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PHYTOCHEMICAL AND PHARMACOLOGICAL EVALUATION OF **PHYSALIS ANGULATA LINN BERRIES**

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ABSTRACT

Herbs include crude plant material such as leaves, flowers, fruit, seed, stems, wood, bark, roots and rhizomes or other plant parts, which may be entire, fragmented or powdered.^[1] Herbal medicine is still the mainstay of about 75 - 80% of the world population, mainly in the developing countries, for primary health care. Herbs contain a variety of chemical compounds that act upon the body and are used to prevent or treat disease or promote health and well-being. Herbal drugs have increasingly been used worldwide during the last few decades as evidenced by rapidly growing global and national markets of herbals and its preparations.^[2-5] Based on the therapeutic values of herbs, we attempted for the herbal extract from Physalis to analyte Phytochemicals compound to demonstrate various therapeutic and pharmacological activities. [6]

KEYWORDS: Medicinal plants; *Physalis angulata*, Extraction, Phytochemical screening: Antiulcer, Anti-Inflammatory.

INTRODUCTION

Developing countries like India depends on herbal medicines for their healthcare as per WHO. India having rich heritage of herbal resources and the traditional systems of medicines still in practice for various healthcare need by the people. Because of the least side effects and cost effectiveness, herbal medicines are in high demand and their acceptance level has

been increasing day by day. In Worldwide, there are 50,000 to 80,000 flowering plants are used medicinally.^[7]

Peptic ulcer

Peptic ulcer is an acid-peptic disease characterized by the rupture of the protective barrier of the epithelial mucosa of the esophagus, stomach or duodenum. Peptic ulcer occurs in that part of the gastrointestinal tract (GIT) which is exposed to gastric acid and pepsin, i.e. the stomach and duodenum. The etiology of peptic ulcer is not clearly known. The most common etiologies of gastric ulcer include a bacterial infection with Helicobacter pylori and gastric prostaglandin loss associated with non-steroidal anti-inflammatory medications. Less common etiologies include hypergastrinemia, viral infections such as CMV, chemotherapy and radiation, gastric outlet obstruction, gastric infiltrative disorders such as malignancy, cigarette smoking and Crohn's disease. [8-12]

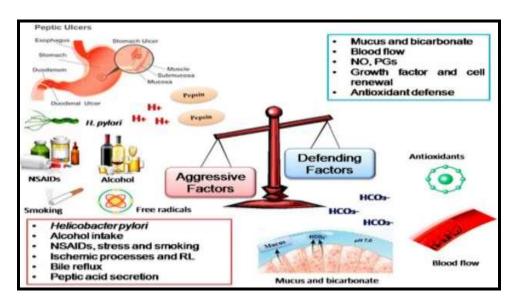


Fig. 1: MOA OF ULCER.

Inflammation

Inflammation is a complex set of interactions among soluble factors and cells that can arise in any tissue in response to traumatic, infectious, post-ischaemic, toxic or autoimmune injury. The process normally leads to recovery from infection and to healing, However, if targeted destruction and assisted repair are not properly phased, inflammation can lead to persistent tissue damage by leukocytes, lymphocytes or collagen. [13-14]

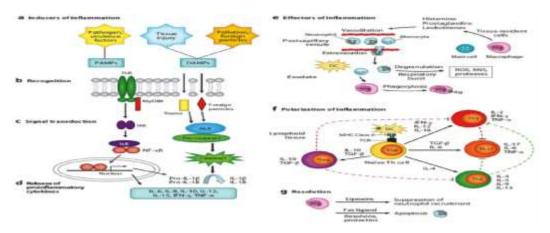


Fig. 2: Primers of the inflammatory cascade.

Plant Description



Fig. 3: Physalis angulate.

Botanical name: Physalis angulata Linn.

Taxonomical classification

Kingdom: Plantae

Subkingdom: Tracheobionta

Division: Magnoliophyta

Class: Magnoliopsida

Order: Solanales

Family: Solanaceae

Genus: Physalis

Species: angulata

Vernacular Name

Tamil: Sodakku Thakkali

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English: Balloon cherry, Wild gooseberry

Malayalam: Njottanjodiyan, Mottaampuli

Habit

It can be found on most continents in the tropics, including Africa, Asia, and the Americas.

Parts used

Berries are most useful part.

Constituents

Flavonoids, alkaloids and plant steroids known as physalins, B, D, F and G, withanolides.

Medicinal uses

This plant is traditionally used for the treatment of malaria, asthma, fever, sore throat, abdominal pain, malaria, externally the entire plant has been used in traditional herbal medicine systems for skin sores, rashes, pruritis and earaches.

Pharmacological action

Physalis angulata and its main constituent possess antimicrobial, antiprotozoal, antimycobacterial, antiviral, anticancer, antioxidant, antiallergic, anti-inflammatory, antihyperglycemic, antiulcer and antinociceptive activities.^[30,31]

MATERIALS AND METHODS

Extraction

The berries were dried in shade and were ground to a coarse powder. In this process, the whole or coarsely powdered crude drug is placed in a stoppered container with the solvent and allowed to stand at room temperature for a period of at least 3 days with frequent agitation until the soluble matter has dissolved. The mixture then is strained, the marc (the damp solid material) is pressed, and the combined liquids are clarified by filtration or decantation after standing.^[15-17]







Fig:5 Dried berries of

Fig:6 EAEPAB

Fig:7EEPAB

Physalis angulata Linn

Pharmacological evaluation of EEPAB

1. Anti-ulcer activity - Procedure - Acid Neutralizing Capacity

The acid neutralizing capacity value for solution of Sample (EEPAB) mixture using different concentration (500, 250, 100, 50 and 10 µg/ml) was compared with the standard antacid Aluminium hydroxide + Magnesium hydroxide (50 mg/ml). To the 5 ml quantity of this mixture, water was added to make up the total volume 70 ml and then mixed for one minute. There after 30 ml of 1.0 N HCl was added into standard and test preparation and stirred for 15 minutes, drops of phenophthalein solution was added and mixed. The excess HCl was immediately titrated with 0.5 N Sodium hydroxide solution drop wise until a pink color is attained.[18-21]

The moles of acid neutralized is calculated by,

Moles of acid neutralized = (vol. of HCl ×Normality of HCl) - (vol. Of NaOH × Normality of NaOH)

Acid neutralizing capacity (ANC) moles of HCl neutralized per gram of antacid Grams of Antacid/Extract

2. Anti-inflammatory activity - Inhibition of albumin denaturation - Procedure

Denaturation of proteins is the main cause of inflammation. Inhibition of protein denaturation was evaluated by the method of Mizushima and Kobayashi and Sakat et al. with slight modification. 500 µL of 1% bovine serum albumin was added to EEPAB (500, 250, 100, 50 and 10 µg/mL) of test sample. This mixture was kept at room temperature for 10 minutes, followed by heating at 51°C for 20 minutes. The resulting solution was cooled down to room temperature and absorbance was recorded at 660 nm. Acetyl salicylic acid was taken as a positive control. The experiment was carried out in triplicates and percent inhibition for protein denaturation was calculated using:

% Inhibition=100-((A1-A2)/A0)*100)

Where A1 is the absorbance of the control, A2 is the absorbance of the test sample and A0 is the absorbance of the positive control. A dose response curve was plotted to determine the IC50 values. IC50 is defined as the concentration sufficient to obtain 50% of a maximum scavenging capacity. All tests and analyses were run in triplicate and averaged. [22-24]

RESULTS AND DISCUSSION

1. Preliminary Phytochemical Screening.

| Phytochemicals | EAEPAB | EEPAB |
|------------------------------|---------------|--------------|
| Alkaloids | - | + |
| Carbohydrates | - | - |
| Flavonoids | - | + |
| Anthraquinone glycosides | - | - |
| Cardiac glycosides | + | - |
| Coumarin glycosides | - | - |
| Saponins | - | + |
| Tannins (Phenolic compounds) | - | + |
| Proteins | - | - |
| Steroids | - | - |
| Triterpenoids | _ | + |
| Gums & Mucilage | _ | _ |
| Lignins | - | - |

Presence (+), (-) – **Absence**

Table 1: Preliminary Phytochemical Screening of Crude extracts of Physalis angulata Linn

2. Pharmacological screening

A) Anti-ulcer activity of Physalis angulata Linn





Fig. 8: Before & After the process of titration of 1 N HCl containing sample (EEPAB 500,250,100,50,10 μ g/ml) and control.

Table 2: In vitro Acid neutralizing capacity of EEPAB.

| S.NO | Name of the sample | Name of the sample concentration | Reading a burette | Moles of acid neutralized | Acid neutralizing capacity (ANC)/ antacid (g) |
|------|--------------------------|----------------------------------|-------------------------|---------------------------|---|
| 1. | | 500 μg/ml | 2.6 | 2.7 | 54 |
| 2. | | 250 μg/ml | 3.0 | 1.5 | 30 |
| 3. | EEPAB | 100 μg/ml | 3.1 | 1.45 | 29 |
| 4. | EEFAD | 50 μg/ml | 3.2 | 1.4 | 28 |
| 5. | | 10 μg/ml | 3.4 | 1.3 | 26 |
| 6. | | Control | 3.5 | 1.25 | 25 |

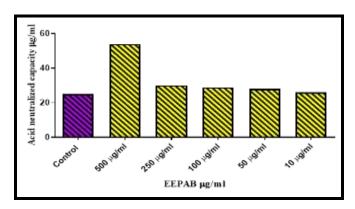
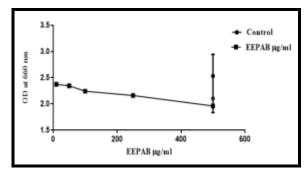


Fig. 9: Graphical representation of Effect of EEPAB on *Invitro* acid neutralizing capacity.

B) ANTI INFLAMMATORY ACTIVITY OF PHYSALIS ANGULATA LINN



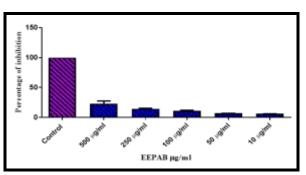


Fig. 10: Graphical represention of OD EEPAB at 660nm denaturation

Fig:11 Graphical representation of value of Inhibition percentage of albumin (%) by EEPAB

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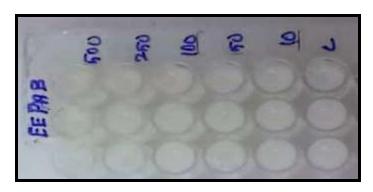


Fig: 12Microtiter plate containing 500 μ L of 1% BSA with EEPAB (500, 250, 100, 50 and 10 μ g/mL in PBS) and Control.

Table 3: OD Value of EEPAB at 660 nm Control Mean OD value: 2.533.

| S. No | Tested sample concentration (µg/ml) | OD Value at 660 nm (in triplicates) | | |
|-------|-------------------------------------|-------------------------------------|-------|-------|
| 1. | Control | 2.290 | 2.298 | 3.011 |
| 2. | 500 μg/ml | 2.011 | 2.055 | 1.817 |
| 3. | 250 μg/ml | 2.144 | 2.179 | 2.158 |
| 4. | 100 μg/ml | 2.226 | 2.265 | 2.238 |
| 5. | 50 μg/ml | 2.339 | 2.340 | 2.360 |
| 6. | 10 μg/ml | 2.360 | 2.371 | 2.391 |

Table 4: Inhibition percentage of albumin denaturation (%) by EEPAB.

| S. | Tested sample | Inhibition percentage albumin | | | Mean |
|----|-----------------------|-----------------------------------|----------|-----------|----------|
| No | concentration (µg/ml) | denaturation (%) (in triplicates) | | Value (%) | |
| 1. | Control | 100 | 100 | 100 | 100 |
| 2. | 500 μg/ml | 20.60797 | 18.8709 | 28.26688 | 22.58192 |
| 3. | 250 μg/ml | 15.35728 | 13.97552 | 14.80458 | 14.71246 |
| 4. | 100 μg/ml | 12.12002 | 10.58034 | 11.64627 | 11.44887 |
| 5. | 50 μg/ml | 7.658902 | 7.619424 | 6.829846 | 7.369391 |
| 6. | 10 μg/ml | 6.829846 | 6.395578 | 5.606001 | 6.277142 |

Table 5: IC50 value of EEPAB for protein denaturation.

| log(inhibitor) vs. normalized | | |
|-------------------------------|------------------|--|
| response Variable slope | | |
| Best-fit values | | |
| LogIC50 | 2.265 | |
| HillSlope | -1.866 | |
| IC50 | 183.9 | |
| 95% CI (profile likelihood) | | |
| LogIC50 | 2.118 to 2.404 | |
| HillSlope | -3.298 to -1.157 | |
| IC50 | 131.3 to 253.4 | |
| Goodness of Fit | | |
| Degrees of Freedom | 13 | |
| R squared | 0.8527 | |
| Sum of Squares | 3166 | |
| Sy.x | 15.61 | |
| Number of points | | |
| # of X values | 15 | |
| # Y values analyzed | 15 | |

IC50 Value of tested sample: 183.9 μg/ml

DISCUSSION

Preliminary qualitative test is useful in the detection of bioactive principles and subsequently may lead to drug discovery and development. The preliminary phytochemical analysis indicates the nature of phytoconstituents present in different solvent extracts. The qualitative chemical examination showed the presence of alkaloids, flavonoids, tannins (phenolic compounds), triterpenoids and saponins in EEPAB and presence of cardiac glycosides in EAEPAB. Results presented in Table no:1. Phytoconstituents and therapeutic activity usually linked due to its antioxidant role and it plays a significant role in disease prevention. The Plant berries contain alkaloids, flavonoids, tannins (phenolic compounds), triterpenoids and saponins may act synergistically in prevention of major problems like inflammation and associated ulcer complications.

The EEPAB was tested for both *in vitro* anti-ulcer & anti-inflammatory activities support the above statement and the literature also stating the usage of this berries based extract found useful in treating inflammation. Table 2 & Fig:10,11,12 showed the dose dependent activity of the extract in the ulcer treatment 500 µg showed good response Significant reduction in the inflammation pattern with the extract with doses tested 10 µg, 50 µg, 100 µg, 250µg and 500 μg. Out of 5 doses, 500 μg showed maximum response. This indicate the extract at the

dose of 500 µg/ml showed a good therapeutic activity with the experiments conducted to justify the same.

CONCLUSION

Based on the above results, the plant extract Physalis angulata Linn, found to possess significant antiulcer and anti-inflammatory property. As this extract contains group of phytoconstituents that may synergistically support the effectiveness of the therapeutic claim based on the literature.

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