

Muscle and Peripheral Nerve Response to Laser Phototherapy

Review of 30-years experience

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ABSTRACT

Posttraumatic nerve repair and prevention of muscle atrophy represent a major challenge of restorative medicine. Considerable interest exists in the potential therapeutic value of laser phototherapy for restoring or temporarily preventing denervated muscle atrophy as well as enhancing regeneration of severely injured peripheral nerve.

Low power laser irradiation (laser phototherapy) was applied for treatment of rat denervated muscle in order to estimate biochemical transformation on cellular and tissue levels, as well as on rat sciatic nerve model after crush injury, direct or side-to-end anastomosis and neurotube reconstruction. Nerve cells' growth and axonal sprouting were investigated on embryonic rat brain cultures. The animal outcome allowed clinical double-blind, placebo-controlled randomized study which measured the effectiveness of 780-nm laser phototherapy on patients suffering from incomplete peripheral nerve injuries for 6 months up to several years. In denervated muscle, animal study suggests that the function of denervated muscles can be partially preserved by temporary prevention of denervation-

induced biochemical changes. The function of denervated muscles can be restored, not completely but to a very substantial degree, by laser treatment, initiated at the earliest possible stage post-injury.

In peripheral nerve injury, laser phototherapy has an immediate protective effect, maintains functional activity of the injured nerve for a long period of time, decreases scar tissue formation at the injury site, decreases degeneration in corresponding motor neurons of the spinal cord and significantly increases axonal growth and myelination. In cell cultures, laser irradiation accelerates migration, nerve cell growth and fiber sprouting.

In a pilot, clinical, double-blind, placebo-controlled randomized study in patients with incomplete long-term peripheral nerve injury, 780-nm laser irradiation can progressively improve peripheral nerve function, which leads to significant functional recovery.

780-nm laser phototherapy temporarily preserves the function of a denervated muscle, accelerates and enhances axonal growth and regeneration after peripheral nerve injury or reconstructive procedures. Laser activation of nerve cells, their growth and axonal sprouting can be considered

as potential treatment of neural injury. Animal and clinical studies show the promoting action of phototherapy on peripheral nerve regeneration, which makes it possible to suggest that the time for broader clinical trials has come.

INTRODUCTION

When muscles are denervated, in cases of complete peripheral nerve injury, they deteriorate progressively. Although some muscle regeneration does occur [1], it is at a level insufficient to replace the degenerative loss. There is a need to find effective methods for muscle preservation and nerve regeneration enhancement, especially after surgical nerve repair [2,3]. Surgical repair is the preferred modality of treatment for the complete or severe peripheral nerve injury [4-10]. In most cases, the results can be successful if the surgery is performed in the first six months after injury, in comparison to long-term cases where surgical management is less successful. Nonetheless, in related literature, there are several publications of surgical treatment of long-term injuries (most of which were severe, incomplete and with minimal or partial preservation of muscle activity) of the brachial plexus and peripheral nerve [11-14]. The reason for early surgical intervention has to do with the fact that between 1 and 3 years post-injury, denervated muscle undergoes progressive degeneration, which leads to loss of muscle fibers and their replacement with fat and fibrous connective tissue. For most patients who suffer from long-term peripheral nerve injuries, spontaneous recovery is often unsatisfactory. The usual results after such an injury are degeneration of the distal axons and retrograde degeneration of the corresponding neurons of the spinal cord, followed by a very slow regeneration. Recovery may eventually occur, but it is slow and frequently incomplete. The secondary effects of peripheral nerve injury are wasted muscles and high incidence of pressure sores. Therefore, numerous attempts have been made to enhance and/or accelerate the recovery

of injured peripheral nerves and decrease or prevent atrophy of the corresponding muscles. Among the various proposed methods for enhancing nerve repair, phototherapy has received increasing attention over the last two decades. The term phototherapy refers to the use of light for producing a therapeutic effect on living tissues. Although a pioneering report on the effects of laser phototherapy on the regeneration of traumatically injured peripheral nerves was published in the late 1970s [15], it is only since the late 1980s that scientific interest was kindled in this therapeutic approach for neural rehabilitation, leading to the publication of a number of studies that have shown positive effects of phototherapy on peripheral nerve regeneration [16,17]. The possible mechanism of action of phototherapy on the nervous tissue with respect to peripheral nerve regeneration has been provided by the *in vitro* studies which showed that phototherapy induces massive neurite sprouting and outgrowth in cultured neuronal cells [18], as well as Schwann cell proliferation [19]. Also, it has been suggested that phototherapy may enhance recovery of neurons from injury by altering mitochondrial oxidative metabolism [20], and guide neuronal growth cones *in vitro*, perhaps due to the interaction with cytoplasmic proteins and, particularly, to the enhancement of actin polymerization at the leading axon edge [21]. Phototherapy alters nerve cell activity, including upregulation of a number of neurotrophic growth factors and extracellular matrix proteins known to support neurite outgrowth [22]. A possible molecular explanation was provided by demonstrating an increase in growth-associated protein-43 (GAP-43) immunoreactivity in early stages of rat sciatic nerve regeneration after phototherapy [23]. Another study [24] showed that application of phototherapy upregulates calcitonin gene-related peptide (CGRP) mRNA expression in facial motor nuclei after axotomy. By altering the intensity or temporal pattern of injury-induced CGRP expression,

phototherapy may thus optimize the rate of regeneration and target innervation and neuronal survival of axotomized neurons. In this paper we report the results of an experimental study aimed at investigating how laser phototherapy affects long-term denervated muscles by examining acetylcholine receptors (AChR), which play a special role in neuromuscular transmission, and creatine kinase (CK) content, which is an important enzyme for supplying a source of energy to the muscle. The results of this investigation supplement our previous studies [17] pertaining to the effectiveness of laser phototherapy in treating severely injured peripheral nerve after crush injury, neurotomy, side-to-end anastomosis or neurotube reconstruction, based on our 30 years of research.

PHOTOTHERAPY IN DENERVATED MUSCLE PRESERVATION

Using as a model of study the denervated rat gastrocnemius muscle (*in vivo*), we investigated the influence of low power laser irradiation on CK activity and the level of AChR in denervated muscle in order to estimate biochemical transformation on cellular and tissue levels. Much of the literature on the effects of long-term denervation of mammalian skeletal muscle has focused on experimental studies of total sciatic section in rats [25,26]. In our study [27], rats were chosen for investigation in the vast majority of cases due to their availability, good survival record and ease of treatment. For the surgical procedure Wistar rats were anesthetized and complete denervation of the gastrocnemius muscle was done (cut and removal of 1cm segment of the sciatic nerve). After operation, the rats were divided into 4 groups: Group I - denervated non-irradiated group (15 rats); Group II - denervated laser treated group (15 rats); Group III - intact non-irradiated group (15 rats), and Group IV - intact irradiated group (15 rats). The rats underwent laser treatment (HeNe laser, 35mW, 30 minutes) every day, for 14 days. Low power laser irradiation was delivered

transcutaneously to the gastrocnemius muscle of denervated Group II and intact Group IV. Under general anesthesia, the rats were sacrificed and the gastrocnemius muscle was homogenized.

CK activity was measured by the specific spectrophotometrical method using spectrophotometer at 340nm and a Sigma kit [28,29] 7, 30, 60 and 120 days after denervation in both denervated and intact muscles.

Internal and membrane inserted AChR was quantitated by 125I-alpha-bungarotoxin on the same homogenates [30,31] 7, 30, 60 and 120 days after denervation in both denervated and intact muscles. The data obtained was evaluated as cpm of bound 125I-a-BuTX/mg protein. Radioactivity was assessed with Auto-Gamma Counter in denervated and intact muscles.

A. Creatine Kinase (CK) Activity in Intact and Denervated Rat Gastrocnemius Muscle

Muscle contraction and relaxation require the action of CK. Phosphocreatine, formed by the reaction of this enzyme, constitutes a reservoir of high energy phosphate which is available for quick resynthesis of ATP. This high concentration of ATP is then accessible for muscle contraction. Following muscle denervation, the level of CK and muscle weight decreases [32]. Like others [33], we found [32] that in the control non-irradiated group, denervation of the gastrocnemius muscle reduces creatine kinase activity. The decrease of CK activity in both groups (non-irradiated and laser-treated) progresses to a similar value for 7 days after denervation and is followed by a sharp fall in the non-laser treated group in comparison to the delayed and attenuated decrease of the CK activity in the laser-irradiated group. Thus, in the control non-irradiated group, 30 days after denervation, the amount of CK decreased markedly to 41% of the normal value (intact muscle). In the same time delayed and attenuated decrease of the CK activity was observed in the laser treated group. The CK activity of the laser

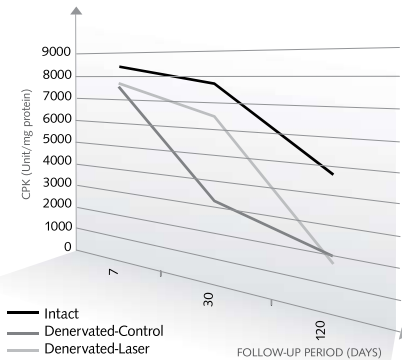


Fig. 1. Graph illustrating the results of study evaluating creatine kinase (CK) activity (unit/mg protein) in intact and denervated rat gastrocnemius muscle. Graph showing content CK (unit/mg protein) during 7, 30 and 120 days in intact and denervated muscles with and without laser treatment.

treated denervated muscle decreased only by 17% of the normal value. The analysis of CK activity in the denervated laser treated group, compared to the control denervated group showed statistically significant difference ($p = 0.008$). After the 30-day period the CK activity gradually began to decrease in both groups and 4 months after denervation it reached similar levels (Fig. 1).

It's known that in denervated muscle the protein degradation rate is accelerated [32]. The temporary prevention of denervation-induced biochemical changes may be prompted by a trophic signal for increased synthesis of CK, thus preserving a reservoir of high energy phosphate available for quick resynthesis of ATP. This data supports Bolognani [34], which shows that laser irradiation increased ATP production in the mitochondria.

B. Acetylcholine Receptors (AChR) Synthesis in Intact and Denervated Rat Gastrocnemius Muscle

Acetylcholine receptors (AChR), which play a special role in neuromuscular transmission, are concentrated at the neuromuscular junction of the adult muscle. A nerve impulse triggers the release of acetylcholine, producing a much larger end-plate potential, which excites the muscle membrane and leads to muscle contraction. The amount of AChR in neuromuscular junction appears

to increase their number and to cover the entire extra-junctional area following muscle denervation. In the denervated muscle, the amount of AChR increases prior to the muscle degeneration [35].

In the control non-irradiated group, 7 days after muscle denervation, as expected, the amount of AChR increased to 161% of the normal value (intact muscle). In contrast, the amount of AChR of the laser irradiated denervated muscle remained near normal value. Thirty days after denervation in laser-treated group amount of AChR increased to 180% compared to 278% in non-laser group. It is interesting that 4 months after denervation, in spite of progressive muscle atrophy, the amount of AChR in laser-treated group remains 53% of normal value compared to only 27% in the non-irradiated group. Statistical analysis showed borderline significance ($p = 0.056$) between denervated laser-treated and non-irradiated denervated muscles (Fig. 2).

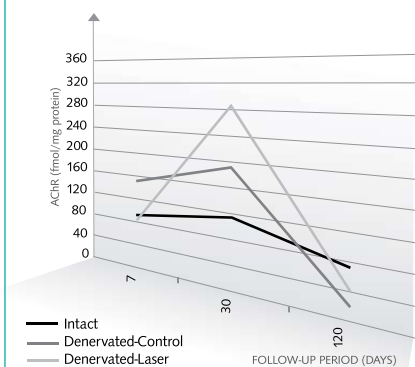


Fig. 2. Graph illustrating the results of study evaluating the level of acetylcholine receptors (AChR) in intact and denervated rat gastrocnemius muscle. Graph showing content AChR (fmol/mg protein) during 7, 30 and 120 days in intact and denervated muscles with and without laser treatment.

Our findings suggest that in early stage of muscle degeneration laser treatment may temporarily preserve the denervated muscle close to its physiological status before injury and during progressive stages of muscle degeneration partially maintain the amount of AChR in the denervated muscle compared to the non-laser treated muscle.

C. Is Laser Phototherapy Damaging to the Muscle?

During 4 months of follow-up period we found no evidence of laser induced damage after irradiation. Moreover, in laser irradiated intact muscle group we found significant increase in CK activity 60 days into the follow-up period ($p=0.008$) and increasing amount of AChR ($p=0.0008$) compared to non-irradiated intact muscle. These findings suggest a possible positive therapeutic effect of laser phototherapy on the muscle.

D. Can Laser Phototherapy Prevent Denervation Muscle Atrophy?

Late denervation has been widely studied in animal models. In rats it has been shown that for the first 7 months after denervation myofibers exhibit a net loss of nuclear domains followed by nuclear groupings [36]. If not re-innervated, the regenerating myofibers undergo atrophy and degeneration [37]. For decrease or temporary prevention of this process, especially in cases of complete peripheral nerve injury, where affected nerve is reconstructed by grafts, tube or primary anastomosis, laser phototherapy can be an effective tool that preserves denervated muscle until nerve sprouting into the muscle occurs. This experimental study suggests that the function of denervated muscles can be restored, not completely but to a very substantial degree, by laser treatment, initiated at the earliest possible stage post-injury. These findings could have direct therapeutic applications for possible treatment of denervated muscles.

PHOTOTHERAPY IN PERIPHERAL NERVE REGENERATION

Posttraumatic nerve repair continues to be a major challenge of restorative medicine. Although enormous progress has been made in surgical techniques over the past three decades, functional recovery after severe lesion of a major nerve trunk is often incomplete and sometimes unsatisfactory.

A. Incomplete Peripheral Nerve Injury

1. Experimental Peripheral Nerve Crush Injury

Under general anesthesia the rat sciatic nerve was exposed and crushed with applied pressure of 6.3 ± 0.7 MPa of an ordinary closed hemostat for 30 seconds. Studies investigating the effects of low power laser irradiation on injured peripheral nerves of rats have found that it provides:

- 1) immediate protective effects which increase the functional activity of the injured peripheral nerve [38];
- 2) maintenance of functional activity of the injured nerve over time [39];
- 3) decrease or prevention of scar tissue formation at the site of injury (Fig. 3) [40];

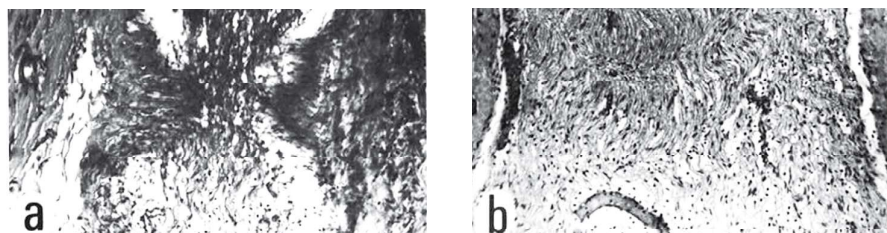


Fig. 3. Histological section of the crush area of the rat sciatic nerve showing the response of the nerve to laser phototherapy. **a.** Nonirradiated nerve. Note of the scar of fibrous tissue; **b.** Laser-treated nerve shows no visible scar. H & E, original magnification X 150.

- 4) prevention or decreased degeneration in corresponding motor neurons of the spinal cord (Fig. 4) [41];

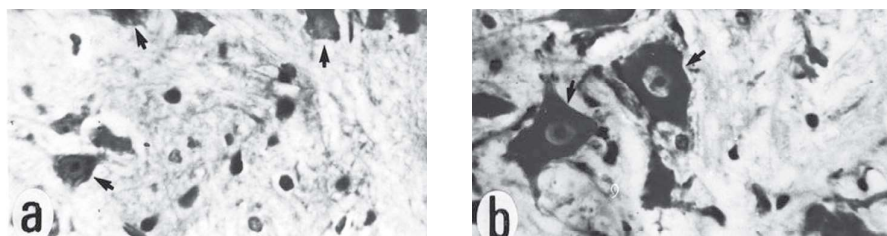


Fig. 4. Paraffin section from the anterior horn of corresponding segments of the rat spinal cord 14 days after crush injury to the sciatic nerve, showing the spinal cord response to laser treatment of the injured peripheral nerve.

a: Section from a control animal shows extensive chromatolysis and cytoplasmic atrophy found in 40% of the motor neurons (arrows). **b:** Section from a laser-treated animal shows minimal degenerative changes found in 20% of the motor neurons (arrows). Stained by cresyl fast violet, magnification X800.

- 5) increase in rate of axonal growth and myelination (Fig. 5) [39]. Moreover, direct laser irradiation of the spinal cord improves recovery of the corresponding injured peripheral nerve [42]. These results, as those of Anders [43], suggest that laser phototherapy accelerates and improves the regeneration of the incomplete injured peripheral nerve.

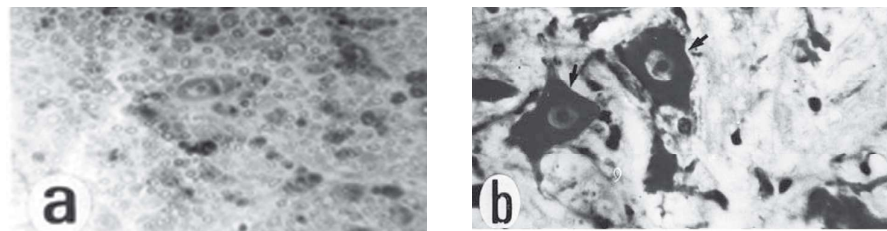


Fig. 5. Photomicrographs of semithin sections stained with toluidine blue showing the axonal response to laser treatment of the injured (crushed) peripheral nerve in rat. One group of rats was treated using laser phototherapy for 20 consecutive days after injury. Twenty-one days after injury the nerves were excised and stained. **a:** Site of crush injury in an untreated nerve showing nerve fibers that appear to be smaller and mostly nonmyelinated. Numerous macrophages and phagocytes are observed. **b:** Site of crush injury in a laser-treated nerve demonstrating that most axons are ensheathed with myelin and a very few infiltrating macrophages are observed. Magnification X300.

B. Complete Peripheral Nerve Injury

1. Regeneration of the Transected Sciatic Nerve in the Rat after Primary

Anastomosis

In acute cases where a peripheral nerve is completely transected, the treatment of choice is direct anastomosis. Means of enhancing regeneration are essential, since degeneration is always inevitable in severely damaged peripheral nerves.

The therapeutic effect of 780-nm laser irradiation on peripheral nerve regeneration after complete transection and direct anastomosis of rat sciatic nerve was evaluated in double-blind randomized study [44]. After surgery, 13 of 24 rats received post-operative laser treatment, applied transcutaneously for 30 minutes on a daily basis, for 21 consecutive days - 15 minutes to the injured sciatic nerve and 15 minutes to the corresponding segments of the spinal cord. Positive somato-sensory evoked responses were found in 69.2% of the irradiated rats ($p=0.019$), compared to 18.2% of the non-irradiated rats.

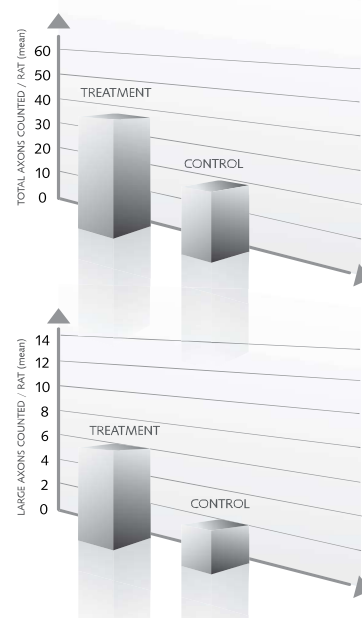


Fig. 6. Bar graphs illustrating the results double-blind randomized study evaluating regeneration of the transected rat sciatic nerve after suturing and postoperative low power laser treatment.

a. Graph showing a statistically significant increase in the total number of axons in the laser-treated group ($p=0.026$), compared with the nontreated control group.

b. Graph showing a statistically significant increase in large diameter axons in the laser-irradiated group ($p=0.021$), compared to the non-irradiated control group.

Immunohistochemical staining in the laser-treated group showed an increased total number of axons ($p=0.026$) and better quality of regeneration process, which became evident by an increased number of large diameter axons ($p=0.021$), compared to the non-irradiated control group (Fig. 6). The study suggests that postoperative laser phototherapy enhances the regenerative processes of peripheral nerves after complete transection and anastomosis.

2. Regeneration of the Sciatic Nerve in the Rat after Complete Segmental Loss and Neurotube Reconstruction

In cases where peripheral nerve is injured and complete segmental loss exists, the treatment of choice is nerve reconstruction using an autogenous nerve graft. The use of a regenerating guiding tube for the reconstruction of segmental loss of a peripheral nerve has some advantages over the regular nerve grafting procedure.

This double-blind randomized study was done to evaluate the efficacy of 780-nm laser phototherapy on the acceleration of axonal growth and regeneration after experimental peripheral nerve reconstruction by guiding tube [45]. The 5mm segment of the right sciatic nerve was removed and proximal and distal parts were inserted into an artificial neurotube (Fig. 7).

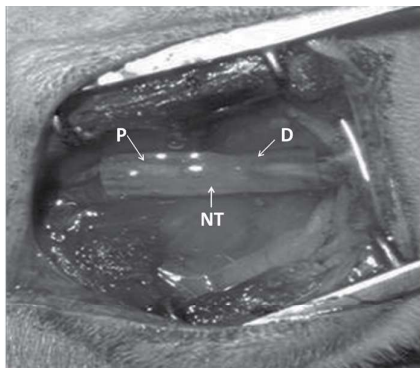


Fig. 7. Intraoperative photograph of the neurotube (NT) reconstruction procedure. A neurotube placed between the proximal (P) and the distal (D) parts of the rat sciatic nerve for the reconnection of 0.5 cm nerve defect.

The rats were divided into two groups - laser treated and non-laser treated. Postoperative low power laser irradiation was applied transcutaneously for 30 minutes: 15 minutes on the transplanted peripheral nerve area and 15 minutes on corresponding segments of the spinal cord, during 14 consecutive days. Conductivity of the sciatic nerve was studied by stimulating the sciatic nerve and recording the somato-sensory evoked potentials (SSEP) from the scalp.

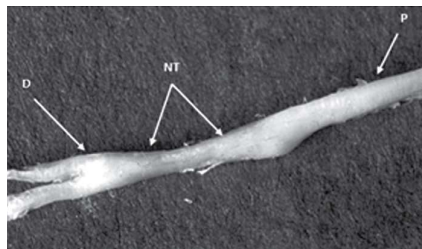


Fig. 8. Photograph of the sciatic nerve of an adult rat 3 months after neurotube (NT) reconstruction. The neurotube recreated the anatomical connection of the previously transected and divided nerve, and a distance of 0.5 cm was recreated.

Three months after surgery SSEP were found in 70% of the rats in the laser-treated group in comparison with 40% of the rats in the non-irradiated group. Morphologically, the transected nerve had good reconnection in both groups and the neurotube had dissolved (Fig. 8). The growth of myelinated axons, which crossed through the composite neurotube, was found and the continuation of axonal sprouting through the area of the tube to the distal part of the nerve was recognized. The laser treated group showed more intensive axonal growth compared to the non-irradiated control group.

PHOTOTHERAPY ON NERVE CELL GROWTH IN VITRO AS A POTENTIAL PROCEDURE FOR CELL THERAPY

Neuronal loss and degeneration resulting from peripheral nerve injuries has led us to explore the possibility of using laser phototherapy on cells, as a method of preventing or decreasing this phenomena. Rochkind [46] investigated the effect of 780-nm laser phototherapy on sprouting and cell size of embryonic rat brain cells,

which were grown on microcarriers (MC) and embedded in neurogel.

Cell cultures: Whole brains were dissected from 16-days old rat embryos (Sprague Dawley). After mechanical dissociation, cells were seeded directly in neurogel, or suspended in positively charged cylindrical MC. Single cell-MC aggregates were either 780-nm laser irradiated within one hour after seeding, or cultured without irradiation.

Neurogel (hyaluronic acid and laminin) was enriched with growth factors: BDNF and IGF-1 [47].

780-nm Low Power Laser irradiation of 10, 30, 50, 110, 160, 200 and 250 mw were used to optimize energy density for activation of nerve cell cultures. Dissociated cells or cell-MC aggregates embedded in neurogel, were irradiated for 1, 3, 4 or 7 min.

Fluorescent staining: Cultures were fixed with 4% paraformaldehyde and incubated with antibodies against neural cell marker: Mouse anti Rat microtubule associated protein. Cells were then washed and incubated with Texas Red conjugated goat anti mouse IgG.

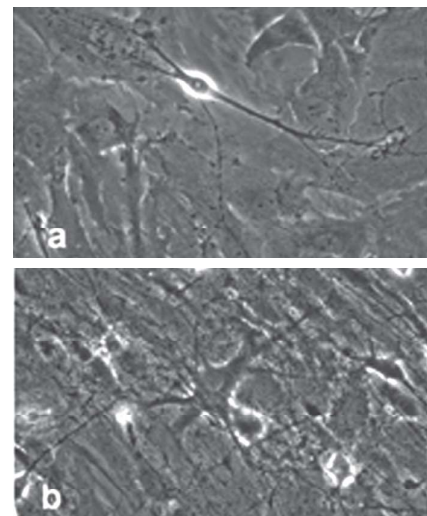


Fig. 9. Photographs of the effect of 780-nm laser irradiation treatment on perikarya and fibers of nerve cells derived from rat embryonic brain.

Dissociated brain cells were embedded in NeuroGel and were either exposed to single radiation dose of 50 mW for 3 min (b), or were non-irradiated controls (a). Large neural cells exhibiting thick fibers were observed after 8 days in vitro irradiated cultures (b). Original magnification: 200X. (Lasers Surg Med, in press, 2009)

A rapid sprouting of nerve processes from the irradiated cell-MC aggregates was detected already within 24h after seeding. The extension of nerve fibers was followed by active neuronal migration. Differences between controls, and irradiated stationary dissociated brain cultures, became evident at about the end of the first week of cultivation - several neurons in the irradiated cultures exhibited large perikarya and thick elongated processes (Fig. 9). Furthermore, during the next 2-3 weeks of cultivation, neurons in the irradiated cultures developed a dense branched interconnected network of neuronal fibers. The sprouting of long processes from large cell body was mainly observed in immunofluorescent MAP-2 staining (Fig.10).

This study suggests that laser phototherapy may play a role in prevention of neuronal loss and accelerate axonal regeneration.

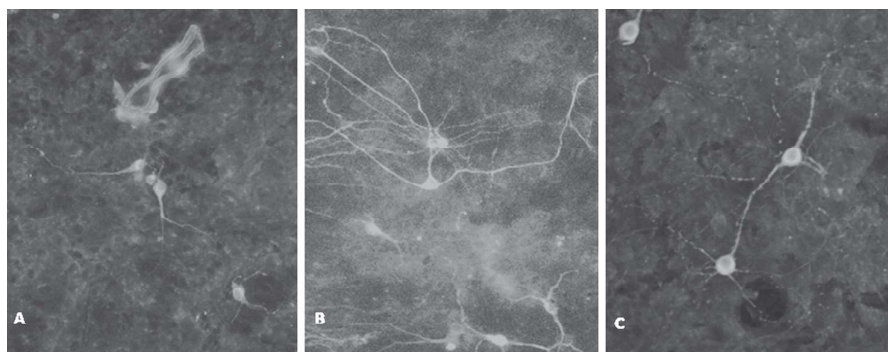


Fig. 10. Immunofluorescent staining of controls and laser irradiated neuronal brain cells migrated from cell-MC aggregates in NeuroGell. **a:** non-irradiated control. **b:** 50 mW 1 min irradiation. **c:** 50 mW 4 min irradiation. In the irradiated cultures note large perikarya bearing long interconnected fibers, which are positively stained for the neuronal marker MAP-2. Original magnification: 200X.

780-nm LASER PHOTOTHERAPY IN CLINICAL TRIAL

Based on the outcome of animal studies, a clinical double-blind, placebo-controlled randomized study was performed to measure the effectiveness of 780-nm low power laser irradiation on patients who had been suffering from incomplete peripheral nerve and brachial plexus injuries for 6 months up to several years [48]. Most of these patients were discharged by orthopedics, neurosurgeons and plastic surgeons without further treatment. In this study 18 patients with a history of traumatic peripheral nerve / brachial plexus injury (means duration from injury to treatment: 7 months in laser-treated group and 11.5 months in placebo group) with a stable neurological deficit and a significant weakness, were randomly divided to receive either 780-nm laser or placebo (non-active light) irradiation. The laser or placebo (non-active light) treatment was applied transcutaneously;

each day for 21 consecutive days, 5 hours daily (3 hours to the injured area of the peripheral nerve and 2 hours to the corresponding segments of the spinal cord). The laser or placebo device were placed approximately 40 cm from the skin treated point, focused on the injured area of the peripheral nerve or corresponding level of the spine (area of corresponding segments of the spinal cord).

Laser dosage:

Spinal cord area - laser irradiation was performed transcutaneously directly above the projection of the corresponding segments of the spinal cord, which was divided into 2 intravertebral levels. Each level was irradiated for 60 minutes a day (150 J/mm²), totaling in 120 minutes a day (300 J/mm²).

Peripheral nerve area - laser irradiation was performed transcutaneously directly above the projection of the injured nerve, which was divided into 3 parts: proximal,

injured area and distal. Each section was irradiated for 60 minutes a day (150 J/mm²), totaling in 180 minutes a day (450 J/mm²). The irradiating spot size was 3x2 mm (6 mm²). The penetration of the near-infrared 780-nm wavelength was approximately 4 cm. Analysis of results of this trial in the laser-irradiated group showed statistically significant improvement in motor function in the previously partially paralyzed limbs, compared to the placebo group, where no statistical significance in neurological status was found (Fig. 11).

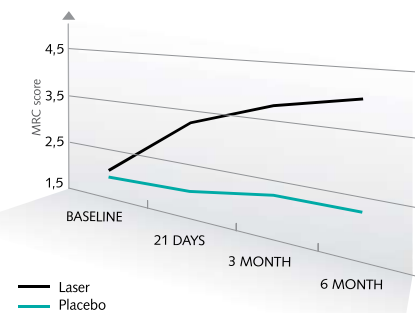


Fig. 11. Graph of the motor function follow-up in injured patients who underwent 780-nm laser phototherapy or placebo treatment.

Mean motor function (+SD) of all affected muscles was examined in injured patients using the Medical Research Council (MRC) Grading System. The analysis of the results showed that at baseline the 780-nm laser-treated and placebo groups were in clinically similar conditions ($p=0.887$). The analysis of motor function during the 6-month follow up period compared to baseline showed statistically significant improvement ($p=0.0001$) in the laser-treated group compared with the placebo group.

Mean motor function of the most influential (functionally dominant) muscle for movement of the affected limb showed a statistically significant improvement in the laser-irradiated group compared to the placebo group. The function was improved mostly by increasing power of the dominant muscles and not intrinsic muscles. Electrophysiological observation during the trial supplied us with important diagnostic information and helped to determine the degree of functional recovery in nerve-injured patients. The electrophysiological analysis also showed statistically significant improvement in

recruitment of voluntary muscle activity in the laser-irradiated group, compared to the placebo group (Fig. 12).

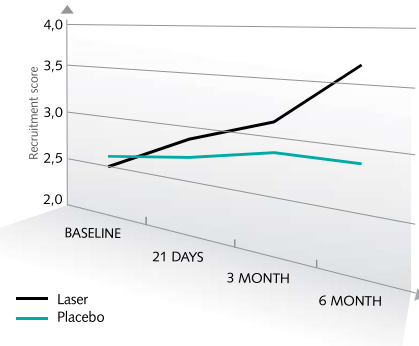


Fig. 12. Graph of the motor unit recruitment in injured patients who underwent either 780-nm laser phototherapy or placebo treatment. Motor unit recruitment, the mean of all examined muscles (+SD), was monitored in injured patients. The 780-nm laser-treated and placebo groups were in similar conditions at baseline ($p=0.934$). In the laser-treated group, statistically significant improvement ($p=0.0006$) was found in motor unit recruitment during the 6-month follow up period, compared with the placebo group.

This study is not the ultimate word regarding 780-nm laser phototherapy in peripheral nerve injured patients. The disadvantages of this study are the small amount of patients, different nerves and etiology of injury. Nevertheless, this pilot study suggests that in peripheral nerve injured patients 780-nm low power laser irradiation can progressively improve peripheral nerve function. That leads us to continue this study with perspective to multicenter trial.

CONCLUSIONS

Results of the experimental study on denervated muscles suggests that laser treatment can restore its function to a substantial degree when initiated at the earliest possible post-injury stage. These findings could have direct therapeutic applications for preserving the function of denervated muscle after peripheral nerve injury.

The extensive review of published articles reported in this paper as well as in previous ones published in *Muscle and Nerve* [16] and *Neurosurgical Focus* [17], revealed that most of experimental studies showed phototherapy to promote the recovery of the severely injured peripheral nerve. This review makes it possible to suggest that a time for broader clinical trials has arrived. The significance of the experimental and clinical studies is the provision of new laser technology for treatment of severe nerve injury.

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