Double Layer Nerve Conduit for Peripheral Nerve Regeneration
Using Grooved & Conductive Yarns

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Introduction:
Every year several 100,000 people are affected by peripheral nerve defects, and many of these patients remain disabled for the rest of their lives. Currently, artificial nerve conduits (NCs) are considered as a promising therapy. However, some commercial NCs are too rigid or fail to maintain a mechanically stable architecture during the regeneration process, which can result in a chronic inflammatory response and compression of the NC [1]. Some NCs fail to meet the desired range of pore size and permeability, resulting in either infiltration of inflammatory cells into the conduit, or inhibition of nutrients & oxygen exchange [2]. The overall goal of this study was to design and fabricate a flexible, conductive and semi-permeable tubular structure with improved compression resistance using a double layer micro-braiding technology with either round or grooved fibers aligned longitudinally inside. It was anticipated that this double-layer nerve conduit design would promote Schwann cell attachment and migration for the regeneration of functional peripheral nerves.

Materials and Methods:
The first objective was to braid a two layer structure with a dense outer layer and a porous inner layer using 170-denier poly (L-lactic acid) (PLA) yarn on a 16-spindle braiding machine (Fig. 1).

![Fig. 1. (a) Winding of yarns for braiding (b) Side view and (c) Front view of 16-spindle braiding machine](image)

In order to get the desired pore size range, various yarns and feeding rates were used to braid 1.5mm and 2mm outer diameter tubes. To fabricate the inner layer, we first braided the 170-denier PLA yarn around a core of collagen coated grooved fibers and 10 sutures (O.D. 1.5mm). Then we braided the outer layer (O.D. 2.0mm) around the braided inner core layer with additional sutures. To stabilize the structure, the nerve conduits (NC) were heat-set in an oven at 65°C for 15 minutes. Finally, the sutures were removed to form a double layer tubular nerve conduit with aligned grooved fibers inside the central core. Tenacity, compression resistance and suture retention strength testing were performed to evaluate mechanical properties of the NCs. Schwann cells were seeded at one end of the grooved fibers. MTT assay, SEM analysis and confocal microscopy were used to observe the cell performance and the rate of migration along the grooved and round fibers.

Results:
The average pore size decreased as the yarn feeding rate increased and the diameter of the NC decreased. Figure 2 shows the compression resistance of the four different NC structures with and without round and grooved (4DG) fibers.

![Fig. 2. Results for compression resistance](image)

Schwann Cells were seeded on the collagen coated grooved and round fibers and cultured for 14 days. At Day 14, the Schwann cells were stained with DAPI and were observed in a laser confocal microscope to evaluate their attachment to the fibers (Fig. 3).

![Fig. 3. LCSM image of DAPI stained cells seeded on collagen-coated grooved fibers at 14 days (a). 2D longitudinal image showing cell distribution along fibers; (b) 3D image of cell distribution at seeded end of collagen coated grooved fibers.](image)

Conclusions: Tubular NCs with the desired range of pore size and diameter can be fabricated by micro-braiding technology. The double layer NC design significantly improved the compression resistance, and the larger surface area of the grooved fibers enhanced cell attachment. In the future work we plan to introduce conductive carbon nanotubes [3] together with the grooved fibers to add some level of conductivity for axons to migrate along the channels and encourage Schwann cells to proliferate and form functional “Bands of Büngner” and the regeneration of peripheral nerves.

References: