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Article

THE EFFECTIVENESS OF INTRADERMAL SUTURE TECHNIQUE ON HYPERTROPHIC SCAR PREVENTION IN RATS

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ABSTRACT

Introduction : Hypertrophic scarring is a complication that occurs in post-surgical wounds. There are many ways to prevent scarring, but there have been no satisfactory methods yet. Moreover, no studies investigated the effectiveness of intradermal sutures. Polypropylene thread can be used to repair a scar after surgery. This study aimed to prove the effectiveness of intradermal sutures using polypropylene thread on surgical scar quality.
Method : The study was conducted using a randomized, controlled trial, and post-test only with 20 rats (*Rattus norvegicus*) as animal subjects. The wounds were made on the back, approximately 6 mm x 2 cm. Then, the rats were divided into two groups: a control group without intradermal sutures (K) and treatment group with intradermal sutures (I). Histopathological examination using Hematoxylin and Eosin (H&E) staining was used to identify the fibroblast number on the 21st day. The fibrocytes number were also observed on the 12th week and the Vancouver Scar Scale (VSS) was used to assess the scar quality.
Result : The number of fibroblasts and fibrocytes in the control group (K) was significantly smaller (53.60 ± 14.571; 3.20 ± 0.447) than the treatment (I) (243.20 ± 75.334; 171.40 ± 13.221). The VSS value was significantly greater in the control than the treatment.
Conclusion: The intradermal sutures using polypropylene thread produced a better-quality scar after surgery compared to wounds without intradermal sutures.
***Keywords**: Fibroblasts; Fibrocytes; Hypertrophic scars; Intradermal sutures; Polypropylene*

Latar Belakang: Jaringan parut hipertrofik merupakan komplikasi yang terjadi pada luka pasca operasi. Ada banyak cara untuk mencegah jaringan parut, tetapi belum ada metode yang memuaskan. Selain itu, tidak ada penelitian yang menyelidiki efektivitas jahitan intradermal. Benang polipropilen dapat digunakan untuk memperbaiki bekas luka setelah operasi. Penelitian ini bertujuan untuk membuktikan efektivitas jahitan intradermal menggunakan benang polipropilen terhadap kualitas bekas luka operasi.
Metodologi: Penelitian dilakukan secara randomized controlled trial, dan post-test dengan subjek hewan 20 ekor tikus *(Rattus norvegicus)*. Luka dibuat di bagian punggung, kurang lebih 6 mm x 2 cm. Kemudian tikus dibagi menjadi dua kelompok yaitu kelompok kontrol tanpa jahitan intradermal (K) dan kelompok perlakuan dengan jahitan intradermal (I). Pemeriksaan histopatologi menggunakan pewarnaan Hematoxylin dan Eosin (H&E) digunakan untuk mengidentifikasi jumlah fibroblas pada hari ke-21. Jumlah fibrosit juga diamati pada minggu ke-12 dan Vancouver Scar Scale (VSS) digunakan untuk menilai kualitas scar.
Hasil: Jumlah fibroblas dan fibrosit pada kelompok kontrol (K) nyata lebih kecil (53,60 ± 14,571; 3,20 ± 0,447) dibandingkan dengan kelompok perlakuan (I) (243,20 ± 75,334; 171,40 ± 13,221). Nilai VSS secara signifikan lebih besar pada kelompok kontrol dibandingkan kelompok perlakuan.
Kesimpulan: Jahitan intradermal menggunakan benang polipropilen menghasilkan kualitas bekas luka yang lebih baik setelah operasi dibandingkan dengan luka tanpa jahitan intradermal.
***Kata Kunci:** Fibroblas; Fibrosit; Bekas luka hipertrofik; jahitan intradermal; Polipropilena*

Conflicts of Interest Statement:
The author(s) listed in this manuscript declare the absence of any conflict of interest on the subject matter or materials discussed.

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INTRODUCTION

Scarring, in simple terms, is a mark resulting from a wound. Clinically, a scar is a natural defect obtained during the wound healing process. The disruption of the wound healing process causes excessive or abnormal scarring, which can either be a keloid or hypertrophic scar.[1,2] Scars can also arise from genetic diseases and injuries, which become a challenge in the treatment process. One of the top treatments, like major skin grafts, still had limitations such as disfigurement, psychological trauma, and the loss of functionality.[3]

The range of abnormal scarring incidence varies from 40-70% for hypertrophic scars and 6-16% for keloid scars. Preventing a continuing injury to develop into an abnormal scar is important and more efficient than treatment.[2,4-6] Hypertrophic scarring causes itching, pain, discomfort, and aesthetic issues as well as affects joint movement when it occurs in the joint area.[7-9]

Preventing abnormal scarring is possible with good surgical techniques and proper wound management to prevent infection. Removing as little tissue as possible, eliminating the dead space, avoiding hematoma, tissue adaptation and approximation, supporting sutures to maintain the stretch using non-absorbable threads, and removing the cutaneous sutures after strengthening the wound attachment are some good surgical techniques for this case.[1] On the other hand, the bad surgical technique can cause a permanent scar to form and has a large risk of scar recurrence.[3,10]

Intradermal suturing is one technique to prevent hypertrophic scarring. It has been widely implemented and recommended by plastic surgeons. A study by Yang et al. showed better cosmesis results using an intradermal technique with synthetic absorbable sutures.[11] Another study by Elsharkawy demonstrated that the unremoved thread after suture could increase the wound infection, hypertrophic, and keloid scar formation.[12] Polypropylene is a non-absorbable thread used in a suture technique, which distributes the wound stress, maintains wound density, and ensures wound eversion.[3,13-14] This study was important because the proper wound closure methods are essential to prevent hypertrophic or keloid scar formation. This study aimed to examine the effect of intradermal sutures on surgical scar quality.

METHOD

Study Design

The study was carried out using a randomized, controlled trial, and post-test only design. The study was conducted at the Pharmacology Laboratory, Faculty of Medicine, Brawijaya University, Malang. The samples were 20 rats (Rattus norvegicus). This value was obtained based on the Biswas and Charan calculation method.[15] Ten experimental animals with intradermal sutures were used as the treatment group (I) and others with complex sutures were used as the control group (K). Each experimental animal underwent anesthesia using an intramuscular injection of Ketalar 100 mg/kg FW. The hair of the rat's back was shaved and treated with antiseptics. The wound was inflicted in the middle of the experimental animal's back into the inner elliptical fascia by 6 mm wide and 2 cm in length. The treatment group (I) was given the intradermal suture addition using Prolene® 7-0 thread, followed by complex sutures using Prolene 5-0® thread. The control group (K) was given the suture in the skin using Prolene 5-0® thread. The experimental animal was given 1 mg/kg FW metamizole three times per day in two days. The suture was removed on the 7th day.

Sample Observation

The sample was observed using histopathology examination. The samples were taken on the 21st day to identify the number of fibroblasts and in the 12th week to identify the number of fibrocytes and assess the scar quality using the Vancouver Scar Scale (VSS). The VSS score is used to evaluate the scar morphologically by considering four aspects: 1. vascularization; 2. pigmentation; 3. consistency/flexibility; 4. height. The histopathological preparations were carried out using the Hematoxylin and Eosin (H&E) staining technique by Rong et al.[16] A 4-µm

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skin tissue sample section was cut from each animal. After the sections were deparaffinized and rehydrated through a series of alcohol concentrations, the hematoxylin and eosin (H&E) staining was performed based on the standard protocols (Sigma, San Francisco, USA).

Statistical Methods

All data were analyzed using SPSS 21.0 (IBM, USA). The difference was statistically significant if the p-value <0.05.

Ethical Considerations

This study was conducted on experimental animals. The treatment was carried out with anesthesia using a catheter. This study was declared ethically decent by the Health Research Ethics Commission of dr. Saiful Anwar Malang General Hospital with a Certificate of Ethical Eligibility No.189/EPK/X/2010, October 10, 2010.

RESULTS

The Fibroblast, Fibrocytes Number, and VSS Result

The statistical analysis result of fibroblasts, fibrocytes, and VSS showed the difference values between the control (K) and treatment (I) (Table 1). These results indicated that there were significant differences (p <0.05) in fibroblasts, fibrocytes, and VSS values between the two groups. The fibroblasts and VSS in the treatment group (I) were lower than control (K), while the fibrocytes were higher (Table 1).

Table 1. The Differences in Fibroblasts, Fibrocytes Number, and VSS Values Between Control (K) and Treatment (I)

Variable	Groups	Mean ± SD	p
Fibroblast	Control (K)	527.80 ± 26.157	0.001
	Treatment (I)	243.20 ± 75.334	
Fibrocytes	Control (K)	53.60 ± 14.571	0.001
	Treatment (I)	171.40 ± 13.221	
VSS	Control (K)	3.20 ± 0.447	0.001
	Treatment (I)	1.00 ± 0.000	

VSS = Vancouver Scar Scale

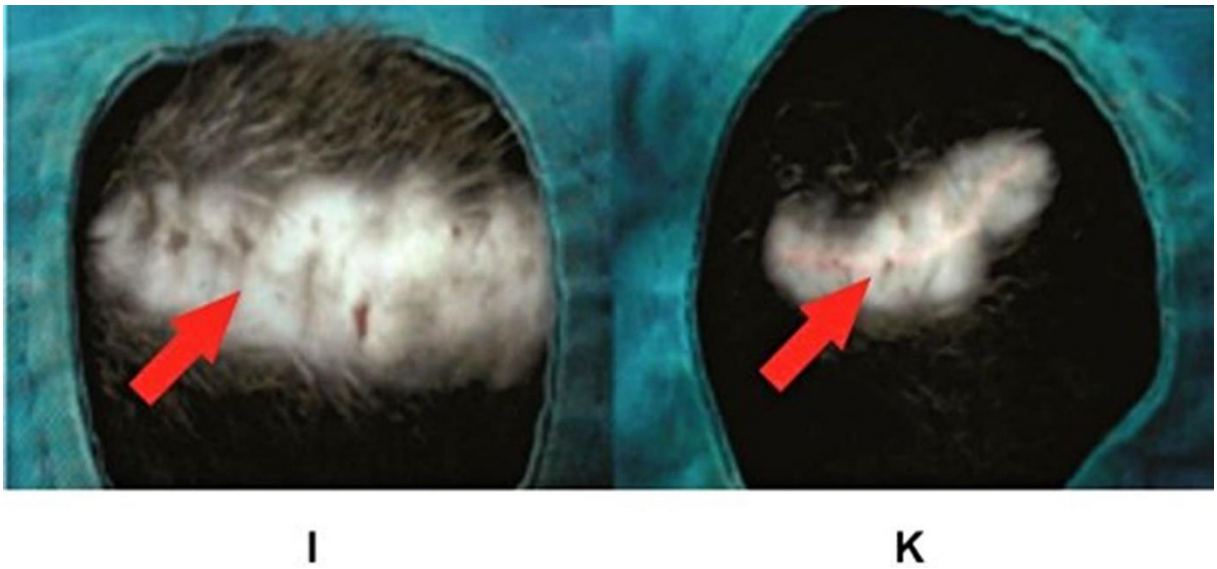

Figure 1. *The wound scar in the control group (K) had a reddish color with less than 2 mm protrusion. The treatment group (I) also had the same color but no protrusion.*

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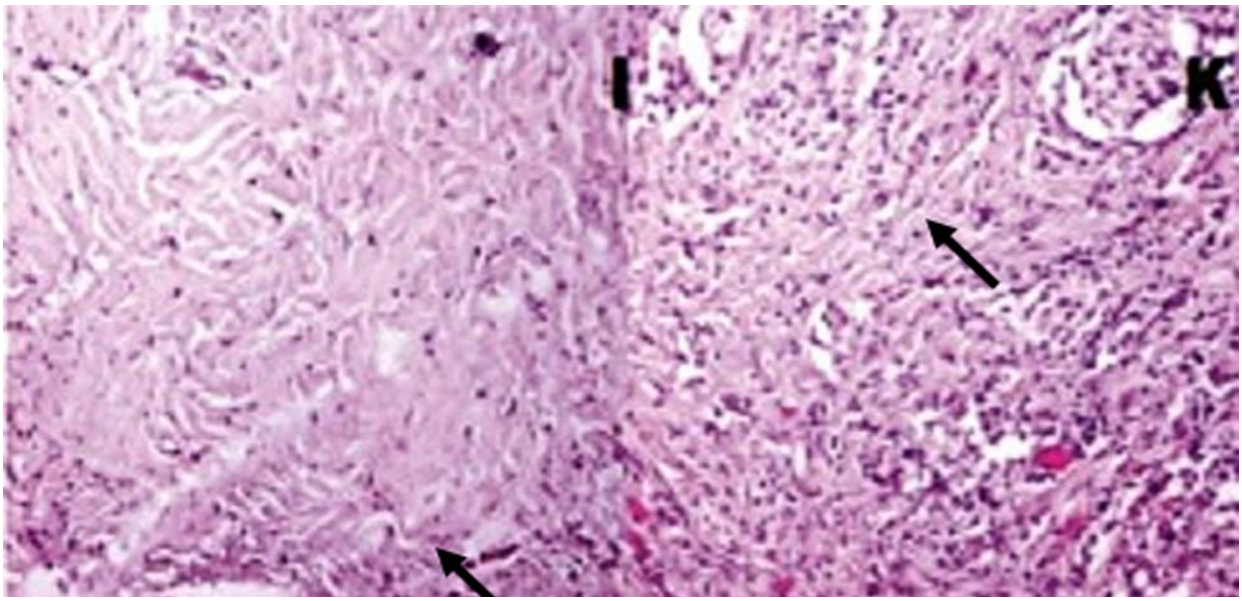

Figure 2. *The collagen density in the treatment group (I) was smaller compared to control (K) on the 21st day.*

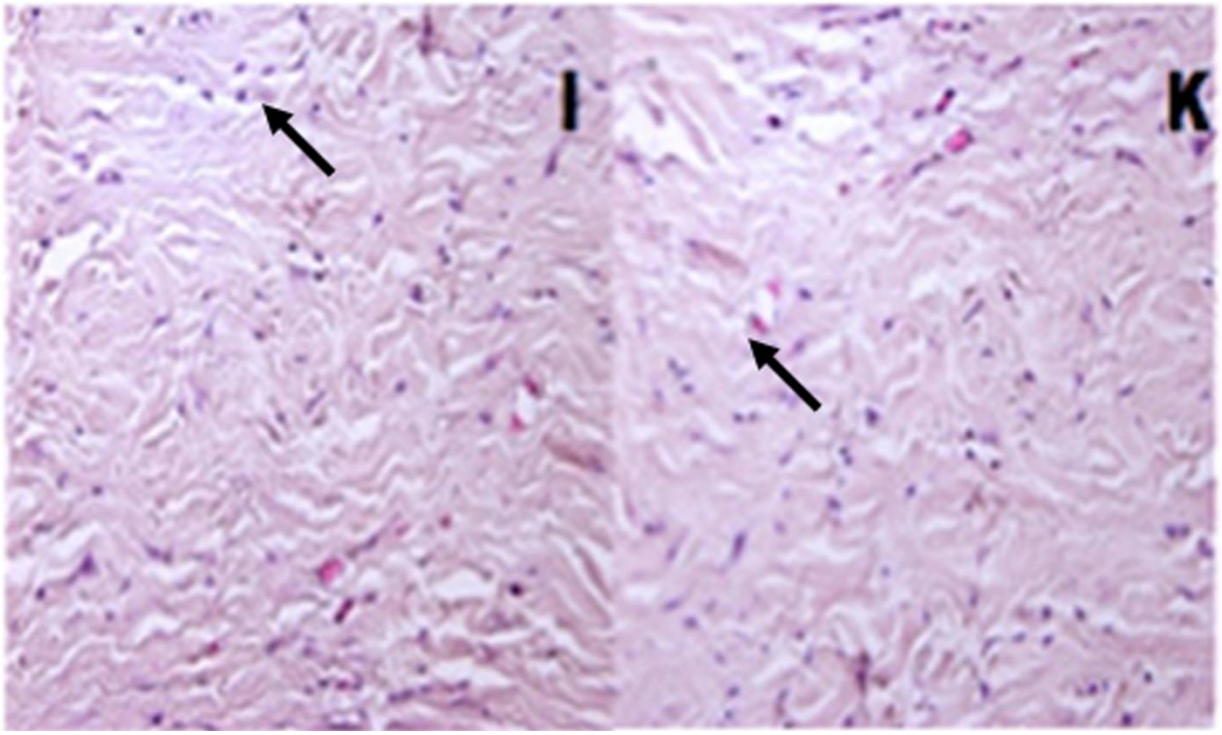

Figure 3. *The collagen density in the treatment group (I) was smaller compared to control (K) on the 12th week.*

The VSS on surgical scar observation at the 12th week had a similar value in the treatment, which was 1. The control group (K) had diverse scores, ranging from 3 to 5. This study did not find fistulation or signs of infection in the surgical wound. The wound scar in the control group (K) had a reddish color with less than 2 mm protrusion. The treatment group (I) also had the same color but no protrusion (Figure 1). The microscopic observation of wound scarring

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showed differences between the treatment and control. The collagen density in the treatment group (I) was smaller compared to control (K) on the 21st day (Figure 2) and the 12th week (Figure 3). These results indicated the reduced fibroblast activity in the intradermal sutures treatment group.

DISCUSSION

The suturing technique is the most important factor to produce a cosmetically good surgical scar. According to Kudur et al. wound sutures can generate a maximal wound eversion and maintain tensile strength during the wound healing process.[17] We argues that all criteria can be fulfilled by intradermal sutures.

The histopathological result showed a significant difference in the fibroblast number on the 21st day. The group with the intradermal sutures had a smaller number of fibroblasts compared to the control. Meanwhile, the VSS in the intradermal sutures was smaller than control at the 12th week. Based on our study, the intradermal sutures were effective when used on a rat's wound. The effect after intradermal sutures technique process includes (1) local hypoxia due to the compression in the wound sutures. Hypoxia can inhibit angiogenesis; (2) the intradermal suture will trigger blood vessels pinch, microvascular and endothelial damage, and vascular endothelial growth factor cascade disruption; (3) the control groups without intradermal sutures treatment can have excessive wound stretch, triggering the recurrent inflammation, and activating the mast cell signal to produce fibroblasts, which leads to collagen accumulation.[2,3,15]

The fibroblasts accumulation occurs due to hypoxia and mechanical load, which induces more collagen production. Wound healing requires an adequate oxygen supply. Enhancing the oxygen supply will increase collagen accumulation, stimulate angiogenesis, and ultimately accelerate wound healing.[18-19] The condition around the wound is always in a hypoxic state and angiogenesis stimulation is greater in the ischemic tissue compared to non-ischemic. The metabolic crisis in the wound will occur when a blood vessel splits, which can cause hypoxia and acidosis conditions. The cells in the worst hypoxia and acidosis conditions will die or have metabolic alteration; the production of chemical signals initiating the wound healing process.[20-21] Meanwhile, the wound stretching as a mechanical load can trigger a stimulus that is converted into chemical activity or activates the Protein-kinase B (Akt) pathway. Mechanical load occurs because of the mechanical stress on the wound.[22] The Akt pathway produces B-cell lymphoma (Bcl) 2-associated death promoter (BAD) phosphorylation in phospho-BAD (Ser136) and the activation of nuclear factor kappa-light-chain-enhancer of activated B cells (NF-κB) through IκB kinase (IKK).[23] The combination of hypoxia and mechanical load can decrease fibroblast apoptosis, increase collagen synthesis, and decrease collagen degradation.[24-26]

CONCLUSION

In this study, surgical wounds with intradermal sutures had decreased fibroblasts and VSS value compared to those without intradermal sutures. Meanwhile, there were more fibrocytes in the surgical wounds with intradermal sutures than without intradermal sutures. The finding of this study indicates that intradermal sutures using polypropylene thread produce a better clinical result.

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