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A method for intramedullary fixation of fractures and segmental grafts in the rat tibia

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Introduction

The rat is the most frequently used animal for studies of fracture healing and bone grafting. The majority of experiments have been done in the femora (*Bonnarens & Einhorn* 1984, *Ekeland et al.* 1981, *Ekeland et al.* 1982, *Grundnes & Reikerås* 1991, *Grøgaard et al.* 1990, *Hou et al.* 1991, *Huo et al.* 1991, *Mølster et al.* 1987, *Pelker et al.* 1989, *Reikerås* 1990, *Reikerås et al.* 1989). The femoral diaphysis has a straight, wide medullary canal suitable for intramedullary nailing and reaming. The cross-sectional geometry of the femur is close to an ellipse, which makes calculation of area- and polar moment of inertia fairly easy. The femur has a thick layer of muscle covering in all directions, reducing the risk of osteomyelitis. The femur is however, loaded horizontally during ambulation of the rat, which is in contrast to the vertical loading of the weight-bearing long bones in humans. The loading pattern strongly influences the orientation of collagen and mineral (*Boyde & Riggs* 1990), and thus the physiological and pathological response of the bone. The tibia in rats is loaded vertically. Intramedullary nailing of the tibia which secures perfect alignment of the fractures or segmental grafts, have been difficult to perform because the medullary canal is slight curved and conical converging in the distal direction.

In our laboratory a new technique for intramedullary nailing of the rat tibia has proved useful. The technique, biomechanical properties of the nail and our experience with it is described in this article.

Method

Wistar/Han/Mol SPF rats (Møllegård, Copenhagen) were used in studies with the modular nail. 25 animals, weight 199–207 grams were used in one fracture study (*Nordsletten et al.* 1991), and 34 rats, weight 180–200 g, in another fracture study (*Berg-Larsen & Kirkeby* 1990). 72 rats, weight 180–200 g, were studied in a grafting experiment (*Kirkeby et al.* 1992). The experiments conformed to the Norwegian Council of Animal Research Code for the Care and Use of Animals for Experimental Purposes.

The animals were anaesthetized with a combination of Hypnorm (fluanisone 5 mg/ml-fentanyl citrate 0.1575 mg/ml, Jansen Pharmaceutica BV, Beerse, Belgium) and Dormicum (midazolam 2.5 mg/ml, Hoffmann La Roche, Basel, Switzerland). 0.2 ml/100 g body weight were given subcutaneously. Postoperatively, the animals were given buprenorphine (Temgesic, Reckitt & Colman, Hull, England), 0.2 mg/kg b.w. at 12 hour intervals for two days. In all three experiments the animals apparently walked on the operated limb from the first postoperative day.

The animals were operated on under aseptic surgical conditions. The right lower leg was shaved, and a longitudinal incision made parallel to the anterior margin of the tibia. A 18 G cannula (Asik A/S-Rødby-Denmark)-outer diameter 1.23 mm (measured with a sliding calliper, accuracy of ± 0.01 mm) was inserted into the medullary canal of the tibia, either through the distal patellar ligament (Fig. 2), or through the proximal tibial

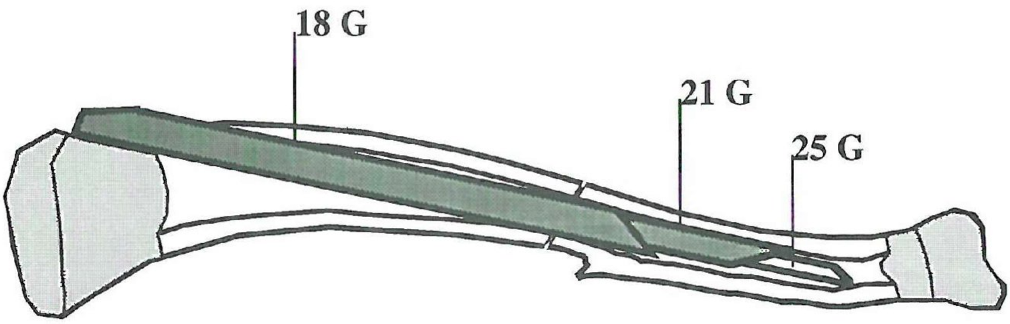

Fig. 1. Fractured tibia with modular intramedullary nail, consisting of three parts threaded into and past each other until jamming in the medullary canal.

crest (Figs. 1, 3). With rotation and axial pressure the angled, sharp tip of the cannula was easily inserted. With the cannula inserted the tibia was fractured, or osteotomies for segmental grafting made. The cannula was then advanced distal to the fracture, or through the graft into the distal tibia until it jammed in the medullary canal. The hollow mandrin of a 18 G Venflon (Viggo AB, Helsingborg, Sweden), diameter 21 G, measured to 0.9 mm, and the mandrin of a spinal needle (Yale spinal, Becton Dickinson SA,

Fig. 2. Radiograph of fractured tibia on the 24th postoperative day from study by *Nordsletten et al.* (1991). The fibula was intact.

Fig. 3. Radiograph from study on segmental cortical grafts (*Kirkeby et al.* 1992) after 12 weeks. The fibula was intact. Abundant callus formation proximally.

Table 1.

	Intramedullary nail	Tibia (230 g)	Tibia (340 g)
Stiffness (Nm/°x10^{-3})	16.1 (±0.6)	19.4 (±3.3)	24.8 (±5.0)
Moment at 20°* deflection (Nmx10^{-2})	29.1 (±0.7)	25.3 (±1.3)	38.5 (±4.6)
Yield moment (Nmx10^{-2})	21.14 (±0.8)		

Biomechanical values (Mean ± SD) for intramedullary nail (tested alone) and for intact tibias of rats weighing 230 grams and 340 grams (*Nordsletten & Ekeland* 1991). The proximal part of the nail, consisting of all three cannulae, and the tibias were all tested in the same three point bending apparatus at deformation rate 0.095 rad/sec.
* Intact tibias fractured at 19.25° (230 g) and 21.15° (340 g).

Madrid, Spain-25 G), measured to 0.4 mm, were inserted inside the 18 G cannula and both advanced past each other until jamming in the distal fragment (Figs. 1, 2 and 3). The cannulae were cut with pliers, and the wound closed in two layers.
The proximal part of the nail consisting of all three cannulae were tested in three-point ventral bending at a rate of 0.095 rad/sec. The testing machine has been described in detail previously (*Engesæter et al.* 1978). The distance from the fulcrum of the testing machine to the cam of the rotating disc was 21.11 mm, and the distance from the clamp to the cam was 26.70 mm. The biomechanical values of the modular intramedullary nail were compared to intact tibias of young and adult male Wistar rats tested under the same conditions (Table 1).

Experiments performed with this technique
Fractures
The technique has been used in experiments on fracture healing (*Berg-Larsen & Kirkeby* 1990, *Nordsletten et al.* 1991). After insertion of the 18 G cannula into the medullary canal, the anterior cortex was sectioned with a scalpel 4 mm distal to the tibial tuberositas. The fracture was completed by manual breaking of the posterior cortex, leaving the fibula intact.
In the study by *Nordsletten et al.* (1991) the fractured legs were radiographed after 24 days. Radiograms disclosed malalignment of the fracture due to insufficient advancement of the intramedullary nails in five animals. These animals were excluded from testing. On day 25 the fractured tibias with intact soft tissues were tested in three-point ventral bending. The nails were removed prior to testing. The removal caused epiphysiolysis proximally in the tibia in four animals.
Berg-Larsen & Kirkeby (1990) evaluated the effect on the ipsilateral femora of tibial fractures fixed with the nail (n = 20). They experienced no problem with fracture alignment or epiphysiolysis.

Segmental grafting
The technique has been used in a study on incorporation of allogeneic and syngeneic segmental, cortical bone grafts (*Kirkeby et al.* 1992). A total of 72 recipient rats were used. Grafts were harvested through a longitudinal incision anteriorly over the tibia. Oblique osteotomies were made with a dental rotating saw under direct vision and continuous saline irrigation. The osteotomies were made 1 mm and 10 mm proximal to the tibiofibular synostosis. The graft was placed in sterile saline in room temperature and transplanted within 30 minutes. After insertion of the 18 G cannula into the tibial canal, a segment of the left tibia was removed as described for the donors, and the donor segment was inserted into the defect. Care was taken to leave the fibula intact. The cannulae were advanced through the

graft as described under method. All tibias with grafted segments were clinically stable before wound closure. After 12 weeks the nails were removed without complications, and the grafts tested in three-point bending. There were no failures of the osteosynthesis.

Discussion

A few papers on fracture and osteotomy experiments in the rat tibia have been reported (*Bak & Andreassen 1989, Greiff* 1978, *Nielsen et al.* 1991, *Paavolainen et al.* 1989). *Mølster* (1985) started his studies on intramedullary nailing in the tibia with a 1.4 mm nail with stiffness equal to the intact tibia, but changed to the femora due to technical difficulties. *Greiff* (1978) made a closed fracture of the tibia after intramedullary nailing with a 0.8-mm stainless-steel wire. *Greiff* did not present the biomechanical parameters of this nail. Due to the small diameter of the nail he was able to get it far down into the medullary canal. However, 14 out of 42 fractures healed with more than 5° angulation. With a small nail diameter the wide upper part of the medullary canal will allow movement of the fracture. The nail reported in the present article has a bigger diameter proximally (1.23 mm), and therefore stabilizes the fracture better. *Bak & Andreassen* (1989) used a 0.8 mm stainless steel tube, with ultimate load 1/6th and bending stiffness 1/9th of the intact tibia in 2 year old rats. In 36 old rats they excluded 10 animals due to bending or fracture of the nail, which shows that the strength and stability of the 0.8 mm tube was insufficient for older animals (*Bak & Andreassen* 1989). In a later experiment in old rats they had changed to a 0.89 mm K-wire (*Bak & Andreassen* 1991), which gave osteosynthesis failure only in four of 57 animals. The rather small diameters of these nails would be expected to cause problems with angulation of the fractures, and specially for segmental grafting it would give unacceptable instability since it would not fill the medullary cavity neither at the proximal, nor at the distal osteotomy.

The mechanical properties of the modular nail in the present article were tested in the proximal part constituting all three cannulae. In case of short advancement of the 18 G cannula the functional stiffness of the nail will be less than reported. However, as Fig. 2 illustrates the 21 G cannulae and the mandrin were strong enough to retain alignment, and avoid fatigue fracture when only the tip of the 18 G cannulae had been advanced past the fracture.

The conical shape of the modular nail fits the medullar cavity of the tibia better than a nail with a uniform outer diameter. This contributes to the stability of the fixation. However, packing the medullary cavity with nails many be disadvantageous to the blood supply (*Wilson* 1991), and thereby affecting the healing process.

The use of the present technique for intramedullary nailing is simple and easily learned when attention is directed to some essential steps. Firstly, the nail must be inserted into the medullary cavity prior to fracture or osteotomy. Preferently the nail should be inserted through the patellar tendon, but it is possible to enter the medullary cavity through the tibial crest. Secondly, the second and third part of the nail must be inserted with adequate force, otherwise the nail will not extend far enough into the distal tibia. In the fracture study by *Nordsletten et al.* (1991) there was malalignment of five tibias due to this "surgeon failure". However, the surgeons were not aware of this problem when they started the study. Since the diameter of the second and third part are small, we had no problems with splintering of the distal tibia, as experienced with wider nails (*Mølster* 1985). Thirdly, the cutting of the nails at the insertion must not be done to close to the bone. In the fracture study by *Nordsletten et al.* (1991) epiphysiolysis occurred during removal of the nail in four tibias. The nail did not protrude from the cortex, therefore bone had to be removed, and rather violent force applied to extract the nail. The nail should, however, not be cut to far apart from the bone, because it is flattened during cutting, and this provides some rotational stability as the three parts of the nail are themselves locked to each other. Additional rotational stability can be achieved by leaving the fibula intact.

In conclusion, a new technique for intramedullary nailing of the rat tibia that provides several advantages over earlier applied techniques is presented.

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Summary
A new technique for intramedullary nailing of the tibia in rats is described. The results of fracture and segmental graft fixations with the nail are presented. The modular nail consists of three parts that are threaded into and past each other, giving it a conical outer diameter which fits the tibial medullary canal. The biomechanical properties of this nail are similar to those of intact tibias of 230 g male rats, which makes it suitable for experiments with rats weighing more than 200 g.

Sammendrag
En ny metode for margnagling av rotte tibia beskrives. Resultater fra fraktur og bentransplantasjons studier der naglen er benyttet presenteres. Naglen består av tre deler som tres inni og føres forbi hverandre slik at den ytre formen av naglen passer til margkanalen på rotte tibia. De mekaniske egenskapene til naglen gjør den velegnet til studier med rotter som veier mere enn 200 gram.

Yhteenveto / P. Pelkonen
Artikkeli kuvaa uuden ydinnaulaustekniikan rotalla. Kolmiosainen naula rakentuu ydinkanavaan sylinterimäisesti. Naulan biomekaaniset ominaisuudet vastaavat 230-grammaisen urosrotan tibian ominaisuuksia, ja se sopii kokaisiin, joissa rotta painaa yli 200 g.

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