Ten Year Experience of Using Cryopreserved Arterial Allografts for Distal Bypass in Critical Limb Ischaemia

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WHAT THIS STUDY ADDS
Recent long-term results of arterial allografts used for distal bypasses in the setting of chronic ischaemia are lacking. This study confirms that the use of modern cryopreserved arterial allografts allows for an excellent limb salvage rate, at the cost of a relatively high re-intervention rate. This material should be considered as a valuable alternative to prosthetic material when no venous material is available.

Objective/Background: In critical limb ischaemia (CLI), current guidelines recommend revascularisation whenever possible, preferentially through endovascular means. However, in the case of long occlusions or failed endovascular attempts, distal bypasses still have a place. Single segment great saphenous vein (GSV), which provides the best conduit, is often not available and currently there is no consensus about the best alternative graft.
Methods: From January 2006 to December 2015, 42 cryopreserved arterial allografts were used for a distal bypass. Autologous GSVs or alternative autologous conduits were unavailable for all patients. The patients were observed for survival, limb salvage, and allograft patency. The results were analysed with Kaplan—Meier graphs.
Results: Estimates of secondary patency at one, two and five years were 81%, 73%, and 57%, respectively. Estimates of primary patency rates at one, two and five years were 60%, 56%, and 26%, respectively. Estimates of limb salvage rates at one, two and five years were 89%, 89%, and 82%, respectively. Estimates of survival rates at one, two and five years were 92%, 76% and 34%, respectively. At 30 days, major amputations and major adverse cardiac events were one and zero, respectively. Six major amputations occurred during the long-term follow up.
Conclusion: Despite a low primary patency rate at two years, the secondary patency of arterial allografts is acceptable for distal bypasses. This suggests that cryopreserved arterial allografts are a suitable alternative for limb saving distal bypasses in the absence of venous conduits, improving limb salvage rates and, possibly, quality of life.

INTRODUCTION

Peripheral artery disease has now become a concerning global health issue, affecting 5—18% of the population, and continues to grow, especially in the ageing population.[1,2] According to the European Society of Vascular Surgery guidelines, revascularisation should be attempted whenever possible in patients suffering from critical limb ischaemia (CLI).[3] Although the endovascular approach has gained popularity over the last 10 years and is now considered the technique of choice for distal revascularisation, some debate remains and bypass still has a place. In the

multicentre randomised Bypass vs. Angioplasty in Severe Ischaemia of the Leg (BASIL) trial, endovascular procedures and open bypass surgery grafts demonstrated similar one year limb salvage rates in CLI, ranging from 85% to 90%.[4] For below knee disease, the European Society of Cardiology guidelines suggest an individual approach, with bypass first strategies for long occlusions in patients fit for surgery or after endovascular failure.[3] Although some controversy remains concerning the expected outcome of bypass surgery after failed endovascular attempts,[5] there is increasing evidence showing that this strategy does not affect bypass patency, limb salvage, or patient survival.[6]

The great saphenous vein (GSV) is the conduit of choice for infra-inguinal bypass graft procedures.[7] However, owing to prior limb or coronary bypasses, varicosities, or inadequate vein diameter, 15—40% of patients have no usable GSV.[8] In those cases, other venous alternatives should be

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considered, such as arm veins, lesser saphenous vein, composite conduits, or allogenic cryopreserved venous material.[9–13] The use of prosthetic material is known to be particularly unsuccessful, especially for infrageniculate reconstruction, with a five year secondary patency of only 40% and poor limb salvage rate.[14] Moreover, this material carries a high infection risk, especially in patients with leg ulcers and/or gangrene.[15] A recent Cochrane review reported no difference in the primary or secondary patency between expanded polytetrafluoroethylene (ePTFE) and Dacron grafts.[16] One study compared ePTFE alone with ePTFE with vein cuff; owing to low quality evidence, any effect or lack thereof on either primary or secondary patency could not be demonstrated.[17]

Since the first use of arterial allografts at the beginning of the twentieth century,[18] the technique of harvesting and conservation has improved. Nowadays, cryopreserved arterial allografts have shown to be a viable option in specific circumstances, such as peripheral bypass infection, with up to 75% limb salvage at five years.[19] However, there is a notable lack of studies in the literature that evaluate the long-term results of this material for distal bypass in the setting of critical ischaemia. Only a few and relatively old articles have reported arterial allograft patency rates, with varying results. The reported secondary patency varies between 74% at 17 months and 25% at five years, according to Albertini *et al.* and Alonso *et al.*, respectively.[10,11] The aim of this study is to report a 10 year experience of distal bypass using arterial cryopreserved allografts.

MATERIALS AND METHODS

Study design and population

In this retrospective observational cohort study, the medical records of all patients who underwent a distal bypass at the Department of Vascular Surgery, CHUV, Lausanne, Switzerland from January 2006 to December 2015, were reviewed. During this period, 269 distal bypasses were performed. Inclusion criteria for the study were bypass with the distal anastomosis performed to the below knee popliteal artery or more distally with one or more cryopreserved arterial allografts, in patients with CLI or acute ischaemia. The data had been prospectively stored in a database since 2013 (Secutrial; interActive Systems, Berlin, Germany) and retrospectively reviewed. Personal medical data, such as age, comorbidities, American Society of Anaesthesiologists' score, and operative indication were collected for each patient. The following comorbidities were also retrieved: diabetes (fasting glycaemia $\geq$7 mmol/L, or random glycaemia $\geq$11.1 mmol/L, or glycated haemoglobin $\geq$6.5%), cardiac disease (atrial fibrillation or valvulopathy), ischaemic heart disease (defined as a stenosis >50% in $\geq$1 coronary artery(ies) and/or positive stress tests and/or angina and/or myocardial infarction [MI]), cerebrovascular disease (defined as a stenosis >50% of a carotid artery, and/or presence of transient ischaemic attack or stroke), hypertension (arterial pressure >140/90 requiring antihypertensive therapy), dyslipidaemia (low density lipoprotein

cholesterol $\geq$2.6 mmol/L or triglycerides $\geq$5.0 mmol/L), chronic renal failure (defined as glomerular filtration rate <60 mL/min/1.73 m^2), obesity (body mass index >29), and active or previous smoking habits. Operative data such as duration of operation, site of each anastomosis, number of homografts, and length were also collected.

The study protocol was in accordance with the Declaration of Helsinki[20] and approved by the local ethics committee (ref. number 2016–02017).

Surgical strategy, allografts, and surgical procedure

At CHUV, the general strategy for treating patients suffering from below knee arterial disease is increasingly focused on endovascular techniques. However, the final decision is based on the extent of the arterial lesions, the patient's comorbidities and preferences, the surgeon's skills and preferences, and the emergency. When the decision to perform a bypass has been taken, the material of choice is always the vein, with the GSV as the first choice, followed by the small saphenous vein and arm veins. Venous mapping is always performed before such an operation. When no veins are available, the next choice is cryopreserved allografts, if the intervention is planned, or other prosthetic material, generally ePTFE, if the intervention is done in an emergency setting.

Cadaveric arterial homografts used in Switzerland are a product of the European Homograft Bank (EHB) in Brussels, obtained from heart beating or non-heart beating donors. Recovered arteries are transported to the EHB and cryopreserved. They can then be ordered by surgeons for implantation.[21] In Switzerland, the delay for obtaining an allograft varies from 24 to 72 h.

The allografts were brought to the operating room the day of the intervention in liquid nitrogen at −150 °C. In the operating room, the arterial allografts were defrosted following the official EHB protocol. The first surgical team checked the allografts and all the collaterals, and any holes were sewn using 6.0 polypropylene sutures. When more than a single allograft was required, they were sutured together to obtain the final bypass length. Simultaneously, a second surgical team prepared the operating sites. Once these steps were completed and the arteries were ready to be clamped, unfractionated heparin 50 IU/kg was administered intravenously. According to the authors' experience, the administration of a bolus of 50 UI/kg of heparin before arterial clamping allows a therapeutic activated partial thromboplastin time to be reached in most patients. After the operation, a prophylactic dose of intravenous heparin or low molecular weight heparin was administered continuously until the patient was discharged.

Post-operative parameters

Complications were distinguished as (i) post-operative complications, representing all complications occurring in the 30 days after the intervention; and (ii) long-term complications, occurring after 30 days. Complications included stenosis (involving >50% of the vessel lumen) or occlusion of the

bypass, and bleeding or wound complication (dehiscence necessitating surgical repair). The post-operative mortality rate was also analysed. In terms of anticoagulation, the individualised decision for starting long-term oral anticoagulation after a distal bypass mainly depends on the quality of the outflow and on each surgeon's preference. Usually, oral anticoagulation is introduced when only one runoff vessel is patent distal to the bypass.

The follow up program consisted of radiological examination (Doppler ultrasound of the lower limbs) and routine radiological monitoring with Doppler ultrasound at seven days, one, six and 12 months, and yearly thereafter. The results of radiological monitoring are represented by plethysmography. A plethysmography value is the measure of the arterial pressure of the toe. It is considered normal when it exceeds 50 mmHg.

For long-term complications, various treatments such as angioplasty and/or stenting, thrombolysis, surgical thrombectomy, new bypass, and minor/major amputation were recorded. The long-term mortality rate was also reported. The date of the end of the follow up was 30 August 2018. The completeness of follow up was defined as the Follow up Index (FUI), as described by von Allmen et al.[22]

Definitions

CLI was defined as ischaemic rest pain or tissue loss combined with an ankle pressure <50 mmHg (or toe pressure <30 mmHg) for >2 weeks, according to the TASC II definition. Primary patency was defined as the time between the creation of the bypass and the first event, such as stenosis or occlusion, i.e., the event free time following the operation. Secondary patency is the time between the creation of the bypass and its definitive occlusion. Limb salvage was defined as the absence of major amputation (above the ankle). Major adverse cardiac event (MACE) is a cardiac event such as a MI.

Outcomes

The primary outcome was the long-term secondary patency rate. Secondary outcomes were primary patency, limb salvage, and survival rates, as well as complications rate.

Statistical analysis

Continuous variables are given as the mean or median and range, and qualitative variables as absolute numbers and percentages. The mean value was used when the distribution was normal and median when it was skewed. Kaplan–Meier graphs were performed to obtain limb salvage and patency rates using GraphPad Prism (version 7.0d for Mac; GraphPad Software, La Jolla CA, USA).

RESULTS

Population

During the study period, from 2006 until 2015, 42 bypasses with one or more allografts were performed in 38 patients (14 men, 24 women), representing 17% of the total number of distal bypasses performed at CHUV. Study population characteristics are summarised in Table 1. The integrative database has been in place since 2013. From 2013 until the end of 2015, 178 infrainguinal bypasses were created, of which 13 were composite. In 126 cases, the GSV (66%) was used (of which nine were composite: five with an arm vein and four with ePTFE). In 26 (14.6%), arm veins were used (of which six were composite: five with GSV, one with ePTFE). In 19 (10.6%), arterial allografts were used (of which two were composite: one with ePTFE and one with Omniflow). In 11 (6.2%), Omniflow II bioprostheses were used (of which two were composite: one with arterial allograft and one with ePTFE) and in nine (5%), ePTFE prostheses were used (of which seven were composite: four with GSV, one with Omniflow, one with arterial allograft, one with arm vein).

Operative indications for distal bypasses were Rutherford stage 4 in 11 patients (26%), stages 5 and 6 in 24 patients (57%), and acute limb ischaemia in seven patients (17%).

Operative parameters

Of the 42 bypasses, the proximal anastomosis was performed to the femoral artery in 41 cases (98%): 27 to the common femoral artery (64%) and 14 to the superficial or deep femoral artery (32%; $n = 42$). One proximal anastomosis was on the external iliac artery. The distal anastomosis was performed to crural arteries in 21 cases (50%), to the tibioperoneal trunk in nine cases (21%), and to the distal popliteal artery in 12 cases (28%). The mean bypass length was 49 cm (range 35–68 cm). Either one (in 12%), two (in 76%), or three (in 12%) homografts per bypass were used. In total, 84 homografts were used to create the 42 bypasses. Of the allografts, there were 71 common femoral arteries, two common and external iliac arteries, one internal iliac artery, and 10 were missing data. The mean diameter of the allografts was 4.69 mm (range

Table 1. Characteristics of population treated with cryopreserved arterial allografts for distal bypass

Variable	Patients ($n = 38$)
Mean age (range) — y	77 (43–96)
Male	14 (37)
Female	24 (63)
Medical history	
Arterial hypertension	35 (92)
Hypercholesterolaemia	23 (61)
Diabetes	12 (32)
Obesity	3 (11)
Active smoker	4 (11)
Previous smoker	14 (37)
Non-smoker	20 (53)
Cardiac disease	17 (45)
Ischaemic heart disease	16 (42)
Cerebrovascular disease	10 (26)
Stroke	3 (8)
Chronic renal failure	14 (37)
Previous revascularisation at the same level	33 (79)
Other revascularisation	24 (57)

Data are n (%) unless otherwise indicated.
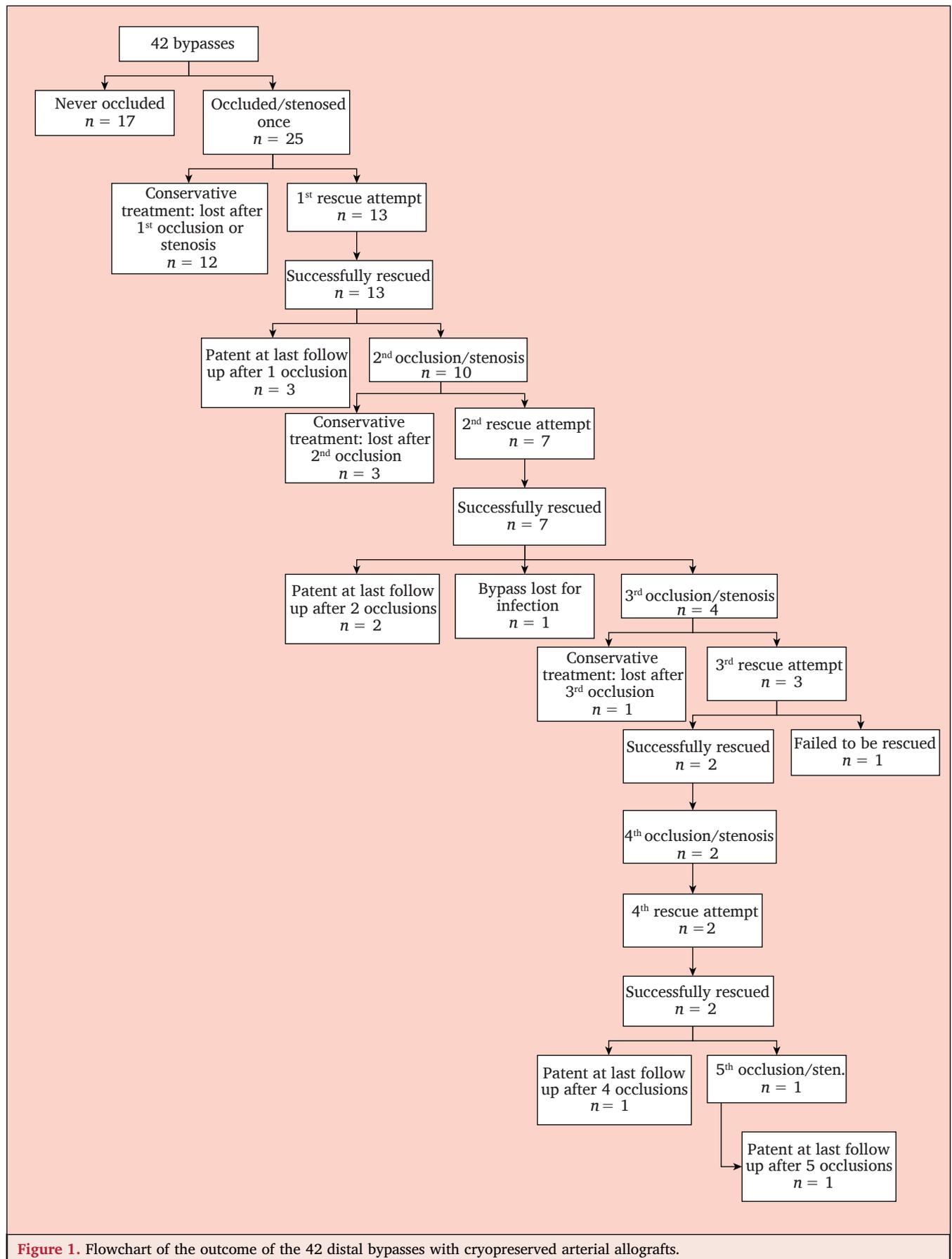
Figure 1. Flowchart of the outcome of the 42 distal bypasses with cryopreserved arterial allografts.

Figure 2. Kaplan—Meier estimates of patency rate of 42 cryopreserved arterial allografts for distal bypass.
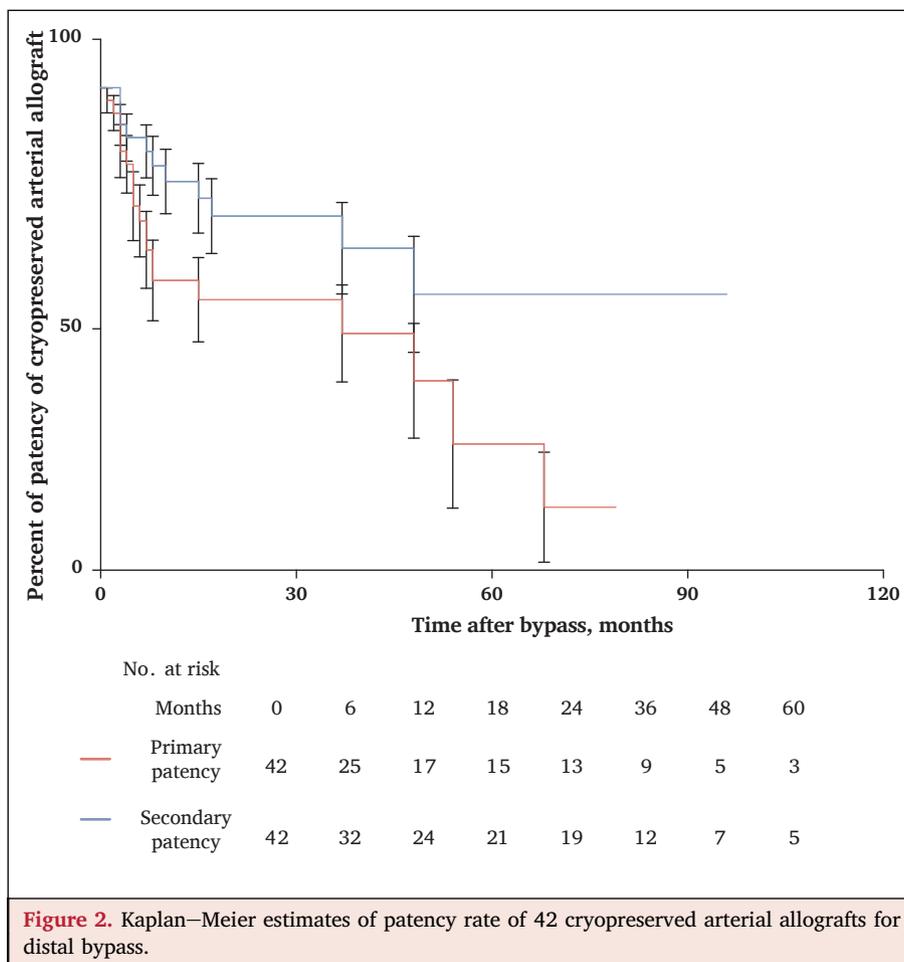
3.38—7.25 mm). The median surgical duration was 158 min (range 74—407 min).

Post-operative time

The median length of hospital stay was 23 days (range 9—226 days). In the post-operative period there were 15 complications in 12 patients (six occlusion, one stenosis, four bleeding, and five wound dehiscences), requiring 13 re-interventions. There were three thrombectomies (twice complemented by bovine pericardial patches and once with bypass extension), four haemostasis, and five re-operations for wound dehiscence. There was one major amputation as an immediate complication in a patient with an occluded bypass. No MACE were observed. The median post-operative plethysmography value was 70 mmHg (range 0—160 mmHg) with a median improvement of 50 mmHg (range 0—130 mmHg).

Post-operatively, all patients were treated with low dose antiplatelet therapy, but the decision to start oral anti-coagulation was individualised as described previously.

Follow up

The mean follow up was 950 days (range 106—2872 days). No patient was lost to follow up. The mean FUI was 0.75. During the follow up there were 30 occlusions, seven
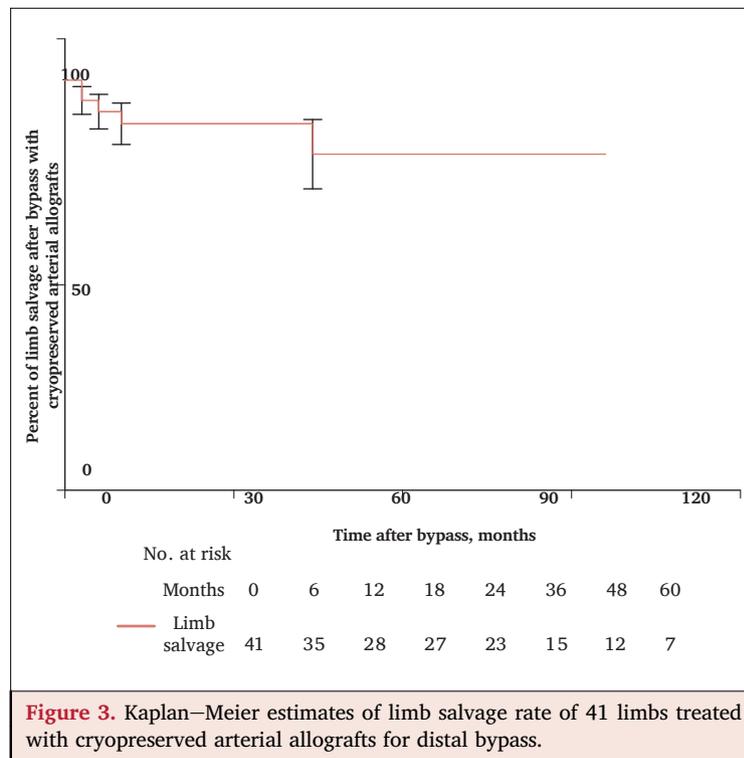
stenoses, one infection, and one bypass degeneration with an anastomotic pseudoaneurysm in 25 bypasses. Twenty-eight endovascular procedures were performed, and 13 thrombolysis, six thrombectomies, two distal extensions of the bypass, two patches, and six major amputations, on 25 different bypasses. The degeneration and pseudoaneurysm complication was corrected with a Viabahn prosthesis. These complications are summarised in Fig. 1.

Estimates of secondary patency at one. two and five years were 81%, 73%, and 57%, respectively. Estimates of primary patency at one, two and five years were 60%, 56%, and 26%, respectively. Estimates of limb salvage at one, two and five years were 89%, 89%, and 82%, respectively. Estimates of survival at one, two and five years were 92%, 76%, and 34%, respectively. Kaplan—Meier graphs are presented in Figs. 2—4

DISCUSSION

The aim of this study was to assess the long-term results of arterial homograft distal bypasses in patients with CLI. It was observed that in patients requiring a distal bypass and lacking suitable veins, allografts had acceptable secondary patency at five years (57%) and a good limb salvage rate at five years (82%).

The ideal vascular bypass graft replicates the mechanical properties of the native artery perfectly in order to maximise patency. In particular, it should demonstrate visco-elasticity for

Figure 3. Kaplan—Meier estimates of limb salvage rate of 41 limbs treated with cryopreserved arterial allografts for distal bypass.
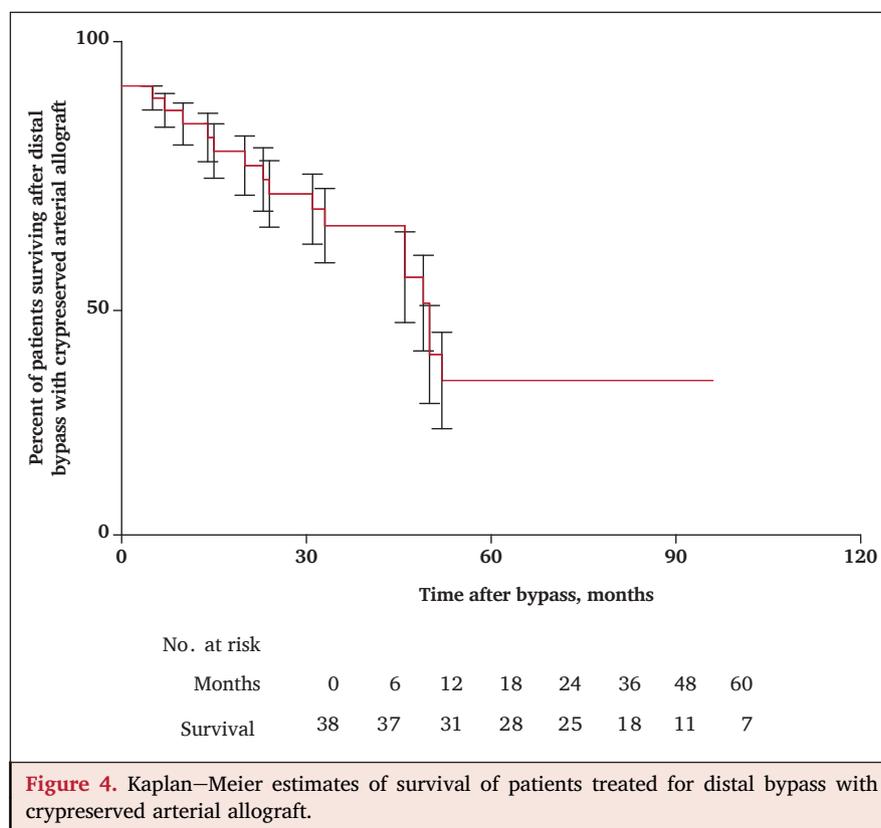
efficient pulsatile flow, matched compliance to prevent intimal hyperplasia, and have a burst pressure well above the physiological range of haemodynamic pressures. Walden *et al.* showed that matched elastic properties between the graft and host conduit led to better results in terms of graft patency.[23] In addition, the graft should be porous enough to promote transmural cell growth and re-endothelialisation. However, excessive porosity could cause fibrous infiltration with subsequent loss of compliance.[23–27] In infrainguinal bypasses, there is a positive correlation between graft compliance and patency.[23] The mechanical properties of the different materials used for distal bypasses are summarised in Table 2. Native arteries have a higher compliance than ePTFE. Importantly, cryopreservation maintains the elastic properties of human arterial allografts, with an elastic modulus that is similar to native arteries.[28] Furthermore, cryopreservation preserves endothelial cells, thus limiting the risk of turbulent flow induced thrombosis. Therefore, based on these mechanical features, arterial allografts could theoretically achieve a better patency and lower thrombosis rate than synthetic grafts.

Arterial allografts have been used and results have been reported since 1999. Reported secondary patency rates at this time were variable, between 74% at 1.5 years and 25% at five years. The patency was low at that time but with an acceptable limb salvage rate of 75%.[10,11] Since then, the quality of cryopreservation, introduced by O'Bryan in 1975, has continuously improved. Nowadays, the EHB in Brussels ensures optimal allograft quality by strict selection of donors and professional processing and distribution of the

products.[26] Recently, two retrospective studies confirmed that open arterial revascularisation with arterial homografts was an acceptable treatment option. In the context of critical ischaemia, Naoum *et al.* reported a primary patency rate of 58% at 18 months, but the study had a small sample size and was performed over a short time period (18 months).[12] Lejay *et al.* showed excellent results using arterial allografts in peripheral prosthetic bypass infection with a primary patency rate of 59% at five years.[19] This patency rate is higher than that observed in the present study, but Lejay *et al.* had only three (11%) distal bypasses. However, both limb salvage rates at five years were comparable (89%).

In general, ePTFE shows poor mid- and long-term patency rates. Reported secondary patency rates vary between 57% at four years and 40% at five years.[14,31] Although these results cannot be directly compared with the present results, a higher secondary patency rate at five years of 57% is reported herein. Only a controlled randomised study could confirm the superiority of allografts over ePTFE. Despite these results, using ePTFE grafts for distal bypass remains a tempting option, particularly in patients who will not tolerate prolonged surgery. Nevertheless, the present study showed an acceptable mean operative time of 174 min, especially when considering that around 80% were re-interventions. In order to achieve such reduced operating times, two surgical teams are required. One team should be dedicated to the allografts preparation as the other prepares the anastomosis sites. Although it could be difficult to ensure the availability of two surgical teams in every

Figure 4. Kaplan−Meier estimates of survival of patients treated for distal bypass with crypreserved arterial allograft.

hospital, it should be explained that allograft preparation does not require advanced technical skills and can be done by a fellow. In addition, patients with critical ischaemia often suffer tissue loss, and are therefore at a higher risk of infection. This condition should also discourage the use of prosthetic material. Whereas the primary outcome (secondary patency) is acceptable and similar to previous series, it should be mentioned that this result was obtained by means of a relatively high re-intervention rate (represented in Fig. 1), compared with the BASIL study, for example.[4] This high re-intervention rate was mainly related to bypass occlusions and not graft degeneration. Indeed, the late fate of arterial allografts, especially aneurysmal degeneration, is questioned in the literature.[32] Only one allograft degeneration occurred among the 42 created bypasses. This result is comparable to that of Albertini et al.,[11] in which only four of 165 bypasses degenerated during follow up. Lejay et al.

reported a 33% allograft related re-intervention rate at five years.[19] This high percentage could be a result of differences in the cryopreservation technique, or to antibiotic treatment administered in the series of Lejay et al. Wound complication related re-intervention (five of 42 bypasses) is similar to the reported rate of wound complications after distal bypass of 10%.[33]

In terms of immediate post-operative complications, there were no MACE and one major amputation in a patient with an occluded bypass. These results were excellent given an expected MACE rate after major vascular surgery of >5%.[33]

In terms of long-term anticoagulation, besides antiplatelet therapy that is routinely administered to every vascular patient, an individual decision of starting anticoagulation was made for every patient, based on the surgeon's preference and the presence of only one patent runoff vessel distal to the bypass. This approach is still

Table 2. Summary of the mechanical properties of the material used for infrainguinal bypass

Parameter	Native artery	Cryopreserved artery	Vein	Expanded polytetrafluoroethylene	Reference
Compliance (%/Pa)	$6.86 \pm 1.79 \times 10^{-5}$	$6.66 \pm 1.8 \times 10^{-5}$	$3.84 \pm 0.81 \times 10^{-5}$		Pukacki et al. (2000)[28]
Elastic modulus (MPa)	1.54 ± 0.33	1.69 ± 0.61	3.11 ± 0.65		Pukacki et al. (2000)[28]
Compliance (%/mmHg)	0.08		0.03	0.016	Kidson (1983)[29]
Elastic modulus (dyn/cm^2)	$4.81 \pm 1.01 \times 10^{-6}$	4.45 ± 1.33		$687.59 \pm 121.52 \times 10^{-6}$	Armentano et al. (2006)[30]
Viscous modulus (dyn × s/cm^2)	$8.53 \pm 1.5 \times 10^{-4}$	10 ± 3.12		0.06 ± 0.01	Armentano et al. (2006)[30]

Data are mean ± standard deviation, unless otherwise indicated.

debated. However, a benefit of secondary patency of infrapopliteal bypasses has been suggested by Liang *et al.*[34]

The survival rate of the present sample was 34% at five years, mainly owing to cardiac related deaths. It could be explained by the high rate of comorbidities of these frail patients suffering from severe vascular disease, known to be a major mortality risk factor. This mortality rate, reported to be 40–50% at five years, is usual for this category of poly-morbid patients who suffer from CLI.[1]

Some limitations exist in this study. It is intrinsically limited by its retrospective design. The mean FUI was 0.75. According to von Allmen *et al.*,[22] this indicates the mortality is underestimated, compared with complete follow up, which is non-negligible. Missing dates of death are most likely the cause of this underestimation. Furthermore, a cost analysis is needed, considering the relatively high re-intervention rate and the fact that cryopreserved arterial allografts tend to be a rather expensive material.

CONCLUSION

This study showed that despite a low rate of primary patency at two years, the secondary patency of arterial allografts is acceptable by means of a relatively high re-intervention rate. Distal homograft bypasses offer lower patency rates than can be obtained with GSV and alternative vein bypasses but could possibly be a suitable alternative for limb saving distal bypass in the particular setting of absence of venous material in patients with critical ischaemia.

CONFLICT OF INTEREST

None.

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None.

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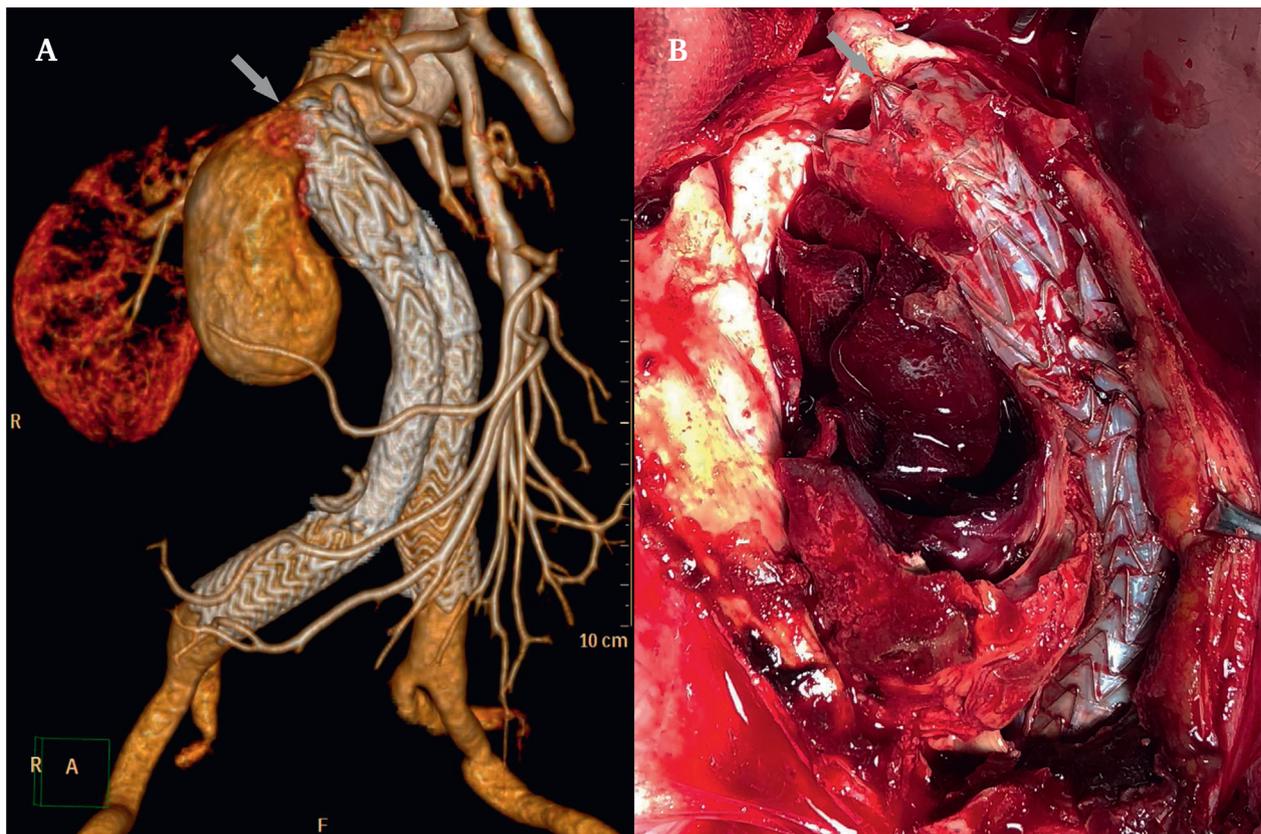
COUP D'OEIL

Type 1a Endoleak Secondary to Graft Migration

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A 58 year old woman who underwent endovascular aneurysm repair one year previously for a 76 mm abdominal aortic aneurysm (AAA) with 70° neck angulation using locally made stent grafts (Percutek Therapeutics Co Ltd, Beijing, China), presented with severe acute abdominal pain. Computed tomographic angiography showed type 1a endoleak secondary to graft migration with AAA sac enlargement (panel A, grey arrow). The patient underwent emergency stent graft explantation (panel B) and aortic reconstruction with a 16 × 8 × 8 mm PTFE (poly-tetra-flour-ethylene) vascular prosthesis (GORE-TEX®, WL Gore & Associates, Flagstaff, USA). The patient made an excellent recovery with no complications at three-month follow-up.

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