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ANTIOXIDANT POTENTIAL OF BARK AND LEAVES EXTRACTS OF MANGROVE PLANT *AEGICERAS CORNICULATUM* L

Bidve S. C.[1], Kadam V. B.*[1], Shindikar M. R.[2] and N. P. Malpathak[3]

[1]P.G. Department of Botany and Research Centre, K.T.H.M. College, Nashik-02.

[2]Department of Applied Science, COEP College Pune.

[3]P.G. Department of botany, Savitribai Phule Pune University, Pune.

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***Corresponding Author**
Dr. Kadam V. B.
P.G. Department of Botany and Research Centre, K.T.H.M. College, Nashik-02.

ABSTRACT

The present study investigated antioxidant activities in leaves and bark of (*Aegiceras corniculatum* L.) mangrove plants. Mangroves are salt-tolerant plants of tropical and subtropical intertidal regions of the world. Nearly 65 species of mangrove reported from world and approximately 59 species from 29 families are reported in India. The dried, powdered plant material of leaves and bark were used for extraction separately. The range of solvent from non-polar to polar was selected for extraction. Total five solvents were used for extraction viz. Petroleum ether, Chloroform, Ethyl acetate, Methanol and Water separately. Total ten extracts were obtained. The extraction was carried out by cold extraction method**.**

KEYWORDS: Antioxidant activity, Mangrove, Petroleum ether, Chloroform, Ethyl acetate, Methanol and Water.

INTRODUCTION

Oxidation is one of most important processes, which produce free radicals in food, chemicals, and even in living systems. Free radicals have an important role in processes of food spoilage, chemical materials degradation and also contribute to more than hundred disorders in humans. Antioxidants are defined as substances that even at low concentration significantly delay or prevent oxidation of other compounds. These reactive oxygen species cause destructive and irreversible damage to the components of a cell, such as lipids, proteins and DNA (Umadevi et.al, 2013). Although normal cells possess antioxidant defense systems against ROS, the continuous accumulation of damage to the cells induces diseases such as

cancer and aging (Maestri et al, 2006). Fruits, vegetables, juices and spices have gain importance throughout history of mankind as a source of natural antioxidants. Along with it dietary constituents such as vitamin C, vitamin E, and carotenoid are potential source of antioxidants (Dorman et al, 2003).

The application of antioxidant is industrially widespread in order to prevent oxidative degradation of polymers, discolourisation of synthetic and natural pigments, etc. Antioxidants are very important in food industries also. Synthetic antioxidants such as butylated hydroxyl toluene (BHT), Tetra butyl hydro quinone (TBHQ), butylated hydroxyl anisole (BHA) used in food industry causes serious problems due to their volatile nature, instability at higher temperature and toxicity. Therefore, replacement of synthetic antioxidant with natural antioxidant has gained enormous importance. Plants served as a potential candidate as a source of natural antioxidant in area of medicine, food and cosmetics.

Aegiceras corniculatum is one of important species of mangrove. It is commonly known as Black Mangrove or River Mangrove. It is a species of shrub or tree mangrove in the Myrsinaceae family with a distribution in coastal and estuarine areas ranging from India through South East Asia to Southern China, New Guinea and Australia. It grows in mud in estuaries and tidal creeks, often at the seaward edge of the mangrove zone. It has salt glands in the leaves for secretion of excess salt and show tolerance to the changes in salinity gradients**.** *Aegiceras corniculatum* grows as a shrub or small tree upto 7 m high, though often considerably less. Its leaves are alternate, entire, leathery and minutely dotted. Its flowers produced in umbellate cluster and flowers are small, fragrant and white in colour. The fruit is curved and cylindrical or horn-shaped, light green to pink in colour. Traditionally *Aegiceras corniculatum* is used in treatment of asthma, diabetes, rheumatism also in fish poisoning.

In mangrove species phenolics are main constituent and important in protection from herbivores. Along with phenolics a wide range of metabolites like tannins, alkaloids, flavonoids, carotenoids, steroids, saponins are identified from mangrove plants. Phenolics are efficient free radical scavengers can potentially interact with biological systems and play important role in preventing oxidative stress induced diseases (Huang et al, 2009). Beula et al, (2012) reported the antioxidant properties of mangrove plant from south east coast of India. Wei et al, (2011) also reported the presence of tannins from *Aegiceras corniculatum* and studied antioxidant activity of freeze dried extracts of stem bark, leaves and root bark powder. They have reported that methanolic extracts were showing good antioxidant activity

and more concentration of phenolic compounds as compared to water and ethyl acetate. Extracts and chemicals from mangroves are used mainly in folkloric medicine (e.g. bush medicine) for treatment of various diseases like hepatitis, asthmas, ulcer diabetes and diarrhea, as well as in treatment of skin disorders like leprosy, boils, wounds etc. The mangrove extracts also used as insecticides and pesticides. Mangrove extracts have been used for diverse medicinal purposes and have a variety of antibacterial, antiherpetic and anthelminthic activities. (Bandaranayake, 1998).

MATERIALS AND METHODS

The plant material was collected from Pawas region of Ratnagiri district of Maharashtra state (south-west costs of India). The collected plant material was first separated as per plant parts (leaves, bark, flowers and fruits). The separated plant parts were clean and dried in shade and ground into fine powder. The dried, powdered plant material of leaves and bark were used for extraction separately. The range of solvent from non polar to polar was selected for extraction. Total five solvents were used for extraction viz. Petroleum ether, Chloroform, Ethyl acetate, Methanol and Water separately. Total ten extracts were obtained. The extraction was carried out by cold extraction method**.**

Cold extraction method

10 gm of powdered plant material was mixed with 50 ml of solvent separately. The mixture was sonicated at 33 KHz for 40 min and allowed to stand for at least 12 hrs. The extraction was carried out repeatedly until solvent got colorless. The extracts were filtered and concentrated in rotary evaporator to dryness. All obtained extracts were dissolved in 0.1% dimethyl sulfoxide (DMSO) and diluted to yield various working concentrations. Overall ten extracts were obtained.

Antioxidant activity

The dried extracts were dissolved in appropriate volume of dimethyl sulphoxied (DMSO) to obtain 10 mg/ml concentration. Antioxidant activities of extracts were determined at 10, 50, 100, 150 and 200 µg/ml concentrations.

a) DPPH (2,2-diphenyl-1-pyerilhydrazil hydrate) radical scavenging assay

The ability of the plant extracts to scavenge the stable free radical DPPH was assayed by the method of Brand-Willams et al, (1995).

b) ABTS$^+$ scavenging effect

The ability of *Aegiceras corniculatum* to scavenge the free radical ABTS (2,2-azino-bis 3-ethyl benzothiazoline-6-sulfonic acid) was studied using the method adopted by Teow *et al*. (2007).

c) Hydrogen Peroxide Scavenging Effect

The scavenging activity of hydrogen peroxide by the plant extracts was determined by the method of Ruch *et al*. (1989).

d) Phosphomolybdenum reduction

The total antioxidant capacity of extracts was determined by Phosphomolybdenum Reduction assay carried out by Prieto et al, (1999) method.

e) Ferric Reducing Antioxidant Power (FRAP)

The reduction power of extracts was determined by FRAP assay carried out by Wei et. al., (2010) / Benzie and strain (1996) method.

f) Estimation of Total Phenol

The content of total phenolics compounds plant extract was determined by modified method of Folin Ciocalteu method (1927).

g) Total flavenol

The total flavonolic components in plant extract were determined by modified method of Ordon et al. 2006.

h) Total flavonoids

The total flavonoid components in plant extract was determined by modified method of Ordon et al. 2006.

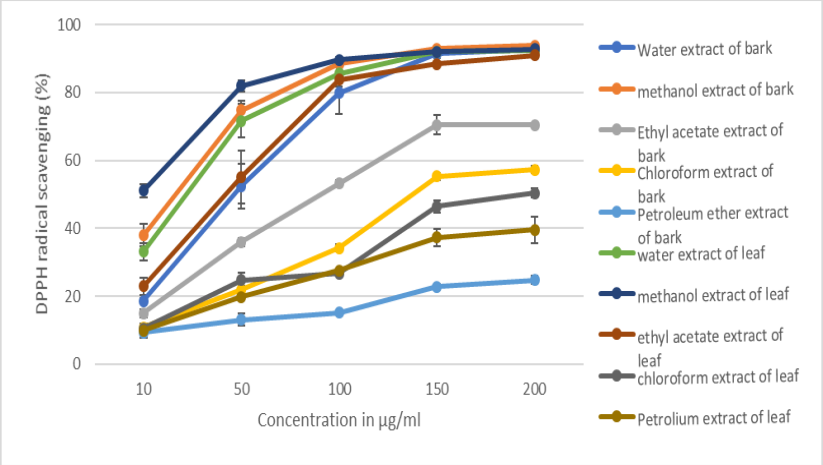
RESULT AND DISCUSSION

4.2.1 Antioxidant activity

The antioxidant activities were studied by using radical scavenging assay- DPPH, ABTS$^+$ and Hydrogen peroxide radical scavenging activity, Phosphomolybdenum reduction assay, Ferric Reducing Antioxidant power, Estimation of total phenolic, flavonoids and flavanols.

A: DPPH radical scavenging activity

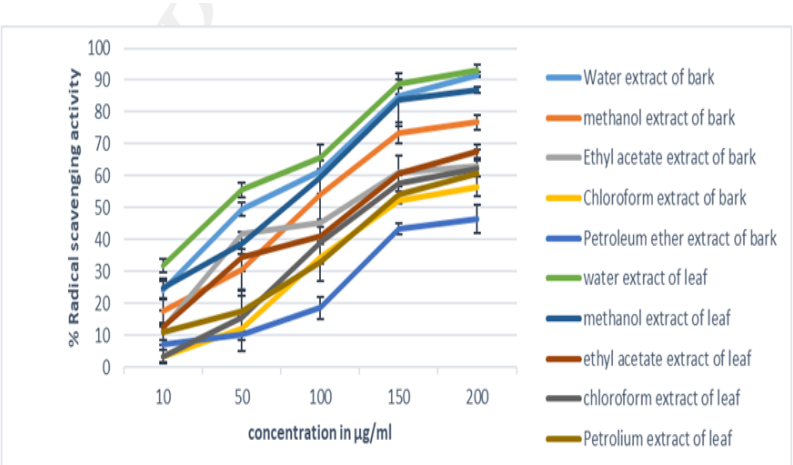
Highest antioxidant activity was showed by methanolic, water and ethyl acetate extract of leaf (EC50 value: 55.77,58.43.59.73 µg /ml). Also, methanolic and water extract of bark shows good antioxidant activity (EC50: 80.73, 82.02 µg /ml). Petroleum ether extracts of bark and leaf showing considerably low antioxidant activity (EC50; 328and 201 µg /ml) as seen in graph 1.



Graph. 1: DPPH radical scavenging activity of *A.corniculatum* extracts.

B: ABTS^{+} radical scavenging activity

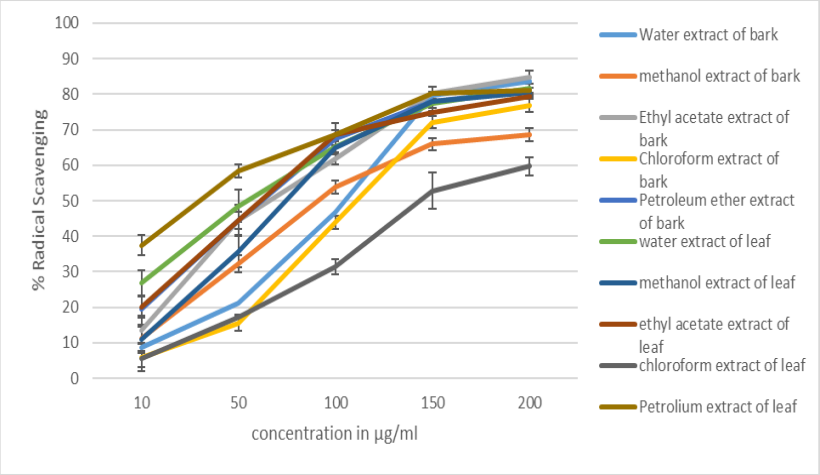
Highest antioxidant potential was showed by water extract of leaf and bark (EC50: 84.38 and 87.89 µg /ml) Also methanolic extract of leaf and bark shows good antioxidant capacity (EC50: 89.33 and 102.34 µg /ml) As shown in graph 2.



Graph. 2: ABTS radical scavenging activity of *A.corniculatum* extracts.

C: Hydrogen peroxide radical scavenging activity

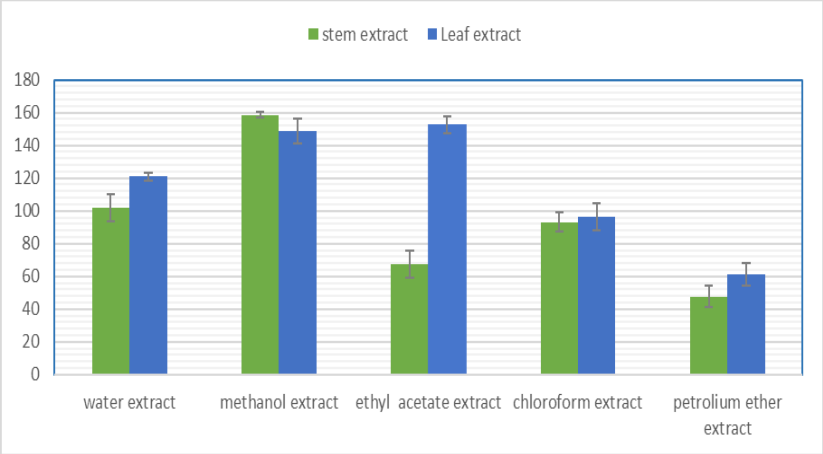
H_2O_2 is weak oxidizing agent that inactivates few enzymes directly, usually oxidation of thiol (-SH) groups. It can cross cell membrane rapidly; once it enters inside cell, it can probably react with Fe^{2+} and Cu^{2+} ions to form hydroxyl radicals and this may be origin of many of its toxic effects. From the results, it was seen that H_2O_2 scavenging was less as compare to ABTS and DPPH scavenging assays. The highest H_2O_2 scavenging was showed by petroleum ether extract of leaf and ethyl acetate extract of bark. (EC50:93.59, 93.44µg/ml). Also, water and petroleum ether extract of bark (EC50:94.19 and 96.09µg/ml) and water and methanol extract of leaf (EC 50: 97.07, 95.05 µg /ml) showed good radical scavenging activity. This activity was very less as compare to standard antioxidant ascorbic acid (EC50: 0.83 µg /ml). As seen from graph 3.s.



Graph. 3: H_2O_2 radical scavenging activity of *A.corniculatum* extracts.

D: Phosphomolybdenum reduction assay

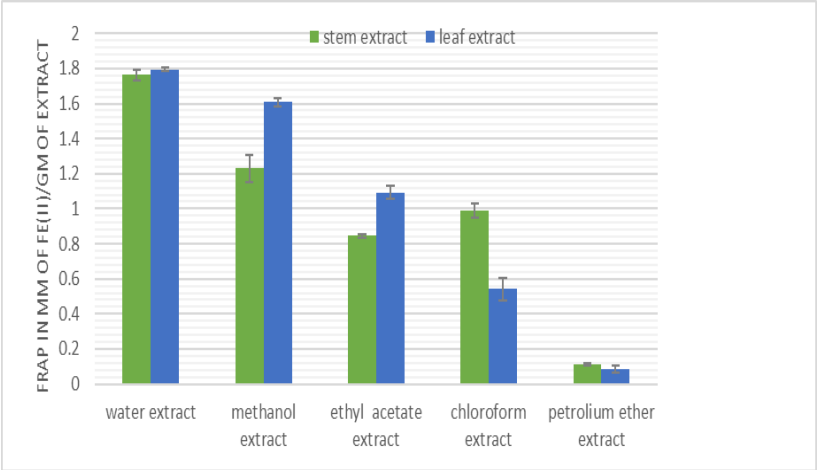
The antioxidant potential could be measured by formation of green Phosphomolybdenum complex. The method is based on reduction of Mo (VI) to Mo(V) by antioxidant compounds and formation of green Mo (V) complex. The different extracts at a concentration of 50ug/ml was used for assay. The strongest reducing potential was shown by methanolic extract of bark and leaf and ethyl acetate extracts of leaves (158.57, 148.6, 152.32 equivalents of ascorbic acid in µg /gm of extract). The reducing capacity of extracts was ranging from 61.27-158.57 equivalents of ascorbic acid in µg/gm of extract (Graph 4.)



Graph. 4: Antioxidant capacity in equivalents of ascorbic acid in ug/gm of *A. corniculatum* extracts.

E: Ferric Reducing Antioxidant power

The FRAP assay treats the antioxidants contained in the sample as reluctant in redox –linked colorimetric reaction and the value reflects the reducing power of antioxidant. This assay is widely used in evaluation of antioxidant component in dietary polyphenols. The reducing power of extract could be estimated from their ability to reduce TPRZ-Fe(III) complex to TPRZ-Fe(II). The reducing power of extracts was ranging from 1.79-0.084 mM of Fe(II) /gm of extract. The highest reducing power was shown by methanolic extract of bark and leaf i.e. 1.76 and 1.79 mM of Fe(II)/gm of extract (Graph 5).

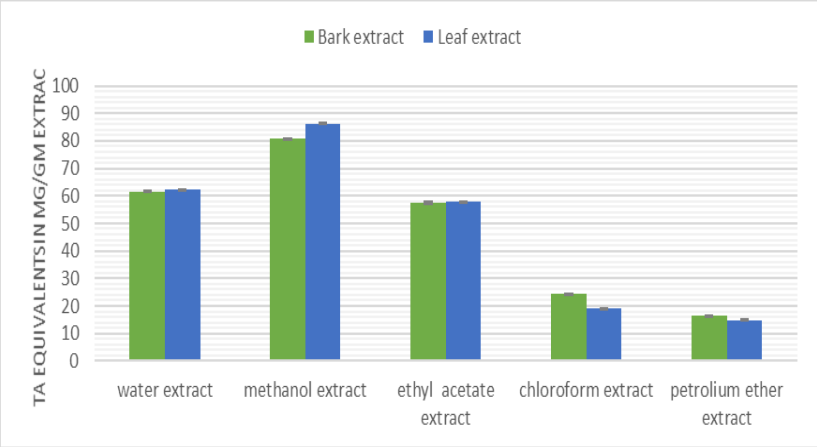


Graph. 5: Reducing power of *A.corniculatum* extracts.

F: Estimation of Total phenol

The total phenolic contents from various extracts of leaves and bark of *A. corniculatum* were analyzed by folin-coicalteu method. As seen in graph 6, the total phenolic contains was

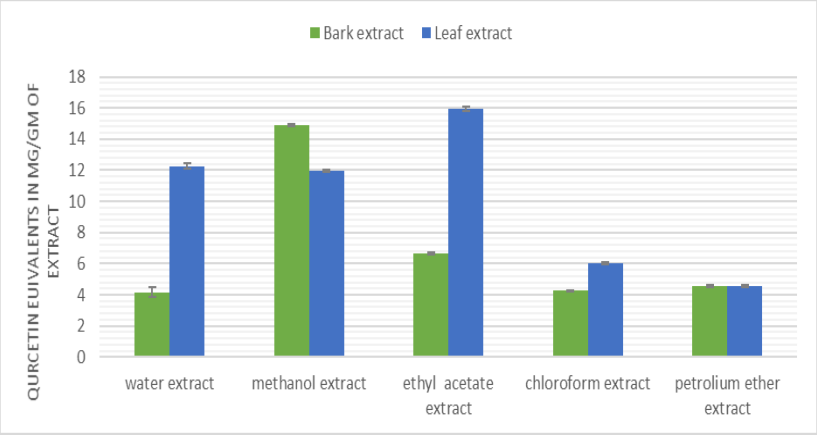
varied from 86-14 mg TAN/gm of dry weight. The methanolic extract of leaves and bark showed highest phenolic contents (86.30 and 80.88mg TAE/gm of dry weight) while petroleum ether and chloroform extracts showed lowest phenolics (14.23, 16.20 and 18.56, 24.84mgTAE/gm dry weight) Ethyl acetate and water extracts showed moderate amount of phenolics (57-62 mg TAE/gm of dry weight) (Graph 6).



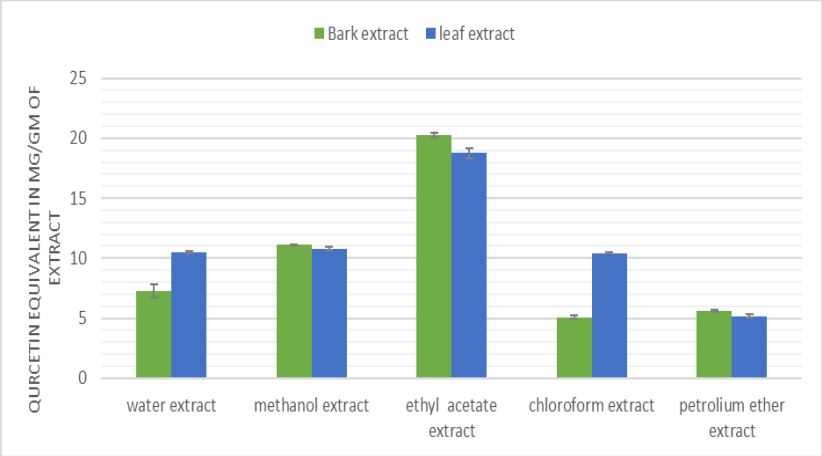
Graph. 6: Total amount of phenolic contents of extracts of *A. corniculatum*.

G: Total flavonol and flavonoid

As showed in graph 7 and graph 8 the highest flavonol and flavonoid contents were observed in ethyl acetate extracts of leaves and bark while chloroform and petroleum ether extracts showed lowest. The ethyl acetate extract of leaves showed 15.9 and 20.25 mg QUR/gm of dry weight of flavonol and flavonoid respectively. The petroleum ether extracts lowest flavonol and flavonoid concentration (4.5 and 5.15 mg QUR/gm dry weight). The water and methanolic extracts showed considerable concentration of flavonol (12.20 and 10.32 mg QUR/gm dry weight) but flavonoid concentration was very low (Graph 7).



Graph. 7: Total amount of flavonol contents of extracts of *A. corniculatum*.



Graph. 8: Total amount of flavonoid contents of extracts of *A. corniculatum*.

CONCLUSION

The correlation was studied between antioxidant activities (at EC_{50} value or 50 µg/ml concentrations) and total phenolics, flavonoids and flavanol from different extracts. It was observed that the total phenol content showed strong correlation with DPPH, ABTS radical scavenging assay and FRAP, Phosphomolybdenum assay. While total flavonoid content showed partial correlation with theses assays and flavanol content showed moderate correlation. The hydrogen peroxides radical scavenging assay showed negative correlation with total phenols, flavanols and flavonoids content.

The antioxidant activity of different extracts was studied by different antioxidant assay includes radical scavenging assays (DPPH, ABTS, H_2O_2) and antioxidant power assay (FRAP and Phospho-molybdenum assay). Results obtained in present study showed the methanolic and ethyl acetate extracts showed highest antioxidant capacity except in hydrogen peroxide scavenging assay. The water extracts also showed good antioxidant activity. The chloroform extracts showed moderate while petroleum ether extracts showed very poor antioxidant activity. The methanolic extracts showed high phenolic content which was strongly correlated to antioxidant activity.

REFERENCE

1. Bandaranayake W. M. Bioactivities, bioactive compounds and chemical constituents of mangrove plants Wetlands Ecology and Management, 2002; 10: 421–452.
2. Benzie I and strain J The Ferric Reducing Ability of plasma (FARP) as a measure of Antioxidant Power: FRAP Assay Analytical Biochemistry, 1996; 239: 70-76.

3. Beula Margaret J. Ganadesigan M., rajkumar Banerjee P., Ravikumar S., Anand M., Antiviral, Antioxident and toxicological evaluation of mangrove plant from south east coast of India Asian Pacefic Journal of Tropical Biomedicine, 2012; S352-S357.
4. Bhandarnayake W. M. Traditional and medicinal uses of mangrove Mangrove salt Marshes, 1998; 2: 133-14.
5. Brand-Williams, Cuvelier M.E., Berset C.L. Use of free radicla method to evaluate antioxidant activity LWT-Food Sci Techanology, 1995; 28: 25-30.
6. Choong C. Teow, Van Den Truong, Rager F. McFeeters, Roger L. Thompson, Kenneth V. Pecota, G. Craig Yencho, Antioxidant activities of phenolic and B-carotene contents of sweet potato genotype with varying flesh colour Food Chemistry, 2007; 103: 820-829.
7. Dormann P., Fiehn O., Kopka J., Altmann T., Trethewey R.N. Willmitzer L. Metabolite profiling for plant functional genomics Nat. Biotechnol, 2000; 18: 1157-1161.
8. Ferguson P., Kurowask E., Freeman D., Chebers A. and Koropatnick Flavonoid fractions from caranberry extracts inhibites proliferation of human tumor cell lines J. Nutrition, 2004; 134(6): 1529-1535.
9. Folin O., Ciocalteau V. On tyrosine and tryptophan determination of proteins. J. Biol.Chem., 1927; 73: 424-427.
10. Huang Xiao-Ying, Wang Qi, Liu Hai-Li, Zhang Yu, Xin Guo-Rong, Shen Xu, Dong Mei-Ling, Guo Yue-Wei 2009 Diastereoisomeric macrocyclic polydisulfides from the mangrove Bruguiera gymnorrhiza Phytochemistry xhxx: xxx–xxx.
11. Huang Xiao-Ying, Wang Qi, Liu Hai-Li, Zhang Yu, Xin Guo-Rong, Shen Xu, Dong Mei-Ling, Guo Yue-Wei 2009 Diastereoisomeric macrocyclic polydisulfides from the mangrove Bruguiera gymnorrhiza Phytochemistry xhxx: xxx–xxx.
12. Maestri D.M., Nepote V., Lamarque A.L. and Zygadlo J, A. 5- Natural products as Antioxidants Phytochemistry, Advances in Research, 2006; 76: 105-135.
13. Ordon Ez, Gomez J.D., Vatuone M.A. and Isla M.I., Antioxidant activities of Sechium edule (Jacq) swart extracts Food chemistry, 2006; 45: 67-70.
14. Prieto P Pineda M. and Aguilar M. Spectrophotometric quantification of antioxidant capacity through formation of Phosphomolybdenum complex: specific application to determinat of vit. E Analytical biochem, 1999; 269: 337-341.
15. Ruch R.T., Cheng S.J., Klaunig J.E Prevention of cytotoxicity and inhibition of intracellular communication by antioxidant catechins isolated from Chinese green tea. Carcinogenesis methods in enzymology, 1984; 105: 198-209.

16. Sundaram Ravikumar, Ganesan Ramanathan, Inbaneson Samuel Jacob and Andy Ramu Parasitol Antiplasmodial activity of two marine polyherbal preparations from Chaetomorpha antennina and Aegiceras corniculatum against Plasmodium falciparum Res., 2011; 108: 107–113.
17. Umadevi M., Kumar Sampath, Bhowmik Debjit, Duraivel S. Traditionally Used Anticancer Herbs In India Journal of Medicinal Plants Studies, 2013; 1(3): 56-74.
18. Wei Shu-Dong, Zhou Hai-Chao and Lin Yi-Ming, Antioxidant activities of extracts and fractions from the hypocotyls of mangrove plant Kndelia candel, Int. J. Mol. Sci., 2010; 11: 4080-4093.
19. Wei Shu-Dong, Lin Yi-Ming, Liao Meng-Meng, Zhou Hai-Chao, Li Yuan-Yue Characterization and Antioxidative Properties of Condensed Tannins from the Mangrove Plant Aegiceras corniculatum Journal of Applied Polymer Science, 2011; 12: 2463–2472.