A Comparative Study on the Effects of *Lactobacillus Acidophilus*, Chlorella and Esomeprazole on Indomethacin-Induced Gastric Ulcer in Adult Albino Rats

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By

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Summary

Current treatment of gastric ulcer is associated with several side effects, which emphasizes the need for new therapeutic modifies. it is believed that the therapeutic outcome of *Lactobacillus Acidophilus* and Chlorella can be more effective than traditional lines of treatment.

This work aimed to study the protective effect of *Lactobacillus Acidophilus* and Chlorella in comparison to Esomeprazole on Indomethacin- induced gastric ulcer in adult male albino rats, monitored by histological, immunohistochemical and morphometric methods.

This study was carried out on twenty-five adult male albino rats, weighing 220-280 gm. Rats was randomly divided into five groups (5 rats each):

1- **Group I (Control):** Each rat received 1 ml distilled water for 11 days.
2- **Group II (Indomethacin only-treated group):** Each capsule (25 mg) was dissolved in 1 ml distilled water so, each rat received single oral dose (100 mg/kg, 1 ml/rat) of indomethacin, by the use of intragastric gavage tube on the 11^{th} day of beginning of the experiment. Rats were scarified 6 hours after indomethacin administration.
3- **Group III (Esomeprazole pre-treated group):** Each capsule (40 mg) was dissolved in 32 ml distilled water. So, each rat received daily oral dose of (5 mg/kg, 1 ml/rat) by the use of intragastric gavage tube for 10 consecutive days and 2 hours before INDO administration at the aforementioned dose and period in group II.

4- **Group IV (Chlorella pre-treated group):** 500 mg powder of chlorella was dissolved in 2 ml distilled water. So, each rat received daily oral dose of (2000 mg/kg, 2 ml/rat) by the use of intragastric gavage tube for 10 consecutive days and 2 hours before INDO administration at the aforementioned dose and period in group II.
5- **Group V (*Lactobacillus acidophilus* pre-treated group):** Each sachet (10 billion CFU) was dissolved in 1 ml distilled water. So, each rat received daily oral dose of (10 billion CFU /rat, 1 ml/rat) by the use of intragastric gavage tube for 10 consecutive days and 2 hours before INDO administration at the aforementioned dose and period in group II.

On the 11th day of the experiment rats were deprived of food but had free access to water 24 hours before ulcer induction. Gastric ulceration was induced by giving the rats a single oral dose of indomethacin after two hours of last dose of the used prophylactic drugs. Six hours after the INDO administration, animals of all groups were euthanized by cervical dislocation and stomach specimens were collected, washed by saline and opened along greater curvature. Stomach was placed with the mucosal surface directed upwards and was photographed with a digital camera of iPhone 14 pro, then ulcer index was counted and finally fixed immediately in 10% buffered formalin for histological and immunohistochemical studies.

Morphometric measurements were done to measure mean area percentage of PAS positive reaction, mean area percentage and optical density of NF-κB positive reaction and mucosal length. The results were then statistically analysed.

Gastric ulcer induced by indomethacin administration caused significant increase in the ulcer index as compared to the control rats. A significant decrease in the ulcer index in all treatment groups as compared to INDO group was observed.

Hematoxlin & Eosin stained fundic sections of INDO group showed a loss of organization of the fundic mucosa with cellular exfoliation. The surface columnar mucus-secreting cells were lost. There was disorganization of the normal architecture of the fundic glands. Other areas showed erosions of mucosa. There were apoptotic changes in parietal cells and basal chief cells. Haemorrhage and hemosiderin deposition also, were observed.

Examination of the fundic sections of ESP+INDO group showed restoration of the normal architecture of mucosa with residual area of erosions. Normal appearance of surface columnar mucus-secreting cells was revealed. Many parietal cells with small darkly stained nuclei and vacuolated cytoplasm were noticed. Relatively restored arrangement of bases of fundic glands with some parietal cells with small darkly stained nucleus and vacuolated cytoplasm and normal chief cell were noticed.

Examination of the fundic sections of Chlorella+INDO group showed normal regular arrangement of tubular fundic glands. Many parietal cells with eccentric small darkly stained nuclei and vacuolated cytoplasm were noticed. Restored arrangement of bases of fundic glands with normal appearance of chief cells and parietal cells with congestion of blood vessels in lamina propria was observed.

Examination of the fundic sections of LB+INDO group showed apparently normal regular arrangement of tubular fundic glands, apparently normal appearance of surface columnar mucus-secreting

cells and parietal cells. Restored arrangement of bases of fundic glands with normal appearance of parietal cells and chief cells were noted.

In PAS stained fundic sections of INDO group showed significant decrease in area percentage of PAS reaction as compared to control and other pre-treated groups. Chlorella+INDO group showed significant increase in area percentage of PAS reaction as compared to INDO and ESP+INDO groups. Moreover LB+INDO group showed significant increase in area percentage of PAS reaction as compared to INDO, ESP+INDO and Chlorella+INDO groups.

Regarding NF-κB immunoreaction INDO group showed significant increase in area percentage and optical denisty of NF-κB reaction as compared to control and other pre-treated groups. While ESP+INDO group showed significant decrease in area percentage and optical density of NF-κB reaction as compared to INDO group, but significant increase as compared to Chlorella+INDO and LB+ INDO groups. Moreover Chlorella+INDO group showed significant increase in area percentage and optical density of NF-κB reaction as compared to LB+ INDO group.

Regarding mucosal length there was a significant decrease in length in INDO group as compared to control and other pre-treated groups. There was a significant increase in mucosal length in Chlorella+INDO group as compared to the control group, but no significant change was detected in mucosal length in both ESP+IND and LB+INDO groups.