MICROVASCULAR TRANSPLANTATION OF PHYSEAL ALLOGRAFTS

MARTIN I. BOYER, JAYNE S. DANSKA, LIANA NOLAN, AHMET KIRAL, C. VAUGHAN A. BOWEN

From The Hospital for Sick Children, Toronto, Canada

We compared growth in vascularised allograft transplants, autografts and in non-operated physes in rabbits immunosuppressed with cyclosporin A and in non-immunosuppressed animals. Molecular haplotyping was undertaken before operation to ensure allogenicity. Postoperative bone scans and fluorochrome labelling were used to confirm physeal vascularity. The animals were killed at three or five weeks.

Proximal tibial physeal autografts, with or without cyclosporin A, or allografts with cyclosporin A, grew at similar rates to the physes of non-operated rabbits. All the operated physes grew at rates significantly greater than their contralateral controls. 99mTc-MDP bone scans accurately predicted the viability of the epiphyseal plate. Quantitative histomorphological analysis of the heights of the physeal proliferative and hypertrophic zones showed that successful physeal transplants have a normal appearance, but when unsuccessful have thickened hypertrophic zones compatible with physeal ischaemia.

We discuss the significance of these results in relation to the transplantation of physes in children.


Received 12 September 1994; Accepted 23 December 1994

Musculoskeletal deformities caused by trauma, tumour, infection or congenital malformation or absence of the physeal plate are currently managed by a variety of reconstructive procedures including autogenous or allogeneic cancellous bone grafts (Phemister 1947; Bosse and Robb 1992; Lindner and Henry 1992), non-vascularised cortical allografts (Enneking and Mindell 1991; Stevenson, Li and Martin 1991; Stevenson and Horowitz 1992), vascularised bone and composite tissue transfers (Weiland 1981; Han et al 1992), the Ilizarov technique of distraction osteogenesis (Ilizarov and Deviatov 1969; DeBastiani et al 1987; Bell, Boyer and Armstrong 1992) and commercially manufactured implants.

In children these techniques do not necessarily address the actively growing nature of the abnormality and may have high complication rates. The development of micro-surgical techniques has enabled free tissue transplantation to be done successfully, and vascularised physeal transplantation has become a biologically attractive solution to the problem of reconstruction of defects or deformities of the immature appendicular skeleton. Experimental studies on vascularised orthotopic and heterotopic transplantation of autografts and allografts have been reported since 1979 (Donski, Carwell and Sharzer 1979; Donski and O'Brien 1980; Zaleske et al 1982; Brown et al 1983; Bowen and O'Brien 1984; Nettelblad, Randolph and Weiland 1984, 1985; O'Brien et al 1984; Bowen 1986, 1988; Bowen et al 1987, 1988a,b, 1995; Nunley et al 1987; Paskert et al 1987; Randolph et al 1987; Bowen, O'Brien and Gumnely 1988; Pho et al 1988; Zhong-Wei and Guang-Jian 1988; Nolan and Bowen 1993; Boyer, Bray and Bowen 1994) and some clinical cases have now been described (O'Brien et al 1984; Nunley et al 1987; Zhong-Wei and Guang-Jian 1988), but the procedure is not common because of the difficulty of obtaining suitable donor tissue.

Allograft transplants are attractive because cadaver donor tissue can be used. Vascularised allograft transplantation of epiphyseal plates has not been attempted in man since transplant recipients require systemic immunosuppression to prevent rejection of the composite tissue transplant (Yaremchuk, Sedarac and Schiller 1983; Yaremchuk et al 1985; Gottfried et al 1987; Paskert et al 1987, 1988; Randolph et al 1987; Gornet et al 1991; Innis et al 1991; Lee et al 1991b). Experiments which allow assessment of the function of physeal allografts after vascularised transplantation are necessary so that when drugs which are less toxic become available, surgeons will be ready to perform transplants with confidence.

We describe an experiment in which we examined the rate of growth of successful allograft transplants and the quantitative histomorphology of the successfully vascu...
Table I. Experimental design showing CsA-treated and CsA-untreated groups and operated and non-operated physes

<table>
<thead>
<tr>
<th>Group</th>
<th>Allograft immunosuppressed</th>
<th>Operated and non-operated physes</th>
<th>3 weeks: n = 8</th>
<th>5 weeks: n = 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Autograft immunosuppressed</td>
<td>Operated and non-operated physes</td>
<td>3 weeks: n = 9</td>
<td>5 weeks: n = 5</td>
</tr>
<tr>
<td>2</td>
<td>Autograft, non-immunosuppressed</td>
<td>Operated and non-operated physes</td>
<td>3 weeks: n = 11</td>
<td>5 weeks: n = 6</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>(B) Control</th>
<th>Immunosuppressed</th>
<th>Operated physes only</th>
<th>3 weeks: n = 10</th>
<th>5 weeks: n = 5</th>
</tr>
</thead>
<tbody>
<tr>
<td>4</td>
<td>Non-immunosuppressed</td>
<td>Operated physes only</td>
<td>3 weeks: n = 10</td>
<td>5 weeks: n = 5</td>
</tr>
</tbody>
</table>

Molecular haplotyping. Donor and recipient rabbits undergoing vascularised allograft physeal plate transplantation were tissue-typed using restriction fragment-length polymorphism (RFLP) analysis of the major histocompatibility complex (MHC) class-I and class-II genes (Boyer, Bowen and Danska 1995). To be considered allogeneic, and thus eligible for transplantation, donor and recipient pairs needed to have differing RFLP patterns at two of the loci.

Operative technique. A modification of the method of Zaleske et al (1982) was used. Under aseptic conditions, a medial incision was made from mid-thigh to mid-leg. The division of the femoral vessels into the popliteal and saphenous branches was located between the quadriceps and adductors. The popliteal vessels were then dissected as far as their branches above the knee. The quadriceps were divided just above the patella and reflected proximally for 2 cm. The adductors and hamstrings were divided close to their insertions and reflected proximally. The muscles of the anterior, lateral and posterior compartments of the leg were divided close to their origins and reflected distally for 2 cm. The anterior and posterior tibial vessels were identified and ligated about 1 cm distal to the proximal tibial epiphyseal plate, leaving the knee covered by its capsule and vascularised by the popliteal vessels through the nutrient artery, metaphyseal periosteal arteries and the epiphyseal circulation (Fig. 1). Osteotomies were made 2.5 cm proximal and distal to the joint line, and the cut bone ends were plugged with wax to prevent blood loss. The popliteal vessels were then divided and the knee was removed and kept moist in saline at room temperature. The distal part of the limb was then amputated, leaving a stump 10 cm long with the knee joint intact.

Table II. Experimental procedure

<table>
<thead>
<tr>
<th>Procedure</th>
<th>Details</th>
</tr>
</thead>
<tbody>
<tr>
<td>14 days preoperatively</td>
<td>7-week-old female New Zealand white rabbits arrive</td>
</tr>
<tr>
<td>12 days preoperatively</td>
<td>5 ml of blood taken for RFLP typing (transplant donors or recipients only)</td>
</tr>
<tr>
<td>7 days preoperatively</td>
<td>Cyclosporin A dosing begins (for immunosuppressed controls, replants or transplants)</td>
</tr>
<tr>
<td>4 days preoperatively</td>
<td>RFLP analysis completed; donor and recipient pairs are mismatched at class-I and class-II major histocompatibility loci</td>
</tr>
<tr>
<td>Day 0</td>
<td>Surgery (tibial screw, replant or transplant), blood drawn for CsA level, initial radiograph taken postoperatively</td>
</tr>
<tr>
<td>Day 3</td>
<td>CsA assay result returns, allowing adjustment of dosage as required</td>
</tr>
<tr>
<td>Day 7</td>
<td>$^{99m}$Tc-MDP bone scan, radiography, blood drawn for CsA level</td>
</tr>
<tr>
<td>Day 10</td>
<td>CsA assay result returns allowing adjustment of dosage as required</td>
</tr>
<tr>
<td>Day 21</td>
<td>Radiography, blood drawn for CsA level; tetracycline and killing of rabbits for three-week follow-up</td>
</tr>
<tr>
<td>Day 24</td>
<td>CsA assay result returns allowing adjustment of dosage as required</td>
</tr>
<tr>
<td>Day 35</td>
<td>Radiography, blood drawn for CsA level, tetracycline and killing of rabbits for five-week follow-up</td>
</tr>
<tr>
<td>Day 38</td>
<td>CsA assay result returns</td>
</tr>
</tbody>
</table>

CsA assays for blood levels are drawn as required at any time during the course of the experiment if the rabbit shows any clinical signs of toxicity or wasting.
remained well vascularised through the saphenous system, and care was taken to preserve the major trunk of the tibial nerve.

Skeletal reconstruction was carried out with a 4-hole AO mini-fragment plate for the tibia and a threaded 2.8 mm Kirschner wire for the femur (Fig. 1), followed by microvascular anastomosis of the popliteal vein and artery, repair of the musculotendinous units and closure of the skin.

Sterile dry dressings were applied after a screw had been placed in the left tibia as a marker for growth of the contralateral proximal tibial epiphyseal plate.

All rabbits were allowed to move about their cages freely, and were fed a standard diet of rabbit food pellets and bottled water.

**Radiological follow-up.** Standardised teleoradiographs were taken immediately after operation and at one, three and five weeks (Fig. 2). The increase in distance (mm) between the proximal end of the tibial eminence and the proximal end of the mini-fragment plate used for tibial fixation was calculated to measure growth of the proximal tibial physis. Calculation of growth in the contralateral control proximal tibial physis in the non-transplanted or replanted controls was made from measurements from the proximal end of the tibial eminence to the proximal end of the marker screw.

**Fig. 1**
Diagram to show femoral fixation with a threaded intramedullary Kirschner wire drilled anterograde through the distal fragment and then retrograde through the osteotomy site. The tibia is fixed by a mini-fragment plate and four screws. After skeletal fixation, microvascular anastomosis of the popliteal vein and artery is performed (A, adductor magnus muscle; B, reflected head of gastrocnemius muscle; C, lateral head of gastrocnemius muscle; D, anterior tibial musculature; E, quadriceps muscles; F, femur; G, tibia; H, tibial nerve; I, popliteal artery and vein; J, saphenous artery and vein; K, threaded intramedullary Kirschner rod; and L, AO mini-plate and screws).

**Fig. 2a**
Teleoradiographs taken immediately postoperatively and at five weeks of the non-operated (a,b) and operated (c,d) tibiae of a rabbit which had vascularised allograft transplantation of the right proximal tibia. The transplanted physis grew to a greater extent than the non-operated side.
Photomicrographs of the successfully (a) and unsuccessfully (b) revascularised proximal tibial epiphysial plate. Note the normal architecture of the epiphysial plate. The unsuccessfully revascularised physis has increased thickness, disorganisation and hypercellularity of the hypertrophic zone (A, epiphysial bone; B, epiphysial vasculature; C, resting zone; D, proliferative zone; E, hypertrophic zone; F, metaphysial zone) (X40).

Bone scanning. A quantitative $^{99m}$Tc-methylene-diphosphonate (MDP) bone scan was done one week after operation to measure the radionucleide uptake of the proximal tibial physis (Fig. 3).

Maintenance of cyclosporin A levels. Cyclosporin A (CsA) was administered daily by subcutaneous injection at a dose of 10 mg/kg/day. Blood levels of 200 to 300 μg/l were maintained. CsA levels were determined at the time of operation, and at 1, 3 and 5 weeks postoperatively, using the Incstar Cyclotrak SP (Incstar, Stillwater, Minnesota) whole blood radioimmunoassay (Wong and Ma 1990; Lee et al 1991a; Wong 1991; Wong 1992).

Histomorphometry. One hour before killing, each rabbit was given an intraperitoneal injection of tetracycline (Milch, Rall and Tobie 1958; Frost 1969) at a minimum dose of 50 mg/kg. The animals were killed at either three or five weeks after operation by injection of 2 ml of a pentobarbital-containing solution (Euthanyl; MTC Pharmaceutical, Cambridge, Canada) into a marginal ear vein. Both right and left proximal tibiae were removed, placed in 50% ethanol and stored at 4°C.

Histological sections of the proximal tibial growth plate were cut in the sagittal plane, 1 mm lateral to the midline. All sections were stained with safranin O (Fig. 4). Separate unstained thick sections were examined for tetracycline fluorescence (Fig. 5). All epiphysial plates were examined using a Leitz microscope with a Vario-Orthoplan 2 camera attachment (Leica, Germany).
Fluorescence micrograph showing fluorochrome uptake (arrow) in the region of the metaphysis confirming its vascular supply immediately before killing (×40).

The sectioned and mounted growth plates were divided into anterior, middle and posterior sections, corresponding to the regions of the growth plate supplied by the metaphyseal periosteal arteries (anterior and posterior sections) and the nutrient artery (middle section). For each epiphyseal plate, 10 × 15 cm photomicrographs were taken of the central area and each of the two peripheral areas at a standardised orientation and uniform magnification ×60 (Fig. 6). Both the experimental and the contralateral limb control physes were photographed.

The average height of the proliferative and hypertrophic zones of cell columns was determined from the photographs. Each photograph was overlaid with a sheet of clear plastic and the margins of the germinal, proliferative and hypertrophic zones were separately determined and traced by an observer who did not know the state of the physis. Their areas were manually counted using a clear plastic grid of squares measuring 2.5 × 2.5 mm. By keeping the magnification and orientation of the photomicrographs constant, the area measured was mathematically related to the average height of the proliferative and hypertrophic zones of the growth plate within the area of the photomicrograph using the formula height = area/width. Zone heights of the epiphyseal plate were measured in micrometers.

**Statistical analysis.** Comparisons of growth data for the operated physes from each of the five groups was by one-way ANOVA and post-test Tukey analysis. Comparison between operated and non-operated physes for each of the three transplant groups (groups 1, 2 and 3) was by repeated-measures ANOVA and Tukey analysis.

These methods were also used to compare the heights of the resting, proliferative and hypertrophic zones of both the operated and non-operated legs of all groups, including those with failed anastomoses.

**RESULTS**

Seventy rabbits were operated on; 22 died before the ⁹⁹ᵐTc-MDP bone scan could be performed one week after operation and were excluded from further analysis.

**Major histocompatibility complex haplotypes.** All donor and recipient rabbits were allogeneic for the rabbit MHC based on RFLP haplotyping (Marche et al 1989; Boyer et al 1995).

**Longitudinal growth.** The mean longitudinal growth (±SD) of the allografts, autografts (CsA-treated and CsA-untreated) and both control groups (CsA-treated and CsA-untreated) is summarised in Tables III and IV. Box-whisker diagrams showing the median and relevant centile values

---

**Table III. Longitudinal growth (mm; mean ± sd) of experimental and control physes at three weeks**

<table>
<thead>
<tr>
<th>Group*</th>
<th>Operated leg</th>
<th>Non-operated leg</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 (n = 8)</td>
<td>6.06 ± 1.08</td>
<td>3.75 ± 0.89</td>
</tr>
<tr>
<td>2 (n = 9)</td>
<td>5.56 ± 1.79</td>
<td>2.83 ± 1.15</td>
</tr>
<tr>
<td>3 (n = 11)</td>
<td>6.36 ± 1.21</td>
<td>3.73 ± 1.47</td>
</tr>
<tr>
<td>4 (n = 10)</td>
<td>4.72 ± 1.00</td>
<td>NA</td>
</tr>
<tr>
<td>5 (n = 10)</td>
<td>6.95 ± 1.79</td>
<td>NA</td>
</tr>
</tbody>
</table>

* see Table 1

**Table IV. Longitudinal growth (mm; mean ± sd) of experimental and control physes at five weeks**

<table>
<thead>
<tr>
<th>Group*</th>
<th>Operated leg</th>
<th>Non-operated leg</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 (n = 3)</td>
<td>8.50 ± 1.32</td>
<td>6.67 ± 0.58</td>
</tr>
<tr>
<td>2 (n = 5)</td>
<td>9.70 ± 3.23</td>
<td>4.80 ± 1.35</td>
</tr>
<tr>
<td>3 (n = 6)</td>
<td>11.08 ± 2.15</td>
<td>8.50 ± 1.41</td>
</tr>
<tr>
<td>4 (n = 5)</td>
<td>7.30 ± 1.40</td>
<td>NA</td>
</tr>
<tr>
<td>5 (n = 5)</td>
<td>11.50 ± 1.00</td>
<td>NA</td>
</tr>
</tbody>
</table>

* see Table 1
The growth of orthotopic allograft proximal tibial physes was not significantly different from that of either autograft physes (CsA-treated and CsA-untreated) or the control group at three weeks ($F_{df = 4.42} = 2.97$, $p = 0.03$) and at five weeks ($F_{df = 4.19} = 2.61$, $p = 0.068$). Tukey analysis of the growth data at three weeks showed no significant intergroup difference between the transplanted physes of the allograft groups and the operated physes of the other groups of rabbits ($p > 0.05$).

The operated physes grew at rates significantly greater than the contralateral non-operated physes in each of the three transplant groups at three weeks ($F_{df = 1.25} = 137.12$, $p < 0.0001$) for between-leg effect) and at five weeks ($F_{df = 1.11} = 45.30$, $p < 0.0001$ for between-leg effect). There was no significant between-group effect on repeated-measures ANOVA.

**Qualitative radionuclide uptake.** All epiphyseal plates showing a positive uptake on $^{99m}$Tc-MDP bone scans one week after operation had continued longitudinal growth at both three and five weeks. Epiphyseal plates which showed no radionuclide uptake at one week had no longitudinal growth after three weeks and on gross examination at postmortem all these knees appeared dead.

No differences were seen between the $^{99m}$Tc-MDP uptake of the proximal tibial epiphyseal plates of the immunosuppressed and non-immunosuppressed groups of control rabbits.

**Qualitative histomorphology.** All transplanted or replanted physes which showed longitudinal growth had positive fluorochrome uptake in the subepiphyseal metaphysis, indicating a patent metaphyseal vascular supply with active calcification of osteoid.

No histological evidence of rejection was seen in the immunosuppressed recipients of successful vascularised allograft transplants.

**Quantitative histomorphology.** The average thickness of the operated (transplanted, replanted and unrelated control) proximal tibial epiphyseal plates decreased significantly between three and five weeks, from $440.8 \mu m$ to $358.9 \mu m$ ($t_{df = 33} = 2.98$, $p = 0.005$) but the non-operated epiphyseal plates showed no significant decrease in height ($423.0 \mu m$ to $397.2 \mu m$, $t_{df = 33} = 1.0432$, $p = 0.30$).

The thickness of the anterior, middle and posterior regions of the hypertrophic zones of failed transfers was significantly greater than for all other groups of transplants, replants and controls ($F_{ant \ df = 5.18} = 10.46$, $p = 0.0001$; $F_{mid \ df = 5.18} = 13.84$, $p = 0.0001$; $F_{post \ df = 5.18} = 8.89$, $p = 0.0002$).

Tukey analysis showed no significant differences in the thickness of the three regions of the resting, proliferative or hypertrophic zones between groups of successfully vascu-
Vascularised CsA-treated transplants, CsA-treated or non-treated replants or CsA-treated or non-treated controls at either three or five weeks.

Some significant regional variations were seen. The anterior region of the resting zone was significantly thicker than both the middle and posterior regions of the resting zone at both three (66.76 mm > 46.24 mm and 44.22 mm) and five weeks (82.25 mm > 53.36 mm and 52.59 mm). The proliferative and hypertrophic zonal thickness did not vary significantly with respect to region at either three or five weeks.

**DISCUSSION**

Vascularised physeal allografts have many possible clinical applications. They may replace absent physes or those damaged by tumour, infection, trauma or congenital malformation (Zuker and Bowen, personal communication, 1990), facilitate augmentation of skeletal growth (Bowen et al 1988c), replace interpositional fat, cranioplast or silastic after physeal bony bridge excision (Peterson, personal communication, 1993) or be a component of whole-limb transplantation in massive trauma in children (Nolan and Bowen 1993). We have examined four aspects of the transplantation of vascularised orthotopic physeal allografts. Each will be considered separately.

This is the first study of vascularised transplantation of solely physeal allografts using precise molecular typing to demonstrate donor-recipient pair allogenicity. Allogenicity must be defined, not assumed, if conclusions are to be drawn about the comparative behaviour of vascularised allograft physeal transplants as compared with syngeneic autografts or controls.

**Longitudinal growth.** There have been few studies of vascularised allograft epiphyseal plate transplants in immunosuppressed hosts other than that of Ford, Gerber and Green (1987).

Our results confirm those of Ford et al in that at both three and five weeks longitudinal growth of vascularised proximal tibial physeal allografts was not significantly different from that of the vascularised autograft physes (CsA-treated and CsA-untreated) and the non-operated control physes (CsA-treated and CsA-untreated). The administration of CsA did not depress longitudinal skeletal growth in either autografts or allografts which had been successfully revascularised.

The left proximal tibial physes, which did not undergo operation and acted as a control, grew significantly less than that on the operated right side. The overall growth of the successfully vascularised physeis is therefore likely to be due to both stimulatory and inhibitory influences.

Historically, physeal stimulation has been attempted by various means with the aim of increasing the blood flow to the physeis, and thus enhancing longitudinal growth of the limb. Sympathectomy, osteotomy and fixation and periosteal stripping have all been tried with varying degrees of success to increase blood flow to an affected limb segment. Each of these procedures was carried out as part of the transplant operation itself. These factors not only play a role in the growth of physes after transplant, but also in the clinical setting. Similar operations to those carried out on the rabbit knees would be performed with human joint transplants; comparison is therefore reasonable.

The blood flow to the operated limb is increased as a result of a major operation, as demonstrated by the increased overall uptake of 99mTc-MDP on the bone scan, and is likely to contribute to the accelerated growth of the proximal tibial physeis. Although the animals did eat and drink normally, however, their weight gain was less and they tended to dehydrate more readily than control rabbits not undergoing operation. This was particularly so in the CsA-treated animals, although none of the experimental animals was infected on postmortem examination. The observation that the growth of the operated physes in groups 1, 2 and 3 was not significantly different from that of non-operated physes of control groups may therefore be due to a balance of both stimulatory and inhibitory influences.

**Radionucleide uptake on qualitative 99mTc-MDP bone scans.** Radionucleide uptake is due to a variety of factors including local blood flow and osteogenic activity. All scans of the operated proximal tibial physes (CsA-treated and CsA-untreated) showed measurably greater uptake than those of the contralateral non-operated physes. All successfully vascularised physes had also grown more than their contralateral controls at three and five weeks. Since all physes showing no 99mTc-MDP uptake at one week had no longitudinal growth at three weeks then 99mTc-MDP bone scans reliably predict growth after transplantation.

**Comparative quantitative histomorphology.** Previous investigators have shown that successfully revascularised heterotopically or orthotopically replanted autogenous physes show normal histomorphological features and zonal architecture. We have shown that this is also true for successfully revascularised allograft epiphyseal plates.

We did not find any statistically significant differences in the thickness of the anterior, middle or posterior regions of the resting, proliferative or hypertrophic zones in successfully revascularised allograft transplants and the other groups of autografts and controls.

Failed transplants showed highly significant increases in overall physeal thickness localised to the hypertrophic zone. The thickness of the proliferative zone remained unchanged, as has been previously shown (Trueta and Amato 1960; Trueta and Little 1960; Trueta and Morgan 1960; Trueta and Trias 1961; Trueta 1963). These authors demonstrated that the hypertrophic zone of cells thickens in the absence of metaphyseal blood supply and that it becomes hypercellular and disorganised before physeal death occurs; hypertrophic zone cells normally grow and differentiate in an environment of low oxygen tension.

No histological signs of rejection were seen in the
metaphysis, physis or epiphysis of any of the allograft transplants. Treatment with CsA was certainly responsible for this since it has been previously shown that vascularised allograft musculoskeletal tissue undergoes vigorous rejection in the absence of immunosuppression (Fritz et al 1984; Gottfried et al 1987; Paskert et al 1988; Hutchinson 1991). This is due to the abundant class-I antigen on all vascular endothelium as well as the class-II molecules present on the cells in the transplanted marrow.

CONCLUSIONS

Cyclosporin A and related immunosuppressant drugs have revolutionised solid organ transplantation. When revascularised by operation at the time of transfer, experimentally transplanted epiphysial plates survive and continue to grow in the presence of cyclosporin A (Ford et al 1987; Paskert et al 1988).

Microvascular transplantation of cadaver epiphyseal plate allografts is in its infancy, both clinically and experimentally. Immunosuppression of the recipient is required to prevent allograft rejection due to the highly-expressed class-I and class-II major histocompatibility complex antigens on the vascular endothelium and marrow cells.

At present, the risks of non-specific systemic immunosuppression still outweigh the benefits of this form of biological limb reconstruction in children, but the use of specific immunosuppressive techniques will allow the successful transplantation of vascularised epiphyseal plates.

The authors wish to thank Ms Nancy Fahrner and Mr Derek Stephens for their help with the statistical analysis, Mr Ajai Kumar for his anaesthetic expertise and Ms Normi Pettel for processing the histological sections.

No benefits in any form have been received or will be received from a commercial party related directly or indirectly to the subject of this article.

REFERENCES


