plant species. The use of medicinal plants plays an essential role in convening the main health needs in developing countries and these plants may offer a modern source of antibacterial, antifungal and antiviral agents with significant activity against infective microorganisms (Shadomy et al., 1985; Odds, 1989). Oliveira et al. (2007) reported the antimicrobial activity of *Syzygium cumini* extract from leaves. Kumar and Vaidhyalingam (2010) reported the antibacterial and antifungal activity of *Rubus racemosus* extract from aerial parts. Alagesaboopathi (2011) reported the phytochemical screening and antimicrobial potential of *Andrographis ovata* extract from leaves.

The antifungal activity of acetone, ethanol, methanol, petroleum ether and chloroform extracts of leaf and stem of *A. lineata* were evaluated by measuring the diameters of zones of growth inhibition of the fungal colonies and the results are given in Table 4. Antifungal activity denoted that the tested fungal strains are most susceptible to acetone extract. Antifungal activity results of acetone extract recorded the diameter of inhibition zones ranging from 7.20 to 13.40 mm with the highest inhibition zone observed against *P. pinophilum* (13.40 mm). Minimum inhibition zone was noticed against *F. solani* (7.20 mm). Where it has no activity against *A. niger* in stem extract. The investigation made on ethanol extract showed a highest activity against *P. pinophilum* (11.48 mm), *Alternaria alternata* (10.31 mm), *A. flavus* (10.24 mm) and *F.solani* (10.18 mm) and the minimal activity against *A. alternata* (6.03 mm) in stem extract. It has no activity against *A. niger* in stem extract.

The extract using methanol showed maximum activity against *A. niger* (12.15 mm), *P. pinophilum* (10.70 mm) and *A. alternata* (10.70 mm). Least inhibition zone was showed against *A. alternata* (5.90 mm) in stem extract. Petroleum ether extract pointed out maximum activity against *P. pinophilum* (12.11 mm), *A. niger* (10.70 mm) and *A. flavus* (9.11 mm) and the minimum activity against *F. solani* (5.14 mm) in stem extract. No activity was noticed against *P. pinophilum* in stem extract. Observation made from chloroform extract showed a highest activity against *A. flavus* (11.38 mm), *P. pinophilum* (10.36 mm) and *A. niger* (9.75 mm) and the minimum activity against *F. solani* (4.70 mm).

Several workers have screened a large number of plants belonging to angiosperms and gymnosperms for their fungitoxic activities. Mainly the aqueous extracts of expressed juice of plants have been used to evaluate their fungitoxicity (Sawant, 1999; Thapliyal et al., 2000; Alagesaboopathi and Balu, 2000). However some of the researchers have used organic extracts.

The use of 50% ethanol ensures extraction of highest compounds as well as facilities further purification of active fractions (Dhar et al., 1973).

The results of the current study revealed that antibacterial efficacies of acetone, ethanol, methanol, petroleum ether and chloroform extracts varied in effectiveness which may be attributed to the presence of saponins and tannin. The presence of phenolic compounds in the plant indicates the antimicrobial properties. In the present research, the author also observed the antibacterial assay, which agrees with the findings of Ofokansi et al. (2005). The phytochemicals are

Known to have antimicrobial properties (Gupta et al., 2010). Hence, these plants can be used to discover bioactive natural products that may lead to the development of new pharmaceuticals research activities.

REFERENCES

Abe H, Odashima S, Arichi S (1981). The effects of saikosaponins on biological membranes. Ultra structure studies on the effects of saikosaponnins on the cell structure. Planta. Med., 42: 356–363.

Ahmedullah M, Nayar MP (1986). Endemic Plants of the Indian Region, Botanical Survey of India, Calcutta. I: 143–146.

Alagesaboopathi C (1993). Studies on the biology of some medicinally potential species of *Andrographis* Wall of the Shevaroy Hills of Tamilnadu, India. Ph.D. Thesis, Bharathidasan University, Tiruchirappalli, Tamilnadu, India.

Alagesaboopathi C (2011). Phytochemical screening and antimicrobial potential of *Andrographis ovata* Clarke. Afr. J. Biotechnol., 10(25): 5033-5036.

Alagesaboopathi C, Balu S (2000). Antifungal activity of some species of *Andrographis* Wallich ex Nees on *Helminthosporium orazye* Breda Dehann. J. Econ. Tax Bot., 24: 705–707.

Al-Bayati, FA, Al-Mola HF (2008). Antibacterial and antifungal activity of different parts of *Tribulus terrestris* L. growing in Iraq. J. Zhejiang Univ Sci,, B9:154-159.

Ayyanar M, Sankarasivaraman K, Ignacimuthu S (2008). Traditional herbal medicines used for the treatment of diabetes among two major tribal groups in South Tamilnadu, India. Ethnobot. Leaflets, 12: 276–280.

Balu S, Alagesaboopathi C (1993). Anti-inflammatory activities of some species of *Andrographis* Wall. Acanthaceae, Anc. Sci. Life, 13:180–184.

Balu S, Alagesaboopathi C (1995). Antivenom activities of some species of *Andrographis* Wall. Anc. Sci. Life, 14: 187–190.

Balu, S, Alagesaboopathi C, Elango V (1993). Antipyretic activities of some species of *Andrographis* Wall. Anc. Sci. Life, 12: 399–402.

Bishnu J, Sunil L, Anuja S (2009). *Ocimum sanctum, Cinnamomum zeylanicum, Xanthoxylum armatum and Origanum majorana.* Kathmandu Uni. J. Sci. Eng. Technol,, 5:143–150.

Chen IN, Chang CC, Wang CY, Shyu YT, Chang TL (2008). Antioxidant and antimicrobial activity of Zingiberaceae plants in Taiwan, Plant. Foods Hum. Nutr,, 63: 15–20.

Costa ES, Hiruma-Lima CA, Limo EG, Sucupira GC, Bertolin AO,.Lolis SF, Andrade FD, Vilegas W, Souza-Brito AR (2008). Antimicrobial activity of some medicinal plants of cerrado, Brazil. Phytother Res., 22: 705 707.

Dhar ML, Dhawan BN, Mehtotra BN, Roy C (1973). Screening of Indian Plants for biological activity. Indian J. Expt. Biol., 6: 232–247.

Fabricant DS, Fansworth NR (2001). The value of plants used in traditional medicine for drug discover. Environ. Health Perspect., 109: 69-75.

Faziy-Bazzaz BS, Khajehkaramadin M, Shokooheizadeh HR (2005). *In vitro* antibacterial activity of *Rheum ribes* extract obtained from various parts against clinical isolates of Gram-negative pathogens. Iranian J. Pharm. Res., 2: 87–91.

Gamble JS (1924). Flora of the Presidency of Madras. Vol.II. Botanical Survey of India, Calcutta. pp.1045 – 1051.

Gupta C, Garg AP, Gupta S (2010). Antimicrobial and phytochemical studies of fresh ripe pulp and dried unripe pulp of *Mangifera indica* (AMCHUR). Middle – East J. Sci. Res., 5: 75–80.

Hari KP, Vijaya BRM, Kesava RM, Gunasekar D, Caux C, Bodo B (2003). Flavonoids from *Andrographis lineata*. J. Phytochem., 63:457-461.

Ikegami F,.Fujii Y, Ishihara K, Satoh T (2003). Toxicological aspects of Kampo medicines in clinical use. Chem. Biol. Interact., 145: 235-250.

Kadhirvel K, Rajivaganthi P, Narayanan G, Govindaraj V (2010). Investigations on antidiabetic medicinal plants used by tribal inhabitants of Nalamankadai, Chitteri Reserve Forest, Dharmapuri, India. Ethnobot. Leaflets, 14: 236–247.

Karuppusamy S (2007). Medicinal plants used by Paliyan tribes of Sirumalai Hills of Southern India. Natural Prod. Radiance, 6: 436–442.

Kirtikar KR, Basu BD (1975). Indian Medicinal Plants, Bishen Singh Mahendra Singh, New Delhi. 3: 1884–1886.

Kokate CK, Purohit AP, Gohale SB (2003). Pharmacognosy Nirali Prakashan Publishers, Pune, India. pp. 1-624.

Kumar PR, Vaidhyalingam V (2010). Antibactorial and antifungal activity of aerial parts of *Rubus racemosus*. Der. Pharmacia. Lettre. 2(6): 16-20.

Kumar RA, Sridevi K, Kumar NV, Nanduri S, Rajagopal S (2004). Anticancer and immunostimulatory compounds from *Andrographis paniculata* J. Ethnopharmacol,, 92: 291–295.

Margineanu VC, Cucu V, Grecu L, Parvu C (1976). Anticandidal action of saponin from *primula* spp. Planta. Med., 30: 35 – 38.

Mishra, US., A. Mishra, R. Kumari, PN. Murthy, BS. Naik. (2009). Antibacterial activity of ethanol extract of *Andrographis paniculata*. Indian J. Pharm. Sci., 71: 436–438.

Moreman DE (1998). Native American Ethnobotany, Timber Press, Orgo. pp. 473–475.

Odds FC (1989). Antifungal activity of saperconazole (R.66905) *in vitro*. J. Antimicrobiol. Chemother., 24: 533-537.

Ofokansi KC, Esimore CO, Anele CK (2005). Evaluation of the in vitro combined antibacterial effect of the leaf extracts of *Bryophyllum pinnatum* and *Ocimum gratissium*. Plant Prod. Res. J., 9: 23–27.

Okwu DE (2004). Phytochemicals and vitamin content of indigenous spices of Southeastern Nigeria. J. Sustain. Agric. Environ., 6: 30–37.

Okwu DE, Josaiah C (2006). Evaluation of the chemical composition of two Nigerian medicinal plants. Afr. J. Biotechnol., 5(4): 357-361.

Oliveira GF, Furtado N, Filho AA, Martins JK, Cunha WR, Silva ML (2007). Antimicrobial activity of *Syzygium cumini* (Myrtaceae) leaves extract. Br. J. Microbiol., 38: 1517-1525.

Perumalsamy R, Ignacimuthu S (2000). Antibacterial activity of some folklore medicinal plants used by tribals in Western Ghats of India. J. Ethnopharmacol., 69: 63–71.

Sangameswaran B, Ilango K (2010). Evaluation anti-hyperglycemic and anti-hyperlipidaemic activities of *Andrographis lineata* Nees on Streptozotocin induced diabetic rats. Jordan J. Biol. Sci., 3: 83–86.

Sangameswaran B, Chandrakanth T, Reddy BJ (2007). Diuretic activity of leaf extracts of *Andrographis lineata*. Nig. J. Nat. Prod. Med. 11: 61–63.

Sangameswaran B, Chandrakath TR, Jayakar B (2008). Hepatoprotective effect leaf extract of *Andrographis lineata* Nees on liver damage caused by Carbon Tetrachloride in rats. Phytother. Res., 22: 124-126.

Santhi R, Alagesaboopathi C, Rajasekarapandiyan M (2006). Antibacterial activity of *Andrographis lineata* Nees and *Andrographis* echioides Nees of the Shevaroy Hills of Salem District, Tamilnadu, Adv. Plant Sci., 19: 371–375.

Sawant GG (1999). Studies on biofungicidal properties of some plant extracts on growth of *Alternaria solani* and *Colletotrichum langenarium in vivo*. Indian J. Environ. Taxicol., 9: 46–48.

Shadomy S, Espinel-Ingrof A, Gebhar RJ (1985). *In vitro* studies with sf 86-327, new orally active allylamine derivatives. I. Med. Vet. Mycologia., 23: 125-132.

Sharma B, Sharma UK (2009). Hepatoprotective activity of some indigenous plants, Int. J. Pharm Tech Res., 4: 1330–1334.

Sheeja, K. Kultan G (2007). Activation of cytotoxic and lymphocyte responses and attenuation of tumor growth *in vivo* by *Andrographis paniculata* extract and andrographolide. Immunopharm. Immunot., 29: 81-93.

Silva O, Duarte A, Pimental M, Viegar M, Barroso J, Machado I, Pires J, Cabrita EG (1997). Antimicrobial activity of *Terminalia macroptera* root. J. Ethnopharmacol., 57: 203–207.

Sivaperumal R, Ramya S, Veera AR, Rajasekaran C, Jayakumari R (2010). Ethnopharmacological studies on the medicinal plants used by tribal inhabitants of Kottur Hills, Dharmapuri, Tamilnadu, India. Environ. We Int. J. Sci. Tech., 5: 57–64.

Sofowara A (1993). Medicinal plants and traditional medicine in Africa, Spectrum Books Ltd. Ibadan, Nigeria. p. 289.

Thaplial M, Ghosh M, Bennet SSR (2000). Screening of six medicinal plants for their antifungal protein activity. Asian J. Microbial. Biotechnol. Environ. Sci., 2: 215–218.