

Angiographic evaluation of arterial configuration after acute tibial shortening

Zafer Atbasi · Bahtiyar Demiralp · Erden Kilic ·
Ozkan Kose · Mustafa Kurklu · Mustafa Basbozkurt

Received: 9 June 2013 / Accepted: 24 September 2013 / Published online: 5 October 2013
© Springer-Verlag France 2013

Abstract

Introduction Tibial nonunion with bone and soft tissue loss is a challenging orthopedic problem. Acute tibial shortening is a well-defined technique; however, arterial configuration following acute tibial shortening has not been demonstrated by a clinical study.

Materials and methods Sixteen patients with tibial nonunion and one patient with acute tibial fracture accompanied by bone and soft tissue loss were treated by acute shortening followed by compression or relengthening between 2004 and 2010. Circulation was monitored by intraoperative Doppler ultrasound and hallux pulse oximetry. Arterial configuration was examined by digital subtraction angiography at the seventh postoperative day and by CT angiography at the second year. Bone healing and functional results were evaluated according to Paley's classification, and complications were evaluated according to Dahl's classification.

Results Mean amount of bone loss was 3 ± 1.4 cm (range 1–6 cm); mean size of the soft tissue defects was 7×6.8 cm (range 3×3 cm– 10×10 cm). The mean follow-up period was 38 ± 11.3 months (range 24–57 months). The average amount of acute shortening was 5.4 ± 1.6 cm (range 3–8 cm). Average lengthening was

6 ± 1.8 cm (range 4–8 cm), and the mean external fixation index was 1.4 months/cm (range 0.1–3.7 months). There was no detectable change in the arterial configuration of patients with acute shortening up to 4 cm. Minimal arterial bending was observed in patients that 4–6 cm of shortening was performed. Arterial configuration of the patients that 8 cm acute shortening was performed showed increased tortuosity, but the patency was maintained.

Conclusion Acute shortening of tibia in nonunions with soft tissue defects allows for primary closure or reduces the need for grafting and secondary operations. Although the amount of acute shortening depends upon intraoperative assessment with Doppler ultrasound and hallux pulse oximetry, acute compression up to 8 cm can be attained in proximal tibia. More than 4 cm of acute shortening leads to increased tortuosity of major arteries rather than kinking, and this new arterial configuration is maintained for up to 2 years with no problem in circulation.

Keywords Acute tibial shortening · Tibial nonunion · Angiography · Open tibial fracture

Introduction

Tibial nonunion with bone and soft tissue loss is a challenging orthopedic problem. Accompanying osteomyelitis, deformity and limb-length discrepancy may complicate the treatment and increase the risk for loss of the limb [1]. Reconstruction of the bone (both length and alignment), soft tissue closure and eradication of infection should all be accomplished to obtain a functional limb [2]. Acute tibial shortening followed by gradual lengthening of the bone at another level by Ilizarov method is a well-defined technique which provides a complete solution to this set of

Z. Atbasi · B. Demiralp · E. Kilic · M. Kurklu · M. Basbozkurt
Department of Orthopaedics and Traumatology, Gulhane
Military Medical Academy, Ankara, Turkey

O. Kose
Orthopaedics and Traumatology Clinic, Antalya Education
and Research Hospital, Antalya, Turkey

O. Kose (✉)
Kultur mah. Durukent sit. F Blok, D:22 Kepez, Antalya, Turkey
e-mail: drozkankose@hotmail.com

complex problems [3–11]. While acute shortening facilitates the soft tissue coverage and promotes fracture union, distraction compensates the limb-length discrepancy. In addition, the deformity can be simultaneously corrected by Ilizarov method.

Traditionally, up to 3–4 cm acute shortening is recommended as the safe limit for acute tibial shortening without disturbing the circulation of the leg. If additional compression is required, gradual shortening of 2–3 mm per day is recommended [6, 7]. Venous and lymphatic stasis and arteriolar occlusion that may lead to swelling and eventual necrosis are major concerns that impose limits of acute shortening [6–9]. However, no clinical study has yet demonstrated the arterial configuration following acute tibial shortening.

The purpose of this study was to determine the vascular changes by comparing preoperative and postoperative angiographies following acute shortening of tibia in patients with tibial nonunion or acute injury accompanied by substantial bone and soft tissue loss and try to outline a safe limit for acute compression considering the clinical picture of the patient together with intraoperative monitoring of dorsalis pedis and tibial arteries by palpation, Doppler ultrasonography and hallux pulse oximetry.

Materials and methods

Patients

This retrospective study was carried out according to the Declaration of Helsinki, and the local ethics committee approved the study protocol. All patients gave informed consent prior to their inclusion in the study. Sixteen

patients with tibial nonunion and one patient with acute injury accompanied by bone and soft tissue loss were treated in our department by either acute shortening and compression, or acute shortening followed by relengthening at another level by Ilizarov method, between October 2004 and February 2010. Operative technique is shown in Fig. 1. All patients were male with an average age of 26.7 ± 7.6 years. The original mechanism of injury was motor vehicle collision ($n = 5$), land mine injury ($n = 8$) and high-velocity gunshot injury ($n = 4$). Contralateral leg was amputated in five cases. The mean number of previously failed surgical procedures for union and soft tissue reconstruction per patient was 3.2 ± 0.7 (range 1–9). The mean duration of time since the index injury was 11.8 months (range 1–96 months).

Preoperative evaluation and planning

Preoperative full-length radiographs of the affected leg were taken to determine the amount of bone loss and level and type of nonunion. All nonunions were of atrophic type. Defects were located in the proximal third in five patients, in the middle third in six patients and in the distal third of tibia in the remaining six patients, which also included one acutely injured case. Mean amount of bone loss was 3 ± 1.4 cm (range 1–6 cm) at the initial radiographic examination. According to the classification of Paley et al. [1], bone loss was type B1-1 in seven, type B1-2 in five and type B3 in five patients. Mean size of the soft tissue defect was 7×6.8 cm (range 3×3 cm– 10×10 cm) and was typically located on the anteromedial or anterolateral aspects of the tibia. Eleven patients had infection at the time of intervention. Methicillin-resistant *Staphylococcus aureus* was identified as the predominant organism in six

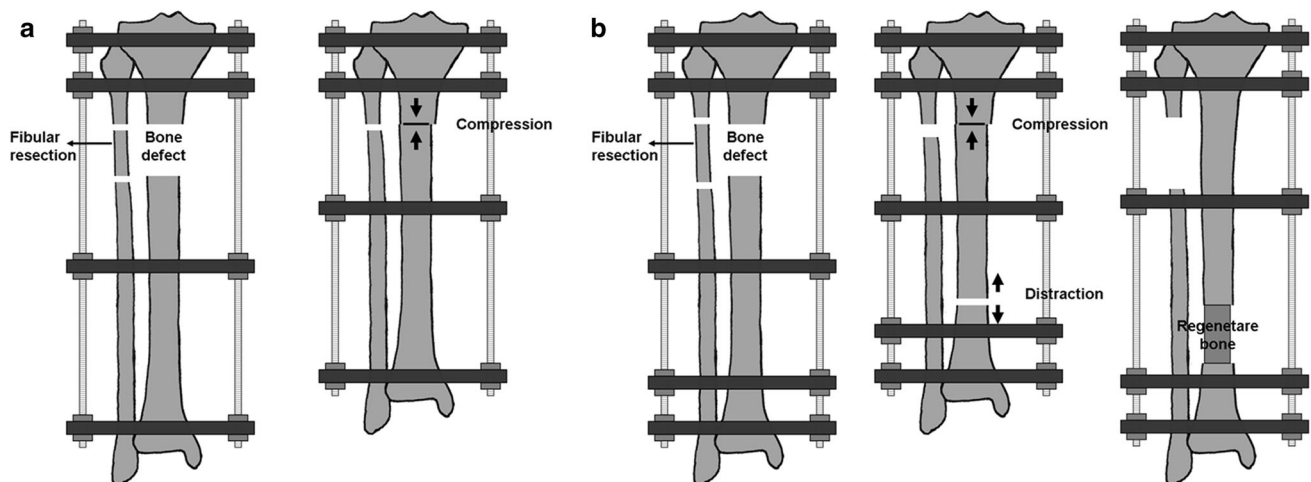


Fig. 1 Illustration showing the operative technique. **a** Acute shortening and compression was utilized in patients with contralateral leg amputation. **b** Acute shortening followed by relengthening at another level

Table 1 Summary of all data

Case	Age	Type of injury	Fracture site	Concomitant injuries	Initial treatment	Gustilo Anderson classification	Infection
1	26	Land mine	Proximal third	Left tibia type B3 open fracture, pelvic fracture, bladder injury	Bilateral tibia EF, pelvis ORIF, free flap for left leg, multiple skin grafts	3B	–
2	23	Land mine	Distal third	Maxillofacial injury, lung contusion, hemothorax	Right tibia EF, 4 weeks in ICU	3B	MRSA
3	28	Land mine	Middle third	Right above-knee amputation, left peroneal nerve injury	Right above-knee amputation, left tibia EF + local flap, nerve repair, skin graft	3B	–
4	31	RTA	Distal third	Head trauma, cerebral edema	3 weeks in ICU, below-knee splint	2	–
5	46	RTA	Middle third		ORIF with plate-screws	3A	Pseudomonas
6	22	Land mine	Proximal third	Left above-knee amputation, left eye enucleated multiple phalangeal fractures in both hands, scrotum laceration	Left above-knee amputation, left eye enucleated, right tibia EF	3B	
7	23	Land mine	Distal third	Left above-knee amputation, scrotum rupture	Left below-knee amputation, right tibia EF, bilateral orchiectomy	3B	MRSA
8	21	Land mine	Middle third	Left above-knee amputation, right thumb amputation, right peroneal nerve lesion	Left above-knee amputation, right tibia EF + fasciotomy, right thumb amputation	3B	
9	31	RTA	Distal third	Cerebral edema, T5 vertebral compression fracture, right clavicle fracture	Left tibia EF, T5 anterior corpectomy + cage	2	
10	40	RTA	Middle third		ECF + 3 cm acute shortening	3B	Pseudomonas
11	35	RTA	Distal third	Right peroneal nerve injury	Right tibia EF + skin graft	3B	MRSA
12	27	Gunshot	Distal third	Injury of anterior tibialis artery	EF + anterior tibial artery repair	3C	Serratia
13	21	Land mine	Proximal third		ECF	3B	Serratia
14	20	Gunshot	Proximal third	Right distal radius fracture, L4 vertebral compression fracture	Left tibia unilateral EF, long-arm cast for right wrist, brace for vertebral fracture	3B	MRSA
15	20	Gunshot	Proximal third	Ischial fracture, L2-3 compression fracture	Left tibia unilateral EF, brace for vertebral fracture	3B	MRSA
16	21	Land mine	Middle third	Left below-knee amputation, right knee multiple ligament injury	Right tibia unilateral EF, left below-knee amputation	3B	Serratia
17	20	Land mine	Middle third	Right below-knee amputation, coccygeal fracture, left pubic fracture	Left tibia unilateral EF, right below-knee amputation	3B	MRSA

Case	Classification for defective pseudarthrosis	Bone defect (cm)	Soft tissue defect (cm)	Bone debridement (cm)	Acute shortening (cm)	Distraction (cm)	Duration of treatment (months)	Bone results	Functional results	Complication
1	B1-1	3	7 × 7	3	6	*	4	Excellent	Excellent	
2	B1-1	2	3 × 3	2	4	**	3	Good	Excellent	
3	B1-1	2	–	4	6	***	4	Poor	Excellent	Refracture
4	B1-1	1	–	3	4	4	8	Excellent	Excellent	
5	B3	2	10 × 5	3	5	5	10	Excellent	Excellent	
6	B1-1	6	10 × 10	2	8	***	7	Excellent	Excellent	

Table 1 Summary of all data

Case	Classification for defective pseudarthrosis	Bone defect (cm)	Soft tissue defect (cm)	Bone debridement (cm)	Acute shortening (cm)	Distraction (cm)	Duration of treatment (months)	Bone results	Functional results	Complication
7	B1-2	2	3 × 3	3	5	5	4	Excellent	Excellent	
8	B1-1	2	–	2	2	***	11	Excellent	Excellent	
9	B1-1	2	–	1	3	**	4	Excellent	Excellent	
10	B3	2	5 × 5	3	5	8	12	Excellent	Good	
11	B3	2	5 × 8	3	5	7	8	Excellent	Excellent	
12	B1-2	4	10 × 10	4	5	–	3	Poor	Fair	Amputation
13	B1-2	5	10 × 8	3	8	**	8	Good	Good	
14	B3	4	8 × 5	2	6	4	10	Good	Good	Pin tract infection
15	B1-2	3.5	5.5 × 6	3.5	7	5	9.5	Poor	Fair	Pin tract infection
16	B1-2	5	9 × 4.5	2	7	***	6.5	Excellent	Fair	
17	B3	4.5	8 × 3	2	6.5	***	6	Excellent	Good	

EF external fixator, ORIF open reduction internal fixation, ICU intensive care unit, MRSA methicillin-resistant *Staphylococcus aureus*

* Only 6 cm of acute shortening was performed due to 16 cm defect in the contralateral leg

** Distraction was delayed to secondary surgery (see text)

*** Contralateral extremity is amputated

patients, *Serratia marcescens* was identified in three patients, and *Pseudomonas aeruginosa* was identified in two patients. Remaining six patients had no infection at the time of surgery. Patients with accompanying osteomyelitis were evaluated with Indium-111 leukocyte scintigraphy to determine the affected bone segment. Arterial configuration was documented by digital subtraction angiography (DSA) before acute shortening in all patients. Distraction was planned for eight patients; the details of the remaining nine patients that distraction was not planned are presented in Table 1.

Surgical technique and follow-up

One gram of first-generation cephalosporin was administered 1 h before surgery for antimicrobial prophylaxis. Tourniquet was not used in order to check the circulation throughout the surgery. Transverse or Z-shaped incisions were used at the level of soft tissue defects. Scar tissue and fistula were excised if present. Infected and necrotic bone was completely resected as directed by scintigraphic involvement and clinical evaluation of viability during surgery, which was followed by aggressive soft tissue debridement and copious irrigation. The average amount of bone debrided during surgery was 2.6 ± 0.8 cm (range 1–4 cm). Adequate length of fibula was resected to allow compression of proximal and distal tibial fragments. Pre-operatively constructed Ilizarov frame was applied under fluoroscopic control for alignment. During surgery, dorsalis

pedis and tibialis posterior arteries were monitored by a portable Doppler USG together with oxygen saturation at the hallux with pulse oximetry. If distraction was planned, an osteotomy was performed by a Gigli wire depending on the defect level—at the proximal metaphyseal level in six patients with defects at the distal tibia and at the distal metaphyseal level in two patients with defects at the proximal tibia. All soft tissue defects were closed primarily, only two patients required split-thickness grafts (Figs. 2 and 3). Distal circulation was checked every 2 h for the first 24 h and every 6 h for the next 3 days after operation with a portable Doppler USG. DSA was performed at the seventh postoperative day to visualize the arterial configuration of the leg and to determine the early postoperative changes, if present.

Distraction was initiated at the seventh day, on a schedule of 0.25 mm every 6 h a day (1 mm/day in total). Full weight bearing was allowed for patients that only shortening was planned, and distracted patients were encouraged to partially bear weight with crutches after the third postoperative day until the end of the distraction period, followed by full weight bearing. Follow-up radiographs were taken monthly during the frame time and at 6-month intervals after the removal of the frame. At the postoperative second year, all patients underwent CT angiography to assess the new arterial configuration. Bone healing and functional results were evaluated according to the criteria by Paley et al. [1]. Complications were evaluated according to the classification by Dahl et al. [12].

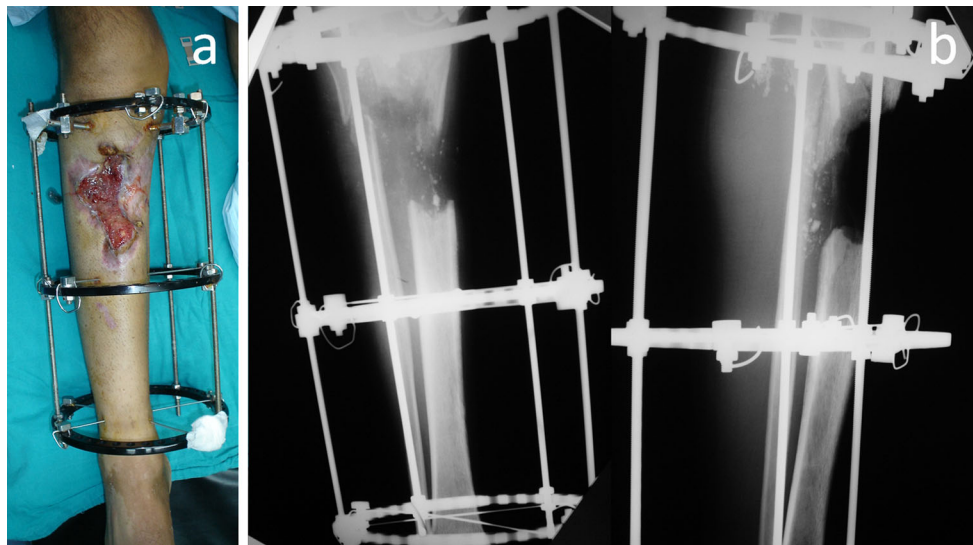
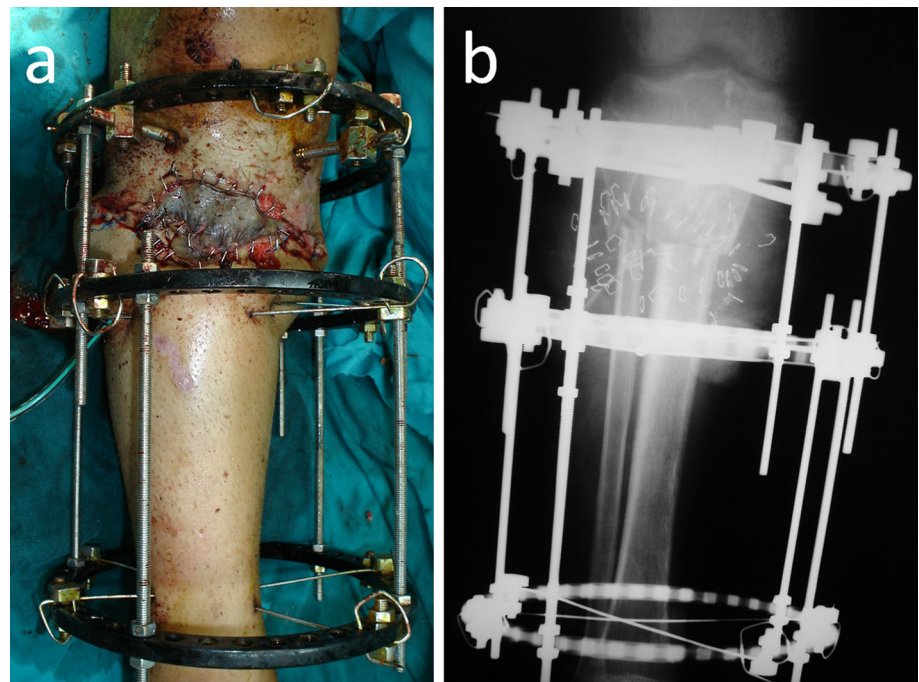


Fig. 2 Clinical appearance of a patient with anterior soft tissue loss (a) and the preoperative radiographs b showing the underlying bone defect

Fig. 3 Intraoperative appearance of the same patient in Fig. 2 soon after debridement and acute tibial shortening (a) and early postoperative radiograph (b)



Results

The mean follow-up period was 38 ± 11.3 months (range 24–57 months). The average amount of acute shortening (to compensate initial bone loss plus resection at surgery) was 5.4 ± 1.6 cm (range 3–8 cm). Total bone loss could be compressed in 16 of 17 patients. In one patient, dorsalis pedis flow ceased after 6 cm of acute shortening; thus, shortening was reduced to 5 cm, and the remaining 1 cm was shortened gradually (2 mm/day). Average lengthening was 6 ± 1.8 cm (range 4–8 cm), and the mean external fixation index (EFI) was 1.4 months/cm (range 0.1–3.7

months). Mean duration of treatment was 6.9 ± 2.9 months (range 3–18 months). Complete union was obtained in all patients without the need for further bone grafting. Limb length was equalized in six of the eight distracted cases; 2 cm of limb-length discrepancy remained in one patient in which distraction was terminated due to severe pain. The other patient underwent amputation before distraction was started. No recurrence of infection was observed during follow-up. We obtained eleven excellent, three good and three poor results in terms of bone assessment. The functional results were excellent in ten patients, good in four and fair in three. Soft tissue healing was

excellent in all patients with no dystrophic changes, hypersensitivity or insensitivity. No patient had more than 7 degrees of valgus or varus deformity, or more than 5 degrees of recurvatum deformity.

Vascular changes

Distal circulation was confirmed to be patent in all patients by DSA performed at the postoperative seventh day. There was no detectable change in the arterial configuration of patients with acute shortening up to 4 cm. Minimal arterial bending was observed in patients that 4–6 cm of shortening was performed. Arterial configuration of the patients that 8 cm acute shortening was performed showed increased tortuosity, but the patency was maintained. Comparison of the images taken before and immediately after the procedure and 2 years after distraction has demonstrated the capability of arteries to adapt to the new length of the extremity. In contrary to what previously proposed, vascular adaptation occurred in the form of increased tortuosity rather than kinking. New configurations seemed to be maintained at least for 2 years (Figs. 4, 5 and 6).

Complications

Refracture occurred in one patient 2 months after the removal of the frame. One patient with acute injury that primary arterial repair was performed (grade IIIC)

underwent a below-knee amputation at the sixth postoperative day. Pin tract infection (grade II) developed in three patients. In one patient that had severe osteomyelitis and large soft tissue defect, limb-length discrepancy of more than 5 cm remained because distraction was not planned due to high amputation risk. Three other patients had multiple injuries which required multiple consecutive operations; thus, they did not consent to distraction.

Discussion

Acute tibial shortening followed by distraction is an effective method in the treatment of tibial nonunions with significant bone and soft tissue loss. However, safe limit of tibial shortening has not been clearly stated in the literature; most recommendations are based on author's personal experiences. In their study on dogs, Rahal et al. [13] shortened the tibia and fibula by 30 % and observed the circulation and edema of the extremity and reported that arterial kinking due to shortening did not lead to tissue necrosis in any of the dogs. Sen et al. [6] successfully performed acute shortening at the fracture site of bone defects up to 3 cm to oppose the fragments in grade III open tibia fractures with bone and soft tissue loss. They recommended gradual shortening at a rate of 2 mm/day for patients with bone defects more than 3 cm. Similarly, Saleh and Rees suggested acute shortening for tibial

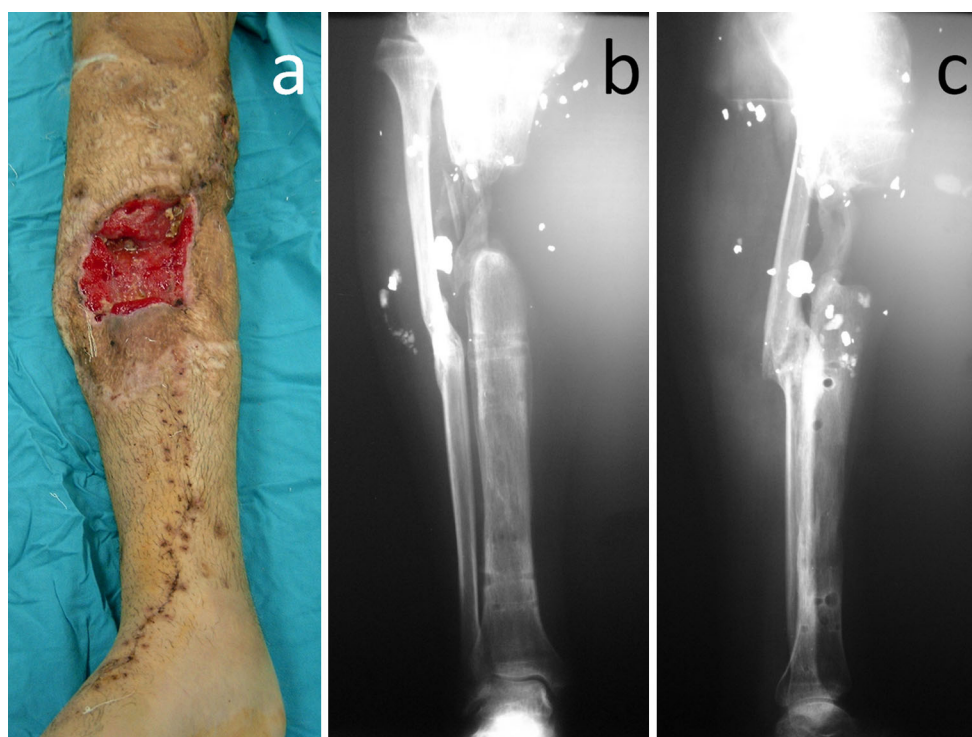


Fig. 4 Clinical appearance (a) and the radiographs (b, c) of the patient with bone and soft tissue loss

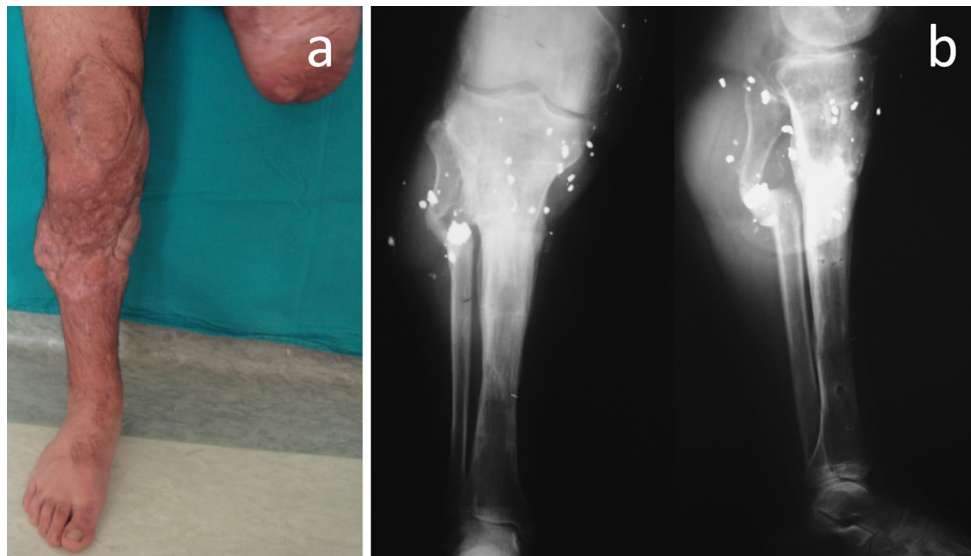


Fig. 5 Final clinical appearance (a) and the radiographs (b) of the patient in Fig. 4

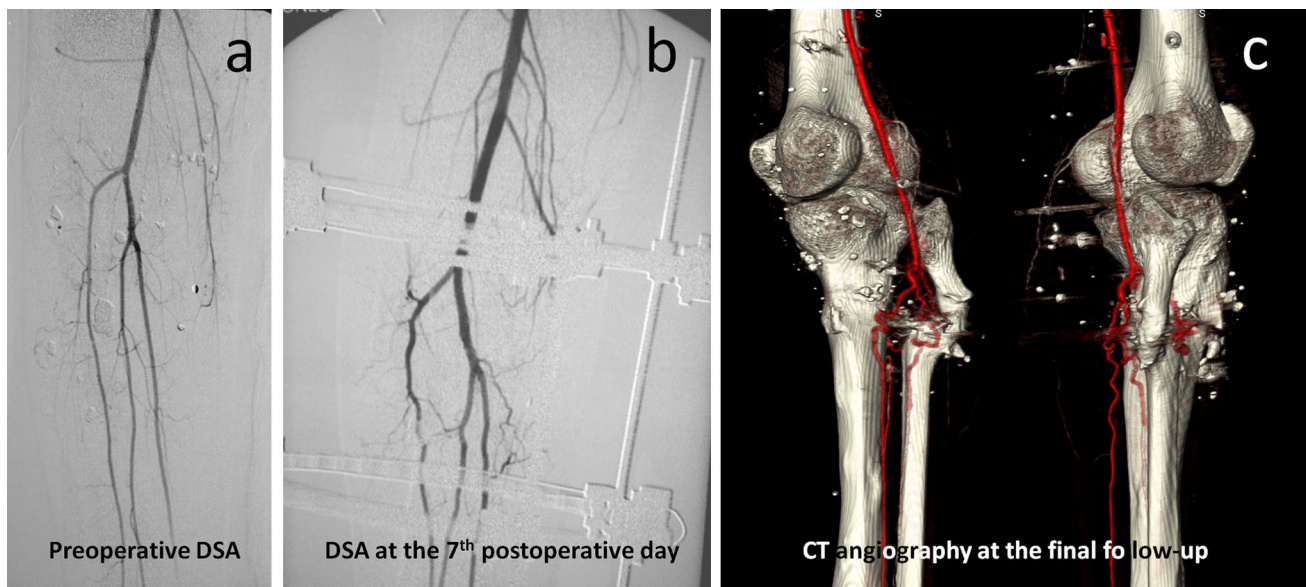


Fig. 6 Preoperative (a), postoperative seventh day (b) and final follow-up angiography (c) of the same patient. Note increased tortuosity of the arteries after 8 cm of acute shortening

defects less than 3 cm [8]. In another study, Sen et al. [7] were able to perform acute shortening for tibial nonunions with bone loss up to 4 cm, if tibia had normal alignment with no rotational deformity. Rozbruch and S. Ilizarov also stated that acute shortening of defects up to 3–4 cm could be safely accomplished; however, over 4 cm, the risk for the development of tortuous vasculature increases, which may lead to a low flow state with detrimental consequences [14]. As already been stated, these cutoff limits have been randomly set and do not depend on specific findings.

There are studies in the literature that claim acute tibial shortening more than 4 cm can be safely performed. In

their study comparing the results of acute shortening and lengthening to those of bone transport in the management of tibial nonunions of 18 patients, Mahaluxmivala et al. [4] were able to shorten the tibia 4–6 cm in five cases with no vascular compromise; however, no information regarding the levels of nonunions was presented. El-Rosasy et al. [10] treated 21 patients with tibial fractures and nonunions complicated with bone and soft tissue loss. They proposed considering the level of the defect to determine the amount of acute shortening: Due to vascular compromise and tissue buckling, they recommended up to 3 cm shortening for upper third of the leg, 3–5 cm for middle third and less

than 6 cm for the lower third. Yokoyama et al. [15] used acute shortening as a salvage procedure in six patients with type B3 open tibial acute fractures and performed shortening up to 9.5 cm at the proximal and 10.3 cm shortening at the midlevel of tibia with no vascular complication. However, they did not mention about the decision-making process to determine the amount of shortening or the method they used to check the circulatory status. Up to our knowledge, the figures presented by Yokoyama et al. are the maximum amounts of acute shortening reported in the literature. The authors suggested acute shortening for tibial defects more than 4.5 cm, but noted that acute shortening exceeding 25 % of the total length of bone yielded poor functional results. Generally speaking, bone and functional results of our cases after acute shortening seemed to be satisfactory. Particularly, results were in good to excellent range in patients with amputation of contralateral leg, which may be attributed to shorter treatment period due to the lack of distraction phase. In contrast to previous reports, level and amount of acute shortening did not seem to affect the results. This may be due to the amount of acute shortening, which was <25 % of the total bone length. In our series, we achieved an average of 7.3 cm shortening for the upper third of tibia, 5 cm for the middle and 4 cm for the distal third. Our results together with the figures reported by Yokoyama et al. suggest that more acute shortening than El-Rosasy reported can be attained at the proximal and middle third of tibia. However, we agree that there is no absolute safe limit for every patient, and decisions should be based on intraoperative monitoring of circulation with Doppler USG and hallux pulse oximetry. Close follow-up of the circulation is mandatory, and in any suspicion of vascular compromise, amount of shortening can be reduced in the postoperative period, which is also possible with Ilizarov frame.

We performed acute tibial shortening over 6 cm in eight cases with no vascular compromise and evaluated the vascular structure and patency by DSA angiographies performed at the seventh postoperative day. After 2 years, CT angiographies of patients that had acute shortening of 4 cm or less did not show a significant structural change compared to preoperative DSA angiographies, but in two patients to whom 8 cm of acute shortening was performed (case 6 and 13), posterior tibial and anterior tibial arteries showed increased tortuosity rather than kinking. In our study, lack of circulatory problem with acute shortening up to 8 cm may be due to young age of our patients, and lower safe limits may be expected from older patients due to increased fragility of arteries. As one of our patients required amputation after acute shortening, previous primary arterial repair (grade IIIc open fracture) should also be considered during decision-making.

Our study has some limitations. A limited number of patients were included; however, this is a very specific type of nonunion, and advances in fracture management and microsurgery decreased the number of such patients. We have used subjective assessment for arterial configuration, as we were unable to determine an established way to assess the structural changes in the vasculature.

In conclusion, acute shortening of tibia in nonunions with soft tissue defects allows for primary closure or reduces the need for grafting and secondary operations. Acute appositioning of the fracture ends promotes bone healing and helps to fight infection as residual space is minimized. Acute shortening also reduces the duration of the treatment and minimizes complications, thus increasing patient satisfaction. Based on our experience, the limit of acute shortening can be increased until distal circulation is impeded as documented by Doppler ultrasound and hallux pulse oximetry. Although the amount of acute shortening depends upon intraoperative assessment, acute compression up to 8 cm can be attained in proximal tibia. More than 4 cm of acute tibial shortening leads to increased tortuosity of major arteries rather than kinking. This new arterial configuration is maintained for up to 2 years with no problem in circulation.

Conflict of interest Authors have no conflict of interest to disclose.

References

1. Paley D, Catagni MA, Argnani F et al (1989) Ilizarov treatment of tibial nonunions with bone loss. *Clin Orthop* 241:146–165
2. Naggar L, Chevalley F, Blanc C et al (1993) Treatment of large bone defects with the Ilizarov technique. *J Trauma* 34:390–393
3. Giebel G (1991) Primary shortening in soft tissue defects with subsequent callotaxis in the tibia. *Unfallchirurg* 94:401–408
4. Mahaluxmivala J, Nadarajah R, Allen PW, Hill RA (2005) Ilizarov external fixator: acute shortening and lengthening versus bone transport in the management of tibial non-unions. *Injury* 36:662–668
5. Meffert RH, Inoue N, Tis JE, Brug E, Chao EY (2000) Distraction osteogenesis after acute limb-shortening for segmental tibial defects. Comparison of a monofocal and a bifocal technique in rabbits. *J Bone Joint Surg Am* 82:799–808
6. Sen C, Kocaoglu M, Eralp L, Gulsen M, Cinar M (2004) Bifocal compression-distraction in the acute treatment of grade III open tibia fractures with bone and soft-tissue loss: a report of 24 cases. *J Orthop Trauma* 18:150–157
7. Sen C, Eralp L, Gunes T, Erdem M, Ozden VE, Kocaoglu M (2006) An alternative method for the treatment of nonunion of the tibia with bone loss. *J Bone Joint Surg Br* 88:783–789
8. Saleh M, Rees A (1995) Bifocal surgery for deformity and bone loss after lower limb fractures. *J Bone Joint Surg* 77:429–434
9. Sales de Gauzy J, Vidal H, Cahuzac JP (1993) Primary shortening followed by callus distraction for the treatment of a posttraumatic bone defect: case report. *J Trauma* 34:461–463
10. El-Rosasy MA (2007) Acute shortening and re-lengthening in the management of bone and soft-tissue loss in complicated fractures of the tibia. *J Bone Joint Surg Br* 89:80–88

11. Lerner A, Fodor L, Soudry M et al (2004) Acute shortening: modular treatment modality for severe combined bone and soft tissue loss of the extremities. *J Trauma* 57:603–608
12. Dahl MT, Gulli B, Berg T (1994) Complication of limb lengthening. A learning curve. *Clin Orthop* 301:10–18
13. Rahal SC, Volpi RS, Vulcano LC (2005) Treatment of segmental tibial defects using acute bone shortening followed by gradual lengthening with circular external fixator. *J Vet Med A Physiol Pathol Clin Med* 52:180–185
14. Rozburch SR, Ilizarov S (2007) Limb lengthening and reconstruction surgery. Informa Healthcare, New York
15. Yokoyama K, Itoman M, Nakamura K, Uchino M, Tsukamoto T, Suzuki T et al (2006) Primary shortening with secondary limb lengthening for Gustilo IIIB open tibial fractures: a report of six cases. *J Trauma* 61:172–180