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IMPLANT MATERIALS USED AS SCAFFOLD FOR RAT MESENCHIMAL STEM CELLS GROWTH: AN IN VITRO STUDY.

Hypothesis / aims of study

Aim of this in vitro study is to test mesh materials normally used for prolapse surgery as scaffolds for the growth of rat Mesenchimal Stem Cells (rMSCs).

Implants have been introduced in pelvic surgery to provide an additional support for the correction of the prolapse. Implant materials can be classified according to the source in synthetic, bio-derived and hybrid. So far an ideal implant material for pelvic surgery has not been identified. Most side effects related to permanent synthetic implant materials result from shrinking, erosion or deformation of the mesh. Biologic materials have been introduced as an alternative to synthetic ones because of their biocompatibility. Potential advantages include in-vivo tissue remodelling, histologic similarity and reduced inflammatory reaction. Bio-grafts however, have potential limitations, including their limited supply, low tissue strength and unpredictable behaviour on medium and long-term.

Mesenchimal Stem Cells (MSCs) have been widely used in tissue engineering to repair and regenerate damaged tissue (1). MSCs are relatively easy to culture, to process and to store. MSCs need a scaffold to grow, and the differentiation process is driven by the micro-environment at the site of implant. In vivo the presence of a layer of MSCs surrounding the surface of the implant can improve its biocompatibility at the interface with host native tissue. This could mean a milder inflammatory reaction against synthetic materials and a quicker remodelling process with improve of the mechanical integration for the biologic materials.

In definitive the presence of a MSCs layer surrounding the implant and at the interface could theoretically modify the host reaction with an improvement of the biologic and bio-mechanic properties of the graft.

Study design, materials and methods

We tested four different implant materials: three biologic (Pelvisoftl® Hydrix® and Surgisis®) and one hybrid (Pelvitex®). Rat MSCs (rMSCs) were isolated from Spague Dawley female rats bone marrow. 1 x 10^6 rMSCs were pipetted and/or injected in sterile conditions on each mesh sample placed in Multiwell culture plates. Microscopic analysis was performed on samples after 7, 14 and 21 days respectively. Samples were cut, embedded and stained. Slides from Pelvisoft, Hydrix and Surgisis were stained with Hematossilin and Eosin and observed at light microscopy, while Pelvitex was stained with DiI and observed at confocal microscopy. and scored on a semiquantitative scale in the case of the viability. Proliferation rate has been classified on a semiquantitative scale as low, moderate or high. Cell viability and proliferation pattern were observed and noted.

Results

No growth of rMSCs was observed on Pelvisoft and Hydrix at any time point. Rat MSCs initially were uniformly distributed on the external surface of Surgisis while along the time it was observed a progressive penetration of rMSCs in the thickness of the material. Rat MSCs at 7 days proliferated on whole surface of Pelvitex, while at 14 and 21 days proliferation was observed mainly at the interstices and around the fibres. When proliferating, rMSCs always showed good viability.

Interpretation of results

In this study it has been showed that biologic materials have different characteristics despite their common origin: both Pelvisoft and Surgisis are porcine-derived and appear like a compact layer of acellular collagen. They differentiate because of source (Pelvisoft derives from skin while Surgisis derives from intestinal submucosa) and cross-linking (Pelvisoft differently from Surgisis is cross-linked). Hydrix is a homogeneous membrane constituted of not cross-linked acellular collagen derived from bovine pericardium. The looser structure of Surgisis, more than the presence of pores or cross-linking seems to be essential for the growth of rMSCs. In our study we tried also to inject rMSCs with an insulin needle inside the biologic materials at different depths without any difference in terms of cell proliferation when compared with pipetting technique. Cells progressively “soaked” Surgisis, with a centripetal proliferation from periphery to the centre.

We considered interesting to test a hybrid implant (Pelvitex) because hybrids belongs to the last generation of mesh materials. The presence of hydrophilic reabsorbable collagen surrounding the polypropilene structure revealed to be essential for the growth of rMSCs.

Rat MSCs were found only at the interstices and around the single fibres of the mesh when the collagen filling the pores was absorbed (mean absorption time: 14 days).

We could conclude that in vitro Surgisis represents a good scaffolds for rMSCs. In our agenda we already planned to check the remodelling processes of Surgisis when used as scaffold for rMSCs in the rat model.

Pelvitex showed good characteristics and we will focus our attention on the assessment of the inflammatory reaction versus this material when implanted with a covering layer of rMSCs.

Concluding message

Certain implant materials normally adopted in urogynecology can be used as scaffolds for the growth of MSCs. Next step will be an in vivo study aimed to check the biological behaviour of this combination.

References

1. Biomaterials (2008) 29; 1017-1027

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