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EVALUATION OF ANALGESIC AND ANTI-ULCERACTIVITY BY HERBAL PLANT EXTRACTS

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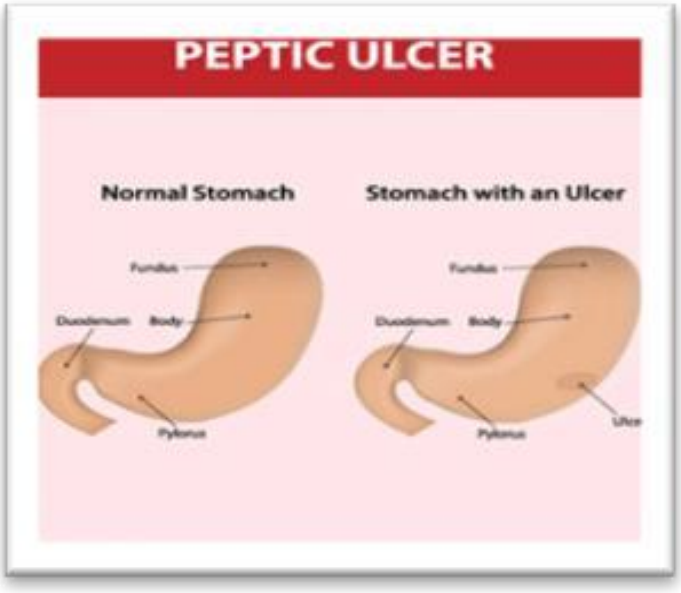
ABSTRACT

The unrivalled substitute to synthetic medicines, available, for relieving, pain andulcers, are found in natural products of plants. They are known to exhibit a varietyof activities. The objective of the present study is screening of *M. piperita & M. Charantia* for their analgesic and anti-ulcer activity. The extract of leaves of *M. piperita & M. Charantia* were tested in the Eddy's hot plate method and in pyloric ligation. The treatment of *M. piperita & M. Charantia* at dose 200mg/kg for analgesic activity reduced the pain induced by the Hot Plate method and the potency was found to be equivalent as compared to standard drug Aspirin. The treatment of *M. piperita & M. Charantia* at dose 500mg/kg reduced the formationof ulcers induced by the pyloric ligation method and the potency was found to beequivalent to the standard drug Ranitidine. The presence of alkaloids, tannins andflavonoids may be accountable for the analgesic and anti-ulcerative activity of *M.piperita and M. charantia.* The results further suggest that *M. piperita and M. charantia* possess analgesic and anti-ulcer activities.

INTRODUCTION TO ULCER

Injury to the oral mucosa may result in a localized defect of the surface in which the covering epithelium is destroyed leaving an inflammation area of exposed connective tissue. Such defects are called ulcers or erosions (term commonly used for superficial ulcer). This may either follow molecular death of surfaces epithelium or its traumatic removal. Ulceration is themost common lesions of oral mucosa and is the manifestation for many local and genetic disorders.

Peptic ulcer is a condition where the first part of the small intestine has painful sores or ulcers develop in the lining of stomach. Generally, a thick layer of mucus protects the stomach liningfrom the effect of its digestive juices. But many things can reduce this protection layer allowingstomach acid to damage the tissue.



Definition of ulcer

Ulcer is a break in skin or mucous membrane with loss of surface tissue , disintegration and necrosis of epithelial tissue and often pus (or) it is something that festers and corrupts like anopen sore.

Etiology of ulcersymptoms

- Burning stomach pain
- Feeling of fullness, bloating or belching
- Intolerance to fatty foods
- Heartburn
- Nausea

The most common peptic ulcer symptom is burning stomach pain. Stomach acid makes the pain worse, as does having an empty stomach. The pain can often be relieved by eating certain foods that buffer stomach acid or by taking an acid-reducing medication, but then it may come back. The pain may be worse between meals and at night.

Many people with peptic ulcers don't even have symptoms.

Less often, ulcers may cause severe signs or symptoms such as:

- Vomiting or vomiting blood — which may appear red or black
- Dark blood in stools, or stools that are black or tarry
- Trouble breathing
- Feeling faint
- Nausea or vomiting
- Unexplained weight loss
- Appetite changes

Causes

Peptic ulcers occur when acid in the digestive tract eats away at the inner surface of the stomach or small intestine. The acid can create a painful open sore that may bleed.

Your digestive tract is coated with a mucous layer that normally protects against acid. But if the amount of acid is increased or the amount of mucus is decreased, you could develop an ulcer.

Risk factors

In addition to having risks related to taking NSAIDs, you may have an increased risk ofpeptic ulcers if you:

- Smoke. Smoking may increase the risk of peptic ulcers in people who are infected with H.pylori.
- Drink alcohol. Alcohol can irritate and erode the mucous lining of your stomach, and it increases the amount of stomach acid that's produced.
- Have untreated stress.
- Eat spicy foods.

Alone, these factors do not cause ulcers, but they can make ulcers worse and more difficult toheal.

Introduction to analgesics

Analgesics are a drug which relieves pain without altering sensory awareness and consciousness or blocking the conduction of nerve impulses. Analgesics are also known as anti-inflammatory drugs, due to its action to reduce local inflammatory responses.

Analgesics are drugs that eliminate or alleviate the feeling of pain that accompanies many pathologic conditions. It is difficult to list all the situations in which it is necessary to use analgesics, for example, muscle aches and headaches, and where there is no possibility of becoming addicted. Analgesics are divided into two groups such as opioids, which predominantly influence the central nervous system (CNS), and Nonopioids, which act predominantly on the peripheral nervous system. Opioids are further subdivided into three large subgroups according to their action on opioids receptors such as agonists, mixed agonists/antagonists, and antagonists. Opioid agonists act first and foremost on μ-receptors.

Reaction of agonists with opioid μ-receptors leads to an increase in the flow of potassium ions from the cell, simultaneously making it difficult for calcium ions to flow into the cell, which makes neurons less excitable. Opioid antagonists on the other hand also bind to opioid receptors but do not activate them. These compounds are not used for analgesia. Their therapeutic value is in relieving side effects that result from either absolute of relativeoverdoses or intolerance of drugs by patients, and also in treating cases of Opioid dependency.

Uses of analgesics

Analgesics are used to relieve pain and inflammation. For example:

- After surgery.
- Due to injury, such as a fractured bone.
- For acute (sudden, short-term) pain, such as a twisted ankle or headache.

Mechanism of pain killers

There are two major groups of analgesics: anti-inflammatory analgesics and opioids. Anti-inflammatory drugs work by reducing inflammation (swelling) at the site of the pain.

Examples include

- Acetaminophen.
- Aspirin.
- COX inhibitors.
- Nonsteroidal anti-inflammatory drugs (NSAIDs), such as ibuprofen and naproxen.

Mechanism of Action of Analgesics

Local tissue injury releases prostaglandins. Prostaglandins have two major actions:

1. Sensitize pain receptors and lower the threshold for painful stimuli
2. Intensify the activation of the nerve endings by other

inflammatory mediators such as bradykinin, serotonin, and histamine

NSAIDs work by inhibiting the production of prostaglandins by inhibiting two types of cyclooxygenase enzymes

- COX-1
- COX-2

COX-1 is present in all cells, while COX-2 is induced in the presence of inflammation.

Traditional NSAIDs inhibit both COX enzymes and thus relieve pain, reduce fever and inflammation. Selective COX-2 inhibitors (coxibs) inhibit the COX-2 enzyme primarily andthus exerts anti-inflammatory effects.

LITERATURE REVIEWS

Literature reviews for Mentha Piperita

1. **Aishwarya Balakrishnan(2015):** She made a study on the effect of peppermint in the treatment of non-obstructive dyspepsia and also showed the anti-emetic effect of peppermint oil by performing the experiments on Wistar rats.
2. **Mohammad Mahdi Zangeneh Saman Salman Akram Zangeneh (2019):** In this study 60 Wistar rats were used and were divided into 6 sub-groups. 3 of these sub groups were received M. Piperita at different concentrations. The other 3 were classified as negative healthy group, untreated negative control receiving distilled water and one test group receiving drug Omeprazole. After 14 days the ulcers were formed by ethanol and 4 hours after administration, the rats were dissected and blood, stomach, and duodenal samples were collected. These samples were further tested andit was proved that M. Piperita decreases the levels of constituents which cause ulcer inwhile compared to control group.
3. **G. Vimala, F. Gricilda Shoba (2014):** The experimental parameters used for antiulcer activity were cold resistant stress – induced ulcer model , Diclofenac – induced ulcer model in rats, (HCl-ethanol) – induced ulcer in mice and water immersion stress – induced ulcer in rats. This article reviews drugs derived from medicinal plants more commonly used in the world for peptic ulcer and , if reported,the antiulcer activity . With the help of this study, the researchers have given a literature review about the effect of M. Piperita in the treatment of ulcer by differentinducing sources.
4. **El-Sayed S. Abdel – Hameed; Mahmood S. Salman (2017):** They studied the effect of extraction methods ; hydro distillation, and solvent free microwave extraction of Mentha piperita L . Taif, on the yield and the chemical composition of their essential oils were investigated. Furthermore, the oils were in Vitro investigated as antimicrobial and anticancer agents.
5. **Ganesan Mahendran, Laiq -Ur Rahman (2020):** The scientific studies provide awareness on the use of M. piperita for biological effects such as antioxidant, antimicrobial, anticancer, anti-diabetic activity been ascribed. A wide spectrum of bioactive phytochemicals such as flavonoids, phenolic lignans and essential oils are expected to be responsible for the aroma effects. In this they provided an extensive overview of the traditional medicine, phytochemical and multiple biological activitiesof this peppermint.

Acetaminophen, or paracetamol, acts by a different mechanism by inhibiting the COX-3 enzyme in the brain. It produces analgesia by altering the pain perception in the brain but hasa little anti-inflammatory effect.

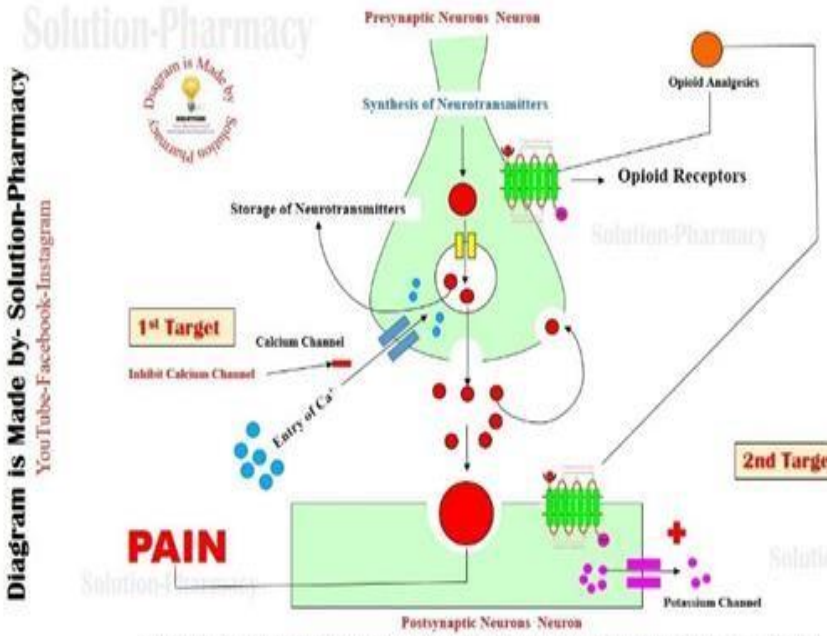


Diagram- Schematic Diagram Explaining MOA of Opioid Analgesic (Reference- Lippincott)

Analgesic opioids (also called narcotics) work by changing the brain's perception of pain. Anopioid can be any drug, natural or manmade. Many are similar to morphine, but newer, unrelated opioids have been created in the laboratory, too. Examples include:

- Codeine.
- Fentanyl.
- Hydrocodone.
- Meperidine.
- Methadone.
- Naloxone or naltrexone.
- Oxycodone.

Pain relievers are available in many forms, including

- Films you place under the tongue to dissolve.
- Liquid you inject into your body with a syringe (needle).
- Liquid you swallow.
- Nasal spray that goes up the nose.
- Patches you place on the skin.
- Pills, tablets or capsules you swallow.

Risks of using pain killers

Anti-inflammatory analgesics are generally safe. But they can cause side effects and complications, if you use them too often, for too long or in very large doses:

- Damage to internal organs, such as the liver or kidneys.
- Diarrhea or constipation.
- Heart problems.
- Hypersensitivity response, which is like an allergic reaction.
- Nausea, upset stomach or heartburn.
- Ringing in the ears, or even deafness.
- Stomach ulcers.
- Trouble forming clots in the blood, which can lead to excessive bleeding.

Opioid analgesics can cause many of the same side effects and complications. Opioids are tightly controlled because they can cause physical dependence and are prone to abuse. The problem, which doctors now call substance abuse disorder, can be mild, moderate or severe, so it isn't always recognized right away.

Some tell-tale signs a problem may be developing include

- Often taking the medication in larger amounts than were intended.
- Unsuccessful efforts to cut down.
- Repeating failure to fulfill major obligations at work, school or home.
- Continued use despite having persistent problems.
- Giving up important social activities.
- Using even in dangerous situations (driving).
- tolerance.
- Withdrawal.
- For aches and pains like menstrual cramps or muscle soreness.
- For chronic painful conditions such as arthritis, cancer or back pain.

Types of Analgesics

Analgesics include opioid and opiate substances and nonopioid medications, which may be prescription or over the counter drugs. Many nonopioid and nonopioid-opioid preparations also work as antipyretics and anti-inflammatory agents for a desirable therapeutic effect.

Opioid and opiate medication are strong analgesics capable of reducing pain of any origin. Nonopioids may require a prescription or may be bought. Non Opioids may also be used in co analgesia or as adjuvant therapy. Coanalgesic such as codeine and acetaminophen are most often used for chronic pain but may be used for acute pain that requires opioid use. Adjacent medications such as diazepam, given with opioids are not true analgesics but are used with analgesics to potentiate pain relief and reduce pseudoaddiction.

There are three types of OTC analgesics. One is salicylates, which includes aspirin, one of the most common analgesics. While aspirin is generally safe for most adults, use by children under 16 has been linked to Raye's syndrome, a rare liver disorder. Long-term use in adults can cause damage to the stomach lining, causing abdominal pain and bleeding. It can also prevent the blood to clot, making it useful for patients suffering from heart attacks and stroke.

Opioids and Opiates

- Opioids and opiates are derivatives of opium or opium like chemicals that produce similar results to elevate pain thresholds and alter pain perception.
- Opiates and opioids have antitussive effects and may cause respiratory depression especiallyin the elderly.
- Opioids and opiates are used for acute pain of moderate to severe intensity and in terminalillnesses.
- Addiction and psychological dependence may occur with use of strong analgesics forchronic pain. These analgesics are effective and safe for short term usage.
- Around the clock administration of opioids and opiates is used for severe, acute pain and chronic pain of terminal illnesses.

Uses of Analgesics

Opioids and Opiates are used to treat acute pain of moderate to severe intensity, alter the perception of pain by mimicking endorphins to block neurotransmission of painful impulsesand increase the pain threshold.

The World Health Organization (WHO) has described a three step analgesic ladder in pharmacologic treatment of pain, using adjuvant analgesics in conjunction with opioids andopiates with each type of pain.

1. Mild pain – Use acetaminophen, aspirin or other nonsteroidal anti inflammatory drugs around the

clock.

2. Moderate pain – If pain persists or increases a mild opioid such as codeine or hydrocodone.

Severe pain – If pain persists or if it is moderate to severe at outset, give a strong opioid or opiate such as morphine, fentanyl or meperidine. A non opioid medication may also be continued to assist with pain control or discontinued.

Mentha Piperita

PLANT PROFILE

Fig. no 5: Mentha Piperita.

Systemic position of the plant
Kingdom: Plantae
Plantae division: Tracheophytes
Order: Lamiales
Family: Lamiaceae
Genus: Mentha
Species: Mentha x piperita

PLANT INFORMATION
Scientific name: *Mentha piperita*
Synonyms: Pudina, mint, peppermint etc.
Biological source: Mint consists of the dried leaves and flowering tops of Mentha piperitaand Mentha spicata.
Family: Lamiaceae
Geographical source: Mentha piperita is indigenous to Europe and the Middle East.
Appearance: Peppermint is a fast-growing herbaceous plant with square stems that can reach a height of 30-90cm. It has dark green leaves that are oblong or lance shaped, with toothed edges. The leaves are arranged oppositely on the stem and emit a strong aroma when crushed.

Growing conditions: Peppermint thrives in cool, moist environment and prefers partial shade to full sun. It requires well drained soil with a pH of 6.0-7.5.

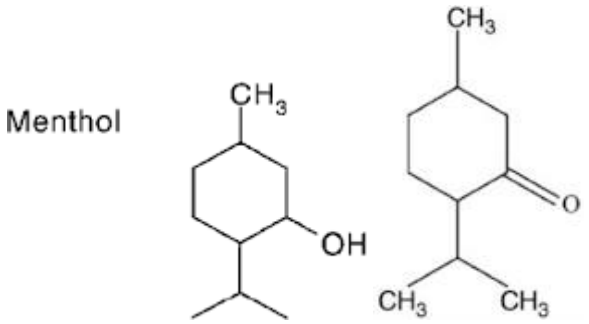
Propagation: Peppermint can be propagated from seeds, although it is more commonly propagated through vegetative means such as stem cutting or root divisions. Stem cuttings taken from healthy plants can be rooted directly in water or in soil.

Cultivation: While cultivating peppermint, adequate spacing is recommended as they tend to spread vigorously through underground runners called stolon's.

Chemical constituents
Mentha piperita is a popular herb that is used for its medicinal and culinary properties. The essential oil derived from peppermint consists of several chemical constituents that contributeto its characteristic aroma and therapeutic effects.

The primary constituents found in Mentha piperita are

- Menthol: Menthol is a major constituent of peppermint oil, constituting of 30-55% of the oil, it provides a cooling sensation and if often used topically for its analgesic properties. Menthol also has spasmodic effects on the smooth muscles of the gastrointestinal tract.
- Menthone: Menthone is a significant component of peppermint oil usually present at 10-30%. It contributes to the minty aroma of the oil and possesses antimicrobial properties.



Menthol and menthone structures

- Methyl acetate: Methyl acetate is a minor constituent of peppermint oil, typically found in 3-10%. It has a fruity aroma and is used in perfumes and flavorings.

1,8-Cineole (Eucalyptol): Peppermint oil contains small amounts of 1,8-Cineole ranging from 2-10%. This compound is also found in eucalyptus oil and contributes to its characteristic strength. This has expectorant and mucolytic properties and it is often used for respiratory conditions

There are three types of OTC analgesics. One is salicylates, which includes aspirin, one of the most common analgesics. While aspirin is generally safe for most adults, use by children under 16 has been linked to Raye's syndrome, a rare liver disorder. Long-term use in adults can cause damage to the stomach lining, causing abdominal pain and bleeding. It can also prevent the blood to clot, making it useful for patients suffering from heart attacks and stroke.

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4. Moderate pain – If pain persists or increases a mild opioid such as codeine or hydrocodone.

Severe pain – If pain persists or if it is moderate to severe at outset, give a strong opioid or opiate such as morphine, fentanyl or meperidine. A non opioid medication may also be continued to assist with pain control or discontinued

Medicinal Uses

Apart from being used in traditional cooking purposes, Mentha piperita is also known to have major medicinal uses. Some of which are as follows:

1) **DIGESTIVE AID:** Peppermint has been used for centuries to support digestive health. It can help relieve symptoms of indigestion such as bloating, gas, and stomach discomfort. Peppermint oil is often used to alleviate irritable bowel syndrome (IBS) symptoms, including abdominal pain and spasms.
2) **HEADACHE AND MIGRAINE RELIEF:** Peppermint oil can be applied topicallyor inhaled as a steam and can help relief tension headaches and migraines. It has a cooling and soothing effect that can reduce pain and promotes relaxation.
3) **RESPIRATORY SUPPORT:** Peppermint has expectorant properties that can help alleviate congestion and promote easier breathing. It is commonly used to relieve symptoms of common cold, sinusitis, and bronchitis.
4) **PAIN RELIEF:** Topical application of peppermint oil can provide relief from muscle pain, joint pain, and soreness. It has a numbing effect and can help reduceinflammation.
5) **ORAL HEALTH:** Peppermint oil is often used in oral care products like toothpaste and mouthwash due to its antibacterial properties. It can help freshen breath, prevent tooth decay, and soothe gum inflammation.
6) **NAUSEA AND MOTION SICKNESS:** Peppermint has been used to relieve nausea and vomiting for centuries. It can be consumed as a tea or inhaled as an essential oil to help alleviate symptoms of motion sickness and morning sickness.
7) **MENTAL CLARITY AND ALERTNESS:** The aroma of peppermint has stimulating effects of the mind and help improve focus, concentration and mental clarity. It is often used in aromatherapy and as a natural remedy for mental fatigue.
8) **SKIN IRRITATIONS:** Peppermint oil has cooling and soothing properties that canhelp relieve itchiness and irritation caused by insect bites, rashes and other skin diseases.

Momordica charantia

Fig. no 6: Momordica charantia.

Systemic position of the plant
Kingdom: Plantae
Plantae division: Tracheophytes
Order: Cucurbitales
Family: Cucurbitaeceae
Genus: Momodica
Species: M. Charantia

PLANT INFORMATION
Synonym: Balsam apple, Balsam pear, Karela, Bitter Goud, Ampayala.

Biological source: It is a topical vine belonging to the genus Momordica.

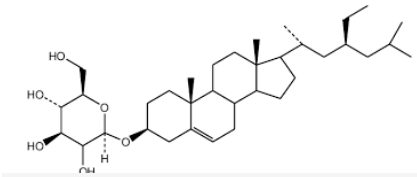
Geographical source: The plant is cultivated medicinally and as a vegetable plant in India, China and South East Asia.

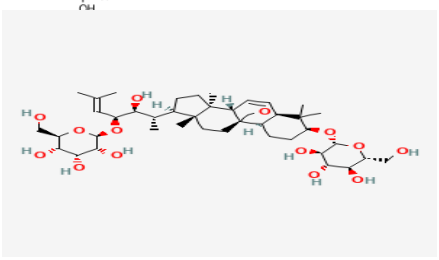
Appearance: Momordica Charantia is a perennial vine that grows vigorously, reaching lengths of up to 5m. The leaves are palmately lobed and have a textured surface with prominent veins. They can vary in shape and size but are generally 5-10cm wide. The flowers are yellow and bell-shaped, usually solitary or in small clusters. They have both male and reproductive parts.

Distribution and habitat: Bitter melon is native to Indian subcontinent, but now is cultivated and naturalized in many tropical and subtropical regions worldwide. It thrives in warm and humid climates and can be found growing in a variety of habitats, including fields, gardens and forest edges.

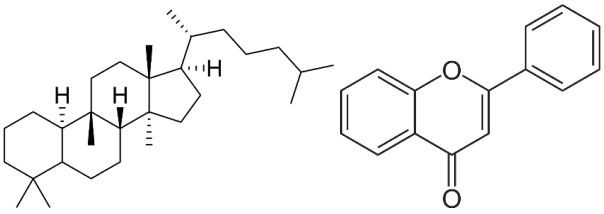
Chemical constituents
The chemical constituents of Momordica Charantia can vary depending on factors such as variety, growing conditions, and maturity of the fruit. Major constituents are as follow:

1) **CHARANTIN:** Charantin is a mixture of steroidal saponins, including charantin I and charantin II. It is one of the key bioactive compounds in bitter gourd and is known for its potential hypoglycemic (blood sugar lowering) effects.
2) **MOMORDICOSIDES:** Momordicosides are a group of triterpenoid saponins found in bitter gourd. They are responsible for the bitter taste of the fruit and possess various pharmacological activities, including antidiabetic, anticancer, antiulcer and modulatory effects.

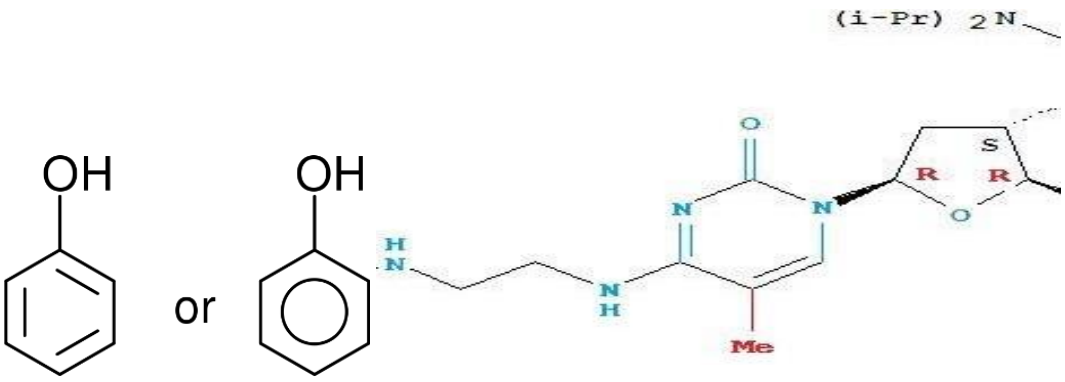




3) **CUCURBITANE-TYPE TRITERPENIODS:** Bitter gourd consists several cucurbitane-type triterpenoids, such as momordicin, momoordenol and momordicosin. These compounds contribute to the medicinal properties of bitter gourd including antidiabetic, anti-viral, antiulcer and anti-inflammatory activities.
4) **FLAVANOIDS:** Bitter gourd contains flavonoids like quercetin, kaempferol, and luteolin. These compounds possess antioxidant properties and may contribute to overall health benefits of bitter gourd.



5) **PHENOLIC COMPUNDS:** Bitter gourd is a rich source of phenolic compounds, including catechins, epicatechins and gallic acid. These compounds have antioxidantsand anti-inflammatory effects.
6) **LECTINS:** Bitter gourd contains lectins, which are carbohydrate-binding proteins. Lectins have been studied for their potential antiviral, immunomodulatory and anticancer activities.



7) **VITAMINS & MINERALS:** Bitter gourd is a good source of various vitamins, including vitamin C, vitamin A, and vitamin E. It also contains minerals such aspotassium, magnesium and calcium.

Traditional uses

Momordica charantia has a long history of traditional use in various cultures for its medicinal properties. Some of them are as follows:

1) **MANAGEMENT OF DIABETES:** Bitter gourd has been traditionally used to help manage diabetes. It is believed to have hypoglycemic properties, meaning it can help lower blood sugar levels. Various components present in bitter gourd are thought to contribute to antidiabetic effects.
2) **DIGESTIVE AID:** Bitter gourd has been used as a digestive aid in traditional medicine. It is believed to stimulate the secretion of digestive enzymes, promotehealthy digestion and relieve constipation.
3) **ANTIPARASITIC PROPERTIES:** Bitter gourd has been used traditionally to treat parasitic infections. It is believed to possess antiparasitic properties and has been used to expel intestinal worms.

MATERIALS AND METHODS

MATERIALS

Apparatus

- Conical flask
- Blender
- Measuring cylinders
- IP syringes
- Oral syringes
- Oral gavages
- Clinical sutures

Equipment's

- Eddy's hot plate
- TLC chamber

Standard drugs

- Aspirin
- Rantidine

Animals

- Wistar Rats (Female) collected from Jeeva life sciences.

PLANT COLLECTION

M. piperita and *M. charantia* were collected from local dhulapally area nearkompally in Hyderabad.

METHODS

Preparation of *M. charantia* powder

The vegetable bitter gourd was collected and washed with distilled water. Then it was chopped carefully into fine pieces and the seeds were removed. It was kept under sun drying for 2 daysand then was finely powdered by a blender and sieved through sieve number 20.

Preparation of mentha piperita powder

Mentha piperita leaves were collected and were washed with distilled water. After that, the leaves were separated from stem and were air dried for 2 days. Then they were finely grinded into a powder with the help of a blender and were sieved to get a fine powder with the help of sieve number 20.

Extraction by using soxhlation- Solvent extraction is the most widely used method. The extraction of natural products progresses through the following stages:

(1) The solvent penetrates into the solid matrix
(2) The solute dissolves in the solvents
(3) The solute is diffused out of the solid matrix
(4) The extracted solutes are collected

3PHYTOCHEMICAL SCREENING

Table 4.1: Procedure for phytochemical screening.

S.No	NAME OF THE TEST	PROCEDURE	OBSERVATION
1.	**Test for alkaloids**		
a)	Mayer's test	Sample +Mayers reagent	Cream ppt/white ppt
b)	Wagner's test	Sample+ Wagner's reagent	Reddish brown ppt
c)	Dragandroff test	Sample+ Dragandroff reagent	Orange brown ppt
d)	Hager's test	Sample+ Hager's reagent	Yellow colour ppt
2.	**Test for Glycosides**		
a)	Killer-killani test	Sample+GAA+$FeCl_3$+conc.H_2SO_4	Reddish-brown colourappears at junction.
b)	Legal's test	Sample+1ml Pyridine+1mlSodium nitroprusside.	Pink or red colour.
c)	Baljeet's test	Sample+ Sodium picrate	Yellow colour
3.	**Test for Phenolics**		
a)	Ferric chloride test	Sample+5%$FeCl_3$	Bluish Black colour
b)	Lead acetate test	Sample+ lead acetate	White ppt
4.	**Test for Flavonoids**		
a)	Shinoda test	Powder sample + 5ml ethanol +Mg turnings	Orange, pink, red orpurple
5.	**Test for proteins:**		
a)	Biuret test	Sample+ Biuret reagent	Violet or pale pink colour
b)	Millon's test	Sample+ Millon's reagent	White ppt, warm ppt turnsto brick red colour
6.	**Test for carbohydrates**		
a)	Fehling's test:	Sample+ Fehling's A and B+Heat	Red colour ppt

PROCEDURE FOR PYLORUS LIGATION METHOD

- Wistar rats of weight 150 – 200 grams were taken and made into four groups of six animals in each group.
- Group 1 was control group and group 2 and group 3 are test groups then group 4 wasstandard group.
- Then after grouping animals were kept for fasting for 24 hours.
- Then under anaesthesia, one inch abdominal midline incision is given below the xiphoid process, pylorus is ligated without damaging the blood supply and then abdominal wall closed with sutures.
- Then control group was not administered with any drug, group 2 animals were administered with ethanolic extract of Mentha piperita quantity of o.5 grams in 50 ml i.e; 1ml of test sample is administered to each rat.
- Then group 3 animals were administered with ethanolic extract of Momordica charantia quantity of 0.5 g in 50 ml i.e, 1ml of test sample is administered to each rat.
- Then standard group animals were administered with standard drug solution ofRanitidine quantity of 20mg\kg.
- Animals after drug administration were kept aside for 17-19 hours and then they aresacrificed, stomach is dissected out and opened along greater curvature.
- Contents of stomach were drained into a graduated centrifuge and acidity isdetermined , then ulcer score index is calculated.

PROCEDURE OF HOTPLATE METHOD

- Wistar rats of weight 150 – 200 grams were taken.
- Then they were separated into four groups named Group 1- control group, Group 2 –test group , Group 3 – test group , Group 4 – standard group.
- Then, control group is not administered with any drug whereas , group 2 – animals were administered with 150 mg \kg of test drug sample i.e ; Ethanolic extract of Momordica . charantia.
- Group 3 animals are administered with 300mg \kg of test drug sample i.e ethanolic extract Momordica charantia.
- Then finally group 4 animals were administered with the 150 mg\kg of aspirinstandard drug solution.
- Then the basal reaction time is observed and recorded by placing the control group animals in the hot plate instrument which is maintained at a temperature of 50 degree Celsius. Then record the time of jumping or licking of paws.
- Then group 2 animals and group 3 animals which are administered with test drug sample and group 4 animals administered with standard drug are placed in hotplate instrument and recorded the time of jumping or licking of paws at the time intervalsof 15 mins, 30 mins, 60 mins, and 90 mins and 120 mins.
- Then compare and calculate the percentage increase

in reaction time at each interval.

Fig. no. 7: Eddy's hot plate.

Group Ⅰ – Control

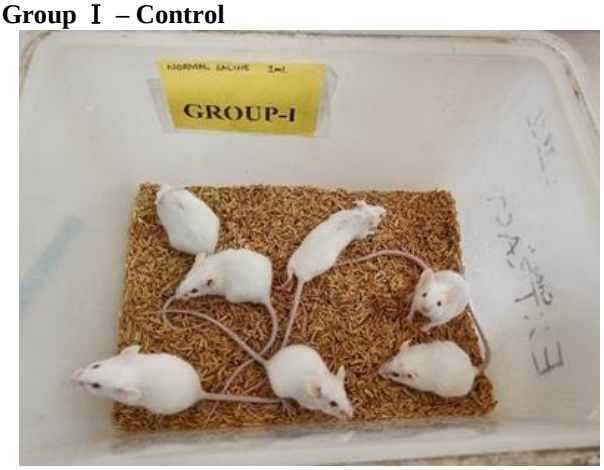


Fig no 8: Control group of animals for analgesic activity.

Group Ⅱ

Fig. no: 9: Standard group II of animals for analgesic activity.

Group Ⅲ

Fig. no: 10 Test 1 Group of Animals (M. Piperita) for Analgesic Activity.

GROUP IV

Fig. no: 11 Test group 2 (M. charantia) for analgesic activity.

RESULTS

The extract of *Mentha piperita* was done and the obtained extract was collected and the colorwas found to be green.

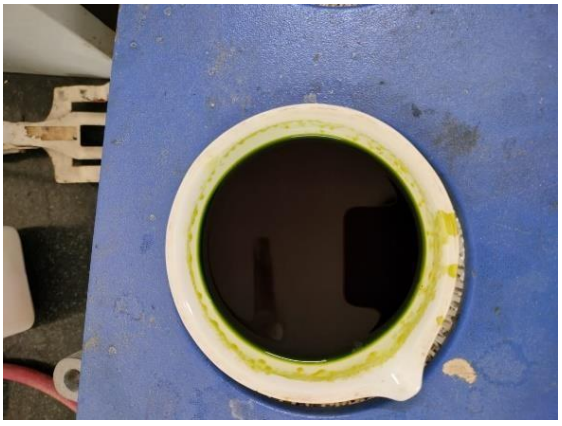


Fig. no. 12: Extract of *M. Piperita.*

The extract of *Momordica charantia* was done and the obtained extract was collected and thecolor was found to be **brown.**

Fig. no 13: Extract of *Momordica charantia.*

Phytochemical Screening

Preliminary phytochemical screening of *M. Piperita* revealed the presence of alkaloids, flavonoids, steroids, terpenoids, phenols, protein and carbohydrates Table 1.

Acute toxicity studies of the alcoholic extracts *M, piperita* did not show any toxicity up to 2g/ kg body weight. Since there was no mortality of animals at high dose, 200mg/kg dose of the extract was taken for the evaluation of analgesic activity.

Table 1: Results of phytochemical screening of *M. Piperita.*

S no	Test	*M. piperita*
1	Carbohydrates	+
2	Alkaloids	+
3	Steroids	+
4	Flavonoids	+
5	Tannins	+
6	Proteins	+

Preliminary phytochemical screening of the extract of *M. Charantia* indicated the presence ofalkaloids, flavonoids, steroids, saponins and carbohydrates. Acute toxicity studies did not show any effect on the animals up to 2g/kg. Since there was no mortality found of the animals at a high dose, hence 200mg/kg dose was taken to evaluate the analgesic activity.

Table 2: Phytochemical screening of *M. Charantia.*

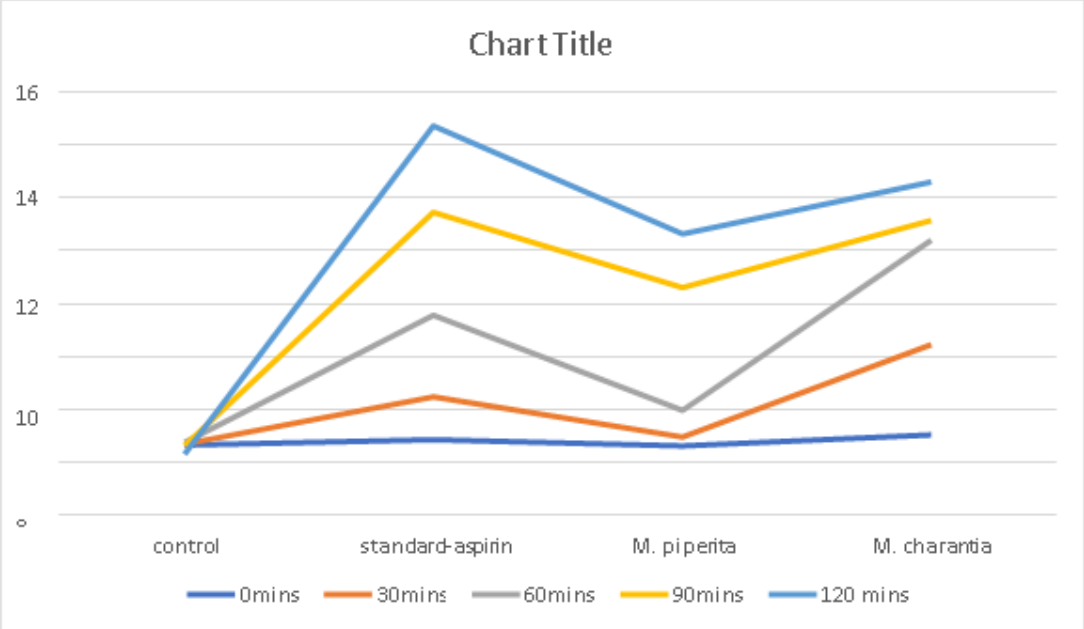
S no	Test	*M. Charantia*
1	Alkaloids	+
2	Flavonoids	+
3	Steroids	+
4	Saponins	+
5	Carbohydrates	+

ANALGESIC ACTIVITY RESULTS USING EDDY'S HOT PLATE

- Results of *M. piperita* & *M. charantia* on the Eddy's hot plate method are represented in table 3. The extracts of the plants are found to exhibit dose dependent increase in the reaction time when compared to the control group.
- Results are expressed as mean SEM from observations compared to control by one way ANOVA method **p<0.05. The difference is considered to be extremely statistically significant.
- The extract of *M. piperita* showed a dose dependent reaction time when compared to standard. As the dose of 200mg was given, the reaction time increased.
- The extract of *M. charantia* showed a dose dependent rection time when compared to standard. As the dose of 200mg was given, the reaction time increased.

Table 3: Reaction time of analgesic activity of the extracts.

Treatment dose mg/kg	Mean reaction time (sec) before & after drug administration				
	0mins	30mins	60mins	90mins	120mins
Normal saline 10ml/kg	2.64±0.13	2.69±0.3	2.75±0.42	2.62±0.43	2.3±0.26
Aspirin 10mg/kg	2.85±0.18	4.48±0.1	7.56±0.22	11.45±0.4	14.72 ±0.13
M. piperita 200mg/kg	2.61±0.18	2.95±0.3	3.96±0.24	8.6±0.08	10.63±0.34
M. charantia 200mg/kg	3.03±0.18	6.45±0.2	10.4±0.45	11.14±0.56	12.6±0.41



Graph 1: Representation of analgesic activity exhibited by samples.

ANTI-ULCER RESULTS

PYLORIC LIGATION IN RATS

- A total of 16 rats were taken and grouped into 4 groups namely CONTROL, STANDARD, TEST 1 AND TEST 2 and were fasted overnight to check the ulceration. Next morning, the standard group was treated with 20mg/kg of ranitidine and left for 4 hours.
- Simultaneously Test group 1 group of animals was treated with *M. piperita* with a dose of 500mg/kg as it did not show any acute level of toxicity.
- Similarly, Test group 2 of animals were treated with *M. charantia* with a dose of 500mg/kg with a dose of 500mg/kg as it did not show any level of toxicity.
- After 5 hours the animals were sacrificed and the stomach contents were emptied to check the ulcer score.

1. Control group

Table 4: Ulcer score in control group.

S. NO	NO. OF ULCERS SEEN
1	3
2	2
3	3
4	4
5	3

ulcer index= 3+2+3+4+3/10 = 15/10 = 1.5

2. Standard (rantidine 20mg/kg)

Table 5: Ulcer score in standard group.

S. NO	NO. OF ULCERS.
1	0
2	1
3	0
4	1
5	0

ULCER INDEX= 0+1+0+1+0/10 = 2/10 = 0.2

3. TEST 1 *M. piperita*

Table 6: Ulcer score in TEST 1 *M. piperita*.

S. NO	NO. OF ULCERS
1	0
2	1
3	1
4	0
5	1

Ulcer index= 0+1+1+0+1/10 = 3/10 = 0.3

4. TEST SAMPLE 2- *M. charantia*

Table 7: Ulcer score in TEST SAMPLE 2- *M. charantia.*

S. NO	NO. OF ULCERS
1	1
2	0
3	1
4	0
5	0

Ulcer index= 1+0+1+0+0= 2/10= 0.2

Effect of *M. piperita and M. charantia* in the prevention of duodenal small, medium, and large ulcers

Duodenal small, medium and large ulcers enhanced significantly in untreated rats compared to the control ones. Administration of *M. piperita & M. charantia* at all doses could significantly reduce ulcers in comparison to the untreated group. No medium and large ulcers were found in Test 1, Test 2, and Ranitidine groups.

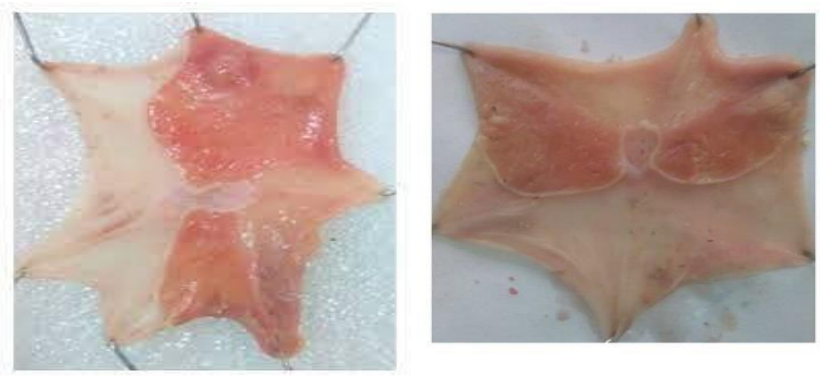


Fig. no 14: Control group results.

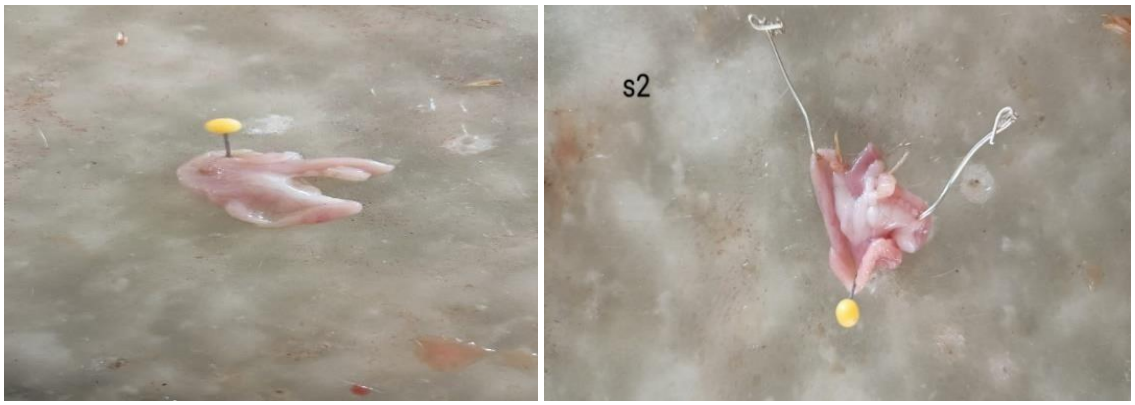
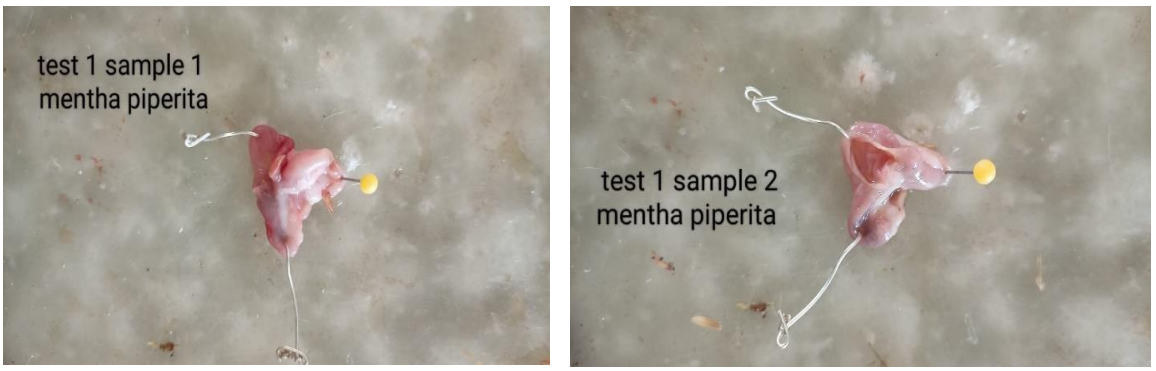


Fig no 15: Standard group results.



Sample 1 **sample 1**

Fig no 16 Test 1 results.

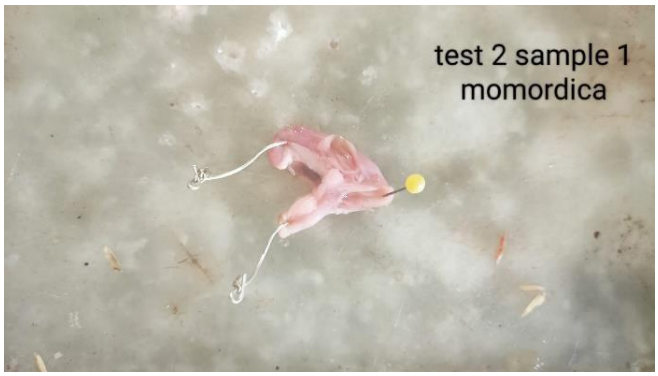


Fig no 17 test 2 results

Fig. Anti-ulcer activity in different groups.

CONCLUSION

In conclusion, pharmacological studies of Mentha piperita and Momordica charantia provides valuable insights into its potential medical uses. The study encompassed various aspects including phytochemical composition, analgesic activity and anti-ulcer activity. The extraction process methanol solvent is used. Phytochemical screening tests were done for the extracts. The phytochemical screening revealed the presence of diverse bioactive compounds in the Mentha piperita extract including alkaloids, flavonoids, steroids, terpenoids, phenols ,protein and carbohydrates. The colour of the Mentha piperita extract obtained was found to be green .Acute toxicity studies of the M.piperita did not show any toxicity up to 2g\kg body weight. Since there was no mortality of animals at high dose , 200mg\kg dose of extract was taken forthe evaluation of analgesic activity . The results of the analgesic activity of M.piperita on Eddy's hot plate method showed a dose dependent reaction when compared to standard. As the dose of 200 mg was given, the reaction time increased. The colour of the M. charantia was found to be brown. The phytochemical screening of the M. charantia extract revealed the presence of various bioactive compounds including alkaloids, saponins, flavonoids, steroids. The results of analgesic activity of M. charantia on Eddy's hot plate method showed a dose dependent reaction time when compared to standard . As the dose of 200mg was given the reaction time increased.

Antiulcer activity was determined by using the pylorus ligation method. The results showed that the administration of Mentha piperita extract and Momordica charantia at all doses could significantly reduce ulcers in comparison to the untreated group. No medium and large ulcers were found in the test1, test 2 and the standard ranitidine groups

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