Introduction

The gastric wall is continuously exposed to various exogenous and endogenous damaging agents. The gastric mucosal barrier is an important structure protecting the gastric wall against damaging agents (1, 2). X-rays possibly slow down the reproduction and regeneration capacity of the vessels in muscle and submucosa in the gastrointestinal tract and of the cells in connective tissues (3, 4). Free oxygen radicals entirely mediate the cytotoxic effects in gastrointestinal epithelium (5). Prostaglandins play a significant role in protecting the gastric mucosal barrier by increasing stimulation of mucus and bicarbonate secretion, mucosal blood flow and phospholipids (6). In addition, in the present study, it is shown that prostaglandin E2 protects the mucosal cells against the fatal effect of radiation (7). Vitamins E and C defend the integrity of the cell membrane against oxidant agents as antioxidants (8, 9). In the studies carried out, it was determined that the vitamin E and C stimulated the prostaglandin synthesis (10, 11).

The purpose of the present study was to investigate whether vitamins E and C protect the gastric mucosal barrier against X-rays.

Materials and Method

Thirty-two Wistar Albino Rats two months old, weighing 200±20 gr were used. They were divided into four groups. The groups were arranged as follows: the first group (n=8) was the sham group; the second group (n=8) was the control group which was irradiated with X-rays, the third group (n=8) to which vitamin C was administered and the fourth group (n=8) to which vitamin E was given. The animals in the groups except the sham group were irradiated with 8.9 Gy X-rays. Twenty-four hours later, the animals were sacrificed by cervical dislocation. The stomach was removed and opened along the greater curvature. The amounts of mucus and phospholipid, which are the components of gastric mucosal barrier, were determined by Corne and Baur's method. The amounts in irradiated rats were found to be lower relative to those in the rats which were not irradiated (p<0.001, p<0.001, respectively). The amounts were observed to be higher in the vitamin C administered group compared with the irradiated control group (p<0.001, 0<0.05, respectively). In the group to which vitamin E was given, the amounts were also found to be higher relative to those in the control group (p<0.001, p<0.05 respectively).

The results indicate that vitamins E and C protect the gastric mucosal barrier against X-rays.

Key Words: X-rays, Gastric mucosal barrier, Vitamin E, Vitamin C.
At the end of 30 days, the rats in the second, third, and fourth groups were irradiated with 8.9 Gy X-rays with a Toshiba DRX 1603 B (0.7mm Al, 125kVp, 200m AS, 10 cm distance) X-ray machine. They were separately placed (ventro-dorsal position) in a Plexiglas box and were kept under controlled temperature (22°C) and a photo period of 12 h light and 12 h dark. The animals were kept hungry for 24 h (but allowed water). The animals were sacrificed by cervical dislocation. The abdomen was opened all along the median line. The stomach was taken out and separated into two pieces along the minor and major curvatures. Amounts of mucus were then determined in one piece according to the method of Corne et al. (12). One piece of stomach, taken out for determination of the amount of mucus was kept for two hours in Alcian Blue solution. After this procedure, the stomachs taken from solution were put into MgCl₂ solution at 37°C and incubated for two hours. The absorbance of MgCl₂ was measured at 605 nm in a spectrophotometer, and the amounts of mucus were obtained through analysis. In the other piece of the stomach, the amount of phospholipid was measured by the method of Baur (13). In order to determine the concentration of phospholipid, 0.2 ml mucosal extract was placed in the tubes. One milliliter of nitric acid was added to each tube and heated by flame until no more nitric acid fumes were observed and then cooled. To each tube was added 1 ml of ascorbic acid –TCA, 0.5 ml of ammonium molybdate (1%) and 1 ml of arsenit cirtare and mixed. After 15 minutes, absorption of the mixture was measured at 700 nm againsta blank. The amount of phospholipid was calculated by the equation below:

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\text{Amount of phospholipide (mg/dl) = \frac{\text{absorption of sample}}{\text{Absorption of standard}}} = \frac{\text{Concentration of standard x dilution coefficient}}{}
\]

The mucosa of the other piece of stomach mucosa was scraped according to this method. The absorbance of supernatant obtained from this scraping, which underwent various extraction procedures, was measured at 700 nm in a spectrophotometer, and the amounts of phospholipid were subtracted. The data were analysed for significance by using the Mann Whitney U test.

**Results**

As seen in Figures 1 and 2, X-ray decreased amounts mucus and phospholipid, which are the components of the gastric mucosal barrier: (p<0.001, p<0.001, respectively). The effects of vitamins E and C on mucus levels in the gastric mucosal barrier in X-ray irradiated rats are seen in Fig.1. The effects of vitamins C and E on phospholipid levels in the gastric mucosal barrier in X-ray
irradiated rats are seen in Fig.2. It was observed that the amounts of mucus and phospholipid were higher in rats given vitamin E relative to those of the control group, which were irradiated with X-rays (p<0.001, p<0.05, respectively). In the rats irradiated with X-rays, vitamin C significantly prevented a decrease in the levels of mucus and phospholipid, which are the components of the gastric mucosal barrier (p<0.001, p<0.05, respectively).

Discussion

According to the results of our study, vitamins C and E significantly reduced the decrease in the levels of mucus and phospholipid, which are the components of the gastric mucosal barrier, in the X-ray irradiated rats. Radiation possibly slows down the reproduction and regeneration of the vessels in muscle and submucosa in the gastrointestinal tract and the cells in the connective tissue (3, 4). In the studies carried out, it was determined that the radiation used experimentally caused ulceration in the small intestines of the rats (14). Acute radiation damage is the result of the death of rapidly renewed epithelium cells in the mucosa (15). Water radiolizes with ionizing radiation, and radicals such O₂⁻, OH, H₂O₂ occur. There is not an intestinal enzyme system which clears OH, the most dangerous radical that is formed. Therefore, free radicals formed in this way deteriorate the integrity and renewal rate of cell membrane, thus leading to ulceration in GIs (16). Prostaglandins play an important role in the strengthening of the gastric mucosal barrier (6). In addition, it has been shown that prostaglandin E₂ protects mucosal cells against the fatal effects of radiation (7). Vitamin E, inhibiting the lipid peroxidation, as a result of free radicals, functions as an antioxidant scavenging free radicals (8). Furthermore, it was found that vitamin E protects the integrity of the cell membrane against oxidant agents (17). In the studies carried out by Lonnier et al. and Soheir et al. it was observed that vitamin E had a protective effect as an antioxidant against the injury in gastrointestinal mucosa resulting from radiation (16, 18). Vitamin E also stimulates synthesis of prostaglandins by activating phospholipase A₂ enzyme (10). However, Behrens et al. (19), in their study in which they aimed to explain whether the effect of vitamin E protecting the gastric mucosa against ulcerogenic agents exhibits its influence by was of prostaglandins or other mechanisms, have shown that vitamin E did not inhibit the occurrence of gastric ulcer caused by indomethacin which is an cyclooxygenase enzyme inhibitor. In the studies carried out, it was determined that vitamin C has a radioprotective effect (18, 20). Clara (11) has found that vitamin C increases the synthesis and secretion of prostaglandins. Charole et al. (21) have suggested that the mucus covering the upper surface of epithelial cells

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**Figure 2.** Effect of vitamin E and C on x-ray irradiated rats on phospholipid component of gastric mucosal barrier.
functions as an antioxidant against free radicals resulting from radiation.

According to our findings, vitamins C and E indicate the effect protecting the gastric mucosal barrier in X-ray irradiated rats. These vitamins are thought to show their effects in two ways; first, they function as an antioxidant against the free radicals as a result of radiation. Second, it may be said that vitamins E and C may indirectly strengthen the gastric mucosal barrier by increasing the synthesis and secretion of prostaglandins, which increase the amounts of mucus and phospholipid, which are important components of the gastric mucosal barrier. However, we are of the opinion that further studies are needed to explain whether other mechanisms play any role in these effects of vitamins E and C or not. As a result, we can say that vitamins E and C protect the gastric mucosal barrier against radiation damage.

References