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The Spine Journal 7 (2007) 61–67

THE SPINE JOURNAL

Posterolateral intertransverse lumbar fusion in a mouse model: surgical anatomy and operative technique

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Received 10 January 2006; received in revised form 7 March 2006; accepted 22 March 2006

Abstract

BACKGROUND CONTEXT: Animal models are frequently used for studying the effect of bone graft substitutes or allogeneic materials on osterolateral lumbar fusion. Transgenic technology in the mouse provides a unique opportunity to further understand the biology of spine fusion.

PURPOSE: To describe pertinent lumbar spine anatomy and formulate a surgical protocol for posterolateral fusion in the mouse model.

STUDY DESIGN: Diagnostic model: development of an animal model for biologic evaluation of posterolateral spine fusion.

METHOD: Ten mice were killed to study relevant lumbar spine anatomy and develop a protocol for lumbar spine fusion. The L4–L6 fusion protocol was validated in 46 mice for ease of exposure, preparation of the posterolateral fusion bed, introduction of bone inductive agents, and perioperative care.

RESULTS: Anatomy and surgical technique for posterolateral intertransverse lumbar fusion in the mouse model are described. A paraspinal approach allows exposure of the transverse processes, decortication, and graft placement at the L4–L6 intertransverse fusion site. Decortication alone did not result in fusion, whereas the use of bone graft resulted in satisfactory fusion rates. Perioperative morbidity and mortality rates were low.

CONCLUSION: The mouse posterolateral lumbar spine fusion model is reproducible, inexpensive, and has low complication rates. Knowledge of the relevant anatomy and adherence to a well-defined surgical protocol provides a reliable and reproducible experimental spine fusion model.

Keywords: Fusion model; Mouse; Transgenic technology

Introduction

Various animals have been used for studying the effect of bone graft substitutes or allogeneic material on posterolateral lumbar fusions [1–11]. The mouse model has been infrequently used, primarily owing to the small size and consequent technical difficulty. The availability of high-quality operating microscopes and microsurgical instrumentation makes the use of the mouse model comparable to larger animals in studying posterolateral spine fusion. Mice are easy to maintain in a laboratory situation and do not have specific or expensive housing care needs. They breed year round with a short generation time, deliver larger litters, and tolerate inbreeding well compared with other mammalian species. Furthermore, the mouse is the only mammal besides man with an established complete genome [12]. Relatedly, transgenic mouse technology offers the potential to evaluate specific gene effects on physiology [13–16], and to follow the fate of the marked donor cells within a recipient mouse, using specialized markers such as beta-galactosidase [17].

To our knowledge, a consistent protocol for lumbar spine fusion in the mouse model has not previously been described. Adherence to a well-defined protocol will result in a reproducible experimental spine fusion model with a low rate of complications. The development of a protocol will minimize variations in technique that may result in inconsistent fusion rates, and will decrease operative

FDA device/drug status: not applicable.

Nothing of value received from a commercial entity related to this manuscript.

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1529-9430/06/$ – see front matter
doi:10.1016/j.spinee.2006.03.004

morbidity and mortality. The aim of this study is: 1) to document anatomy pertinent to a posterolateral approach of the lumbar spine fusion, and 2) to describe a standard surgical technique for performing these fusions in mice.

Materials and methods

The study was done in two parts; the first part involved killing 10 mice to study the pertinent lumbar spine anatomy in a mouse. The second part of the study (reported separately) was used for validation of this fusion model, and involved preparation and decortication of the posterolateral fusion bed in 10 mice, and use of various bone inductive agents to induce fusion in three separate groups of 10 mice each. Appropriate permission was obtained from the institutional animal care and welfare committee.

Anatomic study

Ten skeletally mature mice were killed after their use in an unrelated study. There were eight females and two males, with an age range of 2 months to 8 months. Animal weights varied between 20 and 40 grams. Each cadaver was dissected, and the osseous, soft tissue, and myofascial anatomy pertinent to a lumbar spine fusion was observed and documented. Dissection was limited to the posterior elements of the lumbar spine and proximal pelvis, extending to the lateral tips of the transverse processes.

Fusion study

The posterior-lateral lumbar spine fusion technique was carried out in 46 mice from L4 to L6 levels. The animals were 3 to 4 months in age and weighed from 20 to 40 grams. A single orthopedic surgeon performed all procedures, and worked in conjunction with an animal laboratory supervisor for intraoperative and perioperative monitoring of administered anesthesia.

Anesthesia and preoperative preparation. Sodium pentobarbital solution was administered intraperitoneally at a dose of 50 mg/kg (Fig. 1). The dorsal aspect of the lumbar spine and iliac crest were shaved. Depth of anesthesia was assessed by response to tail, pinnae, or pedal pinch [18–21], supplementing pentobarbital as needed. Normal physiologic parameters are indicated in Table 1 [22].

Surgical technique. Surface landmarks on the dorsum were palpated to identify spinal levels. The iliac crest provided a rough estimate of the interspace between the L5 and L6 spinous processes (Figs. 2 and 3). The entire surgical procedure was carried out using a binocular stereoscopic operating microscope with a 10× magnification power. A paraspinal, trans-sacrospinalis approach similar to the one described by Wiltse in humans was used [23] A 15 mm dorsal midline skin incision was made centered over the L4–L6 spinous process (Fig. 3), and a self-retaining retractor was used to retract the skin edges. The

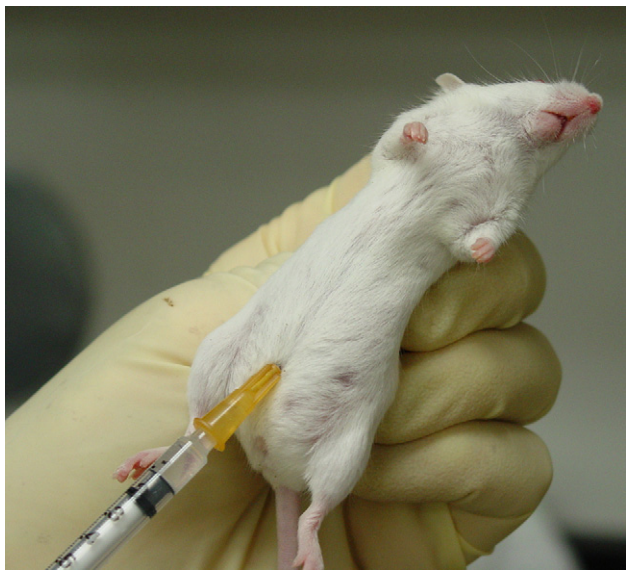

Fig. 1. Technique of intraperitoneal administration of anesthetic agent. Sodium pentobarbital solution diluted 1:1 with water was administered intraperitoneally at a dose of 50 mg/kg.

dorsolumbar fascia covering the paraspinal muscles was the first subcutaneous structure encountered and provided attachment to the latissimus dorsi, external oblique, internal oblique, and the transversus abdominis muscles. The erector spinae (sacrospinalis) is a single large muscle overlying the facet joints and transverse processes. Using a paraspinal approach (15 mm long paramedian longitudinal incision 3–4 mm from the midline) (Fig. 4), the erector spinae muscles were split bluntly, preserving the midline longitudinal ligaments and leading directly to the fusion site. Lateral retraction of the lateral half of erector spinae muscle exposed the transverse processes and the lateral surface of the facet joint (Fig. 5). Morphologically the bony architecture of the mouse lumbar vertebra is similar to that of humans (Fig. 6); the L5 spinous process angles caudally, whereas the L6 spinous process is larger and angles directly dorsally. The mamillary processes are well developed in rodents and provide another useful landmark; they project dorsolaterally from the posterior surface of superior articular process.

The transverse processes begin at the junction of the posterior neural arch and the vertebral body and are angled rostrally and ventrally (unlike in humans where they are more transversely oriented). The transverse processes are tubular structures at their base, tapering into flattened thin triangular plates of cortical bone near the midportion. Decortication of the fusion bed was carried out with

Table 1
Normal physiologic parameters that can help in perioperative monitoring of the mouse

Body temperature	36.5–38.0°C
Respiratory rate	80–230 breaths/min
Heart rate	500–600 beats/min