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Microsurgical approach to the spinal canal in rats

Mortimer Gierthmuehlen[a,*], Thomas M. Freiman[a,1], Dominik Elverfeldt[b,2], Jan Kaminsky[a,3]

[a] Department of Neurosurgery, University of Freiburg, Breisacher Strasse 64, D-79106 Freiburg, Germany
[b] Department of Radiology, MedicalPhysics, University of Freiburg, Hugstetterstraße 55, 79106 Freiburg, Germany

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ABSTRACT

The spinal cord of the rat has become a widely used model for biodynamic, pharmaceutical and neurological experiments. However, no standard procedure to approach the spinal cord in rats has been published in detail. We present a description of a dorsal approach to the spine, spinal canal and myelon of the rat. This approach provides sufficient exposure of the neural structures to perform extended microsurgery at the spinal nerve-roots, the lateral and dorsal myelon and vertebral structures under a surgical microscope. Perioperative management, anaesthesia and anatomical landmarks are discussed and common pitfalls are described.

1. Introduction

Many neurosurgical, neurological and urological experimental studies require surgical interventions in the spinal region. In light of the animal's size, cost and availability, many models have been created for the rat, which has become one of the most important animals for spinal studies (Kwon et al., 2002). But so far, standardized surgical approaches to the spine have not been described. This can be explained by the fact that the surgical procedure usually needs to be summarized in one paragraph in a publication. As a consequence, there is no practical guide how to avoid certain pitfalls during preparation, and each investigator has to develop his own surgical technique. This requires time and resource-consuming training before the actual project commences—a problem we experienced when we planned our study. Furthermore, studies using different approaches are difficult to compare.

It is the aim of this article to describe step by step a standardized surgical procedure with common pitfalls and tricks for the dorsal approach to the spinal canal in rats.

2. Materials and methods

2.1. Animals

The procedure is performed in female Wistar rats weighing between 250 and 300 g. Anaesthesia is done with intramuscular administration of Ketamine 10% (0.75 ml/kg) and Medetomidine (0.15 mg/kg). The instruments we used for surgery are listed in Table 1.

A self-designed OR-table allows stable positioning of the rat with moderate kyphosis for optimal exposure of the spine. It also allows the surgeon to position the hands comfortably on each side of the animal. A heat-lamp and a warming-pad provide constant temperature of the animal during surgery. Sufficient hydration of the animal is ensured by intermittent subcutaneous injection of 0.9% NaCl (3 ml/h). All described procedures on animals were approved by the Regierungspraesidium Baden-Württemberg (G07/37) and the Ethics Committee of the University of Freiburg. The "Principles of laboratory animal care" (NIH publication no. 86-23, revised 1996) were followed for all experiments.

2.2. Anatomy

The lumbar spine of the rat consists of six vertebrae. The landmark for the caudal lumbar spine is the pelvis, the sixth lumbar vertebra can be identified between the two cristae iliacae (Fig. 1). This is the most caudal level of the spinal canal that can be opened safely, since the sacral spinal roots leave the spinal canal dorsally and preparation in that area is extremely difficult.

* Corresponding author. Tel.: +49 761 270 5001; fax: +49 761 156 4773.
E-mail addresses: mortimer.gierthmuehlen@uniklinik-freiburg.de (M. Gierthmuehlen), Thomas.freiman@uniklinik-freiburg.de (T.M. Freiman), Dominik.Elverfeldt@uniklinik-freiburg.de (D. Elverfeldt), jan.kaminsky@uniklinik-freiburg.de (J. Kaminsky).
[1] Tel.: +49 761 270 5001.
[2] Tel.: +49 761 270 7391.
[3] Tel: +49 761 270 5007.

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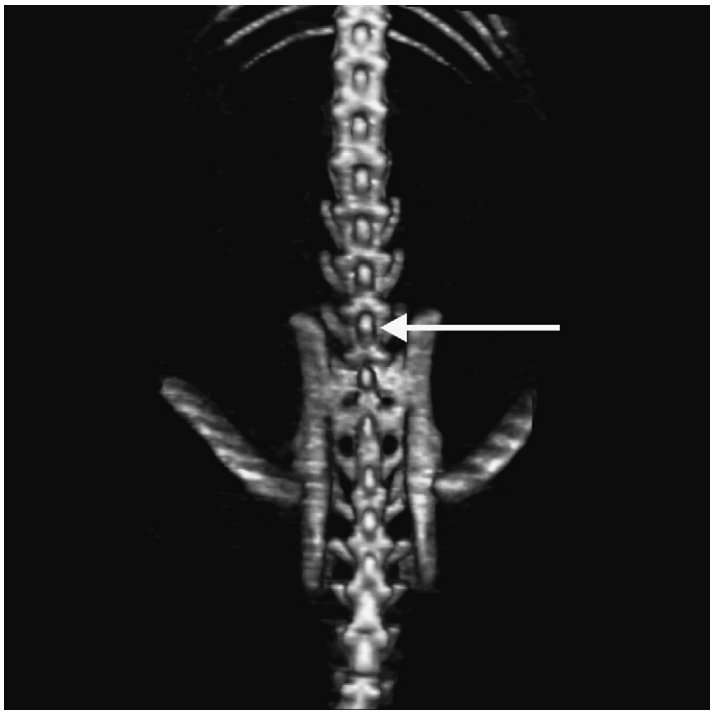

Fig. 1. CT-scan of the lumbar spine of a rat. The 6th lumbar vertebra is located between the cristae iliacae (arrow).

2.3. Approach to the lumbar spinal cord

The coat at the operation field is removed with an electric shaver. After palpating the cristae iliacae, the L6 spineous process is identified marking the most caudal process. After disinfection of the skin, a midline incision is done from 2 cm cranial to 1 cm caudal to the cristae ilacae. The skin is retracted with 2-0 suture, followed by blunt preparation of the subcutaneous tissue. After the subcutaneous connective tissue is carefully lifted with forceps and cut away (Fig. 2), the fascia of the paravertebral muscles becomes visible. The paravertebral tendons connecting to the L6 spinous process can be identified as the last white stripe before the muscles attach directly to the sacral bone (Fig. 3). The fascia is incised superficially and bilaterally to the spineous processes from L3 to L6 (Fig. 3), while a cranio-caudal direction allows cutting the paravertebral tendons. Again, blunt preparation is needed to separate the paravertebral muscles from the spineous processes, the remaining paravertebral tendons are dissected with scissors. Great care should be taken not to go too lateral but to dissect closely to the spineous processes. A lateral and deep preparation may damage the spinal roots emerging from the spinal canal. A small retractor is inserted to hold back the paravertebral muscles, and the interspinous tendons are dissected to make the spinous processes visible (Fig. 4). By holding the spinal process of L6 with forceps and moving the sacrum backward and forward with two fingers, a movement between the L6 and the S1 spinous process can be identified. This again ensures the correct level, as the spinous processes of S1 and S2 do not show any mobility. The spinous processes are now removed with a small rongeur. From this point on, an operation-microscope is used.

A large drilling-head is chosen to clean the operating site (Fig. 5). A combined irrigation-suction-device (Hydroflow®) is helpful in providing good vision. Bleeding mostly occurs in the space between the facet joints and can be coagulated with the bipolar forceps, but since the spinal nerves emerge only a few millimetres below this area, the coagulation power is adjusted to the minimal possible. A cranial-to-caudal direction is chosen to open the spinal canal as its diameter decreases caudally. A smaller drilling-head is now chosen and the drill is held perpendicular to the spine. Otherwise it could

Table 1
Instruments we used for surgery.

Instrument	Manufacturer	Comments
OR-table	Self-made	Providing kyphosis and warming
Scissors	Pfeilring	
Rongeur	Niegeloh	
Foreceps (anat/surg.)	Aesculap	
Needle-holder	Aesculap	
Micro-foreceps	Aesculap	
Micro-scissors	Aesculap	
Micro-needle-holder	Aesculap	
Dura-hook (sharp)	Aesculap	2-0 Suture on top of a dental instrument (Fig. 15)
Nerve-dissector	Self-made	

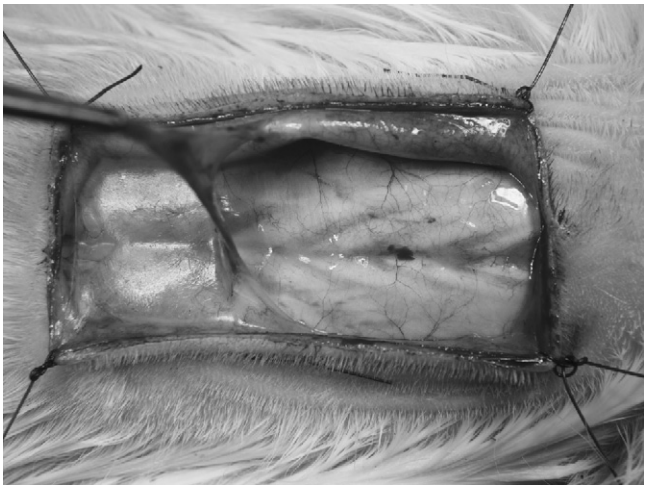

Fig. 2. The edges of the wound are retracted with 2-0 suture attached to small clamps on the operating table. The subcutaneous connective tissue (containing the blood-vessels seen in the photo) should be removed as it may get entangled with the drill. The paravertebral muscles become visible. The spinous process of L6 can be localized as the most caudal insertion-point for paravertebral tendons.