Peripheral Nerve Regeneration – an Appraisal of the Current Treatment Options

Dragoș CINTEZA\textsuperscript{a,b}; Iulia PERȘINARU\textsuperscript{b}; Bogdan Mircea MACIUCEANU ZARNESCU\textsuperscript{a,b}; Dan IONESCU\textsuperscript{a}; Ioan LASCAR\textsuperscript{a,b}

\textsuperscript{a} Department of Plastic and Reconstructive Surgery, Emergency Clinical Hospital, Bucharest, Romania
\textsuperscript{b} “Carol Davila” University of Medicine and Pharmacy Bucharest, Romania

ABSTRACT

During the last decades significant progress was made in the understanding of the physiopathology of the peripheral nerve regeneration. Although the evolution of therapy is not as spectacular, a series of new treatment solutions were developed. The gold standard in therapy remains the use of autografts. We present the current concepts and therapeutic options available.

Keywords: nerve regeneration, autograft, conduit, bioengineering

INTRODUCTION

The peripheral nerve injury, most commonly post-traumatic, has an incidence of 300.000 new cases/year in Europe \cite{1}. After mechanical, chemical or thermic injury, a gap in the nerve structure results with subsequent loss of innervation of the target organ \cite{2}.

The regeneration is influenced by factors depending on the patient’s biological status (co-morbidities, age), on the mechanism of injury and on the level of the injury (more distal injuries have better clinical outcome) \cite{3}.

Although physiopathological mechanism of the nerve regeneration is better understood now than it was 30 years ago, the clinical treatment has not significantly improved, and the clinical outcome is still unsatisfactory \cite{4}.

Depending on the type and the extent of the damage on the peripheral nerve, the injury was classified by Seddon and Sunderland as neuropraxia (the nerve structure is still in continuity; the recovery occurs in days/weeks), axonotmesis (the nerve structure is still in continuity, but the axons are interrupted and surgical intervention is not necessary; the recovery occurs in weeks/years), neurotmesis (complete interruption of the nerve; surgical repair of the nerve is mandatory; complete recovery is never achieved) \cite{5}.

Following the injury, the distal nerve stump undergoes a complex process, known as the Wallerian degeneration, initiated by the Schwann cells deriving from the myelin sheath, which trans-differentiate into a phenotype characterized by phagocytic activity and in-
increased expression of neurotrophic factors (6). After removal of the resulting debris by the Schwann cells and the macrophages, Schwann cells tend to align forming columns of cells known as bands of Büngner which provide an adequate environment for regeneration and serve as guidance for axonal growth (7).

Although the peripheral nervous system has the regeneration capacity, external intervention is mandatory for sustaining it.

Treatment options

The current methods developed for the treatment of the peripheral nerve injuries can be classified into two major groups: direct coaptation and indirect coaptation.

The direct coaptation is the most frequent method used (performed in 82% of cases), and should be performed in the first 24 h post-injury (1,8). This can be applied in an ideal situation, when the gap between the nerve stumps does not exceed 8 mm (1) and the microsurgical repair of the nerve can be performed without any tension in the suture site. When the gap exceeds 8 mm, the tension in the suture site determines an impairment of the blood flow with subsequent inhibition of nerve regeneration (2).

The indirect coaptation implies the interposition of a graft between the nerve stumps which acts as a regeneration chamber. This provides an adequate environment for the growing axons until they reach the distal nervous stump. The grafts used could be autografts, allografts or nerve conduits.

The nerve grafting remains the “gold standard” clinical treatment for peripheral nerve defects, regardless the size of the gap (2). However, several studies demonstrate that for grafts of 4 cm length, only a small number of axons regenerate across the graft, and for those that surpass 10 cm none of the axons from the proximal stump reaches the distal one (9). The standard technique implies the harvesting of a pure sensory nerve, most commonly being used the sural nerve, and its employment to bridge the nerve gap using microsurgical anastomosis. The disadvantage of this method is the morbidity of the donor site, additional intra- and postoperative risks, the limited graft availability and the limitation of use in motor or mixed (motor and sensory), nerve defects. Motor nerve grafts are more suited for these situations, but the benefit does not surpass the disadvantage of sacrificing the motor function (10).

A method that eliminates most of the autograft disadvantages is the use of allografts- nerve grafts harvested from cadavers, but it comes with the price of the associated risks of immunosuppression (11).

Recently AxoGen® claimed that their allograft named Avance® Nerve Graft has no disadvantages related to immunogenicity due to their decellularized and cleansed extracellular matrix. Their on-going study, the Ranger® Study had more than 600 nerve repairs enrolled in January 2015. The preliminary data showed good recovery rates (with an average of over 78%) in a group of 109 subjects, with 151 nerve repairs performed using Avance® Nerve Graft (12).

The nerve conduits were developed in order to overcome nerve grafting inconveniences.

Initial attempts were made to easily achieve a conduit using various types of tissues available at the injury site, such as arteries, veins or skeletal muscle. The inconvenience of this method was that when arteries or veins were used, the conduit collapsed due to the surrounding structures and when only skeletal muscle was used massive fibrous tissue was formed, impairing the regeneration process (2).

<table>
<thead>
<tr>
<th>Nerve grafts</th>
<th>Conduits</th>
</tr>
</thead>
<tbody>
<tr>
<td>Autograft:</td>
<td>Biological conduits:</td>
</tr>
<tr>
<td>• sural nerve</td>
<td>• artery</td>
</tr>
<tr>
<td>• medial cutaneous antebrachial nerve</td>
<td>• vein</td>
</tr>
<tr>
<td>• terminal branch of the posterior interosseous nerve</td>
<td>• muscle</td>
</tr>
<tr>
<td>• lateral cutaneous antebrachial nerve</td>
<td>• composed</td>
</tr>
<tr>
<td>• saphenous nerve</td>
<td></td>
</tr>
<tr>
<td>• superficial branch of the radial nerve</td>
<td></td>
</tr>
<tr>
<td>Allograft</td>
<td>Artificial:</td>
</tr>
<tr>
<td></td>
<td>• biodegradable</td>
</tr>
<tr>
<td></td>
<td>- collagen</td>
</tr>
<tr>
<td></td>
<td>- gelatin</td>
</tr>
<tr>
<td></td>
<td>- fibrin</td>
</tr>
<tr>
<td></td>
<td>- polyglycolic acid</td>
</tr>
<tr>
<td></td>
<td>- polylactic acid</td>
</tr>
<tr>
<td></td>
<td>- polylactide-caprolactone</td>
</tr>
<tr>
<td></td>
<td>• non-biodegradable</td>
</tr>
<tr>
<td></td>
<td>- polyvinyl alcohol</td>
</tr>
<tr>
<td></td>
<td>- silicone</td>
</tr>
<tr>
<td></td>
<td>- poly-tetra-fluoroethylene</td>
</tr>
</tbody>
</table>

TABLE 1. Type of grafts used in peripheral nerve reconstruction.
In order to surpass these inconveniences the vein or the artery were filled with skeletal muscle, the vessel providing the appropriate environment for the regeneration, limiting the interference of adjacent tissues, and the actin/myosin cytoskeleton of the muscle serving as a guide for axonal growth. Several experimental studies on rats were conducted to prove the utility of this composite biological conduit and of its improved versions by adding bone marrow stromal cells or adipose-derived stem cells in its structure (13,14).

Due to the unsatisfactory results achieved by the use of natural conduits, attempts were made to develop a better conduit which can support the adhesion, migration and function of the local cell (15) and can respect as many properties as possible of an ideal nerve conduit, such as (16):

- biocompatibility
- biodegradability
- permeability and porosity
- protection for axonal growth
- adequate size
- adequate flexibility

One of the first artificial conduits used were non-biodegradable, tubes made of biologically inert silicone, which had the advantage of a very good contention due to their impermeability, but they had high rigidity and determined a foreign body-reaction (11). In addition, the patient had to undergo another surgical intervention for removal of the conduit.

Along with the development of tissue bioengineering, the focus was on creating biocompatible and biodegradable conduits. Currently, conduits are being synthetized from natural derived polymers such as animal collagen (usually type I), laminin, fibrin, fibronectin, hyaluronan, polysaccharides derivates such as chitosan, alginate, agarose. Most of the conduits the FDA or Conformit Europe approved for clinical use are made of type I collagen, such as NeuraGen®, NeuroFlex™, NeuroWrap™, but there are also available conduits synthetized of polyglycolic acid and polylactide-caprolactone (Neurotube®, Neurolac®). Other types of polymer are tested for including them in the structure of various conduits: biodegradable glass and magnesium alloys, nanostructured ZnO ceramic, carbon or Al/Al2O3 nanostructures (15). However, studies on nerve regeneration using FDA approved devices show poor results in clinical recovery compared with autologous nerve graft, and their use is limited to defects under 2 cm (17).

The conduits alone are not sufficient for the nervous regeneration which is why for creating the optimal environment, growth factors and different types of cells are packed within the conduit.

The growth factors influence the phenotypic expression of neural cells, supporting the axonal growth. They can be classified into two categories: neurotrophins (brain-derived neurotrophic factor, nerve growth factor, neuregulin, neurotrophin-3) and growth-factors with neurotrophic action (fibroblast growth factor, insulin growth factor-1, ciliary neurotrophic factor) (3,15).

The cellular component of the artificial nerve graft adds trophic support to the regeneration process in order to enhance the outcome. Initially were used the Schwann cells and olfactory ensheathing cells, but they have limited capacities of expansion. That is why researchers appealed to stem cells of different sources, with unlimited capacity of regeneration and possibility of multilineage differentiation. The most attractive type of cells used are the bone marrow mesenchymal stem cells, adipose-derived stem cells and skin-derived precursor cells (15).

There are some alternative methods under study for enhancing the axonal regeneration. Electrical stimulation of the proximal nervous stump in rats stimulates the Schwann cells proliferation and releasing of neurotrophic factors. Administration of β-D-xyloside inhibits the synthesis of chondroitin sulfate proteoglycan, produced by the Schwann cells immediately after injury which retards axonal growth. Within 4 days after de administration the level of chondroitin sulfate proteoglycan reduces by 90%. Studies regarding administration of immunosuppressants (tacrolimus) in lower doses than required show an increased speed of regeneration, the myelin sheath is by 40% thicker, the number of axons that regenerate increases (10).

A new approach on modulating the regeneration process is the interference with the intrinsic growth mechanism, with molecular targeting strategies by use of RNA with the help of genetic engineering (15).
CONCLUSION

Despite major progress in understanding peripheral nerve regeneration, the gold standard for repairing a nerve defect remains autografting. Until now there are no studies to prove better outcomes than nerve grafting. However there are many ongoing trials that show promising results using various artificial conduits, as well “in vitro” as “in vivo”. Bio and genetic engineering will play an essential role in the progress of peripheral nerve repair.

Conflict of interests: none declared.

Acknowledgement: This paper was co-financed from the European Social Fund, through the Sectorial Operational Programme Human Resources Development 2007-2013, project number POSDRU/159/1.5/S/138907 “Excellence in scientific interdisciplinary research, doctoral and postdoctoral, in the economic, social and medical fields -EXCELIS”, coordinator of the University of Economic Studies, Bucharest.

REFERENCES
7. Scheib J, Hoke A – Advances in peripheral nerve regeneration. Nat Rev Neurol 2013;9:668-676