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Comparative Pharmacognostic and Qualitative Study of Some Herbal Drugs

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ABSTRACT

Annonasquamosa is commonly known as custard apple and Sitaphal, belonging to Annonaceae. This research deals with detailed Pharmacognostical, phytochemical, qualitative analysis of plant. The phytochemicals and the biopesticidal components present were ascertained. The results showed That saponins, steroids and terpenes were mostly present, while tannins and glycosides were moderately present, and alkaloids, flavonoids, phenols and oxalic acid were least present.Psidiumguajava (L.) belongs to the Myrtaceae family and it is an important fruit in tropical areas like India, Indonesia, Pakistan, Bangladesh, and South America. The leaves of the guava plant have been studied for their health benefits which are attributed to their plethora of phytochemicals, such as quercetin, avicularin, apigenin, guaijaverin, kaempferol, hyperin, myricetin, gallic acid, catechin, epicatechin, chlorogenic acid, epigallocatechingallate, and caffeic acid.

Caesalpiniabonduc (L) Roxb.is a vital remedy for treating several ailments in Indian traditional systems of medicine. The pharmacognostic study of the anatomical section of the seeds as well as microscopic studies of the powder was conducted to determine their morphological and anatomical features. The qualitative and quantitative microscopy as well as physiochemical properties studied.

This current study is planning to evaluate the pharmacognostical and phytochemical properties of Moringaoleifera Lam leaves. Here the pharmacognostical parameter like Macroscopic, characters were evaluated.

Key Word: Annona squamosal,Psidiumguajava ,Caesalpiniabonduc, Moringaoleifera, **1.0 PLANT PROFILE**

1.1 Annona squamosa L.(Annonaceae)

Figure No.1Seeds of A. squamosa

Taxonomical Hierarchy:
Kingdom:Plantae
Division :Magnoliophyta
Class: Magnoliopsida
Order: Magnoliates
Family: Annonaceae
Genus: Annona

Species :A. Squamosa
Part used: Seed
Common Names:

- Sanskrit : Bahubijika
- Hindi : Sitaphal
- English : Custard apple
- Marathi : Sitaphal

Geographical Distribution: It is native of W. Indies, now cultivated throughout India.
Botanical Description: A tree is about 6 m high.
Leaves: 3.8-7.6 by 1.8-3.7 cm long, oblong-lanceolate or elliptic, obtuse, pellucido-punctate,

International Journal of Pharmaceutical Research and Applications
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glabrous above and pubescent beneath when young.

Flowers: Solitary, leaf-opposed, extra-axillary branchlets; pedicels 12-19mm. long, bracteate below the middle. Sepels are minute, triangular, and pubescent. Petals are pubescent on both surfaces.

Fruit: Globose, 5-10 cm in diameter usually with glaucous bloom on the surface when young, yellowish green when ripe, granulate or tuberculate, 5-6 gonous.

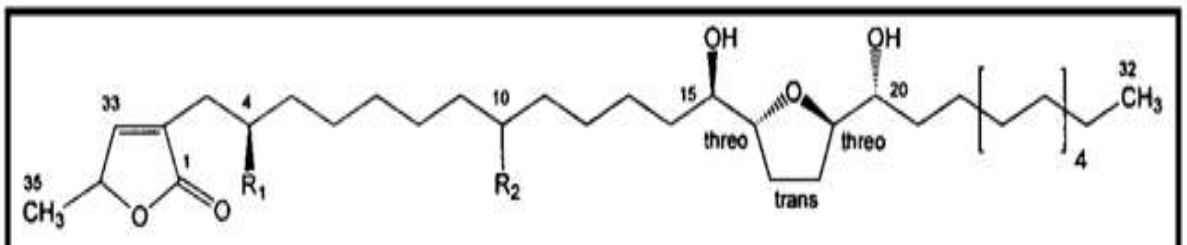
Seeds: Seeds are brownish black, smooth.

Chemical Constituents:

Numerous acetogenins were isolated from seeds of A. squamosa, they are mono- or adjacent bis-THF-rings bearing compounds. Acetogenins belonging to a series of C-35/ C-37, and derived from C-32/ C-34 long chain fatty acids.

A fraction of the MeOH extract contains two adjacent bis-tetrahydrofuranacetogenins named squamocin-O1 (1) and squamocin-O2 (2). Four new annonaceousacetogenins, dieposabadelin, squamocenin, lepirenin and dotistenin were isolated, alongwith sixteen known acetogenins: corepoxylone, diepomuricanins A and B, dieporeticenine, tripoxyrollin, bullatencin, glabrencin B, reticulatains-1, -2, uvariamicins I, II, III, erythrosolamin, annotemoyin-1 and -2 and solamin. The β-aminosquamocin was isolated from seeds ofA.squamosa.

It also contains solamin, corossolin, corossolone, murisolin, annonacin, annonacinone. Ketolactoneacetogenins Bullatacin, 4-Deoxybullatacin, Bullatacinones, 30-OH -bullatacinones, Asimicin, Trilobacin, Bullatalicin, Bullatalicinones, Annonacin, lsoannonacis were isolated. Annotemoyin-1, Annotemoyin-2, squamocin and cholesterylglucopyranoside were isolated from the seeds of A. squamosa.



No	NAME	R_1	R_2
1	SOLAMIN	−H	−H
2	COROSSOLIN	−H	−OH
3	COROSSOLONE	−H	=O
4	MURISOLIN	−OH	−H
5	ANNONACIN	−OH	−OH
6	ANNONACINONE	−OH	=O

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 2993-3008 www.ijprajournal.com ISSN: 2249-7781

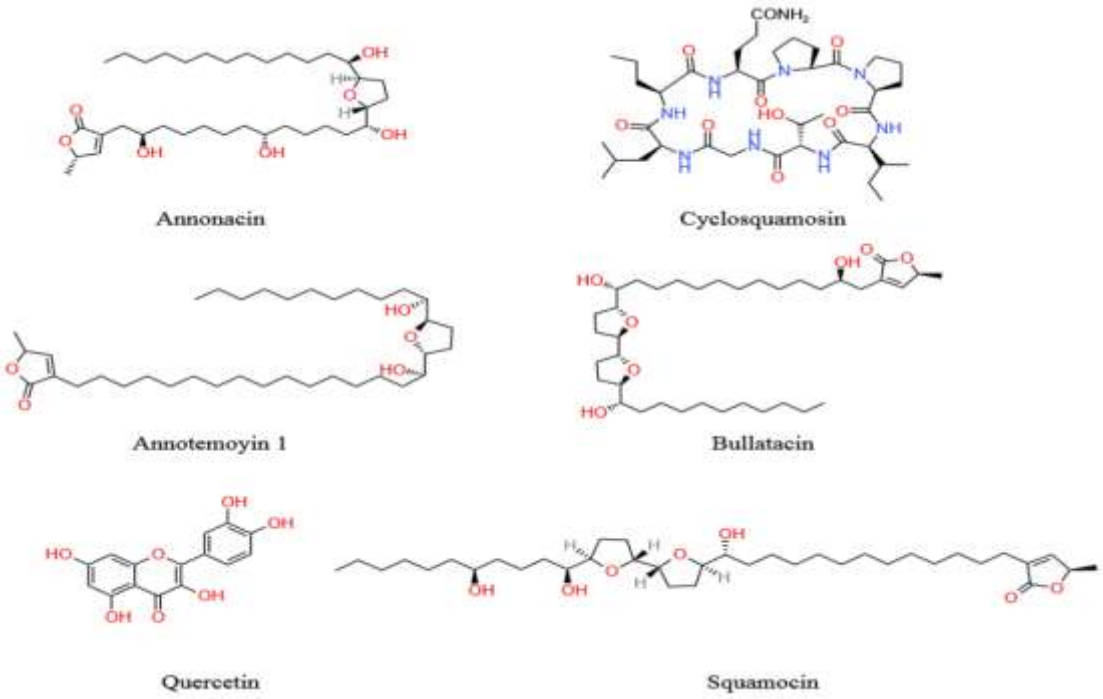


Figure No.2 Chemical constituents from seeds of A. squamosa

Uses:

Ripe fruit bruised and mixed with salt is applied to malignant tumors to hasten suppuration. Powder of seeds mixed with gram is a good hair wash. It is given in diarrhea, dysentery and dyspepsia**.** Seeds, fruits and leaves are effective as insecticide, fish poison, powerful irritant of the conjunctiva and abortifacient.

1.2 Azadirachta indica (Mealiaceae)

Figure No.3 Leaves of A.indica

Taxonomical Hierarchy:
Kingdom:Plantae
Division :Magnoliophyta
Order: Sapindales
Family: Meliaceae
Genus:Azadirachta
Species: A. indica
Part used: Leaves

Common Names:

- Synonym : MeliaAzadirachta
- Sanskrit : Arishta
- Hindi : Nim
- English : Persian/Indian lilac
- Marathi : Limba / Balantnimba

Geographical Distribution: It is distributed all over in India and also cultivated all over the world.

Botanical Description:
Leaves: Leaves are alternate, imparipinnate; leaflets are subopposite serrate, very unequal at base.
Flowers: Hermaphrodite, in axillary panicles. Calyx is 5- lobed. Petals 5 in number, much exceeding the calyx, free, imbricate. Staminal tube is a little shorter than the petals, cylindric, widening above 9-10 lobed at the apex, the lobes truncate, again slightly toothed; anthers within the tube opposite to and shorter than the lobes. Ovary is 3-celled; style is elongate, slender; stigma is shortly cylindric, 3-lobed; Ovules are 2 in each cell, collateral.
Fruits: 1-seeded drupe, endocarp is woody.
Seed: Ellipsoid; cordate at the base; radicle is superior.

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 2993-3008 www.ijprajournal.com ISSN: 2249-7781

Ayurvedic Description:
Rasa - tikta, kasaya
Guna - lagu
Veerya - sheeta
Vipak - katu

Chemical Constituents:

Number of chemicals isolated from leaves like limoniods and cyclic tri and tetrasulphides. It also contains Azadirachtin, Meliantriol and salanin[37]. The leaves contain nimbin, nimbinene, 6-desacetylnimbinene, nimbandiol, nimbolide, quercetin and β- sitosterol.

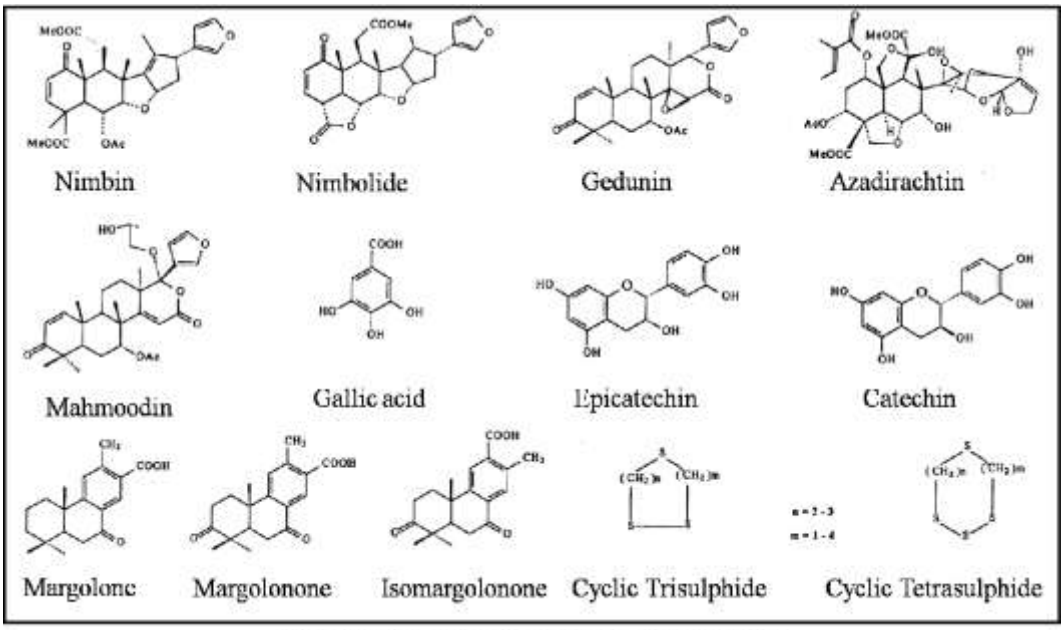


Fig. 1 Structure of bioactive neem compounds

Figure No.4 Chemical constituents from leaves of A. indica.

Uses:

Young leaves are astringent; used in leprosy, skin diseases, rheumatism, leucoderma, piles and reduces inflammation. Young branches are anthelmintic, good for cough, asthma, piles, tumors and urinary discharge. Unripe fruit used in tumors, piles and toothache.

1.3 Psidiumguajava

Linn (guava) is often reffered to as the apple of the tropics; a large genus of tropical and sub-tropical trees and shrubs, native of tropical America. Three species are recorded as cultivated in India, the most important of which is P. guajava (common guava).

Psidium guajava Linn (Myrtaceae)

Fig. 5 Psidium guajava Linn (Myrtaceae)

Scientific classification:
Kingdom:Planteae
Division :Magnoliophyta
Class: Magnoliopsida
Sub class : Rosidae
Order : Myrtales

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 2993-3008 www.ijprajournal.com ISSN: 2249-7781

Family : Myrtaceae
Subfamily : Myratoideae
Tribe : Myrteae
Genus : Psidium
Species : P. guajava L.

Common names:
Synonym : Common guava
Bengal : Goaachhi
Bombay : Jam, Perala, Peru
Gujarat : Jamrud, Jamrukh
Hindi : Amrud, Amrut
Malayalam: Koyya
Marathi: Jamba
Punjab: Anjirzard, Amrud
Sanskrit: Apritthaktvacha, Dridhabija
Tamil: Segappugoyya.

HABITAT:

It is a large evergreen or subdeciduous shrub, sometimes a small tree up to an altitude of 90 cm found throughout native of tropical America, West Indies. In India especially in Uttar Pradesh, Bihar, Maharashtra, Aasam, West Bengal, Andhra Pradesh and Madras**.**

BOTANICAL DESCRIPTION:

Psidiumguajava L. is a large evergrren tree or subdeciduos shrub, sometimes a small tree up to 90 cm girth and 7.5 m high. Stem is irregularly fluted when old. Bark is quite smooth, pale pinkish brown or buff with grey patches, exfoliating in very thin woody plates. Blaze is 2.5-5 mm, cheesy, not fibrous, whitish or pinkish brown usually tinged with chlorophyll outside. Leaves are 10-15 cm long, oblong or elliptic-oblong, entire, glabrous above, pubescent beneath, pellucid-punctate, lateral nerves 10-20 pairs, prominent beneath, strongly curved near the edge and joined by intramarginal veins. Petioles are 2.5-7.5 mm long. Flowers are white in color and 2.5-3.8 cm diameter. Peduncles are 1.3-3.8 cm long, axillary, 1-3 flowered. Calyx-tube adnate to the ovary and produced above it, the upper free portion entire, closed in bud at length bursting irregularly into lobes. Fruit is a globose or pyriform berry 5 cm long or more.

It is a low evergreen tree or shrub 6 to 25 feet high, with wide-spreading branches and square, downy twigs, is a native of tropical America. It is a common vegetation cover by roads and in waste places in Hawaii. Guava is a tropical and semitropical plant. It is well known in the islands for its edible fruit. It is common in the backyards. The branches are crooked, bringing opposite leaves. The flowers are white, incurved petals, 2 or 3 in the leaf axils; they are fragrant, with four to six petals and yellow anthers. The fruit is small, 3 to 6 cm long, pear-shaped, reddish-yellow when ripe.

Leaves:

Leaves are 10-15 cm long, opposite, not dotted, smooth, glossy green, chartaceous, oblong or elliptic-oblong, entire, glabrous above, pubescent beneath, pellucid-punctate, lateral nerves 10-20 pairs, prominent beneath, strongly curved near the edge and joined by intramarginal veins.

Fruit:

Fruits are globose or pyriform berry 5 cm long or more. It have sweet in taste, also green to light yellow, but in some varieties red in color varying in shape and size to a great extent. Sometimes two varieties are broadly distinguished, var. pyriferum and var. pomiferum based on the shape of the fruits, whether pear shaped or globose and ovoid. It is medium, roundish, surface smooth, gloosy, skin colour yellow, also apple like colour, dot small and distinct, apex flat, round, taste sweet, warty, ovate, pulp white, semi hard and pyriform.

Bark:

It is quite smooth, pale pinkish brown or buff with grey patches, exfoliating in very thin woody plates.

Flowers: Flowers are large in size and white in color.

CHEMICAL CONSTITUENTS:

Fruit:

Vitamin–C, Carbohydrate ester, Vitamin-A, Iron, Calcium, Phosphorous, Manganese, Oleanolic acid, Quercetin, Guaijavarin, morin-3-o-alpha-L-lyxopyranoside and morin-3-O-alpha-L-arabopyranoside. The ripe edible fruits three benzophenone glycosides, 2, 6- dihydroxy- 3, 5-dimethyl-4- O- beta- D- glucopyranosyl-benzophenone, 2, 6- dihydroxy- 3- methyl-4-O-(6"-O- galloyl- beta- D- glucopyranosyl)-benzophenone, 2, 6-dihydroxy- 3, 5- dimethyl- 4-O- (6"-O-galloyl- beta- D- glucopyranosyl)-benzophenone were isolated by means of chromatography. Immature fruits contains aldehydes such as (E)-2-hexenal and (Z)-3-hexenal. In mature fruits, esters like Z-3-hexenyl acetate and E-3-hexenyl acetate and sesquiterpenescaryophyllene, α-humulene and β-

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 2993-3008 www.ijprajournal.com ISSN: 2249-7781

bisabollene are present. New components were described for the first time as active aromatic constituents in pink guava fruit (3-penten-2-ol and 2-butenyl acetate).

Leaves:

It contains essential oil 90.56%. The major components were caryophyllene (18.81%), copaene (11.80%), pentacyclictriterpenoidspsidiumoic acid, beta-sitosterol, obtusol, oleanolic acid, and ursolic acid. The new constituents have been characterized as 2 alpha-glycolyl-3beta-hydroxyolean-12-en-28-oic acid, 3beta-p-E-coumaroyloxy-2alpha-methoxyurs-12-en-28-oic acid. Guajadial, a novel Caryophyllene-based Meroterpenoid, morin-3-O-alpha-L-lyxopyranoside and morin-3-O-alpha-L-arabopyranoside, and two known flavonoids guaijavarin and quercetin were identified. Leaves are isolated Quercetin containing compounds Quercetin 3-O-alpha-L-arabinoside (Guaijavarin); quercetin 3-O-beta-D-glucoside (isoquercetin); quercetin 3-O-beta-D-galactoside (hyperin); quercetin 3-O-beta-L-rhamnoside (quercitrin) and quercetin 3-O-gentobioside. Two triterpenoids, 20 beta-acetoxy-2 alpha, 3 beta-dihydroxyurs-12-en-28-oic acid (guavanoic acid), and 2 alpha, 3 beta-dihydroxy-24-p-z-coumaroyloxyurs-12-en-28-oic acid (guavacoumaric acid) along with six known compounds are 2 alpha-hydroxyursolic acid, jacoumaric acid, isoneriucoumaric acid, asiatic acid, ilelatifol D and beta-sitosterol-3-O-beta-D-glucopyranoside are isolated.

Bark:

Bark contains 12-30% of tannins alsoPolyphenols (Burkill, 1997), resin and crystals of calcium oxalate.

Root:

The roots are rich in tannins. It also contains leukocynidins, sterols and Gallic acid. There is high percentage of carbohydrates and salts. Root, stem bark and leaves contain a large percentage of tannic acid.

Seeds:

A seed contains 14% oil in dry weight, 15% protein, 13% starch, Quercetin-3-O-B-D-(2''-O- galloyglucoside)-4'-O-vinyl propionate. The seeds which are very small but abundant in the fruit and have been reported to contain 14% oil on dry weight, with 15% proteins and 13% starch (burkill, 1997). Ten Phenolic and flavonoid compounds including one new acylatedflavonol glycoside were isolated. The structures of the new compound quercetin-3-O-β-D-(2"-O-galloyglucoside)-4'-O-vinylpropionate and of the known compounds were elucidated.

Chemical structure:

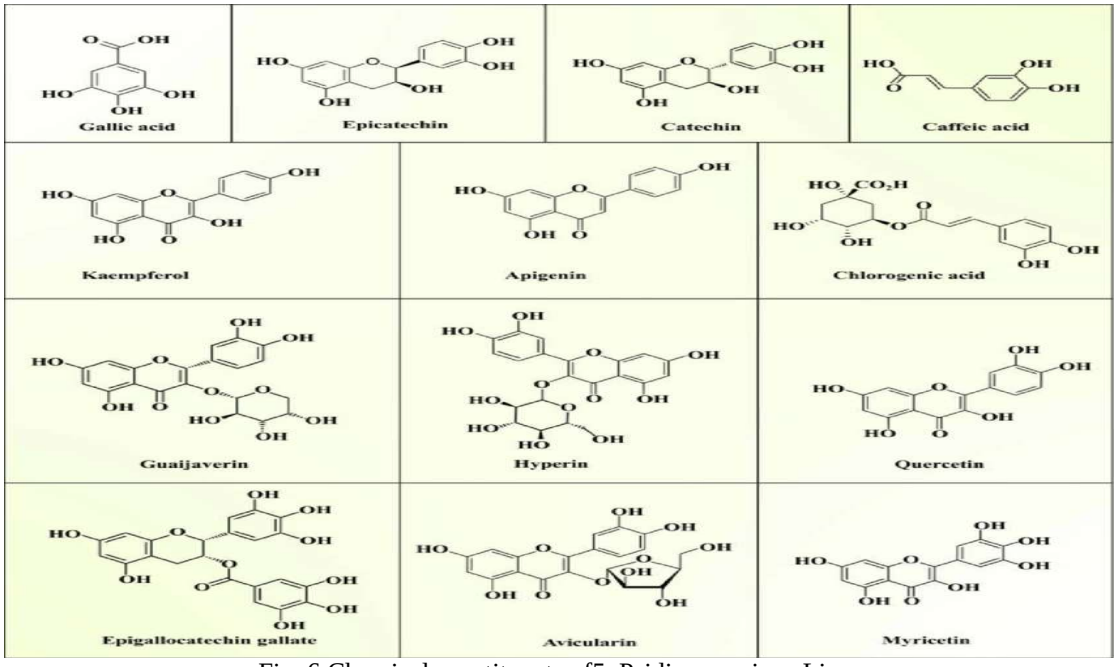


Fig. 6 Chemical constituents of5 Psidium guajava Linn

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 2993-3008 www.ijprajournal.com ISSN: 2249-7781

Uses of Psidiumguajava:

- **Fruit:**

It is tonic, cooling, laxative after food; used in the thirst; good in colic, and bleeding gums. It is also mucilaginous, astringent, and used in the treatment of diarrhea, dysentery and gout.

- **Leaves:**

It is used for the treatment of wounds and ulcer. It is astringent for bowls, Febrifuge, antispasmodic, cerebral affection, nephritis, tonic, cholera, cachexia, rheumatism, epilepsy, uterine hemorrhage, vomiting, and diarrhea and also used for toothache reliever.

- **Bark:**

It is having good Astringent and tonic properties and used in the treatment of diarrhea in the children and for dyeing purposes.

- **Flowers:**

These are used for the treatment of bronchitis, eye sores, and dry wounds (Conway & Peter, 2001).

- **Whole plant:**

P. guajavais mainly known for its antispasmodic and antimicrobial properties in the treatment of diarrhoea and dysentery, hypo-glycaemic agent, antioxidant, hepato-protective, anti-allergy, antimicrobial, antigenotoxic, antiplasmodial, cytotoxic, antispasmodic, cardioactive, anticough, antidiabetic, anti-inflammatory and antinociceptive activities.

1.4 C.bonducella

Fig. 7 C.bonducella

Taxonomical hierarchy
Kingdom:-Plantae
Order:-Febales
Family:-Caesalpiniacaeae
Genus:-Caesalpinia
Species:-C. bonducella

Part used: seeds
Synonyms:

- Kakachika, Karanja and Latakaranja in Sanskrit.
- Kathkaranj in Hindi.
- Fever nut in English.
- Nata in Bengali.
- Gajaga in Marathi.
- Bois canic in French.

Geographical Distribution:

An armed liana, up to 15 m in height, found up to an altitude of 1,000 m in Himalaya and wild throughout the plains of India and; it is also found in deltaic region of western, eastern and southern India 1. Found particularly along the seacoast throughout the hotter parts of India, Burma and Sri Lanka.

Botanical Description
Leaves:

Leaves are with large, leafy, branched, basal Appendages; 30-60 cm. long; petioles prickly; stipules a pair of reduced pinnae at the base of the leaf each furnished with a long mucronate point; pinnae 6-8 pairs, 5-7.5 cm. long, with a pair of hook stipulary spines at the base. Main leaf axis armed with stout, sharp, re-curved spines, divided into 4-8 pairs of secondary branches.

Leaflet:

Leaflets 6-9 pairs, 2-3.8 by 1.3-2.2 cm., membranous, elliptic-oblong, obtuse, strongly mucronate, glabrous above, more or less puberulous beneath; petioloules very short; stipels of short hooked spines.

Flowers:

Flowers in dense (usually) long-peduncled terminal and supraaxillary racemes dense at the top, lax downward, 15-25 cm. long; pedicels very short in bud, elongating to 5 mm. in flower and 8 mm. in fruits, brown downy; bracts squarrose, linear, acute, reaching 1 cm. long, fulvous hair. Calyx are 6-8 mm long, fulvous hairy; lobes

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 2993-3008 www.ijprajournal.com ISSN: 2249-7781

obovate-oblong, obtuse. Petals are oblanceolate, yellow.

Seed coat is hard, glossy, and greenish to ash grey in colour. And is traversed by circular and vertical faint markings of the cracks, forming uniform rectangular to squarishrectulations all over the surface seeds, oblong, lead-colored, 1.3 centimeter long. A raised hilum with remains of the stalk lies in the centre of the dark spot, at the narrow edge of the seed. Adjacent to the hilum, lies a faint coloured circular to oval elevated micropyle. In dry seed, kernel gets detached from the testa. Testa is about 1-1.25 mm in thickness and is composed of three distinct layers, the outermost - thin and brittle, the middle one - broad, fibrous and dark – brown and the innermost – white and papery.

The seed is exalbuminous. The kernel surface is furrowed and ridged, hard, pale yellowish – white, circular to oval, flattened and about 1.23- 1.75 centimeter in diameter. A scar of the micropyle lies at one end of the kernel, from where arises a prominent ridge demarking the two cotyledons of the embryo. Plumule – radical axis is thick, cylindrical and straight. Taste is very bitter and odour is nauseating and unpleasant.

Seeds:

Seeds show a palisade layers which are composed of vertical, columnar, and laterally closed appressed cells. Thickenings are present on the walls of palisade cells which in tangential section appear as 6-10 denticulate projections into the lumen of cells. Then after that there is the layer of bearer cells and a thick zone of parenchymatous cells. The majority of bearer cells are T-shaped, thick walled and non-lignified.

ETHNOMEDICAL CLAIM:

Seed:- Used in the treatment of intermittent fever, asthma, colic . Also used as antiperiodic, in dyspepsia,dentrifice, filariasis.

Roots: Used in the treatment of fever, asthma, colic etc.

Seed kernel:- Used in the treatment of orchitis, ovaritis, scrofula, useful for dispersing swellings, restraining haemorrhage in hydrocele leprosy and keeping off infectious diseases.

Leaves: Used in the treatment of disorders of liver and oil is useful in convulsions, palsy and nervous complaints. Leaves with castor oil or ghee is useful in painful and swollen testicles, deobstrurent and emmonogogue.

Chemical constituent

The seed kernel of plant caesalpiniabonducella mainly contains bonducin, natin, and sulphurcompounds.Theseed also contains fatty acids. The root mainly shows the presence of diosgeninas chemical constituent. The leaf also contains proteins (Anon, 1992).The fruit contains saponinsviz, saponin C and saponin D. The shoot of plant contains 36,47-dihydroxyhenpentacosan-4-one; Triacontanol; 27-cyclohexylheptacosan-7-ol; 16-hydroxy-26-methylheptacosan-2-one; 4-methylheptatriacont-1-en-10-ol; tetracontanol-2; betasitosterol.

1.5Moringo oleifera L. (Moringaceae)

Figure No.8 Leaves of M. Oleifera

Taxonomical Hierarchy:
Kingdom:Plantae
Division :Magnoliophyta
Order: Brassicales
Family :Moringaceae
Genus:Moringa
Species :M. oleifera

Part used: Leaves
Common Names:

- Synonym : M. pterygosperma C.F. Gaertn
- Sanskrit : Shigru
- Hindi : Segva
- English : Horseradish tree, drumstick tree
- Marathi : Shevaga

Geographical Distribution: A tree growing wild in the Sub-Himalayas from Chinab to U.P. It is commonly cultivated in India & Burma.

Botanical Description:
It is a small or middle sized tree. Bark is corky; wood is soft; root is pungent; young parts are tomentose.

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 2993-3008 www.ijprajournal.com ISSN: 2249-7781

Leaves: usually 3- pinnate, sometimes 45 cm long; rhachis is slender, thickened and articulated at the base; pinnae and pinnules opposite, deciduous; leaflets 12-20 by 6-10 mm, elliptic, terminally obovate and slightly larger than lateral ones; petiolules of the lateral leaflets are 1.5-2.5 mm long.
Flowers: white, in large puberulous panicles. Petals are spathulate, veined. Stamens are 5 alternating with 5-7 antherless ones; Overy is oblong, style is cylindric.
Seeds: 3 angled, angles are winged.

Ayurvedic Description:
Rasa - katu, khara
Guna - lagu, rooksha
Veerya - ushna
Vipak- katu

Leaves of M. oleifera contain amino acid, Vitamin A, 3'-O- Methy l- quecetin, Gossypetin, Quercetagetin and Proanthocynidins. The crude leaf extract contains 4-(α-L-rhamnosyloxy) phenylacetonitrile, Niazinin A and B, Niazimicin A and B, Niaziminin A and B, Niazicin A and B, Niazimin A and B, Niazicinin A, Niazirinin and Niazidin.

Ethanolic leaf extract of M. oleiferacontainshexadecanoic acid, ethyl ester (CAS) ethyl palmitate, palmitic acid ethyl ester, 2,6- Dimethyl-1, 7-octadiene-3-ol, 4-Hexadecen-6-yne, (z)-(CAS), 2-hexanone, 3- cyclohexyliden-4-ethyl – E2- Dodecenylacetate, Hi-oleic safflower oil (CAS), Safflower oil[51]. Fractionated leaf extract contains4-(α-L-rhamnosyloxy) benzyl isothiocyanate**(2)**, niazimicin**(3)**, niazirin**(4)**, β-sitosterol**(5)**,glycerol-1-(9-octadecanoate)**(6)**, 3-O-(6'-O-oleoyl-β-D-glucopyranosyl)-β-sitosterol**(7)**, and β-sitosterol-3-O-β-D-glucopyranoside.

Chemical Constituents:

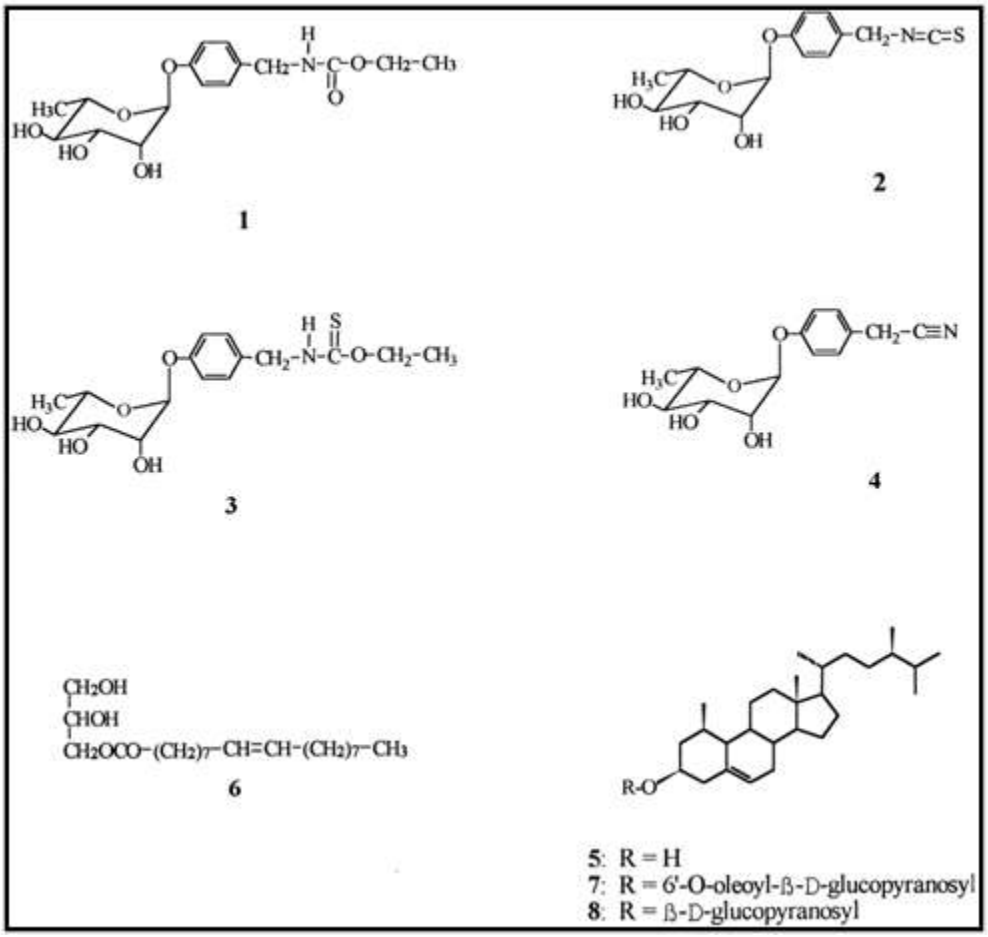


Figure No.9 Chemical Constituents from leaves of M. oleifera

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 2993-3008 www.ijprajournal.com ISSN: 2249-7781

Uses:

The leaves aretasty, remove all kinds of pain; anthelmintic; useful in eye diseases, dry tumors, asthma. Leaves pounded and warmed, are applied to tumors as a resolvent. Fruit and flowers are used in tumors, cures vata and kapha. The ground roots are mixed with salt and applied as a poultice to tumors.

2.MATERIALS AND METHODS

2.1 Plant Material:

Collection and Procurement:

All parts of plant were collected fromShevgaonTahasil ofAhmednagar district (M.S.). The parts of plant were dried under shade away from direct sunlight. The dried parts were cleaned and coarsely powdered in grinder and powder material was passed through 120 mesh to remove fine powders and coarse powder was used for extraction.

2.2 Pharmacognostic Studies:

Macroscopy:

Organoleptic characters, extra feature and macroscopical details of all parts of plantwere carried out.

2.3 Evaluation of Physical Constants:

Determination of foreign organic matter:

Five gm of air dried coarsely powdered drug was spread in a thin layer. The sample was inspected with the unaided eye or with the use of 6X lens. The foreign organic matter was separated manually as completely as possible. Sample was weighed and percentage of foreign organic matter was determined from the weight of the drug taken.

Determination of Moisture content:

Accurately weighed glass Stoppard shallow weighing bottle, and was dried. 2 gm of sample was transferred to the bottle and covered, the weight was taken and sample was distributed evenly and poured to a depth not exceeding 10 mm. Then loaded bottle was kept in oven and stopper was removed. The sample was dried to constant weight. After drying it was collected to room temperature in a desiccator. Weighed and calculated loss on drying in terms of percent w/w.

Ash Value:

Ash value is used to determine quality and purity of crude drug. Ash value contains inorganic radicals like phosphates, carbonates and silicates of sodium, potassium, magnesium and calcium etc. sometimes inorganic variables like calcium oxalate, silica, carbonate content of the crude drug affects 'total ash value'. Such variables are then removed by treating with acid and then acid insoluble ash value is determined.

Determination of Total Ash:

Accurately weighed 2 gm of the air-dried crude drug was taken in a tarred silica dish and incinerated at a temperature not exceeding 450^0C until free from carbon, cooled in a desiccator and weight was taken.The process was repeated till constant weight was obtained. The percentage of ash was calculated with reference to air-dried drug.

Water Soluble Ash:

The ash, obtained as per the method described above boiled for 5 minutes with 25 ml of water, filtered, and collected the insoluble matter in a Gooch crucible, washed with hot water and ignited for 15 minutes at a temperature not exceeding 450^0C and weight was taken. The percentage of water-soluble ash was calculated with reference to air-dried drug.

Acid Insoluble Ash:

The ash obtained as per method described above and boiled with 25 ml of
2M hydrochloric acid for 5 minutes, filtered, and collected the insoluble matter in a Gooch crucible or on an ash less filter paper, washed with hot water, ignited, and cooled in a desiccator and weighed. The percentage of acid-insoluble ash was calculated with reference to the air-dried drug.

Extractive Values:

Different extractive values like alcohol soluble extractive, water soluble extractive values were performed by standard method.

Determination of Water-Soluble extractive value:

Five gm of air dried coarsely powdered drug was macerated with 100 ml of chloroform water in a closed flask for 24 hours and it was shaken frequently during first 6 hours and allowed to stand for 18 hours. Then it was filtered, 25 ml of the filtrate was evaporated in a flat shallow dish and dried at 105^0C and weighed. Percentage of water-soluble extractive value was calculated with reference to air-dried drugs.

Determination of Alcohol-soluble extractive value:

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 2993-3008 www.ijprajournal.com ISSN: 2249-7781

Five gm of air-dried coarsely powdered drug was macerated with 100 ml of ethanol of specified strength in a closed flask for 24 hours and it was shaken frequently during first 6 hours and allowed to stand for 18 hours. Then it was filtered, during filtration precaution was taken against loss of ethanol, 25 ml of the filtrate was evaporated in a flat shallow dish and dried at 105^0C and weighed. Percentage of ethanol soluble extractive value was calculated with reference to air-dried drugs.

2.4 Preliminary Phytochemical Test:

Preliminary phytochemical evaluation of extracts:

1) Test for Carbohydrates:

i) Molish's test

2-3 ml of extract, add few drops of α-naphthol solution in alcohol. Shake and add concentrated sulphuric acid from sides of the test tube. Violet ring is formed at the junction of two liquids which shows presence of carbohydrates.

a) Test for reducing sugar:

i) Fehling's test

Five ml of extract solution was mixed with 5 ml Fehling's solution (equal mixture of Fehling's solution A and B) and boiled. Development of brick red precipitate indicates the presence of reducing sugars.

ii) Benedict's test

Mix equal volume of Benedict's regent and extract solution in test tube. Heat in a boiling water bath for 5 min. Solution appears green, yellow or red depending on amount of reducing sugar present.

b) Test for monosaccharides:

i) Barfoed's test

Mix equal volume of Barfoed's reagent and extract solution. Heat for 1-2 minute in boiling water bath and cool. Development of red precipitate indicates presence of monosaccharides.

2) Test for Proteins:

i) Biuret test

The extract was treated with 1 ml of 10 percent sodium hydroxide solution and heated. A drop of 0.7 percent copper sulphate solution was added to the above mixture. The formation of purple violet color indicates the presence of proteins.

ii) Millon's test

The extract was treated with 2 ml of Millon's reagent. Formation of white precipitate indicates the presence of proteins and amino acids.

3) Test for Amino acids:

i) Ninhydrin test

The extract was treated with Ninhydrin reagent at pH range of 4-8 and boiled. Formation of purple color indicates the presence of amino acids.

4) Test for Steroids:

i) Liebermann-Burchard test

10 mg extract was dissolved in 1 ml of chloroform and 1 ml of acetic anhydride was added following the addition of 2 ml of concentrated sulphuric acid from the side of the test tube. Formation of reddish violet color at the junction indicates the presence of steroids.

ii) Liebermann's test

To 2 ml of the extract a few ml of acetic anhydride was added and gentle heated. The content of the test tube were cooled and 2 ml of concentrated sulphuric acid was added from the side of the test tube. Development of blue color gave the evidence for presence of steroids.

iii) Salkowski test

One ml of concentrated sulphuric acid was added to 10 mg of extract dissolved in 1 ml of chloroform. A reddish brown color exhibited by chloroform layer and green fluorescence by the acid layer suggests the presence of steroids.

5) Test for Glycosides:

i) Anthraquinone glycosides:

a) Borntrager's test

To 3 ml extract add dilute sulphuric acid, boil and filter. To the cold filtrate, add equal volume benzene or chloroform shake well. Separate organic solvent. Add ammonia, the ammonical layer turns pink or red indicates the presence of anthroquinone glycoside.

ii) Cardiac glycoside:

a) Keller-killani test

To 2 ml of extract, glacial acetic acid, one drop 5 % Ferric chloride and concentrated sulphuric acid was added. Presence of cardiac glycosides is indicated by formation of reddish brown color at junction of the two liquid layers and upper layer appeared bluish green.

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 2993-3008 www.ijprajournal.com ISSN: 2249-7781

6) Test for Saponins:

i) Foam formation test

One ml solution of the extract was diluted with 20 ml distilled water and shaken in a graduated cylinder for 15 minutes. The development of stable foam indicates the presence of saponins.

7) Test for Flavonoids:

i) Shinoda test

To the extract 5 ml (95%) ethanol and few drops of con. HCl and 0.5 g of magnesium turnings was added gives pink color indicates presence of flavonoids.

ii) Lead acetate test

Few drops of 10 percent lead acetate are added to the extract. Development of yellow colored precipitate confirms the presence of flavonoids.

8) Test for Alkaloids:

i) Dragendroff's test

0.1 ml dilute hydrochloric acid and 0.1 ml Dragendroff's reagent was added in 2 ml of extracts in test tube. Formation of orange brown precipitate indicates the presence of alkaloids.

ii) Mayer's test

Two ml of extract was taken in a test tube. 0.2 ml of dilute hydrochloric acid and 0.1 ml of Mayer's reagent were added. Formation of yellowish buff precipitate indicates the presence of alkaloids.

iii) Hager's test

Two ml of extract was allowed to react with 0.2 ml dilute hydrochloric acid and 0.1 ml of Hager's reagent. Formation of yellowish precipitate indicates the presence of alkaloids.

iv) Wagner's test

Two ml of extract was treated with 0.2 ml dilute hydrochloric acid and 0.1 ml of Wagner's reagent. Formation of reddish brown precipitate indicates the presence of alkaloids.

9) Test for Tannins and Phenolic compounds:

i) 5 % Ferric chloride test

Five ml of extract solution was allowed to react with 1 ml of 5 percent ferric chloride solution. Greenish black coloration indicates the presence of tannins.

ii) Potassium Dichromate test

2-3 ml of extract solution, mix with 2 ml of Potassium dichromate. The formation of red precipitate indicates presence of tannins.

iii) Bromine Water test

Two ml of extract solution mix with 2 ml of bromine water. Discoloration of bromine water indicates presence of tannins.

iv) Dilute Nitric acid test

Two ml of extract solution was allowed to react with few drops of dilute nitric acid solution. Formation of reddish to yellow color indicates the presence of tannins.

10) Test for Vitamin-C:

1 ml of extract solution with 2 ml of water and add 1 drop of freshly prepared 5 % w/v solution of sodium nitroprusside and 1 ml of dilute sodium hydroxide solution. Add 0.6 ml of HCl, stir it. The yellow color turns to blue indicate presence of vitamin- C.

3. RESULTS

Pharmacognostic Studies:

As per WHO guideline pharmacognostic study of all plant parts like macroscopy, physical parameters, and extractive values were studied.

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 2993-3008 www.ijprajournal.com ISSN: 2249-7781

Macroscopy

Table No. 1: Morphological and Organoleptic Characters

Sr. No.	Parameter	Part of Plant				
		A. squamosa	**A. indica**	**P. guajava**	**C. bonducella**	**M. oleifera**
1.	Color	Brownish black	Green	Greenish yellow	Green	Green
2.	Odor	-	Characteristic	Characteristic	Characteristic	-
3.	Taste	-	-	Sweet	Bitter	-
4.	Size	-	leaflets 9-15in.long,	Fibrous	2-4 cm long,1-2 cm diameter	30-60cm, 12-18mm long
5.	Shape	-	Alternate, imparipinnate,	-	circular	leaflets elliptic-ovate

Ash Value:

TABLE NO. 2: ASH VALUES OFDIFFERENT PARTS OF PLANT

Sr. No.	Parameter	Part of Plant				
		A. squamosa	**A. indica**	**P. guajava**	**C. bonducella**	**M. oleifera**
1.	Total ash	2.0 %	7.9 %	4.5 %	4.9 %	5.5 %
2.	Water- soluble ash	1.7 %	1.9 %	1.5 %	1.3 %	1.6%
3.	Acid insoluble ash	0.8 %	0.7 %	0.5 %	1.2%	2.5%

Extractive Values:

Table No. 3: Extractive values of Different parts of plant

Sr. No.	Parameter	Part of Plant				
		A. squamosa	**A. indica**	**P. guajava**	**C. bonducella**	**M. oleifera**
1.	Alcohol soluble extractive value	10.8 %	10.5 %	10.3 %	4.45 %	14.2 %
2.	Water soluble extractive value	17 %	23.9 %	21.2 %	10.5 %	15 %

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 2993-3008 www.ijprajournal.com ISSN: 2249-7781

Foreign Organic Matter and Moisture Content

Table No. 4: FOM and Moisture content of Different parts of plant

Sr. No.	Parameter	Part of Plant				
		A. squamosa	A. indica	P. guajava	C. bonducella	M. oleifera
1.	Foreign organic matter	00%	0.9%	00 %	00 %	0.4%
2.	Moisture content	1.0%	0.8 %	12.5 %	5.9 %	1.20 %

Preliminary Screening of Extracts

Table No. 5:Summery of Extraction

Extracts	Color	Nature	Percentage Yield (% W/W)
A. squamosa extract	Whitish Brown	Dry powder	5.9 %
A. indica extract	Dark Green	Sticky powder	14.09 %
P. guajava	Dark Green	Semisolid	19 %
C. bonducella	Dark Green	Semisolid	18 %
M. oleifera extract	Green	Sticky powder	23.2%

Phytochemical Evaluation

Preliminary phytochemical tests

Table No. 6: Preliminary phytochemical tests ofdifferent parts of plant

Sr. No.	Test Perform	A. squamosa	A. indica	P. guajava	C. bonducella	M. oleifera
1.	**Test for carbohydrate**					
	Molish's test	+	-	+	-	-
	Fehling test	-	-	-	-	-
	Benedicts test	+	+		-	+
	Barfoed's test	-	-		-	-
2.	**Test for Proteins**					
	Biuret Test	-	-	+	-	-
	Millions Test	-	-	-	-	+
3.	**Test for amino acids**					
	Ninhydrine test	-	-	-	-	+
4.	**Test for Steroids**					
	Salkowski test	-	+	+	+	+

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 3 May-June 2023, pp: 2993-3008 www.ijprajournal.com ISSN: 2249-7781

	Libermann test	-	-	+	+	-
	LibermannBurchard reaction	-	+	+	+	+
5.	**Test for Glycosides**					
	Cardiac	-	-	+	-	+
	Anthraquinone	-	+	+	-	+
6.	**Test for Saponin**					
	Foam test	-	+	_	-	+
7.	**Test for Flavonoids**					
	Shinoda test	+	+	+	-	+
	Lead acetate test	-	+	+	-	+
8.	**Test for Alkaloids**					
	Dragondroff's test	+	+	+	-	-
	Mayer's test	+	+	-	-	+
	Hager's test	-	+	-	+	-
	Wagner's test	+	+	+	-	+
9.	**Test for Tannins and phenolic compounds**					
	5% Ferric chloride test	+	+	+	-	+
	Potassium dichromate test	+	-	-	-	+
	Bromine water test	-	+	+	-	+
	Dil. Nitric acid test	-	-	-	-	-
10.	**Test for Vitamin**					
	Vitamin C	-	-	-	-	-

+ indicates presence of constituents.
- indicates absence of constituents.

4. CONCLUSION

From the obtained results, it can be concluded that:

- squamosaseed, A .indica leaves, P. guajava frits, C. bonducellafruit and M. oleifera leaves extracts were used to prepare formulation.
- The total ash value was greater. Water soluble ash was found higher than acid insoluble ash value.
- The results of extractive value show significant results.
- In preliminary phytochemical tests of the various extracts showed presence of phytosterols, alkaloids, glycosides, flavonoids, saponins, tannins and phenolic compounds.

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