Evaluation of gastro-protective effect of Pep-Up Tablet and Pep-Up Syrup on water immersion plus restraint stress-induced gastric ulcer in rats

Hardik Soni, Sweta Patel, Arindam Paul, Ghanshyam Patel

ABSTRACT

Stress is an adaptive physiological response to disruption of homeostasis. Stress-induced gastric ulceration is a typical example of stress-associated organ injuries. The present study was carried out to evaluate gastroprotective effect of Pep-Up Tablet and Syrup, proprietary Ayurvedic formulations, on water immersion plus restraint stress-induced gastric ulcer in rats. Both test drugs were studied for their effect on ulcer index, gastric wall mucus content, lipid peroxidation level in stomach tissue and tissue anti-oxidant parameters like superoxide dismutase, reduced glutathione and catalase activity. Pre-treatment of Pep-Up Tablet and Syrup showed significant protection against loss of gastric wall mucus content and ulceration. They also significantly prevented depletion of tissue anti-oxidant parameters in comparison of disease control group. From the present study, it can be concluded that Pep-Up Tablet and Syrup are having significant anti-oxidant property which enhanced the stability of gastric mucosa against the damage caused by water immersion plus restraint induced stress. The gastro-protective effect of Pep-Up Tablet and Syrup on stress induced gastric ulceration supports their clinical application to prevent acid-peptic disorders.

Keywords: Pep-Up Tablet, Pep-Up Syrup, ulcer index, gastric wall mucus content, anti-oxidant property, gastro-protective effect

1. Introduction

Stress is an adaptive physiological response to disruption of body’s internal homeostasis. Excessive stress can induce organ injury or contribute to diseases, such as gastric ulcers, hypertension, diabetes, and cancer. Stress-induced gastric ulceration is a typical example of stress-associated organ injuries [1]. Acute gastric mucosal injury is a serious clinical problem worldwide [2]. Since the development of gastric lesions during restraint stress was significantly enhanced by exposure to water immersion. The rise in acid secretion may be an important aggravating factor in the process of ulceration during water immersion [1]. The use of herbal medicine has become increasingly popular worldwide and medicinal plants are believed to be an important source of new chemical substances with potential therapeutic effects [3]. Recently, it has also been reported that many traditional Asian herbal remedies act on gastro-intestinal diseases [4]. Pep-Up Tablet and Syrup are Ayurvedic proprietary formulations manufactured and marketed by Vasu Healthcare Pvt. Ltd., Vadodara.


Majority of ingredients of Pep-Up Tablet and Syrup are well reported in Ayurvedic texts and scientific research publications for gastroprotective effect. However, no such evidence was found which proves efficacy of their combination. The present study was aimed to investigate
gastro-protective effect of Pep-Up Tablet and Syrup on water immersion plus restraint stress-induced gastric ulcer in rats.

2. Materials and methods

2.1 Experimental animals

Adult wistar albino rats of either sex weighing 200-250 g were used and acclimatized to the experimental room having ambient temperature (23±2 °C), controlled humidity (55±5%) conditions, and 12 h light and dark cycle. Animals were caged in polypropylene cages with maximum of three animals per cage. The rats were fed with Chakkan food pellets and water ad libitum. Study was conducted after obtaining approval by Institutional Animal Ethical Committee (IAEC) of Bababur Institute of Pharmacy (M.Pharm Sem-IV/12-13/11) as per the guidance of Committee for the Purpose of Control and Supervision of Experiments on Animal (CPCSEA), Ministry of Social Justice and Empowerment, Government of India.

2.2 Administration of test drug and dosage

The test drugs (Pep-Up Tablet and Syrup) were received from Vasu Healthcare Pvt. Ltd., Vadodara, Gujarat, India. Pep-Up Tablet was administered orally in form of suspension by mixing with vehicle solution of 1% sodium carboxy methyl cellulose (Na-CMC). Pep-Up Syrup was administered orally as it is. Dose of the test drugs were fixed by extrapolating the human dose to laboratory animals, based on body surface area ratio as per the table of Paget and Barnes [33]. Experimental dose of Pep-Up Tablet was 200 mg/kg/day (p.o.) and Pep-Up Syrup was 3 mL/kg/day (p.o.).

2.3 Experimental groups

The selected animals were divided into four groups as each group consisted of six animals.

Group 1: (Normal control): received distilled water as vehicle

Group 2: (Disease control): received distilled water as vehicle and exposed to water immersion plus restraint stress

Group 3: (Pep-Up Tablet treated): received Pep-Up Tablet (200 mg/kg/day, p.o.) and exposed to water immersion plus restraint stress

Group 4: (Pep-Up Syrup treated): received Pep-Up Syrup (3 mL/kg/day, p.o.) and exposed to water immersion plus restraint stress

2.4 Water immersion plus restraint stress-induced gastric ulcer

The selected animals were divided in to four groups as mentioned above. Pep-Up Tablet and Syrup were administered for 7 days through oral route in group 3 and 4 respectively. The method described by Takagi and Okabe [34] was employed with slight modification. After sixth day treatment rats were fasted for 18 h, care being taken to avoid coprophagy. The rats were immobilized in a restrainer and subsequently they were immersed in water up to xiphoid process for 7 h. The temperature of the water was maintained at 24±5 °C. Drugs were given orally 2 h prior to the restraint procedure. After 7 h of immobilization and water immersion the animals were taken out and killed with high-dose anesthetic ether. The stomach was removed and ulcer index with its severity [35] was determined. The samples of stomach tissue were analyzed to determine gastric wall mucus content, reduced glutathione, superoxide dismutase and catalase activity and lipid peroxidation (MDA) [40].

2.5 Statistical analysis

The data were expressed as mean ± standard error of mean (SEM) for six rats per experimental group. Different groups were compared with analysis of variance (ANOVA) followed by post hoc Tukey’s test. A P ≤ 0.05 was considered as statistically significant.

3. Results and discussion

The experimental stress ulcer may be considered equivalent to clinical stress ulcer. An acute gastric hemorrhagic lesion in the glandular stomach characterizes a stress ulcer [1]. In present study, exposure to water immersion plus restraint stress developed gastric ulceration and decreased content of gastric wall mucus. Pretreatment of Pep-Up Tablet and Syrup provided significant gastroprotection against water immersion plus restraint stress-induced gastric ulcer and mucosal damage. Pep-Up Syrup showed more significant protection against gastric ulcer and mucosal damage with respect to Pep-Up Tablet (Table 1).

Table 1: Effect of Pep-Up Tablet and Syrup on ulcer index and gastric wall mucus content

<table>
<thead>
<tr>
<th>Group</th>
<th>Ulcer index</th>
<th>Gastric wall mucus content (µg/mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal control</td>
<td>0.00 ± 0.00</td>
<td>969.47 ± 09.14</td>
</tr>
<tr>
<td>Disease control</td>
<td>1.26 ± 0.11###</td>
<td>631.23 ± 13.47 ##</td>
</tr>
<tr>
<td>Pep-Up Tablet treated</td>
<td>0.78 ± 0.05**</td>
<td>868.08 ± 14.80 **</td>
</tr>
<tr>
<td>Pep-Up Syrup treated</td>
<td>0.54 ± 0.05**</td>
<td>861.72 ± 11.73 **</td>
</tr>
</tbody>
</table>

All the values are expressed as mean ± SEM (n=6) in each group. **P < 0.01 when compared to disease control group; ###P < 0.01, ####P < 0.001 when compared to normal control group.

Anti-oxidant defensive system also plays vital role in recovery of damage that occurs due to free radicals. It converts free radicals into non-toxic compounds. Exposure to water immersion plus restraint stress significantly reduced anti-oxidant parameters of stomach tissue viz. reduced glutathione, superoxide dismutase and catalase activity. Pre-treatment of Pep-Up Tablet and Syrup both showed significant protection against depletion of tissue anti-oxidants in comparison of disease control group however, Pep-Up Syrup was little superior as compared to Pep-Up Tablet (Table 2).
Table 2: Effect of Pep-Up Tablet and Syrup on anti-oxidant biochemical parameters of stomach tissue

<table>
<thead>
<tr>
<th>Group</th>
<th>Reduced glutathione (µg/g of tissue)</th>
<th>Superoxide dismutase (µg/g of tissue)</th>
<th>Catalase enzyme activity (µmole H₂O₂ consumed/min/g of tissue)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal control</td>
<td>893.82 ± 24.83</td>
<td>400.00 ± 28.58</td>
<td>910.99 ± 79.92</td>
</tr>
<tr>
<td>Disease control</td>
<td>496.76 ± 16.07**</td>
<td>176.96 ± 17.64##</td>
<td>407.74 ± 35.46###</td>
</tr>
<tr>
<td>Pep-Up Tablet treated</td>
<td>729.61 ± 18.81*</td>
<td>344.32 ± 17.64**</td>
<td>815.50 ± 67.38**</td>
</tr>
<tr>
<td>Pep-Up Syrup treated</td>
<td>814.41 ± 17.07**</td>
<td>358.24 ± 13.56**</td>
<td>891.60 ± 61.74***</td>
</tr>
</tbody>
</table>

All the values are expressed as mean ± SEM (n=6) in each group. *P < 0.05, **P < 0.01, ***P < 0.001 when compared to disease control group; #P < 0.01, ##P < 0.001 when compared to normal control group.

The observed significant anti-oxidant property of Pep-Up Tablet and Syrup may be attributed to *Withania somnifera* (Ashwagandha) Root, [22] *Zingiber officinale* (Shunthi) Rhizome, [15, 16] *Emblica officinalis* (Amalaki) Fruit, [5-7] *Terminalia chebula* (Haritaki) Fruit, [8, 9] *Piper longum* (Pippali) Fruit [20, 21] and *Piper nigrum* (Kali mirch) Fruit [19]. All these ingredients are well reported for having anti-oxidant property.

As a matter of fact, free radicals also has important role in the pathogenesis of various tissue injury, including the digestive system. Oxygen and hydrogen derived free radicals like hydrogen peroxide, hydroxyl radicals can be responsible for mucosal damage [41]. Pre-treatment of Pep-Up Tablet and Syrup both significantly inhibited lipid peroxidation in comparison to disease control group which indicates their certain effect against free radicals generation and related damage (Table 3) however Pep-Up Syrup was found more effective in comparison to Pep-Up Tablet.

Table 3: Effect of Pep-Up Tablet and Syrup on lipid peroxidation level of stomach tissue

<table>
<thead>
<tr>
<th>Group</th>
<th>Lipid peroxidation (nmole/g of tissue)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal control</td>
<td>226.43 ± 13.77</td>
</tr>
<tr>
<td>Disease control</td>
<td>379.06 ± 19.26##</td>
</tr>
<tr>
<td>Pep-Up Tablet treated</td>
<td>308.23 ± 12.36*</td>
</tr>
<tr>
<td>Pep-Up Syrup treated</td>
<td>259.27 ± 24.72**</td>
</tr>
</tbody>
</table>

All the values are expressed as mean ± SEM (n=6) in each group. *P < 0.05, **P < 0.01 when compared to disease control group; #P < 0.01 when compared to normal control group.

4. Conclusion
From the present study, it can be concluded that Pep-Up Tablet and Syrup both are having significant anti-oxidant property which may be responsible for enhancing the stability of gastric mucosal tissues against damage caused by water immersion plus restraint induced stress. Promising gastro-protective effect of Pep-Up Tablet and Syrup on stress induced gastric ulceration was also found supporting to their acclaimed clinical application.

5. Acknowledgement
Authors are sincerely thankful to the management of Vasu Healthcare Pvt. Ltd. and Babaria Institute of Pharmacy for providing the necessary facilities for conducting the study.

6. References


