World Journal of Pharmaceutical Sciences
ISSN (Print): 2321-3310; ISSN (Online): 2321-3086
Published by Atom and Cell Publishers
Available online at: http://www.wjpsonline.com/
Research Article

Study on anti-ulcer and anti-inflammatory effects of *Vilvathi Lehiyam*

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Received: 01-10-2013 / Revised: 06-12-2013 / Accepted: 25-12-2013

ABSTRACT

The aim of the study, to evaluate the phytochemical, anti-ulcer and anti-inflammatory activities of *Vilvathi Lehiyam.* Anti-ulcer activity of ethanolic extract of *Vilvathi Lehiyam* was investigated on omeprazole induced ulcer model in albino rats. Ethanolic extract of dosage 250 and 500kg/mg produced significant inhibition of gastric lesions induced by Omeprazole induced ulcer. The extract 250 and 500kg/mg showed significant ($p<0.01$) reduction of p^H value of gastric juice compared control. The *Vilvathi Lehiyam* was evaluated for anti-inflammatory activity against the carrageenan induced rat paw oedema at injected sthe doses 500 kg/mg body weight and the study was compared with standard drug Dexamethasone (2mg/kg). The *Vilvathi Lehiyam* has significant anti-inflammatory activity, which support the traditional medicinal utilization of *Vilvathi Lehiyam*. Based on the above results, of *Vilvathi Lehiyam* may be useful as a natural drug for the treatment of ulcer and inflammation.

Key words: GC-MS, Anti-ulcer and Anti-inflammatory, *Vilvathi Lehiyam.*

INTRODUCTION

In siddha pharmacopoeia various herbs and their parts have been widely used for the different types. In siddha system equal importance has been given to internal as well as external medicine because siddha medicine advocates 32 type of internal and 32 type of external medicine with their self-life (Thiyagarajan *et al.*,1952). Medicated *Leyhiyam* from herbs is a common type of internal medicine and is used as a base line treatment for all aliments including ulcer and inflammation. According, it is advised to first pure herbs in the form of liquid, powder, pill or paste. It is not controlling. Then the physicians use mixture of herbs metals, minerals, and animal product in additional to herbs. Another type of Internal medicine is potion which gets absorbed quickly within our system and facilitate faster action that is a greater important in ulcer and inflammation treatment.

There are wide arrays of treatment modalities in siddha system practical by a number of physicians well organized preclinical well and clinical trial evidence are not adequately available in order to advocate their scientific merit and supremacy over the existing therapies. Hence scientific validation of the safety and efficacy of the siddha drugs both individually as well as formulations have to be studied in a systematic and organized manner to complete in the international market. In this study ***Vilvathi Lehiyam*** was taken for analysis. It contains *Aegle marmelos* (Vilvam) *Terminalia chebula* (Kadukkai) *Terminalia belrica* (Thantrikkai) *Mesua ferrea* (Nagamaram), *Piper longum* (Milagu), *Zingiber officinale* (Enchi), *Emblica officinalis* (nellikai). Gingili Oil, Sugar, Honey. Anti-ulcer and Anti-inflammatory action *Vilvathi Lehiyam* was studied in this investigation

MATERIALS AND METHOD

In this study *Vilvathi Lehiyam* was brought from manufactured by Solaimalai, Paramakkudi. It was used for analysis, it was brought from local market in Mannargudi,

Anti-ulcer activity

Animals: Male albino rats of Wister strain approximately weighing 120-150g were used in this study. The animals were housed in spacious polypropylene cages bedded with rice husk. The animal room was well ventilated and maintained under standard experimental conditions

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(Temperature 27 ± 2º C and 12hour light/dark cycle) throughout the experimental period. All the animals were fed with standard pellet diet and water were provided *ad labium*. They were acclimatized to the environment for one week prior to experimental use. The animal feed composition is crude protein (22.3%), crude oil (4.01%), crude fibre (4.02%), Ash (8.02%) and sand silical (1.02%).

Ethanol-induced gastric ulcer: Animals were randomly divided into nine groups each of 6 rats. Gastric lesions were induced with ethanol (96%) at a dose 0.2ml/animal (Moraisa *et al.*, 2010). Group I served as control group received saline orally. Group II animals served as ulcero genic group received ethanol orally. Group III animals received Vilvathi *lehiyam* at a dose of 500 mg/kg orally. Group IV was orally administered 20mg/kg (ip) omeperazole as a standard drug. Forty-five minutes after treatment with *Vilvathi lehiyam* and standard drug, each animal was given orally 0.2mL of ethanol (96%). and they were sacrificed 60 min later. After 60 min, the rats were sacrificed and the stomach was removed. The gastric content was collected and centrifuge for 5 min at 2000 x g and the supernatant was separated. The volume, pH, and total acidity, free acidity of gastric fluid were determined.

The stomachs were excised and were examined for hemorrhagic lesions in glandular mucosa. Immediately after the animals were sacrificed, their stomachs were dissected out, cut along the greater curvature and the mucosa were rinsed with cold normal saline to remove blood contaminant, if any. The surface area of each lesion was counted and scored as described by Tan *et al.* (1996). The ulcer index for each rat was taken as the mean ulcer score.

Anti-inflammatory: activity was evaluated using the carrageenan induced rat paw oedema according to the technique of (Winter *et al.* 1962). after 12hrs fast rats were divided into five groups of six each. Group I served as control group received carrageen an only. Group II to IV animals received *Vilvathi lehiyam* at a dose of 100, 250 and 500 mg/kg orally. Group V was orally administered 2mg/kg Dexamethasone as a standard drug. The animals were pretreated with the *Vilvathi lehiyam* at half an hour before the administration of carrageen an. Acute inflammation was produced by the sub plantar administration of 0.1 ml of 1% carrageen an in normal saline in the right paw of the control and experimental rats. The paw was marked with in at the level of lateral mallows and immersed in mercury up to the mark and measured by mercury volume displacement methods. The paw volume was measured ½, 1, 1½ and 2 hours after injection of carrageen and to each group. The difference between the readings was taken as the volume of oedema and the percentage of anti-inflammatory activity was calculated. (Winter *et al*., 1962 and Ghosh, 2008).

Biochemical estimations

Determination of Gastric Juice volume and pH: The volume and pH of centrifuged gastric juice were measured by pipette and digital pH meter. The volume was expressed (Hirohashi *et al.,* 1993).

Determination of total and free acidity: The total and free acidity were determined by titrating with 0.01N NaOH using phenolphthalein and Topfer's reagent or methyl orange Pipette 1ml of filtered gastric contents into a small beaker, add 2 to 3 drops of Topfer's reagent or methyl orange and titrate with 0.01 N NaOH until all trace of the red colour disappears and the colour is yellowish orange. Note the volume of alkali added that indicate free acidity. Then add 2 or 3 drops of phenolphthalein and continue titrating until a definite red tinge reappears. Note the total volume of alkali added that indicate total acidity (Varley, 1988).

Statistical Analysis: Values were expressed as mean ± SD for six rats in each group and statistically significant differences between mean values were determined by one way analysis of variance (ANOVA) $p< 0.01$ was considered to be significant.

RESULTS AND DISCUSSION

In the present study ethanol *Vilvathi Lehiyam* was investigated for its antiulcer activity against Ethanol induced gastric ulceration in rats. The effect of orally administered *Vilvathi Lehiyam* on gastric damage induced by absolute produced characteristic lesions in the glandular portion of rat stomach which appeared as elongated bands of thick, black & dark red lesions. It was observed that increase the ulcer lesion in ulcer control rats. Significant reduction in ulcer lesion was observed in treatment with *Vilvathi Lehiyam* (Gupta *et al.,* 2007). Ethanol has shown significant protection index of inhibition was 83.73 % with the dose of 500 kg/mg respectively in comparison to control, Omeprazole as reference standard drug was reduction of ulcer 81.40 %. (Table1 and 2). Gastric ulcers result from an imbalance between aggressive factors and the maintenance of the mucosal integrity through the endogenous defence mechanism. The role of free radicals is also reported in the indication of ulcers. Prostaglandins

(PG) offer protection to duodenum through both increases in mucosal resistance as well as decrease in aggressive factors, mainly acid and Pepsin. Ethanol induced gastric ulcers have been widely used for the evaluation of gastro protective activity. Ethanol is metabolized in the body and releases superoxide anion and hydro proxy free radicals. The incidence of ethanol induced ulcers is predominant in the glandular part of stomach.

Ethanol induced gastric lesion formation may be due to stasis in gastric blood flow which contributes to the development of the haemorrhage and necrotic aspects of tissue injury. Alcohol rapidly penetrates the gastric mucosa apparently causing cell and plasma membrane damage leading to increased intra cellular membrane permeability to sodium and water. The massive intracellular accumulation of calcium represents a major step in the pathogenesis of gastric mucosal injury. This leads to cell death and exfoliation in the surface epithelium (Gupta *et al.,* 2003). The increase in volume in the ulcer control rats is undoubtedly due to increase production of hydrochloric acid as evident from the total acidity and decrease pH value of gastric juice. In the present study, the decrease in volume of the gastric juice and concomitant decrease in the acidity and increase in pH proving the antiulcer activity of *Vilvathi Lehiyam* and this result complements the earlier findings reported by Gurbuz *et al.,* (2000). Further evidenced by the reduced the oedema formation and epithelial lifting were observed in morphometric study.

The Anti-inflammatory activity of *Vilvathi Lehiyam* against carrageen induced paw oedema has been showed in Table 2 and the result were comparable to that of reference drug dexamethasone. The *Vilvathi Lehiyam* showed maximum inhibition of 20.24%, 54.57% and 70.72% at the dose of 100, 250 and 500 mg/kg body weight respectively after 2 hours of the *Vilvathi Lehiyam* treatment against carrageen an induced paw oedema whereas the reference drug produced 72.51% inhibition for standard respectively. Carrageen an induced hind paw oedema is the standard experimental model of acute inflammation. Carrageen an is the phlogiston agent of choice for testing anti-inflammatory drugs as it is not known to be antigenic and is devoid of apparent systemic effects. Moreover, the experimental model exhibits a high degree of reproducibility (Winter *et al*., 1962). Carrageen an induced oedema is a biphasic response. The first Phase is mediated through the release of histamine, serotonin and kinas whereas the second phase is related to the release of prostaglandin and slow reacting substances which peek at 2hr (Vinegar *et al.,* 1969). It has been reported that the second phase of oedema is sensitive to drugs like dexamethasone.

The use of study on *Vilvathi Lehiyam* reduced ulcer incidence, when compared to the control as evidenced by decrease in ulcer. The observed gastro protection is possibly mediated to a major extent by an gastric mucosal secretion mechanism as the *Vilvathi Lehiyam* were able to restore the increased volume, acidity and depleted p^H by ethanol almost towards normal levels seen in control.

The potential anti-inflammatory of *Vilvathi Lehiyam* is due to the presence of therapeutic phytochemicals. *Vilvathi Lehiyam intake* protects G.I tract and on treatment with less side effects, was noted. It is less costly, affordable and more effective in the treatment of different types of gastric diseases. The mechanism behind the action of *Vilvathi Lehiyam* was to be analysed in future studies. On the basis of the results of this study, it evenly indicates that *Vilvathi Lehiyam* possesses anti-ulcer and anti-inflammatory activity.

Acknowledgment: We express our gratitude to Dr. V. Divaharan, Secretary S.T.E.T Women's College, Mannargudi, for his support to this research work.

Table 1: Effect of *Vilvathi Lehiyam* on pH, volume Total acidity, Free acidity, Ulcer lesion, % of Ulcer protection in gastric juice and ulcer score of Normal and Experimental rats

Parameters	Volume (ml)	pH	Total acidity	Free acidity	Ulcer lesion	% of Ulcer protection
Group I	0.73 ± 0.08	3.45± 0.81	206± 10.1	183±7.23	1 ± 0.7	-
Group II	1.16± 0.08[a]	1.78± 0.28[a]	303± 18.5[a]	281± 14[a]	2 ± 0.84[a]	-
Group III	0.55± 0.10*	3.81± 0.74*	211± 11*	168± 6.57*	3 ± 0.21*	83.73 ± 5.86*
Group IV	0.51± 0.09*	4.23± 0.47*	181± 9.1*	168± 6.54*	5 ± 0.35*	81.40 ± 5.69*

Values were expressed as mean ± SD for six rats in each group.

[a]Significantly different from Group I (P< 0.01); * Significantly different Group II (P< 0.01)

Table 2: Shows the % of *Vilvathi Lehiyam* on carrageenan induced paw oedema

S.No	Doses	½ hour	1 hour	1 ½ hour	2 hour
1	Group I (%) (Control)	-	-	-	-
2	Group II (%) (100 mg/ml)	24.51 ± 2.20	24.10 ±2.16	22.40 ± 2.01	20.40 ± 1.82
3	Group III (%) (250 mg/ml)	33.95± 3.05	40.14 ± 3.16	53.99 ± 4.85	54.57 ± 4.91
4	Group IV(%) (500 mg/ml)	44.27 ± 3.98	55.65 ±5.0	64.89 ± 5.84	70.72 ± 4.91
5	Group V (%) (Standard) (2mg/kg Dexamethasone)	47.80 ± 4.30	51.87 ± 4.64	65.89 ± 5.87	72.51 ± 6.52

Values were expressed in % of inhibition as mean ± SD for six rats.

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