


Human Journals

Research Article

October 2016 Vol.:4, Issue:4

© All rights are reserved by N.Hariram et al.


Coleus aromaticus Benth Synthesis of Potentially Nanomedicine as High Nutritive Value of Human Health and Immunomodulator



IJSRM

INTERNATIONAL JOURNAL OF SCIENCE AND RESEARCH METHODOLOGY

An Official Publication of Human Journals



Thilagavathi. S, and *N.Hariram

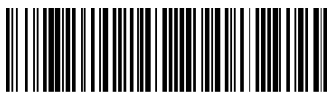
Department of Biotechnology,

Kalasalingam University, Krishnankoil-626 126, India.

Submission: 29 September 2016

Accepted: 7 October 2016

Published: 25 October 2016



HUMAN JOURNALS

www.ijsrm.humanjournals.com

Keywords: Rosmarinic acid, Chlorogenic acid, Coumaric acid, XRD and HPTLC

ABSTRACT

Coleus aromaticus Benth beautiful succulent garden medicinal herb potted plants and Indian traditional medicine has a lot of potentiality to cure much human disease. The broad spectrum of the plants is therapeutic potential of antioxidant, antimicrobial, antitumor, and natural antibiotic was cured. The present study was crystallization (82.3%) of aromatic compounds of *Coleus aromaticus* Benth for the first time from leaf extracts. The crystallized aromatic compounds were characterized by XRD (X-ray Powder Diffraction) and HPTLC for 35 types of compound were expressed, antibiotic activity was analyzed by peak-1, peak-2 and peak-3 specifically. The highest level was yield in *rosmarinic acid*, *chlorogenic acid*, *coumaric acid* and *caffeic acid* in the sample. However, the amount of organic compound was predominantly found in *rosmarinic acid* (90%), *chlorogenic acid* (31.6%), *coumaric acid* (35.6%) and *caffeic acid* (62.1%) for out of 35 different compounds in the crystal. The result shows therapeutic potential of *Coleus aromaticus* which can be used as potential of nanomedicine as high nutritive value of human health and big source of bioactive compounds for immunomodulator.

INTRODUCTION

Coleus aromaticus (Benth) is a commonly available medicinal herb in India. The plant chosen for study *Coleus aromaticus* (Benth), belonging to the Family: *Lamiaceae* is grown as a household herb in Tamilnadu (vernacular Tamil name: ommam or ommavalli). This herb is native to East Indies and is widely cultivated in Africa and almost all tropical countries. At first appearance, *Coleus aromaticus* look as a green, perennial, shrub having oval shaped, and succulent leathery leaves with scalloped edges (Fig-1). The plant grows to around 50 cm tall with horizontal stems up to 180 cm long potted plants. The leaves are highly aromatic with a strong flavor of mixed herbs (Pritima, and Pandian. 2008).

Coleus aromaticus, a plant of medical and food interest is told as “must- have” plant in medicinal herb home garden. It is known to possess antimicrobial antiepileptic, and antioxidant activities (Gao *et al.*, 1999; Williams *et al* 1999 and Osawa, and Namiki 1981). The leaves are also used for treatment of cough, throat infection and nasal congestion. The last decade has witnessed considerable research on *Coleus aromaticus* in different parts of world as evident from the increase in publication during last decade. The leaves of this plant are traditionally used for the treatment of severe bronchitis, asthma, diarrhea, epilepsy, renal and vesicle calculi and fever (Warrier *et al* 1995). *C. aromaticus* has been reported to exhibit antilithiatic, chemopreventive, (Prasad *et al* 2002) antiepileptic (Jose *et al* 2005; Buznego and Perez-Saad. 1999) and antioxidant properties (Padma *et al* 1988). Several scientists reported that synthesis of nanoparticles mainly chemical synthesis like reduction in solutions, chemical and photochemical reactions in reverse micelles, physical synthesis like thermal decomposition, radiation assisted, electrochemical, sonochemical, microwave-assisted process and recently via green chemistry route or biological synthesis using plants. Although, *Coleus aromaticus* Benth have rich in aromatic organic compounds found in the form of either gold and or silver nanoparticles. The present study indicates that the plants under investigations are safe for the consumption as reported in early studies. Therefore, the use of the *Lamiaceae* family is regarded safe for if consumed.

The HPTLC analysis was carried out to confirm the presence of alkaloid sand their derivatives in the plants. Chromatographic fingerprint is a holistic, valid and rapid method. HPTLC is a

valuable quality assessment tool for the evaluation of botanical materials. It allows the analysis of a broad number of compounds both efficiently and cost effectively. HPTLC studies have shown that it is more versatile than ordinary TLC methods, as the spots were well studied. X-ray diffraction data provides information about crystallinity, crystallite size, orientation of the crystallites and phase composition and aid in molecular modeling to determine the structure of the material (Joshi *et al* 2008). Advantages of XRD are simplicity of sample preparation, rapidity of measurement, analyze mixed phases and determine sample purity. Its limitations are requirement of homogenous and powdered material, peak overlays lead to unclear data.

THERAPEUTIC USES

Therapeutically used as various disorders in medicine, such as severe bronchitis, asthma, diarrhoea, epilepsy, renal and vesicle calculi, fever (Warrier *et al.*, 1995), common cold, cough, headache, indigestion, urinary diseases, vaginal discharges (Shamachar *et al.*, 1967), colic, dyspepsia, , convulsions (Kirtikar *et al.*, 1935), stimulates the functions of liver, indicated in kidney and bladder stones, dysentery, cholera, bilious affections, poisonous bites (Kirtikar *et al.*, 1935 & Sivarajan *et al.*, 1994) and vitiated conditions of *Kapha* and *Vata* (Sharma *et al.* 1995). Leaves are anthilithic, antispasmodic, carminative, cathartic, and stimulant, stomachic. However, the sexual disease was curing the urinary diseases, vaginal discharge. The leaves juice carminative when mixed with sugar, given to children in colic. It is beneficial in asthma, calculus, chronic cough, dyspepsia, fever, gonorrhea, piles and in strangury. It is externally used in conjunctivitis and bruised leaves locally applied in headache. The expressed juice is used in epilepsy and other convulsive disorders and plant extracts used in the treatment of gastrointestinal troubles (Chatterjee *et al.*, 2001).

IMMUNOSTIMULANTS

These agents are envisaged to enhance body's resistance against infections (and may be against allergy, autoimmunity, and cancer as well), can act through both the innate and adaptive arms of the immune response. In healthy individuals, the immunostimulants are expected to serve as prophylactic agents such as immune potentiators by enhancing the basic levels of immune response and in individuals with immunocompromised conditions (primary and secondary immune deficiencies) as immunotherapeutic agent.[35-40] These agents do not affect

immunological memory cells. Their pharmacological efficacy fades away quickly and must, therefore, be renewed by administering the drug either in intervals or continuously. [41]

IMMUNOMODULATOR OF COMPOUNDS

Several biological activities have been described for polyphenolic compounds, including a modulator effect on the immune system. The effects of these biologically active compounds on the immune system are associated to processes as differentiation and activation of immune cells. Plants are the biosynthetic laboratory of phytochemicals. Phytochemicals are naturally occurring compound with bioactive potentials. The prefix “Phyto” is a Greek word meaning plant. These chemicals are often referred to as “secondary metabolites”. These are several classes of compounds that include alkaloids, flavonoids, coumarins, glycosides, gums, polysaccharides, phenols, tannins, terpenes, and terpenoids.[51,52] In addition to compounds that are necessary for the growth and reproduction, plant cells synthesize a number of secondary metabolites, which do not appear to be strictly necessary for the survival of the plant. These secondary metabolites or phytochemicals are produced as a response to external stimuli such as infection, nutritional or climatic changes and they may be accumulated in only certain parts of the plant. [53] In plants, phytochemicals act as a natural defense system for host plants and provide color, aroma and flavor. More than 4000 of these compounds have been discovered until date.

Besides, protection of plants these phytochemicals also possess therapeutic potentials such as anti-oxidant, anti-diabetic, memory enhancing, cholesterol lowering effects, adaptogenic property, anticancer, and immunomodulatory activity. The thousands of phytochemicals that have been discovered are grouped based on function and sometimes source. Natural compounds with potential immunostimulating activity can be classified as high- and low-molecular compounds. Terpenoids, phenolic compounds, and alkaloids dominate among low-molecular immunomodulatory compounds, polysaccharides dominate among the high-molecular weight compounds.[29]

MATERIALS AND METHODS

Ornamental

Many species of the *Lamiaceae* are attractive to grow in the garden, for example, the genus *Coleus aromaticus* Benth. *Coleus* is grown as an ornamental in Kalasalingam University, Krishnankoil, Tamil Nadu, India.

Botanical description

Coleus aromaticus Benth is a small plant with certain features that resemble a climber. Its stem is hairy, fleshy and attains a height of 1-3 feet. Leaves are 1-2 inch in length, roundish, dentate, hairy, thick and fleshy resembling heart. Flowers are small and blue or purplish in color with four petals, and it flowers in early summer. Family: *Lamiaceae* Genus: *Coleus* Species: *aromaticus* Botanical name: *Coleus aromaticus* Benth (Fig-1)

Organic compound

Preparation of crude extracts

Fresh leaves were collected, washed with distilled water, and cut into small pieces. These samples were ground and stored at 4°C until further use. Crude extracts (10% w/v) were made using 0.1% alkaline (KOH). The extracts were filtered through fine muslin cloth and the clear filtrate was mixed with 5% HCL precipitate the samples and dryness to form the precipitated sample under the shade condition.

X-Ray diffraction (XRD) study

XRD was done on pure Rutin and RN-Ps in different ratios of drug and PC to see the crystallinity in the substance. Sample was scanned in the angular range of 5°–80° in a PHILIPS XPert Pro X-Ray Diffractometer. Dried powder sample was kept in sample holder (20 mm × 15 mm × 2 mm) which was fitted into the instrument and X-ray was passed through the sample.

Thin Layer Chromatography

The crystal was subjected to thin layer chromatography. Various mobile phases were tried and the one with maximum number of spots was selected. The most suitable mobile phase was found to be Toluene: Dioxan: Glacial acetic acid (90:25:4). The crystal was dissolved in methanol and applied to pre-coated TLC silica gel plates (silica gel 60 F254 luGram, Germany). Chromatograms were developed and were examined under UV and daylight as well as after spraying within is aldehyde sulphuric acid reagent to detect the presence of different phytoconstituents.

Antimicrobial activity of the compound

Bacteria test organism was maintained on stock culture agar inoculated in nutrient broth. The broth seeds were inoculated at 37°C for 24 hours. The antibacterial assay was carried out by disc diffusion technique. Disc was impregnated with peak-1, peak-2 and peak-3, dried and then placed on the inoculated agar. The clear zone of inhibition around the disc was measured in millimeters.

RESULT AND DISCUSSION

Coleus aromaticus Benth medicinal value

It is a large succulent aromatic perennial herb with hispidly villous or tomentose fleshy stem. Leaves are simple, opposite, broadly ovate, crenate and fleshy. Flowers are pale purplish in dense whorls at distant intervals in a long slender raceme. Fruits are orbicular or ovoid nutlets. The leaves are useful in cephalalgia, otalgia, anorexia, dyspepsia, flatulence, colic, diarrhea, cholera, halitosis, convulsions, epilepsy, cough, asthma, hiccup, bronchitis, strangury, hepatopathy and malarial fever (Warrier *et al* 1995).



Fig-1. Shows potted *Coleus aromaticus* plants

Coleus aromaticus Benth plants have contributed a nanomedicine as a nutritive value rich in health to human beings (Table-1). Plant extracts and their bioactive compounds present in them which are responsible for antioxidant, antimicrobial, antitumor, and natural antibiotic anticancer activity have to be screened for their valuable information. A huge reservoir of bioactive compounds exists in many species of plants of Earth, only a small percentage of which have been examined and continued to be an important source of anticancer agents. Worldwide effects are ongoing to identify new anticancer compounds from plants. With the current decline in the number of new molecular entities from the pharmaceutical industry, novel anticancer agents are being sought from traditional medicines. Traditionally, drugs used for cancer chemotherapy are organic compounds, either synthetic or natural products and include alkylating agents, antibiotics, alkaloids, enzymes and hormones compare to our results positively (Praveena Bhatt and Pradeep S. Negi 2012)

Table-1 Shows in nutritive value in *Coleus aromaticus* plant.

Sr.No	Nutrients	Value in %
I	Proteins	0.6
II	Vitamins	
	Ascorbic acid	0.003± 0.1
	Thiamine	0.00008± 0.02
III	Minerals	
	Calcium	0.158± 0.032
	Phosphorus	0.016± 0.11
	Potassium	0.138± 0.12
	sodium	0.0047± 0.10
	magnesium	0.088± 0.001
IV	Trace metals	
	Iron	0.262± 0.05
	Zinc	0.0003± 0.10
	Copper	0.00011± 0.001
	Chromium	0.00002± 0.002
V	Soluble dietary fibers	0.31± 0.002
VI	Insoluble dietary fibers	1.56± 0.001
VII	Phytic acid	0.00092± 0.1
VIII	Soluble oxalate	0.02± 0.1
IX	Chlorophyll a	0.44 ±0.13
X	Chlorophyll b	0.29 ± 0.001
XI	Xanthophylls	0.35 mg/g
XII	β- carotene	0.0035mg/g

Nutritive value of *Coleus aromaticus* Benth

The present study was crystallization (82.3%) of aromatic compounds of *Coleus aromaticus* Benth for the first time from leaf extracts. The crystallized aromatic compounds were characterized by XRD (X-ray Powder Diffraction) and HPTLC for 35 types of compound were expressed, antibiotic activity was analyzed by peak-1, peak-2 and peak-3 specifically. The highest level was yield in *rosmarinic acid*, *chlorogenic acid*, *coumaric acid* and *caffeic acid* in the sample. However, the amount of organic compound was predominantly found in *rosmarinic acid* (100%), *chlorogenic acid* (31.6%), *coumaric acid* (35.6%) and *caffeic acid* (62.1%) for out of 35 different compounds in the crystal. The present study was crystallization (82.3%) of aromatic compounds of *Coleus aromaticus* Benth for the first time from leaf extracts. The crystallized aromatic compounds were characterized by XRD (X-ray Powder Diffraction) and HPTLC for 35 types of compound were expressed, antibiotic activity was analyzed by peak-1, peak-2 and peak-3 specifically. The highest level was yield in *rosmarinic acid*, *chlorogenic acid*, *coumaric acid* and *caffeic acid* in the sample. However, the amount of organic compound was predominantly found in *rosmarinic acid* (90%), *chlorogenic acid* (31.6%), *coumaric acid* (35.6%) and *caffeic acid* (62.1%) for out of 35 different compounds in the crystal. The result shows therapeutic potential of *Coleus aromaticus* which can be used as potential of nanomedicine as high nutritive value of human health.

Coleus aromaticus Benth crystal

This is the first time were precipitated fiber crystal of *Coleus* plant leaves of aromatic compounds (Fig-2 &3).

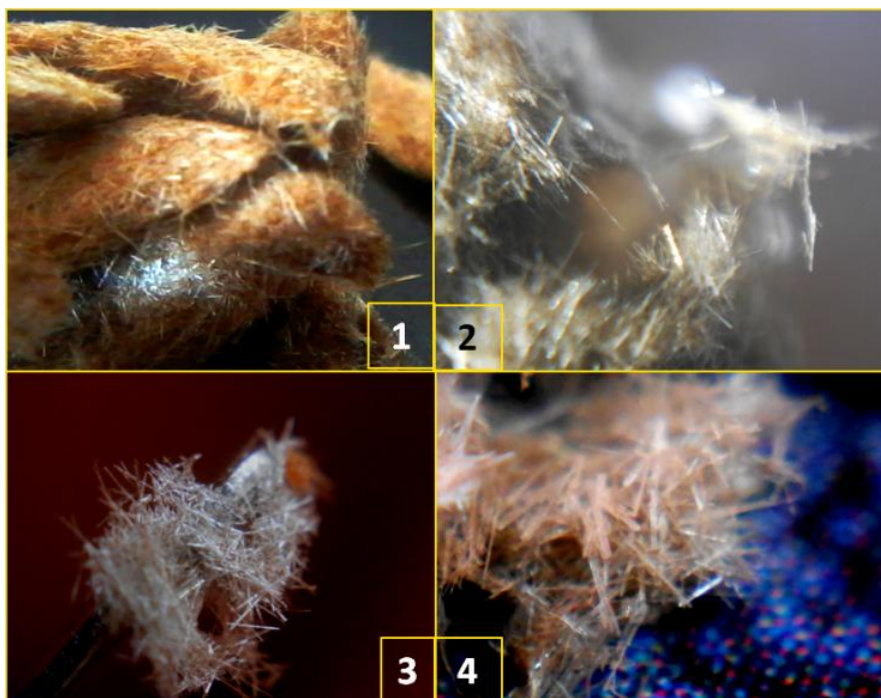


Fig-2. Shows that crystal of organic compound of *Coleus aromaticus* Plate 1-4 (10 times: Mobile photos).

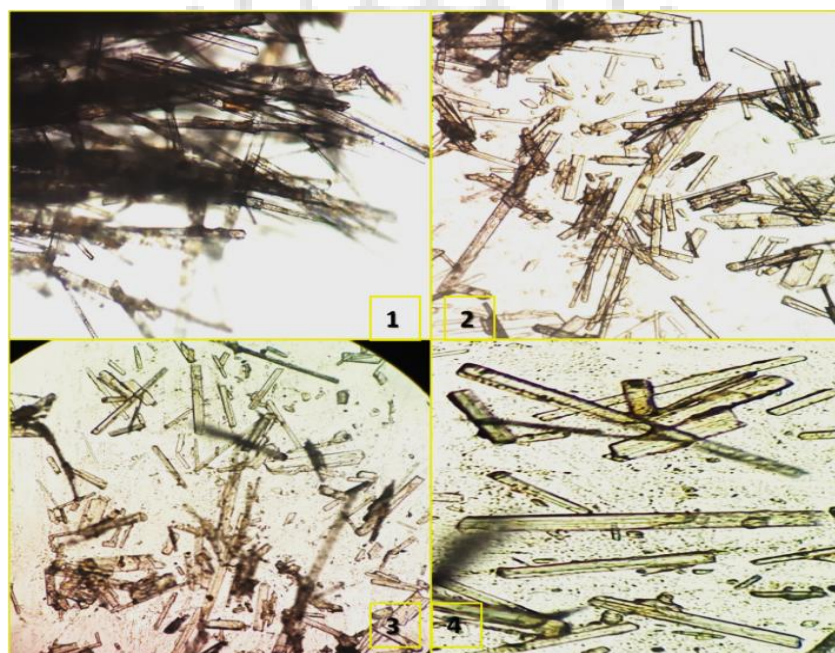


Fig-3. Shows that fiber crystal of organic compound of *Coleus aromaticus* Plate 1-4 (10 x 10 times for binocular basic microscope view).

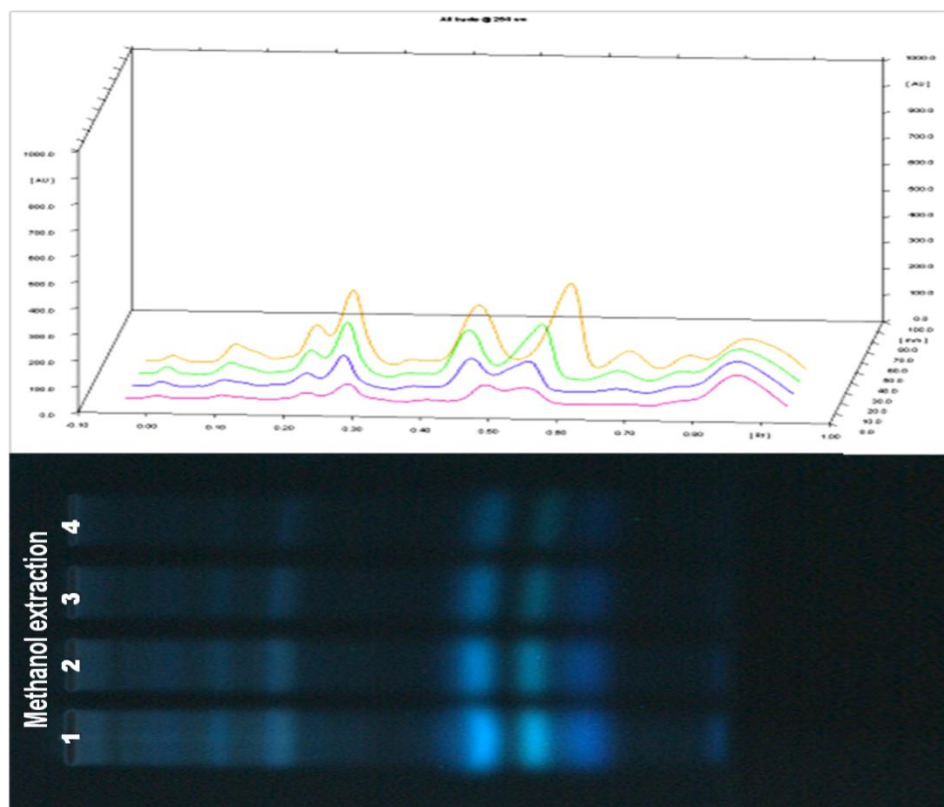


Fig 4- Shows the results of a. sample run the HPTLC and b. Sample run the TLC.

HPTLC

This technique can be used to rectify many qualitative and quantitative analytical problems in a wide range of fields including medicines, pharmaceutical, chemistry, biochemistry and toxicology. In the present study HPTLC analysis was carried out to confirm the presence of alkaloids and their derivatives in MGsSTL using colchicines as standard using ethyl acetate-methanol-water. The alkaloid peak was found to be colchicines in MGsS as its R_f value corresponds to the R_f value of the standard colchicines. Yellow, Orange yellow, Brownish yellow colored zone at Daylight mode were present in the track chromatogram. After derivatization, the chromatogram confirmed the presence of Alkaloid/Nitrogen-containing compound. The alkaloid profile, UV- visible chromatogram pattern, densitogram displays at 254 nm showed the presence of colchicines. The superimposed HPTLC fingerprints (Figure 4) also revealed the presence of alkaloids in MGsSTL. HPTLC fingerprinting is proved to be a linear, precise, accurate method for herbal formulation and can be used further in quality control of non-established herbals.

Thin Layer Chromatography (results)

The solvent system which showed maximum separation of constituents for all the extracts was- Toluene: Dioxan: Glacial acetic acid (90:25:4). In visible light and in UV both short and long wavelengths, the R_f value of quercetin matches with the R_f values of the first spots of all the three extracts. This indicates the presence of quercetin in all the three parts of the plant. In visible light and in UV-both short and long wavelengths leaf extract showed presence of about 10 components, whereas stem and root extract showed fewer components. But when sprayed with an anisaldehyde-sulphuric acid reagent, about 14 spots have been detected in both leaf and stem extracts, whereas about 16 spots detected with root extract. This indicates presence of more numbers of triterpenoids and saponins in the root extract (Figure 2).

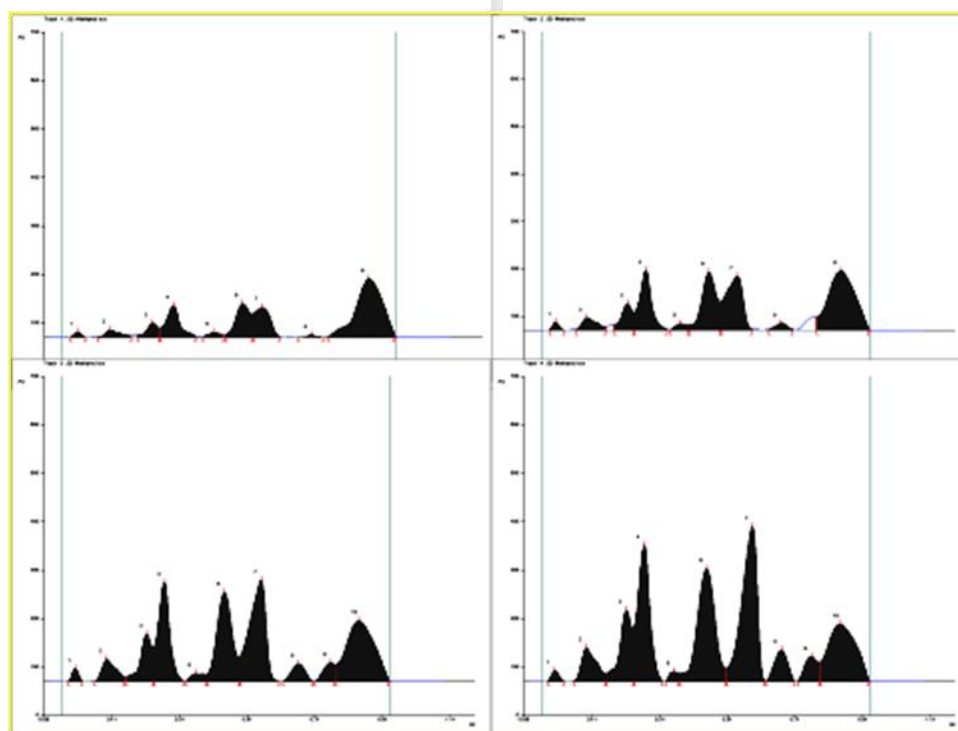
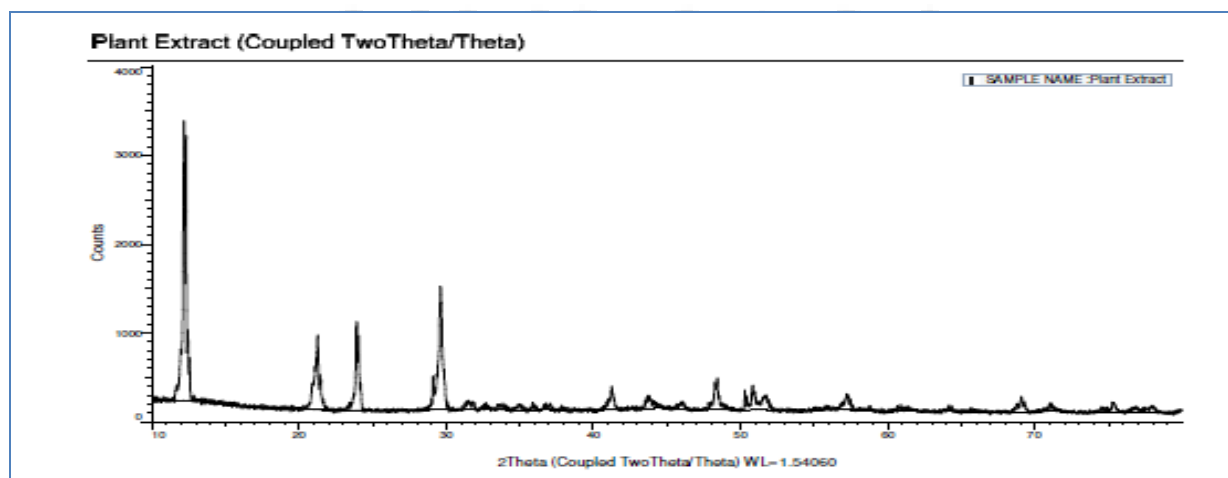


Fig-5. Shows the purification of crystal of *Coleus aromaticus* Benth from leaf extract for 60% of Methanol extract track-3 (1-2) and track-4 (3-4) results on the HPTLC.

Similarly presence of macrocyclic alkaloids- bud munchiamines in methanolic extract of *Albiziaamarawas* reported by Rajkumar and Sinha (2010).Methanol mixture of *Clerodendrum viscosum*vent roots showed bands for flavonoid (Prasanth *et al.*, 2012) in HPTLC analysis. *Spermacoce hispida* using HPTLC showed the presence of alkaloids, flavonoids, phenolics,

steroids, tannins and terpenoids (Rathi *et al.*, 2011). HPTLC analysis of N-hexane extract of the whole plant of *Emilia sonchifolia* confirmed the presence of terpenoids (Sophia *et al.*, 2011). Yamunadevi *et al.*, (2011) showed the presence of 30 different types of steroids in the methanolic extract of stem, leaves, root, flower and seeds of *Aervalanata*. Methanolic extract of *Albizialebeck* leaves demonstrated the presence of 4 different types of alkaloids (Bobby *et al.*, 2012). Lakshmi *et al.*, (2012) showed the presence of quercetin and rutin in the Acacia Catechu ethanolic leaf extract. The presence of alkaloids and phenolic compounds (Quercetin) of *Barleria cristata* Linn. Leaves; Ferulic acid in *Lycopodium clavatum* HPTLC fingerprint were reported by (Narmadha and Devaki 2012; Srivastava *et al.*, 2012). Cancer is the leading cause of death in developing countries like India. As there is an enormous increase in the population day by day, the alternative the rapy in the market is getting its glimpse. The cheap herbal drug is an ideal choice for the rural and poor people to treat effectively the cancers of various types may highly be recommended. Based on that the siddha medicines are coming up in combination with metals and other essential supplements to improve the immune status of the cancer patients in India. The above study revealed the role of *Gloriosa superba* plant and their phytochemicals may be effectively used for cancer treatment. The presence of colchicine, colchicine derivatives, unknown alkaloids and other related phytoconstituents may be responsible for their activity. Further studies are warranted, for the isolation of the constituents responsible for the activity and also to explore the exact mechanism of action.



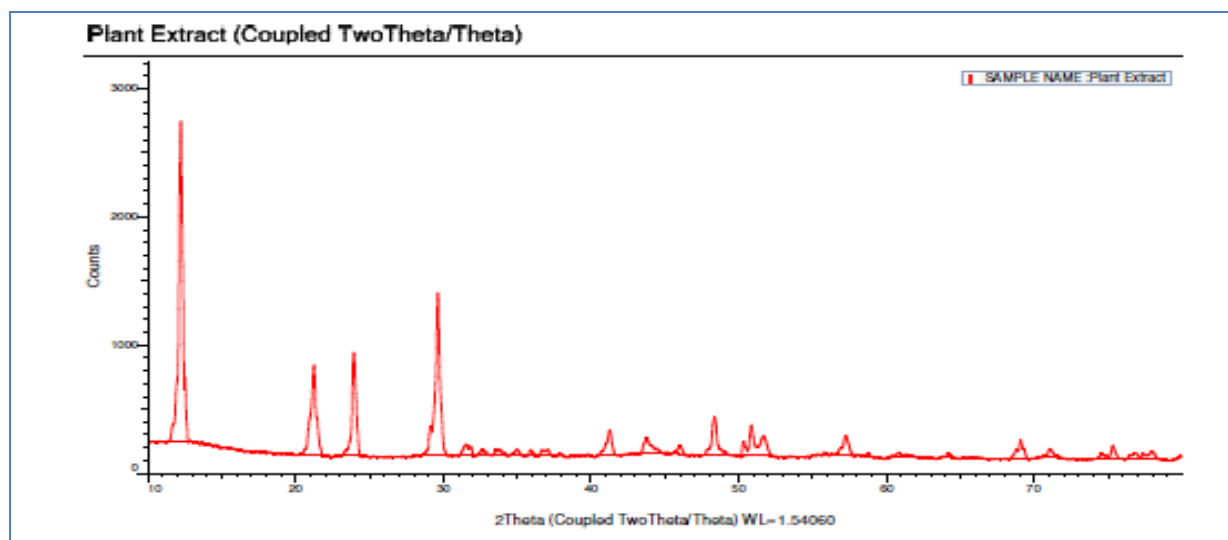


Fig 6. X-Ray Diffraction Analysis. Structural and crystalline nature of the phytochemical like silver nanoparticles has been performed using XRD analysis.

X-Ray diffraction study

XRD of RN shows intense crystalline peaks (Figure 1), indicating higher crystallinity of the drug. The XRD pattern indicates four distinct diffraction peaks at 2θ values of 12.23, 21.23, 23.96 and 29.63 Indexed as (111), (200), (220) and (311).

Coleus aromaticus leaf extract runs the TLC plate and bottom three peaks were analysis of antibacterial activity against was tested *Bacillus* sp. Peak 2 compare to peak-1 and peak-3 was more sensitive to *Bacillus* sp. and peak-3 resistant the bacterial activity.

Crystalline form of leaf extract was carried out by XRD. The biosynthesized organic compounds by employing *Coleus aromaticus* leaf extract was further demonstrated and confirmed by the characteristic peaks observed in the XRD image (Fig), 10.0° and 79.9° can be assigned the plane of 2197, 695, 781, and 1364 respectively indicates the high yield of rosmarinic acid, chlorogenic acid, coumaric acid and caffeic acid for crystalline in nature HPTLC.

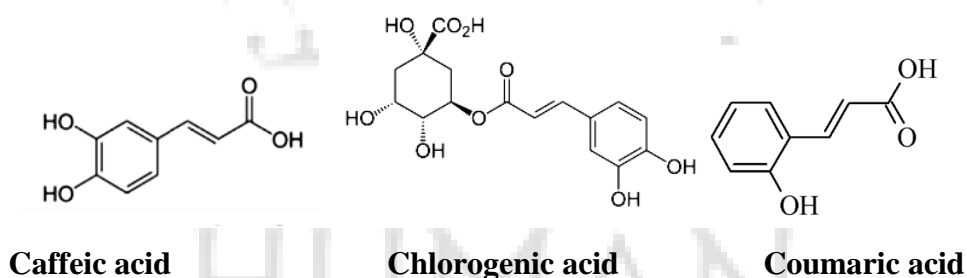
Organic compound

Fresh leaves of *C. aromaticus* were extracted with water & then separated into hexane, ethyl acetate and water fractions. Three components showing strong DPPH radical scavenging activity

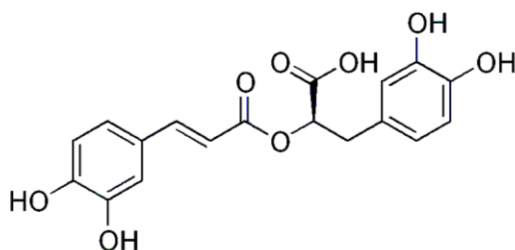
were shown to be rosmarinic acid, Chlorogenic acid, Caffeic acid Rosmarinic acid was major constituent for free radical scavenging potential (Shyama *et al* 2002).

Figure- 6 shows that the biosynthesized silver nanostructure by using *C. aromaticus* leaf extract was demonstrated and confirmed by the four characteristic peaks observed in the XRD image at 2θ values ranging from 30 to 90. The four intense peaks are 38.13° , 46.2° , 64.44° corresponding to the planes of (111), (200), (220), and (311), respectively.

The XRD pattern thus clearly showed that the Ag-NPs are crystalline in nature (Fig-2 &3). Similar report was obtained using cell filtrate of *Streptomyces* sp. ERI3 synthesized extracellular (Subramani, *et al* 1989). Caffeic acid is an inhibitory effect of caffeic acid on cancer cell proliferation by oxidative mechanism in human HT-1080 fibrosarcoma cell line has recently been established and inflammatory activity, antioxidant in vitro and also in vivo (Gurgel *et al* 2009; Castrillo *et al* 2001). Although, high doses of combined antioxidants, including caffeic acid, showed a significant decrease in growth of colon tumors in the rats (Damanik R, Torbangun 2009).



Chlorogenic acid is an important intermediate in lignin biosynthesis. This compound, known as an antioxidant, may also slow the release of glucose into the bloodstream after a meal (Lukhoba *et al* 2006). It is rich in chlorogenic acid found in *Coleus aromaticus* extract, as a food additive used in coffee products, chewing gum, and mints, and also as a stand-alone product. Coumaric acid is a hydroxycinnamic acid, an organic compound that is a hydroxy derivative of cinnamic acid a wide variety of edible plants such as peanuts, navy beans, tomatoes, carrots, garlic and rich in *Coleus aromaticus*. Antioxidant properties and is believed to reduce the risk of stomach cancer (Ragasa *et al* 1999) by reducing the formation of carcinogenic nitrosamines. Coumaric acid is normally present in honey, but not in the substitute nutrients based on high-fructose corn syrup.



Rosmarinic acid

Rosmarinic acid accumulation is maximum found in *Coleus aromaticus* belongs to *Lamiaceae* and shown in hornworts, in the fern family *Blechnaceae* and in species of several orders of mono- and dicotyledonous angiosperms (Khare CP 2007). It is found most notably in many *Lamiaceae* (dicotyledons in the order Lamiales), especially in the subfamily *Nepetoideae* (Lorimer *et al* 2009). It is found in species used commonly as culinary herbs such as *Ocimum basilicum* (basil), *Ocimum tenuiflorum* (holy basil), *Melissa officinalis* (lemon balm), *Rosmarinus officinalis* (rosemary), *Origanum majorana* (marjoram), *Salvia officinalis* (sage), thyme and peppermint (Epling 1981) Orin plants with medicinal properties such as common self-heal (*Prunella vulgaris*) or species in the genus *Stachys*.t is also found in other Lamiales such as *Heliotropium foertherianum*, a plant in the family *Boraginaceae*.

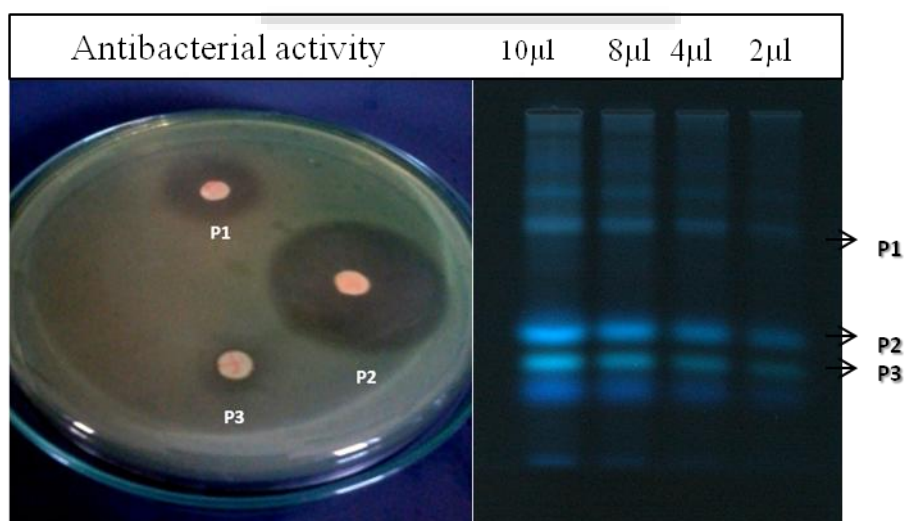


Fig 7. Shows that antibacterial activity against *Coleus aromaticus* leaf extract runs the TLC Plate in high three peaks 1-3 were analysis of antibacterial activity.

Sr.No	TLC plate samples	Zone formation	Diameter of the zone in cm	Organic compound in µg (single band)
1	Peak-1	+	1.2	0.02
2	Peak-2	+	2.7	2.2
3	Peak-3	+	0.2	0.001

Anti-Microbial effects of the plants leave extracts:

Microorganisms used

Clinical isolates of enteric pathogens viz., *E. coli*, *Salmonella typhi* and *Shigella* sp. were obtained from Meenakshi Mission Hospital & Research center, Madurai. The enteric pathogens were cultured in nutrient broth for 24 hours at 37 °C and the fresh inoculums were used for the test against the TLC plate from peak-1, peak-2 and peak-3 samples. (Kambizi, and Afolayan. 2008; Mohana *et al* 2008).

Coleus aromaticus Benth belongs to Lamiaceae had greater on the diameter of the inhibition zone for *S. aureus* than for *E. coli* strain, indicating that the gram-positive strain is more sensitive than the gram-negative strain. All leaves extracts from *Coleus aromaticus* Benth have having inhibitory effect on the gram positive bacteria (*Staphylococcus aureus*) compared to the gram negative bacteria (*Escherichia coli* and *Pseudomonas aeruginosa*). Generally, gram-negative bacteria are more resistant to plants extract compared to gram positive bacteria (Basri *et al.*, 2005). This may be due to the permeability barrier provided by the cell wall. The reason is that the density of the lipopolysaccharide layer in the outer surface of bacterial cell wall, which is greater in the gram-negative bacteria as compared to the gram-positive bacteria (Burn, 1988). It has been postulated that the antibacterial properties are of due to nucleotide damage with an increase in spatial division and condensation of genetic material (Chakraborty *et al.*, 2007). It is surprising that there are differences in the antibacterial activities of the different extracts tested. This could be due to the difference in the phytochemical composition among them (Dikbas *et al.*, 2009). Results show that the quantity of phenol among different *Lamiaceae* species studied does not necessarily affect the ability to stop the microbial growth. Baydar (2004) report that the greater the amount of phenolic compound in the extract would imply the greater inhibitory effect

of a particular extract on the test microorganism involved provided the test microorganism is sensitive to the extract.

Crystal particle size

Average particle size varied between the sizes of the crystals was 0.1 cm to 0.2 cm (Fig 2- 3). This is first was precipitated by acid alkali method.

Antibacterial activity

Not all the phyto compounds act as antibiotic activity, Figure – shows that high zone inhibition of peak-3 mediated biosynthesized than other peak-1 and peak-2 from leaf extract chemically synthesized antibiotic function. Biosynthesized organic compounds were exhibit more peak-3 more zone inhibition in *Bacillus* sp. was 2.7 cm respectively, than peak-1 and 2 for 50 µl of single band samples were loaded single disc. Our study coincided with the report of Mahitha *et al* (2011) and Malini *et al* (2013) they have examined the antibacterial effect of phytocompounds against the gram-positive *Bacillus* sp.

Immunostimulants of photochemical

Commonly are classified according to its chemical structures into nonflavonoids as phenolic acids and phenolic amides or flavonoids, subdivided by their substituents into isoflavones, neoflavonoids, chalcones, flavones, flavonols, flavanones, flavonols, flavanols, proanthocyanidins and anthocyanidins (Tsao, R 2010). Numerous studies have attributed to them a wide range of biological activities including anti-inflammatory (Recio 2012), antioxidant (Eberhardt 2000), cardiovascular protective (Andriantsitohaina 2012) and anti-cancer actions (Spagnuolo 2012), among many others (Vauzour 2010)]. The evidence about the effects of polyphenols on immune function is abundant; however, the mechanisms involved in these actions are not fully understood. Similar effects were obtained in response to treatment of aged rats with polyphenols from *Coleus aromaticus*, increasing splenic T and B cells.

CONCLUSION

To the understanding the nanoparticles from *Coleus aromaticus* leaves have rich in form of aromatic compounds such as *rosmarinic acid*, *chlorogenic acid*, *coumaric acid* and *caffeic acid*

was found and act as a immunomodulator of nanomedicine of human health. Health or medicinal foods have recently received immense interest among the health professionals urban and rural. The medicinal plant *Coleus aromaticus* have rich in secondary metabolites potentially sources of drugs and therapeutic importance. The important advantages claimed for therapeutic uses in various ailments are their safety besides being economical, effective and their easy availability. Because of these advantages, the medicinal plants have been widely used by the traditional medical practitioners in their day-to-day practice. However, the global health market has been swamped with such products claiming to improve health as well as prevent diseases by enhancing immune system. In addition, plant and originated/derived products have a potential application as an immunostimulant, primarily because they can be easily available, not expensive, act against a broad spectrum of pathogens and have no side effect. These are considered as an effective tool for enhancing immune status of cultured organisms too. It contains many immunological active components such as polysaccharides, organic acid, alkaloids, saponins, and glycosides which can enhance immune function. Because of increased commercial exploitation of medicinal foods, all varieties of fruits and vegetables even plants were re-evaluated for their photochemical compositions and health benefits. Recently, there has been increased in immune stimulating function of herbs.

ACKNOWLEDGEMENT

We are sincerely thankful to Director, Chancellor and Vice Chancellor, Kalasalingam University, Krishnankoil, Viruthunagar District, Srivilliputhur – 626 126 Tamil Nadu.

REFERENCES

1. Mahitha B, Deva Prasad Raju B, Dillip GR, Reddy CM, Mallikarjuna K, Manoj L, Priyanka S, Rao KJ, Sushma NJ. 2011. Biosynthesis, characterization and antimicrobial studies of Ag NPs extract from *Bacopamonniera* whole plant. *Dig J NanomaterBiostruct* 6 (1): 135-142.
2. Joshi M, Bhattacharyya A, Ali SW, Characterization techniques for nanotechnology applications in textiles, *Indian journal of fiber & textile research*, 33, 2008, 304–317.
3. M. Malini, G. Abirami, V.Hemalatha and G. Annadurai. 2013 Antimicrobial activity of Ethanolic and Aqueous Extracts of medicinal plants against waste water pathogens, *International Journal of Research in Pure and Applied Microbiology*, 3(2): 40-42.
4. J. Subramani, P. C. Josekutty, A. R. Mehta, and P. N. Bhatt, “Solasodine levels in *Solanum sisymbriifolium* Lam,” *Indian Journal of Experimental Biology*, vol.27,no.2,p.189,1989.
5. Pritima, R.A. and R.S.Pandian. (2008).Antimicrobial activity of *Coleus aromaticus* (benth) against microbes of reproductive tract infections amongwomen *Afr. J. Infect. Diseases*, 1(1): 18–24.

6. Warriar, P.K., V.P. Nambiar and K.Ramankutty. (1995) Indian medicinal plants, IEd, Orient Logman Limited: Madras, Pp. 315.
7. Jose, M.A., L. Ibrahim. and S.Janardhanan. (2005). Modulatory effect of *Plectranthus amboinicus* Lour. on ethylene glycol induced nephrolithiasis in rats. *Indian J Pharmacol.* 37: 43-47.
8. Prasad, S., P. Naik and K.K.Vijayalaxmi. (2002). Efficiency of *Coleus aromaticus* extract in modifying cyclophosphamide and mitomycin-C induced clastogenicity in mouse bone marrow cells. *Indian J ExpBiol.* 40: 1020-5.
9. Buznego, M.T. and H. Perez-Saad. (1999).Antiepileptic effect of *Plectranthus amboinicus* (lour.)Spreng. (*Frenchmarjoram*).*Rev Neurol.*, 29: 388-390.
10. Padma, P.R., V. Bhuvaneshwari. and S.K.Chelvi. (1988). The activities of enzyme antioxidant in selected green leaves. *Indian J Nutr Diet*, 35: 1-3.
11. Kambizi, L. and A.J. Afolayan. (2008).Extracts from *Aloe ferox* and *Withaniasomnifera* inhibits *Candida albicans* and *Neisseria gonorrhoea*. *African J.biotechnol.* 7: 12- 15.
12. Mohana, D.C., S. Satish and K.A.Raveesha (2008). Antibacterial evaluation of Some Plant Extracts against some human pathogenic bacteria. *Advances in Biological Research*, 2(3-4): 49- 55.
13. J. J. Gao, K. Igalashi and M. Nukina, "Radical Scavenging Activity of Phenylpropanoid Glycosides in *Caryopterisincana*," *Bioscience, Biotechnology and Biochemistry*, Vol. 63, No.6, 1999, pp. 983-988.doi:10.1271/bbb.63.983.
14. G. M. Williams, M. J. Iatropoulos and J. Whysner, "Safety Assessment of Butylated Hydroxyanisole and Butylated Hydroxytoluene as Antioxidant Food Additives," *Food Chemistry and Toxicology*, Vol. 37, 1999, pp. 1027-1038. doi:10.1016/S0278-6915(99)00085-X.
15. T. Osawa, and M. Namiki, "A Novel Type of Antioxidant Isolated from Leaf Wax of *Eucalyptus* Leaves," *Agriculture Biology and Chemistry*, Vol. 45, No. 3, 1981, pp. 735-739. doi:10.1271/bbb1961.45.735.
16. Praveena Bhatt and Pradeep S. Negi 2012. Antioxidant and Antibacterial Activities in the Leaf Extracts of Indian Borage (*Plectranthus amboinicus*), *Food and Nutrition Sciences*, 2012, 3, 146-152 <http://dx.doi.org/10.4236/fns.2012.32022>.
17. Warriar PK, Nambiar VP and Ramankutty C. Indian Medicinal Plants (Orient Longman Ltd., Madras) 1995.
18. Shyama PS, P Naik, KK Vijayalaxmi. Efficiency of *Coleus aromaticus* extract in modifying cyclophosphamide and mitomycin-C induced clastogenicity in mouse bone marrow cells. *J ExpBiol*, 2002, 40:1020-1025.
19. Lukhoba CW, Simmonds MSJ and Paton AJ, *Plectranthus*: A review of ethnobotanical uses, J. of Ethnopharmacology, 103: 1–24, (2006).
20. Ragasa CY, Sangalang V, Pendon Z and Rideout JA, Antimicrobial flavones from *Coleus amboinicus* Philippine J. of Sci. 128: 347-351, (1999).
21. Khare CP, Indian Medicinal Plants: an illustrated dictionary Library of congress Control Number: 2007922446 ISBN: 978- 0-387-70637-5 Springer-Verlag Berlin/ Heidelberg, Springer science+ Business Media, LLC (2007).
22. Lorimer M, Kanku JP, Signorini MA, Pardini A, *Plectranthus amboinicus*: an ornamental plant of medical and food interest ASAT Sci-Tec - Vol. 1, (2009).
23. Epling, P. *amboinicus* (Lour.) Spreng. Herbarium specimen held at K collected from the Pacific (Epling 18080). 1981.
24. Gurgel APAD, da Silva JG, Grangeiroa ARS, Oliveira DC, Lima CMP, da Silva ACP, Oliveira RAG and Souza IA, *In vivo* study of the anti-inflammatory and antitumor activities of leaves from *Plectranthus amboinicus* (Lour.) Spreng (Lamiaceae), J. of Ethnopharmacol, 125: 361–363, (2009).
25. Castrillo M, Vizcaino D, Moreno E and Latorraca Z, Chlorophyll content in some cultivated and wild species of the family *Lamiaceae*, *Biologia plantarum* 44(3): 432-425, (2001).
26. Damanik R, Torbangun (*Coleus amboinicus* Lour): a Batakese traditional cuisine perceived as lactagogue by Batakese lactating women in Simalungun, North Sumatera, Indonesia. *J Hum Lact.* 25(1): 64-72, (2009).
27. Warriar, P.K. V.P. Nambiar, Ramankutty (1995). Indian Medicinal Plants, 1st ed, Orient Longman Limited, Madras, Vol IV; 315-317.

28. Shamachar B.R. (1967). Use of Pashanabheda *Coleus aromaticus* in ashmari (Urinary calculi). *Natn. Med. J.*, 9, 475-477.
29. Kirtikar K.R., B.D. Basu (1935). *Indian Medicinal Plants*, Lalit Mohan Basu, Allahabad; India, Vol. III, 1970-1971.
30. Sivarajan V. V., I. Balchandran (1994). *Ayurvedic Drugs & Their Plant Sources*. Oxford & IBH Publishing Co Pvt.Ltd, New Delhi, 234.
31. Sharma P.V. (1995). *Dravyaguna Vijnana*. Chaukhamba Bharati Academy, Varanasi, India, Vol. II, 461-463. *Plectranthus*, (Last accessed on 10.06.2012) Available from: [www. Wikipedia. Org/wiki/ Plectranthus_am](http://www.Wikipedia.Org/wiki/Plectranthus_am).
32. Chatterjee A., S.C. Pakrashi (1997). *The Treatise on Indian Medicinal Plants*. National Institute of Science Communication, New Delhi, Vol. 5, 8-13.
33. Tsao, R. Chemistry and biochemistry of dietary polyphenols. *Nutrients* 2010, 2, 1231–1246.
34. Recio, M.C.; Andujar, I.; Rios, J.L. Anti-inflammatory agents from plants: Progress and potential. *Curr. Med. Chem.* 2012, 19, 2088–2103.
35. Eberhardt, M.V.; Lee, C.Y.; Liu, R.H. Antioxidant activity of fresh apples. *Nature* 2000, 405, 903–904.
36. Andriantsitohaina, R.; Auger, C.; Chataigneau, T.; Etienne-Selloum, N.; Li, H.; Martinez, M.C.; Schini-Kerth, V.B.; Laher, I. Molecular mechanisms of the cardiovascular protective effects of polyphenols. *Br. J. Nutr.* 2012, 108, 1532–1549.
37. Spagnuolo, C.; Russo, M.; Bilotto, S.; Tedesco, I.; Laratta, B.; Russo, G.L. Dietary polyphenols in cancer prevention: The example of the flavonoid quercetin in leukemia. *Ann. N. Y. Acad. Sci.* 2012, 1259, 95–103.
38. Vauzour, D.; Rodriguez-Mateos, A.; Corona, G.; Oruna-Concha, M.J.; Spencer, J.P. Polyphenols and human health: Prevention of disease and mechanisms of action. *Nutrients* 2010, 2, 1106–1131.

