

Wistar Albino Sıçanlarda İleal Anastomoz İyileşmesinde Piroksikamın Etkisi

Effect Of Piroxicam On Ileal Anastomoses In Wistar Albino Rats

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ÖZET

Amaç: Çalışmamızın amacı bir non steroid antiinflamatuvar ilaç olan piroksikamın sıçanlarda ileal anastomoz iyileşmesindeki etkisini incelemektir.

Gereç ve Yöntem: 20 sıçan randomize olarak iki gruba ayrılarak rezeksiyonları ve ileal anastomozları yapıldı. Deney grubuna operasyondan bir gün önceden sakrifiye edilene kadar 2mg/kg/gün piroksikam verildi. Hayvanlar operasyondan sonraki üçüncü ve yedinci günlerde sakrifiye edildi. Dokunun kollajen miktarını göstergesi olarak doku homojenatı HCl içinde hidroliz edilmesine takiben evapore edildi. Evapore edilmiş örnek HCl içinde deproteinize edilerek işlem sonrasında süpernatant fraksiyonu isopropanol içine alınarak hidroksiprolin spektrofotometrik olarak tayin edildi.

Bulgular: Deney grubunda hidroksiprolin konsantrasyonları üçüncü günde hafif düşerken anastomoz sonrası yedinci günde operasyon öncesi seviyeye yükseldi. Diğer yandan kontrol grubunda ileal hidroksiprolin konsantrasyonu üçüncü günde anlamlı olarak azalırken operasyon sonrasındaki yedinci günde operasyon öncesi seviyeye yükseldi.

Tartışma: Sonuç olarak piroksikam yara iyileşmesinin başlangıç fazı süresince hidroksiprolin düzeylerini arttırabilmektedir.

ABSTRACT

Objective: The purpose of our study was to examine the influence of piroxicam, non-steroid anti-inflammatory drug, on healing of ileal anastomoses in rats.

Materials and Methods: 20 rats were divided randomly into two groups and submitted to resection and ileal anastomosis. The test group received piroxicam, 2mg/kg/day, beginning one day before operation until sacrifice. The animals were sacrificed on third and seventh days after the operation. Tissue homogenate, indicator of tissue collagen content, was hydrolyzed with HCl and then evaporated to dryness. Evaporated sample was deproteinized in HCl and after this processes hydroxyproline was measured spectrophotometrically in supernatant fraction in which taken isopropanol.

Results: In test group, hydroxyproline concentrations were slightly reduced on 3rd day but again increased to the preoperational levels on 7th day after anastomosis (p>0.05). On the other hand, in the control group, ileal hydroxyproline concentrations were reduced significantly on 3rd day but increased to preoperational levels on 7th day after the operation.

Conclusion: In conclusion, piroxicam might improve hydroxyproline levels during initial phase of wound healing.

INTRODUCTION

In gastrointestinal surgery, anastomotic complications such as leakage and dehiscence are serious problems (5, 19, 22, 23). Many factors have been implied as causing these complications such as tension over the anastomoses, disturbed blood supply, local trauma, obstruction and infection (6, 9, 13, 26). Postoperative alterations in collagen concentration have a profound effect upon anastomotic integrity. The strength of the intestine at any time depends upon the submucosal fibrous tissue (2). The strength of fibrous tissue in turn, depends upon the quantity and quality of the collagen which it contains. For quantitative description of anastomotic repair, both mechanical and biochemical parameters are employed (8). Biochemical parameters depend on the measuring the levels of collagen components such as hydroxyproline. Some studies suggest that there is relationship between nonsteroid anti-inflammatory drugs (NSAID) and the healing of anastomoses (20, 21).

In the present study, we investigated the effect of piroxicam on the healing of ileal anastomoses in rats.

MATERIAL AND METHODS

Twenty Wistar albino adult male rats, weighting 230-250 g were obtained from Institute for Experimental Medicine Research. They were fed a standard diet and allowed water ad libitum. They were randomly allocated into 2 groups (test and control). Piroxicam, 2mg/kg/day, was given to test group in drinking water, starting one day before anastomosis until sacrifice. All rats were anesthetized with ether. The abdomen was opened through a median abdominal incision. 1 cm of ileal material was resected in each rat and preserved at -20°C for hydroxyproline measurement. The free ends of the ileum were anastomosed by single layer suture technique.

Half of the rats (n=5) in each group were sacrificed on the 3rd days and the other half (n=5) were sacrificed on the 7th day. The ileum samples were collected and preserved at -20°C for hydroxyproline measurement.

Hydroxyproline levels of the tissue sample, regarded as

a measure of collagen content, were measured as follows: Each tissue sample was homogenized in acetate citrate buffer with pH 6.0. Homogenate was mixed with 12 M HCl for hydrolysis, left in a sterilizer at 80°C for 72 h for evaporation. The remaining precipitate was dissolved by 6 M HCl and left in the incubator for 3h. It was centrifuged for deproteinization. The supernatant was mixed with isopropanol. It was centrifuged again and the supernatant was used for hydroxyproline measurement, according to the method described by Bergman (1). The results were expressed as µg/g of dry tissue.

Statistical Analysis

Data were expressed as means ± SD. Differences in hydroxyproline content among groups were calculated using ANOVA test by using a statistical software package (SPSS 10.0). Statistical significance was considered as p < 0.05 levels.

RESULTS

Figure 1 and 2 show the results of ileal hydroxyproline content. In test group, hydroxyproline concentrations were slightly reduced on 3rd day but again increased to the preoperational levels on 7th day after anastomosis (Fig. 1) (p > 0.05).

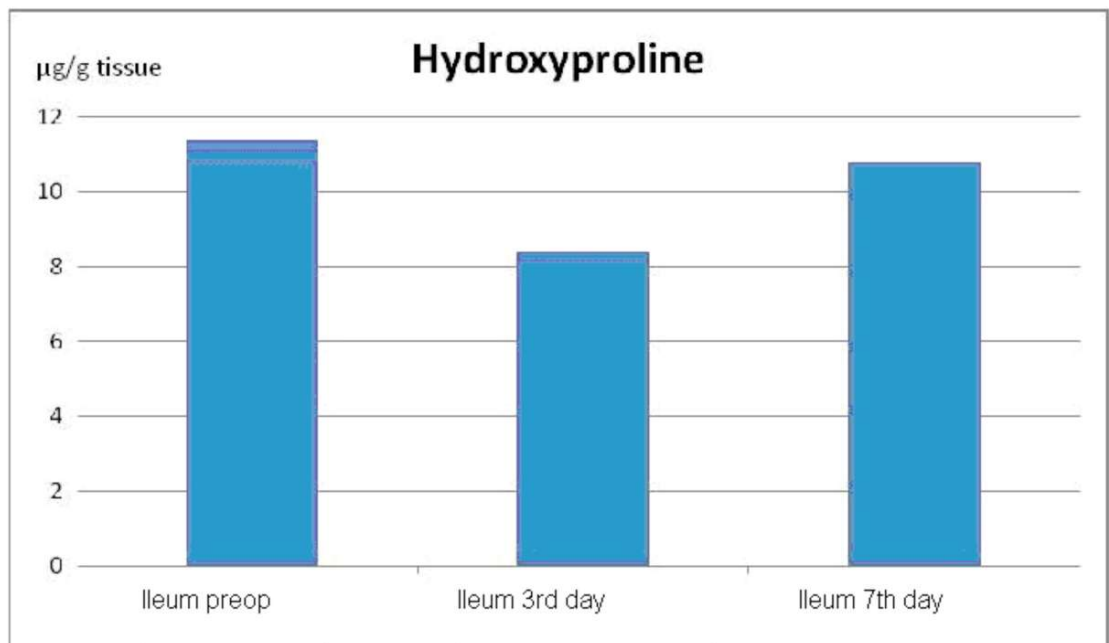


Fig 1. Hydroxyproline concentrations in the test group

On the other hand, in control group, ileal hydroxyproline concentrations were reduced significantly on 3rd day and increased to preoperational levels on 7th day after anastomosis (Fig. 2) (p < 0.05).

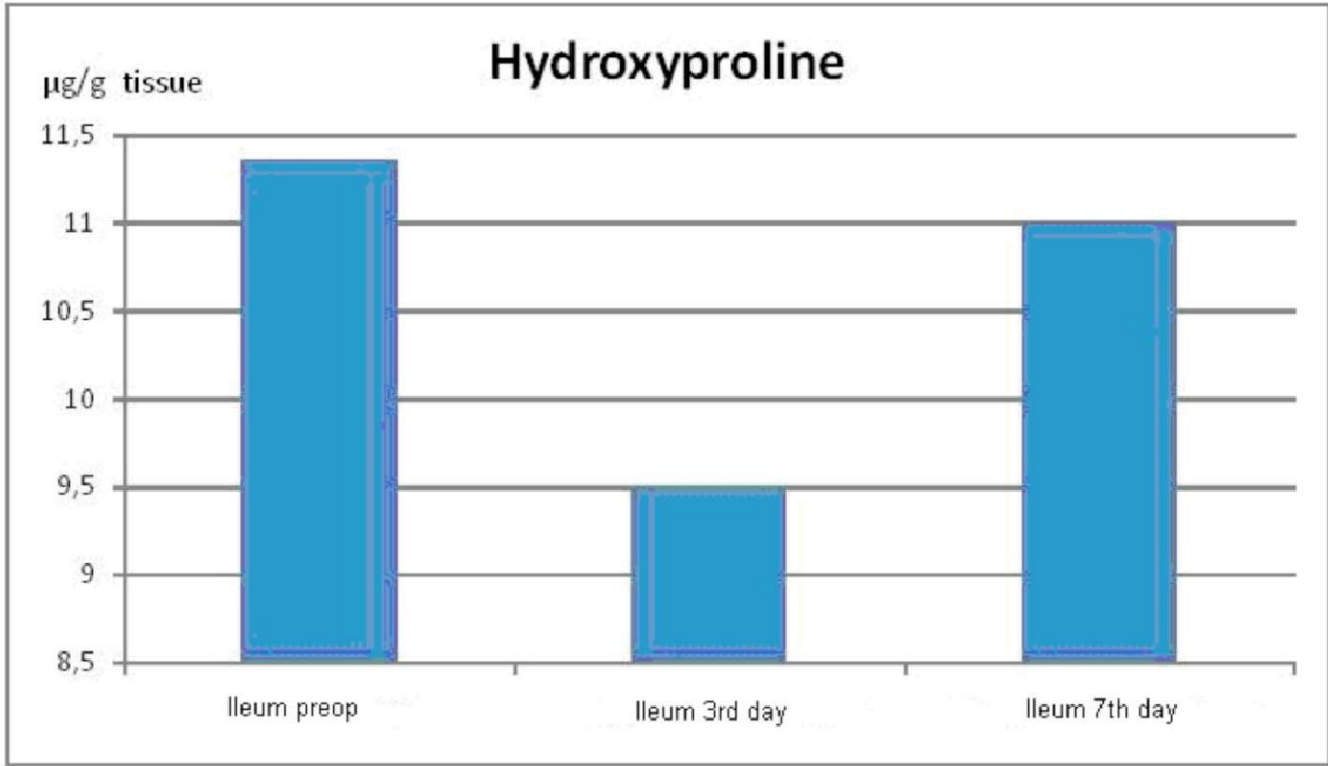


Fig 2. Hydroxyproline concentrations in the control group

DISCUSSION

Healing of intestinal anastomoses is characterized by transient loss of collagen and temporary lowering of intestinal strength. While the occurrence of these changes in experimental colonic anastomoses has been known for some time (7, 14-17, 24), data on ileal anastomoses have only recently become available. A similar loss of bursting strength in dogs (27), and of collagen (25) and breaking strength (18) in rats (10) has been reported.

In the present study, in the control group, hydroxyproline levels on 3rd day were significantly reduced as compared with test group. In contrast, piroxicam significantly prevented the reduction of hydroxyproline levels in test group. In other words, in test group, hydroxyproline was slightly but not significantly reduced on 3rd day as compared with the control group. In both experimental and control groups, hydroxyproline concentrations on 7th day almost reached at the preoperational values. Nevertheless, hydroxyproline levels on 7th day were higher in the test group than control group. Thus, we can speculate that piroxicam given to the animals before the operation might improve healing of intestinal anastomosis. The structural protein collagen is of major importance in maintaining the strength of the intestinal wall. Investigations on the healing of intestinal anastomoses in various animals have shown that massive changes in collagen

concentrations occur during the first week after operation (4, 7, 17). Experimental studies in rats have demonstrated that a significant postoperative decrease in bursting wall pressure, after action of specific enzyme collagenase, is associated with a similar reduction in colonic collagen type I,III,IV, and V, fibronectin, laminin from around the anastomoses using with immunohistochemical antibodies (2). There is a significant increase in the amount of this enzyme produced postoperatively and it is suggested that this is responsible for the decrease in collagen levels and weakness of the collagen molecule (2). It is generally believed that immediately after operation collagen degradation occurs, followed by de novo synthesis. This collagenous equilibrium is thought to be of paramount importance in undistributed healing (12).

Since NSAID are inhibitors of prostaglandins (21), in the present study, the effect of piroxicam on healing might be attributed to the inhibition of the synthesis of prostaglandins, essential molecules involved in inflammatory processes (3, 11) and subsequent suppression of immune responses in the ileal tissue following the operation.

In conclusion, our data suggest that piroxicam impede the decrease of hydroxyproline in healing of anastomoses.

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