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Dorsal root ganglion electrical stimulation promoted intertransverse process spinal fusion without decortications and bone grafting: a proof-of-concept study

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Abstract

BACKGROUND CONTEXT: Periosteum, endosteum, and bone are innervated by sensory nerves expressing calcitonin gene-related peptide (CGRP), which is a known osteoanabolic peptide and plays an important role in fracture healing and spinal fusion. Synthesis and release of CGRP are found in sensory neurons located in the dorsal root ganglions (DRGs) and can be upregulated by electrical stimulation (ES) at DRG.

PURPOSE: To prove our study hypothesis on the potential of precise ES at DRG through implantable microelectrical stimulation system (IMESS) for its effect on promoting spinal fusion in a rat model *without decortications and bone grafting*.

STUDY DESIGN: An experimental animal study.

METHODS: A novel IMESS was developed for stimulating L4–L6 DRG in rats. Sixteen rats were used and divided equally into the control group without ES and the ES group, with a daily 20 minutes ES to DRG for 6 weeks. At the end of 6 weeks, radiography and microcomputed tomography were conducted to evaluate new bone formation and spinal fusion. Bilateral L4–L6 DRGs were harvested for immunohistochemistry and quantification of neurons with upregulated CGRP expression.

RESULTS: In the ES group, rate of radiographic fusion with complete and uninterrupted bony bridging was 100% (8/8) at the right L4/L5 transverse processes and 75% (6/8) at the right L5/L6 transverse processes. Bony callus formation was absent at the left L4–L6 transverse processes in the ES group and in bilateral L4–L6 transverse processes in the control group.

CONCLUSIONS: We proved for the first time that precise ES at DRG through IMESS effectively promoted intertransverse process fusion in rat model *without decortications and bone grafting*. Electrical stimulation at DRG might be an attractive minimal invasive bioengineering approach

FDA device/drug status: Not applicable.

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and an alternative therapy for intertransverse process fusion that is increasingly being used for the treatment of degenerative spine disorders.

Keywords: Animal model; Calcitonin gene-related peptide; Dorsal root ganglion stimulation; Implantable microelectrical stimulation system; Spinal fusion; Minimally invasive

Introduction

Lumbar spinal fusion is increasingly being used for the treatment of degenerative spine disorders, such as degenerative disc disease, spondylolisthesis, spondylosis, spinal stenosis, and scoliosis [1,2]. Traditionally, spinal fusion can be achieved by decortications of articular surface and packing the joint space with bone grafts or biological substitutes [1–3], such as synthesized hydroxyapatite (HA) and bone morphogenetic proteins (BMPs) [4].

The present study was to test a concept of electrical stimulation (ES) at dorsal root ganglion (DRG) via an implantable microelectrical stimulation system (IMESS) to achieve an intertransverse process fusion without bone grafting in a rat model *without decortications*. In DRG, there are neurons synthesizing and releasing calcitonin gene-related peptide (CGRP) that is a sensory neuropeptide sharing the same gene complex encoding with calcitonin [5]. Calcitonin is produced when the gene is expressed in C cells in the thyroid, whereas the CGRP is produced when the same gene is expressed in sensory neurons located in the DRG and is transported peripherally. Numerous studies proved the distribution of CGRP receptors on the membrane of osteoblasts [5–7]. It was reported recently that CGRP stimulates BMP-2 expression and the differentiation of human osteoblast-like cells in vitro [8]. CGRP-expressing nerves are present in the periosteum, bone, and endosteum [9,10]. Rapid proliferation of CGRP-expressing nerves has been observed during healing of rat tibial fracture [11–13]. The important role of CGRP in fracture healing has been evidenced by our findings that sciatic neurectomy in rat resulted in the absence of CGRP-expressing nerve fibers in the fracture site in tibia and delayed fracture healing [14]. Our group has also found rapid growth of CGRP-expressing nerves in spinal fusion callus in rabbit [15,16].

Recent studies further showed that secretion of CGRP could be triggered by ES at DRG [17–19]. We have recently developed an IMESS that provides precise ES to target neurons in rats [20]. The purpose of this study was to examine DRG ES via IMESS for its potential in spinal fusion enhancement in a rat model.

Material and methods

Animals and experimental design

Sixteen Sprague-Dawley rats (female, 3-month-old, weight: 420–450 g) were equally randomly allocated into two groups (n=8 for each group), that is, the control group and the ES group. Electrical stimulation was applied at the right DRGs at the vertebral level L4–L6 of rats in the ES group. The experiment duration was 6 weeks. Dorsal root ganglion stimulation was provided via a set of IMESS implanted at the L4–L6 transverse processes. Rats were housed individually in metal cages and were allowed free access to tap water and standard rodent chew. The environment was kept with 12-hour day-night cycle at 24°C.

All animal care and experimental protocols were approved by the Animal Ethics Committee of The Hong Kong Polytechnic University in Hong Kong.

Implantation of IMESS and electrical stimulation protocol

Electrical stimulation is generated by an IMESS developed by the coauthor from the School of Aerospace, Tsinghua University, China [20,21]. The implanted stimulation system consists of three parts: a body consisting of a circuit board and a button-type battery, three electrodes, and wires connecting the body and the electrodes (Fig. 1). The dimension of the body is 25×20×8 mm and it weighs 5.86 g. To resist the corrosion of components in tissue environment, the body is coated with polyparaxylylen (parylene C) and silicone rubber. The body is connected to three coaxial electrodes. The outer layer of each electrode is a stainless steel tube serving as reference pole. The core is fitted a stainless steel wire coated with a layer of parylene C. The uncoated tip of the core serves as the stimulating pole of negative polarity (Fig. 2). The IMESS is controlled externally with a remote control unit. Treatment parameters, including stimulation frequency, pulse duration, stimulation voltage, and on- and off-time, are adjustable with the remote control unit for various purposes.

Implantation of IMESS was carried out under general anesthesia with intraperitoneal injection of ketamine and xylazine (ketamine: 10% and xylazine: 2%, volume ratio=2:1, dosage=0.1 ml per 100 g body mass). Right transverse processes of L4–L6 were exposed through a dorsal incision and the periosteum was carefully preserved. A hole of 1 mm diameter was drilled at the location of DRG on each right transverse processes (Fig. 3) and an electrode of the IMESS was inserted. Then, the muscle was sutured to cover the transverse processes and to fix the positions of the electrodes. The body of the stimulator was implanted subcutaneously at the left low back area so as to create least restriction on motion of the animal. The electrodes were then connected to the stimulator with wires. The positions of the electrodes and the stimulator were confirmed by computed radiography (CR). The CR images were taken by a conventional X-ray unit (Model TF-6TL-6; Toshiba Corporation, Tokyo, Japan) with the use of CR image