

**XXV National Congress of the "Società Polispecialistica Italiana dei Giovani Chirurghi"
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**A NEW ANGIOGENETIC MODEL FOR THE STUDY OF DERMAL SUBSTITUTES
VASCULARIZATION IN VIVO: OUR EXPERIMENTAL PROJECT**

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Objective: Our first *in vivo* microvascular systems for the study of neo-angiogenesis were limited by several inconveniences due to technical difficulty, high risk of thromboses and position of the regeneration chamber. Here we present our second line of experimental research which aim is to overcome these weaknesses and to practically and definitively assesses the correlation between shear-stress and neo-angiogenesis. The clinical implication of these experiments would be to create new models of prefabricated flaps and bioreactors.

Methods: The hereby present experimental protocol envisages the use minimum of 10 males Wistar rats. After left nephrectomy an end-to-end anastomosis is performed between the renal artery and vein. The microvascular loop is wrapped around by the dermal substitute INTEGRATM folded onto itself and isolated by the outer silicon layer to impede tissue ingrowth. On day 30 after surgery the chamber is exposed. If patency test is positive proximally and distally to the chamber, the latter is send to Pathology for histological analysis (hematoxylin-eosine, Masson Tricomic and immunfluorescence).

Results: Our new microsurgical AV loop experimental model for the study of angiogenesis in the bioengineering templates shows several benefits. Former, the easy way to obtain it thanks to the good caliber of the rat renal vessels and the simplicity of the microsurgical technique. Secondly, the caliber of the vessels and the strong bloodstream prevent thrombosis and allow a fast execution of the sutures. Finally, the regeneration chamber, deeply placed into the abdomen, will be well protected from the rat which will not be able to remove it.

Conclusions: At the current state, results seem to be encouraging. The fact that a new, neo-vascularized tissue is created in a transplantable regeneration chamber offers the attractive possibility of applying microsurgical techniques to prefabricated flap. Besides, the model could be used as a bioreactor for drugs and growth factors release by enriching it of viral gene transfer and bone marrow derived cells. Still it's our opinion that in this model the increase of neo-angiogenesis could be assess just thanks to the shear-stress. It's our purpose to increase the number of cases.

**INTERACTIONS BETWEEN MESENCHYMAL STEM CELLS (MSCs) ISOLATED FROM
PERIPROSTHETIC CAPSULE AND BREAST CANCER CELLS : THE ROLE OF PARACRINE
EFFECT**

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Introduction: MSCs are multipotent cells that can regenerate adult tissue, with potential applications in regenerative medicine. They exert an important paracrine effect on tumor cells by influencing the progression of the tumor with conflicting results, depending on their source of isolation and their degree of differentiation.

MSCs produce soluble factors with regulatory functions. Nowadays breast cancer is the most common malignancy in women. "Oncoplastic surgery" apply great attention to mammary reconstruction with implants after mastectomy. The development of a fibrous periprosthetic capsule is a chronic inflammation, rich in MSCs and essential for the promotion of tumor growth.

Methods: We evaluated interactions between MSCs isolated from periprosthetic capsules of women affected by invasive ductal carcinoma and subjected to breast reconstruction post-mastectomy, and a cell-line of breast cancer (MCF-7). MCF-7 were grown or in presence of MSCs or with the medium previously conditioned by them. We analyzed the proliferative capacity and the expression of genes involved in oncogenic pathways.

Results: MSCs had a double effect on MCF-7: increased the proliferative potential; decreased the aggressiveness of cancer cells. This effect was exercised either directly by the presence in culture of MSCs or by the conditioned medium without MSCs.

Conclusion: This study yields more information about MSCs paracrine effect and provides a preliminary assessment of whether breast reconstruction with implants could influence interactions of MSCs with Tumor Cells. Breast implants cause inflammation that requires a constant presence of MSCs, but they do not seem to promote any recurrence of tumor, that instead appears to be less aggressive. However, the presence of a persistent inflammation suggests to consider periprosthetic capsule as a critical site for the excessive production of soluble factors by Stem Cells. Further studies need to better understand these mechanisms and to correlate the laboratory observations with the clinical outcomes.

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THE PROTECTIVE EFFECT OF CILOSTAZOL ON ISCHEMIA-REPERFUSION INJURY: AN EXPERIMENTAL STUDY IN A RAT TRAM FLAP MODEL

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Objective: Cilostazol is an anti-platelet and direct arterial vasodilator drug, which has also showed other actions: improves endothelial cell function, and reduce the number of activate and/or preconditioned platelets. Recently, its protective effect against ischemia-reperfusion injury has been studied in cardiovascular and neuronal systems. The purpose of the study was to investigate the effect of cilostazol on ischemia-reperfusion injury in a rat transverse rectus abdominis musculocutaneous (TRAM) flap model.

Methods: Forty Sprague-Dawley rats were divided into four groups of 10 animals. The first group was previously undergone to 4 hours ischemic insult then treated with a systemic administration of cilostazol 30-minutes before the flap reperfusion; the second group was treated only with vehicle after 4-hours of ischemic injury; the third group was not undergone to any drug administration after the ischemic insult; the fourth group was not exposed to ischemic injury and the flap was inset after elevation. Four rats from each group was killed to measure flap myeloperoxidase (MPO) activity, malondialdehyde activity, nitric oxide activity at 24 hours after surgery. Laser Doppler blood flow was also measured. At day 7, the remaining rats were evaluated by histological and immunohistochemical analysis and measuring the flap survival rate. Western blot analysis for eNOS was also performed.

Results: The experimental group demonstrated significantly lower MPO activity, higher NO level, but no difference in malondialdehyde activity compared to control group at 24 Hours. A significantly higher flap survival, capillary density and P-eNOS level at postoperative day 7 were also showed. Whereas the treatment group demonstrated a significantly lower ICAM-1 activity at postoperative day 7.

Conclusions: This study demonstrated the protective effect against ischemia-reperfusion injury by down-regulating the expression of adherent molecules such as ICAM-1 and p-selectin, and angiogenic repair of cilostazol in ischaemic limbs through its ability to increase eNOS activation.

LIGASURE FOR CREATING INTESTINAL ANASTOMOSIS

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Objective: To evaluate the feasibility and the effectiveness of LigaSure Forced Triad to create intestinal anastomosis in a ex-vivo porcine models.

Methods: Colon samples were prospectively randomised in two groups (LigaSure Group and Stapler Group). LigaSure Group was divided into nine subgroups according to the power levels of the LigaSure system and the number of radiofrequency application. The anastomoses were tested for early burst pressure. The LigaSure subgroup having the highest burst pressure was compared with Stapler group. Finally the specimen was reviewed by pathologist.

Results: A total of 100 samples were tested. Each LigaSure subgroups (n=9) counted 10 samples; the Stapler group included the remaining 10 samples. The highest burst pressure was obtained in Subgroup g (3 bars-power levels and 1 frequency application) (p<0.001). No significant difference was found between burst pressure of Subgroup-g and Stapler group (42 ± 4.7 versus 42 ± 4.3, respectively, p=0.9). Histological view showed that sealing created by LigaSure was completely formed by collagen sealed without cavitation defects.

Conclusions: Our study seems to confirm the feasibility of creating intestinal anastomosis using LigaSure. If our data are confirmed in vivo models, in the future such technology would undoubtedly have strong clinical implication in this setting.

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**EXPERIMENTAL MODELS OF REPLANTATION IN THE RAT: HOW FAR YOU CAN PUSH THE
MICROSURGICAL TRAINING!**

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Replant surgery requires many technical skills and confidence with different anatomical structures. Bony fixation; tendon repair; venus, artery and nerve anastomosis have to be performed sequentially. The operation is performed in emergency condition and often need to perform numerous anastomoses, venous grafts, free flaps in limb salvage, etc. Thus the conditions for practice are extremely difficult and limited.

Because of the ischemia time it's required to be fast, therefore you can not imagine a successful reimplantation without appropriate training.

In this paper we present different models of replantation performed in experimental microsurgery with an increasing gradient of difficulty and how to prepare to solve common problems that may occur in emergency hand surgery.

We describe the anatomy and the technical execution of the replant of limb, tail and the toe-to-thumb transfer in the rat.

These models represent a microsurgical challenge because of the supramicrosurgical size of the vessels but undoubtedly constitute an excellent training model and really cheap.

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