EVALUATION OF ANTIINFLAMMATORY ACTIVITY IPOMOEA AQUATICA ON EXPERIMENTAL ANIMALS

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ABSTRACT: ***Inflammation is commonly thought of as a swollen, painful, or otherwise unpleasant condition, such as in your joints, sinuses, or bowel. However, most people experience inflammation without any symptoms. Inflammation is traditionally characterised as the body's defensive response to physical or chemical insult; acute inflammatory response occurs soon after cellular injury. The number of chemical compounds known as phytochemicals discovered in the plant kingdom is truly enormous, as is their range of function. Some phytochemicals contained in specific herbs and plants have been shown to have anti-pain and anti-inflammatory activities. By injecting 0.1 ml of 1% carrageenan under the skin of the rat's planter region, edema was caused in the rat's right hind paw.Albino Rat were treated with either EEIA-LD, EEIA-HD or diclofenac at 20 mg/kg dose i.m. with saline. 1 h after the treatments, individual mouse was injected i.v. with 2% Evan's blue solution at 10 mL/kg body weight through the tail vein. After 10 minutes, each rat was injected i.p. with 10 ml of a 6 percent acetic acid solution in saline. MEIA showed a dose dependent significant inhibition of paw edemafollowing carrageenan administration (low dose - $p < 0.01$, high dose – $p < 0.001$). Treatment with EEIA-LD and EEIA-HD showed decrease in Evans blue dye extravasation into the peritoneal cavity of mice ($p < 0.05$ and $p < 0.01$).***
Keywords: Anti –Inflammatory, Ipomoea aquatica, Methanolic extract.

I. INTRODUCTION

In the world right now, inflammation is one of the most important health problems (1, 2). Inflammation is the body's way of getting rid of or stopping the spread of harmful substances. It is the local response of living tissues to damage (1, 2). The main signs to look for are heat, redness, swelling, pain, and loss of function (1-4). This happens in three steps: blood vessels widen and become more permeable, phagocytic cells move in, and tissues heal. [(3,4)].

Inflammation can be short-term or ongoing (3, 4). While chronic inflammation can endure for months or even years, acute inflammation develops quickly and lasts for only a few minutes to a few days (3,4). Without prompt removal of the offending chemical, persistent inflammation can set up (3). Chronic prostatitis, glomerulonephritis, hypersensitivities, pelvic inflammatory disease, interstitial cystitis, etc. are only a few of the inflammatory conditions that have been linked to antibiotic use (4).

To stop or slow the inflammatory disease's spread, doctors can prescribe any number of treatments such nonsteroidal anti-inflammatory drugs (NSAIDs) and corticosteroids (5). Several adverse effects have been linked to NSAIDs, including suppression of bone marrow production, intolerance of the drug in the stomach, kidney failure, retention of water and salt, liver failure, ulcers, and excessive bleeding after injury or surgery (5). Weight gain, skin bruising, osteoporosis, diabetes, swelling in the ankles and feet, cataracts, and other side effects are possible with either high doses or prolonged usage of steroids (6).

As a result, there is a pressing need to create a novel anti-inflammatory composition that is both effective

and safe for use. Curcumin from Curcuma longa L. (Zingiberaceae) rhizomes, polyphenols like resveratrol from grape skin, flavonoids like rutin, quercetin, triterpenoid saponins like avicins from Acacia Victoria (Leguminosae), ursonic acid from Plantago major (Plantaginaceae), etc., are all used to treat acute and chronic inflammation (7).

It is a perennial herb belonging to the Convolvulaceae family that can be found in India, Sri Lanka, Africa, Tropical Asia, and Australia (10). The Unani medical system makes use of I. aquatica as a carminative agent and an anti-inflammatory that helps with a variety of conditions including fever, jaundice, biliousness, bronchitis, and liver ailments (11). I.aquatica is a popular green leafy vegetable that contains numerous beneficial nutrients such vitamins, minerals, proteins, fibres, carotenoids, and flavonoids (12).

However, the absence of documentation and quality control requirements has been a major barrier to the worldwide acceptance of alternative medicines. Therefore, it is crucial to work towards standardization of medicinal plant material. This is why the macroscopy and microscopy of crude pharmaceuticals, powder microscopy, physiochemical parameters, and preliminary phytochemical analysis of I. aquatica leaves are all part of the evaluation process.

The widespread idea that green medicine is safer than synthetic products sparked a renewed interest in natural pharmaceuticals in the last decade. Developed and emerging countries alike are increasingly turning to herbal remedies, fueling the contemporary era's exponential expansion of plant-based health goods in the worldwide market (15, 16). The approximate value of the global herbal market is $61 billion (15, 16). In 2013, the German herbal medicine market was worth around $3 billion, the American herbal medicine market was worth about $1.5 billion, and the Indian ayurvedic industry was worth about $0.8 billion (15,16). About 80% of the global population still relies primarily on herbal medicine for their primary healthcare requirements, especially in poor and under developed nations, where it is more widely available, more widely accepted culturally, and more compatible with the human body (17). New herbal formulations utilising plant extracts are being developed for the treatment of both acute and chronic pathophysiological diseases including inflammation.
This study set out to combine the most potent components of the medicinal plant under study in order to create a novel herbal formulation with anti-inflammatory potential.

Inflammation

Although inflammation can be damaging or even fatal, it is a necessary and protective local response of living tissues to injury (1, 2). It's a natural response that the body has developed to neutralize or contain harmful substances (1, 2). Pain, heat, and redness (erythema) are all symptoms of inflammation (1, 2).

PhenomenonofInflammationResponse

An important part of innate immunity is inflammation (18). There are three distinct stages to the inflammatory response: I blood vessel dilatation and enhanced permeability (ii) phagocyte migration from blood into interstitial fluid (iii) tissue healing (3, 19).

Histamine, kinins, prostaglandins (PGs), leukotrienes, and various components of the complement system all have a role in vasodilation and increased vascular permeability. Dilation of arterioles and increased permeability of capillaries cause the three signs of inflammation: heat, redness, and swelling (3, 19). Neuronal damage, bacterial toxins, and increased pressure due to swelling all contribute to pain (19). Agony is primarily caused by kinins, which act on nerve terminals, and by prostaglandins, which amplify and prolong the pain of inflammation (19).

In the first hour after inflammation begins, neutrophils begin migrating out to eliminate invading bacteria by phagocytosis (3, 19). As the inflammatory response proceeds, monocytes arrive at the site of infection shortly after neutrophils (3, 19). Tissue-resident monocytes undergo a differentiation programme that allows them to become free-roaming macrophages (3, 19). Comparatively, neutrophils are weaker phagocytes than macrophages (3, 19). They can swallow injured tissue and invading microorganisms, as well as wear down neutrophils (3, 19). Macrophages perish within days as well (3, 19). Dead phagocytes, injured tissues, and fluid buildup lead to pus production (3, 19).

TypesofInflammation

Inflammation can be short-lived or ongoing. Fluid and plasma protein exudation and a predominately neutrophilic cell buildup characterize the short-lived inflammation known as acute inflammation. Its onset

can be as quick as a few minutes and its persistence as long as a few days (3, 19, 20). Chronic inflammation is characterized by lymphocyte and macrophage infiltration, vascular proliferation, and fibrosis, and it can be more persistent (lasting days to years) (20). When the harmful chemical is removed and the anti-inflammatory systems kick in, the process winds down and the host organism recovers to its previous healthy state (20). Injuries can lead to persistent inflammation if the offending factor isn't swiftly removed (20).

InflammatoryDisorders

Conditions like rheumatoid arthritis, inflammatory bowel disease, acne vulgaris, autoimmune disease, chronic prostatitis, glomerulonephritis, hypersensitivities, pelvic inflammatory disease, reperfusion injury, sarcoidosis, transplant rejection, vasculitis, interstitial cystitis, and many more are linked to inflammation (20).

CurrentlyUsedAnti-InflammatoryMedicines

Non-steroidal anti-inflammatory drugs (NSAIDs) like aspirin, ibuprofen, celecoxib, diclofenac, indomethacin, ketoprofen, piroxicam; corticosteroids (steroids in tablet form include prednisolone, prednisone, and medrol); and many others can be used to slow or stop the inflammatory disease's progression (20). Inhaled types of pharmaceuticals such as methotrexate, sulfasalazine, leflunomide, anti-TNF therapies, cyclophosphamide, and mycophenolate are available, as are anti-malarial drugs like hydroxychloroquine (20).

AdverseEffectsofCurrentlyUsedMedicines

Several adverse effects have been linked to NSAIDs (20—22). Nausea, vomiting, diarrhoea, constipation, decreased appetite, rash, dizziness, headache, and sleepiness are the most commonly reported negative reactions. Gastric intolerance, decrease of bone marrow, water and salt retention are common side effects of long-term NSAID use (20—22). Kidney failure, liver failure, ulcers, and excessive bleeding after an injury or surgery are among the worst possible outcomes (20—22).

Some people have NSAID allergies and experience shortness of breath after taking one. Asthmatics are more likely to have a severe allergic reaction to NSAIDs. The onset of Reye's syndrome has been linked to the use of aspirin in children and adolescents suffering from chickenpox or the flu (20). Heart attacks, strokes, and other cardiovascular complications may be more likely to occur in those taking NSAIDs (other than low dose aspirin) (20). Long-term use and patients with preexisting risk factors for cardiovascular disease may increase this danger (20, 21). Pain after coronary artery bypass grafting should not be treated with nonsteroidal anti-inflammatory drugs (20).

The severity of steroid-related side effects often increases with increased dosing (21). In contrast to the pill form, inhaled steroids are less prone to cause side effects (21). Sore throat, hoarse voice, and mouth and throat infections are typical reactions to inhaled steroids (21). The lac of steroids can create severe, life-threatening problems, and the high doses of steroids in tablet form (or smaller doses given for long periods of time) may cause bruising of the skin, weight gain, bone weakening (osteoporosis), high blood sugar levels (diabetes), cataracts, swelling of the ankles or feet (21)

PhytoconstituentsANewSourceofAnti-InflammatoryAgents

As a result, there is a pressing need to create a novel, safe anti-inflammatory formulation with minimal side effects, as most existing anti-inflammatory medications have a number of undesirable effects. Curcumin, derived from Curcuma longa L. (Zingiberaceae) rhizomes; polyphenols include resveratrol from grape skin; 5,7,5'-trihydroxy-3,6,2',4'-teramethoxyflavone; scopoletin and centaureidin from Eupatorimbuniifolium (Asteraceae) (22, 23); flavonoids including rutin, quercetin (22, 23).

According to the World Health Organization, around 80% of people still use primarily herbal medicines (17, 23). Because of their low cost and lack of toxicity, plant-based medications are being studied extensively in the present study. These natural plants are also being used to generate new herbal formulation for the treatment of acute and chronic pathophysiological diseases, such as inflammation.

III. MATERIALS &METHODOLOGY

Materials:

Carrageenan (Sigma Chemical Company, St. Louis, MO, USA). Methanol, distilled water and all the other reagents were used of analytical reagent grade purchased from Merck India. diclofenac injections standard non-steroidal anti-inflammatory drugs were purchased from a pharmaceutical shop at Hyderabad Telangana, Gujarat, India.

Instruments:

Histamine chamber, Nebulizer, Glass rod, Beaker, Stop watch and Cages.

Plant material collection

Ipomoea aquatica were collectedwas purchased from Svagro foods

Experimental animals

During the experiment, albino wistar rats weighing 150-200gms were obtained from the jeeva life science animal home. The test animals were kept in typical laboratory conditions, which included a 12-hour light/dark cycle and temperature control. All of the animals were forced to acclimatise to laboratory settings for at least one week before the trial began.

Methodology:

Extraction

Ipomoea aquatica leaves acquired from Svagro foods. The powder was Sieved via a 40-mesh sieve before being extracted with Soxhlet. 800 gm powdered extract was obtained using a methanolic solvent (80 percent methanol in a Soxhlet Apparatus). The residue was dehydrated in a hot water bath at 50^0 degrees Celsius. After drying, the weight of the extract was calculated to be 98 gms, yielding a percentage yield of 15.25 percent.

Finally, preliminary phytochemical screening of the extracted extract was performed to discover several phytochemical components.

Preliminary phytochemical screening

The MEIA *was*evaluatedto check if phytochemicals such as alkaloids, carbohydrates, glycosides, proteins, amino acids, flavonoids and tannin & phenolic compounds are present.

Preliminary Phytochemical Screening

The whole plant part extracts of CR obtained after each successive steps were subjected to qualitative chemical testing for preliminary screening of phytoconstituents. Phytochemical screening were performed using standard procedures.13,14 Phytochemical screening of CR extracts include test for alkaloids, saponins, glycosides and sugar, phenolic compounds and tannins, flavonoids and flavones, coumarin and its derivatives and triterpenoids.

1. Oral toxicity studies

Acute oral toxicity experiments were conducted on albino rats using a methanolic extract of the whole plant of Ipomoea aquatica in accordance with OECD guidelines 425. The animals were fasted for 24 hours. They were first given an oral dose of a methanolic extract of Ipomoea aquaticaforsk (1000mg/kg body weight) dissolved in water. The animals were regularly examined for 3-4 hours for any changes in their general behaviour, morbidity, and eventually, during the toxicity testing period, all animals were healthy (i.e. 2 days).

Because there was no evidence of toxicity at 1000mg/kg, a greater dose of methanolic extract (2000mg/kg body weight) was administered for the evaluation of all biochemical profiles. Some variations in usual behaviour were seen, such as chewing, rashes, and shortness of breath, but no death was recorded. The two dosages were chosen so that the high dose (400mg/kg) was almost double of one-tenth of the highest dose used in acute toxicity studies, while the low dose (200mg/kg) was around one-tenth of the highest dose. Because there was no evidence of toxicity at 1000mg/kg, a greater dose of methanolic extract (2000mg/kg body weight) was administered for the evaluation of all biochemical profiles. Some variations in usual behaviour were seen, such as chewing, rashes, and shortness of breath, but no death was recorded. The two dosages were chosen so that the high dose (400mg/kg) was almost double of one-tenth of the highest dose used in acute toxicity studies, while the low dose (200mg/kg) was around one-tenth of the highest dose.

2. Carragenan induced paw odema in rats

Albino rats (weighing 200-250 g) of both sexes were divided into five groups of six animals each. The following was the treatment's experimental protocol:

Group 1: The control group received saline injections and was treated orally with 0.5 percent CMC.

Group 2: Carrageenan was administered into the control group

Group 3: MEIA-LD, treated with methanolic extract of *Ipomoea aquatica* (200 mg/kg p.o.) in 0.5% CMC

Group 4: MEIA-HD, treated with methanolic extract of *Ipomoea aquatica* (400 mg/ kg p.o.) in 0.5% CMC

Group 5: Standard group, treated with Diclofenac sodium (20 mg/kg dose i.m. with saline)

Animals in groups 2-5 were given their respective medications, followed by 0.1 ml of 1 percent carrageenan subcutaneously administered into the planter region of the right hind paw 1 hour later. This treatment caused paw-edema, and the initial paw volume was measured using a plethysmographic method at 0 hour, 3 hours, and 5 hours following carrageenan treatment. The amount of paw edema (ml) increased was assessed. [24-26]

Acetic acid induced vascular permeability

Albino rats were given either EEIA-LD, EEIA-HD, or diclofenac at a dose of 20 mg/kg dose i.m. with saline. Individual rats were injected intravenously with 2 percent Evan's blue solution at 10 mL/kg body weight through

the tail vein 1 hour after the treatments. After 10 minutes, each rat was injected i.p. with a 0.6 percent acetic acid solution (in saline) at a rate of 10 ml/kg body weight.The rat was euthanized after 30 minutes of acetic acid administration, and the peritoneal cavity was cleaned three times with saline (10 ml). The saline washes were centrifuged for 5 minutes at 3500 rpm. The supernatant was collected, and the absorbance at 590 nm was measured using a plate reader. Evans blue extravasation was quantified using a standard curve and represented in micrograms.

% inhibition was calculated using the formula given below:

$$= \frac{Absorbance_{test} - Absorbance_{control}}{Absorbance_{test}} \times 100$$

In-vitro anti-inflammatory activity.

The Human red blood cell (HRBC) membrane stabilization method

A blood sample of 2ml was collected from a volunteer using a heparinized tube. The sample was then washed twice with phosphate buffered saline and centrifuged at 3000 rpm for 10 minutes. Next, red blood cells (RBCs) were suspended in normal saline and transferred to a 0.5 ml tube. To this, 0.5 ml of extract and 0.5 ml of hypotonic solution were added. The mixture was then incubated at room temperature for 30 minutes. Next, the contents were centrifuged at a speed of 1500 rpm for a duration of 10 minutes. After centrifugation, the supernatant was carefully collected and the absorbance was measured at a wavelength of 560nm. The membrane stabilisation effect was calculated based on the absorbance of the extract and control.

Inhibition of albumin denaturation method

The reaction mixture comprised Leaves extracts and a 1% aqueous solution of bovine albumin fraction. The pH of the reaction mixture was adjusted using 1N HCL. The samples were subjected to incubation at a temperature of 37°C for a duration of 20 minutes. Subsequently, the samples were heated to a temperature of 51°C for another 20 minutes. After allowing the samples to cool, the turbidity was measured using spectrophotometry at a wavelength of 660nm. The experiment was conducted in triplicate. The calculation of the percentage inhibition of protein denaturation was performed using the following method:

Statistical analysis

Prism 5.04 (Graph Pad) was used for statistical analysis. Data was analysed using ANOVA, followed by Tukey's multiple comparison tests. Values are reported as mean standard error of the mean (SEM), with $p < 0.05$ deemed statistically significant.

III. RESULTS

Preliminary phytochemical screening

The methanolic extract of *ipomoea aquatica* was screened for the presence of alkaloids,

Table 1: Results of preliminary phytochemical screening

S.No	Tests	Extract of *Ipomoea aquatica*
1	**Alkaloids**	
	a. Dragendroff's test	+ve
	b. Mayer's test	-ve
	c. Hager's test	-ve
	d. Wagner's test	+ve
2	**Carbohydrates**	
	a. Molisch test	+ve
	b. Fehling's test	+ve
	c. Benedict's test	+ve
	d. Barfoed's test	+ve
3	**Proteins**	
	a.Biuret	-ve
	b.Millions reagent test	+ve
	c.Xanthoprotein test	-ve
	d.Protein containing sulphur test	+ve

4	**Test for steroids**	
	a.Salkowiski test	-ve
	b.Liebermann test	-ve
5	**Test for flavonoids**	
	a.Lead acetate test	+ve
	b.Sodium hydro oxide test	+ve
6	**Tannins and phenolic compound**	
	a. Lead acetate solution	+ve
	b.Iodine solution	+ve
	c.Acetate acid solution	-ve
	d.Nitric acid solution	+ve
7	**Glycosides**	-ve

Effects of MEIA on carrageenan-induced paw edema:

Table 2: Effect of MEIA on carrageenan-induced paw edema.

Treatment	**Increase in paw edema (mL)**					
	After 1 h		**After 3 h**		**After 5 h**	
	Left Leg	**Right Leg**	**Left Leg**	**Right Leg**	**Left Leg**	**Right Leg**
Normal control	0.24 ± 0.003	0.24 ± 0.011	0.24 ± 0.004	0.23± 0.013	0.24 ± 0.006	0.23± 0.009
Carrageenan control	0.23 ± 0.004	0.42 ± 0.0095$^{\alpha}$	0.23 ± 0.004	0.940 ± 0.021$^{\alpha}$	0.23 ± 0.004	0.80 ± 0.024$^{\alpha}$
MEIA (LD)	0.24 ± 0.004	0.34 ± 0.021^{b}	0.23 ± 0.005	0.850 ± 0.022^{c}	0.24 ± 0.005	0.66 ± 0.028^{a}
MEIA (HD)	0.23 ± 0.003	0.29 ± 0.011^{a}	0.23 ± 0.004	0.350 ± 0.022^{a}	0.23 ± 0.005	0.30 ± 0.023^{a}
Standard	0.24 ± 0.004	0.24 ± 0.012^{a}	0.23 ± 0.005	0.220 ± 0.015^{a}	0.24 ± 0.004	0.21 ± 0.012^{a}

Values are expressed as mean ± SEM. $^{\alpha}p < 0.001$, as compared withthe control group, $^{a}p < 0.001$, $^{b}p < 0.01$,$^{c}p < 0.05$in comparison withthe carrageenan control group. The difference between the groups was determined usinga one-way analysis of variance (ANOVA) andTukey's test.

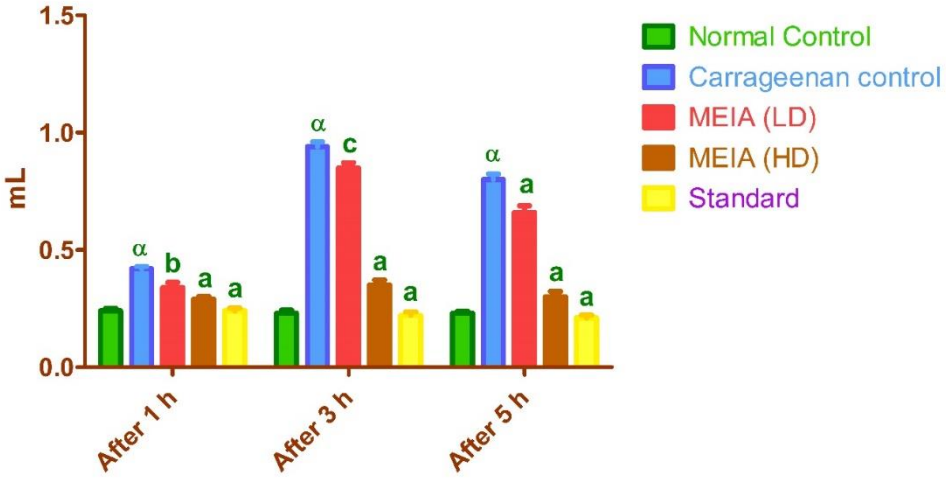

Fig.1. Effect of MEIA on carrageenan-induced paw edema.

Values are expressed as mean ± SEM. $^{\alpha}p < 0.001$, in comparison withthe control group, $^{a}p < 0.001$, $^{b}p < 0.01$, compared to the carrageenan control group. The difference between the groups was determined using a one-way

analysis of variance (ANOVA) and Tukey's test.

Subplantar injection of carrageenan in rats resulted in the development of paw edema. Methanolic extract of *Ipomoea aquatica* showed a dose-dependent significant inhibition of paw edema at time 1 hour 3 hours and 5 hours after carrageenan administration (low dose - $p < 0.01$, high dose – $p < 0.001$). The standard anti-inflammatory drug diclofenac sodium also caused a significant decrease in paw edema ($p < 0.001$) at times 3 hours and 5 hoursafter carrageenan administration (Table 7, Figure 11).

Effect of Methanolic extract of Ipomoea Aquatica (MEIA) on acetic acid induced vascular permeability

Table 3: Effect ofMEIA-LD and MEIA-HD on Evans blue dye extravasation into the peritoneal cavity of rat

Group	Control	MEIA-LD	MEIA-HD	Standard
Dye leakage (µg)	0.029 ± 0.001	0.022 ± 0.002[c]	0.019 ± 0.002[b]	0.0037 ± 0.001[a]
Percentage inhibition	-	31.8 %	52.6 %	21.6%

Values are expressed as mean ± SEM. [a]$p < 0.001$, [b]$p < 0.01$, [c]$p < 0.05$ compared to carrageenan control group. Difference between the groups was analyzed by one-way analysis of variance (ANOVA) followed by turkey's test.

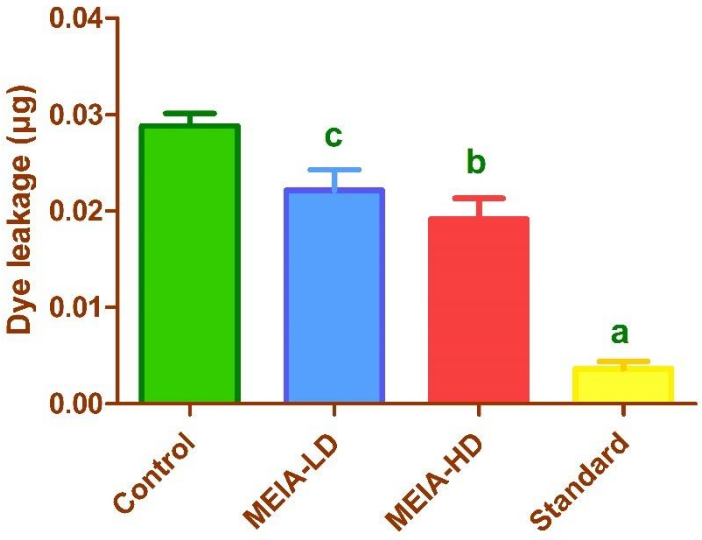

Fig.2.Effect of MEIA-LD and MEIA-HD on Evans blue dye extravasation into the peritoneal cavity of rat

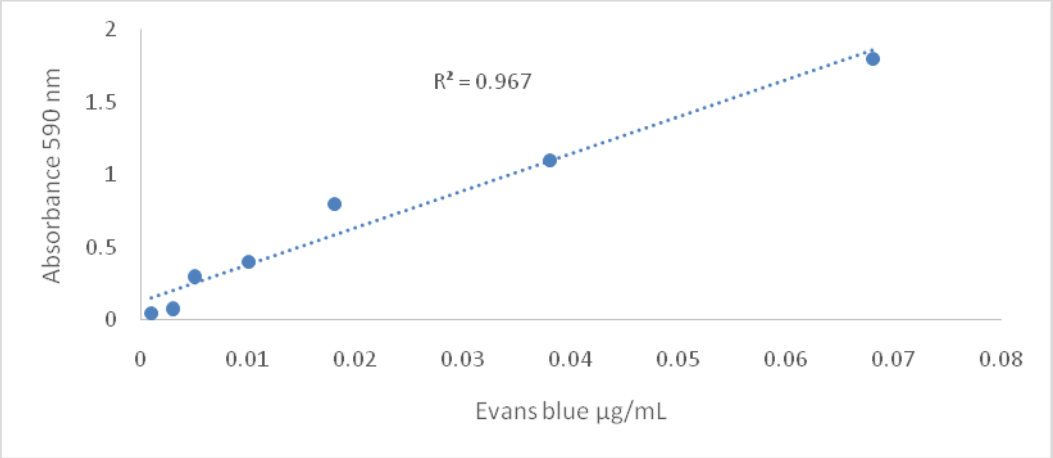

Fig.3. Evan blue dye standard curve

Treatment with MEIA-LD and MEIA-HD showed a decrease in Evans blue dye extravasation into the peritoneal cavity of mice ($p < 0.05$ and $p < 0.01$).

Values are expressed as mean ± SEM. $^{a}p < 0.001$, $^{b}p < 0.01$,$^{c}p < 0.05$ compared to carrageenan control group. The difference between the groups was analyzed by one-way analysis of variance (ANOVA) followed by Tukey's test.

Anti-inflammatoryactivity

InhibitionofAlbumindenaturationmethod

The present study investigated the inhibitory effect of various concentrations of I. aquatica, ranging from 300 to 1500 µg/ml, on the denaturation of egg albumin. The results demonstrated a significant concentration-dependent inhibition of denaturation, as evidenced by the standard concentrations of 300, 600, 900, 1200, and 1500 µg/ml. The in vitro anti-inflammatory activity of I. aquatica, as investigated in our study, is attributed to both its membrane stabilisation activity and its impact on protein denaturation.

Concentration (µg/ml)	***I. aquatica Methanolic extract* % inhibition**	**Diclofenac standard**
300	68.89±0.012	122.12±0.087
600	145.89±0.082	149.23±0.045
900	289.73±0.091	179.78±0.078
1200	398.58±0.036	272.35±0.035
1500	547.12±0.245	319.12±0.025

HRBC Membrane Stabilization Method

The investigation focused on evaluating the membrane stabilisation activity of I. aquatica across a concentration range of 300, 600, 900, 1200, and 1500 µg/ml. The results demonstrated that the erythrocyte membrane was effectively protected against lysis induced by a hypotonic solution in a concentration-dependent manner. The standard concentrations of diclofenac (300, 600, 900, 1200, 1500 µg/ml) were found to provide protection to the HRBC membrane against the damaging effects caused by a hypotonic solution. This study investigates the membrane stabilisation action and inhibitory effect of various concentrations of I. aquatica**.**

Concentration (µg/ml)	***I. aquatica Me*thanolic extract% inhibition**	**Diclofenac standard**
300	58.15± 0.035	89.85±0.045
600	68.26±0.049	102.35±0.048
900	89.23±0.065	108.24±0.036
1200	98.23±0.078	110.25±0.387
1500	125.32±0.136	115.25±0.0361

Table 4: *InVitro* Anti-Inflammatory Activity of ethanolic extract of *ZeaMays* And Aspirin of HRBC Membrane Stabilization method

DISCUSSION

Inflammation is traditionally characterised as the body's defensive response to physical or chemical insult; acute inflammatory response occurs soon after cellular injury. The number of chemical compounds known as phytochemicals discovered in the plant kingdom is truly enormous, as is their range of function. Some phytochemicals contained in specific herbs and plants have been shown to have anti-pain and anti-inflammatory activities.

Preliminary phytochemical screening of Ipomoea aquatica extract revealed the presence of carbohydrates, proteins, amino acids, alkaloids, flavonoids, glycosides, tannins, and saponins. These phytochemical substances may contribute to the plant's anti-asthmatic activity. Dhanasekaran et al. (2010) investigated the antimicrobial and anti-inflammation activity of methanolic extract of Ipomoea aquatica; similarly, in the current study, Methanolic extract of Ipomoea aquatica demonstrated significant anti-inflammatory activity against acetic acid-induced vascular permeability.Following carrageenan treatment, MEIA demonstrated a dose-dependent substantial reduction of paw oedema (low dose: $p< 0.01$, large dose: $p <0.001$). Evans blue dye extravasation into the peritoneal cavity of mice was reduced following treatment with EEIA-LD and EEIA-HD ($p< 0.05$ and $p <0.01$, respectively)

The HRBC membrane stabilisation technique has been employed as a means of investigating the in vitro anti-inflammatory properties, as the erythrocyte membrane bears resemblance to the lysosomal membrane. The observed stabilisation of the HRBC membrane suggests that the extract may have the potential to stabilise the

lysosomal membrane as well. The stabilisation of lysosomes plays a crucial role in mitigating the inflammatory response by inhibiting the release of lysosomal components from activated neutrophils. These components, including bacterial enzymes and proteases, have the potential to exacerbate tissue inflammation and inflict additional damage upon their extracellular release.

The release of lysosomal enzymes during inflammation can result in the development of various disorders. The enzymes' extracellular activity is purportedly associated with the continuum of inflammation from acute to chronic stages. Non-steroidal drugs exert their effects through two mechanisms: inhibition of lysosomal enzymes or stabilisation of lysosomal membranes.

The HRBC method was chosen for the in vitro assessment of the anti-inflammatory property due to the similarity between erythrocyte membranes and lysosomal membranes. The stabilisation of erythrocyte membranes suggests that the extract may also have the potential to stabilise lysosomal membranes. The stabilisation of lysosomal membranes plays a crucial role in mitigating the inflammatory response by inhibiting the release of lysosomal constituents from activated neutrophils. These constituents, including bactericidal enzymes and proteases, have the potential to exacerbate tissue inflammation and inflict additional damage upon extracellular release.

IV. CONCLUSION

Finally, *I. aquatica* is useful in treating anti –inflammation.MEIA showed a dose dependent significant inhibition of paw edema following carrageenan administration (low dose - $p < 0.01$, high dose – $p < 0.001$). Treatment with EEIA-LD and EEIA-HD showed decrease in Evans blue dye extravasation into the peritoneal cavity of rat ($p < 0.05$ and $p < 0.01$). The findings will offer the foundation for future research and application of these plants in the development of new drugs.

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