**The Effect of Platelete Rich Plasma Combined with Microneedling on Full Venous Outflow Compromise in a Rat Skin Flap Model**

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**Introduction:**

Although, the use of free flap in reconstructive surgery has increased over time[1], the complications related to obstructed venous outflow are common in flap transfer and can result in irreversible tissue injury, necrosis, and flap loss [2]. Microneedling aims to induce as many microwounds in the dermis by the needle pricks as possible rolling needles vertically, horizontally and diagonally with pressure over the treated area[3]. Microneedling initiate the normal phases of wound healing. makes it release of many growth factors and cytokines[4]. Platelet rich plasma (PRP) is a rich source of growth factors, has been found effective in accelerating significant tissue repair and regeneration, and releases massive quantities of platelet growth factors[4].

The purpose of our study is to determine the efficiency of microneedling and PRP enriched microneedling method for alleviating the harmful effect of venous congestion.

**Materials And Methods:** Ten adult male Wistar rats were used to obtain platelet rich plasma. A bilateral an epigastric skin flap based on the superficial epigastric artery and vein were harvested. The animals were randomized into five groups (n=8 each group): sham, control, microneedling (M), PRP applied microneedling (M+PRP), platelet poor plasma (PPP) applied microneedling group (M+PPP). Four hours of complete ischemia was induced (except in sham group) and after the end of ischemia, treatments were applied. Flap necrosis, neuronal and non-neuronal levels of Substance P as well as histological changes in the tissues were evaluated at the seventh postoperative day.

**Results:**

Surviving flap area of M+PRPgroup was significantly higher ($p<0,01$) than all the other groups. Microneeding alone increased the survival of the flap area when compared to the control group (Table 1). In M+PRP group, all epidermal layers were clearly organized and dermal integrity was similar to sham dermiş (figure 1). We also observed that microneeding alone markedly decreased Substance P levels

(p=0,001)demonstrating activation of sensory nerve fibers. which might br involved in healing process (graph 1).

**Conclusion**

M+PRP might be an effective treatment modality to reduce congestion, interstitial edema and increase neoangiogenesis in venous congested skin flaps. Our results suggests that growth factors of PRP in addition to M-induced activation of sensory nerve fibers increases tissue survival and regeneration.

**Refernces**

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**FIGURE LEGENDS**

**Table 1.** Extent of necrosis following treatments. M-microneedling ; PRP: Platelet rich plasma, PPP: Platelet poor plasma

**Figure 1.** Histologic analysis with haematoxylin-eosin (H+E). In M+PRP groıp, all epidermal layers were clearly organized and the junction between epidermis and dermis was shown in obviously. Neoangiogenesis was seen in dermal layer (arrows). And inflammatory cells were realized in endovascular area of dermal and hypodermal layers (arrowhead). Also, connective tissue was more organized compare than the other groups of epidermal and dermal area (double arrowheads figure L,M).

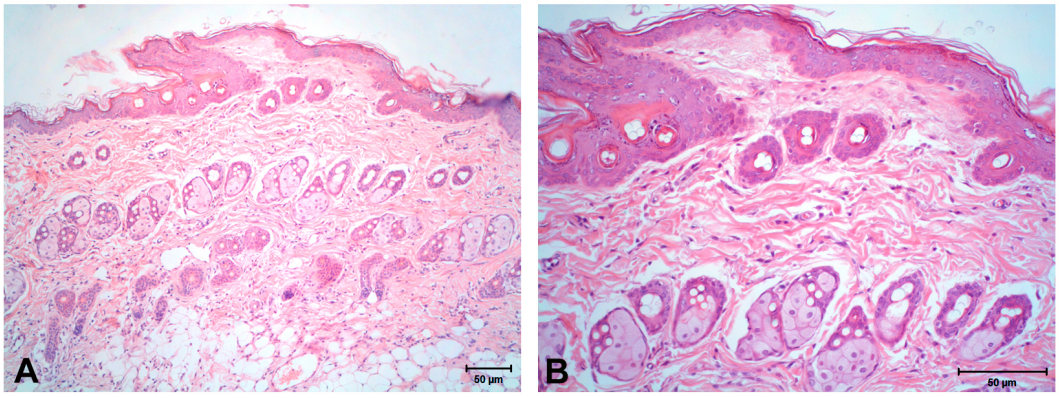
Grapic 1. There was a decrease in the SP levels after 4 hour ischemia applied groups compared with sham operated animals both in first and second extraction ($p<0,001$). Microneedling has trigerred to releasing of SP from both neuronal and non-neuronal tissue. (y axis defines tissue extraction and SP as pg/g).

| Flap Surface Area($cm^2$) | Group | | | | | | | | | | P value |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | Sham | | Control | | M Group | | M+PRP Group | | M+PPP-Group | | |
| | Mean | SD | Mean | SD | Mean | SD | Mean | SD | Mean | SD | |
| **Flap** | 4,50 | 2,45 | 4,50 | 2,45 | 4,50 | 2,45 | 5,00 | 2,74 | 4,50 | 2,45 | 0,991 |
| **Total flap area (Op. Day)** | 11,41 | 1,83 | 8,96 | 1,34 | 11,85 | 2,48 | 12,83 | 1,00 | 11,91 | 2,85 | 0,006* |
| **Intact flap Area (Day 7)** | 9,35 | 0,92 | 3,26 ** | 4,52 | 9,59 | 2,28 | 9,08 | 2,07 | 6,56 | 1,95 | <0.001* |
| **Necrosis (Day7)** | 0,00 | 0,00 | 2,09 ** | 1,90 | 0,71 | 0,56 | 0,00 | 0,00 | 0,63 | 0,66 | <0.001* |
| | | | | | | | | | | | |

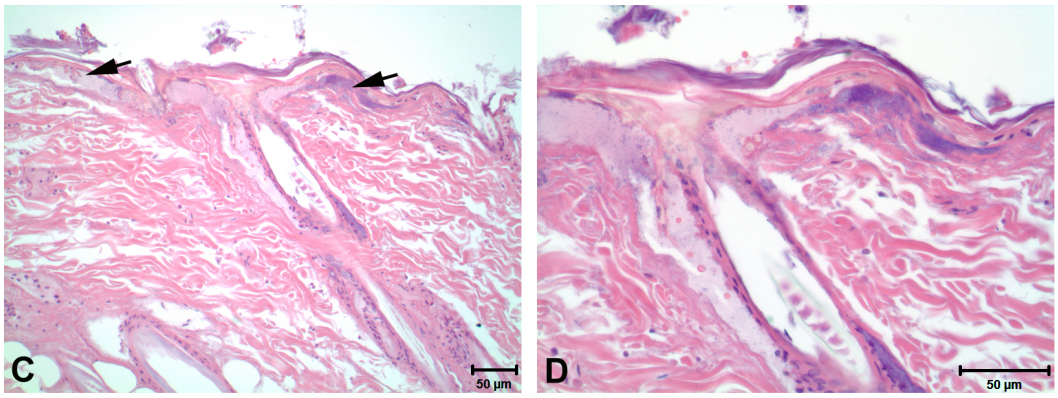
* 0.05 statistically significant
** making difference statistically

## Venous ischemia groups staining with hematoxylin-eosin

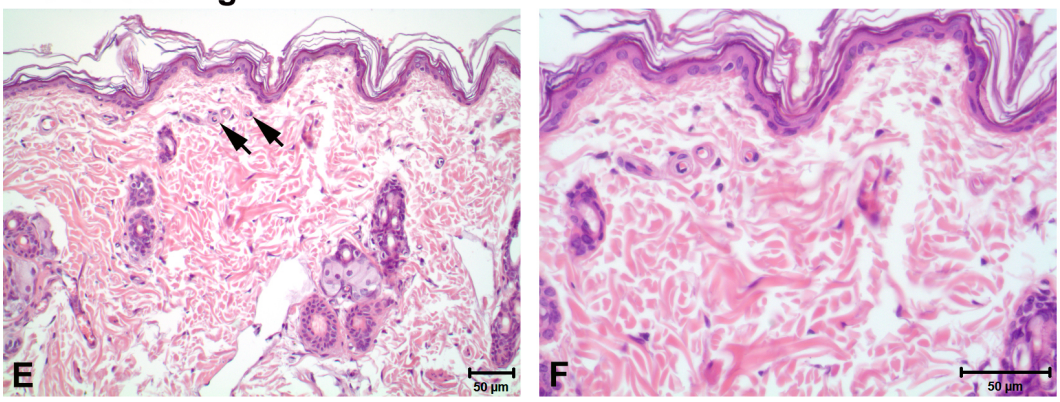
### Sham



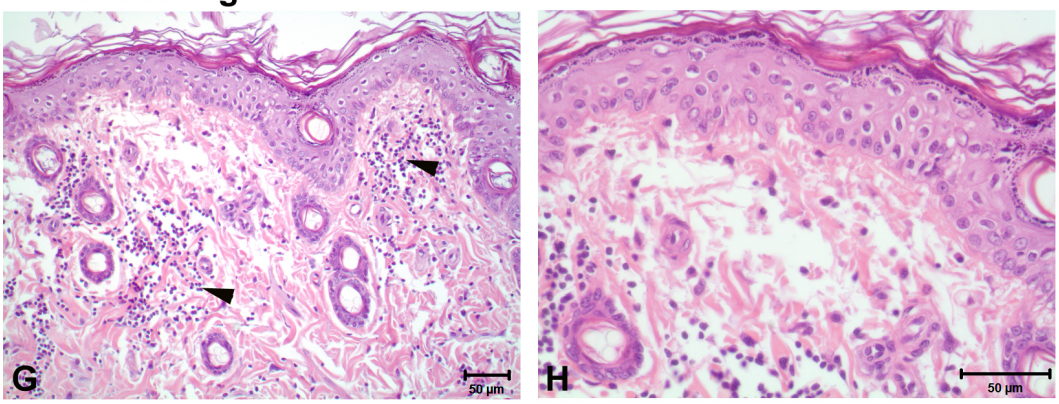

### Control



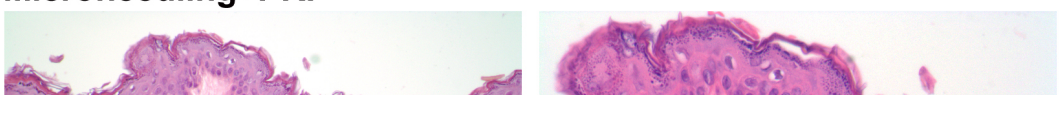

### Microneedling

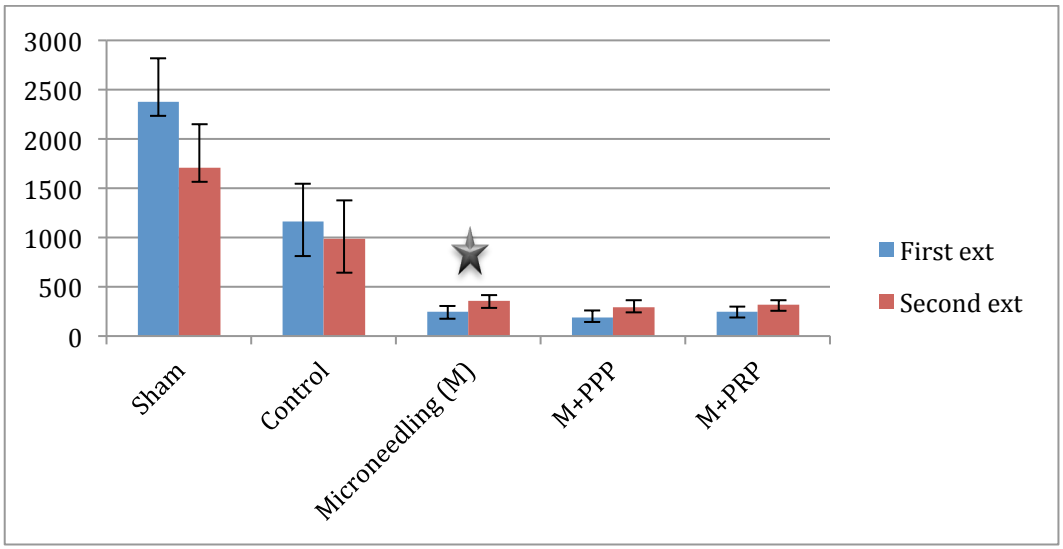



### Microneedling+PPP




### Microneedling+PRP




3000
2500
2000
1500
1000
500
0
Sham
Control
Microneedling (M)
M+PPP
M+PRP
First ext
Second ext