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COMPARISON EFFECT OF MICRONEEDLING WITH A ROLLER DEVICE WITH MCFARLANE METHOD ON THE VIABILITY OF RANDOM SKIN FLAPS IN RATS

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Abstract:

Background: *Flap necrosis is considered an important complication of reconstructive surgery. The current study was conducted to assess the effect of "Dermaroller" on the survival of random skin flap by determining the degree of flap necrosis in the distal area and the degree of vascularity in the dermis.*

Methods: *30 rats were randomized into two groups. In the first group or the case group, the posterior random skin flap was done with dimensions of 3*9 cm. Then, each rat underwent microneedling by dermaroller. In the second group or the control group, the posterior random skin flap was done similar to the first group. In the end of the 7th day after operation, a ruler was inserted and the whole length of the flap was photographed. The area of necrosis of each flap (in cm2) was determined using computer software. Histological analysis was done in two areas (2cm and base of the flap) to determine the number of vessels in these areas. Data were collected and analyzed.*

Results: *30 surgeries were studied. 15 rats were assigned to control group and 15 were assigned to case group. Mean flap necrosis was 37.3 (SD=10.3) in dermaroller group and 39.6 (SD=10.4) in control group. The difference between two groups was not statistically significant (p-value=0.54). Mean number of vessels in 2cm from the base of flap was 0.8 (SD=0.7) in case and 0.6 (SD=0.7) in control group. The difference between two groups was not statistically significant (p-value=0.6).The mean percentages of vessels in the base of the flap were 1.0 (SD=0.8) and 1.2 (SD=0.9) in the dermaroller group. The difference between two groups was not statistically significant (p-value=0.55).*

Conclusion: *We found no statistically significant difference between dermaroller group and the control group. We advise to repeat the study with dermaroller one week before surgery.*

Keywords: *Microneedling, Dermaroller, Random skin flap survival, Rat*

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INTRODUCTION:

Random skin flap is one of the most popular operations in the plastic and reconstructive surgery(1).

One of the complications of these types of flap is flap necrosis, especially in the distal part of the flap. Subdermal arterial plexus is responsible for the skin n flap blood supply. Flap necrosis is still one of the most important complications in the reconstructive surgeries. This issue can increase the length of hospitalization, treatment costs, and failure in obtaining the best results and It can decrease the patient Satisfaction. We can apply some adjustments to facilitate the flap usage and increasing the operation usefulness.

Random skin flap has some limitations like length-to-width ratio constraints, arc of rotation limitations, proximity of the flap to the injury. Unfortunately, distal flap necrosis is irreversible despite of detecting the mechanisms and designing the flap properly, flap necrosis is not always preventable(2,3).

Up to now, many different techniques are used to increase the skin flap survival. Microneedling is a new method which increases the blood flow in the skin derm layer. This technique seems to be a new method for increasing the flap viability but a few researches has been done related to the efficiency of this technique.

Microneedling with dermaroller is a medical technique, specially for facial rejuvenation. Microneedling can rupture dermal small vessels and then platelets are released afterward. These platelets release growth factor. Blood flow is increased in this area subsequent to the inflammation (4). This research is done due to the surveying of skin flap viability and determining the amount if dermal blood vessels and distal necrosis.

METHOD:

This study was performed on on sprague-dawley rats purchased from Razi institute. All rats were male, with same age, with no disease and they all weighed between 350 to 400 grams.

30 rats were used for the study, randomly divided to 2 same groups.

Each group had 15 rats and they were chosen with calculating the sample size.

Study type was interventional. All rats were kept due to the guide of care and use of laboratory animals, 7th edition published by NRC 1997,ARENA/OLAW, institutional animal care and use committee guide book,2nd edition 2002. After the operation, rats were kept separately, to avoid them from licking their wounds, with access to water and food separately. Time of transporting them to the lab and their residence time in the lab until the operation was equal.

After randomly dividing them into 4 equal groups, all rats were anesthetized with intramuscular injection of ketamin 10% with dosage of 90 mg/kg and xylazine 2% with dosage of 9 mg/kg. Anesthetic dosage was repeated if needed. Then hairs of posterior side of the rats were shaved with a rechargeable shaver this place was disinfected with the solution of alcohol and betadine. Then prophylaxis antibiotic cephazoline was intramascularly injected with the dosage of 60 mg/kg. After confirming the depth of anesthesia with withdrawal pinch flexion test, the operation was started all asepsis measures taken.

In the first group or the control group, the posterior random skin flap was done according to the

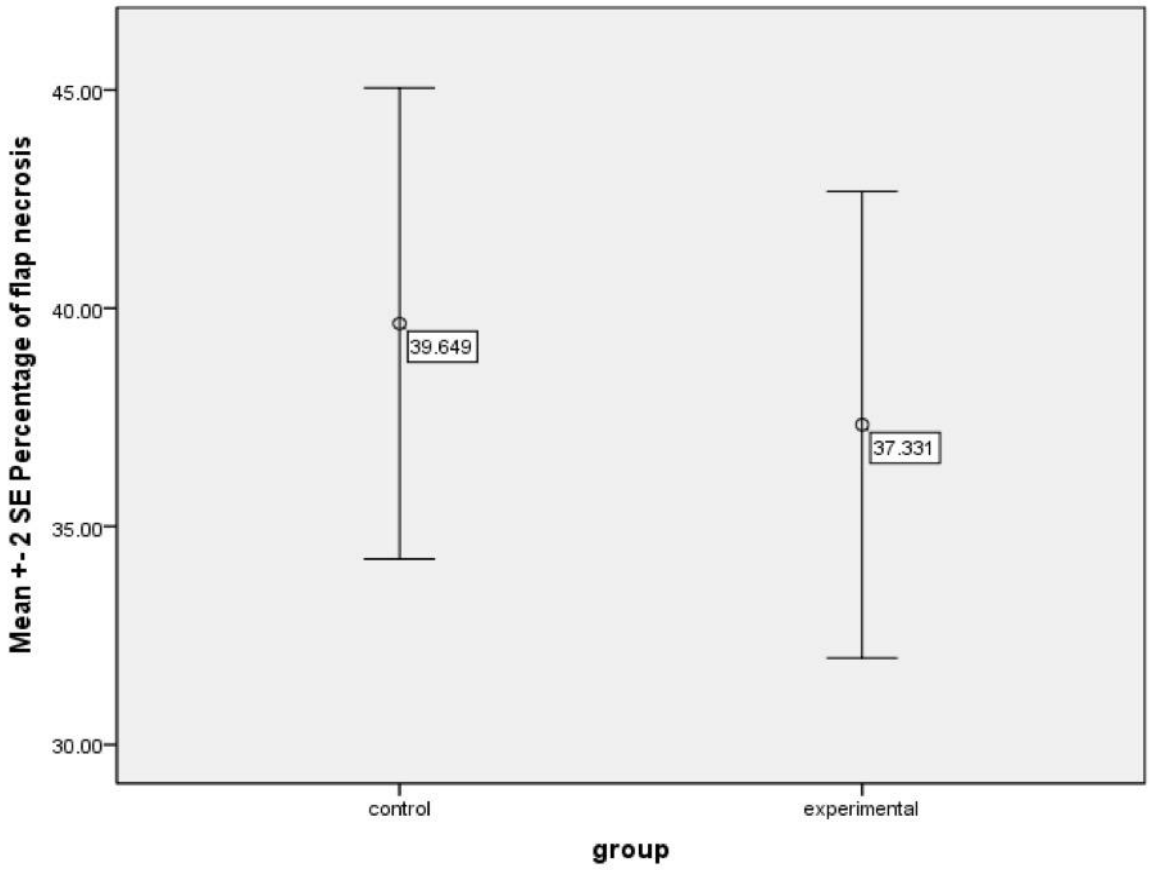
mc. Farlane changed method with using a plastic template with dimensions of 3*9 cm in which every centimeter was determined for the suture place. Hip joint was chosen as the base point of skin flap. Skin cut was performed by the surgical blade number 15 and the akin was cut deep the the parsiculus carnosus. Any peripheral or axial vessel was blocked and a transparent sterilized plate was attached to the wound bed to prevent the graft effect. Then the skin flap was returned to its first place and was sutured with nylon number 04. Then 0.5 cc of normal saline was injected to the central point of the 1⁄3 part of the flap. In the second group , after the operation with the same method , microneedling dermaroller was performed with needle size of 0.5 mm in this order: 3 days before the operation, the operation day, 3 and 6 days after the operation, microneedling with dermaroller was performed 15-20 times in vertical, horizontal and oblique directions until the pointing bleeding was started. At the end of the day 7, all rats were sacrificed with the painless technique(CO2 gas) then topography was done with a digital camera.

After sacrificing all the rats, a transparent flat glass with a deployed ruler beneath the glass(for calibration) was placed on the rat and then a full frame shot was taken all the flap and the ruler above by Nikon digital camera D300 and the Nikon 60mm micro lens with 1:10 magnifying and distance of 80cm. Pictures was transferred to the computer and surveying of the flap and the necrotic part was measured with the NIH , USA ImageJ V.1.40 g app and calibrated as square centimeter. Then histologic evaluation of the flap(necrotic and intact part) was done by optical microscope. Aggregated data was statistically analyzed by the application SPSS 11.5. Normal distribution of the quantitative data was checked. Comparison of quantitative variants between 2 groups was done by independent sample T-test (Mann whitney T-test if needed) and comparison with more than 2 groups was done by

Anova test(kruskl wallis if needed). All researchers were committed the Helsinky treaty. Keeping the mice was as the rules and anesthetics were used in the surgery and sacrificing the rats was completely painless.

RESULTS:

In these research 30 operations was studied. 15 rats were in study group and 15 rats were in the control group. The amount of the necrosis in the study group was 37.3±10.3 and was 30.6±10.40 in the control group. There was no significant differences(p=0.5) (Graph1). The average of newly produced vessels in distance of 2cm from the base of the flap was 0.8±0.7 in the study group and 0.6±0.7 square centimeters in the control group. There was no significant differences(p=0.6) the average of newly produced vessels in distance base of the base if the flap was 1.2±0.9 in the dermaroller group and 1±0.8 in the control group. There was no significant difference. (P=0.55)



Graph1-mean standard deviation of flap necrosis in two control and study group

DISCUSSION:

Owing to our observation, skin flap viability was equal in both with or without using dermaroller. Random skin flap is one of the most popular operations in plastic and reconstructive surgery. Skin flap coordination from the point of color and consolidating and thickness of the flap made this technique more efficient, but distal flap necrosis was always a problem. Angiogenesis increases the flap viability (5). Microneedling effect on skin flap viability was not studied properly before (6,7). Skin microneedling was first used in 1995 by Drentviech for facial wrinkles (6,8). Camirand and Doucet used a tattoo needle for derm abrasion of hypertrophied scars (6,9).

Fernandes used a stamp like needle utensil for collagen making(10,6). there have been a lot of progress in the tools since then. Dermaroller used in this study has multiple needles, penetrating from epiderm to derm(6). Injuries made by this tool cause cell damage which activates keratinocytes, fibroblasts, endothelial cells, macrophages and platelets. At the first stage of healing, activation of immune system and neutrophils and platelets causes the release of cytokines and growth factors like TGF-α and TGF-β ,PDGFs and connective tissue growth factors(6).

These factors make the keratinocytes and fibroblasts grow.

At the second stage of healing, monocytes replace the neutrophils and epithelial cell proliferation, angiogenesis and production of collagen proceeds. Collagen type III, elastins, GAGs and proteoglycans are produced from fibroblasts.Growth factors are released from monocytes at this stage(6). At the third stage of healing, collagenase and protease convert the collagen type III to type type I which stays for 5 to 7 years at its place(11). Recent studies show that growth factors are effective in angiogenesis(12,13).

Kryger study with effect of VEGF on skin flap indicates that VEGF can reduce the ischemia on skin flap (in study group) in comparison with the control group and more angiogenesis in the study group(13). Aust et al. Study shows that subcutaneous collagen induction results in increase of growth factors and collagen I expression(14). Another research from Aust shows that collagen injection results in up-regulation and expression of TGF-β3. In our study, amount of dermal arteries in study didn't have any significant increase in comparison with the control group. Yakakowa et al. Study shows that hypoxia can induce angiogenesis and production of VEGF(12). This study showed that microneedling with dermaroller has no effects on viability of skin flap. Our result was unlike Baris et al.(4) Study result. Maybe monitoring the flaps in a longer time would show the differences more obviously. As a review of our study and comparing our results with other researchers, repetition of the research with dermaroller in the surgery department one week before the operation is advised.

Our study limitations were lack of radio nuclide scientigraphic equipments for more detailed monitoring of blood flow. For more advanced molecular searches, microarray production gene expression analysis is advised, which can facilitate any research in future, make it more convenient and accurate.

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