BIOMECHANICAL STRENGTH OF NON-VASCULARISED AND VASCULARISED DIAPHYSEAL BONE TRANSPLANTS

AN EXPERIMENTAL STUDY

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We studied the healing and torsional strength of non-vascularised (28) and vascularised (28) sections of tibial diaphyses in 56 cats. Both types of graft achieved fracture union in the same period of time, and at 12 and 16 weeks the non-vascularised grafts were as strong as the vascularised grafts.

Bridging large bone defects is one of the most challenging problems in orthopaedics. Such defects arise from bone loss due to severe trauma, 'gap nonunions' following infection in open fractures, resection for bone malignancies and, rarely, after resection for congenital pseudarthrosis. Recently, free vascularised autografts have been advocated as the method of choice (Weiland and Daniel 1979). However, non-vascularised autografts are still commonly and successfully used (Enneking et al 1980).

Whilst a high incidence of stress fractures has been reported with non-vascularised grafts (Enneking et al 1980), they have also been shown to occur with free vascularised grafts (Pho et al 1985).

We report an experimental study, using adult cats, comparing the biomechanical strength of large non-vascularised diaphyseal bone transplants with that of identical vascularised autografts.

MATERIALS AND METHODS

We used 56 adult cats (the epiphyses were shown to be closed on plain radiographs) in two different experiments using 28 cats each. Intraperitoneal pentobarbitone anaesthesia was employed.

In the first experiment, a vertical incision was made on the anterolateral surface of the right shin. Two

osteotomies were made through the tibia using an oscillating saw, the proximal osteotomy being made just below the insertion of the pes anserinus. The posterior muscle pedicle containing the nutrient artery was divided. A 4 cm segment of the tibial diaphysis (at least two-thirds of the shaft) with its periosteum was removed (Fig. 1). The devascularised segment was immediately replaced in its original bed, and the soft tissues were closed with 3-0 chromic catgut sutures. The limb was then immobilised in an above-knee plaster with the knee in 90° of flexion and the foot in plantigrade position. In the second experiment, the procedure was the same except that the posterior muscle pedicle containing the nutrient artery was not divided (Fig. 2).

The healing of the bone segments in each group was

Fig. 1

A large segment (4 cm) of the diaphysis was removed with its periosteum. Below, the devascularised segment repositioned in its muscle bed.

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studied at one, two, four, six, eight, 12 and 16 weeks, using four cats for each period of observation. At sacrifice, the operated and unoperated legs in each cat were retrieved by disarticulation at the knee and ankle and fixed in 10% buffered formalin. The fracture union at both ends of the grafts was assessed clinically as well as radiologically and histologically. The soft tissues were removed from the tibiae to expose bare bone and photographs and radiographs were taken (Fig. 3). For each graft, the diaphyseal bone segment in the operated leg was removed by cutting through the callus at its two ends. An identical diaphyseal bone segment was cut from the tibia of the unoperated leg to serve as the control segment for that cat. All segments were tested within one week of retrieval and fixation.

For fixation to the testing apparatus, each bone segment was embedded in rectangular jigs (moulds) measuring $3.2 \times 2.4 \times 3.2$ cm using quick-setting dental cement, leaving the central 2 cm portion free for biomechanical testing (Fig. 4).

Torsional testing used the Shimadzu universal testing machine autographs DCS series with a torsion test device of 50 kilograms force metre. For standardisation, the proximal end of each segment was fixed to the top jig and the distal end to the bottom jig. A lateral rotation force resulted from applying the torque in a clockwise direction for the bone grafts (right leg) and an anticlockwise direction for the controls (left leg). The specimens were tested to failure at a speed of 0.18 rev/min.

Histological examination of the proximal and distal fractures of the grafts was performed using decalcified specimens. Longitudinal sections $10 \mu\text{m}$ thick were cut and stained with haematoxylin and eosin. Similar sections of the grafts were also studied after torque testing.

The biomechanical parameters studied included maximum torque, torsional stiffness and energy of absorption as calculated from the load-deformation curve obtained for each segment. The maximum values for each of these parameters was expressed as a percentage of the maximum values in the control segment of the same cat. The biomechanical data obtained for the two groups were compared and analysed using the paired Student's *t*-test.

RESULTS

Non-vascularised diaphyseal grafts took about eight weeks to achieve osseous union, by bridging with osteoid (Fig. 5), the same time taken by vascularised grafts to achieve complete union with bony callus. In neither group was there any bony union at six weeks, but at eight, 12 and 16 weeks some union was achieved in all specimens.

Histological examination of non-vascularised grafts revealed the healing processes of bone resorption, cortical new bone formation and 'callus encasement' of the

Fig. 2

Proximal and distal osteotomies completed. The posterior muscle pedicle (P) containing the nutrient artery was left intact.

Fig. 3

At six weeks the non-vascularised bone graft (on the right) was thicker than the unoperated tibia (on the left) and the callus (C) at both fractures could be clearly seen.

segment (Fig. 6). In contrast, in vascularised grafts, the osteocytes remained alive and bone resorption and cortical new bone formation did not occur (Fig. 6).

Figure 7 shows the maximum torque values for both types of bone graft. Non-vascularised grafts initially became weaker, reaching their lowest mean torque strength at four weeks (about 52%). After four weeks, these transplants progressively increased in strength reaching a mean torque strength of 100% by 12 weeks. Vascularised grafts also initially weakened up to two weeks (about 63%) after which the grafts rapidly increased in strength. Torque strength of 100% was achieved by eight weeks, the mean torque strength at four months being about 140%. Comparison between the two groups showed that at two weeks, non-vascularised

grafts were in fact stronger than vascularised ones ($p < 0.005$). At four, six and eight weeks, non-vascularised grafts were weaker than vascularised grafts; this was statistically significant. However, at 12 and 16 weeks, the non-vascularised grafts were as strong as the vascularised transplants. No statistical difference was observed between the two groups.

Figure 8 shows that the torsional stiffness of non-

vascularised grafts also decreased up to four weeks (about 63%), after which it increased with time. The torsional stiffness of non-vascularised grafts approached a mean value of 100% by eight weeks. Statistical analysis revealed no difference between the two groups, except at two weeks, when the value for non-vascularised grafts (about 78%) was greater than that for vascularised grafts (about 58%).

Fig. 4

The bone segment was mounted in rectangular jigs and loaded to failure, the torsion force applied producing an oblique fracture. Only the central 2 cm of the bone segment was left free for torsional testing.

Fig. 5

Osteoid (O) was seen at proximal (a) and distal (b) osteotomy sites in a non-vascularised graft at eight weeks. Similar osseous union was seen both proximally (c) and distally (d) in a vascularised graft at eight weeks.

Fig. 6

Cross-section of a non-vascularised graft (a) at eight weeks showing resorption cavities (RC) and callus encasement (CE). At higher magnification (b) a resorption cavity with new bone formation (NB) has osteocytes (O) trapped in it. A ghost cell (G) with empty lacunae is also seen. Cross-section of a vascularised graft at eight weeks (c). No obvious resorption cavity or callus encasement was visible. Even at a magnification of $\times 200$ (d), no resorption cavity was seen. Living osteocytes (O) were seen in the cortex.

Fig. 7

Graph showing maximum torque of non-vascularised and vascularised grafts.

The energy of absorption for non-vascularised grafts also decreased up to four weeks (about 67%), after which it progressively increased with time (Fig. 9). The value for non-vascularised grafts reached 100% by eight weeks. Statistical analysis between the two groups again revealed no significant difference, except at four weeks, when the value for non-vascularised grafts (about 67%) was significantly smaller than that for vascularised grafts (about 87%).

DISCUSSION

The cat tibia was chosen as the experimental model for this study because, compared with dogs and rabbits, its morphology most closely resembles that of man.

It was interesting to note that non-vascularised grafts took the same period of time to achieve fracture union as vascularised grafts. Similar findings were reported by Dell, Burchardt and Glowczewskie in 1985.

Burstein et al (1975) and Yu et al (1975) showed that the strength of bone was related to its mineral content. In non-vascularised bone grafts, the strength depended on the reparative processes taking place in such segments (Enneking et al 1975). The mineral content varied according to whether bone resorption or new bone formation was predominating.

In the cat tibia, the torsional strength of non-vascularised transplants was reduced up to four weeks, after which it progressively increased in strength. This pattern of biomechanical events was similar to that in adult mongrel dogs described by Enneking et al (1975), with an initial weakening phase due to resorption activity followed by a phase of increasing torsional strength as new bone formation predominated. However, in the feline model, 100% torque strength was reached as early as 12 weeks. This was in marked contrast to the findings in Enneking's series, where the transplants continued to weaken from six weeks to six months and approached 100% torque strength only by about one year. It is difficult to explain such a gross discrepancy, although, in the canine model, the transplants were subperiosteal whereas in our study the periosteum was not removed. However, Dell et al (1985) found no difference in the torsional strength between subperiosteal and periosteum-encased non-vascularised grafts in dogs. Another difference was the fact that in the canine model, the experiments were performed on the fibula, which in the dog is fused to the tibia in the mid-shaft portion and is therefore not fully weight-bearing. In our feline model, the tibia is fully weight-bearing and is therefore subject to more stress. In accordance with Wolff's Law, the return to normal biomechanical function is expected to be faster in the cat tibia.

In this study, vascularised bone grafts were found to achieve 100% torque strength by eight weeks. A review of the literature revealed that this is the first study describing the changes in biomechanical strength of

Fig. 8

Graph showing torsional stiffness of non-vascularised and vascularised grafts.

Fig. 9

Graph showing energy of absorption of non-vascularised and vascularised grafts.

vascularised grafts with time. Since vascularised grafts did not undergo the repair processes of resorption and apposition and retained their normal architecture (Doi, Tominaga and Shibata 1977), it is difficult to explain why they passed through an initial weakening phase. This could be due to disuse porosis following immobilisation, the decrease in mineral content being facilitated by the intact vessels.

Several studies have shown that vascularised grafts are significantly stronger than conventional non-vascularised grafts (Davis, Mazur and Coleman 1982; Moore et al 1984). In contrast, our study shows that non-vascularised grafts are equally strong.

Another interesting feature was that torsional stiffness in non-vascularised grafts reached normal values (100%) even earlier than did torsional strength. In fracture healing, normal stiffness is achieved earlier than normal strength (Henry, Freeman and Swanson 1968; Davy and Connolly 1982). The same phenomenon could also be expected to occur with healing of bone transplants.

It is relevant to point out that, whilst both non-vascularised and vascularised grafts incorporated readily in adult cats, in patients incorporation of such grafts takes a longer time.

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