Does Becaplermin (Platelet-Derived Growth Factor-BB) Reverse Detrimental Effects of Ischemia on Colonic Anastomosis?

Kaya Sarıbeyoğlu, M.D.,* Bilgi Baca, M.D.,* İsmail Hamzaoğlu, M.D.,* Salih Pekmezci, M.D.,* Tayfun Karahasanoğlu, M.D.,* Hülya Hamzaoğlu, M.D.†

From the Departments of *General Surgery, Cerrahpaşa Medical School, Istanbul University, and †Gastroenterology, Marmara Medical School, Marmara University, Istanbul, Turkey

PURPOSE: The aim of this study was to evaluate the effects of becaplermin on normal and ischemic colon anastomoses. METHODS: Forty adult male Wistar-Albino rats were divided into four equal groups of ten. All rats underwent standard left colon resection and end-to-end anastomosis. The groups were as follows: Group 1, normal anastomosis alone; Group 2, ischemic anastomosis alone; Group 3, normal anastomosis and becaplermin therapy; and Group 4, ischemic anastomosis and becaplermin therapy. All animals were killed on postoperative Day 4. Intra-abdominal adhesions were scored, and anastomotic healing was evaluated with both in situ bursting pressure and hydroxyproline levels. RESULTS: According to bursting pressure results, there was a significant difference between Group 2 and Group 4 (P < 0.05), but there was no statistically significant difference between Group 1 and Group 3 (P > 0.05). Hydroxyproline values revealed no statistically significant difference among any of the groups (P > 0.05). With regard to intra-abdominal adhesion scores, Group 3 had significantly lower values than Group 2 and Group 4 (P < 0.05). No statistically significant difference was observed between the other groups. CONCLUSIONS: Detrimental effects of ischemia on colonic anastomotic healing were reversed with becaplermin therapy. However, there was no such benefit on nonischemic anastomoses. Intra-abdominal application of this gel did not lead to an increase in intra-abdominal adhesion formation. [Key words: Becaplermin; Regranex; Colon; Anastomosis; Ischemia]


Anastomotic dehiscence or failure is associated with increased morbidity and mortality in colorectal surgery. Ischemia on the anastomotic line, which impairs the healing process and consequently leads to disruption, has a crucial role in the etiology of anastomotic dehiscence.1–4 Adequate oxygenation is essential for wound healing, normal oxidative function of neutrophils, leukocyte activation, fibroblast production, angiogenesis, and reepithelialization.5,6 The prevention of complications in such risky anastomosis is a dilemma that has yet to be resolved.

Growth factors are biologically active polypeptides that play an important part in the wound-healing process. However, different growth factors contribute to healing of different types of wounds. For instance, diabetic wounds require angiogenesis first, whereas wounds such as abrasions heal primarily with epithelialization.7 Platelet-derived growth factor-BB (PDGF-BB) is produced in platelets, macrophages, vascular endothelium, and fibroblasts and has strong chemotactic and mitogenic effects on fibroblasts, smooth muscle, and inflammatory cells.8 Becaplermin (recombinant human PDGF-BB; Regranex® gel; Ortho-McNeil Pharmaceutical, Inc, Raritan, NJ) is the only United States Food and Drug Administration (FDA)–approved recombinant growth factor so far, and randomized, placebo-controlled clinical trials have reported its efficacy in the treatment of both chronic diabetic and pressure ulcers.9–11 Moreover, there are experimental indications that PDGF-BB promotes the local angiogenic response and impedes ischemic damage.12

The objective of this study was to investigate whether becaplermin has an effect on normal and ischemic colonic anastomosis in a rat model. Postoperative intra-abdominal adhesions were also evaluated with regard to initial intra-abdominal application of this agent.

MATERIALS AND METHODS

Animals

Approval for the study was obtained from the Institutional Animal Care Committee, and the study was
conducted according to accepted guidelines for care and use of laboratory animals for research. Forty adult male Wistar-Albino rats, each weighing 180 to 220 g, were divided into 4 equal groups of 10. The animals were fed according to the standard laboratory regimen and allowed water *ad libitum*. All rats underwent standard left colon resection and end-to-end anastomosis. The groups were as follows: Group 1, normal anastomosis alone; Group 2, ischemic anastomosis alone; Group 3, normal anastomosis and becaplermin therapy, and Group 4, ischemic anastomosis and becaplermin therapy.

**Surgical Procedure**

Animals were anesthetized by intraperitoneal injection of ketamine hydrochloride (50 mg/kg of body weight). The distal colon was found through a midline laparotomy of 3 cm. Then, a 1-cm segment of left colon, 3 cm proximal to the peritoneal reflection, was resected in each animal. The end-to-end anastomoses were performed with interrupted inverting 6-0 polypropylene sutures. In Groups 2 and 4, to establish ischemia in the anastomotic line, the vessels of the mesocolon were ligated 2 cm proximal and 2 cm distal from the line. The muscle and skin layers of the abdomen were closed separately with continuous 3-0 silk sutures. The animals were allowed to feed after 24 hours and were killed on postoperative Day 4.

**Becaplermin Application**

A 15-g tube of becaplermin gel 0.01 (Regranex®; Ortho-McNeil Pharmaceutical, Inc) was used in this study. The dosage was calculated with the suggested formula of \( \text{length} \times \text{width} / 4 \), and 0.5 cm of gel (125 mg) was applied to the anastomotic line just after completion of the anastomoses.

**Evaluation of Intraperitoneal Adhesions**

Animals were killed on postoperative Day 4. Median laparotomy incisions were reopened, and intraperitoneal adhesions were scored by two different observers who did not know to which group the animal belonged or the score of the other observer, as described by Nagler et al.\(^1\) The severity of adhesions was classified as follows: 0, no adhesions; 1, thin, easily separated adhesion; 2, several thin adhesions; 3, thick, broad adhesion; and 4, several thick adhesions or thick adhesions to organs or abdominal wall. The total of the two scores reflected the final score.

**Measurement of Colonic Bursting Pressure**

After intra-abdominal adhesions were scored, the anastomotic line was defined by polypropylene sutures, and bursting pressure was measured *in situ*. Fecal content of the colon was evacuated gently. The colon was cut and divided at 2 cm proximal to the anastomosis. The cannula was inserted into the colonic segment, proximal to the anastomosis. The cannula was encircled and tied by 3-0 silk to avoid air leaks, then it was connected to an infusion pump. A stopcock was attached between the cannula and manometer. The distal part of the anastomosis was ligated, and the abdomen was filled with saline. The colon was insufflated with air *via* the cannula at a constant rate of 6 ml/min. Bursting pressure was noted when the first air bubbles were observed.

**Hydroxyproline Determination**

A 2-cm-long perianastomotic bowel segment was preserved in a \(-70^\circ\text{C}\) freezer. Hydroxyproline measurement was performed by spectrophotometer at 560 nm.

**Statistical Analysis**

Analysis of variance and Tukey-Kramer multiple comparison tests were used for statistical analysis. *P* values less than 0.05 were considered to be significant. Mean values ± standard deviations are given in the Table.

**RESULTS**

All rats survived during the study. In all groups, bursts occurred during the anastomotic lines. Bursting pressure values are given in the Table. Group 2 had the lowest values, and the differences were significant (*P < 0.05*) vs. each of the other groups (Group 2 vs. Groups 1, 3, and 4), whereas there were no statistically significant differences for Group 1 vs. Groups 3 and 4 or for Group 3 vs. Group 4 (*P > 0.05*).

Hydroxyproline values are shown in the Table. No statistically significant differences were found among the groups.

Group 3 had significantly lower intra-abdominal adhesion scores than Groups 2 and 4. No statistically