THE EFFECT OF TRAUMA TO THE LOWER FEMORAL EPIPHYSEAL PLATE
AN EXPERIMENTAL STUDY IN RABBITS
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Forty-four rabbits were operated on when five weeks old; in one group a 2 mm drill-hole was made in the intercondylar portion of the right femur across the central portion of the growth plate up to the diaphysis, while in the other group a similar drill-hole of 3.2 mm was made. At 3, 6, 12 and 24 weeks after operation, specimens from the growth plates of both femora were analysed using radiographic, microradiographic, histological and histomorphometric techniques.

It was found that destruction of 7% of the cross-sectional area of the growth plate caused permanent growth disturbance and shortening of the femur.

The most significant injuries of the growth plate are those which are transepiphyseal. Central epiphysodeis, caused by an undisplaced fracture, temporary transepiphyseal wires or pins, curettage or limited destruction secondary to infections, does not always cause permanent cessation of growth, since the pressure of growth is able to break a minor bone bridge, or rather to tear it from the metaphysis (Ford and Key 1956; Friedenberg 1957; Campbell, Grisola and Zanconato 1959; Siffert 1956, 1966). When longitudinal growth is prevented by rigid fixation of the epiphysis to the metaphysis, the growth plate is eventually destroyed and epiphysodeisis occurs (Siffert 1956; Campbell et al. 1959; Nordentoft 1969). Numerous descriptions of the results of trauma to the growth plate have been reported (Pheimister 1933; Gelbe 1951; Ford and Key 1956; Siffert 1956, 1966; Friedenberg 1957; Campbell et al. 1959; Trueta and Amato 1960; Nordentoft 1969; Porter 1978). However, the quantitative changes due to trauma have not, as far as we know, been analysed. The purpose of our study was to use semi-automatic quantitative histomorphometric analysis in order to determine the changes in the growth plate caused by transepiphyseal injury.

MATERIAL AND METHODS
Forty-four New Zealand White rabbits five weeks of age and weighing over 500 g were used. They were anaesthetised with subcutaneous Hypnorm (Philips-Duphar) 0.3 ml/kg and diazepam (Diapam, Orion) 3–5 mg/kg. Pre-operatively the rabbits were given 100 000 IU of procain penicillin intramuscularly. Both knees were shaved and scrubbed with Neo-Amisept R (Lääke Farmos); a medial parapatellar incision was then made in the right knee, the patella was dislocated laterally and the distal portion of the femur was exposed. A drill-hole of either 2 mm or 3.2 mm in diameter was made in the intercondylar portion of the femur across the central portion of the epiphyseal plate up to the diaphysis. The patella was then replaced and the incision closed in layers by 3–0 polyglycolic acid (Dexon) sutures. A similar arthroscopy was performed in the left (control) knee but no drilling was carried out.

Postoperatively the rabbits were given naloxone 0.3 ml subcutaneously; they were returned to their cages and were given normal food and care. Groups of five rabbits were then sacrificed at 3, 6, 12 and 24 weeks by an overdose of Hypnorm (1.0 ml/kg); two and three days before sacrifice the rabbits had been given Vendarcin (Gist-Brocades) 50 mg/kg subcutaneously for oxytetracycline (OTC) fluorescence studies. Radiographic, microradiographic, OTC-fluorescence, histological and histomorphometric techniques. After sacrifice, both femora were dissected free; they were then measured, radiographed and inspected for signs of infection or deformity (Fig. 1). The distal third of each femur was taken as a specimen and fixed in 70% alcohol and embedded in methylmethacrylate (Schenk 1965). For microradiographic and OTC-fluorescence studies, longitudinal sections 80 μm thick were cut in the coronal
plane (Milch et al. 1958). For the microradiographic examinations (50 kV, 9 mA), film was exposed for 12 minutes at a focal distance of 29.5 cm. For histological and histomorphometric analysis 5 μm sections were cut in the coronal plane with a microtome and stained using Masson–Goldner’s method (Goldner 1938).

For histomorphometric analysis a Leitz microscope was linked via a television camera to a Reichert-Jung MOP Videoplan and a magnification of four was used (Renwell 1983). The microscopic field was displayed on the TV-screen in the computer, and the surface areas of the basement plate, the growth zone and the hypertrophic zone were measured (Ham 1974; Haines 1975). The cellular elements of the growth plate in three standardised areas were measured through the whole thickness of the growth plate, with the width of the screen serving as a limit for lateral directions (Fig. 1).

Measurements were made of four specimens taken from rabbits sacrificed immediately after operation (0 weeks). The percentages of the transverse diameter and the cross-sectional area of the drill-hole in relation to the same measurements taken from the growth plate were determined with the MOP Videoplan. Histological sections used were cut in the transverse plane through the growth plate. For statistical evaluation the Student’s t-test was used.

RESULTS

Before sacrifice all 40 rabbits were able to walk without difficulty, and there were no patellar dislocations or infections.

A drill-hole of 2 mm in diameter accounted for 13% of the transverse diameter of the distal femoral growth plate of a five-week-old rabbit. The cartilaginous injury was 3% of the whole cross-sectional area of the growth plate. The corresponding figures for a drill-hole of 3.2 mm in diameter were 20% and 7%.

At three weeks. A drill-hole of 2 mm caused no inequality of femoral length. Fibrous tissue with a thin strip of fluorescent calcified new bone extended through the growth plate to the metaphysis at the drilling point, and an integrate basement plate was found in all cases. At the point of the drill-hole a dip of the epiphyseal cartilage was seen (Fig. 2).

A drill-hole of 3.2 mm caused shortening of the right femur (mean 2.3 mm), and a dip of the epiphyseal cartilage extending deep to the metaphysis was found in all cases. Microradiographic and OTC-fluorescence studies revealed new bone which bridged epiphyseal and metaphyseal cancellous bone (Fig. 3). The normal appearance of the growth plate from the control side of an eight-week-old rabbit is seen in Figure 4.

In three cases there was intense fluorescence of
calcified tissue extending deep to the hypertrophic zone from the metaphyseal side, involving about one-half of the whole thickness of the epiphyseal cartilage. Histomorphometry showed a constant decrease of the cellular elements of the growth zone (germinal, proliferating and palisading zones). The cellular elements of the hypertrophic zone were slightly increased on the control side and, after a drill-hole of 2 mm in diameter, on the operated side also. The narrowing of the growth plate in the lateral and medial condylar areas after a drill hole of 3.2 mm in diameter approached significance (p < 0.05, d.f. = 8).

At six weeks. After a drill-hole of 2 mm the operated femur was shorter by a mean of 0.2 mm in one out of five cases. Histological studies revealed occasional scanty nests of cartilage cells and blood vessels in the cancellous spaces of the epiphysis at the point of the drill-hole. A dip was seen at the site of injury in the growth plate, and a tiny bony connection between epiphyseal and metaphyseal cancellous bone was found in one case (confirmed by an OTC-fluorescence study and microradiography). A drill-hole of 3.2 mm caused shortening of the operated femur by a mean of 0.8 mm in four of five cases. Histology revealed a bony bridge between the epiphysis and metaphysis, and the margins of the epiphyseal plate cartilage bordering on this bridge were invaginated into the metaphysis. OTC-fluorescence studies showed tiny projections of calcified tissue extending from the basement plate into the epiphyseal cartilage at the drilling point. Histomorphometry revealed increased height of the epiphyseal cartilage in lateral and medial condylar areas after a drill-hole of 3.2 mm (p < 0.05, d.f. = 8), and a decrease of the cellular elements which were replaced by the new bone and granulation tissue in the central area.

At 12 weeks. After a drill-hole of 2 mm the operated femur was shorter by a mean of 0.5 mm than the control one in two of five cases. Histological studies revealed a tiny dip at the drilling point in the epiphyseal cartilage (Fig. 5), and a small nest of cartilage cells was occasionally seen in the cancellous bone of the metaphysis and epiphysis corresponding to the drill-hole. In one specimen, OTC-fluorescence studies showed tiny projections of calcified tissue extending from the basement plate into the epiphyseal cartilage.

A drill-hole of 3.2 mm caused shortening of the operated femur by a mean of 3.8 mm. Histological analysis showed a firm central bony connection between the epiphysis and metaphysis, and epiphyseal cartilage was found at the periphery of the growth plate (Fig. 6). OTC-fluorescence studies revealed tiny bone bridges connecting the basement plate to the metaphyseal cancellous bone. Histomorphometry showed a decrease of the elements of both the growth and hypertrophic zones except in the central area of the operated side after a drill-hole of 3.2 mm; here the height of the epiphyseal cartilage was greatly increased (for comparison, the normal appearance of the growth plate of a 17-week-old rabbit is shown in Figure 7). The basement plate reached its maximum area by 12 weeks.

At 24 weeks. After a drill-hole of 2 mm both femora were equal in length, and histology confirmed the union of the distal femoral growth plate on both the operated and the control sides. At the site of the previous growth plate, microradiography revealed low-density bone formation on the operated side in three cases and on the control side in all cases. OTC-fluorescence studies showed intense fluorescence corresponding to the areas of low-density bone.

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Figure 5 – Twelve weeks after a 2 mm drill-hole had been made a tiny bone bridge is seen connecting epiphyseal and metaphyseal cancellous bone; the palisading structure of the growth plate is normal (× 2.5). Figure 6 – After a 3.2 mm drill-hole a broad bony bridge is seen extending from the epiphysis to the metaphysis; the palisading structure of the growth plate is slightly distorted (× 2.5). Figure 7 – The normal appearance of the growth plate of a 17-week-old rabbit (× 2.5).
A drill-hole of 3.2 mm caused shortening of the operated femur (mean 4.6 mm, range 2 to 9 mm), and, in four cases, the difference in length was greater than 3 mm. Histology revealed union of the distal femoral growth plate on the operated side in all cases, and on the control side in four of five cases. Microradiography revealed low-density bone formation at the periphery of the previous growth plate on the operated side in two cases and on the control side in four cases. OTC-fluorescence studies revealed low-density bone formation, while histomorphometry revealed remnants of cellular elements of the growth zone on the control side. Figures 8 and 9 illustrate the results of histomorphometry analysis; Table I summarises the mean values of femoral shortening after operation for both drill sizes in comparison with controls.
Table 1. Shortening of the femur after operation compared with control side (mean values)

<table>
<thead>
<tr>
<th>Weeks</th>
<th>2 mm drill-hole (mm)</th>
<th>3 mm drill-hole (mm)</th>
</tr>
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<tbody>
<tr>
<td>3</td>
<td>*</td>
<td>2.3</td>
</tr>
<tr>
<td>6</td>
<td>0.2</td>
<td>0.8</td>
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<tr>
<td>12</td>
<td>0.5</td>
<td>3.8</td>
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<tr>
<td>24</td>
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<td>4.6</td>
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* Both femurs equal in length

**DISCUSSION**

A transepiphysal drill-hole of 2 mm in diameter (destroying 3% of the growth plate) caused no permanent growth disturbance in the distal femoral growth plate. Histomorphometry at three weeks confirmed a slight increase of cellular elements in the hypertrophic zone, although this increase was also seen on the control side. The cellular elements of both the growth zone and the hypertrophic zone decreased by 12 weeks, and the height of the epiphyseal cartilage (that is, the sum of the growth zone and the hypertrophic zone) was approximately equal on the two sides.

By contrast, a drill-hole of 3.2 mm (destroying 7% of the growth plate) did cause permanent growth disturbance and shortening of the femur. Previously, Nordentoft (1969) had estimated that a drill-hole affecting 20% of the transverse diameter of the distal femoral growth plate did not cause permanent disturbance of longitudinal growth; further, he estimated that approximately 10% of the growth plate could be destroyed without causing permanent cessation of growth if the basement plate was left intact; however, his estimations were not based on histomorphometric techniques. Another study by Khermosh et al. (1972) of the natural course of femoral growth in the rabbit revealed an average difference in length of 0.6 mm (range ± 2.5 mm).

Osseous bridge formation after a transepiphysal drill-hole of 3.2 mm was seen histologically after three weeks (Fig. 3); by this time the height of the epiphyseal cartilage from both the operated and control sides had decreased in the condylar areas, and this decrease approached significance. There was no increase in the cellular elements of the hypertrophic zone on the operated side, though a slight increase was found on the control side. The narrowing of the epiphyseal cartilage was more prominent on the operated side and was due to the retardation of the growth caused by the central osseous bridging.

Although radiological evidence of thinning of the epiphyseal line was thought to be a warning sign of epiphyscdesis after stapling (Siffert 1956), we found that radiography failed to show any bone bridge and was of little value in predicting growth disturbance. Seinsheimer and Sledge (1981) reported a correlation of the longitudinal growth rate and growth-plate thickness in rabbits at a given age, and we found that growth disturbance correlated well with the narrowing of the growth plate. Our study confirmed also the natural pattern of longitudinal growth rate in the rabbit previously described by Khermosh et al. (1972). The small nests of cartilage cells seen in the cancellous bone of the metaphysis and in the intercondylar area showed that implantation of the cellular elements of the growth plate had occurred, and that cartilage cells thus implanted could survive up to 12 weeks in growing rabbits. The occurrence of blood vessels in the drill-hole may indicate that they play a role in the formation of a bony connection between the epiphysis and metaphysis (Trueta and Amato 1960; Trueta and Morgan 1960). In conclusion, we found that histomorphometry facilitated precise analysis of the changes in the growth plate.

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**REFERENCES**


