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EXPLORING THE THERAPEUTIC POTENTIAL: ANALYSIS OF PHYTOCHEMICALS AND BIOLOGICAL PROPERTIES OF BITTER GOURD (*MOMORDICA CHARANTIA. L*) FRUIT EXTRACTS

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ABSTRACT

Momordica charantia L., commonly known as bitter gourd, is a climbing plant indigenous to Asia, notably prevalent in India, and is identified by various vernacular names such as Karol, spiny gourd, or teasle gourd. Despite its longstanding use as a traditional remedy for diverse ailments, bitter gourd has also been part of dietary consumption owing to its nutritional richness, even though it is not a commonly utilized vegetable. Our study focuses on the fruit extracts of bitter gourd, with the primary objective of scrutinizing its chemical composition and assessing its potential health properties. The extraction process has unveiled a spectrum of beneficial compounds within bitter gourd, including flavonoids, alkaloids, and polyphenols. Laboratory tests have demonstrated significant antioxidant and anti-inflammatory properties in these extracts. Noteworthy is their potential efficacy in the management of diabetes, suggesting prospective applications in the treatment of this condition. This research contributes insights into the medicinal attributes of bitter gourd fruit, accentuating its significance as a source of bioactive compounds with potential therapeutic benefits. Furthermore, it underscores the imperative for continued investigation into the specific chemical constituents of bitter gourd, essential for researchers interested in unlocking the untapped therapeutic potential of the fruit. In addition to the examination of fruit extracts, our investigation extends to the study of methanolic and ethanolic extracts of bitter gourd, entailing an exploration of their chemical properties and the identification of active ingredients. This systematic exploration augments our comprehension of bitter gourd's broader therapeutic roles, laying the foundation for further research in this promising field.

INTRODUCTION

Throughout history, plants and herbal preparations have played a vital role in traditional medicine, offering remedies for a myriad of ailments. Over the last few decades, scientific research has substantiated many claims associated with the use of various plants in traditional medicine. One such plant that has garnered significant attention is *M. charantia* (MC), widely recognized in diverse traditional medicine systems for its therapeutic potential across a spectrum of conditions. The popularity of MC spans a multitude of applications in traditional medicine, where it has been historically employed to address ailments such as diabetes, abortion, anthelmintic needs, contraception, dysmenorrhea, eczema, emmenagogue, antimalarial purposes, galactagogue, gout, jaundice, abdominal pain, kidney stones, laxative effects, leprosy, leucorrhea, piles, pneumonia, psoriasis, purgative properties, rheumatism, fever, and scabies. The versatile applications of MC have drawn significant attention from researchers, prompting extensive investigations into its medicinal properties.

Modern research, employing advanced techniques, has validated the traditional use of MC in the management of diabetes and its complications, including nephropathy, cataracts, and insulin resistance. Furthermore, MC has demonstrated antibacterial and antiviral properties, showcasing potential efficacy against infections, including HIV. Additionally, the plant has exhibited anthelmintic and abortifacient activities. Interestingly, MC has shown promise in the treatment of peptic ulcers, with recent experimental studies highlighting its potential against Helicobacter pylori, a bacterium associated with such ulcers. Notably, recent research has expanded our understanding of MC's potential role in various cancers, including lymphoid leukemia, lymphoma, choriocarcinoma, melanoma, breast cancer, skin tumors, prostatic cancer, squamous carcinoma of the tongue and larynx, human bladder carcinomas, and Hodgkin's disease.

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While clinical studies on the use of MC in diabetes and cancer patients are still emerging, the available reports indicate promising results. The wealth of scientific evidence supporting the traditional uses of MC underscores its importance in modern medicine and suggests a potential avenue for the development of novel therapeutic interventions.

Bitter gourd (*Momordica charantia* L.) has long been recognized for its medicinal and nutritional significance, finding extensive use in traditional medicine and dietary practices, particularly in Asia and notably in India. The plant, known by various names such as Karol, spiny gourd, or teasle gourd, is a climbing vine with a rich history of being employed as a remedy for various ailments and concurrently valued as a vegetable due to its nutritional content. This botanical marvel has recently garnered increased attention for its potential therapeutic applications, prompting comprehensive scientific investigations into its bioactive constituents. The fruit extracts of bitter gourd have been a focus of scrutiny, revealing a diverse array of phytochemicals. These extracts have been found to contain significant concentrations of flavonoids, alkaloids, and polyphenols, each recognized for their potential health-promoting properties of particular interest are the observed antioxidant and anti-inflammatory properties exhibited by these bitter gourd extracts. Moreover, the potential efficacy of bitter gourd extracts in the management of diabetes has emerged as a noteworthy aspect of the investigation. Diabetes mellitus, a prevalent and challenging health concern globally, may benefit from the bioactive compounds found in bitter gourd, potentially influencing glucose metabolism and insulin sensitivity.

As scientific interest in bitter gourd grows, this exploration aims to provide a comprehensive overview of the phytochemical composition of bitter gourd fruit extracts, emphasizing their antioxidant and anti-inflammatory properties. Additionally, we delve into the potential implications of these properties in the context of diabetes management. This research contributes to the expanding body of knowledge surrounding bitter gourd's therapeutic potential, with implications for both traditional medicine and contemporary healthcare practices.

Phytochemical Studies

The substance comprises Lectins, proteins, triterpenes, and vitamins (Naik, 1951). The fruit is rich in vitamin C (Bhuiya, 1977). The fruit is abundant in ascorbic acid and contains iodine (Rao, 2001). The fruit furthermore contains alkaloids, flavonoids, glycosides, and amino acids, as stated by Kushwaha *et. al.* in 2005. *M. charantia* also includes an alkaloid, a fragrant extractive substance, and ash comprising 3 to 4 percent of its composition. Ash includes a small amount of manganese (Data, 2010).

The typical nutritional composition of *M. charantia* per 100 g of edible fruit includes 84.1% moisture, 7.7 g of carbohydrates, 3.1 g of protein, 3.1 g of fat, 3.0 g of fiber, and 1.1 g of minerals. In addition, it included trace amounts of vital vitamins such as ascorbic acid, carotene, thiamin, riboflavin, and niacin (Singh, 2006). The protein content in the leaves and dry weight of aerial plant parts were found to be greater in male defruited and monoecious plants compared to female defruited plants, as reported by Ghosh (2005). 6-methyl tritriacont-50on-28-of and 8-methyl entrant-3-ene were extracted from the fruit of *M. charantia*, together with the previously identified sterol pleuchiol. Momodicaursenol, a previously unidentified pentacyclic triterpene found in the seeds, has been identified as urs- 18 (19)-dien-3 betaol. Phytochemical analyses have shown small amounts of alkaloids and ascorbic acid in fruits. The compounds included in the substance are lectins, b-sitosterol, saponins, glycosides, triterpenes such as ursolic acid, hederagenin, oleanolic acid, aspiranosterol, stearic acid, gypsogenin, as well as two newly discovered aliphatic components (Ali and Srivastava, 1998, Sadyojatha and Vaidya, 1996, Ghosh *et al.*, 1981). Three triterpenes and two steroidal substances were identified from the dried root of *M. charantia*.

The compounds identified were alphaspinasterol octadecanonate (I), alphaspinasterol-3-O-beta-D-glucopyranoside (II), 3-O-beta D-glucuronopyranosyl gypsogenin (III), 3-O-beta-D-glucopyranosyl gypsogenin (IV), and 3-O-beta-D glucopyranosyl hederagenin (V). Constituent III was a novel compound. Fruits have been employed in the management of inflammation resulting from lizard excretion (Sastri, 1962) as well as mental and intestinal issues. The whole plant is renowned for its medicinal properties in treating ocular ailments, poisoning, and febrile conditions (Satyavathi, 1987).

The fruit pulps have been demonstrated to possess hypoglycemic, hepatoprotective, anti-inflammatory, analgesic, and antifeedant properties. The plant's leaves possess antihelminthic and aphrodisiac properties. Additionally, it is employed for treating tridosha, fever, pitta imbalances, jaundice, asthma, bronchitis, piles, hepatic impairments, mental digestive problems, bleeding piles, bowel afflictions, and urinary complaints. An ointment is made by combining the juice of the leaves with coconut, pepper, red sandalwood, and other ingredients. This ointment is then applied to the head to alleviate headaches. Topical application of leaf paste is administered to the skin and taken orally two to three times per day for the treatment of skin ailments. This study focuses on doing a qualitative phytochemical examination of Momordica dioica to determine the presence of tannins, phenols, saponins, alkaloids, flavonoids, anthraquinones, cardiac glycosides, carbohydrates, triterpenoids, and steroids.

MATERIALS AND METHODS

The experimental material, *M. charantia* fruits, was collected in October 2023 in and around the Government College for Women, Jagityal. To ensure the preservation of moisture during transportation to the laboratory, the collected plant material was placed in polyethylene bags. The plant extraction process involved thorough washing of the fruit in tap water, followed by separate shade drying in the open air. The dried fruit was mechanically ground to obtain a powder. Approximately 100 grams of each dried powder were soaked individually in 100 ml of various solvents, such as methanol, ethanol, chloroform, pet ether, and hot water, in conical flasks. The mixtures were then agitated on a rotary magnetic shaker for 72 hours.

After the three-day agitation period, the plant extracts underwent filtration using No. 42 Whatman filter paper individually. The concentrated extracts were carefully preserved in sterilized, air-tight, labeled bottles and stored in a refrigerator at 4°C until needed for further use.

To refine the extract, it underwent filtration under reduced pressure using a rotary flash evaporator. The resulting concentrated extracts were then subjected to preliminary phytochemical tests for identification. The methods used for these tests followed the procedures described by Mohan *et al.* (2015).

Table 1 Photochemical screening test of fruit extracts of *M. charantia* L.

Sl No	Chemical components	Methanol	Ethanol	Chloroform	Pet Ether	Water
1	Alkaloids	+	+	+	+	+
2	Terpenoids	+	+	+	+	+
3	Flavonoids	-	+	+	-	-
4	Anthraquinones	-	_	-	-	+
5	Tannins	+	_	-	-	-
6	Saponins	+	+	-	-	+
7	Glycosides	-	+	-	-	+
8	Reducing sugars	+	+	+	+	+
9	Steroids	+	+	+	+	+
10	Cardiac glycosides	+	+	+	+	+
11	Phenol	-	-	-	-	-

Test for identification of Alkaloids:

A test tube containing about 0.5 grams of methanol extract was diluted and mixed well with 10 ml of distilled water. The mixture was then dissolved in 20 ml of a diluted hydrochloric acid solution and cleared by filtering. The filtrate underwent testing using Drangendroff's and Mayer's reagents. The solution that underwent treatment exhibited the occurrence of precipitation, which manifested as a white or creamy color.

Test for identification of Terpenoids

To create a layer, 5 ml of the methanol extract was combined with 2 ml of chloroform and 2 ml of strong sulfuric acid. The presence of terpenoids was indicated by the interface having a reddish-brown tint.

Test for identification of Flavonoids

Approximately 0.5 grams of extract was added to a test tube containing 10 ml of ethyl acetate. The mixture was then boiled in boiling water for 1 minute. Subsequently, the concoction underwent filtration. Approximately 4 ml of the filtered substance was mixed with 1 ml of a 1% solution of aluminum chloride and allowed to incubate.

Test for identification of Anthraquinones and Glycosides

Bromine test*:* is used to identify anthraquinone glycosides in general. Its procedure is: to two ml of the extract add an equal volume or an excess of freshly prepared solution of bromine. Record the color.

Nitric acid test: is a specific identification of the anthraquinone glycosides. To 5 ml solution add 2 ml of concentrated nitric acid. Record the result.

Test for identification of Tannins

To extract five grams of the pulverized powder, ten ml of ammonical chloroform and five ml of chloroform were used. After the mixture was filtered, the filtrate was mixed with ten drops of sulphuric acid that had a concentration of 0.5 M. Tannins were found manifesting themselves as a white precipitate with a cream-like consistency.

Test for identification of Saponins:

A test tube was filled with about 0.5 grams of methanol extract, and then 5 millilitres of distilled water was added to the mixture. A vigorous shake was performed on the solution, and persistent foam was monitored. Following the vigorous shaking of the frothy mixture, which was then combined with three drops of olive oil, the mixture was examined to see whether an emulsion had formed.

Test for identification of Reducing sugars

Benedict's test is a chemical test that can be used to check for the presence of reducing sugars in each analyte. Therefore, simple 2 ml of extract with this test. The test is based on Benedict's reagent (also known as Benedict's solution), which is a complex mixture of sodium citrate, sodium carbonate, and the pentahydrate of copper (II) sulfate. When exposed to reducing sugars, the reactions undergone by Benedict's reagent result in the formation of a brick-red precipitate, which indicates a positive Benedict's test. An image detailing the changes in the color of Benedict's reagent (from clear blue to brick-red) that are triggered by exposure to reducing sugars is provided.

Test for identification of Steroids

After placing approximately 0.5 grams of methanol extract in a test tube, 2 ml of acetic anhydride was added to it. Then, 2 ml of sulfuric acid was poured by the sides of the test tube. The color of the solution was seen to change to either blue-green or violet color

Test for identification of Cardiac glycosides:

An extract solution of glycosides is treated with a small amount of Kedde reagent (Mix equal volumes of a 2% solution of 3, 5 dinitrobenzoic acid in menthol and a 7.5% aqueous solution of KOH). Development of a blue or violet colour that faded out in l to 2 hrs shows it presence of cardinoloids.

Test for identification of Phenols

A test tube containing about 0.5 grams of extract was combined with 100 ml of distilled water and subjected to gentle heating. Subsequently, 2 ml of ferric chloride solution was introduced and monitored for the occurrence of a green or blue color.

RESULTS AND DISCUSSION

The current research makes a significant contribution to the body of knowledge concerning the bioactive chemicals found in *M. charantia*, a plant that is utilized in a wide range of traditional medical practices. The qualitative analysis of *M. charantia* reveals that extracts of the plant in methanol, ethanol, chloroform, pet ether, and water contain bioactive compounds. These compounds include phenols, saponins, alkaloids, flavonoids, anthraquinones, cardiac glycosides, carbohydrates, triterpenoids, and steroids, except for tannins and anthraquinones (Table-1 Fig-1). A wild link between *M. charantia* and other organisms has been established by the current investigation. This relationship may be further investigated to determine the comparative biological activity for additional confirmation. When it comes to biological activity, steroids and flavonoids are both well-known. These substances are known to be harmful to microorganisms and should be avoided. It is believed that the relative toxicity of the phenol group to microorganisms is related

to the location of the hydroxyl groups on the phenol group as well as the quantity of hydroxyl groups. It is likely that enzyme inhibition by oxidized chemicals, which may occur because of a reaction with sulfhydryl groups or through a more general interaction with proteins, is the mechanism that is responsible for the toxicity of phenolic compounds to microorganisms. Similarly, the phytochemical examination of the extracts in our research revealed the presence of bioactive substances such as steroids, fatty acids, saponin glycosides, and triterpenes. The presence of these chemicals may be the cause of the antibacterial action described above.

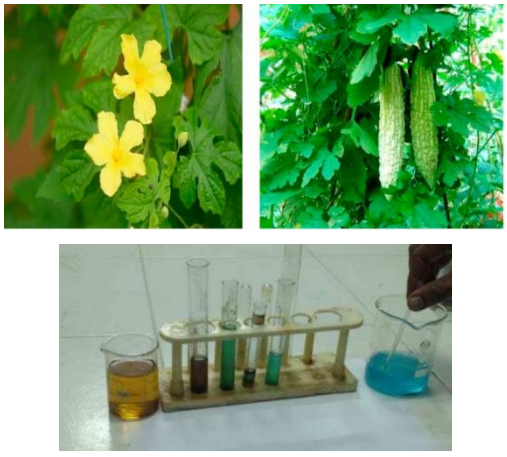

Fig 1 Studies on Phytochemical and Biological Activities of *M. charantia*: a) Showing the plant with flowers and fruits b): Showing the results of Phytochemical Analysis is kept in the test tubes with stand.

Biological activities

The plant Momordica dioica, commonly known for its various medicinal uses, has been traditionally employed in the treatment of eye diseases, fever, snake bites, and inflammation caused by lizard bites. Additionally, it is recognized as a remedy for diabetes. During an investigation into the spermatogenic properties of the ethanolic extract derived from the fruit of M. dioica on animals, behavioral observations unexpectedly revealed sedative activity associated with the extract.

Limited research is available concerning the pharmacological activities of this plant. Comparable sedative and anxiolytic activities have been reported in other plants such as Passiflora actinia, Aloysia polystachya, *Euphorbia hirta*, Kigelia Africana, and Coriandrum sativum. Notably, *M. charantia* possesses essential nutrient compounds vital for bodily functions, including Calcium (0.5 mg/g), Sodium (1.5 mg/g), Potassium (8.3 mg/g), Iron (0.14 mg/g), Zinc (1.34 mg/g), Protein (19.38%), Fat (4.7%), Total phenolic compound (3.7 mg/g), Phytic acid (2.8 mg/g), and an ash value of 6.7%. Studies by Jain et al. have highlighted the antioxidant and hepatoprotective activities of ethanolic and aqueous extracts of *M. charantia*. The ethanolic extract exhibited superior hepatoprotective effects, and both extracts displayed positive antioxidant and free radical scavenging activities, possibly attributed to the presence of flavonoids. Furthermore, the plant's roots have demonstrated antiallergic activity in alcoholic extracts. The vegetable seed oil extracted from Small bitter gourd (*M. charantia*) has been evaluated as a grain protectant against *Callosobruchus chinensis* in stored legume-pulse grains, as reported by (Mishra *et al.* 2002), (Thirupathi *et al.* 2000) also reported a protective effect of Momordica dioica against hepatic damage caused by carbon tetrachloride in rats.

M. charantia exhibits diverse medicinal properties, ranging from antiallergic and hepatoprotective activities to antioxidant effects, making it a valuable subject for further research and exploration in the field of natural medicine.

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