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PHYTOCHEMICAL EVALUATION OF MANGO BARK

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ABSTRACT

The Mango Bark Extraction Project explores the sustainable utilization of mango tree bark for its medicinal and industrial properties. This initiative focuses on the systematic collection, processing, and extraction of bioactive compounds from mango bark, particularly targeting tannins, polyphenols, and flavonoids, which have demonstrated antimicrobial, antioxidant, and anti-inflammatory effects. The present study investigates the phytochemical composition of Mangifera indica *(mango) bark extract obtained through Soxhlet extraction using ethanol as the solvent. The project outlines a detailed methodology involving the drying, pulverizing, and solvent extraction of the bark, followed by quality analysis and potential application in pharmaceuticals, cosmetics, and natural preservatives. With an emphasis on environmental conservation and resource optimization, the project aims to add value to agricultural by-products while promoting eco-friendly extraction practices and opening avenues for commercial-scale production.*

KEYWORDS: *Mangifera , Phytochemical, Soxlet Extraction, Antioxidant, Research.*

INTRODUCTION

There has been a tremendous pressure on medicinal plants for their extensiveUtilization as sources of raw materials for the pharmaceutical industries. Demands forMedicinal plants are rapidly increasing not only indeveloping countries but also in theDeveloped ones. Medicinal plants have various effects on living systems. Some areSedatives, analgesic, antipyretics, cardio-protective, anti-inflammatory, antioxidants,Antispasmodics, and immunomodulatory functions.The research for the novel compounds effective against plasmodium strainsResistant to widely used synthetic drugs, coronary arteriosclerosis, cardiovascular, Neurological, endocrine, respiratory, immune and self-immune, ischeamia, gastric disorder, Tumor progression and carcinogensis has led to increased interest in new and existing Information about the remedies of these diseases from natural sources 1-5. Plants form the Major part of treatments used by traditional healers in many societies.

Mangifera indica, commonly known as mango, is an evergreenspecies of flowering plant in the family Anacardiaceae. It is a large fruit tree, capable of growing to a height and width of 30 m (100 ft).There are two distinct genetic populations in modern mangoes – the "Indian type" and the "Southeast Asian type".

SCIENTIFIC CLASSIFICATION

Kingdom:Plantae
Clade:Tracheophytes
Clade:Angiosperms
Clade:Eudicots
Clade:Rosids
Order:Sapindales
Family:Anacardiaceae
Genus:Mangifera
Species:M. indica

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Binomial name: Mangifera indica
Synonyms :

- Mangifera amba Forssk.
- Mangifera anisodora Blanco
- Mangifera austroyunnanensis Hu
- Mangifera balba Crevost & Lemarié
- Mangifera cambodiana (Pierre) Anon.
- Mangifera domestica Gaertn.
- Mangifera equina Crevost & Lemarié
- Mangifera gladiata Bojer
- Mangifera kukulu Blume
- Mangifera laxiflora Desr.
- Mangifera linnaei Korth. Ex Hassk.
- Mangifera maritima Lechaume
- Mangifera mekongensis (Pierre) Anon.
- Mangifera montana B.Heyne ex Wight & Arn.
- Mangifera oryza Crevost & Lemarié
- Mangifera rostrata Blanco
- Mangifera rubra Bojer
- Mangifera sativa Roem. & Schult.
- Mangifera siamensis Warb. Ex Craib
- Mangifera viridis Bojer

The mango tree (Mangifera indica Linn Anacardiaceae) is naturalized in West Africa. The tree originated from India to West Africa. The bark and leaves have Astringent prosperities and are used in Nigeria as lotion to relieve toothache, sore gums, Sore throat or as an infusion in malaria, diarrhoea and dysentery treatment . All the organs Of the plants are rich in tannins and flavonoids5,7. In the leaves of the West African Species, four anthocyanidins (3 – monosides of delphinidin, paeonidin, and cyanidin)Leucoanthocyanins, catechic and gallic tannins, mangiferin (flavoric heteroside), Kaempferol and quercitin (both free and glycosides) were reported to be phytoconstituents Of West African species. The mango tree is rich in phytochemicals, which are vital in Health promotion, disease prevention and drug production. Phytochemicals act as Antioxidant, stimulate the human system, induce protective enzymes in the liver or block Damage to genetic materials . Phytochemicals exhibit a wide range of biological Functions due to their antioxidant properties. Several types of polyphenols (phenolic acid, Hydroly-sable tannins and flavonoids) show anti-carcinogenic and anti-mutagenic Effects. Polyphenols interfere in many steps of malignant tumors, inactivating Carcinogens, inhibiting the expression of mutagens and the activity of enzymes involved in The activation of procarcinogens. Polyphenols activate enzymatic systems, which are Responsible for the detoxification of xenobiotics . Polyphenolic flavonoids inhibit the Initiation, promotion and progression of tumors. Recently, plant flavonoids have attracted The attention of researchers as potentially important dietary cancer chemo-protective and Preventive agents. Naturally occurring flavonoids are potentially antiallergic,Anticarcinogenic, anti-viral and antioxidant. Phytochemicals regulates, protects and Control prostrate and testicular cancer and semen quality in men. Isoflavones, which are Effective phytoestrogens modulate estrogen levels in humans. They are of clinical value in Low astrogens states like menopause, or imbalanced and toxic estrogen – sensitive Conditions such as breast, uterine and prostrate tumor growth. Isoflavones prevent Breast cancer, cystic ovaries and endometriosis among women.

Mango extracts from leaves, fruit, seed kernel, fruit pulp, roots, bark and stem bark Have been used extensively for medicinal purposes in many countries. The ethnomedical Use of mango stem bark aqueous extract in Cuba has been documented widely. It has Been extensively used in cancer, diabetes, asthma, infertility, lupus, prostatisis, prostatic Hyperplasia, gastric disorders, arthralgies, mouth sores and tooth pain.

The phytochemical profile of mango stem barks has been reported. Mangiferin is The major component in mango stem bark. Other flavonoids and flavonol constituents Include quercetin, catachin and epicatechin. The phytochemical screening of the Raw material showed thatpolyphenols, terpenoids, sugars and saponins were present in Mango stem bark. Many phenolic constituents, benzoic acids and its propylester ,Three free sugars (galactose, glucose and arabinose) and three polyalcohols (sorbitol, Myoinositol and xylitol) were identified and tested from the mango fruits and stem bark Related constituents of mango stem barks. Volatile components of mango stem bark wereExtracted, and the major constituents identified were β-relemens, aromandrene, α-guaiene, B-endesmol, β-sitrosterol and β-campesterp . The amount of sesquiterpenoid Hydrocarbons was higher than the oxygenated compounds. The composition of the fatty

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Acid fraction was also determined as their trimethylsilyl esters derivatives and the major Constituents were palmitic, oleic and linoleic acids with a minor proportion of myristic and Estearic acids . Many polyunsaturated and dicarboxylic acids of biological importance Such as eicosatriencoic, succinic and malonic were found to be present in mango stem bark In trace levels. Mangifera indica stem bark has antioxidant, anti-inflammatory and Immunoduletory properties and have been developed in Cuba as nutritional supplement or Functional food in several formulations (antioxidant) or anti-inflamatory, analgesic and Immunomodulator (tablet, capsule, syrup and cream) and have been extensively used to Prevent disease progress or improve the patient's quality of life in diseases like HIV/AIDS, Cancer, asthma, gastric and dermatological disorders

The present investigation was undertaken to quantify the percentage constituent of Crude phytochemicals, vitamins and minerals present in the leaves and stem bark of Mangifera indica and consequently assess the potential raw materials for drug formulation.

BOTANICAL DISCRIPTION

MI is a large evergreen tree in the anacardiaceae family that grows to a height of 10-45 m, dome shaped with dense foliage, typically heavy branched from a stout trunk. The leaves are spirally arranged on branches, linear-oblong, lanceolate – elliptical, pointed at both ends, the leaf blades mostly about 25-cm long and 8-cm wide, sometimes much larger, reddish and thinly flaccid when first formed and release an aromatic odour when crushed. The inflorescence occurs in panicles consisting of about 3000 tiny whitish-red or yellowish – green flowers. The fruit is a well known large drupe, but shows a great variation in shape and size. It contains a thick yellow pulp, single seed and thick yellowish – red skin when ripe. The seed is solitary, ovoid or oblong, encased in a hard, compressed fibrous endocarp.

HABITAT

It is native tropical Asia and has been cultivated in the Indian subcontinent for over 4000 years and is now found naturalized in most tropical countries.
Parts used: Roots, bark, leaves, fruits, seeds, flowers and kernels are used.

PHYTOCHEMISTRY

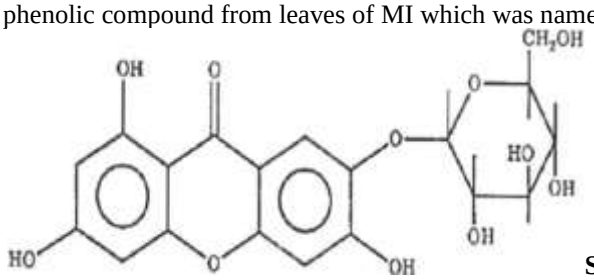
Chemical constituents of MI are always of an interest. The different chemical constituents of the plant, especially the polyphenolics, flavonoids, triterpenoids. Mangiferin a xanthone glycoside major bio-active constituent, isomangiferin, tannins & gallic acid derivatives. The bark is reported to contain protocatechic acid, catechin, mangiferin [Figure 1], alanine, glycine, γ-aminobutyric acid, kinic acid, shikimic acid and the tetracyclic triterpenoids cycloart-24-en-3β,26diol, 3-ketodammar-24 €-en-20S,26-diol, C-24 epimers of cycloart-25 en 3β,24,27-triol and cycloartan-3β,24,27-triol.[1]

Indicoside A and B, manghopanal, mangoleanone, friedelin, cycloartan-3β-30-diol and derivatives, mangsterol, manglupenone, mangocoumarin, n-tetacosane, n-heneicosane, n-triacontane and mangiferolic acid methyl ester and others isolated from stem bark of MI.[2] Mangostin, 29-hydroxy mangiferonic acid and mangiferin have been isolated from the stem bark together with common flavonoids.[3] The flower yielded alkyl gallates such as gallic acid, ethyl gallate, methyl gallate, n-propyl gallate, n-pentyl gallate, n-octyl gallate, 4-phenyl gallate, 6-phenyl-n-hexyl gallate and dihydrogallic acid.[4] Root of mango contains the chromones, 3-hydroxy-2-(4'-methylbenzoyl)-chromone and 3-methoxy-2-(4'-methyl benzoyl)-chromone. The leaf and flower yield an essential oil containing humulene, elemene, ocimene, linalool, nerol and many others. The fruit pulp contains vitamins A and C, β-carotene and xanthophylls.[5] An unusual fatty acid, cis-9, cis-15-octadecadienoic acid was isolated from the pulp lipids of mango.[6] Phenolic Antioxidants, Free Sugars and Polyols isolated and analyzed from Mango (MI) Stem Bark. All structures were elucidated by ES-MS and NMR spectroscopic methods. Quantitative analysis of the compounds has been performed by HPLC, and mangiferin was found to be the predominant component.[7]

Polyphenols have been characterized in mango puree concentrate by HPLC with diode array and mass spectrometric detection.[8] A rapid method was developed for quantitative determination of beta-carotene, including cis-isomers, in dried mango.[9] HPLC method was developed to determine carotenoids in Taiwanese mango.[10] 5-Alkyl- and 5-alkenylresorcinols, as well as their hydroxylated derivatives, extracted from mango (MI) peels, purified on polyamide and characterized by high-performance liquid chromatography/atmospheric pressure chemical ionization mass spectrometry (HPLC/APcI-MS) for the first time.[11] Xanthophyll esters, carotenes, and tocopherols has been identified and quantified in the fruit of seven mexican mango cultivars by liquid chromatography-atmospheric pressure chemical ionization-time-of-flight mass spectrometry [LC-(APcI (+))-MS].[12] A simple, precise, and rapid HPTLC method was established for quantitative determination of the bioactive marker compound mangiferin in the stem bark & leaves of MI. The method was validated for selectivity, linearity, precision, accuracy, and robustness.[13] The natural C-

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glucoside xanthone mangiferin [2-C-β-Dgluco-pyranosyl-1,3,6,7-tetrahydroxyxanthone; C19H18O11; Mw, 422.35; melting point, anhydrous 271°C[14] has been reported in various parts of MI leaves,[15] fruits, stem bark, heartwood and roots. The presence of a phenolic compound from leaves of MI which was named as homomangifirin.[16]



Structure of Mangiferin

PHARMACOLOGY

Although a lot of pharmacological investigations have been carried out based on the ingredients present but a lot more can still be explored, exploited and utilized. A summary of the findings of these studies is presented below.

Anti-oxidant: Reactive oxygen species (ROS) possess a strong oxidizing effect and induce damage to biological molecules, including proteins, lipids and DNA, with concomitant changes in their structure and function. The major nutritional antioxidants, vitamin E, vitamin C and β-carotene, may be beneficial to prevent several chronic disorders considerable interest has arisen in the possible reinforcement of antioxidant defenses, both for chemoprevention and treatment purposes. The extract showed a powerful scavenging activity of hydroxy radicals and acted as a chelator of iron. It also showed a significant inhibitory effect on the peroxidation of rat brain phospholipid and prevented DNA damage caused by bleomycin or copper-phenenthroline systemsThe interaction of Vimang (MI extract) with Fe (III) was studied and the results justify the high efficiency of Vimang as an agent protecting from iron-induced oxidative damage.The work has been carried out to investigate the pulp composition of four mango cultivars (Haden, Tommy Atkins and Ubá) at the ripening stage in relation to three components with antioxidant potential (total phenolics, carotenoids and ascorbic acid). These results corroborated previous information that mangoes are a good source of antioxidants in human diet.In vitro antioxidant and free radical scavenging properties of a stem bark aqueous extract of mango tree (MI), whose formulations are used in Cuba as food supplements under the brand name of Vimang, Luminol-enhanced chemiluminescence was used to elucidate the effect of this extract on the generation of reactive oxygen species in PMA- or zymosan-stimulated human polymorphonuclear leukocytes and on superoxide radicals generated in the hypoxanthine–xanthine oxidase reaction. Part of this MI extract antioxidant activity could be ascribed to the presence of mangiferin as its main component] The iron-complexing ability of Vimang as a primary mechanism for protection of rat liver mitochondria against Fe2+-citrate-induced lipoperoxidation was reported. The results are of pharmacological relevance since Vimang could be a potential candidate for antioxidant therapy in diseases related to abnormal intracellular iron distribution or iron overload. The protective abilities of MI stem bark extract (Vimang) 50-250 mgkg(-1), mangiferin 50 mgkg(-1) and selected antioxidants (vitamin C 100 mgkg(-1), vitamin E 100 mgkg(-1)and beta -carotene 50 mgkg(-1)) against the 12-O-tetradecanoylphorbol-13-acetate (TPA)-induced oxidative damage in serum, liver, brain as well as in the hyper-production of reactive oxygen species (ROS) by peritoneal macrophages was compared.

Anti-diabetic: A 50% ethanolic extract of the leaves of MI produced a significant hypoglycemic effect at a dose of 250 mg/kg, both in normal and streptozotocin-induced diabetic animals. The stimulation of β-cells to release insulin was thought to be part of the mechanism of action.The effect of the aqueous extract of the leaves of MI on blood glucose level in normoglycaemic, glucose – induced hyperglycaemic and streptozotocin (STZ)-induced diabetic rats has been assessed. The results indicate that the aqueous extract of the leaves of MI possess hypoglycaemic activity. This action may be due to an intestinal reduction of the absorption of glucose.The leaves of MI used for antidiabetic properties using normoglycaemic, glucose-induced hyperglycaemia and streptozotocin (STZ) induced diabetic mice. The aqueous extract of the leaves of MI possess hypoglycaemic activity.The effect of mango (MI) ingestion on blood glucose levels of normal and diabetic rats has been studied. The results from this research suggest that mango flour can possibly help in the treatment of diabetes.The stem-bark of aqueous extract of MI was used to examine the antiinflammatory, analgesic and antidiabetic properties. The different chemical constituents of the plant, especially the polyphenolics, flavonoids, triterpenoids, mangiferin, and other chemical compounds present in the plant may be involved in the observed antiinflammatory, analgesic, and hypoglycemic effects of the plant's extract. The results of this experimental animal study lend pharmacological credence to the suggested folkloric uses of the plant in the management and control of painful, arthritic and other inflammatory conditions, as well as in the management of adult-onset type 2 diabetes mellitus in some rural African communities.Investigations were carried out to evaluate the effect of MI on glucose absorption

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using a rat intestinal preparation in situ. The ethanol extracts of stem-barks reduced glucose absorption gradually during the whole perfusion period in type 2 rats. In glucose-loaded normal rats, mangiferin induces a significant improvement in oral glucose tolerance but without alteration of basal plasma glucose levels these studies show that mangiferin (10 and 20 mg/kg, i.p.) exhibits potent antidiabetic, antihyperlipidemic, antiatherogenic and antioxidant properties without causing hypoglycaemia; mangiferin would then offer a greater therapeutic benefit for the management of diabetes mellitus and diabetic complications associated with abnormalities in lipid profiles. It has been reported that long standing hyperglycaemia with diabetes mellitus leads to the formation of advanced glycosylated end-products which are involved in the generation of ROS, leading to oxidative damage, particularly to heart and kidney.
Antiviral activity: In vitro the effect of mangiferin was studied against Herpes simplex virus type 2; mangiferin does not directly inactivate HSV-2 but inhibits the late event in HSV-2 replication.[34] In vitro mangiferin was also able to inhibit HSV-1 virus replication within cells[35] and to antagonize the cytopathic effects of HIV.

Anthelmintic and anti-allergenic activity : Anthelminthic and antiallergic activities of MI stem bark components Vimang and mangiferin was investigated in mice experimentally infected with nematodes, Trichinella spiralis. The study was carried out to find out anti-allergic properties of vimang and mangiferin, a C-glucosylxanthone isolated from extract of MI. The results constitute the anti-allergic properties of Vimang on allergic models, as well as suggesting that this natural extract could be successfully used in the treatment of allergic disorders. Mangiferin, the major compound of Vimang, contributes to the anti-allergic effects of the extract.

Antiparasitic activity: In a neonatal mouse model, mangiferin at 100 mg/kg has a similar inhibitory activity on Cryptosporidium parvum than the same dose (100 mg/kg) of an active drug, paromomycin.

Antibone resorption : Four water extracts of Kampo formulae were screened for their inhibitory effect on bone resorption induced by parathyroid hormone in organ culture of neonatal mouse parietal bones. Mangiferin isolated and tested in vitro showed a significant inhibitory effect on this model.

Anti-tumor-anti-HIV: The significant cytotoxic activities has been demonstrated by the stem bark extract of mango against the breast cancer cell lines MCF 7, MDA-MB-435 and MDA-N, as well as against a colon cancer cell line (SW-620) and a renal cancer cell line (786-0). The ethanol/water (1:1) extract of dried aerial parts of mango administered intraperitoneally to mice at a dose of 250.0 mg/kg was inactive on Leuk-P388. In vitro, mangiferin dose- and time-dependently inhibited the proliferation of K562 leukemia cells and induced apoptosis in K563 cells line, probably through down-regulation of bcr/abl gene expression.These results suggest that mangiferin has a potential as a naturally-occurring chemopreventive agent.

Antispasmodic and antipyretic activity : The stem bark extract of MI was evaluated for antiplasmodial activity against Plasmodium yoelii nigeriensis. The extract was also screened for antipyretic activity in mice. The extract exhibited a schizontocidal effect during early infection, and also demonstrated repository activity. A reduction in yeast-induced hyperpyrexia was also produced by the extract.[45] The in vitro antimalarial activity of chloroform: methanol (1:1) extract of MI was evaluated. The extract showed a good activity on P. falciparum in vitro with a growth inhibition of 50.4% at 20 µg/mL.

Immunomodulatory: Immunomodulatory activity of alcoholic extract of stem bark of MI was investigated in mice. It is concluded that test extract is a promising drug with immunostimulant properties. Mangiferin mediates the down-regulation of NF-xB, suppresses NF-xB activation induced by inflammatory agents, including tumor nuclear factor (TNF), increases the intracellular glutathione (GSH) levels and potentiates chemotherapeutic agent-mediated cell death; this suggests a possible role in combination therapy for cancer. It is likely that these effects are mediated through mangiferin ROS quenching and GSH rising; increased intracellular (GSH) levels are indeed known to inhibit the TNF-induced activation of NF-κB.

Anti-diarrhoeal : The potential anti-diarrhoeal activity of methanolic (MMI) and aqueous (AMI) extracts of seeds of MI has been evaluated in experimental diarrhoea, induced by castor oil and magnesium sulphate in mice. The results illustrate that the extracts of MI have significant anti-diarrhoeal activity and part of the activity of MMI may be attributed to its effect on intestinal transit.

Anti-inflammatory : An ethanolic (95%) extract of the seed kernel of MI exhibited significant anti-inflammatory activity in acute, subacute and chronic cases of inflammation. The MI leaf extract exhibited antibacterial activity against Bacillus subtilis, staphylococcus albus and Vibrio cholerae. Analgesic and anti-inflammatory effects of MI extract (Vimang) has studied. The polyphenols found in the extract were found to account for the activity reported In vivo and in vitro anti-inflammatory activity of MI extracts (VIMANG) was investigated. MI extract, administered topically (0.5-2 mg per ear), reduced ear edema induced by arachidonic acid (AA) and phorbol

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myristate acetate (PMA, ED50 = 1.1 mg per ear) in mice. The results represent an important contribution to the elucidation of the mechanism involved in the anti-inflammatory and anti-nociceptive effects reported by the standard MI extract VIMANG.
Anti-bacterial and antifungal activity : In an in vitro agar diffusion technique, mangiferin showed activity against 7 bacterial species, Bacillus pumilus, B. cereus, Staphylococcus aureus, S. citreus, Escherichia coli, Salmonella agona, Klebsiella pneumoniae, 1 yeast (Saccharomyces cerevisiae) and 4 fungi (Thermoascus aurantiacus, Trichoderma reesei, Aspergillus flavus and A. fumigatus).
Anti-microbial : The antimicrobial activities of methanolic extracts of P. guajava and MI have been investigated. The results show that P. guajava and MI extracts exhibited antimicrobial activities at a concentration of 20 mg/ml. Overall, P. guajava extract show mo

USES
Mango stem bark has been traditionally used for various medicinal purposes, including treating menorrhagia, diarrhea, syphilis, and diabetes. It's also used for scabies, cutaneous infections, and anemia. The bark is astringent and believed to have a tonic effect on mucous membranes. Additionally, mango stem bark extract (MSBE) has shown antioxidant properties and has been used in clinical trials for HIV/AIDS, geriatrics, and skin disorders.

Traditional and Modern Uses of Mango Stem Bark
Ethnomedical Evidences
Mango stem bark has been used in various countries for the treatment of various ailments, including menorrhagia, diarrhea, syphilis, diabetes, scabies, cutaneous infections, and anemia.

Production and Chemical Composition
Mango trees, particularly certain varieties, can provide a reliable source of stem bark with a reproducible chemical composition, according to research conducted in Cuba.

Pre-clinical Evidences
The MSBE has shown antioxidant effects both in vitro and in vivo, suggesting potential therapeutic benefits.

Clinical Evidences
Clinical trials with MSBE formulations have demonstrated improvements in patient quality of life in conditions like HIV/AIDS, geriatrics, and skin disorders.

Other Traditional Uses
The bark is also used for treating toothaches, internal hemorrhages, bronchitis, and catarrh. The gum is used for cracked feet and scabies, and the kernel is used to make flour after soaking in water to remove astringent principles.

Astringent Properties
The bark's astringent properties are believed to help with diphtheria and rheumatism.

Anti-syphilitic
The bark is also considered anti-syphilitic.

Rheumatism and Diphtheria
The bark contains mangiferin and is used in India for treating rheumatism and diphtheria.

Antioxidant Properties:
MSBE has shown antioxidant effects, protecting against cell damage and potentially preventing degenerative diseases like cardiovascular diseases and diabetes.

Radioprotective Effects
Mangiferin, a key component of MSBE, has shown radioprotective effects, potentially reducing DNA damage from radiation exposure.

Hepatoprotective Effects
Vimang, an MSBE formulation, has demonstrated hepatoprotective effects in rat hepatocyte cultures.

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Aim and objective of mango bark extraction

Aim

To extract and analyze the bioactive compounds from mango (Mangifera indica) bark for potential therapeutic or industrial applications and to investigate the phytochemical composition and bioactive properties of Mangifera indica (mango) bark through extraction.

Objectives

1. To Collect and Authenticate Mango Bark Samples

Collect mature, disease-free bark samples from Mangifera indica trees.

Use proper botanical identification methods (morphological features, botanical keys) to ensure sample authenticity. This ensures consistency and accuracy in research outcomes.

2. To Prepare the Bark Material for Extraction

Process the collected bark for efficient extraction.

The bark will be cleaned to remove dirt and debris, shade-dried to prevent degradation of heat-sensitive compounds, and ground into fine powder to increase the surface area for better solvent penetration.

3. To Select Appropriate Solvents for Extraction

Choose solvents of different polarities (e.g., water, ethanol, methanol, chloroform, hexane) to extract a wide range of phytochemicals.

Different solvents dissolve different types of compounds. Using a range of solvents ensures maximum recovery of active ingredients like phenols, flavonoids, alkaloids, tannins, etc.

4. To Optimize the Extraction Method and Conditions

Standardize the method (e.g., maceration, Soxhlet extraction, ultrasonication) and optimize parameters like solvent-to-solid ratio, extraction time, and temperature.

Experiment with different conditions to maximize yield without damaging sensitive bioactive compounds. For instance, high temperatures might destroy some antioxidants.

5. To Perform Preliminary Phytochemical Screening

Qualitatively identify the classes of phytochemicals present in the extract.

Standard chemical tests (like Ferric chloride test for phenols, Shinoda test for flavonoids) will be used to determine which types of compounds are present.

6. To Quantify Major Phytochemical Groups

Quantitatively estimate total phenolic content (TPC), total flavonoid content (TFC), and total tannin content (TTC).

Use colorimetric assays such as:

Folin–Ciocalteu method for phenols

Aluminum chloride colorimetric method for flavonoids

Vanillin-HCl method for tannins

METHODOLOGY

Materials

1 Plant Material

Fresh bark of Mangifera indica L. (Family: Anacardiaceae) was collected from mature trees located within the botanical garden of [Insert Institution Name], [Insert City and Country], during the month of [Insert Month, Year]. The trees selected were disease-free and exhibited no visible signs of pest infestation.

2 Chemicals and Reagents

Ethanol (analytical grade, 70% v/v)

Distilled water

Other laboratory consumables: muslin cloth, Whatman No.1 filter paper, aluminum foil

3 Instruments and Apparatus

Mechanical grinder

Rotary evaporator

Soxhlet extractor setup

Hot air oven

Vacuum desiccator

Analytical weighing balance (accuracy ±0.001 g)

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Airtight amber glass containers

Preparation of Plant Material

1 Initial Processing

Upon arrival at the laboratory, the collected mango bark was thoroughly cleaned. Physical impurities like soil, dust, and epiphytes were first removed by washing under running tap water for 5 minutes. The samples were then rinsed with distilled water to ensure complete removal of contaminants and to prevent microbial growth during the drying phase.

2 Drying

Cleaned bark samples were initially air-dried under shade at ambient temperature (25 ± 2°C) with good air circulation. Shade drying was preferred over sun-drying to prevent thermal degradation and photolysis of thermolabile phytoconstituents like flavonoids and phenolic compounds.

The drying process continued for approximately 14 days until the samples were free of moisture, which was checked manually (by brittleness) and confirmed by weight stability. If climatic conditions were humid, supplementary drying was carried out in a hot air oven at 40°C for an additional 24 hours.

3 Grinding and Sieving

After complete drying, the bark samples were manually broken into smaller fragments and ground using a mechanical grinder. The resulting powder was passed through a 40-mesh sieve to ensure uniform particle size, which enhances surface area and solvent penetration during extraction. The fine powder was stored in airtight amber glass containers to protect against oxidation, moisture absorption, and photodegradation until further use.

Extraction of Mango Bark

1 Selection of Extraction Method

Preliminary studies and literature reviews indicated that hydroethanolic extraction (ethanol:water, 70:30 v/v) is ideal for isolating a wide range of phytochemicals, including tannins, flavonoids, polyphenols, and alkaloids. Ethanol is a semi-polar solvent that efficiently extracts both polar and non-polar constituents, and the presence of water increases the extraction of hydrophilic compounds.

Thus, soxhlet extraction approaches were adopted

Maceration Procedure(Alternative Method)

An accurately weighed 100 g sample of mango bark powder was transferred into a sterile 2-liter conical flask. A solvent-to-solid ratio of 10:1 (v/w) was maintained by adding 1000 mL of 70% ethanol.

The flask was sealed with aluminum foil to prevent evaporation and solvent loss. The mixture was allowed to stand at room temperature (25 ± 2°C) for 72 hours with occasional shaking at 12-hour intervals to facilitate better solute-solvent interaction.

After the maceration period, the mixture was filtered first through four layers of muslin cloth to remove coarse debris, followed by filtration through Whatman No.1 filter paper to obtain a clear filtrate. The residue (marc) was subjected to two additional rounds of maceration with fresh solvent under identical conditions to ensure exhaustive extraction. The combined filtrates were collected for concentration.

Soxhlet Extraction

For comparative purposes, Soxhlet extraction was performed using 50 g of mango bark powder placed into a cellulose thimble. Extraction was carried out in a Soxhlet apparatus with 500 mL of 70% ethanol as solvent.

The extraction continued for 6 hours until the siphoning solvent in the thimble became colorless, indicating exhaustion of soluble constituents.

Compared to maceration, Soxhlet extraction offers the advantage of continuous hot percolation, ensuring more complete extraction, but it poses a risk of thermal degradation of sensitive compounds due to prolonged heating.

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5 Concentration and Drying of Extract
The pooled filtrates from both extraction methods were concentrated separately under reduced pressure using a rotary evaporator set at a bath temperature not exceeding 45°C. The use of vacuum distillation minimized solvent loss and prevented thermal degradation.

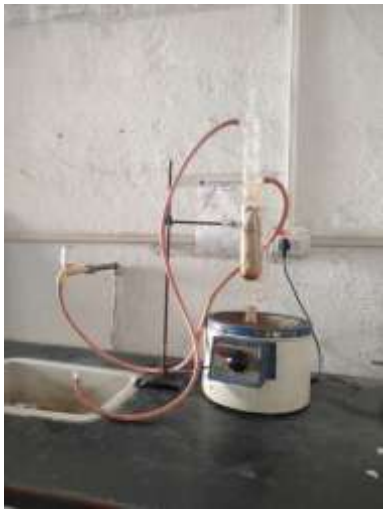
The concentrated extracts were further dried in a vacuum desiccator containing anhydrous silica gel to remove residual moisture until a constant weight was obtained. Dried extracts were carefully scraped, weighed, and transferred into amber-colored glass vials, sealed tightly, and stored at 4°C until subsequent analyses.

6 Determination of Extraction Yield
The percentage yield of the dried extract was calculated using the following formula:
\text{Extraction Yield (\%)} = \left(\frac{\text{Weight of Dried Extract (g)}}{\text{Weight of Initial Plant Material (g)}} \right) \times 100

This parameter provided an estimate of extraction efficiency and was used to compare the performance of different extraction techniques.
7 Preliminary Phytochemical Screening

Qualitative phytochemical screening was performed on the crude extracts to detect the presence of various secondary metabolites, including alkaloids, tannins, saponins, flavonoids, glycosides, steroids, and phenolic compounds. The following standard procedures were used:





Alkaloids: Mayer's test, Dragendorff's test
Tannins: Ferric chloride test
Flavonoids: Shinoda test
Saponins: Frothing test
Phenolics: Lead acetate test

Screening was essential to establish the preliminary phytochemical profile of the mango bark extract, which could guide future bioactivity studies.

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Flowchart for Mango Bark Extraction Process.

Collection of Mango Bark
↓
Authentication by Botanist
↓
Cleaning (Tap Water → Distilled Water)
↓
Shade Drying (7–14 days) / Oven Drying (40–45°C)
↓
Grinding into Fine Powder (40-mesh sieve)
↓
Storage in Airtight Amber Containers
↓
Extraction:
└— Soxhlet Extraction (6 hours, 70% Ethanol)
↓
Filtration (Muslin Cloth → Whatman No.1 Filter Paper)
↓
Concentration (Rotary Evaporator, 45°C)
↓
Drying (Vacuum Desiccator)
↓
Storage of Dried Extract (Amber Vials at 4°C)

Preliminary PhytoChemical Screening

The extracted material from various solvent was subjected to systematic phytochemical screening for the presence of phytoconstituents using standard chemical tests.

Test for Tannins

(a) Acetic Acid Test:- Alcoholic extract is mixed with acetic acid solution gives red color solution.

Test for Steroids

(a) Salkowski Reaction:- To 2ml of extract, add 2ml chloroform and 2ml concentrated H2SO4 shake well. Chloroform layers appears red and acid layer shows greenish yellow fluorescence.

Detection of Carbohydrates

(a) Fehlings Test:- To the test solution, Fehling A and B reagent was added and heated on a water bath, appearance of brick red color or green color indicate the presence of carbohydrates.

Test for Proteins

(a) Biuret Test:- To 3ml test solution add 4% NaoH and few drops of 1% cuso4 solution voilet or pink color appears.

Test for Glycosides

(a) Cardiac Glycosides Test.

Plan of Work

Introduction

Mango bark extraction is a process of isolating bioactive compounds from the bark of mango trees. The bark is rich in phytochemicals, which have potential applications in pharmaceuticals, cosmetics, and food industries.

Extraction Process

The extraction process involves several steps:

1. Collection and preparation of mango bark
2. Selection of extraction method (e.g., solvent extraction, microwave-assisted extraction)

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3. Optimization of extraction parameters (e.g., temperature, time, solvent-to-bark ratio)
4. Filtration and concentration of the extract

Phytochemical Analysis and Biological Activity
The extract is then analyzed for its phytochemical composition and biological activity:
1. Qualitative and quantitative analysis of phytochemicals (e.g., alkaloids, flavonoids, phenolic compounds)
2. Evaluation of biological activity (e.g., antioxidant, antimicrobial, anti-inflammatory)

Soxhalation Process Cycle
The soxhlation process involves a cycle of 62 iterations, which includes:
1. SoxhlationCycle: A process of extraction and purification that involves multiple cycles of solvent extraction, filtration, and concentration.
2. Number of Cycles:62 cycles are performed to optimize the extraction process and obtain high-quality extract.
3. Benefits: The soxhlation process with 62 cycles helps to increase the yield and purity of the extract, making it suitable for various applications.

The soxhlation process cycle involves the following steps:
1. Solvent extraction
2. Filtration
3. Concentration
4. Purification
5. Repeat cycles (62 times)




By incorporating the soxhlation process with 62 cycles, the extraction process can be optimized to obtain high-quality mango bark extract with potential applications.

Solvent Extraction Process for Mango Bark
Overview
Solvent extraction is a method used to extract bioactive compounds from plant materials, such as mango bark. The process involves using a solvent to dissolve and separate the desired compounds from the plant material.

Steps
1. Sample preparation: Grind the mango bark into a fine powder to increase surface area.
2. Solvent selection: Choose a suitable solvent (e.g., ethanol, methanol, water) based on the desired compounds.
3. Extraction: Mix the ground mango bark with the solvent in a suitable ratio.
4. Incubation: Allow the mixture to incubate for a specified period, enabling the solvent to extract the bioactive compounds.
5. Filtration: Filter the mixture to separate the solvent containing the extracted compounds from the plant material.
6. Concentration: Concentrate the extracted compounds using techniques like evaporation or lyophilization.

Factors Affecting Extraction
1. Solvent ratio: The ratio of solvent to plant material can impact extraction efficiency.
2. Temperature: Temperature can influence the extraction process and stability of bioactive compounds.
3. Time: Extraction time can affect the yield and quality of extracted compounds.

Advantages
1. Efficient: Solvent extraction can be an efficient method for extracting bioactive compounds.

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2. Flexible: Various solvents can be used to target specific compounds.

Disadvantages
1. Solvent toxicity: Some solvents may be toxic or hazardous.
2. Residual solvents: Residual solvents may remain in the extracted compounds, requiring additional purification steps.

By optimizing the solvent extraction process, high-quality extracts with desired bioactive compounds can be obtained for further analysis or application.

Filtration Process in Mango Bark Extraction
Overview
Filtration is a crucial step in the extraction process, separating the liquid extract from the solid plant material.

Types of Filtration
1. Gravity filtration: Uses gravity to separate liquids from solids.
2. Vacuum filtration: Uses vacuum pressure to accelerate filtration.
3. Membrane filtration: Uses semipermeable membranes to separate particles based on size.

Steps
1. Preparation: Prepare the extract-solvent mixture for filtration.
2. Filtration: Pass the mixture through a filter medium (e.g., paper, membrane).
3. Collection: Collect the filtered extract.

Importance
1. Removes impurities: Filtration removes solid particles and impurities.
2. Improves clarity: Filtration improves the clarity and appearance of the extract.
3. Enhances quality: Filtration ensures the quality of the final product.

Applications
1. Labor
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Applications
1. Laboratory settings: Filtration is used in laboratory-scale extractions.
2. Industrial settings: Filtration is used in large-scale commercial extractions.

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By using appropriate filtration techniques, the quality and purity of mango bark extracts can be ensured.

Concentration Process in Mango Bark Extraction

Overview

Concentration is a critical step in the extraction process, where the extracted compounds are concentrated to remove excess solvent and enhance the potency of the extract.

Methods

1. Evaporation: Uses heat and/or vacuum to evaporate the solvent.
2. Lyophilization (Freeze-drying): Removes solvent by freezing and then reducing pressure.
3. Rotary evaporation: Uses a rotating flask and vacuum to evaporate the solvent.

Importance

1. Enhances potency: Concentration increases the concentration of bioactive compounds.
2. Reduces volume: Concentration reduces the volume of the extract, making it easier to handle and store.
3. Improves stability: Concentration can improve the stability of the extract by reducing the risk of degradation.

Applications

1. Pharmaceuticals: Concentration is used to produce high-quality extracts for pharmaceutical applications.
2. Food and beverages: Concentration is used to produce flavorings, extracts, and other products.
3. Cosmetics: Concentration is used to produce high-quality extracts for cosmetic applications.

Considerations

1. Temperature control: Temperature control is crucial to prevent degradation of bioactive compounds.
2. Solvent removal: Complete removal of solvent is essential to ensure the quality and safety of the final product.

By using appropriate concentration techniques, high-quality mango bark extracts can be produced for various applications.

Purification in Mango Bark Extraction

Overview

Purification is a crucial step in the extraction process, where the concentrated extract is further refined to remove impurities and enhance its quality.

Methods

1. Chromatography: Separates compounds based on their interactions with a stationary phase.
2. Crystallization: Involves the formation of crystals from a solution.
3. Distillation: Separates compounds based on differences in boiling points.

Importance

1. Removes impurities: Purification removes impurities and contaminants.
2. Enhances quality: Purification improves the quality and potency of the extract.
3. Ensures safety: Purification ensures the safety of the final product.

Applications

1. Pharmaceuticals: Purification is critical for producing high-quality extracts for pharmaceutical applications.
2. Food and beverages: Purification is used to produce safe and high-quality flavorings and extracts.
3. Cosmetics: Purification is used to produce high-quality extracts for cosmetic applications.

Considerations

1. Method selection: Choosing the appropriate purification method depends on the specific compounds and desired level of purity.
2. Quality control: Quality control measures are essential to ensure the purity and quality of the final product.

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By using appropriate purification techniques, high-quality mango bark extracts can be produced for various applications.

Repeat Cycles in Mango Bark Extraction

Overview

Repeat cycles, also known as repeated extraction or multiple extraction, involve repeating the extraction process multiple times to maximize the yield of bioactive compounds.

Benefits

1. Increased yield: Repeat cycles can increase the overall yield of bioactive compounds.
2. Improved efficiency: Repeat cycles can improve the efficiency of the extraction process.
3. Enhanced quality: Repeat cycles can help ensure that the maximum amount of bioactive compounds is extracted.

Considerations

1. Solvent usage: Repeat cycles may require additional solvent, which can increase costs and environmental impact.
2. Time and labor: Repeat cycles can be time-consuming and labor-intensive.
3. Optimization: Optimization of extraction conditions and number of cycles is crucial to achieve maximum yield and efficiency.

Applications

1. Industrial-scale extraction: Repeat cycles are often used in industrial-scale extraction processes.
2. Research and development: Repeat cycles can be used in research and development to optimize extraction protocols.

By optimizing repeat cycles, the yield and quality of mango bark extracts can be maximized.

RESULT AND DISCUSSION

Prelimilary Phytochemical Investigation

Sr. No.	Test	Chloroform	Ethanol
1	Glycoside Test	+	-
2	Tannins Test	-	+
3	Salkowsai Test	+	-
4	Carbohydrate Test (Fehling Test)	+	-
5	Protein Test	+	-



CONCLUSIONS AND SUMMARY OF MANGO BARK STEM EXTRACTION

Conclusions

The extraction of mango bark stem has several conclusions based on recent studies.

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- Antimicrobial Activity: Mango plant extracts and formulations exhibit effective antimicrobial activity due to the synergy of various phytochemical constituents. Specifically, mangiferin isolated from ethanol extract of mango stem bark shows antibacterial activity against certain Gram-positive and Gram-negative bacteria.

- Antioxidant Properties: Both cold and hot water extracts of mango stem bark demonstrate potent antioxidant activity, with hot water extracts showing a marked increase in antioxidant ability.

- Anti-Diabetic Potential: Mango stem bark contains flavonoids and polyphenols that may contribute to its anti-diabetic activity, although more research is needed to fully understand its effects.

- Phytochemical Composition: Mango stem bark extracts contain bioactive phytochemicals, including mangiferin, quercetin, catechin, and epicatechin, which may be responsible for its medicinal properties.

Summary

Mango bark stem extraction is a promising area of research, with potential applications in:
- Medicinal Properties: Antibacterial, anti-inflammatory, analgesic, and antioxidant properties make mango bark stem extracts valuable for pharmaceutical and therapeutic uses.

- Pharmaceuticals: Development of new medicines or supplements leveraging the medicinal properties of mango bark stem extracts.

- Cosmetics: Antioxidant and anti-inflammatory properties may be beneficial in skincare products.
- Food Industry: Potential use as natural preservatives or additives.

Overall, the extraction of mango bark stem offers opportunities for sustainable utilization of mango tree byproducts, with potential economic benefits for the mango industry.

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