Biochemical studies evaluating the antiulcerogenic potential of UL-409, a herbal drug formulation on experimental peptic ulcer in rats

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This study was designed to evaluate the cytoprotective effect of UL-409 in experimental peptic ulcer. Male Wistar rats orally intubated with HCl–ethanol mixture for the induction of ulcer were pretreated with UL-409, orally for 30 days. The volume and total acidity of the gastric juice were decreased in pretreated rats. The mucosal protein, mucosal sugars and sialic acid levels which were decreased in ulcer group were maintained to near normalcy after the pretreatment.

AGGRESSIVE acid/pepsin secretion and lowered mucosal defense are critical factors involved in the pathogenesis of peptic ulcer. Defect in gastro-duodenal defense mechanisms include alteration in prostaglandin biosynthesis and their release, mucous and bicarbonate secretion and the luminal hydrophobic layer. Correction of possible deficiency in mucosal defense/resistance may be a useful and practical principle in management of patients with peptic ulcer[1].

There are many reports from different studies carried out by different authors evaluating the various pharmacological properties of herbs which are the ingredients of the ayurvedic formulation, UL-409 (Table 1). *Tinospora cordifolia* has been reported to cure various stomach disorders when used in combination with herbs such as *Ailanthus triphysa*, *Helictrea erora*[2]. It is also said to protect against stress[3]. *Saussurea lappa* has been used in the treatment of osteoarthritis[4]. The fruit decoction of *Emblica officianalis*[5] and *Foeniculum vulgaris*[6] has been used to combat hypertension. Aqueous extract of *Emblica officinalis* was found to be a potent inhibitor of lipid peroxide formation[7]. Aqueous extract of *Santalum album* is said to cure vaginal infections[8]. Roots and leaves of *Aegle marmelos* were reported to have versatile medical value[9]. *Rosa damascena* was found to have hepatoprotective potential[10]. *Glycorrhiza glabra* has been studied to evaluate its anticancer effect[11].

Hence an attempt has been made to evaluate antiulcerogenic potential of the herbal formulation UL-409 in rats.

The herbal drug formulation named UL-409 was obtained from Himalaya Drug Company, Bangalore.

This report does not constitute an endorsement of the efficacy of the herbal drug formulation UL-409.

Table 1. Composition of UL-409

ingredients	Parts used	Percentage
Glycorhiza glabra	Root	18
Benincasa hispida	Fruit juice/pulp	16
Tinospora cordifolia	Stem	11
Saussurea lappa	Root	11
Emblica officinalis	Fruit	11
Santalum album	Stem	18
Aegle marmelos	Fruit	7
Jasad Bhasma	(Ayurvedic mineral preparation)	4
Zaharmora Bhasma	(Ayurvedic mineral preparation)	4
Processed in		
Aloe vera	Leaves	
Foeniculum vulgaris	Seed	
Rosa demascena	Floor petals	

Male Wistar rats weighing 150–200 g obtained from Fredrick Institute of Plant Protection and Toxicology, Padappai, Madras were maintained on a commercial pelleted food (M/s Hindustan Lever Foods, Bangalore) and water *ad libitum*.

The experimental animals were divided into three groups with six animals in each group. Group I served as control. Group II animals were ulcer induced. Group III animals were pretreated with UL-409 (600 mg/kg body wt/day) orally in aqueous suspension. Each animal received 1.0 ml of aqueous suspension for 30 days before the induction of ulcer. After 30 days of pretreatment, all the three groups were deprived of food for 24 h. Group II and Group III animals were intubated with 1.5 ml HCl-ethanol mixture containing 150 mM solution in 70% v/v ethanol (1:1) to induce peptic ulcer[12]. Ulcer would be developed in 1 h. After 1 h, the rats were subjected to mild ether anaesthesia and the abdomen was opened through a midline incision. The pylorus was secured and ligated with silk sutures after which the wound was closed and the animals were allowed to recover from anesthesia. Gastric juice was collected for a period of 4 h (ref. 13). Water was withheld after anesthetization. After 4 h, the rats were killed by cervical decapitation and the stomach was removed after clamping the oesophagus. The gastric juice was collected, centrifuged and the volume was noted. The total acidity was determined by titrating with 0.1 N sodium hydroxide using phenolphthalein as an indicator. The mucosal tissue was scraped from the stomach and used to estimate the activity of pepsin[14], protein[15], hexose and hexosamine[16] and sialic acid[17]. The values were expressed as mean ± S.D. The statistical significance difference as analysed by Student's *t* test, *p* values were expressed. An analysis of variance is also indicated.

Experimental peptic ulcer may be assessed on the basis of number of gastric mucosal lesions. Ethanol causes hemorrhagic and necrotic tissue injury[18] and the

Table 2. Volume and total acidity of gastric juice, activity of pepsin, levels of protein and glycoprotein in gastric mucosa of the three experimental groups

Parameter	Group I	Group II	Group III	ANOVA (*f* value)
Gastric juice				
Volume ml/100 g	3.35 ± 0.11	7.50 ± 0.24^{c}	3.50 ± 0.09^{z}	36.80
Total acidity μ Eq/4 h	180.0 ± 7.02	362.4 ± 9.06^{c}	178.2 ± 6.21^{z}	25.43
Gastric mucosa				
Pepsin μmol tyrosine liberated/4 h	648 ± 25.31	600 ± 20.42^{a}	630 ± 20.82^{x}	0.98
Protein mg/g wet tissue	25.8 ± 0.78	11.8 ± 0.30^{c}	24.3 ± 0.67^{z}	64.30
Hexose mg/g wet tissue	15.6 ± 0.60	7.60 ± 0.30^{c}	14.3 ± 0.54^{z}	40.67
Hexosamine mg/g wet tissue	9.32 ± 0.25	4.33 ± 0.14^{c}	8.99 ± 0.29^{z}	82.99
Sialic acid mg/g wet tissue	2.53 ± 0.06	0.74 ± 0.02^{c}	2.12 ± 0.07^{z}	15.62

$n = 6$, Values are expressed as the mean ± SEM.
$^{c}p < 0.001$ Group II vs Group I.
$^{z}p < 0.001$ Group III vs Group II.
$^{a}p < 0.1$ Group II vs Group I.
$^{x}p < 0.1$ Group III vs Group II.

number of lesions are indicative of severity[19]. Our previous study with UL-409 has showed that the drug decreases the number of ulcerative lesions induced experimentally[20], suggesting its combating action towards ulcer.

Our present study has revealed an increase in volume and acidity of the gastric juice of Group II rats. This may be due to the increased production of hydrochloric acid. Gastric mucosal barrier provided by gastric epithelial cells, luminal surfaces and intercellular tight junction provide an almost completely impermeable gastric mucosal barrier. This barrier can be interrupted by bile acids, salicylates, ethanol and acid. This results in the back diffusion of H^{+} ions from lumen into gastric tissues which cause release of histamine from mast cells, further secretion of acid and ulceration[21]. Since acid is one of the aggressive factors in the production of ulcer, acid inhibition accelerates ulcer healing[22]. The decrease in volume of the gastric juice with simultaneous decrease in acidity in Group III suggests the effect of UL-409 in ulcer healing.

Our results showed no significant change in the activity of mucosal pepsin in both Group II and Group III rate when compared to the control rats. Ethanol has been reported to inactivate pepsinogen or affect its activation to pepsin and it also alters the activity of pepsin[23]. The lack of effect on the activity of pepsin in Group II rats could be due to the effect of ethanol used for the induction of ulcer which correlates with the above report. There is no significant change in the peptic activity in Group III when compared with Group II.

From Table 2 it is also inferred that the levels of gastric mucosal protein and mucosal sugars of Group II rats were decreased well when compared with that of Group I rats and Group III rats. It has been reported that ulcerative lesions of gastrointestinal tract may be associated with increased loss of protein[24] and alcohol increases the loss of protein in the presence of hydrochloric acid[25]. These reports support our observation of decreased gastric mucosal protein in Group II. But maintenance of near-normal levels of protein in Group III rats suggested the cytoprotective nature of UL-409 by virtue of its ability to reduce the mucosal lesions[20].

Mucus is said to protect the gastrointestinal tract from infective, chemical and physical insults. The glycoprotein component of mucus which is responsible for the characteristic viscous gel-forming property is believed to be important for the functional role of mucus[26]. This mucosal barrier lessens the stomach wall friction during contraction, improves the buffering acid and acts as an effective barrier to back diffusion of hydrogen ions[27]. The carbohydrate side chains of mucosal glycoprotein comprise over 80% by weight of the glycoprotein molecule. Each chain consists of sugars such as fucose, hexose, amino sugars and sialic acid[26]. Our observation of decrease in levels of gastric mucosal sugars and sialic acid in Group II correlates with the report which showed rapid loss of mucosal glycoprotein when exposed to ethanol[28]. Such a decrease of mucosal hexose, hexosamine and sialic acid was not encountered in Group III after the prior treatment with UL-409. By preventing the protective mucus from being degraded by HCl–ethanol insult, UL-409 establishes itself as a cytoprotective drug.

It has been concluded from the above observations that since UL-409 regulates acidity and protects the mucosal barrier, it could be categorized as an antiulcerogenic drug.

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Outbreak of falciparum malaria in submerged villages of Narayanganj PHC, district Mandla due to Narmada Irrigation Project, Central India (Madhya Pradesh)

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On receipt of a report about high prevalence of malaria and deaths in submerged villages of Narayanganj Primary Health Centre (PHC) of district Mandla, Central India (Madhya Pradesh) due to Bargidam in October–November 1996, an investigation into the causes was carried out in 20 villages. Blood smears from fever cases and contacts of deceased patients were collected. Slide positivity rate was over 70%, of which more than 90% was *Plasmodium falciparum*. Mass blood surveys of infant and pregnant women revealed 39% and 62% parasite prevalence rate respectively. More than 80% children (2–9 yrs) had enlarged spleen. Such high malaria prevalence appeared to be maintained by *Anopheles culicifacies* and *An. fluviatilis* which could not be suppressed by intensive surveillance, prompt radical treatment with 1500 mg chloroquine and 45 mg primaquine and two rounds of special focal spray with DDT in October 1996 and January 1997. There is, therefore, an urgent need to develop suitable malaria control strategy by replacement of insecticides in conjunction with prompt and effective radical treatment.

NARMADA, an inter-state monsoon river, is the fifth largest river of India. Since bulk of the runoff water is generated during the monsoon months (June–September) and that too in a few spells of intense and heavy rainfall, conservation of water is important to meet the ever-growing water needs of the society, as more than 80 lakh population of Madhya Pradesh (Central India) lives along Narmada river and its tributaries. To utilize Narmada waters, it is proposed to construct a series of projects for irrigation and power generation. Bargi is the first completed hydro-electric-cum-irrigation project on river Narmada in Jabalpur district in the series. The dam is 826.9 m in length and 68.9 m in height. The water reservoir is 14556 km^2 and command area is 2.544 lakh ha. The construction of this multipurpose dam (1974–1988) has resulted in the submergence of 162 villages of three districts, viz. Jabalpur, Mandla and Seoni. Water has been stored in this dam to full capacity since 1990. In October–November 1996, there were reports of 109 deaths due to clinical malaria in some submergence villages of district Mandla. An investigation into the cause of deaths was carried out in 20 villages of Narayanganj Primary Health Centre (PHC) from November 1996 to March 1997. Results of this study are presented in this paper.

The whole region is a cobweb of peaks of the Maikal hills, which forms a broad plateaus of about 2200 km^2, mostly forest inhabited by tribals. Since the region is a hilly-tract, its elevation is very irregular which renders the whole area highly undulating. The region is best known for the magnificent forests of sal (*Shorea robusta*) and teak (*Tectona grandis*) as seen in Figure 1. Study villages are sandwiched between forest and dam reservoir as shown in Figure 2. Wild life was rare but lion, tiger, bison, etc are still present in the jungle and seen occasionally in villages. These villages are generally located on the slopes of hillocks or on hill