Modified Bovine Heterografts for Arterial Replacement

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The accumulated results of various types of vascular repair have led to some uniformity of opinion and standardization of diagnostic technics, operative indications, and technical procedures. Thus, there is now a consensus favoring replacement of the aorta and iliac vessels by knit Dacron prosthetic tubes while the patient's saphenous vein appears best for arterial repairs of the femoro-popliteal segment. However, inavailability of the saphenous vein, for one reason or another, in a small percentage of patients and dissatisfaction with prosthetic Dacron tubes for such procedures have led to our continued interest in alternatives.

Report of a new modification in the preparation of heterologous grafts from the carotid arteries of cows by Rosenberg, Henderson, Lord, and Bothwell in 1964 [49] suggested a need for further evaluation of this material which was said to be nonantigenic, well accepted by the host, and unlikely to form aneurysms. During the past four years we have completed 103 animal experiments and recently 12 heterografts have been placed in human patients with promising early results. While all problems of arterial grafts are not yet solved, this material is a distinct improvement over grafts prepared by other methods and offers sufficient promise to warrant further study in the search for optimal methods of repair in vascular surgery.

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History

Experimental. Pioneers in experimental peripheral vascular surgery recognized that it would be desirable to utilize vessels from other species as vascular replacements in man. The summary in Table 1 shows that there were some experimental heterologous replacements performed as early as 1903, but successful results were always clouded by other experiments in which the grafts either thrombosed or became aneurysmal with subsequent rupture.*

New interest occurred following World War II but again results of heterograft vascular replacement were unpredictable and it appeared that heterologous vessels either thrombosed or formed aneurysms and disrupted in too many instances to warrant clinical use. Various methods of preparation and preservation failed to alter the poor results and led to the statement in 1954 that "the fate of heterografts is that of gradual deterioration rather than replacement and restoration." [11]

The initial report on enzyme-modified heterografts was by Rosenberg, Gaughran, Henderson, Lord and Douglas [47] in 1956. They described experiments with bovine carotid arteries partially digested by the proteolytic enzyme ficin with subsequent formaldehyde fixation. Enzymatic removal of most of the elastic and muscle fibers caused increases in diameter and length in

* The reports in Table 1 are listed to furnish the interested reader with as complete a background as possible. Many are preliminary and incomplete studies.

TABLE 1. *Summary of*

Authors	Year	Heterograft Source and Recipient	Graft Prep.
Hopfner[25]	1903	rabbit and dog → dog femoral	
Stich[59]	1907	rabbit, cat and human → dog	
Guthrie[21]	1907	cat and rabbit → dog carotid	fresh
Klatz, Permar and Guthrie[34]	1907	rabbit → dog carotid	fresh
Carrel[5, 6, 7]	1907 1908	dog → cat aorta	cold storage
	1912	human → dog	
Ward[62]	1908	rabbit and cat → dog carotid	
Gross, Bill and Pierce[20]	1949	hog, baboon and human → dog abdominal aorta	cold storage
Gauthier-Villars[19]	1950	pig and calf → dog iliac	cold storage
Colombo, Teich and Costa[9]	1951	? → dog femoral, carotid and aorta	cold storage or frozen
Juszczynski[31, 32]	1950	pig, calf, sheep → to dog abdominal aorta	cold storage
Oeconomos and Hewitt[43]	1952	human, calf, sheep, horse and pig → dog abdominal aorta	various
Hufnagel, Rabil and Reed[26]	1953	pig, calf, lamb and human → dog aorta	sterilized by ethylene oxide; freeze dried
Zannini, Cocchia, and Angrisani[66]	1953	lamb, horse, and hog → thoracic aorta	cold storage
Sautot, Bost, Touraine, Martin and Feroldi[2, 55, 56]	1954	calf → dog abdominal aorta	frozen
Creech, DeBakey, Self and Halpert[11]	1954	sheep, calf, hog → dog	formalin
Pate[46]	1954	pig → dog	freeze dried
Kimoto, Sugia, and Tsunoda[33]	1954	?	fresh
Rosenberg, Gaughran, Henderson, Lord and Douglas[47]	1956	cow → abdominal aorta	ficin-digested, formalin treated and lyophylized
Sauvage and Wesolowski[57]	1955	human, dog → young pig thoracic aorta	fresh; freeze dried
Hardin[22]	1955	monkey → dog	fresh; frozen
Donati, Compani, and Martino[18]	1956	calf, horse, pig → dog	formalin
Bost, Joubert and Sautot[3]	1956	calf → dog aorta	lyophylized
Martino, Campani, Guagliano and Scarabelli[39]	1956	calf, horse, dog → dog aorta	formalin
Szilagyi, Shonnard, Lopez and Smyth[60]	1956	cow → dog thoraco-abdominal	lyophilized
Wesolowski and Sauvage[63]	1957	human, pig → dog aorta	freeze dried
Inahara, Menendez, Shaw and Linton[28]	1957	cow → dog aorta	frozen and irradiated

Total No. Grafts	No. Patent	No. Thrombosed	No. Aneurysms	No. Bled	Comments
1		1			Complete failure, number unstated, first attempt made
1	1				First successful attempt
2	2				Vaseline coated
1	1				Dilated with calcium deposit 7 months
5	4	1			
1	1				Slight dilatation at 4 yr
2	2				
8	3	3	2	1	One marked intimal sclerosis
14	5	7		2	Some aneurysms
8	7	1			7 had mural thrombosis
15	9	6			
			some		Thromboses of fresh grafts delayed by fixing solution
21	18	2		1	2 partially thrombosed. High percentage successful in pigs and calves
14	12		1	1	3 had mural thrombosis
?	?	?	?	?	No failures, number not stated
40	32	7			15 constricted and 8 dilated, but patent
					37/40 acceptable
?	50%		50%		Failure due to necrosis
33	4		1		Initial report of present graft
114	57	9	45	3	
27	7	16	1	3	
20	20				Condition of graft not otherwise stated
14	14				2 dilated
24	20	4			
20	10		1	6	3 still living and well
54	49	4	3	1	Marked ulceration in many
7	5	2			All survivors had abundant thrombus at 30 days

TABLE 1.

Authors	Year	Heterograft Source and Recipient	Graft Prep.
DeMuylder and Hennebert[15]	1957	human, calf → dog abdominal aorta	formalin
Kraljevic, Magazinovic, Piscevic, Ginzberg and Vajs[36]	1957	calf → dog abdominal aorta	freeze dried
Papo, Ginzberg, Vajs, Kraljevic, Magazinovic and Piscevic[45]	1957	calf → dog aorta	lyophilized and freeze dried
Cornet, Kerneis, Dupon and Coiffard[10]	1958	ox → dog thoracic aorta	formalin
Morton and Mahoney[41]	1958	pig → dog abdominal aorta	Fresh and cold storage
deTakats, Thompson and Dolowy[16]	1959	cow → dog thoracic aorta	ficin digested
Henly, Crawford, DeBakey and Halpert[23]	1959	horse → dog abdominal aorta	freeze dried
Rykowski[54]	1960	human → dog abdominal aorta	fresh
Johnson, Easling Nemir[29]	1960	cow → dog aorta	plasma storage
Bonilla-Naar and Alvarez-Vazquez[1]	1961	human, pig → dog thoracic aorta	proteolytic eynzyme treated and alcohol stored
Rosenberg, Henderson, Lord, Bothwell and Gaughran[50]	1962	cow → dog thoraco-abdominal and abdominal aorta	ficin digested +; formalin, polyacrolein "stabilized" formalin or dialdehyde starch treated
Rosenberg, Henderson, Lord and Bothwell[49]	1964	cow → dog thoracic and thoraco-abdominal aorta	ficin and dialdehyde starch
Tamames and Sanchez[61]	1964	lamb → dog thoracic and abdominal aorta	cold storage
Kovanov and Bilenko[35]	1964	? → dog carotid and femoral	fresh or lyophilized
Breslau, Schwartz, Smith and Rob[4]	1965	cow → dog IVC	ficin +, dialdehyde starch or polyacrolein
Silver, Kaye, Aquilizan and Hurwitt[58]	1965	cow → dog abdominal aorta	ficin digested polyacrolein
Wimberly, Lewis and Dale[64]	1968	cow → dog carotid and femorals	ficin and new methods

(*Continued*)

Total No. Grafts	No. Patent	No. Thrombosed	No. Aneurysms	No. Bled	Comments
22	12	7	1	2	3 thromboses seen
40	12	18		10	
					24/40 survived but only 4 grafts "completely trouble-free"
18	14	2		2	One graft composite with ivalon
23	7	7		9	Grafts wrapped with polyvinyl sponge to prevent aneurysms
43	26	2	8	7	Early form of heterograft discussed in present article
15	4	1	10		
10	10				Contaminated in bacterial cultures and irradiated to sterilize
28	16	6	6		short grafts
20				20	Some grafts lined with autogenous pericardium
33	33				Modification of the heterograft under discussion
26	24	1	1		New modification of present graft
25	19	*			* 6 grafts ulcerated or thrombosed
20	6	14			6 had mural thromboses
14	6	8			
16	12	4			
30	22	8			Used as lateral patches

TABLE 2. *Summary of Heterografts Placed in Humans*

Author	Reported	Description	Result
Hufnagel, Rabil and Reed[26]	1953	aorto-iliac graft of composite calf and pig vessel calf heterograft to radial artery, popliteal aneurysm and iliac aneurysm	Patent six months 3/3 successful
Oudot[44]	1954	fresh dog arteries grafted to femoral arteris in three patients	Patent by arteriogram
Creech, DeBakey, Self and Halpert[11]	1954	freeze-dried hog's artery replaced the iliac artery following cancer resection	Dilated to twice size on angiography at five months. Recently thrombosed at autopsy 7 months postoperative
Sautot, Bost, Touraine, Martin and and Feroldi[56]	1954	calf artery in a patient with arterial injury patients with obliterative disease	Good result Good results in some
Kimoto, Sugia and Tsunoda[33]	1954	aneurysm right iliac artery grafted with dog's artery stored in 70% alcohol	Uncertain, but limb appeared normal
		aneurysm right carotid artery replaced with dog graft	Patient died pulmonary edema second day. Graft patient without thrombus
		two femoral arteries replaced with dog grafts	Patent on arteriogram 16 and 25 days postoperatively
		aneurysm abdominal aorta replaced with sheep graft	Patent on aortogram 35 days postoperative. Patient clinically well seven months postoperative
Oudot[44]	1954	80 grafts prepared by Gross' technique	Three leaks. Used aortic bifurcation with 10/12 good results
Leger and Oudot[37]	1954	dog heterograft to femoral artery for injury	Blocked on arteriogram one month postoperatively
Hardin[22]	1955	pig heterograft frozen and irradiated replaced femoral artery aneurysm	Patent at nine months angiographically
Donati, Campani and Martino[18]	1956	heterografts to replace femoral arteries in unstated number of patients	Satisfactory results. One arteriogram showed patency at three months
Newton, Ray and Butcher[42]	1958	equine carotid arteries modified by partial peptic digestion used to bypass femoral artery occlusive disease	
		six patients	Immediate thrombosis
		two patients	Thrombosed 3 and 6 months
		seven patients	Multiple aneurysms formed requiring excision from 3 weeks to 18 months postoperatively
		one patient	Multiple aneurysms and thrombosis

TABLE 2. (*Continued*)

Author	Reported	Description	Result
Rosenberg, Martinez, Sawyer, Wesolowski, Postlethwait and Dillon[53]	1966	bovine carotid arteries treated with ficin and tanned with dialdehyde starch	
		aorta to right profunda femoral (Dacron upper end)	Functioning 32 months
		2 ilio-femoral bypass	1 functioning at death 1 month postoperatively, 1 functioning 12 months postoperatively
		13 femoro-popliteal bypass	8 functioning at last follow-up or death 2 weeks to 16 months (one required revision for thrombosis) 5 failed; 28 months, 7 months, 2½ months, and 2 immediately postoperatively
		end-to-end popliteal	Functioning arteriographically 4 months postoperatively

the order of 35%, but the remaining collagen appeared quite resistant to mechanical stress.[48] However, further animal investigation showed that these grafts still frequently degenerated into aneurysms as noted by deTakats, Thompson and Dolowy in 1959.[16]

Review of the processing technic indicated that the enzyme ficin was highly resistant to elution, that digestion had, in fact, been continuing throughout processing, and that the formaldehyde bonding and tanning was being reversed during sterilization and storage. As a result, an unnecessary weakening of the collagen remaining in the tube occurred.

The process was therefore modified to a more rigidly controlled digestion period, with de-activation of enyme and subsequent crosslinking with dialdehyde starch. Heterografts prepared by this method have appeared to retain their strength for long periods of time without evoking antigenic responses.[49, 50]

Clinical. As has been noted, the few untreated heterologous arteries which were placed in humans in the 1940's and 1950's did not appear any more promising than predicted by the experimental results although some successes were reported (Table 2).

In 1966 Rosenberg, Martinez, Sawyer, Wesolowski, Postlethwait and Dillon reported the first results of human arterial replacement by the dialdehyde starch-modified bovine grafts, indicating that 13

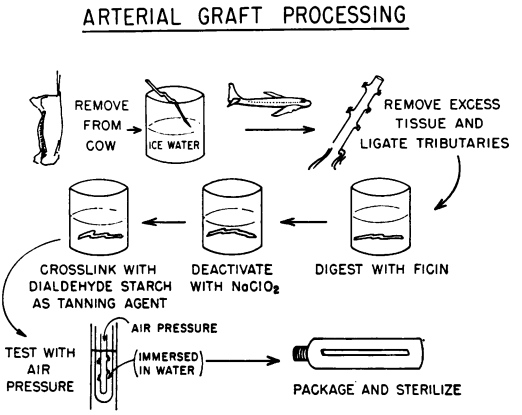

FIG. 1. Steps in preparation of the modified bovine carotid heterografts are discussed in the text.

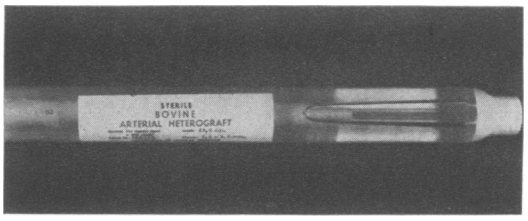

FIG. 2a. Following processing, the modified bovine heterografts are stored on glass mandrels in tubes containing 50% ethanol in water.

of 18 had remained patent for as long as 32 months without development of aneurysm.[58] This work led to a rekindling of interest in the heterograft technic and further intensive study in animals and in humans.

Experimental Background

Method of Preparation. Carotid arteries are removed from the carcass in the slaughter house, immersed immediately in iced water and air shipped to the laboratory for processing. There they are washed, stripped of surrounding tissue, and the tributaries are ligated with 000 surgical silk (Fig. 1). The muscle and elastic tissues are substantially removed by treatment for two and one half hours at 37° C. in a 1% solution of ficin * buffered with citrate to pH 5.5. The arteries are again washed and any further enzymatic action stopped by immersion in 1% aqueous sodium chlorite for 18 hours. Tanning then is accomplished by placing the vessels for 24 hours in 1.3% aqueous dialdehyde starch solution buffered to pH 8.8 with saturated sodium bicarbonate. This dialdehyde starch improves crosslinking of collagen and serves to add stiffness in the wall and to reduce susceptibility to biological attack and resorption when used as a graft.

* Ficin is a proteolytic enzyme obtained from the latex of fig trees growing wild in the tropical rain forests of the Amazon valley.

TABLE 3. *Summary of 103 Heterografts in Dogs*

	Patent/ Total	Comment
Aorta: end-end (9 had renal artery transfers)	17/19	2 aneurysmal dilatation
composite with dacron	9/11	2 ruptured and bled
Femoral: end-end or cross-over	0/19	1 ruptured and bled
lateral patch	22/30	
"diminished"	2/11	
Shunt: to renal	0/6	
"diminished" to renal	1/2	1 ruptured anastomosis at 41 days
IVC: end-end	0/5	

Following tanning the arteries are again washed and tested for security of tributary ties and possible leaks and for focal weakness with air pressure equivalent to 240 mm. of mercury. Those that pass all tests are slid onto glass mandrels and placed in tubes containing a sterilizing solution composed of 50% etanol in water plus 1% propylene oxide. The units are held at 100° F. for 14 days for sterilization. The glass container serves as a package until the grafts are used (Fig. 2).

Animal Experiments. One hundred and three bovine grafts prepared in this way have been placed in dogs. The experiments are summarized in Table 3. The only operative difficulty was a tendency for bleeding to occur at the suture line unless the graft were maintained on constant firm tension during placement, and there was a slight tendency of the suture to "hang" in the adventitia of the graft during placement. Nevertheless, these grafts were found as easy to handle and place as any other presently available material.

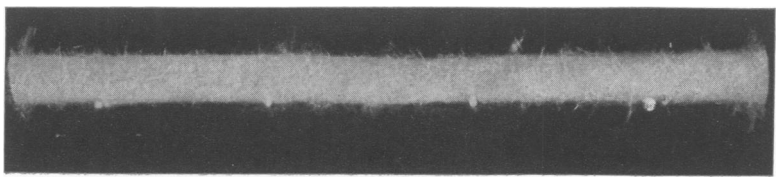

FIG. 2b. The shaggy appearance of the adventitia and the ligated branches are shown.

FIG. 3a. Heterograft replacement of the canine aorta with transplantation of both renal arteries.

FIG. 3b. Autopsy specimen of the same canine aortic replacement with orifices of renal arteries clearly visible 197 days later. Glistening whitish neointima extends from both anastomotic lines and from the renal orifices.

The technic of placement was as follows. The lumen as well as the exterior of the graft was washed thoroughly with normal saline prior to placement. Continuous 5-0 Dacron sutures were used for the anastomoses. While the interior of the opened vessels was irrigated with heparinized saline, no anticoagulant was given during or after operations. Each procedure was conducted as similar to a human operation as possible. The animals were returned to cages and no particular postoperative care was given. No antibiotics were used.

Aortic replacement grafts maintained patency in 26 of 30 experiments. In nine animals the cut ends of both renal arteries were anastomosed to the sides of the bovine grafts and also maintained patency (Fig. 3). Eleven aortic grafts were performed using a composite of Dacron prosthesis sutured to a section of bovine graft (Fig. 4).

Femoral bypasses and cross-overs were failures in 19 animals, probably because of disproportion between the large grafts (10 to 11 millimeter luminal diameter) and smaller femoral vessels (three to five millimeter luminal diameter).

Since luminal disproportion appeared to influence patency unfavorably, 13 grafts were prepared by diminishing the size of the graft to five or six millimeters by placing a double row of metallic staples down the center of the graft. These "diminished" grafts were used as femoral replacements nine times and as obligatory renal shunts

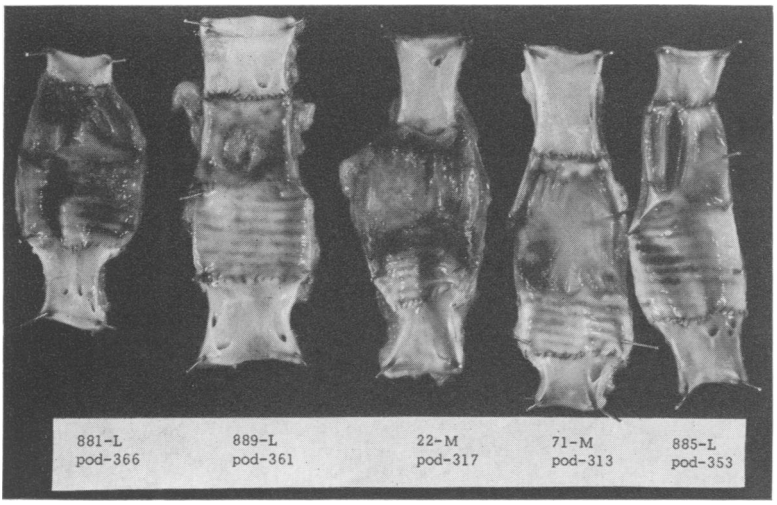

FIG. 4. Five composite heterograft-knit Dacron preparation removed more than 300 days postoperatively. In each instance the short segment of Dacron is well covered with whitish neo-intima to which patches of thrombus are adherent occasionally. 889-L shows almost complete covering except for a central area. 71-M emphasizes the ingrowth of whitish neo-intima from the anastomotic lines with incompleteness centrally.

twice. Three instances of long-term patency resulted (seven months in two and 41 days in another animal). These findings suggested the desirability of using small diameter grafts.

Results of 30 lateral patch grafts placed in the carotid and femoral arteries (by a standard technic previously reported for patch graft evaluation [12]) have been previously reported.[64] The patency rate of heterograft patches was almost as good as that of autogenous vein patches.

Five inferior vena caval replacements by bovine heterografts were uniformly unsuccessful.

Discussion of Experimental Results. Experimental results indicated that the host dog accepted the modified bovine graft without antigenic response or rejection in any instance. Microscopic examinations of multiple sections of implanted grafts showed a minimal infiltration with round cells, and there was a relatively small amount of fibrosis about the graft (Fig. 5). The perivascular fibrotic reaction uniformly appeared less dense than about synthetic tubes placed in either animals or humans.

The inner surface of bovine grafts at approximately a year following operation was uniformly an irregular yellowish smooth surface, appearing to be the original graft, alternating with irregular areas in which reddish thin fibrinous material was adherent to the wall. In renal implants, a thin glistening whitish internal neo-intima appeared to extend from the suture lines at either end from the orifices of the implanted renal arteries for short distances (Fig. 3b). Several very short grafts were completely covered by this neo-intima but complete internal coverage did not occur in longer grafts. Geoffrey Lord believes that the internal surface undergoes a constant dynamic variation between resolution and deposition of fibrinous material,[38] but such an observation is incapable of proof at present.

Comparison of the luminal surface of the graft with the attached Dacron tubes in nine surviving composite replacements of the aorta indicated some similarities between the two in that both had irregular areas of covering alternating with areas of incomplete intimization (Fig. 4). In these relatively short segments it appeared that synthetic grafts had more complete coverings of thin whitish "neo-intima" than did attached bovine grafts, but no quantitative evaluation of this was made. Whether final complete intimal covering is of functional importance or not is indeterminate, since the majority of synthetic grafts recovered

from humans do not show complete lining surfaces even though function has been excellent and without thrombosis or embolization. While these experiments do not show complete anatomic covering, the function of the grafts has been good.

Two instances of aneurysmal dilatation of a portion of the wall of the bovine graft have been encountered. Neither of these was very large but both appeared to indicate a ballooning of a localized part of the graft.

The importance of luminal disproportion in production of thrombosis (perhaps by means of eddy currents and stasis) is emphasized by the failure of patency of relatively large bovine grafts when applied to small canine host arteries in both femoral and renal arteries. There appears to be a tendency for internal thrombosis to adjust the lumen of disproportionate areas to the same size in human aneurysms (where the clotting of most of the outer portion of the aneurysm is well known), of lateral patch graft (where clotting has been shown to occur if the graft is too loosely applied), and in these bovine graft experiments where luminal disproportion occurred. It may be postulated that luminal disproportion encourages the beginning of thrombosis in the larger diameter segment with continuation of the thrombosis so that total occlusion of the vessel is likely.[52]

The failure of patency of inferior vena cava replacements confirm reports of Breslau, Schwartz, Smith and Rob,[4] and Silver, Kaye, Aquilizan and Hurwitt.[58] Our efforts to replace inferior vena cava with a variety of substances has consistently failed except when autogenous jugular vein is used.[13]

These experiments, viewed in the light of reports of others, indicate that modified bovine graft is an improvement over earlier preparations by virtue of lack of antigenicity,[14] absence of inflammatory response, excellent incorporation into the host, and relative lack of aneurysmal degeneration. The grafts are easy to handle and excellent

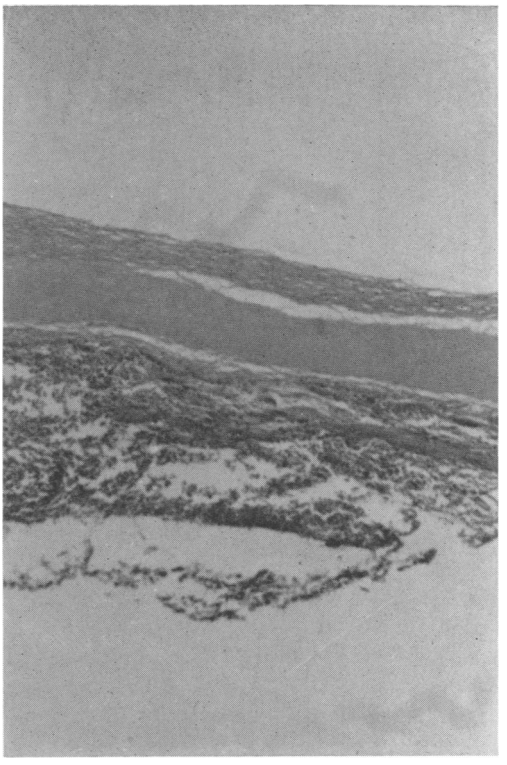

FIG. 5. Microscopic section of a modified bovine heterograft placed in the dog aorta shows minimal round cell infiltration and fibrosis with excellent preservation of collagen architecture.

patency results when grafts are applied to vessels of similar caliber. The grafts appeared to function well for reasonable periods of time. Hence, cautious clinical applications were carried out.

Clinical Results

Modified bovine grafts were placed 12 times in lower extremities of ten patients as summarized in Table 4 and Figure 6. Seven of the 12 are patent to date.* Four thrombosed in the early postoperative periods and one thrombosed 15 months after operation.

There was no technical difficulty with placement of the graph in any instance and the time of operation was less than that usual with autogenous vein grafts. Blood loss and tissue trauma also appeared to be

* December 5, 1968.

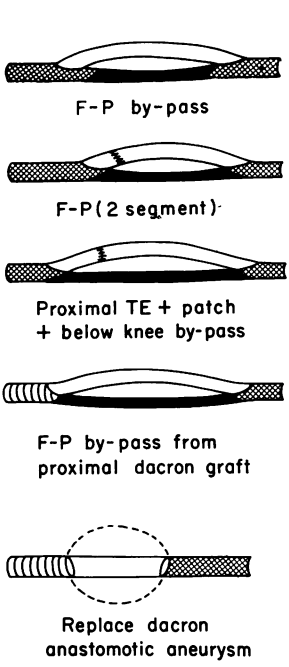

FIG. 6. Summary of results of 12 grafts placed in humans.

less than with venous replacement. In each instance the graft was placed in a subcutaneous tunnel where it was easily palpable. There did not appear to be any unusual amount of overlying or nearby inflammatory response.

Two grafts were thrombosed within eight hours of conclusion of the operation and were cleaned out by immediate thrombectomy and thereafter remained patent. The cause of these thromboses is not clear and both are open (18 months and one month, respectively). No technical errors occurred. Whether failure of the two-segment (sutured end to end) grafts is significant or coincidence is unknown. The only complication aside from thromboses was venous thrombosis in the calf of one patient which did not appear related to the graft.

Case Reports

Case 1. W. B., a 71-year-old man, had a right femoro-popliteal autogenous saphenous venous bypass placed to relieve incapacitating claudication July 2, 1965. Convalescence was uneventful and the graft was palpable until it suddenly thrombosed six months later. Need for secondary repair was indicated by return of incapacitating claudication and a bypassing modified bovine graft was placed on December 23, 1965. This graft remains patent 35 months following operation.

Seventeen months after the bovine graft was placed, for incapacitating claudication in the left leg another bovine graft was placed as a femoropopliteal bypass on that side on April 24, 1967. It remained patent for 15 months when thrombosis occurred. Claudication returned but the extremity has continued viable.

Comment: The right femoral arteriogram in Figure 7 was made 14 months postoperatively and shows the patent graft. The lower part of the angiogram does not reproduce clearly and is omitted but showed irregularities of lumen similar to that seen in the upper portion. While these irregularities have been of concern, this first graft placed has remained patent 35 months. The

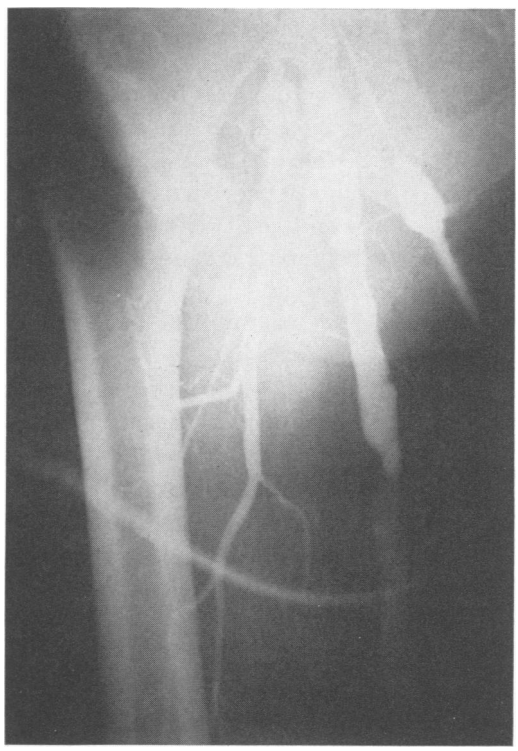

FIG. 7. Case 1, femoral arteriogram 17 months after operation shows irregularity suggesting adherent internal thrombi. The graft has remained patent 35 months.

TABLE 4. *Twelve Modified Bovine Heterogra ts in 10 Patients*

Patient	Sex/ Age	Site	Indication	Results	Complications
1. D. J.	59/M	femoro-popliteal	Failure of previous thromboendarterectomy	Patent 6 months	Venous thrombosis of calf
2. W. K.	64/M	2 segment femoro-popliteal	Failure previous vein graft	Early thrombosis	Above-knee amputation later
3. W. B. (R)	71/M	femoro-popliteal	Failure previous vein graft	Patent 35 months	
4. (L)	74/M	femoro-popliteal	Primary choice	Thrombosed after 15 months patency	
5. W. P.	68/M	femoro-popliteal	Saphenous vein not suitable	Patent 2 months	Wound healed slowly; died myocardial infarction 2 months postoperatively
6. J. B.	40/M	femoro-popliteal graft added to old aortic-iliac dacron graft	Saphenous vein not suitable	Patent 18 months	Thrombosis that same night cleared by catheter
7. W. P. (L)	48/M	aneurysm at femoral dacron anastomosis	Primary choice	Patent 9 months	
8. (R)	48/M	same	Primary choice	Patent 12 months	
9. E. C.	80/F	femoro-popliteal	Saphenous vein not suitable	Early thrombosis	Sudden death 9 days postoperative of myocardial infarction
10. R. C.	59/M	2 segment femoro-popliteal	Failure previous vein graft	Early thrombosis	
11. W. N.	79/M	femoro-popliteal	Saphenous vein not suitable	Patent 1 month	Thrombosis same night cleared by catheter
12. W. F.	61/M	proximal thrombo-endarterectomy and long graft to anterior tibial	Saphenous vein not suitable	Early thrombosis	

cause of delayed thrombosis at 15 months in the contralateral side is unexplained.

Case 2. J. B., a 40-year-old man, on November 21, 1966, had a knit Dacron Y graft placed between the aorta and both femoral arteries for relief of aorto-iliac occlusion causing ischemia and constant pain in the right foot. The same evening secondary operation was required because the right limb of the Dacron graft thrombosed. This limb was shortened by resection and anastomosis since it appeared to have buckled due to excessive length. At the same time thromboendarterectomy of the superficial femoral artery was done and was reconstructed with a saphenous venous patch graft. Thereafter excellent pulses were maintained in both femoral arteries, although there was no pulse in the foot because of a known block in the right superficial femoral artery.

Five months later the pain and numbness in the right foot was not completely relieved and was disabling. A graft was placed from the side of the distal end of the right limb of the previous Dacron graft down to the popliteal artery just above the knee. Since a portion of the saphenous vein had been previously used and was not avail-

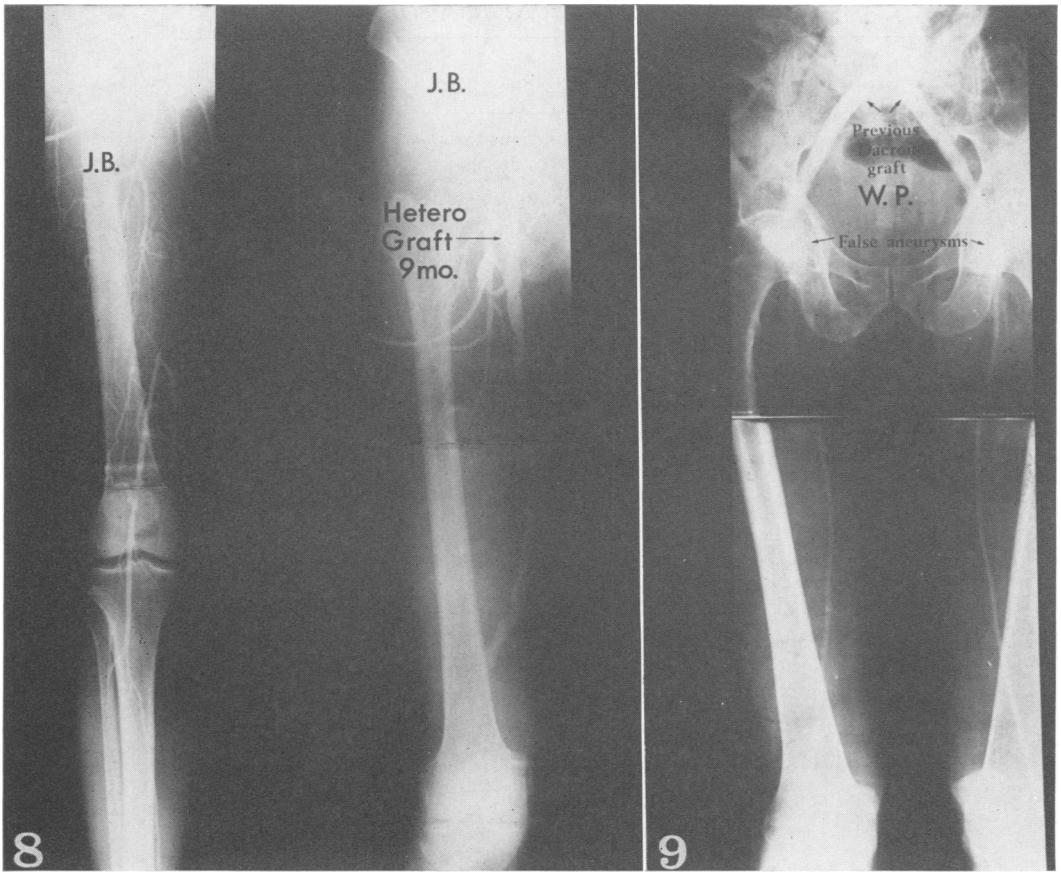

FIG. 8. Case 2, *left,* preoperative femoral arteriogram shows femoral occlusion with popliteal reconstitution; *right,* femoral arteriogram nine months postoperatively showing luminal irregularities suggesting adherent thrombi. Graft has remained patent 18 months.

FIG. 9. Case 3. Bilateral false aneurysms 38 months after a Dacron Y graft was placed between the aorta and both femoral arteries. These were replaced at two different operations by modified bovine heterografts and have remained patent 12 and nine months, respectively.

able, a modified bovine graft was implanted. It has remained patent since May 26, 1967, 18 months.

Comment: Angiograms in Figure 8 illustrate this repair. Again, the lumen of the bovine graft is irregular and it appears that there is an irregular deposition of fibrinous material similar to that seen in experimental animals. Function, however, has continued to be splendid.

Case 3. W. P., a 68-year-old man, had an onlay knit Dacron Y graft placed between the aorto and both femoral arteries on September 22, 1964, for relief of bilateral incapacitating calf claudication. Pulses returned to both feet and convalescence was uneventful. Twenty-eight months later he was found to have small aneurysms where limbs of the Y graft were attached to the femoral arteries. These bilateral false aneurysms are shown in Figure 9. On November 27, 1967, the right aneurysm was resected and replaced with an end-to-end modified bovine graft between the Dacron tube and the common femoral artery and six weeks later, on January 5, 1968, the other side was similarly replaced. Both grafts continue to function well during the 12 and 10 month follow-up periods.

Comment: Bovine graft was chosen as a replacement in the hope that it would be more pliable and less stiff than a synthetic tube and therefore less likely to pull away

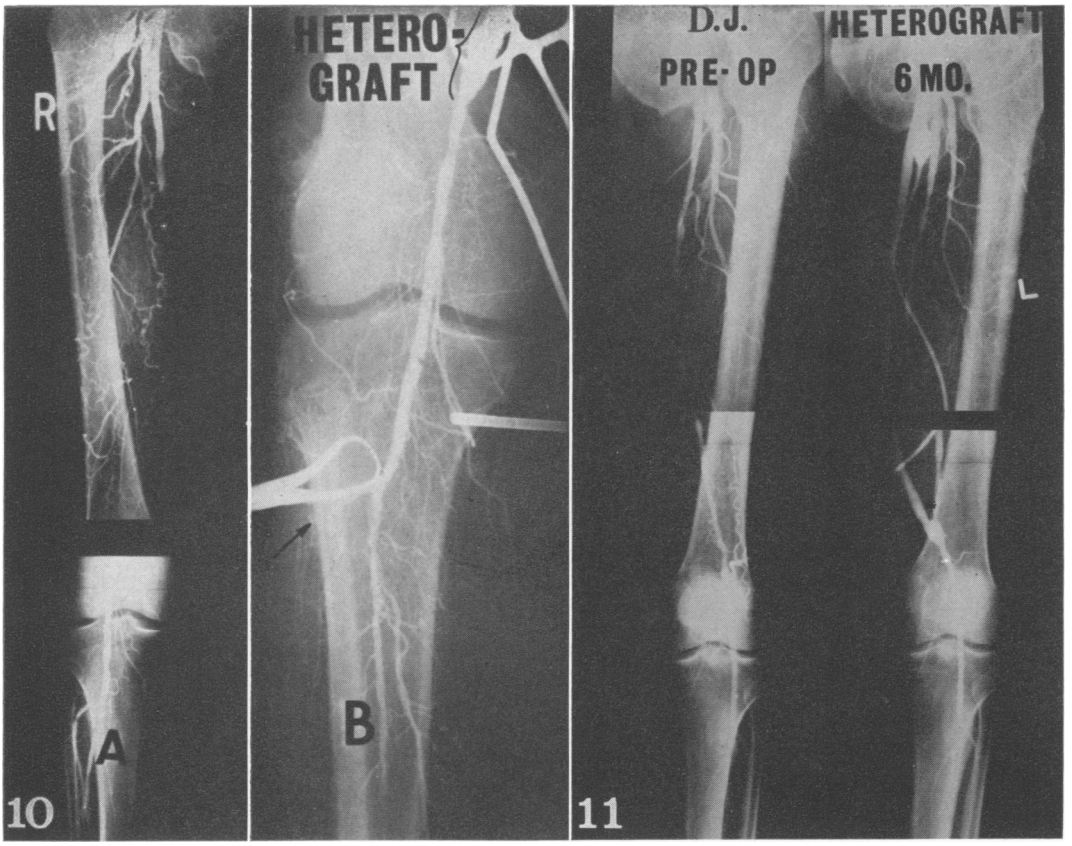

FIG. 10. Case 4, *A*, shows femoral occlusion with popliteal reconstitution preoperatively; *B*, shows arteriogram immediately following secondary operation to clear thrombosed heterograft. Although anterior tibial artery failed to opacify (arrow) the dorsalis pedis pulse returned immediately and has remained palpable throughout the one month follow-up period. The anastomosis of heterograft to popliteal artery is indicated by the bracket at top.

FIG. 11. Case 5, 6 months after operation, the femoro-popliteal heterograft is smooth and patent.

slowly from the femoral arterial wall with aneurysm formation. Both original silk suture lines were intact in the wall of the false aneurysms. The graft was placed using Dacron suture material.

Case 4. W. N., a 79-year-old man, required a femoro-popliteal bypass graft for ischemia with continuous pain at rest in the foot and toes. The saphenous vein was formed of three small tributaries half-way down the thigh and was too small for use as an arterial bypass. A bovine graft was placed October 17, 1968, with immediate return of a foot pulse. Four hours later the foot was cool and pulseless. Secondary operation upon the lower end of the graft showed it to be completely filled with clot. This was removed in both directions with Fogarty catheters. Arteriogram (Fig. 10) immediately afterward showed patency of the popliteal, tibial and peroneal vessels but lack of filling of the anterior tibial artery. Despite this radiographic appearance all pulses became palpable and have remained so in the one month follow-up period.

Comment: The cause of this early postoperative thrombosis is not clear, nor is the failure of opacification of the anterior tibial artery at a time when the dorsalis pedis pulse was palpable. The immediate thrombectomy has, however, resulted in satisfactory convalescence to date.

Case 5. D. J., a 59-year-old man, had previously undergone femoral thromboendarterectomy elsewhere for relief of claudication. This repair

TABLE 5. *Current Preferences for Arterial Repair*

	Short Block	Long Block or Aneurysm
large artery (aorta, iliacs)	thrombo-endarter-ectomy	knit dacron tube
small artery (femoro-popliteal, carotid)	thrombo-endarter-ectomy	1) autogenous vein 2) bovine heterograft 3) knit dacron tube

failed so the entire area was bypassed with the modified bovine graft from femoral to popliteal artery. This has remained patent for six months with relief of symptoms.

Comment: Figure 11 shows the smooth and patent bovine graft six months after placement as a femoro-popliteal shunt.

Discussion

Autogenous venous bypass graft is the material of choice for replacement of the femoro-popliteal system (Table 5). Modified bovine graft may prove to be a second choice and at present is preferred to either a long thromboendarterectomy or a Dacron tube graft when the saphenous vein is unavailable or not suitable. Four of 12 extremities had bovine grafts placed because of failure of previous repairs (three using vein graft and one having previous thromboendarterectomy). In five other extremities the indication for bovine grafts were non-suitability of the saphenous vein. Femoral anastomotic aneurysms were replaced with bovine grafts rather than secondary synthetic tubes since this material had already failed.

The important question regarding modified bovine grafts is whether aneurysms (or any other form of graft degeneration) will result as with previous heterologous arterial replacements. The answer will depend on long term follow-up studies. This new bovine graft has done well in laboratory animals. Geoffrey Lord states that aneurysms have not occurred in dogs followed as long as seven years.[38] Our experiments have been terminated at a year or sooner. There were two instances where aneurysmal dilatations occurred. Whether these would have enlarged and ruptured after a longer period is not known. Follow-up in humans have not disclosed aneurysmal degeneration 35 months after placement.

The second question is tissue reaction and antigenicity. Neither in experiments nor in humans has there been evidence of tissue reaction in the vicinity of the implanted graft. No graft has yet become available for histologic study from humans.

The appearance of some postoperative arteriograms is disturbing because irregularities which could be internal mural thrombi have appeared. Internal linings of experimental grafts are irregular and angiograms suggest the same in patients. A single late thrombosis (at 15 months) occurred among eight grafts and seven remain patent for periods between one and 35 months.

It has been suggested [38] that a true "neo-intima" does not form in modified bovine grafts and that deposits of fibrinous material seen at post-mortem vary from time to time and are not constant. It was also suggested that intimization differs from that with textile grafts in which intimization occurs through the organization of interstitial thrombi extending along the internal surface of the prosthesis. "Neo-intima" is therefore organized mural thrombus, the fibrotic elements of which extend to the external fibrous sheath through interstices in the prosthesis. It has also been suggested that there is a thin internal membrane covering the yellowish inner surface of the modified bovine graft and that this can be demonstrated by careful histologic study. However, no migration of fibroblasts through the wall of the bovine graft occurs.

The present report of experiments and of the twelve patients operated upon should

not be construed as a statement that modified bovine graft is the best grafting material. Autogenous materials should be used when available. Bovine graft appears to be an improvement over previous heterologous arterial grafts, and warrants further study in humans.

Summary

Modification of bovine carotid arteries provides an essentially collagen tube without antigenicity which is an improvement over previous heterologous arterial grafts. Review of experimental and clinical results with other heterografts indicates a high failure rate due to thrombosis, rupture, and aneurysm formation. Results with 103 canine preparations using the new heterograft indicated satisfactory function if size disproportion was avoided. This modified bovine heterograft has been used as femoral or popliteal replacement in 12 patients with favorable preliminary results. Follow-up to 35 months has shown no aneurysm formation and patency in seven of the 12 grafts placed.

Addendum

On April 1, 1969, when the galley proofs were reviewed four additional bovine heterografts had been placed as femoro-popliteal shunts with continued patency in each. Thus 11 of the total 16 are patent at present.

Acknowledgment

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Discussion

Dr. Nicholas Kouchoukas (Birmingham): Since a modified heterograft such as Dr. Dale described can be used safely in the cardiovascular system, their ready availability would make them extremely attractive.

At the University of Alabama we have been evaluating another kind of heterograft, the formalin fixed and mounted pig aortic valve. The diseased mitral valves of 13 patients have been replaced with such valves, using a technic described by Dr. Unescu, of Leeds, England.

(Slide) This is the frame designed by Dr. Unescu, in which the trimmed pig aortic valves are mounted.

(Slide) This is one of the grafts sewn onto the frame and fixed in formalin. Formalin fixation allows excellent coaptation of the valve cusps, and may decrease the antigenic potential of such grafts, although definite information in this regard is not available at present.

(Slide) This is the undersurface of the same valve, which faces the left atrial chamber after the valve is sutured into the mitral annulus. A piece of Dacron cloth is used to cover the muscle tissue which overlies the portion of the right coronary cusp in the pig valve.

The 13 patients in this group are a small percentage of the patients who have undergone mitral valve replacement in our hospital. They were carefully selected patients in whom it was felt that long-term anticoagulation would have been particularly disadvantageous. Three of the 13 grafts became incompetent, and were removed to 2½, and 6 months after insertion. Structural defects resulting in prolapse or tears of the cusps were responsible for the incompetence in all three instances. It is possible that such complications can be prevented in the future, as our experience increases. The valves were replaced with Starr-Edwards prostheses, and all patients have been well.

(Slide) This is one of the grafts removed 2½ months after insertion. The incompetence was the result of a tear, as you can see, in the right coronary cusp, due, we believe, to a replacement of a suture in the cusp tissue itself. Excellent healing of the graft and rings to the microannulus was noted in all three valves removed. No thrombus was observed in any of the grafts, nor was there any calcification.

(Slide) This is the ventricular surface of the same valve, showing no thrombus formation, and soft, pliable cusp tissue.

The longest follow-up in the entire group is 6 months. There have been no thromboembolic complications, although long-term anticoagulation therapy has not been employed. There has been no operative mortality.

Certainly longer follow-up is necessary. The early results are encouraging, and we are continuing to use such grafts in selected patients.

Dr. Dennis Rosenberg (New Orleans): Although the main homograft has become the treatment of choice for arterial reconstruction below the inguinal region, and vascular surgeons have lost some of their earlier enthusiasm for plastic prostheses, we have been interested and using and evaluating the modified bovine heterograft, as described by Dr. Dale.

We have used such a graft now in over 40 patients, and have 31 grafts below the inguinal ligament. The others are for aortorenal anastomoses and bypasses, and in the carotid.

Our indications have not only been for salvage alone, but also in patients with cortications severe enough to interfere with the ordinary activities of life and work. We have used these grafts as a primary choice, so that we can compare them with the saphenous vein grafts as used on the other surgical services.

In our experience, the modified bovine heterograft has been an eminently successful and satis-