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ORIGINAL PAPER

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A prospective randomized trial of percutaneous marrow injection in a series of closed fresh tibial fractures

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Abstract We performed a prospective, randomized study on 40 patients with fresh closed fractures of the tibial shaft to determine the effect of autologous bone marrow injection on the union rate. Forty patients were randomized to two injections with 15 ml of autologous bone marrow injections at the fracture site and casting or conventional casting. Fracture union measured by absence of localized tenderness and mobility and bridging of three out of four cortices at the fracture site on plain roentgenograms was assessed at 3, 4, and 5 months of treatment. All fractures receiving bone marrow injections united in 3.65±0.49 months; 19/20 fractures treated conventionally united in 4.31±0.48 months (p=0.0004). Other possible determinants of union, complication rates, and cost incurred in the treatment were similar in the two groups.

Résumé Nous avons exécuté une étude prospective et randomisé sur 40 malades avec des fractures diaphysaires tibiales fermées fraîches pour déterminer l'effet d'injection de la moelle osseuse autologue sur le taux de consolidation. Quarante malades ont été randomisés avec deux injections de 15 ml de moelle osseuse autologue à l'emplacement de la fracture et contention par plâtre ou contention conventionnelle par plâtre. La consolidation a mesuré par l'absence de mobilité localisée et par la fusion de trois des quatres corticales sur des clichés ordinaires réalisés à 3, 4 et 5 mois de traitement. Tout les fractures qui ont recus des injections de moelle ont consolidé en 3.65±0.49 mois; 19/20 fractures traitées conventionnellement ont consolidé dans 4.31±0.48 mois (p=0.0004). Les autres déterminants possibles de la consolidation, la fréquence des complication et le coût du traitement était semblable dans les deux groupes.

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Introduction

Autologous bone grafting is a standard procedure for treatment of delayed union and nonunion of fractures. However, operative harvesting and implantation of the fracture site with the graft is not without complications at the donor site and the recipient site. Painful scar, hematoma, neuroma, infection, fracture or subluxation, and gait disturbance have been reported among the problems at the donor site. In addition, the need to open the fracture site has added to the risk of infection or devascularization of the fracture end where healing is already impaired. Therefore, a simple method of providing cellular reinforcement to the fracture site is expected to enhance the probability of union [3].

The technique of percutaneous aspiration and injection of bone marrow may offer the advantage of treating fractures without operative exposure of either the donor or recipient site. The procedure is safe, simple, less time consuming, and less costly as compared to autologous bone grafting. It can be performed as an out patient procedure.

A number of animal studies have demonstrated the osteogenic property of bone marrow stromal or stem cells in fresh fractures [9, 11, 12]. Autologous bone marrow injection has also been used successfully in the treatment of delayed union and nonunion of tibia fractures in human beings [2, 4]. However, there are no clinical studies demonstrating the role of bone marrow injection in fresh fractures.

Various other methods such as ultrasound and bone morphogenetic protein have been successfully used to stimulate healing in fresh fractures. Heckman et al. demonstrated significant decrease in union time in tibial fractures treated by ultrasound stimulating device as an adjunct to conventional treatment with a cast [7]. Govender et al., in a prospective randomized controlled trial in 421 open tibial fractures, showed that application of recombinant human bone morphogenetic protein-2 (rhBMP2)-impregnated absorbable collagen sponge over the fracture along with intramedullary nail fixation was

significantly superior to intramedullary nail fixation alone in reducing the frequency of secondary intervention (because of delayed union) and accelerating fracture and wound healing [5]. But these techniques are expensive and may not be available at most centers in third world countries.

The purpose of this study was to determine whether bone marrow injected percutaneously in closed tibial fractures leads to increased bone production and reduces the time required for union or increases the chances of union. Additional costs and risks were also compared between the experimental and the control groups.

Materials and methods

After informed consent and approval by the institutional research and ethical committee, 40 consecutive cases of fresh (<7 days old) closed fractures of the tibial shaft in skeletally mature patients treatable conservatively who attended the outpatient or emergency department during May-December 2002 were included in the study. They were randomly allocated using a number-generation technique to receive marrow injection in 20 cases as an adjunct to conventional treatment (experimental group) and conventional treatment alone in the other 20 cases (control group). Thirty cases not treatable conservatively were excluded on grounds of being open fractures (25), ipsilateral fracture of the femoral shaft (two), and old maluniting fractures (three).

All patients in both groups were treated by closed manipulation and immobilization in above-knee plaster of Paris cast under general or regional anesthesia with no toe touch, two-crutch-based three-point gait mobilization at 72 h. At 6 weeks, a patellar tendon weight bearing (PTB) walking cast was given till clinicoradiological union. In addition, the experimental group received at the fracture site (determined by palpation) aseptically 15 ml of bone marrow aspirated in 3–5 ml aliquots (to avoid venous dilution) by a 20 cc disposable syringe through a 16G bone marrow aspiration needle from four different sites, 1 cm. apart on the posterior part of the iliac crest. This was administered at initial manipulation and 6 weeks (at the time of change to PTB plaster as it is logistically more convenient to the patients, especially in Nepal's mountainous terrain with poor transport facilities). The amount of 15 ml of bone marrow was chosen as it is the minimum effective volume reported in the literature [4]. Plain roentgenograms of the fracture site—anteroposterior and lateral views—were done at 6 weeks and the 3rd, 4th, and 5th months of injury in both groups.

Union was measured clinically by absence of local tenderness and mobility and painless weight bearing, and radiologically by bridging callus across three out of four cortices at the fracture site. All criteria were necessary before pronouncement of union [1, 6, 8]. All patients were followed for 5 months after the initial treatment.

Bone marrow injection, assessment of clinical union, and assessment of radiological union were done by three different investigators blinded to each other to prevent observer bias. Randomization was tested by comparing the two groups on variables like age,gender, duration of injury, type of fracture, site of fracture, and associated fracture fibula considering both the magnitude and significance of difference. Student's t test was used for continuous normally distributed variables, Kruskal Wallis-H (KW) statistics for not normally distributed variables, and κ^2 for categorical data. KW statistics were used if the Bartlett's test of homogeneity of variance showed inequality of population variances. Difference in mean union time was then tested between the two groups.

Table 1 Comparison of variables in both groups

Variables	Experimental group (n=20)	Control group (n=20)	P value
Mean age (years)	37.7±16.31	43.65±17.10	0.23
Gender	13 male, 7 female	17 male, 3 female	0.97
Duration of fracture (hours)	9.15±12.70	15.80±36	0.80
Mechanism of injury			0.43
Road traffic accident	6	11	
Fall from height	8	6	
Playground injury	4	2	
Trivial injury	2	1	
Fracture site			0.64
Proximal third	3	5	
Middle third	6	4	
Distal third	11	11	
Fracture type			0.63
Transverse	8	11	
Oblique	7	5	
Comminuted	5	4	
Fracture fibula	14	14	0.63

Results

The mean age was 40.6±16.76 years. The mean age in the experimental group was 37.7±16.31 and in the control group 43.65±17.10 years. The two groups were comparable for age, gender distribution, duration of fracture, mechanism of injury, fracture site, fracture type, and associated fracture fibula (Table 1) suggesting successful randomization.

In the experimental group, 7/20 fractures united at 3 months and the remaining thirteen united at 4 months with a mean union time of 3.65±0.49 months (Fig. 1). In the control group, none of the fracture showed union at 3 months. Thirteen fractures united at 4 month, six at 5 months, and one remained ununited (Fig. 2). The mean union time in the control group was 4.31±0.48 months. The union time was significantly less in the experimental group as compared to the control group (p=0.0004). The ununited transverse fracture at the junction of middle and distal third of the tibia with segmental fracture fibula in the control group was treated by open reduction and internal fixation with intramedullary nailing supplemented with autogenous corticocancellous iliac crest bone grafting. The fracture subsequently united.

Asymptomatic anterior angulation (<10°) was observed in two cases each in both experimental and control groups. Asymptomatic posterior angulation (<10°) was seen in four cases in the experimental group and two cases in the control group. Asymptomatic varus angulation (<5°) was present in two patients in the experimental group. There was no valgus angulation. The two groups were comparable on each of the above grounds.

There was full range of knee movement at the last follow-up at 5 months in both groups. The average cost during the 5 months of treatment in the experimental group was US$79.00, whereas in the control group, the cost was US$77, which was comparable. The cost of

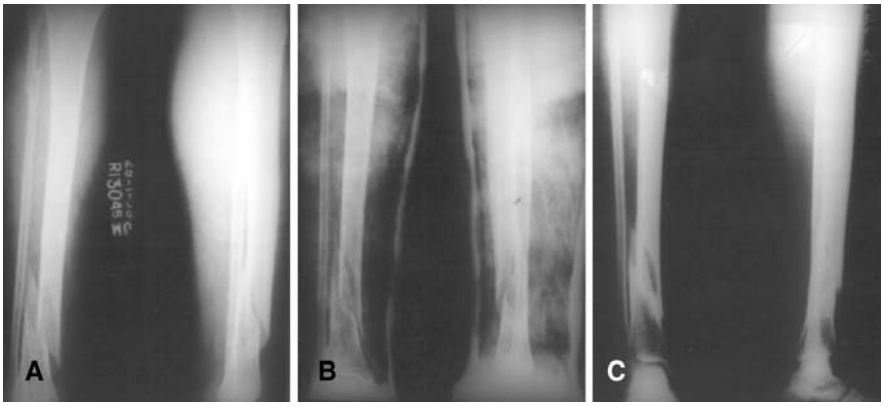

Fig. 1A–C X-ray of both bones, right leg (experimental group). **A** Prereduction X-ray, **B** immediate postreduction X-ray, **C** union at 3 1/2 months

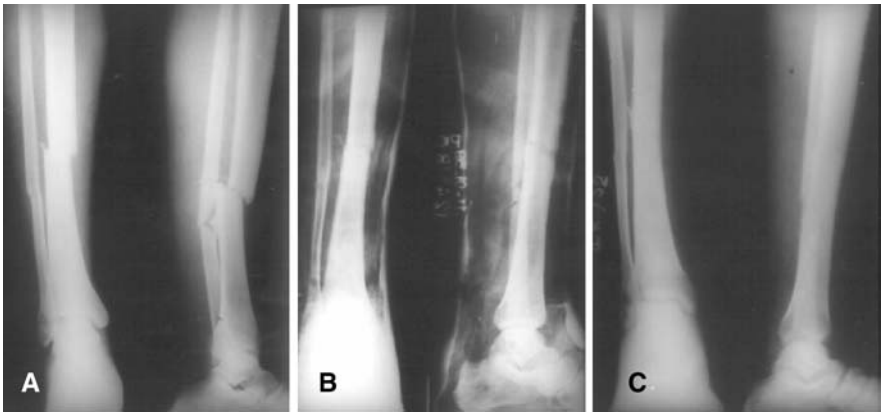

Fig. 2A–C X-ray of both bones, right leg (control group). **A** Prereduction X-ray, **B** immediate postreduction X-ray, **C** union at 4 1/2 months

treatment was low in both groups as this hospital provides government-subsidized health care facilities.

Discussion

McGaw and Harbin, in 1934, were among the first to demonstrate the osteogenic activity of bone marrow [9]. They grafted bone defects in dog fibulae with bone marrow and compared this with contralateral ungrafted defects. Only the bone-marrow-grafted defects filled the gap with bone. Since then, there has been much active interest in the osteogenicity of marrow. Various other experimental studies in rabbits have also successfully demonstrated the osteogenic property of bone marrow in fresh fractures [11, 12].

Composite grafts of autologous bone marrow and allogenic demineralized bone matrix have shown better union rates as compared to either bone marrow or bone matrix alone in large bone defects in experimental animals [13, 14, 15]. Demineralized bone matrix has osteoinductive (bone morphogenetic protein) and osteoconductive properties, and the transplanted marrow cells provide the osteogenic precursor cells [10].

Excellent clinical results have been reported using percutaneous autologous bone marrow grafting along with external immobilization in the treatment of delayed union and even established nonunion of tibial fractures. Connolly demonstrated union in 18 of 20 cases of delayed union and nonunion of tibial fractures after 3–6 months of 100–150 ml of autologous bone marrow injection along with external immobilization [2].

Garg et al. obtained union in 17 out of 20 cases of ununited tibial fractures at 5 months of 15–20 ml of autologous bone marrow injection twice at the nonunion site within an interval of 3 weeks, in combination with immobilization in plaster [4].

The osteogenicity of bone marrow has been traced to bone marrow stromal and endosteal cells. Two types of osteoprogenitor cells (OPC) have been demonstrated, one induced to produce bone OPC (IOPC) and the other that has been determined to produce bone OPC (DOPC). The former (IOPC) exists in all connective tissues and is thought to be an undifferentiated mesenchymal cell. The latter (DOPC) is found only in marrow and is already differentiated into a bone-producing line. Inducible osteoprogenitor cells respond by producing bone to local stimuli (eg., fracture, bone graft, etc.), i.e., osteoinductive stimuli. Because bone marrow is the only tissue that contains an abundance of both DOPC and IOPC, it is considered to be a logical graft choice [11]. Thus, autologous bone marrow injection is a simple method of providing cellular reinforcement to the fracture site and is expected to enhance the probability of union without associated donor or recipient site morbidity [3].

This is the first clinical randomized controlled trial that goes on to prove this experimentally demonstrable osteogenicity of bone marrow injection in fresh fractures of the human tibia. In the present study, the mean union time in the experimental group (3.65±0.49 months) was significantly less than in the control group (4.31±0.48 months) with a p value of 0.0004, thus validating the high osteogenic potential of autologous bone marrow.

Since the nonunion rates in the control group were relatively low, a very large sample size will be required to detect the difference in nonunion rates among the groups. There was no significant difference in complication rates and cost incurred in the two groups.

The technique of autologous bone marrow grafting along with external immobilization can significantly affect union rate in closed, fresh, long bone, conservatively treatable fractures. It is safe, easy, practical, economical, and involves minimal trauma. It is recommended as a useful adjunct to conservative treatment of fresh, closed, long-bone fractures in clinical practice, especially where osteogenic potential of the callus needs to be supported. The technique could be particularly useful in third world countries and other areas where expensive agents such as bone morphogenetic protein or bone stimulatory devices are not available.

References

1. Apley AG, Solomon L (1993) Principles of fracture. In: Apley's system of orthopaedics and fractures, 7th edn,, Butterworth Heinemann, Newton, MA, pp. 519–521
2. Connoly JF (1995) Injectable bone marrow preparation to stimulate osteogenic repair. Clin Orthop 313:8–18
3. Conolly JF, Guse R, Tiedeman J (1991) Autologous marrow injection as a substitute for operative grafting of tibial nonunions. Clin Orthop 266:259–269
4. Garg NK, Gaur S, Sharma S (1993) Percutaneous autogenous bone marrow grafting in 20 cases of ununited fracture. Acta Orthop Scand 64:671–672
5. Govender S, Csimma C, Valentin-Opran A, Genant HK (2002) Recombinant human bone morphogenetic protein-2 for treatment of open tibial fractures. J. Bone Joint Surg [Am] 84:2123–2134
6. Harding RAJ, David HR (1985) Fractures—general principles. In: Bailey and Love (eds), Short practice of surgery 9th edn, English Language Book Society, pp. 220–222
7. Heckman JD, Ryaby JP, McCabe J, Frey JJ, Kilcoyne RF (1994) Acceleration of tibial fracture-healing by non-invasive, low-intensity pulsed ultrasound. J. Bone Joint Surg [Am] 76:26–34
8. Joseph AB, Thomas AE, Mark EB, Richard LC (1996) Healing of the musculoskeletal tissue: In: Rockwood and Greens (eds), Fracture in adults, vol 1, 4th edn, Lipincott-Raven, Baltimore, pp. 276–279
9. McGaw WH, Harbin M (1934) The role of bone marrow, an experiment study of bone marrow and endosteal transplants and endosteum in bone regeneration. J. Bone Joint Surg [Am] 16:816–821
10. Nade S (1977) Osteogenesis after bone and bone marrow transplantation II. The initial cellular events following transplantation of decalcified allografts of cancellous bone. Acta Orthop Scand 48:572–576
11. Paley D, Young MC, Wiley AM, Fornasier VL, Jackson RW (1986) Percutaneous bone marrow grafting of fractures and bony defects: an experimental study in rabbits. Clin Orthop 208:300–312
12. Sharma S, Garg NK, Veliath AJ, Subramanian S, Srivastava KK (1992) Percutaneous bone-marrow grafting of osteotomies and bone defects in rabbits. Acta Orthop. Scand. 63:166–169
13. Takagi K, Urist MR (1982) The role of bone marrow in bone morphogenetic protein-induced repair of femoral massive diaphyseal defects. Clin Orthop 171:224–231
14. Tiedemen JJ, Connolly JF, Strates BS, Lippiello L (1991) Treatment of nonunion by percutaneous injection of bone marrow and demineralized bone matrix: an experimental study in dogs. Clin Orthop 268:294–301
15. Wittbjer J, Palmer B, Rohlin M, Thorngren KG (1983) Osteogenetic activity in composite grafts of demineralized compact bone and marrow. Clin Orthop 173:229–238