SRI LANKAN MEDICINAL PLANT MONOGRAPHS AND ANALYSIS

VOL – 9

PIPER BETLE

Lakshmi Arambewela

Sachintha Alagiyawanna

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Lakshmi Arambewela and Sachintha Alagiyawanna

Edited by

Dilmani Warnasuriya

Industrial Technology Institute

(Ceylon Institute of Scientific and Industrial Research)

363, Bauddhaloka Mawatha

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Preface

Studies on medicinal plants of Sri Lanka have been carried out in the Herbal Technology Division of Industrial Technology Institute (former Ceylon Institute of Scientific and Industrial Research) for almost two decades. This monograph which is the nineth in this series incorporates information collected from literature surveys, researches and also experiences of the Herbal Technology Division staff. This monograph is intended for a varied reading public, herbal drug manufacturers who need to identify their herbal raw materials, Ayurvedic physicians who need some scientific information on medicinal plants, research workers requiring some quick background information on a plant, industrialists or entrepreneurs pondering on commercial ventures and the inquiring lay readers. We hope this monograph fulfils some requirements of each of them.

The authors wish to thank the members of the Herbal Technology Division for their contribution, the Information Service Center for providing information, Department of Plant Sciences and Department of Zoology of the University of Colombo for assisting in anatomical studies, Food Technology Division of the Industrial Technology Institute for helping in the analysis of powdered plant materials and the Microbiology Laboratory for photographing the slides. They also gratefully acknowledge the sponsor National Science Foundation for the research grant (RG / 2004 / TM / 01).

Herbal Technology Division
Industrial Technology Institute
P.O.Box 787
Colombo 07
Sri Lanka.

Piper betle Linn.

Family
Piperaceae

Synonyms
Chavica betle Miq., *Piper siriboa* Linn., *Chavica siriboa* Miq., *Piper betle* var. *siriboa* Cas. DC., *Piper peepuloides* Wall., *Piper amisumolens* Blanco., *Piper anisudorum* Blanco., *Piper philippinense* Cas. DC., *Piper blancoi* Merr., *Piper bathycarpum* Cas. DC., *Piper puberulinodum* Cas. DC., *Piper carnistylum* Cas. DC[1].

Selected Vernacular Names
Sinhala – Bulath, Bulath wel[1]

English – Betel, Betel vine, Betel leaf vine, Betel pepper[1,2,3,4,5]

Tamil – Ilaikkodi, Mellilai, Sukkli, Sulini, Sivanagini, Nirvalli, Pachadam, Tambulam, Vellilai, Vettilai, Vettilaichurul[1,3,4]

Hindi – Pan, Tambuli[1,4,5]

Sanskrit – Bhakshypatra, Bhujangalata, Bhujangavalli, Divabhishta, Kalaskanda, Nagavalli, Nagavallika, Nagini, Parna, Parnalata, Phanivalli, Saptalata, Tambulavallari, Tambuli, Tambulavalli, Vitika[1,4,5].

Arabic – Tanbol[2,3,4]

Burma – Kun, Kunyoe, Kwan, Kwanynet, Kwonrwet[2]

Cambodia – Mluw[2]

Chinese – Chu chiang[2]

French – Betel, Betre[2]

Java – Siri utan[2]

Malay – Sirih, Sirih china, Siri hudang, Siri malaya[2]

Philippines – Hojas de buyo, Poro[2]

Persian – Bargatanbol, Tambol[2]

Spanish – Betel, Betel de la India, Buyo de Filipinas[2]

Pharmacopoeia

Ayurveda Pharmacopoeia[5,6]

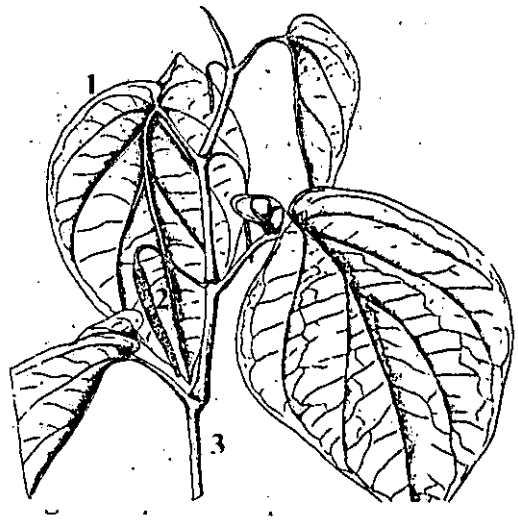
Pharmacopoeia Indica[7]

Distribution

Indigenous to and cultivated in Sri Lanka, India, Malay Peninsula, Philippine Islands and East Africa. It is commonly cultivated in home gardens in the low country of Sri Lanka[1,3].

Morphology[1]

A perennial vine climbing by many short adventitious rootlets up to about 20m. Stems semi-woody, very stout, much thickened at nodes, papillose when young, entirely glabrous; leaves simple, alternate, large, 15 – 20 cm long, some broadly ovate, cordate and symmetrical at base known as female leaves and others narrower and oblique called male leaves, thin, brittle, very acuminate, cordate at base, entire but margin often rather undulate, usually 7 veined, glabrous, thick, bright green and shining on both surfaces, petioles 1.8-2.5 cm long, stout, stipules membranous, enclosing the bud; flowers naked, unisexual, dioecious in dense cylindrical spikes, 2.5 – 5 cm long, pendulous, bracts triangular-rotundate, peltate, yellow, rachis pilose; ovary superior, unilocular with a solitary erect ovules, stigmas 5 or 6, spreading stellately; fruit sparingly produced, quite immersed in the fleshy spike forming nodosites and pendulous.



1. Betel leaf 2. Inflorescence 3. Stem

(Source – Jayaweera, D. M. A., (1981). Medicinal plants used in Ceylon Part iv)

Part Used [3,5,7]

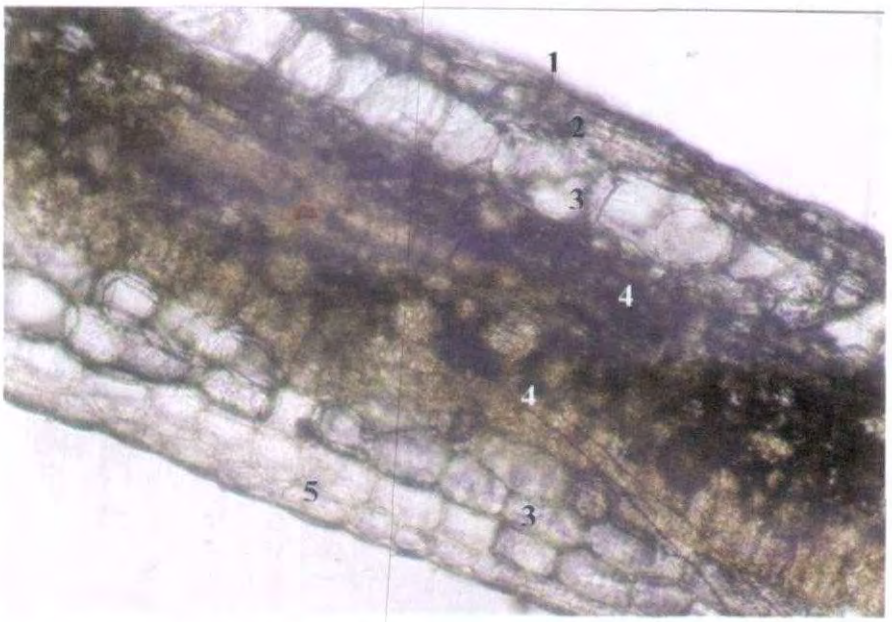
Whole vine; leaves, roots and inflorescence.

Official Drug [5,25]

Leaves, juice of leaves and oil of leaves.

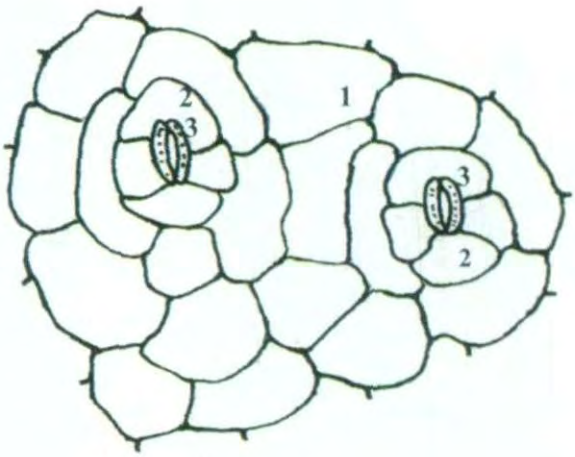
Pharmacognostic Features*

Anatomy



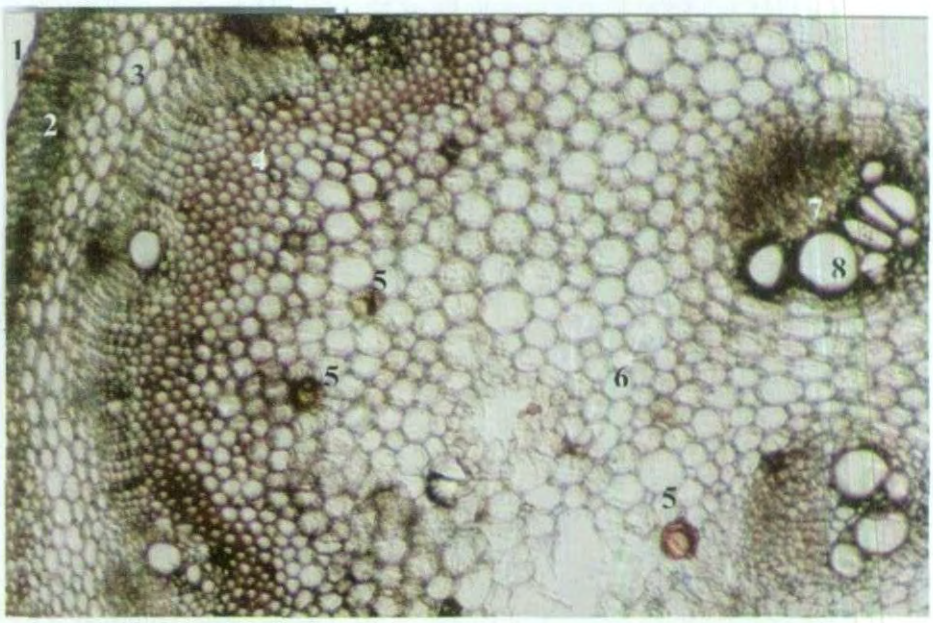
Fig– 2. Cross section of *Piper betle* leaf (stained with safranine (10 x 10))

1. Cuticle 2. Upper epidermis 3. Spongy parenchyma 4. Palisade cells 5. Lower epidermis



Fig– 3. Schematic diagram of epidermis of leaf

1. Epidermis cells 2. Subsidiary cells 3. Guard cells



Fig– 4. Cross section of *Piper betle* stem (stained with safranine (10 x10))

1. Epidermis 2. Collenchyma 3. Cortex parenchyma 4. Sclerenchyma

5. Oil vesicles 6. Pith 7. Phloem 8. Xylem

Powder analysis

Part analyzed – Leaf

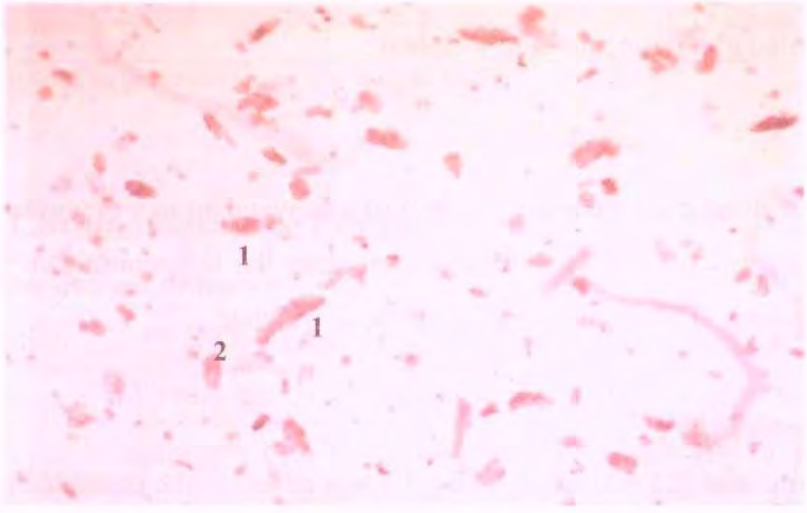
Organoleptic properties

Colour – Pale green

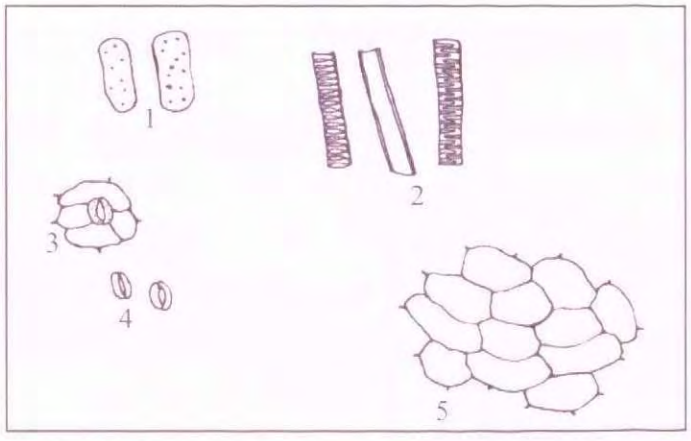
Odour – Aromatic

Taste – Pungent

Microscopic characters



Fig– 5. Powdered plant material (leaf) under the microscope (10 x 4)
1. Parenchyma cells 2. Vessel segment



Fig– 6. Schematic diagram of powder microscopy

1. Palisade cells 2. Vessel segments with different thickenings 3. Stomata with subsidiary cells 4. Stomata 5. Epidermal cells

* These analysis were carried out by the authors at Industrial Technology Institute and the Dept. of Plant Sciences and Dept. of Zoology of University of Colombo.

Physico-chemical Analysis[108]

Extractable matter

Crushed, air dried plant material (about 4 g) was weighed to a glass-stoppered conical flask. Solvent (100 mL) was added, weighed, shaken well and allowed to stand for 1h. Then it was boiled for 1h and cooled. The weight was readjusted with specified solvent and filtered. Filtrate (25 mL) was taken, solvent was evaporated and oven dried at 105 ^{0}C for 6 h, cooled in a desiccator and weighed.

Total ash

Crushed, air dried plant material (about 4 g) was weighed to a previously ignited crucible. The material was ignited by gradually increasing the temperature to 550 ^{0}C until it was free from carbon. The crucible was cooled and weighed.

Acid insoluble ash

Hydrochloric acid (25 mL, conc. ~70 g/L) was added to the crucible containing total ash, covered with a watch glass and boiled gently for 5 min. The insoluble matter was collected on an ashless filter paper and washed with hot water until the filtrate was neutral. The filter paper containing the insoluble matter was transferred to the original crucible and ignited to a constant weight.

Water soluble ash

Water (25 mL) was added to the crucible containing total ash, covered with a watch glass and boiled gently for 5 min. The insoluble matter was collected on an ashless filter paper and washed with hot water. The filter paper containing the insoluble matter was transferred to the original crucible and ignited for 15 min. at a temperature not exceeding 450 ^{0}C. Water soluble ash is the calculated difference in weight between the total ash and the residue remaining after treatment of the total ash with water.

Moisture content of the samples was estimated and all the calculations were done on dry weight basis.

Table 1. Physico-chemical parameters of *Piper betle* leaf [**]

Physico-chemical parameter	Amount %
1. Water extractable matter	20.8 - 22.5
2. Ethanol extractable matter	14.9 - 16.2
3. Total ash	13.9 - 14.5
4. Water soluble ash	8.5 - 9.3
5. Acid insoluble ash	0.07 - 0.08

(Results are expressed as percentages on dry weight basis)

Thin Layer Chromatographic Profile[**]

***Piper betle* water extract of leaves**

Sample preparation : *P. betle* leaves (4 g) were boiled for one hour with water (100 mL) and the extract was filtered and evaporated to dryness. Ten microliters (10 µL) of the diluted extract (0.1 g in 5 mL) was spotted on TLC plate.

Absorbent : Silica gel GF_{254}

Solvent system : Methanol : Butanol : Hexane (1 : 1 : 1)

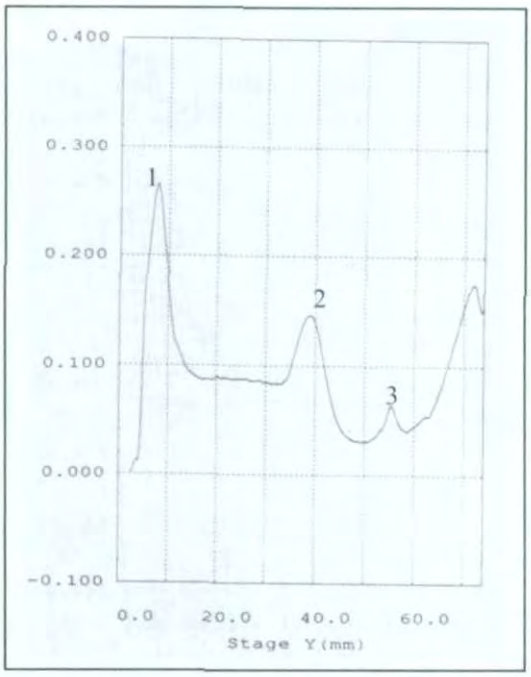
Detection

Direct evaluation : UV_{254} nm, R_f values – 0.58, 0.91

Scanning : Densitometer at 254 nm (before spraying) and 450 nm (after spraying)

Spray reagent : Vanillin sulphate

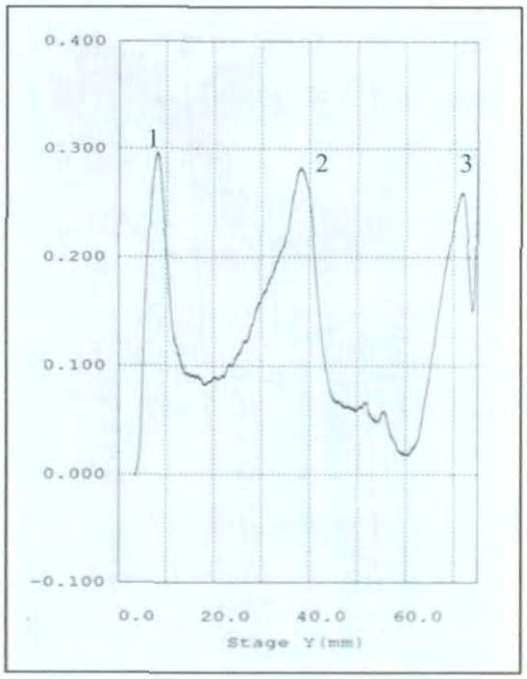
Fig– 7. TLC finger print profile of water extract of *Piper betle* leaves



Fig– 8. Densitogram of TLC finger print profile of water extract of *Piper betle* leaves at 254 nm

Table 2. Description of densitogram (Fig-8)

Peak no.	Y (mm)	Relative area %
1	7.71	68.21
2	38.94	27.74
3	55.44	4.05



Fig– 9. Densitogram of TLC finger print profile of water extract of *Piper betle* leaves at 450 nm

Table 2. Description of densitogram (Fig-9)

Peak no.	Y (mm)	Relative area %
1	8.13	27.96
2	38.20	56.53
3	71.58	11.57

***Piper betle* ethanol extract of leaves**

Sample preparation	: *P. betle* leaves (4 g) were boiled for one hour with 95% ethanol (100 mL) and the extract was filtered and evaporated to dryness. Six microliters (6 µL) of the diluted extract (0.06 g in 3 mL) was spotted on TLC plate.
Absorbent	: Silica gel GF_{254}
Solvent system	: Methanol : Chloroform : Cyclohexane (1 : 10: 14)

Detection

Direct evaluation	: UV_{254} nm, R_f values – 0.07, 0.23, 0.49, 0.81, 0.91
Scanning	: Densitometer at 254 nm (before spraying) and 450 nm (after spraying)
Spray reagent	: Vanillin sulphate

Fig– 10. TLC finger print profile of ethanol extract of *Piper betle* leaves

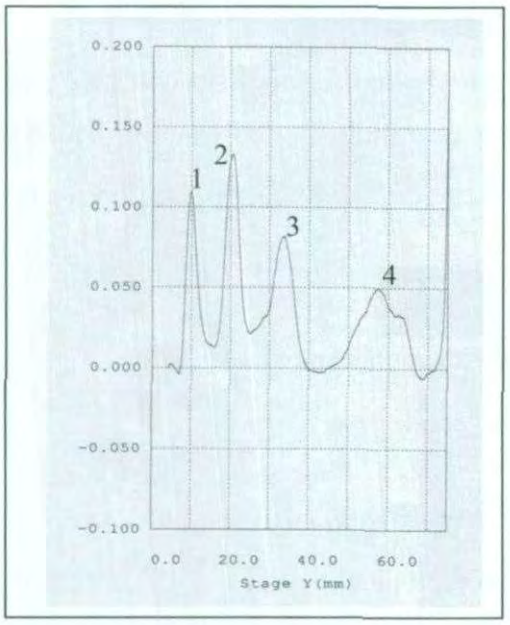


Table 4. Description of densitogram(Fig-11)

Peak no.	Y (mm)	Relative area %
1	9.91	17.75
2	20.56	26.67
3	33.54	27.25
4	57.46	28.33

Fig– 11. Densitogram of TLC finger print profile of ethanol extract of *Piper betle* leaves at 254 nm

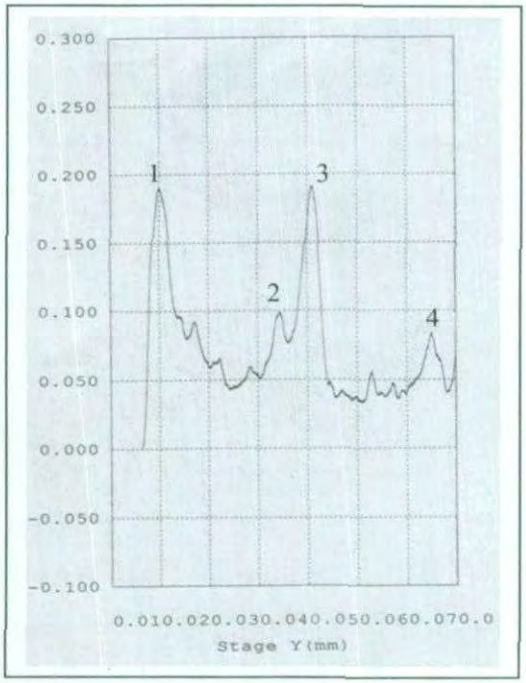


Table 5. Description of densitogram(Fig-12)

Peak no.	Y (mm)	Relative area %
1	10.02	33.68
2	34.47	10.01
3	40.98	26.64
4	65.05	5.93

Fig– 12. Densitogram of TLC finger print profile of ethanol extract of *Piper betle* leaves at 450 nm

High Pressure Liquid Chromatographic Profile**

Piper betle water extract of leaves

Sample preparation : *P. betle* leaves (4 g) were boiled for one hour with water (100 mL) and the extract was filtered and evaporated to dryness. The diluted extract (11.5 mg in 5 mL) was purified using Sep-pak C18 cartridge.

Injection volume : 20 µL

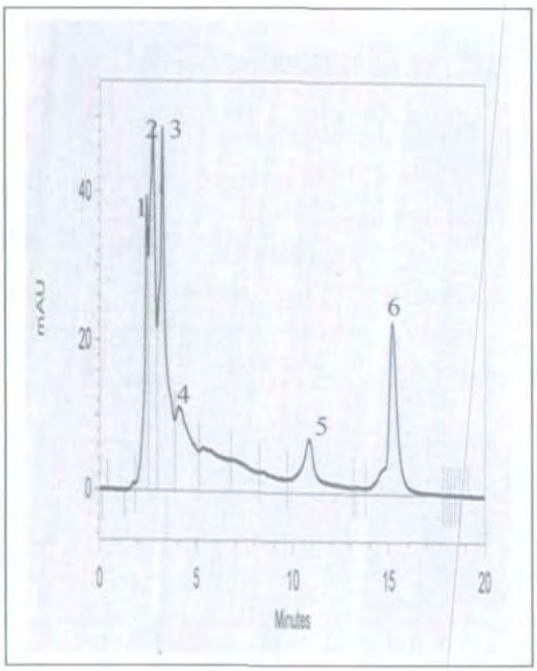
Apparatus : Shimadzu LC – 10 ADvp pumps and Shimadzu SPD – M 10 Avp uv / vis photodiode array detector.

Column : Inertsil 5U ODS – 2 reverse phase column, (250 mm x 2.6 mm)

Solvent system : Acetonitrile : Water (30 : 70)

Flow rate : 1 mL/min

Detection : 254 nm



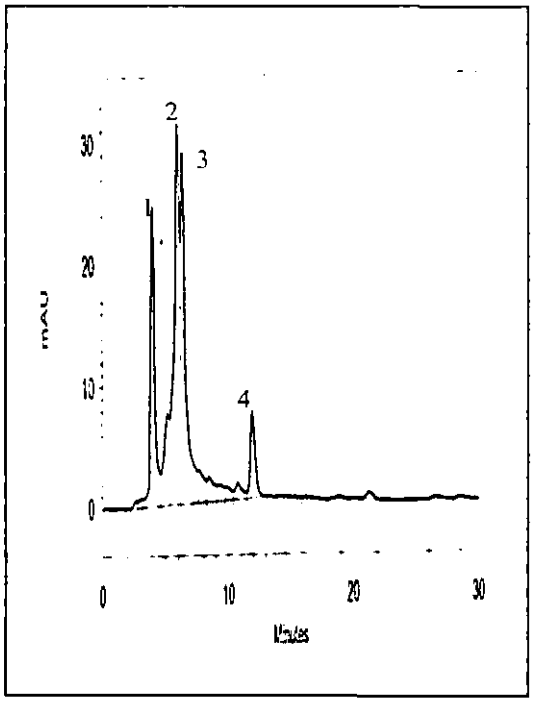
Fig– 13. HPLC finger print profile of water extract of *Piper betle* leaves

Table 7. Retention times of main peaks

Peak no.	Retention time (min)	Relative area %
1	2.42	8.42
2	2.74	17.87
3	3.23	21.26
4	4.06	10.93
5	10.86	8.74
6	15.19	14.48

***Piper betle* ethanol extract of leaves**

Sample preparation	: *P. betle* leaves (4 g) were boiled for one hour with 95% ethanol (100 mL) and the extract was filtered and evaporated to dryness. The diluted extract (5 mg in 5 mL) was purified using Sep-pak C18 cartridge.
Injection volume	: 20 µL
Apparatus	: Shimadzu LC – 10 ADvp pumps and Shimadzu SPD – M 10 Avp uv / vis photodiode array detector.
Column	: Inertsil 5U ODS – 2 reverse phase column, (250 mm x 2.6 mm)
Solvent system	: Methanol : Water (80 : 20)
Flow rate	: 0.6 mL/min
Detection	: 254 nm



Fig– 14. HPLC finger print profile of ethanol extract *Piper betle* leaves

Table 8. Retention times of main peaks

Peak no.	Retention time (min)	Relative area %
1	3.94	19.66
2	5.94	27.90
3	6.32	26.68
4	11.90	6.05

**These analyses were carried out by the authors at the Industrial Technology Institute.

Phytochemistry

The following chemical constituents have been identified in volatile oil (betel oil) of betel grown in Sri Lanka[8,9].

Leaf oil – β-phellandrene, γ-muurolene, α-humulene, γ-selinene, β-cadinene, allyl pyrocatechol diacetate.

Stem oil – α-pinene, 4-terpineol, safrole, eugenol, chavibitol acetate, γ-muurolene, camphene, α-elemene, β-phellandrene, α-humulene, γ-selinene, β-cadinene, allyl pyrocatechol diacetate.

Stalk oil – 4-terpineol, safrole, eugenol, chavibitol acetate, γ-muurolene, α-humulene, γ-selinene, β-cadinene.

Fruit oil – α-pinene, camphene, β-phellandrene, γ-muurolene, α-humulene, γ-selinene, β-cadinene, allyl pyrocatechol diacetate, 4-terpineol, ρ-cymene, limonene, safrole, eugenol, chavibitol acetate, β-caryophyllane.

Root oil – α-pinene, camphene, β-phellandrene, 1,8-cineol, limonene, α-terpineol, safrole, eugenol, chavibitol acetate, α-elemene, β-caryophyllane, γ-selinene, β-cadinene, allyl pyrocatechol diacetate.

In Indian betel varieties α-thujene, campene, sabinene, β-myrcene, β-ocimene, bornylene, β- pinene, trans 3-ocimene, γ-terpinene, terpinolene, α-terpinene, β-phellandrene, limonene, ρ-cymene, α- pinene γ-cadinene, Δ- cadinene, α-cadinene, β-selinene, β-elemene, γ-elemene, caryophyllene, aromadendrene, α-cubenene, β- cubenene, linalool, α-terpineol, α-castol, Δ-cadinol, decanal, stearaldehyde, geraniol, hexadecanoic acid, 1,8-cineol, carophyllene oxide, eugenol, isoeugenol, chavibetol, methyleugenol, methylchavicol, anethole, eugenol acetate, methyl benzoate have been reported in leaves[10,11,12,13]. The leaves also contain alkaloids, arachene, terpenes, sesquiterpenes amino acids, vitamins (B and C), carotene and minerals[10,19].

The following chemical constituents have been found in Taiwan varieties[14,15,16]

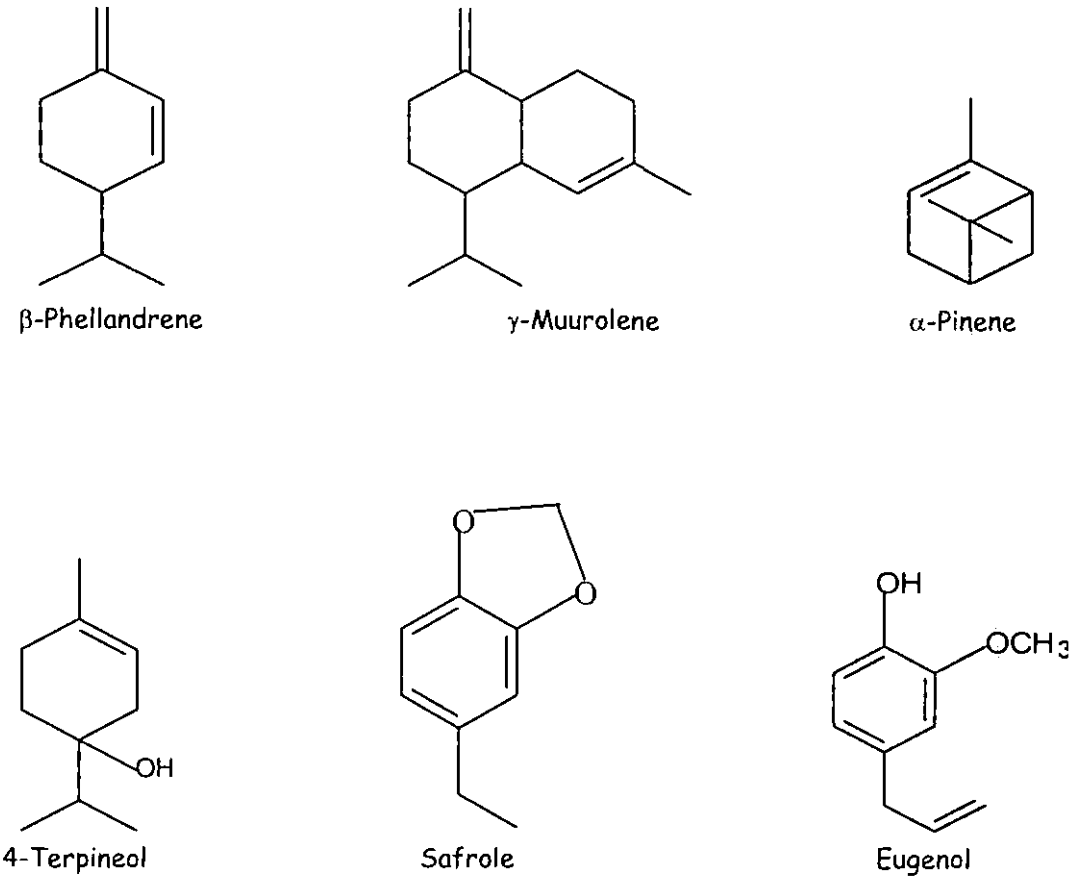
Leaf – chavibetol

Flower essential oil – hydroxy chavicol

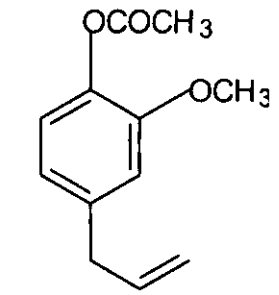
Inflorescence – eugenol, myrcene, allyl pyrocatechol, safrole, tocopherol.

In Philippines varieties camphene, caryophyllene, chavibetol, chavibetol acetate, 1,8-cineol, ρ-cymene, eugenol, eugenol methylester, limonene, α-pinene, β-pinene, allyl pyrocatechol, allyl pyrocatechol monoacetate, allyl pyrocatechol diacetate, safrole have been reported[17].

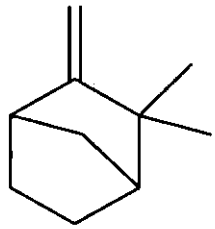
In Pakistan betel varieties β-sitosterol, ursonic acid, ursolic acid 3-β-acetyl have been reported in leaves[18].



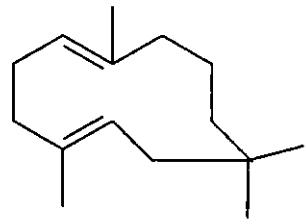
Fig– 15. Compounds present in *Piper betle*



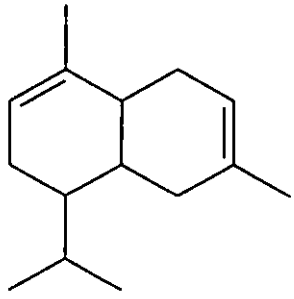
Chavibetol acetate



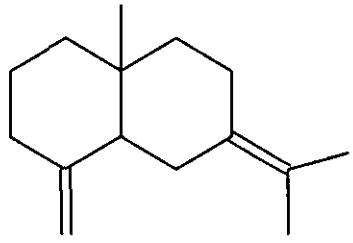
Camphene



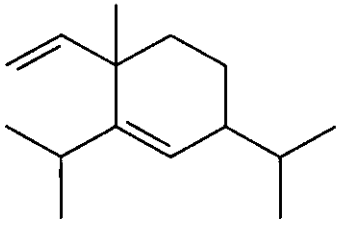
α-Humulene



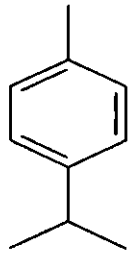
β-Cadinene



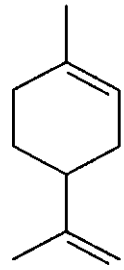
γ-Selinene



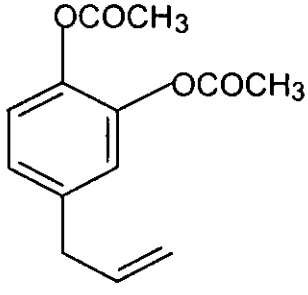
α-Elemene



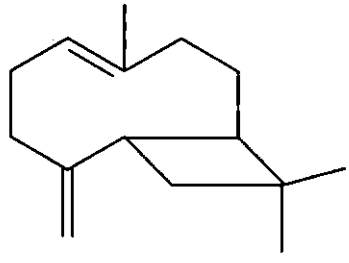
ρ-Cymene



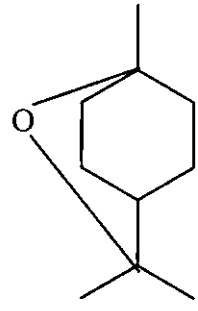
Limonene



Allyl pyrocatechol diacetate

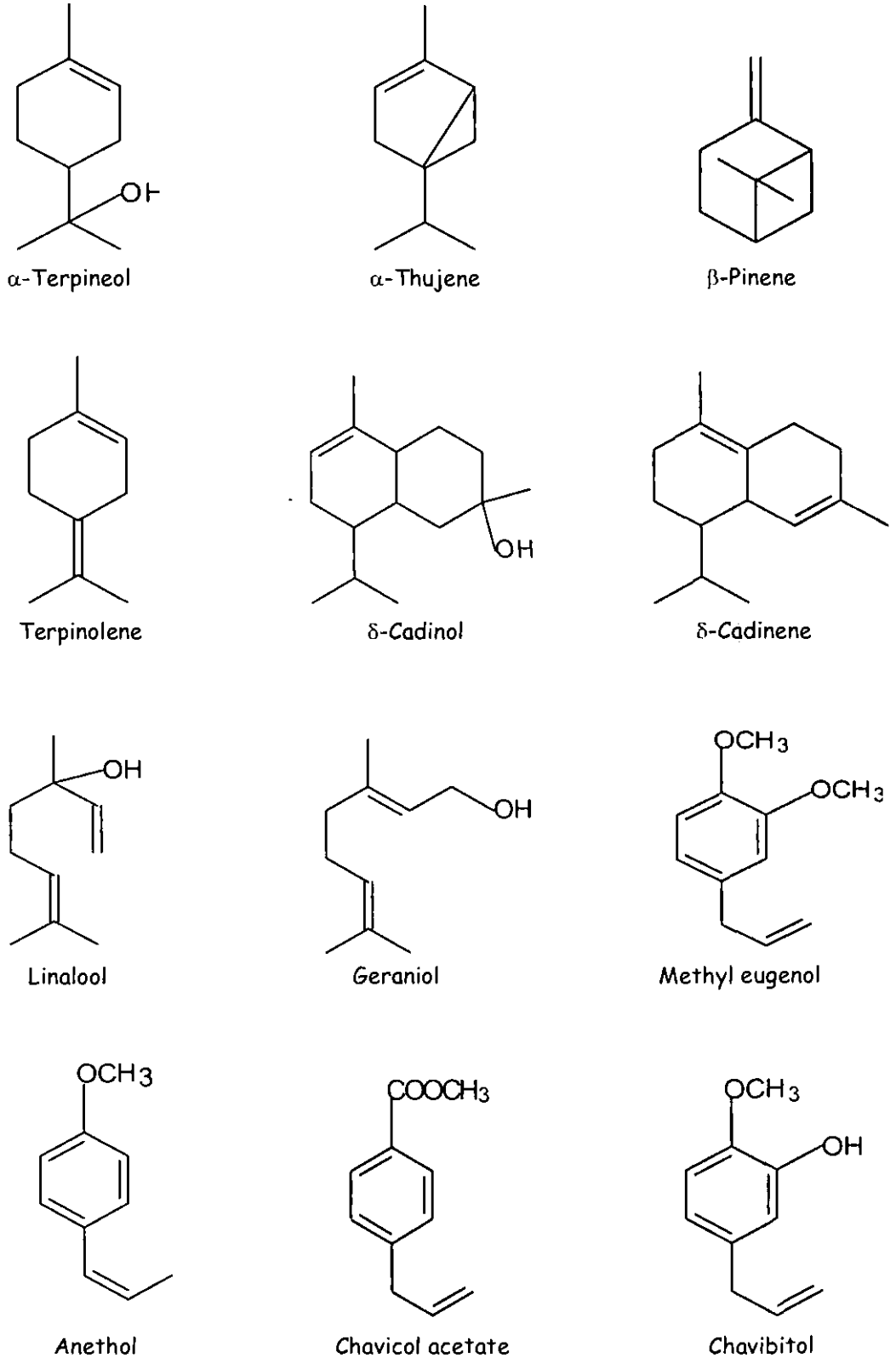


β-Caryophyllene



1,8-Cineol

Fig– 16. Compounds present in *Piper betle*



Fig– 17. Compounds present in *Piper betle*

Gas Chromatographic Profile of Essential Oil of *Piper betle* [8]

Method of distillation of oil

Air dried leaves (251.9 g), stems (175.8 g) fruits (176.7 g), roots (160.7 g) and leaf stalks (245 g) were separately hydrodistilled (2 L of water) for 4 hours. A Clevenger-type oil arm was used to collect the oils of leaves (1.05%), stems (0.08 %), fruits (3.78 %), roots (0.09 %) and leaf stalks (0.93 %). The oil contents are expressed on dry weight basis.

Details of the gas chromatographic operating conditions

Chromatograph – Hewlett-Packard 5890 Series II

Detector – Flame ionization detector

Column – DB-5 MS capillary column (30m x 0.25 mm id., 0.25 μm film)

Initial oven temperature – 40 ^{0}C

Final oven temperature – 280 ^{0}C

Program rate – 10 ^{0}C/min

Injector temperature – 550 ^{0}C

Detector temperature – 550 ^{0}C

Carrier gas – Argon

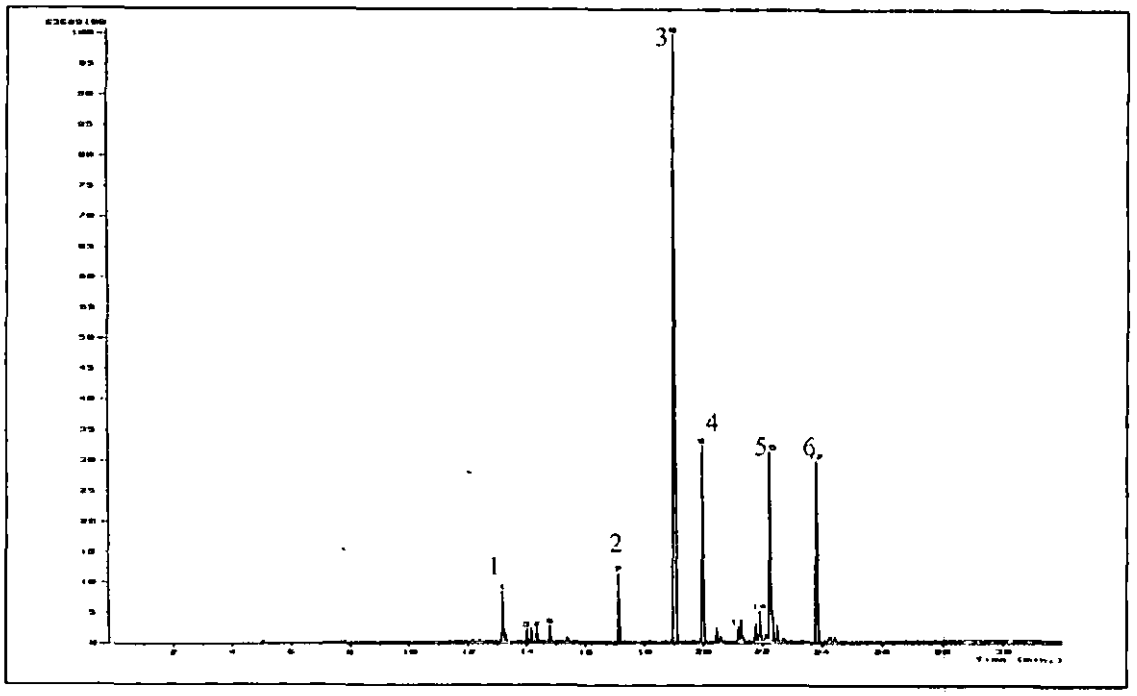
Flow rate – 30 mL/min

Hydrogen flow rate – 25 mL/min

Air flow rate – 30 mL/min

Sample size – 1 μL

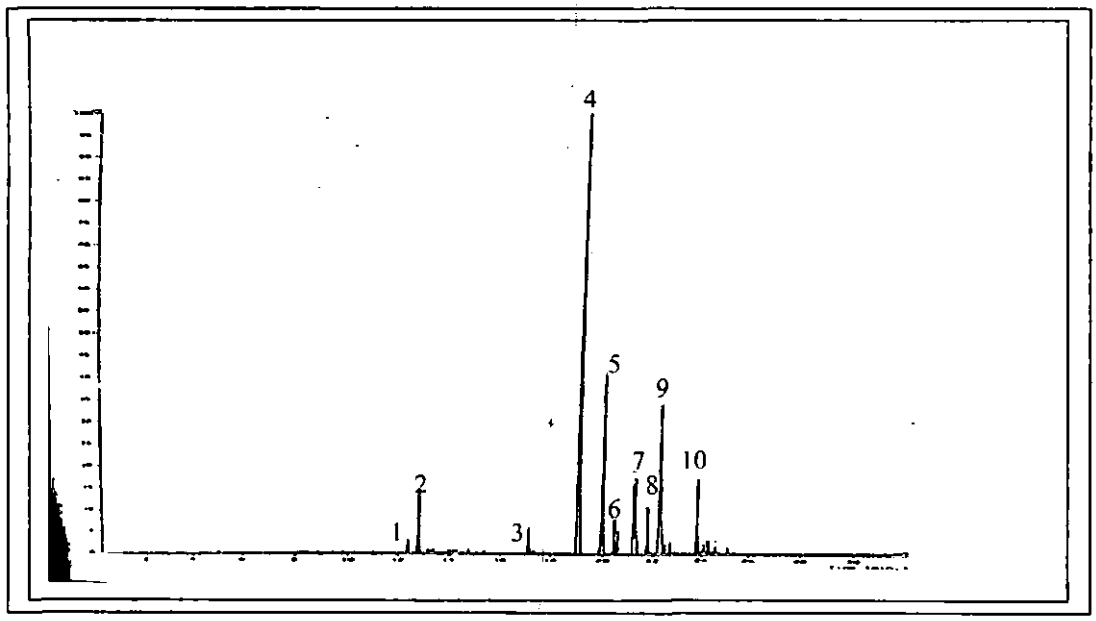
The concentration of the compounds was determined by comparing the peak area of the compound with the total area of the peaks in the chromatogram.



Fig– 18. Gas liquid chromatogram of common betel leaf oil[8]

Table 2. *Description of the gas liquid chromatogram of common betel leaf oil*

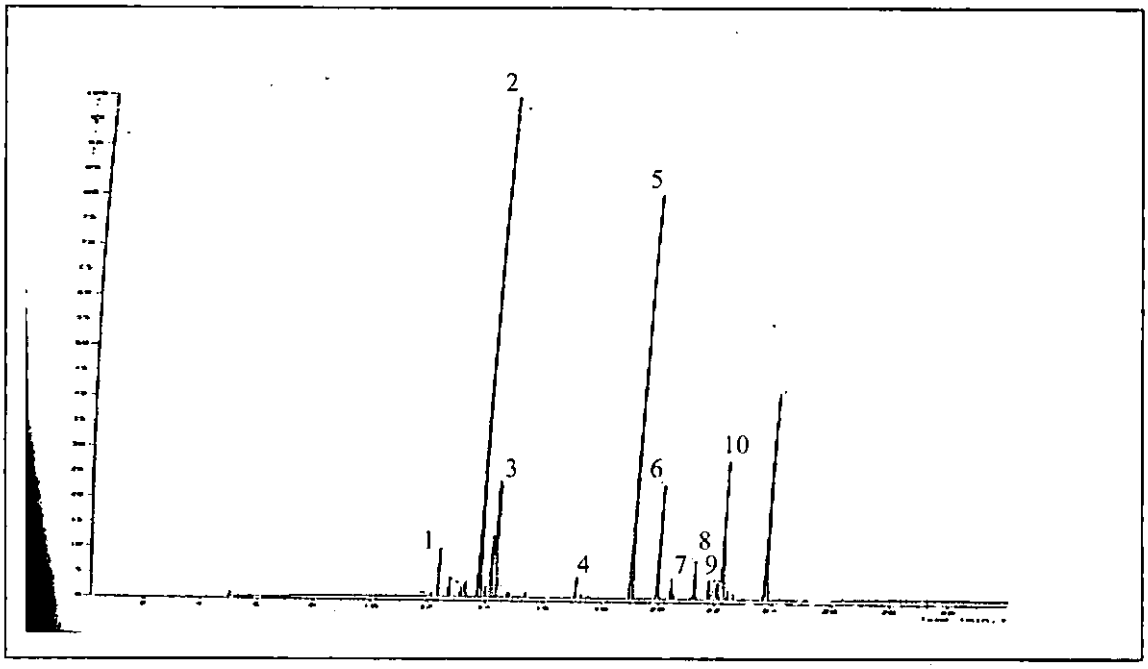
Peak no.	**Compound**	**Relative area %**
1	β-Phellandrene	2.58
2	4-Terpineol	3.61
3	Safrole	48.69
4	Eugenol	11.93
5	Chavibitol acetate	12.55
6	Allyl pyrocatechol diacetate	11.34



Fig– 19. Gas liquid chromatogram of common betel stem oil[8]

Table – 3. Description of the gas liquid chromatogram of common betel stem oil

Peak no.	Compound	Relative area %
1	α-Pinene	0.82
2	Camphene	4.06
3	4-Terpineol	1.51
4	Safrole	41.74
5	Eugenol	13.19
6	γ-Muurolene	2.15
7	β-Caryophyllene	5.03
8	α-Humulene	2.97
9	Chavibitol acetate	10.30
10	Allyl pyrocatechol diacetate	5.14



Fig– 20. Gas liquid chromatogram of common betel fruit oil[8]

Table 4. Description of the gas liquid chromatogram of common betel fruit oil

Peak no.	Compound	Relative area %
1	α-Pinene	2.31
2	β-Phellandrene	25.04
3	Limonene	5.29
4	4-Terpineol	0.98
5	Safrole	22.16
6	Eugenol	6.11
7	γ-Muurolene	0.98
8	β-Caryophyllene	1.83
9	Chavibitol acetate	7.24
10	Allyl pyrocatechol diacetate	0.45

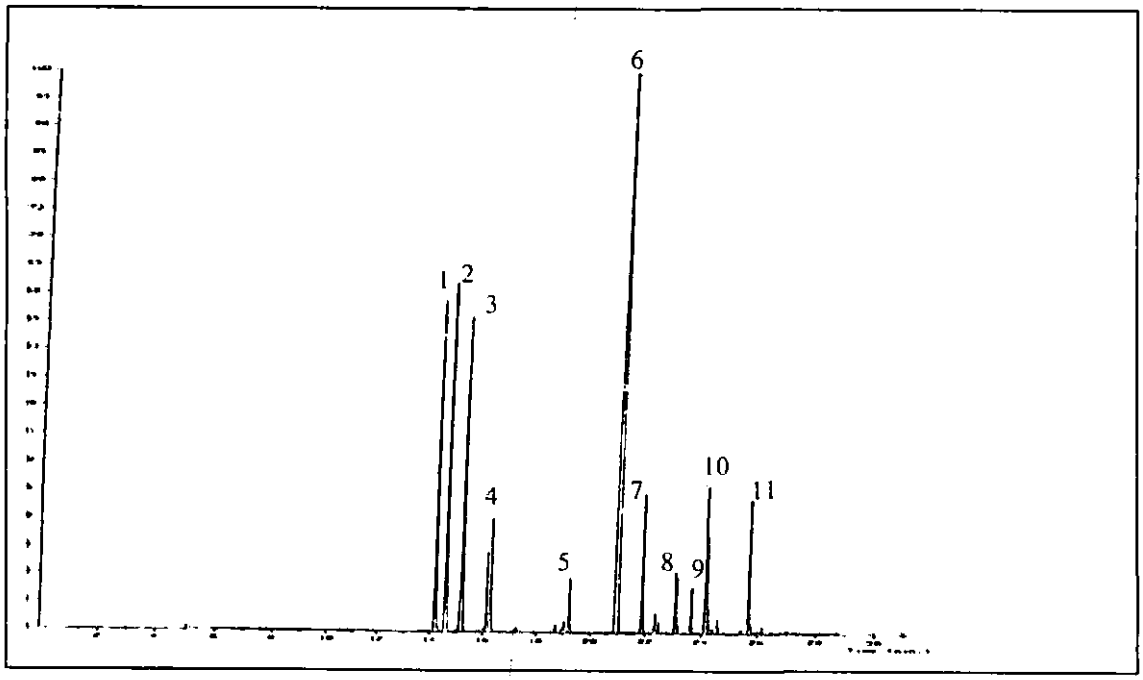


Fig - 21. Gas liquid chromatogram of common betel root oil[8]

Table 5. Description of the gas liquid chromatogram of common betel root oil

Peak No.	Compound	Relative area %
1	α-Pinene	12.86
2	Camphene	13.48
3	β-Phellandrene	12.72
4	1,8-Cineole	3.19
5	α-Terpineol	1.51
6	Safrole	31.81
7	Eugenol	4.50
8	α-Elemene	1.80
9	β-Caryophyllene	1.57
10	Chavibitol acetate	4.74
11	Allyl pyrocatechol diacetate	4.25

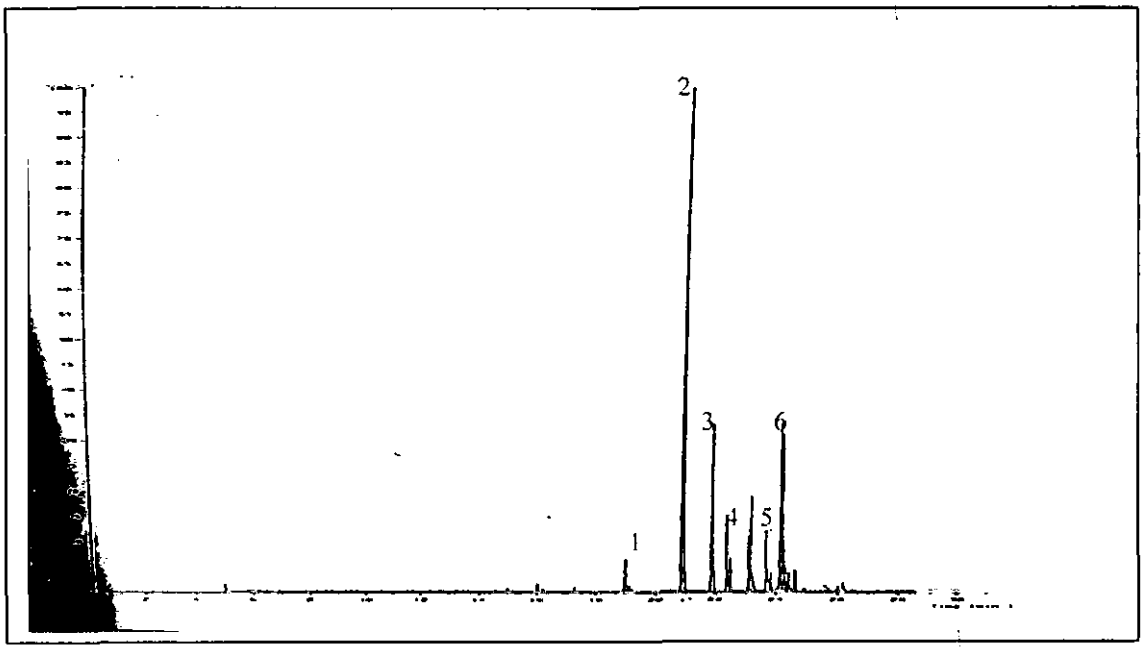


Fig - 22. Gas liquid chromatogram of common betel stalk oil[8]

Table 6. Description of the gas liquid chromatogram of common betel stalk oil

Peak No.	Compound	Relative area %
1	4-Terpineol	1.85
2	Safrole	40.47
3	Eugenol	11.48
4	γ-Muurolene	4.76
5	α-Humulene	4.48
6	Chavibitol acetate	11.63

Medicinal Uses

Uses described in pharmacopoeia and other traditional systems of medicine

Betel leaf is pungent and acrid[2,3,20]. It improves taste and appetite[2,20], lessens thirst, clears the throat and is vulnerary and styptic[2,21,22]. It act as a tonic to brain, heart and liver[2,21]. Betel leaves are considered as being stimulatory[1,6,7,21,23], sialagogue[1,2,3], carminative[1,3,7,21,23], stomachic[3,20,21] and anthelmintic[2,3,20]. It is also considered as a tonic[2,3,20,21], laxative[2,3,20,21], astringent[1,3,4,7,23], expectorant, febrifuge[3,20], antiseptic[4,7] and aphrodisiac[1,2,3,4,7,20]. It is useful in ozoena[2,21], bronchitis[2,3,20,21], asthma[3,7,20], catarrh, cough, leprosy, dyspepsia, syncope, fever, halitosis, diarrhoea[3,20], conjunctivitis, skin diseases[3] and elephantiasis of the leg[2,21]. It is known to produce a primary stimulation of the central nervous system[24]. Betel leaves are a constituent of a polyherbal oil to treat toxicity effects caused from snake, rat and dog bites[25].

The juice of betel is stomachic and is a febrifuge[1]. It is used as an administrating agent of traditional medicine; that is the pills are rubbed into an emulsion with the juice of betel leaves and licked up[5,25,23]. The juice is also used in decoctions for severe fevers after childbirth[26]. It is given to children for cough and administered into the eye for night blindness in adults[1,3]. It is also used to relieve cerebral congestion and satyriasis[2].

The juice of betel flower is used in a paste to be applied for eye diseases[27].

Roots of *P. betle* are used in a polyherbal oil to treat chest pains, muscle pains and pains in joints[25] and also used in polyherbal preparation to treat vomiting and stomachic diseases[26].

The tender stalk of leaf dipped in oil is introduced into the rectum of children suffering from simple constipation[5,23].

Uses in folk medicine

Betel leaf is a folk remedy for adenopathy, asthma, bronchitis, bruises, cancer, catarrh, colic, congestion, cough, diphtheria, edema, gastroenteritis, inflammations and wounds[22]. The bruised leaves are applied as an antiseptic on cuts and wounds and as a poultice on boils[1]. For foul ulcers, leaves are used as a dressing which seems to improve under them. Leaves are applied to the temples for relieving pain during headache and to painful and swollen glands for promoting absorption[23]. Warmed betel leaves with oil form a valuable application to the chest in case of bronchitis, during breathing difficulties, cough and asthma. The same application has been recommended in congestion and other affections of liver[24]. Betel leaves are warmed by fire and placed over the breasts to promote the secretion of milk[5]. Warmed betel leaves or warmed juice of leaves is also applied on abscesses and on axilla, groin and breasts for inflammations[28].

Leaves pounded with small quantities of sulphur, camphor, pepper and garlic are roasted in sesame oil and applied to the skin to treat eczema and scabies[29].

For dyspepsia, indigestion and stomachaches a few slices of ginger, *Piper nigrum* seeds and salt are chewed with a betel leaf and swallowed [28].

The forehead is covered with a piece of cotton cloth wet with juice of betel leaves for headache[28]. The juice of leaves is dropped into the ear to relieve earache and into the eye for painful eye-infections[24]. Fresh juice of leaves is used to relieve throat pain and used in digestive complaints with juice of *Zingiber officinale*[30].

Dried petioles are used as an aphrodisiac[31,32].

A preparation of betel roots with black pepper is given to women to bring about sterility[33] and plant juice with black pepper is used as a contraceptive in India[34]. Also *P. betle* tubers, tuber of a wild variety of Cucumis and another unspecified plant are mixed together with little water to make a paste and used for birth control[35]. The root is said to be used to prevent child bearing in Orissa[2].

Betel also has an effect of breath sweetening and removes all foulness from the mouth[19,23].

In Konkan, the fruit is employed with honey as a remedy for cough[2].

In Philippine Islands the leaves are used to cure diseases in children such as indigestion, colic, diarrhoea, pulmonary catarrh, and laryngitis[2]. Heated betel leaves are applied to the abdomen to relieve constipation [36]. The leaves are given for gastric and lung disorders[1]. Fresh leaves are used as a purgative for new borns; the leaf is rolled up, covered with oil and used as a suppository[37].

In East Africa the juice of leaves is given for catarrh and diphtheria[1].

In Cambodia the pounded leaves are used for the preparation of lotions and baths for patients suffering from protracted fever, smallpox, enlarged glands and lymphangitis[1].

In Borneo the decoction from betel leaves is used to cure spleen fever[38].

In Brunei betel is used to treat stomachache and to induce childbirth. Leaf juice is used for fatigue, diarrhoea and asthma[39].

In Malaysia juice of betel leaves are used to treat sore eyes[40].

In Fiji fresh juice of betel is used for ulcers, coughs, colds and sore throat[41].

Other Uses

Chewing of betel leaves with various adjuncts (betel quid) is an ancient practice in many South Asian countries including Sri Lanka[19,22].
In Sri Lanka betel leaves are used in many traditional occasions.
In Thailand betel leaves are eaten as a food[42].

Ayurvedic/Traditional Medicinal Preparations

Betel is used as a constituent of the following Ayurvedic preparations[3,25].
Kumburukohombadi quata, Neelaraja taila, Vata taila, Shwasakasa chintamani rasa, Nagavalli rasa, Pushpadhanva rasa, Kalkuta rasa.

Activity Studies

Antioxidant activity

P. betle leaves possess antioxidant activity[43, 44, 45,46] and it is shown that activity is due to the presence of phenolic compounds[46]. Aqueous extract of *P. betle* inflorescence posses antioxidative effects and the aqueous components of the extract are potential reactive oxygen species scavengers[47]. The effect of betel leaf extract on control of autooxidation of fat in dry fish has been studied and the sample prepared by dipping the fish in the extract immediately after salting and then drying as usual, had better keeping qualities[48]. The daily administration of an aqueous suspension of *P. betle* for 30 days provides good antioxidant potential and protection against tissue lipid peroxidation in streptozotocin induced diabetes rats[49].

Antimicrobial activity

The ethyl acetate and ethanol extracts have shown significant antibacterial activity against *Vibrio cholerae ogawa, Stapylococcus aureus, Diplococcus pneumoniae*, and *Klebsiella aerogenes*[50]. The aqueous extract, ether extract and pet-ether extracts have shown activity against *Aerobactor aerogenes, Escherichia coli, Klebsiella pneumoniae, Proteus vulgaris, Salmonella enteritidis, Serratia marcescens* and *Shigella flexneri*[51]. It has been found that essential oil and extract of leaves are effective against several Gram positive and Gram negative bacteria such as *Escherichia coli, Streptococcus pyogens, Pseudomonas aerugenosa, Staphylococcus aureus*[8], *Micrococcus pyogenes, Bacillus subtilis*, and *B. megaterium*, *B.* pumilus, *Diplococcus pneumoniae, Salmonella typhosa, Vibrio comma, V. cholera, Shigella dysenteriae, Proteus valgaris, Staphylococcus aureus, Pseudomonas solanacaerum, Sarcina lutea, Erwinia carotovora, Klebsiella species, Enterobactor species* and *Mycobaterium tuberculosis*[19,52,53]. The ether fraction of betel leaves showed significant antibacterial activity against *Fusobacterium nucleatum, Porphyromonas gingivalis* and *Peptostreptococcus anaerobius*, which are the organisms known to be responsible for halitosis (production of volatile sulphur compounds) in human oral cavity[54].

The essential oil and the leaf extracts possess anti-fungal activity against *Aspergillus niger* and *A. oryzea, A. candidus, A. flavus, Arthroderma benhamiae, Curvularia lunata* and *Fusarium oxysporum*[19]. The effect of *Piper betle* on the growth and aflatoxin

production by *Aspergillus parasiticus* was studied. Results have shown that betel leaf powder exhibit higher antimycotic activity than fruit[55]. Chloroform and ethanol extracts also possess antimytotic activity against *Aspergillus parasiticus*[55]. The essential oil of betel was shown to possess antifungal activity against *Corynospora cassicola, Colletotricum* sp., and *Rigidoporous* sp. which cause diseases to rubber plant[52]. Aqueous extract of *P. betle* has shown activity against number of Aspergillus species such as *Aspergillus auricomus, A. fischeri, A. flavus, A. fumigatus, A. nudulans, A. niger, A. sydowi, A. terreus, A. tericola, A. ustus,and A. versicolar*[56].

Anthelmintic activity

Anthelmintic activity was observed against *Ascaridia galli* in essential oil of *P. betle*[57]. Essential oil also possess antiascaric activity against *Ascaris lumbricoides*[58] and anticestodal activity agains *Taenia solium*[57]. The essential oil of betel leaves showed dose-dependent larvicidal effects on *Chrysomea megacephala* larva which is known to cause myiasis in man and animals [59]. Antinematodal activity was observed against *Bunostomum trigonocephalum* in essential oil and the plant alcoholic extracts have antinematodal activity against the pine wood nematode, *Bursaphelenchs xylophilus*[60].

Insecticidal activity

Betel essential oil possesses ant-repelling activity[52].

Betel leaf oil in ethanol has promising mosquitocidal activity against several mosquito species, *Aedes egypti*, *Anopheles tessellatus* and *Culex quinquefasciatus*. It was found that this oil solution is also effective against larvae of these mosquito species[52].

Leaf oil of betel showed knockdown and mortality effects against house fly (*Muca domestica*)[52].

Leaf oil has shown insecticidal effects on Rice weevil (*Sitophilus oryzae*) and Ticks (*Ixodes pacificus*), the external parasites[52].

Neuroprotective effects

Aqueous extract of betel has shown neuroprotective effect against alcoholic toxicity in the brain of rats[61].

Antifertility activity

Aqueous extract of leaves reversibly inhibits the fertility of male rats[62] and inhibits the masculine sexual behavior of male rats[63]. It has antimortality effect on washed human spermatazoa[42,64]. The alcoholic crude extract of leaf stalks of betel had shown pregnancy interceptive effect on female rats[65]. Chronic administration of extract of stalks of betel suggested the antifertility activity in both sexes of Albino rats[66]. The ethanol extract of betel showed spermicidal effects[31].

Antiinflammatory activity

Triterpenes and β-sitosterol isolated from *P. betle* have shown antiinflammatory effects[18].

Antitumor activity

Studies have indicated that extracts from *P. betle* leaves posses antitumor promoting activity[67]. The aqueous extract of leaves of *P. betle* has inhibited the initiation phase of 7,12-diethylbenz[a]anthracene (DMBA) induced mammary carcinogenesis in rats. But no appreciable degree of inhibition of tumor growth was noticed when the extract was fed to rats bearing DMBA-induced mammary tumor after 8 weeks[68]. Ether extract of dried leaves has shown carcinogenisis inhibition activity in mammary tumors of female rats[69].

Gastrointestinal function and ulcers

The influence of leaves of two betel varieties (the pungent Myrose and non-pungent Ambadi) has been examined. The results indicated that they have significant stimulatory influence on pancreatic lipase activity and decreasing influence on pancreatic trypsin and chymotrypsin activities. The Ambadi variety has positive stimulatory influence on intestinal digestive enzymes lipase, amylase and disaccharidases whereas a slight lowering in the activity of these enzymes when Myrose variety was administered[70]. *P. betle* extracts showed hepatoprotective effect in ethanol treated rats. Ethanol extract of betel leaves showed healing effect on peptic ulcers induced by NAID in albino rats[71]. Hot water extract and cold ethanol extracts exhibit gastroprotective properties as evidenced by its significant inhibition in the formation of gastric lesions induced by absolute ethanol[43].

Antinociceptive activity

Hot water extract and cold ethanol extracts of *P. betle* exhibit antinociceptive activity[43].

Wound healing properties

The effectiveness of *P. betle* ointment on wound healing in rabbits has been studied with reference to the wound contraction, epithelisation as observed visually and granulation tissue formation. The ointment has significantly healed the wounds and wound contraction and epithelisation were the dominant factors[72]. An ointment prepared using betel extract has shown wound healing activity[52].

Hypotensive and cardiovascular effects

The acute administration of betel inflorescence extract may activate C-fiber-evoked parasympathetic and sympathetic cardiovascular reflexes in rats[73]. Myocardial depressant activity was observed in essential oil of betel[74]. Methanol extract of betel leaves showed vasorelaxing effects[75]. The aqueous, ether and acetone extracts were shown to possess hypotensive activity. The results indicated that angiotensin converting enzyme inhibition may be an alternative mechanism leading to the hypotensive activity of betel[76]. Essential oil of betel has showed hypotensive activity[74].

Antidiabetes activity

Antidiabetes activity of *P. betle* was studied and it is shown that hot water and cold ethanol extracts have significantly reduced the blood glucose levels in rats. These two extracts showed comparable results. Moreover hot water extract failed to inhibit the glucose absorption by the intestine[43,77].

Antimutagenic activity

Water and acetone extracts of betel leaves have suppressed the mutagenicity of betel quid mutagenes in dose dependent manner. Furthermore acetone extract is more potent than water extract in inhibiting mutagenicity of environment mutagenes. It was suggested that hydroxychavicol and eugenol isolated from betel leaves have suppressed the formation of mutagenic N-nitrosomethylurea[78]. In contrast some studies have demonstrated the formation of mutagenic compounds in aqueous extracts of *P. betle* fruit and leaves when these extracts were nitrosated[79].

Carcinogenisis inhibition

P. betle leaves possess carcinogenesis inhibition effects[80,81]. Ether extract[82] and ethanol extract[83] have shown carcinogenisis inhibition activity in mouse. Methanol extract has shown antitumor promoting activity[42].

Plant germination and root growth inhibition effects

Betel ethanol extract showed germination inhibition effects on bajiri seeds[52]. Ethanol extract has inhibited the growth of Allium roots[84].

Vitamin activity

Vitamin A level in liver and plasma of mice treated with n-nitrosonornicotine and 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone was increased by the administration of the ethanol extract[83].

Radioprotective activity

The radioprotective activity of *P. betle* ethanol extract has been studied. The extract effectively prevented gamma-ray induced lipid peroxidation as assessed by measuring thiobarbituric acid reactive substrates, lipid hydroperoxide and conjugated diene. Likewise, it prevented radiation-induced DNA strand breaks in a concentration dependent manner[84].

Effect of betel quid

Chewing of betel quid mainly affects the central and autonomic nervous systems[86]. It has been shown that excessive indulgence in chewing betel leaves with tobacco and hydrated lime for long periods is liable to produce dental caries, oral sepsis, dyspepsia, palpitation and neurosis. Sometimes it leads to carcinomatous growth in the mouth but it is considered that the use of tobacco with betel leaves may be responsible for it[19]. Some studies had identified an independent effect of betel quid without tobacco in the causation of oral cancer[87]. Safrole was isolated from betel quid ingredients, specially from betel inflorescence. It has been found that safrole forms stable safrole-DNA adduct in human oral tissue following betel quid chewing, which may contribute to oral carcinogenesis[88]. Some experiments indicated that betel quid contains not only genotoxic and cytotoxic agents but also compounds which stimulates cell proliferation. These compounds may act

synergistically in the pathogenesis of oral submucous fibrosis and oral cancer in betel quid chewers[89]. Some studies have shown that betel quid ingredients may stimulate the arachidonic acid and collagen induced platelet aggregation, indicating its potential regulatory effects on local and systematic circulation system in a dose dependent manner[90].

Safty Evaluation

The water and the ethanol extracts of *P. betle* were well tolerated in terms of percentage weight gain, food and water intake, morbidity, aversive behavour, mortality, blood haematology, serum analysis and organ weights except the weight of the spleen. Both extracts significantly increased the weight of the spleen which suggested lymphoproliferative activity[91].

Studies on pre-clinical toxicity were undertaken to determine the haematological and biochemical changes after administration of alcoholic extract of leaf stalk of *P. betle* in rats. The results have indicated that the extract was devoid of toxicity[107].

Weak cytotoxic activity was observed in aqueous extract of betel[89].

Patent Information

- Herbal based composition for treating acute and chronic myeloid leukemia[92].
- Herbal composition for treating CD33+ acute and chronic myeloid leukemia and a method thereof[93].
- A herbal composition for the treatment and remedy for the bronchial respiratory difficulties[94].
- Betel leaf extract consisting of antileishmanial activity[95].
- The use of betel leaf extract to induced IFN-gamma production from human peripheral blood T cells and as a Th 1 type immunomodulator[96].
- Analgesic and refreshing herbal composition and a process for preparing the same[97].
- *P. betle* leaf extract consisting of antibacterial activity[98].
- Method of instantaneous removal of warts and moles using slaked lime and stem of betel leaf[99].
- Incapacitating composition and a device for its use[100].

Products Developed from *Piper betle*

- Instant herbal tea[102].
- Asthmacare (A preparation to treat asthma)[103].
- A novel polymeric gel for Periodentis[104].
- Health drink[105].
- Betel leaf oil[106].
- Analgesic herbal liquid[97].
- A herbal composition to be taken internally to treat bronchial respiratory difficulties[94].

Products developed at Industrial Technology Institute[52]

- Betel toothpaste.
- Betel mouth wash.
- Betel face cream.
- Betel pellets.
- Betel lotion for wound myiasis.
- Wound healing ointment.
- Instant betel quid.

Propagation[19,101]

Number of *P. betle* varieties can be found in Sri Lanka showing variation in size, shape and the colour of the leaves. Maha maneru, Rata dalu, Kaha bulat, Kaha maneru, Kalu bulat, Gal dalu, Gata todu, Kaha keeriya, Val bulat, Garandi maneru, Nagawalli and Mati pala are the main varieties and some of them are suitable for export and local consumption.

The betel vine grows best under tropical climate having considerable humidity and good supply of soil moisture. It can be successfully grown at elevations up to 1000 m from sea level. Betel can be grown successfully in all types of soil found in Sri Lanka and the laterite soil in Kurunegala and Gampaha districts is more favourable. Although the plant prefers bright sunlight, the intermediate and wet zones are better for betel cultivations. A

flat land with proper drainage and sunlight is suitable for betel cultivation. The weeds are first removed from the land and burned. Then the beds are prepared to the size of 4 x 25 ft. The space between two beds should be about 5 ft. The soil on beds is mixed thoroughly and is kept open to sunlight for about a week. Before the planting the beds should be sterilized. The most economical method is burning of few layers of hay and rice husk on moistened beds. To maintain a proper drainage a channel should be dug around the area that beds are prepared.

Betel vine is propagated only vegetatively by cuttings taken from healthy mature vines. The vine is trimmed into sections containing 3 nodes, the above two containing leaves. The trimmed sections are washed with a fungicide and they are planted in such a manner that two nodes are buried in the soil, but the lowest leaf should be at the same level of the land. Betel vine needs a support for its growth. It is provided by growing some sticks from plants like *Sesbania grandiflora* and *Moringa oleifera.* These sticks are first placed and cuttings are then placed near the sticks, two cuttings for one stick. After planting, the beds should be covered with coconut leaves. The crop needs a daily water supply and fertilizer should be supplied as required.

The harvesting can start after the vine has reached up to 4 ft. Once it has grown up to 6 ft. harvesting can be done every two weeks. The best harvest can be obtained in two years after planting the crop and the cultivation can be maintained up to 6 years. It is best to recultivate the crop after 6 years.

Diseases and Pests [19,101]

Betel vine is subjected to attack by various diseases and pests. Leaf spot, Foot rot, Wilt and Powdery mildew are commonly found diseases and bugs, aphids and mites are common among insect pests.

Leaf spot is caused by both bacteria (bacteria leafspot) and fungi (fungi leafspot). Bacteria leafspot is caused by *Gloeosporium* and *Santhomonas* bacterii. Brown or black spots with perceptible concentric rings appear on the leaves. The spots have an oily appearance and they may extend to the entire leaf and also to the stem which starts rotting

downwards. Treatment with bordeaux mixture or Perenox prevents the spread of the disease to the stems. Fungal leaf spot is caused by fungi, *Phytopthera* and *Choletroticum*. Brown or black spots appear on leaves, but they have dry appearance and sometimes have a yellow ring around the spot.

Wilt is caused by *Sclerotium* sps. In this, the stem turns dark, the leaves drop and finally the plant withers and dies. The disease can be controlled by deep ploughing and application of green manuring.

Powdery mildew is caused by *Oidium piperis*. The disease is characterized by the formation of circular grayish white powdery patches first on the lower surface of the leaves and then on both sides. This is followed by the yellowing of the surface and the leaves becoming brittle and finally falling off. The growing shoots are also affected and in severe cases the whole crop may be destroyed. Application of finely powdered sulfur once or twice act as a preventive measure in controlling the disease.

Foot rot is caused by fungal species like *Schlerotium, Rhizoctonia* and *Phytophthera*. These fungi live in soil and attack the root, stem and leaves. It produces a large number of spores at low temperatures, under moist conditions. These spores are carried from vine to vine through water. The first sign of the disease is the yellowing of the lowermost leaves followed by drooping. The roots also start rotting, followed by the drying up of the entire vine. This disease is not commonly found in Sri Lanka.

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