FRESH SKELETAL MUSCLE FIBRES AS INTERNAL FILLER OF CHITOSAN TUBES FOR PRIMARY AND DELAYED NERVE REPAIR

Benedetta Elena Fornasari[1,2 *], Giulia Ronchi[1,2], Stefania Raimondo[1,2], Alessandro Crosio[3], Claudia Alexandra Budau[1], Pierluigi Tos[4], Isabelle Perroteau[1], Bruno Battiston[3], Stefano Geuna[1,2], Giovanna Gambarotta[1]

[1] Department of Clinical and Biological Sciences, University of Turin, Italy

[2] Neuroscience Institute Cavalieri Ottolenghi, University of Turin, Italy

[3] Microsurgery Unit, AOUS Città della Salute e della Scienza, CTO,Turin, Italy

[4] UO Microchirurgia e Chirurgia della Mano, Ospedale Gaetano Pini, Milan, Italy

INTRODUCTION

Nerve fibre regeneration and complete functional recovery after peripheral nerve injury do not always occur and can be influenced by many factors, including time interval that elapses before performing surgical repair. The poor outcome occurring after a long delay can be due to loss of neuron ability to regenerate, loss of Schwann cell ability to support regeneration and the consequent ineffective support for nerve regeneration. One of the most important factors regulating Schwann cell action is Neuregulin1 (NRG1), which is highly up-regulated following acute nerve injury, while it is strongly down-regulated during chronic nerve degeneration. Taken together these data suggest that providing a source of soluble NRG1 might be a good strategy to improve the outcome after delayed nerve repair.

METHODS

Median nerve of adult Wistar rats was repaired with autologous nerve grafts or 10mm hollow chitosan tubes or 10 mm chitosan tubes filled with a longitudinal piece of *pectoralis major* muscle (muscle-in-tube).

In a preliminary study, the median nerve was transected and immediately repaired. Regenerating nerve samples were harvested at early (up to 28 days) and late (3 months) time-points for biomolecular and morphological analyses and for stereological analysis respectively.

In a subsequent study, rats were denervated for 3 months: the median nerve was transected and sutured on itself to prevent spontaneous reinnervation of the target organ. After 3 months of denervation, the median nerve was repaired with the same repair procedures of the preliminary study. Functional tests were performed. Regenerating nerve samples were harvested 6 months after the delayed repair for morphological and stereological analyses.

RESULTS

By biomolecular analysis, we verified that fresh skeletal muscle produces and releases high levels of soluble isoforms of NRG1 and for this reason we chose it to enrich hollow chitosan tubes. Biomolecular analysis confirmed NRG1 expression *in vivo* at early time-points after muscle-in tube repair. Functional assay and stereological analysis, performed on the distal part of regenerated nerve 3 months after nerve repair, demonstrate that the muscle-in-tube promotes nerve regeneration well as the hollow chitosan tube, in primary nerve repair of short gaps (10 mm).

After a delayed repair, functional recovery was assessed for the three experimental groups. Morphological and stereological analyses are still in progress to evaluate the number and the size of regenerated myelinated fibers in the nerve distal stump.

CONCLUSIONS

For short gaps (10 mm), primary repair with hollow chitosan tube is effective. The enrichment of the hollow tube with fresh skeletal muscle might improve peripheral nerve regeneration after delayed repair, thanks to the release of NRG1.

KEYWORDS

peripheral nerve repair, chitosan, Neuregulin1