INTRODUCTION:
Muscle injuries are the most common musculoskeletal injuries encountered by physicians. Muscle undergoes time-dependent phases of healing, which ultimately result in residual fibrosis in the injured area. Fibrous tissue impedes the muscle healing process by forming a physical barrier that limits myoblast migration and fusion, which in turn creates an unsuitable environment for regenerating myofibers (1). This process predisposes the muscle not only to diminished function, but also to re-injury. Although researchers have investigated the prevention of fibrosis through the prophylactic use of anti-fibrosis agents (e.g., decorin and suramin), clinical scenarios likely would require the treatment of fibrous tissue after its development (2,3). The therapeutic use of exogenous matrix metalloproteinases (MMPs), a family of naturally occurring proteases involved in cell migration and extracellular matrix maintenance (4,5), may improve recovery after muscle injuries by promoting the digestion of existing fibrous tissue and releasing local growth factors, thereby enhancing the growth of regenerating myofibers and their migration into the area of injury.

MATERIALS AND METHODS:
In this study, bilateral gastrocnemius (GM) lacerations were created in 21 SCID mice (42 muscles). Twenty-five days after injury (i.e., at the time of peak posttraumatic fibrosis), 5x10^5 C2C12 cells (myoblasts) transduced with the LacZ reporter gene were injected along with 100ng of exogenous MMP-1 directly into the right GMs at the site of injury (21 muscles); the same type of cells (5x10^5) was injected along with PBS (control) at the site of injury in the left GMs (21 muscles). Fourteen days after transplantation, all GMs were examined histologically via X-gal, hematoxylin and eosin (H&E), and trichrome staining. Only muscles with LacZ-positive cells (i.e., those exhibiting evidence of therapeutic cell delivery into the injured area), as confirmed by X-gal staining, were used for analysis. The average number of regenerating (centronucleated) myofibers within the area of injury was determined by H&E staining of the muscle sections (20X magnification). Trichrome staining was used in combination with image analysis software to calculate the percent cross-sectional area of fibrous tissue present within the zone of injury.

RESULTS:
We analyzed 16 muscles treated with MMP and 17 control muscles. The MMP-treated limbs contained significantly more regenerating myofibers in the area of injury than did the control limbs (MMP: 170±100 fibers, Control 62±50 fibers; p<0.001; Figs. 1 and 2). We also observed significantly less fibrous tissue within the injury zone in MMP-treated muscles than in control muscles (MMP: 23±11% of the total injury area, Control: 35±14% of the total injury area; p<0.05; Fig. 3). In addition, the area occupied by regenerating myofibers (i.e., the site of myoblast migration) appeared to be larger in the MMP therapy group than in the control group.

CONCLUSIONS:
These results, generated in a muscle laceration model, suggest that the direct injection of MMP-1 into the zone of injury during fibrosis can enhance muscle regeneration by increasing the number of myofibers and decreasing the amount of fibrous tissue. The use of MMPs may expand the therapeutic options that are clinically available for treatment of severe skeletal muscle injuries and may ultimately enable improved functional recovery of injured muscles by inhibiting the formation of fibrous tissue within injured skeletal muscle.

ACKNOWLEDGMENTS:
This research was supported in part by the National Institutes of Health (R01 AR 47973-01), the Henry J. Mankin Endowed Chair at the University of Pittsburgh, the William F. and Jean W. Donaldson Chair at Children’s Hospital of Pittsburgh, and the Hirtzel Foundation.

REFERENCE: