



STUDIES ON THE PHYTOCHEMICAL AND PHARMACOLOGICAL PROPERTIES OF *ANNONA SQUAMOSA* L.

Chandraiah. G

Head & Assistant Prof of Botany

Govt. Degree College for Women Jagtial -505327

Alternative medicine and natural remedies have been used since ancient times for the treatment and well-being of humans. Medicinal plants are considered to be effective and most important for the above purposes. Mother Nature has provided us with a huge count of flora and fauna. Some of the natural medicinal plants are so common that we use them in daily life without knowing their medicinal importance. *Annona squamosa* L. is the best example of it. The fruit of this plant is commonly known as the custard apple which is eatable *A. squamosa*. and human beings were also providing updated facts of phytochemical and pharmacological approaches of the above plant in the present 21st century. *A. squamosa* L. is a multipurpose shrub tree that is distributed throughout tropical countries and eminently a desert fruit in India. The plant belongs to the family Annonaceae which in Hindi is known as Sitaphal or Sharifa which is utilized for its medicinal properties. The ethnobotanical traditional uses include wound healing, lice repellent, and treatment of dysentery and urinary tract infections. Phytochemicals include alkaloids (benzoxyquinazoline, salsolinol, coclaurine), terpenoids (annomosin A, *annosquamosa* in A), glycoside (quercetin-3-glucoside), essential oil (bicyclogernacrene, T-cadinol), flavonoids (kaempferol, farmarixetin) tannins, and much more responsible for pharmacological action of plant parts are fruits, seeds, leaves, bark, and flower. Information on medicinal uses and organoleptic properties of various pharmacologically active parts is also provided. The leaves are used as a vermicide, for treating cancerous tumors and are applied to abscesses, insect bites, and other skin complaints. Scrapings of root bark are used for toothache. Powdered seeds are used to kill head lice and fleas but care should be taken that the powder does not come in contact with the eyes as this causes great pain. The present article discusses the updated information regarding distribution, plant parts used, chemical constituents, and morphological and pharmacological importance of this plant hoping for exploring better medicinal value.

Keywords: *Annona squamosa*, Custard Apple, Phytochemistry alkaloids, phytochemical screening, ethanol, extract, antidiabetic.

Introduction:

The genus name, 'Annona' is from the Latin word 'anon', meaning 'yearly produce', referring to the production of fruits of the various species in this genus. Annonaceae, the custard apple family (NRCS2008) is a family of flowering plants consisting of trees, shrubs, or rarely lianas (GRIN 1997). With about 2300 to 2500 species and more than 130 genera, its type genus is *Annona*. The family is concentrated in the tropics, with few species found in temperate regions. About 900 species are Neo-tropical, 450 are Afro tropical, and the other species Indo Malayan. Under the Annonaceae family 130 genera (Raj Sobiya *et al.*, 2009) are available, and out of that genera are widely available (Jonathan *et.al.*, 1994). *Annona*, *Anonidium*, *Rolliania*, *Uvaria*, *Melodorum*, *Asimina*, *Stelechocarpus*

As the *Annona* genus is widely available in India this genus has been selected for the study. The bark, leaves, and roots of some species are used in folk medicines. The strong bark is used for forests ((Raj Sobiya *et al.*, 2009) and for wooden implements, such as tool handles and pegs. The wood is valued as firewood (Jonathan *et.al.*, 1994), with Yellow and brown dyes. A recent study suggests that alcoholic seed extract contains anti-cancer compounds (Cochrane *et.al.*, 2008). Leaf extract has an antinociceptive effect (Gonzalez-Trujano *et. al.*, 2009).

Roots were found to have an anticonvulsant effect (Eva and Gonzalez-Trujano 2006). In Mexico, juice is used for chills and fever [9]. Pulp was found to have mutagenic properties (Ecology and evolutionary biology plant growth facilities. 5th Edn CBS Publishers.2007). It is mainly used as an ornamental plant and it is cultivated along with banana plantations. It is an orange skin fruit native to Brazil, and it is rarely available (Oliveira L *et.al.*, 2010).

It is found to have *In vitro*, *In vivo* studies exhibiting anti-tumor activity. Fruit and fruit juice are taken for worms and parasites, to cool fevers, to increase the mother's milk after childbirth, and as an astringent for diarrhoea and dysentery. The crushed seeds are used against internal and external parasites, head lice, and worms. The bark leaves and roots are considered sedative, and ulcer treatment for various disorders towards those effects. Roots containing acerogenins proved to have an anti-carcinogenic effect by inhibiting DNA synthesis.

Leaves are used to treat hysteria and fainting spells. Leaf decoction is used in the treatment of colds, coughs, intestinal infections, and acidity conditions. bark decoction is used in diarrhoea. Roots are used in dysentery. Fruit is used in the making of ice creams and milk beverages (Nehey pandya 2011). Crushed leaves are used in internal and external wounds and boils, leaf decoction is used in gastritis

Leaf, the juice is used as a vermifuge. Unripe dried fruits are used in diarrhoea and dysentery treatment. Root, the bark is used in toothache. Seeds leaves and young fruits have got insecticidal activity. Seeds are used in folk for their insecticidal activity, and parasitic activity (International center for research in agroforestry. 2008)]. Roots are reported to have apomorphine alkaloids: Reomerine, Antonine, and Dehydro reomerine produce a skeletal muscle relaxant effect. The yellow resin extracted from seeds exhibits sympathetic action such as dilatation of the pupil, dryness of mouth, and decreases secretions. From the folklore usages three plants of the Annonaceae family, *Annona* genus- *Annona squamosa* Linn, were selected. Leaf parts of these plants were taken for the study.

TAXONOMICAL CHARACTERISATION AND DESCRIPTION:

Various indigenous and traditional sources of medicinal plants have been extensively used for treatments. Various parts of plants such as the leaves, fruits, bark, roots, and even seeds are being used for the preparation of medicines. *Annona squamosa* Linn is also been extensively used as traditional medicine in various cultures.

Taxonomical characterization of *Annona squamosa* Linn.

Kingdom: Plantae

Order: Magnoliales

Family: Annonaceae

Genus: Annona

Species: Squamosa

Vernacular names: "Ata"

English: Custard apple

Annona squamosa is a small well-branched tree or shrub that bears edible fruits called sugar-apples, species of the genus *Annona* and a member of the family Annonaceae more willing to grow at lower altitudes than its relatives *Annona reticulata* and *Annona cherimola*. A custard apple tree does not and will do well if watered regularly, along with enough light for it to grow. It grows well in hot dry climates and adjusts in any kind of soil, a job that is a little difficult for other plants in its family. If you have sowed the plant's seeds, they will bear fruits in 2 to 3 years. The fruits are generally conical or round in shape and will take around 3 to 4 months to ripen.

Materials and Methods: -

Plant Material

The leaves and fruits of the *Annona squamosa* plant were collected from College Ground Govt Degree College for Women Jagityal District Telangana state located in the eastern area in September 2022. The plants were identified by Dr. V. Narasinha Murthy Botany Department of Botany Satavahana University Karimnagar. The voucher specimen was deposited in the Research Laboratory Department of Botany Govt Degree College for Women Jagityal. The plant organs were cut into small pieces and shade dried at room temperature (20°C) for two weeks, finely powdered plant materials were stored in airtight polythene bags protected from sunlight until use.

Chemical Composition

It has been determined that over 100 chemicals from various categories exist:

- Alkaloids: Benzoxyquinazoline, samoquasine A, N-nitrosoxylopine, roemerolidine, duguevalline, meleagrine, chrysogine, dopamine, salsolinol, coclaurine, liriodenine, oxoanalobine.⁸⁻¹¹
- Flavanoids: Rutin, isoquercitrin, bullatacin, quercetin, kaempherol, farmarixetin, isorhamnetin^{8, 12}
- Entkaurediterpenoid: Annomosin A Annosquamosin A, annosquamoannosquamosin D, annosquamosin E, annosquamosin F, annoquamosin.
- Glycoside: quercetin-3-glucoside¹³

- Essential oil: germacrene, β -elemene, α and β pinene, sabinene, bicyclogernacrene, T-cadinol, T-muciololspathulenol, bornylacetate, borneol, verbenone¹⁴.
- Other compounds include Glucopyranoside, coumarinoligans annotemoyin-1, annonacin, annonacin A, and annonastatin, as well as Squamocin B to Squamocin N, annotemoyin-2, Squamocin, and cholesteryl, etc. Fig. 1 shows the Phytochemical Screening of Leaf Extract various constituents of *Annona squamosa* plants.

Organoleptic Characters

Annona Squamosa L. has irregularly spreading branches with a height of 3-7m Lanceolate to oblong-lanceolate, pale green on both sides, and almost globose leaves are found singly, measuring 6–17 × 3-6 cm. They belong to the category of a semi-deciduous tree having light brown bark with smoothies fissured in plates.

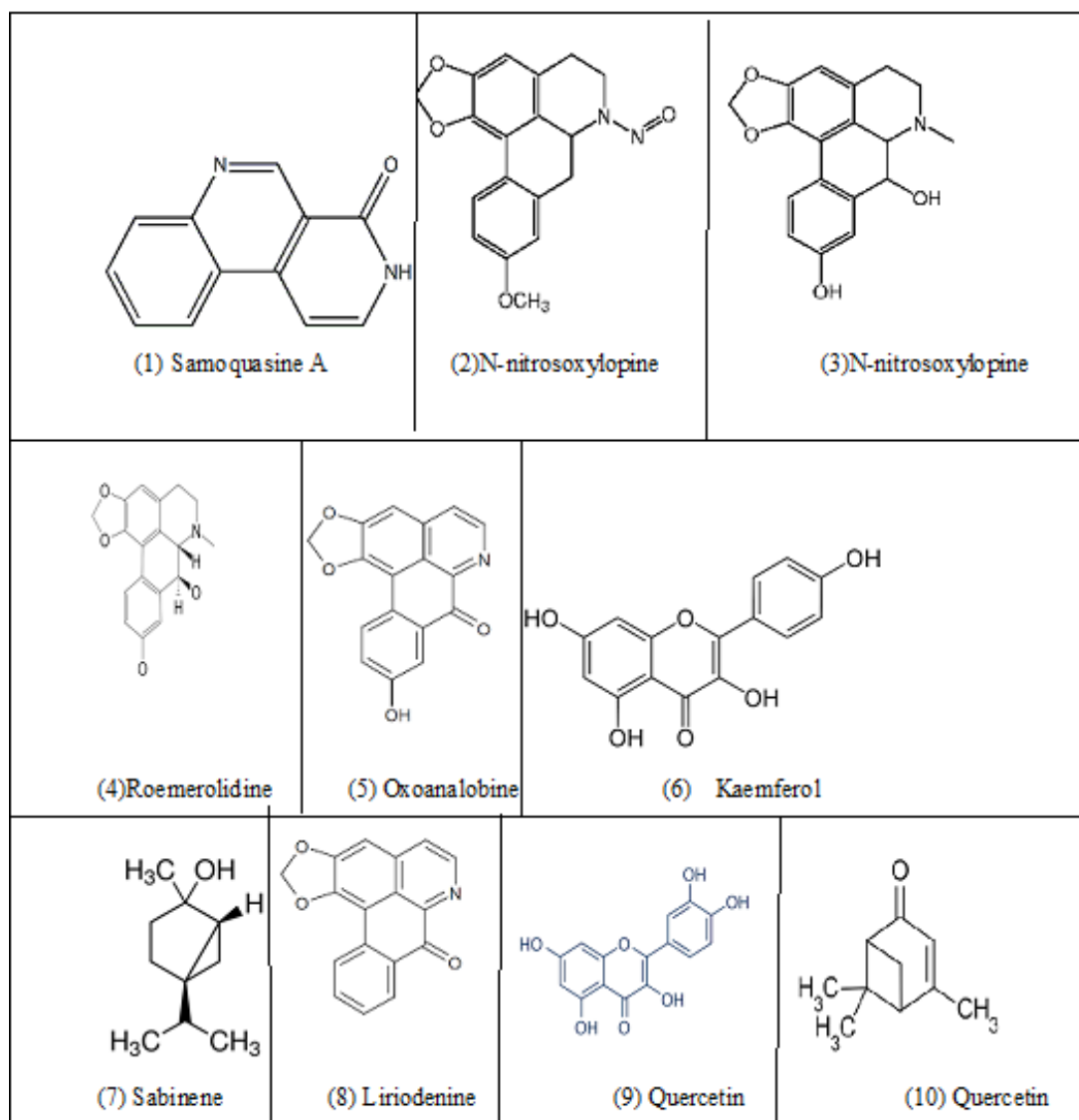


Fig.1 Showing various constituents of *Annona squamosa* L. plants

Organoleptic Characters

Annona Squamosa L. has irregularly spreading branches with a height of 3-7m Lanceolate to oblong-lanceolate, pale green on both sides, and almost globose leaves are found singly, measuring 6–17 × 3-6 cm. They belong to the category of a semi-deciduous tree having light brown bark with smoothies fissured in plates.

Table 1. Organoleptic characters various parts of the plant *A. squamosa*

Characters	Leaves	Fruits	Seeds	Stem	Roots
Taste	Bitter	Sweet	No Taste	Slight bitter	Bitter
Odor	Characteristic	Characteristic	No Odour	No odor	No odor
Color	Green	Pulp White inside and green outside	Black	Brown	Brown

Flowers are greenish yellow with fragrance on slender hairy stalks produced singly or in short clusters in 2.5cm length where sepals in 16mm length are pointed, hairy, and green in color. The three outer petals are yellow-green, slightly hairy, thick, and oblong at the tips whereas inner petals are ovate and pointed scale. Flowers are greenish-yellow with fragrance, produced singly or in short lateral clusters of 2-4 flowers, about 2.5 cm long, and 2.5 cm wide; sepals are pointed, hairy, green, about 16 mm long; the three outer petals are oblong, thick and rounded at the tips, fleshy, 1.6-2.5 cm long, and 0.6 cm wide; they are yellow-green, slightly hairy, and the interior is light yellow with a purplish or red Scales on the three-minute inner petals are ovoid. Table 1.1 showing the morphological features of the seeds, leaves, stem, roots, and fruits of *A. squamosa*.

Fruits are green-yellow in color, round, ovate, irregular, and heart-shaped having sizes 5-10 cm in diameter. When ripen, the pulp becomes edible, white, and sweet. Seeds are oblong, shiny, black, 1.3-1.6cm long numerous in each carpel. Ovaries are light green, style is white and crowded and stamen are numerous, white, and crowded in 16mm long. (Fig-2, a, b, c, and d) show respective fruits, flowers, seeds, and whole plants.

Extraction, Fractionation, and Isolation:

Shade-dried *A. squamosa* was soaked in methanol for 5 days. The extract was concentrated using an electric water bath at 500⁰ C The solvent extract was concentrated under reduce pressure at 400⁰C using rotavapor, the extracts obtained were suspended in water and successively partitioned with n-hexane, chloroform, ethyl acetate, and methanolic fraction. The chloroform fraction was subjected to column chromatography which led to the isolation of two known compounds β- Sitosterol (1) and stigmasterol (2).

The structure of these compounds was elucidated by comparing their physical and spectra data with already reported data in the literature.



Fig-2 Morphological characters of *A. squamosa* L. a) Flowers b) Fruits on the plant c) Individual Fruits d) Opened Ripen Fruit with seeds

Two methods of extraction were used in this study; the first was Precipitation by acetone and the second was extraction by ether.

1. Extraction of *A. squamosa* Seeds by Acetone:
2. This method of extraction was carried out as follows: -Five grams of seeds were ground in a seed mill.
3. The powder was extracted with 100 ml of PBS (prepared by dissolving 8.2 g Na_2HPO_4 and 8.82 g NaCl in 1000 ml distilled water and kept pH at 7.2 by stirring overnight at 4°C. Then was filtrated and made up to 100 ml with phosphate buffer.
4. The obtained extract was centrifuged at 30 g for 1 hour.
5. The supernatant was incubated at 70°C for 15 minutes and then cooled to 4°C, before centrifugation at 10000 g for 15 minutes.
6. The supernatant was cooled in an ice bath to 0°C, followed by a slow addition of 23 ml of acetone at 0°C, and left for 10 minutes, before centrifugation at 100g for 10 minutes.
7. After that, 161 ml of acetone was added to the supernatant and then centrifuged.
8. The precipitate of the last centrifugation was dissolved in 75 ml buffer saline.

9. The solution was dialyzed overnight against a 5-liter phosphate buffer at 4°C. Afterward, the sample for the biochemical test was taken while the remaining was poured in a Petri dish and dried at 37°C.
10. The dried extract was scrubbed off the Petri dishes, placed in labeled, tightly sealed plastic tubes, and stored in deep freeze at -2°C.
11. In order to destruct the seeds, fifteen grams of seeds were ground 15-20 times with diethyl ether in a blender for 15 minutes each time.
12. The mixture was filtered by Whatman No.1 filter-paper.
13. The resulting precipitate (powder) was extracted by stirring overnight at 4°C with 10 volumes of cold PBS (prepared by dissolving 0.82 g Na₂HPO₄ and 11.76 g NaCl in 1000 ml distilled water and keeping pH at 7.2).
14. The solution was centrifuged in a cooled centrifuge at 400 rpm for 30 min.
15. The volume of supernatant was estimated and fractionated with (NH₄)₂SO₄ ammonium sulfate at 0-40% [the determination of ammonium sulfate weight occurred according to the volume of supernatant and depending on the saturation table] at 4°C.
16. The solution was centrifuged in a cold centrifuge at 4000 rpm for 30 minutes.
17. The volume of supernatant was estimated and fractionated with ammonium sulfate at 40-60% at 4°C.
18. After centrifugation by cooled centrifuge, the supernatant was fractionated with ammonium sulfate at 60-100%.
19. The obtained precipitate was re-dissolved in ordinary phosphate buffer and dialyzed against five liters of ordinary PBS at 4°C. Then sample for the biochemical test was taken, while the remains were poured off in the Petri dishes for drying at 4°C.
20. The dried extract was scrubbed off the Petri dishes, placed in labeled, tightly sealed plastic tube, and stored in deep freeze at -20°C. Solutions of Biochemical Test Reagents Test for Lectin One drop of the extract was added to one drop of human blood on the slide. Agglutination should appear if lectin was present.

Phytochemical Profiling:

The chemical tests were performed on the hexane, chloroform, ethyl acetate, and methanolic extracts of *A. squamosa* using the standard procedures to identify the bioactive secondary metabolite.

Test for alkaloids: -

Alkaloids salts: the aqueous extract of each organ of the plant (25 ml) was stirred with 15 ml of 10 % HCl in a steam bath for 30 minutes. The mixture was extracted then three times with diethyl ether. 1 ml of the aqueous layer was treated with two drops of Wagner's reagent. The formation of a brownish precipitate was regarded as evidence of the presence of salts and alkaloids in the extract. Or 0.2g of each fraction was warmed with 2% H₂SO₄ for 2 min. The reaction mixture was filtered and added a few drops of Dragendorff reagent were to each filtrate. Orange-red precipitate indicates the presence of alkaloids.

Free Alkaloids:

10 ml of organic layer (diethyl ether) was evaporated to dryness. The residue was then dissolved in 1.5 ml of HCl 2 % and treated with two drops of Mayer's reagent. Turbidity and formation of creamy white precipitate were regarded as evidence for the presence of free alkaloids in the extract and all results were compared with blanks.

Test for Saponins:

2 g of the powdered leaves or fruit was introduced into a beaker containing 100 ml of distilled water; the mixture was boiled in a water bath and filtered. The filtrate was completed and then to 100 ml with water. In ten test tubes were introduced the following volumes (1, 2 ... 10 ml) of the mother solution. Then the final volume was readjusted to 10 ml with distilled water. All tubes were vigorously shaken for 15s; the formation of froth indicated the presence of saponins.

Test for Glycosides:

Each extract was hydrolyzed with HCl and neutralized with NaOH solution. A few drops of Fehling solutions A and B were added. The red color indicates the presence of Glycosides.

Table 2: Preliminary Phytochemical screening of leaf extract of *Annona squamosa*

Constituents	Pet ether extract	Chloroform extract	Methanol extract	Water extract
Steroids	+	+	-	-
Triterpene	-	+	-	-
Glycoside A	+	+	+	+
Alkaloids	-	-	+	+
Flavonoids	-	-	+	+
Saponins	-	-	+	+
Phenolic compounds	+	+	+	+

Key words: present: +, absent: -

Test for Flavonoids: 0.2g of each extract was dissolved in diluted NaOH and a few drops of HCL were added. Yellow solutions that turn colorless indicate the presence of flavonoids.

Test for Steroids: 2ml of acetic anhydride was added to the mixture of 0.5 g of each extract and H₂SO₄ (2 ml). The color from violet to green in some samples indicates the presence of steroids.

Test for Terpenoids: 0.2 g of each extract was mixed with 2 ml of chloroform and concentrated (3ml) H₂SO₄ was carefully added to form a layer. The formation of reddish-brown coloration at the interface indicates the presence of terpenoids.

Test for phenols: A small amount of the ethanolic extract was taken with 1 mL of water in a test tube and 1 to 2 drops of Iron III chloride (FeCl₃) was added. A blue, green, red, or purple color is a positive test

Test for Cardiac glycosides:

The Keller-Killani test was performed to assess the presence of cardiac glycosides. 5 ml of each extract was treated with 2 ml of glacial acetic acid containing one drop of ferric chloride solution. This was under layered with 1 ml of concentrated sulphuric acid. A brown ring of the interface indicates a deoxy sugar characteristic of cardenolides. A violet ring may appear below the brown ring, while in the acetic acid layer, a greenish-blue color ring may form just gradually throughout the thin layer indicating the presence of cardiac glycosides.

Results and Discussion: -

The results obtained from the phytochemical screening of *A. squamosa* are presented in (Table -2). The *n*-hexane fraction exhibited the presence of terpenoids, tannins, and saponins while the rest of the phytochemical was not detected. The chloroform fraction showed the accumulation of terpenoids, tannins and saponins, glycosides, reducing sugar, and amino acids. When ethyl acetate was tested for various phytochemical tests, it confirmed the presence of terpenoids, tannins, and saponins, reducing sugar, beta cyanin, and amino acids. The antibacterial effect of various solvent fractions of *A. squamosa* is presented in (Table 2). The medicinal value of the plant can be correlated to the presence of various bioactive secondary metabolites. The *n*-hexane and ethyl acetate fractions and methanolic extract of the *A. squamosa* showed the presence of terpenoids which exhibits antiviral and antibacterial activities,

Extensive research has been carried out since 1925 for the isolation and characterization of the total alkaloid contents in *A. squamosa* species (Berkov *et al.*, 2006; Dovelana *et al.*, 2006). Vitale *et al.* (1995) have shown that the total alkaloid content in *A. squamosa* varies from 0.02 to 0.52% and scopolamine from 0.0029 to 0.32% relative to the dried material, depending on the geographical area, the part of the plant studied and the stage of growth.

My findings correlate with the observations of previously screened medicinal plants for antimicrobial activity, where most of the active plants showed activity against selected bacterial strains. The pharmacological activity of *A. squamosa* was confirmed by the antimicrobial assay of various fractions and methanolic extract. Among the tested bacteria, *Klebsiella pneumonia* and *Staphylococcus aureus* exhibited complete resistance against all the tested solvent fractions except *n*-hexane. The chloroform fraction exhibited 16-, 12-, 10- and 10-mm zone of inhibition against *Klebsiella pneumonia*, *Staphylococcus aureus*, *S. Typhimuriun*, and *Bacillus subtilis* respectively. While the ethyl acetate was tested against these bacterial strains, it caused activity only against *Staphylococcus aureus* 10 mm zone on inhibition. Methanol fraction demonstrated moderate antibacterial effect against *Klebsiella pneumonia*, *S. Typhimuriun*, *Staphylococcus aureus*, and *Bacillus subtilis* with a zone of inhibition ranging from 10-12. Our results suggest that further work is needed to locate the active principles from the various extracts or fractions and such efforts could result in the discovery of new compounds possessing a wide range of bioactivity for the treatment of infectious diseases.

Antifungal activity

Using the agar well diffusion method, the effects of methanol, chloroform, and an aqueous extract of *Annona squamosa* L. leaves were assessed. Inhibitory minimum concentration for the broth microdilution method using various strains of fungi *Aspergillus niger*, *Microsporium canis*, *Candida albicans*, *Alternaria alternate*, and *Fusarium solani* was investigated. The comparison of antimicrobial activity between various parts of *A. squamosa* such as root, leaf, seeds, and cotyledon was made. The antimicrobial activity of methanol extract of the above-mentioned plant parts was evaluated against four fungi *Trichophyton rubrum*, *Aspergillus niger*, *Candida albicans*, and *Aspergillus flavus*. Agar well diffusion was used to examine the effects of methanol, chloroform, and an aqueous extract of *A. squamosa*. leaves. the smallest inhibitory dose.

Anticancer Activity

Evaluation of anticancer on peel, seed, and pulp was performed using MTT assay method where sulfated polysaccharide, tannins, flavonoid, and triterpenoid were also calculated using RP-HPLC.²⁸ Lebanese *Annona squamosa* L. seeds were used in the phytochemical screening and cytotoxic activity of the extracts, which demonstrated antioxidant, anti-diabetic, and anti-proliferative properties due to the presence of several secondary metabolites such as flavonoids and phenol.²⁹ Table 1.3 showing anticancer activity of *Annona Squamosa*.

The findings of an evaluation of *A. squamosa* seeds oil's anticancer efficacy revealed that the oil inhibits the growth of H22 solid tumors, which may be because of the oil's unsaturated fatty acid composition and level of total annonaceous acetogenins (ACGs). Numerous pathways, including vascular, adipose tissues, inflammation, structural, and physiological, are involved in cellulite. Cellular lipid accumulation, anti-platelet aggregation, and microcirculation were evaluated for three plants where *Rosmarinus officinalis*, and *A. squamosa* L. showed a reduction of lipid accumulation whereas *Rosmarinus officinalis* showed inhibition in platelet aggregation and *Zanthoxylum clavaherculis* showed a reduction of the recto-anus coefficient by 79.6% which result in the improvement in microcirculation. This results in a formulation with a standardized composition which suggested its usage for cellulite.

Immunostimulant Activity

Hematological tests were used to assess the immunostimulant activity of *A. squamosa* aqueous extract in which phytochemical analysis was performed, and the findings showed that alkaloids, steroids, tannins, phenols, reducing sugar, saponin, and flavonoids were present. The findings supported the plant's immunomodulatory properties. Gas chromatography-mass chromatography (GC-MS) and gas chromatography-flame ionization detector (GC-FID) was used to evaluate the chemical makeup of the essential oil of *A. squamosa* L. leaves from northern India. The majority of sesquiterpenoids and oxygenated sesqui terpenoids were obtained. (E)-caryophyllene, epi- α -cadinol, β -elemene, (Z) caryophyllene, α -humulene, viridiflorene, caryophyllene oxide, spathulenol, (2Z,6Z)-farnesal and many more sesquiterpenoids were also obtained. The monoterpenoids present in the essential oil were p-cymene, limonene, and bornyl acetate. The major constituents of sesquiterpenoids were (E)-caryophyllene, epi- α -cadinol, (Z)-caryophyllene, α -humulene, γ -cardinene, viridiflorene and γ -muurolene.

Significant impact especially in relation to an average of antibody titer where gumbo (like HIV) affects the profile of antibody titers from ethanol extract of leaves whereas fruits ethanol extract showed cell-mediated and humoral immunity in mice as compared to control animal which confirms the good potential of the immunomodulatory activity.

Antimalarial activity

In recent research on *Annona squamosa* L., all compounds have shown modest effectiveness against a chloroquine-sensitive strain and a chloroquine-resistant strain of *Plasmodium falcifarum*. Isolation of antimalarial compounds including N-Nitrosoxylopine (1), Roemerolidine (2) and Duguevalline which was isolated from bark showed moderate activity against chloroquine resistant strain.

Anti-inflammatory property

In order to study the effects of an ethanolic extract of *Annona squamosa* L. seeds, indomethacin was utilized as the reference medication and a technique to produce hind paw edema with carrageenan was employed. The results of the phytochemical screening indicated the presence of amino acids, reducing sugar, phenolic compounds, alkaloids, and tannins. A 100mg/kg dose of *Annona squamosa* L. seeds resulted in substantial anti-inflammatory effects.⁵¹

Novel Formulation Approaches *Annona squamosa* L.

Plant extracts and isolates have improved over past years in the creation of novel medication delivery methods. *Annona squamosa* L. showed significant antimicrobial activity, antibacterial, insecticidal activity, anticancer Activity, hepatoprotective, anti-genotoxic, anthelmintic activity, antimalarial activity, Antidiabetic activity, anti-inflammatory property, and many more pharmacological activities. To justify various therapeutic activities and effective therapeutic treatment involving better absorption of the drug with minimum risk of complications wh, their basis for the production of herbal formulation.

Tetranychus urticae was tested for fatal toxicity after a formulation of *Annona squamosa* L. extract microencapsulation. The microencapsulation was shown to be effective in killing the two-spotted spider mite and might be utilized to control it.

In streptozotocin-induced diabetic rats, the effects of a polyherbal formulation containing *Annona squamosa* L. and *Nigella sativa* on blood sugar, plasma insulin, tissue lipid profile, and lipid peroxidation were examined. Emulgel loaded with *Annona squamosa* L. extract with and without penetration enhancer where the formulation was evaluated at various storage conditions. The result showed good pharmaceutical stability that helps to utilize in the cosmetic and pharmaceutical and cosmetic industry.

Conclusion

Medicinal plants are considered rich sources of phytochemicals that can be utilized for drug formulation and development. Natural medicines are a reliable and safe approach for the treatment and prevention of various diseases compared to synthetic drugs. Additionally, herbal drugs are economical due to their low cost compared to synthetic drugs.

Custard apple is the popular name for the medicinal plant *A. squamosa*. (Annonaceae). It has a long history of usage as a purgative, vermicide, insecticide, and therapy for abscesses, infertility, and cancer. The reported pharmacological properties include antidiabetic activity, hepatoprotective activity, antimalarial activity, antibacterial, antiulcer, wound healing, anti-inflammatory, anti-microbial, and several other medicinal properties. Phytoconstituents investigations considered as alkaloids i.e. samoquasine A, N-nitrosoxylopinine, etc., flavonoids i.e. rutin, quercetin, bullaracin etc., entkaurene diterpenoid i.e. Annomosin A, Annosquamosin etc., glycoside i.e. quercetin-3glucoside, essential oil i.e. germacrene, β -elemene, etc. as secondary metabolites. According to research and development, novel drug delivery and polyherbal formulations have a number of advantages over conventional formulations, including improved solubility, bioavailability, and protection from toxicity, pharmacological activity, stability, improved tissue macrophage distribution, sustained delivery, and defense against physical and chemical deterioration. The future investigation of phytochemistry and pharmacological studies of the plant will have a fruitful scope with justified evidence-based herbal formulation as phytotherapeutic medicine for the purpose of prevention and treatment of severe disease. In further future, *A. squamosa* L. plant needs to explore investigation in various emerging areas with possible clinical studies that can be useful for the welfare of mankind.

References: -

- Berkov S, Zayed R, Doncheva T (2006).** Alkaloid pattern in some varieties of *Datura stramonium*. *Fitoterapia* 77:179-182.
- CDC (2002).** Suspected Moonflower Intoxication. *United States Media Report*. Ohio.
- Natural Resources Conservation Service (NRCS). Plants profile *Annona squamosa* L. United States Department of Agriculture. 2008: 4-17.
- Germplasm Resources Information Network (GRIN) (1997).** *Taxonomy for Plants*. USDA, ARS, National Genetic Resources Program.:7-11.
- Raj Sobiya. D, Vennila Jannet. J, Aiyavu. C, Panneerselvam (2009).** The hepatoprotective effect of alcoholic extract of *Annona squamosa* leaves on experimentally induced liver injury in swissalbino mice. *International Journal of Integrative Biology*; 5(3);pp 182-186.
- Jonathan H. Crane, Carlos F(1994).** Balerdi, and Ian Maguire. Sugar Apple Growing in the Florida Home Landscape. 4: 4-19.
- Cochrane C B, Nair P K, Melnick S J, Resek A P, Ramachandran C (2008).** Anticancer effects of *Annona glabra* plant extracts in human leukemia cell lines. *Anticancer Research* ; 28 (2A): 965–71.

Gonzalez-Trujano M E, Lopez-Meraz L, Reyes-Ramirez A, Aguilon M, Martinez A (2009) Effect of repeated administration of *Annona diversifolia* Saff extracts and palmitone on rat amygdala kindling. *J. pubmed.* 16(4):590-5.

Eva M, Gonzalez-Trujano (2006) Anticonvulsant effect of *A. diversifolia* and *Palmitone epilepsy*, Blackwell Publishers. 47(11):1810–1817.

Ecology and evolutionary biology plant growth facilities. 5th Edn CBS Publishers. 2007 :56-59.

Oliveira L C, Hankerson S J, Dietz J M, Raboy B E (2010). Key tree species for the golden-headed lion tamarin and implications for shade-cocoa management in southern Bahia, Brazil, *Animal Conservation.* 2010; 13: 60.

Nehey pandya (2011). Pharmacological and phytochemical review on *Annona squamosa*. *Int .J. res.in pharm and biomedical sciences.* 2011; 2(4):1404-1414.

Current name: *Annona cherimola*. Agro-Forestry Tree Database. International center for research in agroforestry. 2008:78

Taha SA (1984). Datura intoxication in Riyadh. *Trans. R Soc. Trop. Med. Hyg.*, 78(1): 134 -135.

Vitale AA, Acher A, Pomilio AB (1995). Alkaloids of *Datura ferox* from Argentina. *J. Ethanopharmacol.* 49:81-89.

Yamazaki Z, Tagaya I (1980). Antiviral effects of atropine and caffeine. *J. Gen. Virol.* 50:429-431.

