EVALUATION OF TRADITIONAL HERBAL FORMULATION IN EXPERIMENTALLY INDUCED GASTRIC ULCERS IN RATS.

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Summary

This formulation was selected to exploit the medicinal plant potential for the treatment and management of ulcers. Since the formulation is effectively used for the treatment of ulcers in human beings. Formulation contains powdered seeds of *Solanum niagram*, *Physalis alkekengi*. *Pistacia integerimmia* and *Tribulus terrestris*. The anti-ulcerogenic activity has been performed in Indomethacin induced ulcers model and Pylorus ligation ulcer model in rats.The ethanolic extract of Traditional Herbal Formulation in the dose of 300 mg/kg body weight showed significant protective effect in both the models. The results revealed significant activity when compared to control and standard groups.

Introduction

In the current years, peptic ulcer is one of the commonest diseases of the alimentary tract. It affects particularly the working years of a patient's life and its social implications are therefore considerable and this disease is a perennial problem encountered by the clinicians around the world. In the Western Hemisphere, like in the United States about 5-10% of the population develops peptic ulcer diseases.[1] The peptic ulcer refers to an ulcer found in the lower and of the esophagus, the stomach and the duodenum, in the small intestine after surgical anatomosis to the stomach, or rarely at the junction of a Meckel's divertificulum with the small intestine. Although the immediate cause of peptic ulceration's is digestion of mucosa by acid and pepsin of the gastric juice, the sequence of events that leads to the development of the ulcer are unknown.[2]

Theoretically, peptic ulceration may be caused due to either increase in gastric ulceration or if the resistance of the mucosa is reduced. It is convenient to consider the several factors believed to be involved in the development of chronic peptic ulcers according to how they may alter either side of the balance. [3]

An ulcer occurring in the stomach and duodenum may be acute or chronic, the difference being that a chronic ulcer penetrates the muscularis mucosae, where as an acute ulcer or erosion doesn't. Chronic ulcers occur with remarkable regularity in certain sites; in the stomach on the lesser curvature just above the angulus or less frequently at or near the pylorus on the anterior or posterior wall. Acute lesions are frequently multiple, and are less regularly distributed. Benign ulcers occur only rarely on the greater curvature or on the anterior wall of the stomach.

There are three objectives in the treatment or management of peptic ulcer, namely the alleviation of symptoms, the healing of the ulcer and the prevention of its recurrence. A number of **Ayurvedic formulations** have been advocated in traditional system of medicines to overcome above disorders and diseases. [4]
In recent days, there has been a large volume of work aimed at scientific validation of efficiency of herbal drugs used in the traditional systems. Among those one of the above formulation is widely used for the treatment and management of ulcers.

Materials and Methods

Formulated by:
This formulation was a gift sample from ***Dr. M.A. Samadkhan*** G.C.U.M, G.C.I.M. Meraj clinic, Hyderabad, Andhrapradesh, India. Herbal drug formulation contains,

Scientific name	English name	Parts used
Solanum nigram	Black night shade	Seed
Physalis alkekengi	Alkekengi	Seed
Pistacia integerrima	Pistacia galls	Seed
Tribulus terrestris	Gokhru	Seed

Experimental Studies

Extraction:
150gm of the powder were extracted with 95% ethanol in a soxhelt extractor and concentrated under reduced pressure in a rotary flash evaporator. The residue was dried in a desicator over sodium sulphite. The percentage yield of ethanol extract was found to be 6.72% w/w.

Preparation of the dose:
The suspension of the ethanol extract of formulation was prepared using 2% gum acacia.

Acute toxicity study:
Acute toxicity studies are designed to determine the nature and extent of untoward effects, which might follow the administration of a single dose within 24 hr. No drug is absolutely safe and for any new drug toxicity studies are necessary to fix the effective dose. Acute toxicity study of ethanolic extract of the formulation was carried out by **"UP and DOWN"** or **"STAIRCASE"** method. Healthy albino mice of either sex weighing between (20-25g) maintained under standard conditions were selected, which were fed with pellet diet and water provided *ad libitum*. The ethanolic extract cause mortality when administered up to a dose of 3000 mg/kg b. w and accordingly dose was fixed as 300 mg/kg b. w.[5]

Indomethacin Induced ulcers model:
Wistar rats of either sex were deprived of food for 24 hr before the experiment. The test drug or reference drug or the control vehicle was administered in two doses at an interval of 15 hr. Indomethacin (10 mg/kg b. w) was administered by gavage needle in two doses after thirty minutes of administration of each dose of test compound. 1hr after the second dose of Indomethacin all rat were sacrificed.

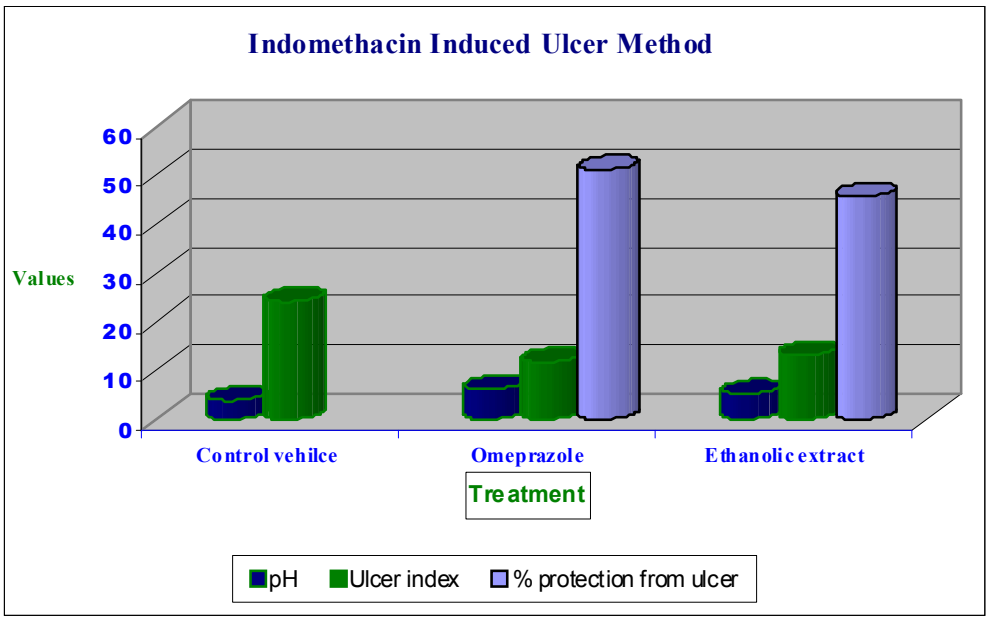
The numbers of ulcer spots in the glandular portion of the stomach were counted in both control and extract treated animals and the ulcer index was calculated.[6]

Table: 01 Indomethacin induced gastric mucosal damage in experimental rats.

S.NO	Design of Treatment	Dose (mg/kg b. w)	P^H	Ulcer Index	% Protection from Ulcer
01	Control	Vehicle	3.95±0.031	24.17±0.21	-
02	Omeprazole	8	6.17±0.019*	11.84±0.84*	51.1
03	Ethanolic extract	300	5.30±0.054**	13.16±1.36**	45.55

The results are expressed as Mean±SEM. Statistical significance on comparison with normal control are indicated by *marks. Control vs Standard *P<0.0001, Control vs Ethanolic extract **p<0.0001

Fig.01 Histogram showing Indomethacin induced ulcer model



Pylorus ligation ulcer model in rats:

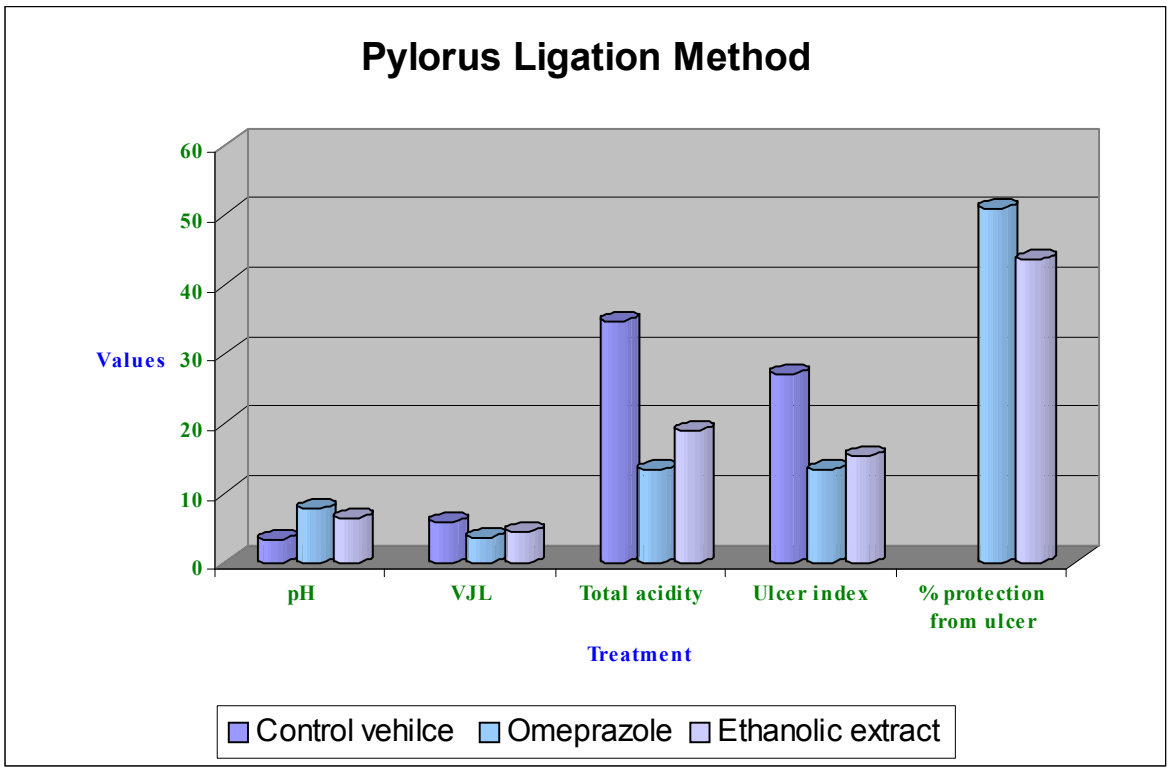
Albino rats were fasted in individual cages for 24 hrs. Extract or reference drug or control vehicle was administered 30 mins prior to pyloric ligation under light ether anesthesia, the abdomen was opened and the pylorus was ligated. The abdomen was then sutured. At the end of 4 hrs after ligation, the animals were sacrificed with excess of anesthetic ether, and the stomach was dissected out. Gastric juice was collected and it's volume, pH, free and total acidity was measured. The glandular portion was then exposed and examined for ulceration. Ulcer index was determined.[7-9]

Table no: 02 Effect of Ethanolic extract of formulation on pylorus ligation model.

S.No	Design of treatment	Dose mg/kg b. w.	pH	VGJ (ML/100mg)	Total acidity (mEq/L)	Ulcer index	% Protection
01	Control	Vehicle	3.36±0.12	5.92±0.07	34.67±0.61	27.17±0.210	--
02	Omeprazole	8	7.68±0.02	3.68±0.07*	13.33±0.42*	13.34±0.63*	50.90
03	Ethanolic extract	300	6.37±0.07	4.40±0.4**	19.00±0.68**	15.33±0.30**	43.57

The results are expressed as Mean±SEM. Statistical significance on comparison with normal Control are indicated by *marks. Control vs Standard *P<0.0001. Control vs Ethanolic extract **p<0.0001.

Fig.02 Histogram showing Pylorus ligation method



Results

Preliminary phytochemical screening of herbal formulation reveled the presence of glycosides, steroids, carbohydrates, phytosterols, saponins and tannins. The dose ethanolic extract determined by "Up and Down" method was found to be 300 mg/kg body weight in mice. The ulcers were induced in rats by Indomethacin and Pylorus ligation model.[10] Controlled animals showed increase in ulcers, after treatment with formulation drastic decrease in the ulcers was observed. The percentage protection of ulcers when compared with controlled animals shows 50%.

Discussion

Gastric ulceration is related with integrity of mucosal layer and is mainly dependent on the arachidonic acid metabolism. The ulcerogens like NSAID's induced the effect by interfering with the cyclo-oxygenase pathway. Indomethacin can affect the mucosal blood flow; platelet thrombi and can increase the production of free radicals leading to increased lipid peroxidation, which can damage the cell and cell membranes.[11] while pylorus ligation increases the presence of acid and pepsin in the stomach. The ethanolic extract of formulation is reported to contain flavonoids like Quercetin. Phytopharmacological studies of flavonoids have opened new vistas in ulcer research. Quercetin is reported to have free radical scavenging and dose dependent anti-ulcer activity.[12] Thus it can be concluded that the antiulcerogenic activity of the formulation may be due to cytoprotective and healing property of flavonoids. Determination of exact mode and screening for various models is subjected of our further research interest.

References

1. Gerard Tortora J., Sandra Reynolds Grabowski, 1996, "Principles of Anatomy and Physiology", Hapercollins College Publishers, 8th ed, 847-870.
2. Guytor M.D., "Text Book of Medical Physiology", Prism Books (Pvt) Ltd, B'lore., India, 2nd ed, 273-300.
3. Kiritikar K.R., Basu B.D., Indian Medicinal Plants, International Book Distributors, Dehradun, India, 1995, Text Vol-II, 1385-1386.
4. Yoganarasimhan Y.N., "Medicinal Plants of India", Interline Publishing Pvt., Ltd., 1996, Vol-I, 460.
5. Jckurian., "Plants That Heal", Oriental watchman Publishing House, 1st ed., Pune, India. 1995, 296-297.
6. The Wealth of India, A Dictionary of Indian Raw Materials and Industrial products, Publications and Information Directorate, CSIR, New Delhi, 1976, Vol-X(SP-W), 109-112.
7. J.Singh, A.K.Singh, S.P.S. Khanuja, 2003, "Medicinal Plants India's Opportunity", February-April 2, 59-63.
8. Vogal, Gerjard H, Wolfgand H Vogal, 1996, "Drug Discovery and Evolution", New York, Springer-Verlag, Berlin, Heidel Berg, 35, 37, 74-78pp.
9. Rudraprabhu V.Sauadi, 1995, "Phytochemical and Pharmacological Investigations of Achyranthes aspera Linn", M. Pharm Dissertation submitted to Karnataka University, Dharwad, 44-54pp.
10. John D.Baur(Ed), 1982, "Acid base regulation laboratory methods". The C.V.Mosby Company St Louis Toronto, London.
11. S.B.P.Board Consulatants and Engineers, 2002, "S.B.P. Hand Book of Ayurvedic Medicines and Herbal Formulations", S.B.P. Publication Division, New Delhi, 4-7.
12. A.U.Ahmed ., Z.Ali., S.R. Hussaiaini., F.Iqbal., M.Zahid., M.Abbas., 1999, " Flavonoids of *Tephrosia purpurea*", Fitoterpia, 70, 443-445.