Host Stem Cell Mobilization for *In Situ* Muscle Tissue Regeneration

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Statement of Purpose: Skeletal muscle defects represent a component of injury in a major percentage of traumatic patients. Current treatment options for restoring large skeletal muscle tissue defects due to trauma or tumor ablation are limited by host muscle tissue availability and donor site morbidity resulting from muscle flap implantation. In this study, we sought to utilize muscle satellite/progenitor cells residing in host muscle to regenerate muscle tissue using a target-specific scaffolding system. The objectives of this study were to evaluate the potential of various signaling molecules (myogenic factors) to induce muscle cell migration, proliferation, and differentiation *in vitro* and to investigate the possibility of using an appropriate biomaterial to initiate cell mobilization and recruitment *in vivo*.

Methods: In order to investigate whether muscle satellite/progenitor cells can be migrated into an implanted biomaterial, nonwoven poly(L-lactic acid) (PLLA Scaftex®; density 43 mg/cc, thickness 4 mm) scaffolds were implanted in the leg muscle of SD rats. PLLA scaffolds have a porous fiber structure with high interconnectivity to allow host cell migration. The implanted scaffolds were retrieved after implantation to characterize infiltrating host cells. To evaluate the myogenic factors that affect muscle cells, we selected a series of myogenic factors and applied these to cells *in vitro* to determine whether they could induce cellular migration and proliferation. To evaluate effects of myogenic factors *in vivo*, we implanted porous gelatin-based scaffolds or gelatin microspheres containing myogenic factors. The myogenic factor-containing scaffolds were implanted in the lower leg muscle of rats and were retrieved at 1, 2, and 4 weeks after implantation to evaluate muscle stem/progenitor cells recruitment and remodeled skeletal muscle tissue.

Results: The retrieved scaffolds showed progressive tissue ingrowth over time. By the fourth week after implantation, the scaffolds were completely infiltrated by host cells, including inflammatory cells and stromal mesenchymal-like cells. Pax7 was expressed within the implanted scaffolds at all designed time points. These findings indicate that host muscle satellite/progenitor cells are able to migrate into the implanted scaffolds. In addition, the myogenic factors we tested effectively promoted myogenic cell migration and proliferation in vitro. In addition, the number of cells expressing Pax7 and newly formed muscle fibers (Fig. 1) increased within the implanted biomaterials that contained myogenic factors, suggesting that these factors can be used to mobilize muscle progenitor cells and to remodel muscle tissue within an implant in a muscle injury animal model.

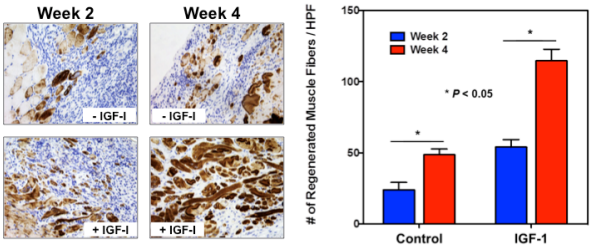

Fig. 1. Immuohistochemistry for myosin heavy chain (MHC) of the implanted IGF-1-immobilized gelatin microspheres.

Conclusions: This study suggests that it may be possible to use the body's biologic and environmental resources for in situ muscle tissue regeneration. We demonstrate that cells expressing muscle satellite/progenitor cell markers can be mobilized into an implanted biomaterial and that these cells are capable of differentiating into muscle cells. Therefore, it may be possible to enrich the infiltrate with specific cell types and control their fate, provided the proper substrate-mediated signaling can be imparted into the scaffold. Thus, in situ regeneration of functional muscle tissue through host cell recruitment may be possible.

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References:

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