Original Article

Cukurova Anestezi ve Cerrahi Bilimler Dergisi

Role of Tacrolimus in Preventing Intra-Abdominal Adhesions

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Abstract

Aim: The study aimed to identify whether tacrolimus has preventive effects on intraperitoneal adhesions. **Methods:** Thirty-four 8–10-week-old male Wistar Albino rats were used. They were randomized into 3 groups; control group, abdominal lavage group, and systemic administration group. The features of abdominal adhesions were evaluated macroscopically, and the degree of adhesion within rats was graded by using Nair and Knightly adhesion grading scales. **Results:** Adhesion grades were significantly lower in tacrolimus administered groups. However, route of tacrolimus administration did not affect the reduction of adhesion formation. **Conclusion:** Tacrolimus has been observed to potentially mitigate the incidence of intraperitoneal adhesion formation without significantly compromising wound healing processes.

Keywords: Tacrolimus; intraperitoneal adhesion; wound healing; rats

1. Introduction

Peritoneal adhesions frequently occur after abdominal surgery[1]. Chronic pelvic pain, intestinal obstruction, and female infertility are some well-known serious conditions related to postoperative adhesions[1-3]. While approximately 30% of intestinal obstructions are due to intra-abdominal adhesions, postoperative peritoneal adhesions can occur in 9 out of 10 patients when all laparotomies are considered[4]. Despite advanced surgical techniques and medical approaches, postoperative adhesions remain a problem.

To prevent abdominal adhesions, some methods have included modifications in surgical techniques and approaches and the use of surgical adjuvants such as fibrinolytic agents, anticoagulants, anti-inflammatory agents, and mechanical separation agents (carboxymethyl cellulose)[5,6]. Clinical observations have indicated significant reduction in adhesion formation in patients received immunosuppressive therapy for organ transplantation; however, experimental and clinical data demonstrating the effect of immunosuppressive agents on adhesion formation remain insufficient[7]. This study aimed to research the preventive effects of tacrolimus on intraperitoneal adhesions using experimental rat models.

Despite the plethora of experimental studies that have investigated pharmacological strategies for preventing postoperative intra-abdominal adhesions, the potential role of tacrolimus in adhesion prevention, particularly in terms of the comparative effects of

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systemic and intraperitoneal administration methods, remains to be fully elucidated. Therefore, the objective of this study was to evaluate the effect of tacrolimus in preventing intraperitoneal adhesion formation and to compare the effectiveness of systemic and intraperitoneal administration routes in an experimental rat model of intra-abdominal inflammation.

2. Materials and Methods

Our study was performed at Ankara University, Laboratory of Experimental Animals, and the research and the animal protocols. The experimental procedures were carried out in accordance with the guidelines established for the use of vertebrates in experimental settings (UN, Stroudsburg, 20 February, 1985).

Thirty-four 8–10-weeks-old male Wistar Albino rats, weighing 250–350 mg, were used. They were randomized into three groups as outlined in Table 1, kept under 12-h light/12-h dark cycles, and provided with food ad libitum.

2.1. Surgical Procedure

The tacrolimus dose (0.5 mg/kg) was selected based on previous experimental studies evaluating the antifibrotic and immunomodulatory effects of tacrolimus in animal models, where similar doses were shown to exert biological activity without causing significant systemic toxicity. All rats were placed under general anesthesia via intramuscular injection of 60 mg/kg ketamine (Ketalar, Pfizer, NY, USA) and 5 mg/kg xylazine (Rompun, Bayer Turk Chemical Industry Limited Company, Istanbul, Turkey). The procedures were performed by only one surgeon for standardization in accordance with the principles of asepsis and antisepsis. The rats were placed in the supine position, and after laparotomy, cecal ligation and puncture of cecum were performed on all rats in all groups. A median incision was made in the abdomen, approximately 3 cm from the xiphoid process following which cecum was ligated with 3/0 silk suture (Sterisilk, SSM, Istanbul, Turkey) without blocking passage from the intestine to colon. Thereafter, a 22-gauge needle was punctured into the cecum on one side and through the cecal wall on the opposite side, and contamination was achieved by feces extralumination. The blind bowel ligation and perforation (CLP) model is a widely used experimental approach to simulate two conditions closely associated with excessive inflammatory responses in the abdominal cavity: namely, intra-abdominal sepsis and polymicrobial peritonitis. The inflammatory environment has been demonstrated to encourage fibrin deposition, impaired fibrinolysis, and subsequent adhesion formation. Consequently, the CLP model provides a clinically relevant framework for evaluating the therapeutic potential of agents designed to reduce adhesion formation under inflammatory intra-abdominal conditions. The CLP model was selected for this procedure due to its ability to address these requirements. The full-thickness of the abdominal incision was sutured continuously with 4/0 polypropylene sutures (Prolene, Ethicon Ltd, USA). Imipenem (Tienam, Merck Sharp & Dohme, New York, USA) was administered intramuscularly to all subjects at 15 mg/kg to reduce the mortality rate caused by sepsis. Further, 10 mL 0.9% sodium chloride (NaCl) solution was injected subcutaneously for fluid resuscitation. In the postoperative eighth hour under general anesthesia, relaparotomy was performed and necrotic cecum was resected. Abdominal lavage was administered with 20 mL 0.9% NaCl solution, and the full-thickness abdominal incision was sutured continuously with 4/0 polypropylene sutures.

Group 1 (Control Group): All rats in this group underwent the standard adhesion induction procedure.

Group 2 (Abdominal Lavage Group): Abdominal lavage was performed with 0.5 mg/kg tacrolimus added to 20 mL 0.9% NaCl for all rats in this group.

Group 3 (Systemic Administration Group): Tacrolimus was injected 2 h before surgical intervention, and daily tacrolimus was administered subcutaneously at the same dose until the tenth postoperative day.

All rats were euthanized with intracardiac injection of 100 mg/kg thiopental (Pentothal, Abbott Company, Chicago, IL, USA), and relaparotomy was performed. The peritoneal cavity of rats was exposed through U-shaped incisions, providing maximal exposure to quantitatively evaluate adhesions using the Nair and Knightly adhesion scoring systems (Table 2)[8,9]. The features of abdominal adhesions were evaluated macroscopically, and the degree of adhesion within rats was graded from 0 to 4 (Figure 1). Macroscopic adhesion evaluation was conducted by an independent investigator who was blinded to the group allocation in order to minimize potential observer bias.

2.2. Statistical Analysis

The data were analyzed using SPSS version 23.0 (SPSS Inc., Chicago, IL, USA). The Non-parametric Kruskal–Wallis H-test was used to determine whether there were differences between the groups. If significant differences were found, multiple comparison tests were used to determine which groups were different. When statistically significant differences were identified, a post-hoc test, specifically the Dunn–Bonferroni method, was employed to perform pairwise comparisons. A p-value < 0.05 was accepted as significant.

2.3. Ethics Approval

This study was approved by Ankara University, Local Ethics Committee for Animal Experiments (protocol number: 2009-38-182, date: 2009/03/04).

Table 1. Features of the study groups.

Groups	Number of rats	Surgical procedure (Laparotomy, cecal ligation, and puncture)	Method of tacrolimus administration
Group 1	12	+	None
Group 2	11	+	Abdominal lavage
Group 3	11	+	Systemic administration

Table 2. Nair and Knightly adhesion grading scales.

Grade	Nair	Knightly
Grade 0	No adhesion	No adhesion
Grade 1	Single band of adhesion between viscera to abdominal wall	A single, thin, easily separable adhesion
Grade 2	Two adhesive bands either between viscera or viscera to abdominal wall	Less extensive but weak adhesions which stool traction poorly
Grade 3	More than two bands between viscera or viscera to abdominal wall or intestines, forming a mass without being adherent to the abdominal wall cohesive attachment	Numerous extensive visceral adhesions, which involved the parietal extensions
Grade 4	Dense and complex adhesive bands between viscera or viscera to abdominal wall, or visceral adhesion to abdominal wall directly	Numerous extensive dense adhesions, which involved the adjacent mesentery, intestine, and omentum, extending to the abdominal wall

3. Results

Throughout the study process, four rats were lost (group 1, two rats; group 2, one; and group 3, one). The cause of mortality was identified as sepsis in all four rats, and there

were no differences between the groups in terms of exitus (Pearson $\chi^2 = 0.807$). A total of 30 rats were sacrificed on tenth postoperative day.

Wound infection was observed in five rats (group 1, two rats; group 2, one; and group 3, two), but there were no significant differences between the groups (Pearson $\chi^2 = 0.787$).

The mean Nair and Knightly adhesion scores of the study groups are shown in Table 3. We compared the Nair and Knightly adhesion grades between the groups and determined that both grades were significantly lower in groups 2 and 3 than in group 1 ($^P < 0.05$; Table 4). There were no significant differences in the scores between groups 2 and 3 ($^P > 0.05$; Table 4).

Table 3. Kruskal–Wallis test results for adhesion formation in the study groups.

Study Group	Nair Adhesion Score Mean ± SD	Knightly Adhesion Score Mean ± SD
Group 1	2.2 ± 0.79	2.0 ± 0.67
Group 2	1.2 ± 0.76	1.1 ± 0.74
Group 3	1.1 ± 0.74	1.1 ± 0.74

Group 1: Control Group, Group 2: Abdominal Lavage Group, Group 3: Systemic Administration Group

Table 4. Paired statistical comparisons of adhesion formation between the study groups.

Study Groups	P-value for Nair Adhesion Scores	P-value for Knightly Adhesion Scores
Group 1/Group 2	0.006	0.015
Group 1/Group 3	0.006	0.015
Group 2/Group 3	1.000	1.000

Group 1: Control Group, Group 2: Abdominal Lavage Group, Group 3: Systemic Administration Group

Figure 1. Representative adhesions after surgical procedure. (A) Grade 0 adhesion (B) Grade 3 adhesion.

4. Discussion

Postoperative intra-abdominal adhesions are one of the most important factors affecting long-term morbidity, and preventive strategies against adhesion formation have been the goal of numerous studies. Local or systematic administration of several drugs and materials, such as mechanical barriers and physical, chemical, and pharmacologic

agents, has been used for prevention of adhesion formation[10,11]. However, there is no approved standard procedure or therapy despite many promising investigations[12].

Peritoneal inflammation characterized with extravasations of serum and cellular elements is the first response to trauma, ischemia, infection, and foreign bodies[13,14]. Extravasation of fibrinogen molecules with postoperatively increased vascular permeability causes a fibrous exudation. Inflammatory cells, particularly phagocytes, infiltrate the injured area by the first day, and then fibroblasts begin to produce collagen fibrils on the third day. The inflammatory response depends significantly on the local release of proinflammatory cytokines[10,15]. Transforming growth factor (TGF)-β plays a key role in tissue recovery and triggers tissue fibrosis. It is known that excessive amounts of TGF-β are released by the peritoneum in the acute inflammatory phase and cause increased adhesion formation. In the period of peritoneal recovery and adhesion formation, plasmin activates not only latent TGF-β but also other cellular mediators of adhesion formation[16].

Nagano et al. studied the effects of tacrolimus in rats with lung fibrosis induced by bleomycin and they showed that tacrolimus inhibited the TGF-β-dependent collagen synthesis and decreased lung fibrosis[17]. Studies regarding tacrolimus have focused mainly on its immunosuppressive effects, and a limited number of studies have researched the antifibrinogenic and antiproliferative effects of tacrolimus. Tacrolimus is a calcineurin inhibitor-like cyclosporin A (CsA) but differs from CsA with antiproliferative effects on human gingival fibroblast[18].

Studies of adhesion formation have focused on the similarities in cytokine production during responses to rat and human peritoneal injury[19]. Animal models can provide the same conditions in some experimental studies[19]. Wichterman et al. first performed cecal ligation and puncture in rats to generate an experimental model of polymicrobial sepsis in 1980[20]. Experimental cecal ligation and puncture is a research model technique suitable for modifications and manipulations. In this model, the diameter of the cecal puncture is directly related to the mortality of the rats. Using a 22-gauge needle for puncture in this model results in a 27% mortality rate, whereas a puncture with 0.5 cm diameter causes a 95% mortality rate. Varying the diameter of the puncture can adjust the severity of sepsis in experimental studies and gives the advantage of being able to obtain clinical features resulting in different types of models, such as pure sepsis and adhesion models[21].

Tuzuner et al. investigated the effects of hyaluronan-based agents on intra-abdominal adhesion formation in a model of sepsis and identified more extensive adhesions in septic groups[22]. Previous studies already confirmed that abdominal sepsis is a predictive factor for intra-abdominal adhesions[23].

Treatment strategies preventing adhesion formation should aim to control cellular mediators and cytokines in the peritoneal fluid at the beginning of adhesion formation[24]. Some of these mediators are tnf-α, IL-1, IL-6, TGF-β, and epidermal growth factor[25-27].

Wasserberg and colleagues conducted a study in which they performed small intestine transplants on four groups of rats. A substantial decrease in the total adhesion score was observed in animals treated with tacrolimus, both in allogeneic and syngenic combinations[28]. Peker et al. utilized a rat model to comparatively assess the efficacy of systemically and intraperitoneally administered common immunosuppressive medications, including mycophenolate mofetil, tacrolimus, and cyclosporine, in the prevention of postoperative intraperitoneal adhesions[7]. They reported a decrease in fibrosis with all immunosuppressive agents. Further, they mentioned that the decrease in fibrosis was most predominant in rats that received intra-peritoneal tacrolimus. Furthermore, a reduction in vascular proliferation was observed in all experimental groups except those receiving systemic tacrolimus treatment. The most significant reduction in vascular proliferation was observed in rats treated with intraperitoneally administered tacrolimus[7].

4.1. Limitations

It is imperative to acknowledge the limitations of this study when interpreting the findings. Firstly, the relatively modest sample size may constrain the generalizability of the findings. Secondly, adhesion assessment was performed using macroscopic scoring systems. However, histopathological confirmation of fibrosis or inflammatory activity was not performed. Additionally, the evaluation of cytokine levels and molecular mediators implicated in adhesion formation was not conducted. Subsequent studies with augmented sample sizes and comprehensive histopathological and biochemical assessments may offer a more exhaustive understanding of the mechanisms by which tacrolimus affects adhesion formation.

5. Conclusion

Tacrolimus seems to reduce intraperitoneal adhesion formation significantly without any prominent adverse effect on wound healing in the therapeutic dose range. Immunosuppressive drugs may offer an effective solution for preventing postoperative intraperitoneal adhesions and their potentially serious complications.

genAI: No artificial intelligence-based tools or generative AI technologies were used in this study. The entire content of the manuscript was originally prepared, reviewed, and approved by all authors.

Statement of Ethics: This study was approved by Ankara University, Local Ethics Committee for Animal Experiments (protocol number: 2009-38-182, date: 2009/03/04).

Scientific Responsibility Statement: The authors declare that they are responsible for the article's scientific content, including study design, data collection, analysis and interpretation, writing, and some of the main line, or all of the preparation and scientific review of the contents, and approval of the final version of the article.

Animal and Human Rights Statement: All procedures performed in this study were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards.

Data Availability: The datasets used and/or analyzed during the current study are not publicly available due to patient privacy reasons but are available from the corresponding author on reasonable request.

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References

1. Fatehi Hassanabad A, Zarzycki AN, Jeon K, et al. Post-Operative Adhesions: A Comprehensive Review of Mechanisms. Biomedicines. 2021;9(8):867. https://doi.org/10.3390/biomedicines9080867

2. Howard FM. The role of laparoscopy in the chronic pelvic pain patient. Clin Obstet Gynecol. 2003;46(4):749-766. https://doi.org/10.1097/00003081-200312000-00004

3. D'Cruz OJ, Uckun FM. Targeting mast cells in endometriosis with janus kinase 3 inhibitor, JANEX-1. Am J Reprod Immunol. 2007;58(2):75-97. https://doi.org/10.1111/j.1600-0897.2007.00502.x

4. Becker JM, Stucchi AF. Intra-abdominal adhesion prevention: are we getting any closer?. Ann Surg. 2004;240(2):202-204. https://doi.org/10.1097/01.sla.0000133118.38686.d0

5. Kamel RM. Prevention of postoperative peritoneal adhesions. Eur J Obstet Gynecol Reprod Biol. 2010;150(2):111-118. https://doi.org/10.1016/j.ejogrb.2010.02.003

6. Lim R, Stucchi AF, Morrill JM, et al. The efficacy of a hyaluronate-carboxymethylcellulose bioresorbable membrane that reduces postoperative adhesions is increased by the intra-operative co-administration of a neurokinin 1 receptor antagonist in a rat model. Surgery. 2010;148(5):991-999. https://doi.org/10.1016/j.surg.2010.01.024
7. Peker K, Inal A, Sayar I, et al. Prevention of intraabdominal adhesions by local and systemic administration of immunosuppressive drugs. Iran Red Crescent Med J. 2013;15(12):e14148. https://doi.org/10.5812/ircmj.14148
8. Nair SK, Bhat IK, Aurora AL. Role of proteolytic enzyme in the prevention of postoperative intraperitoneal adhesions. Arch Surg. 1974;108(6):849-853. https://doi.org/10.1001/archsurg.1974.01350300081019
9. KNIGHTLY JJ, AGOSTINO D, CLIFFTON EE. The effect of fibrinolysin and heparin on the formation of peritoneal adhesions. Surgery. 1962;52:250-258.
10. Kaptanoglu L, Kucuk HF, Yegenoglu A, et al. Effects of seprafilm and heparin in combination on intra-abdominal adhesions. Eur Surg Res. 2008;41(2):203-207. https://doi.org/10.1159/000134919
11. Kesting MR, Loeffelbein DJ, Steinstraesser L, et al. Cryopreserved human amniotic membrane for soft tissue repair in rats. Ann Plast Surg. 2008;60(6):684-691. https://doi.org/10.1097/SAP.0b013e31814fb9d2
12. Hellebrekers BW, Trimbos-Kemper GC, van Blitterswijk CA, et al. Effects of five different barrier materials on postsurgical adhesion formation in the rat. Hum Reprod. 2000;15(6):1358-1363. https://doi.org/10.1093/humrep/15.6.1358
13. Saed GM, Munkarah AR, Diamond MP. Cyclooxygenase-2 is expressed in human fibroblasts isolated from intraperitoneal adhesions but not from normal peritoneal tissues. Fertil Steril. 2003;79(6):1404-1408. https://doi.org/10.1016/s0015-0282(03)00257-7
14. Guvenal T, Cetin A, Ozdemir H, et al. Prevention of postoperative adhesion formation in rat uterine horn model by nimesulide: a selective COX-2 inhibitor. Hum Reprod. 2001;16(8):1732-1735. https://doi.org/10.1093/humrep/16.8.1732
15. Gilroy DW, Tomlinson A, Willoughby DA. Differential effects of inhibitors of cyclooxygenase (cyclooxygenase 1 and cyclooxygenase 2) in acute inflammation. Eur J Pharmacol. 1998;355(2-3):211-217. https://doi.org/10.1016/s0014-2999(98)00508-1
16. Chegini N. TGF-beta system: the principal profibrotic mediator of peritoneal adhesion formation. Semin Reprod Med. 2008;26(4):298-312. https://doi.org/10.1055/s-0028-1082388
17. Nagano J, Iyonaga K, Kawamura K, et al. Use of tacrolimus, a potent antifibrotic agent, in bleomycin-induced lung fibrosis. Eur Respir J. 2006;27(3):460-469. https://doi.org/10.1183/09031936.06.00070705
18. Gagliano N, Moscheni C, Dellavia C, et al. Immunosuppression and gingival overgrowth: gene and protein expression profiles of collagen turnover in FK506-treated human gingival fibroblasts. J Clin Periodontol. 2005;32(2):167-173. https://doi.org/10.1111/j.1600-051X.2005.00654.x
19. Cantürk NZ, Sayek İ. Karın içi adezyon oluşturma teknikleri ve önleme modelleri. In: Cantürk NZ, Sayek İ, editors. Cerrahi araştırma: Nobel Tıp Kitabevleri; 2005. p. 421-8.
20. Wichterman KA, Baue AE, Chaudry IH. Sepsis and septic shock--a review of laboratory models and a proposal. J Surg Res. 1980;29(2):189-201. https://doi.org/10.1016/0022-4804(80)90037-2
21. Otero-Antón E, González-Quintela A, López-Soto A, et al. Cecal ligation and puncture as a model of sepsis in the rat: influence of the puncture size on mortality, bacteremia, endotoxemia and tumor necrosis factor alpha levels. Eur Surg Res. 2001;33(2):77-79. https://doi.org/10.1159/000049698
22. Tüzüner A, Kuzu MA, Akin B, et al. The effect of hyaluronan-based agents on adhesion formation in an intraabdominal sepsis model. Dig Dis Sci. 2004;49(6):1054-1061. https://doi.org/10.1023/b:ddas.0000034571.30602.36
23. Reijnen MM, Bleichrodt RP, van Goor H. Pathophysiology of intra-abdominal adhesion and abscess formation, and the effect of hyaluronan. Br J Surg. 2003;90(5):533-541. https://doi.org/10.1002/bjs.4141
24. Cheong YC, Laird SM, Shelton JB, et al. The correlation of adhesions and peritoneal fluid cytokine concentrations: a pilot study. Hum Reprod. 2002;17(4):1039-1045. https://doi.org/10.1093/humrep/17.4.1039
25. Saba AA, Kaidi AA, Godziachvili V, et al. Effects of interleukin-6 and its neutralizing antibodies on peritoneal adhesion formation and wound healing. Am Surg. 1996;62(7):569-572.
26. Hershlag A, Otterness IG, Bliven ML, et al. The effect of interleukin-1 on adhesion formation in the rat. Am J Obstet Gynecol. 1991;165(3):771-774. https://doi.org/10.1016/0002-9378(91)90326-m
27. Naiki Y, Maeda Y, Matsuo K, et al. Involvement of TGF-beta signal for peritoneal sclerosing in continuous ambulatory peritoneal dialysis. J Nephrol. 2003;16(1):95-102.
28. Wasserberg N, Nunoo-Mensah JW, et al. The effect of immunosuppression on peritoneal adhesions formation after small bowel transplantation in rats. J Surg Res. 2007;141(2):294-298. https://doi.org/10.1016/j.jss.2006.12.541