Histopathologic analysis of endovascular stent grafts from patients with aortic aneurysms: Does healing occur?

Claudie McArthur, MD, Victoria Teodorescu, MD, Leon Eisen, MD, Nicholas Morrissey, MD, Peter Faries, MD, Larry Hollier, MD, and **Michael L. Marin, MD,** *New York, NY*

Background: **Research with animal models has demonstrated tissue healing of endovascular grafts in both native arterial segments and in experimentally created arterial aneurysms. Fundamental to the successful clinical use of endovascular grafts for the treatment of aneurysmal disease is the creation of a permanent hemostatic seal between the graft ends and the arterial wall. Characteristics of this healing process in patients with aneurysmal disease have not been fully studied. In this study, we analyzed the macroscopic and histopathologic changes of the arterial wall after endovascular repair of aortic aneurysms.**

Methods: **Over a 7-year period, 313 patients were treated with endovascular grafts to exclude arterial aneurysms of the thoracic and abdominal aorta. Of these patients, 11 had their endovascular grafts recovered for analysis. Five graft specimens were recovered during subsequent open aortic surgery. Six grafts were recovered at autopsy after the death of the patient of causes unrelated to the patient's endovascular graft. All specimens were fixed in formalin. Histologic analysis included light microscopy with hematoxylin and eosin and trichrome stains. Well-preserved specimens were selected after light microscopic examination and postfixed in 3% buffered glutaraldehyde for electron microscopy. The aortas from autopsy specimens were removed en bloc and fixed in formalin; representative regions of each graft were sectioned for analysis. Adherence of the graft to the vessel wall was categorized as densely adherent or easily separated after graft explantation. Traction applied to the graft-aortic anastomosis was equal to traction generated by suspending a standardized 2-kg weight. Infrarenal graft specimens were obtained with supraceliac aortic clamping, longitudinal aortotomy, and graft sampling before endograft revision.**

Results: **In eight patients, endograft fixation was found to be firmly adherent to the arterial wall. A translucent film of fibrinous material was consistently seen across the entire luminal surface of the endograft. Light and electron microscopy failed to demonstrate an endothelial layer or organized pseudointima at the graft-artery interface.**

Conclusion: **Despite suggestive experimental data regarding endograft healing in animals, minimal graft incorporation was apparent in the stent grafts recovered in this study. A greater emphasis on the construction and mechanism of fixation of endograft attachment systems will be important for long-term device function. (J Vasc Surg 2001;33:733-8.)**

The ability to achieve complete, long-term aneurysm exclusion with protection from rupture is essential to the long-term success of endovascular graft therapy for the treatment of aortic aneurysms. The healing properties of conventionally sutured arterial grafts for the treatment of arterial lesions in human beings have been studied over three decades.[1-4] Prosthetic vascular grafts placed endoluminally have only recently been explored for clinical use. Studies with animal models to evaluate healing related to endovascular grafts have demonstrated some improvement in endothelialization with less intimal hyperplasia, when compared with similar animal models after conventional grafting.[2,5-9] The incorporation and healing of endovascular grafts for the treatment of aortic aneurysms in humans have not been established. Currently, data are limited to the study of a finite number of explanted human grafts, of which there have been a few reports.[10-13] In this study we analyze specimens recovered over a 7-year period from patients who had undergone endoluminal repair of aortic aneurysms.

From the Division of Vascular Surgery, Department of Surgery, Mount Sinai School of Medicine.

Competition of interest: nil.

Presented at the Twenty-eighth Annual Symposium of the Society for Clinical Vascular Surgery, Rancho Mirage, Calif, Mar 15-19, 2000.

Reprint requests: Michael L. Marin, MD, Henry Kaufmann Professor of Vascular Surgery, Department of Surgery, Mount Sinai School of Medicine, One Gustave L Levy Place, New York, NY 10029 (e-mail: michael.marin@mountsinai.org).

0741-5214/2001/$35.00 + 0 **24/6/113980**
doi:10.1067/mva.2001.113980

METHODS

Over a 7-year period, 313 patients were treated with endovascular grafts to exclude arterial aneurysms of the thoracic and abdominal aorta. Of these patients, 11 had their endografts recovered for subsequent analysis. Five graft specimens were obtained after subsequent open aortic surgery for the correction of an endograft-related problem (1 distal endoleak, 2 proximal endoleaks, 1 infected graft, 1 proximal aneurysmal degeneration). The remaining six grafts were obtained at autopsy after the death of the patient of causes unrelated to the patient's endograft procedure (Table I).

All specimens were fixed in formalin. Histologic analysis included light microscopy with hematoxylin and eosin and trichrome stains. Well-preserved specimens were selected after light microscopic examination and postfixed

Table I. Patient information

Patient	*Age (y)/sex*	*Aneurysm size (cm)*	*Graft*	*Construct**	*Time of explant (mo)*	*Recovery method*	*Cause of death/surgery*
1	79/M	7.0	Talent	A	4	Postmortem examination	MI
2	86/M	6.5	Talent	A	18	Postmortem examination	MI
3	83/M	6.5	Megs PTFE	B	8	Graft explant	Graft infection
4	91/M	7.6	Megs PTFE	B	2	Graft explant	Proximal endoleak
5	73/F	6.5	Megs PTFE	B	2	Graft explant	Distal endoleak
6	82/M	7.0	Talent	C	1	Graft explant	Proximal endoleak
7	83/M	6.0	Gore	A	6	Postmortem examination	MI
8	81/M	7.0	Talent	B	4 d	Postmortem examination	MI
9	50/F	5.0	Talent	A	3	Graft exploration	Proximal aneurysm degeneration
10	46/F	5.1†	Talent	A	6 d	Postmortem examination	CVA
11	76/M	6.0	Talent	A	24 d	Postmortem examination	Fungal sepsis

**A* = tube graft, *B* = aorto–uni-iliac graft, *C* = bifurcated graft.
†Traumatic aortic rupture.
CVA, Cerebrovascular accident; *F,* female; *M,* male; *MI,* myocardial infarction; *PTFE,* polytetrafluoroethylene.

Table II. Pathologic condition

Patient	*Gross analysis*	*Microscopic**
1	Densely adherent	N/A
2	Easily removed	N/A
3	Densely adherent	a
4	Easily removed	a
5	Easily removed	b
6	Densely adherent	b
7	Densely adherent	b
8	Densely adherent	b
9	Densely adherent	N/A
10	Densely adherent	a
11	Densely adherent	a

*a = Compacted fibrin over interface; b = scant cellular or fibrous tissue at interface.
N/A, Not applicable.

in 3% buffered glutaraldehyde for electron microscopy. The aortas from autopsy specimens were removed en bloc and fixed in formalin; representative regions of each graft were sectioned for analysis. In our study, adherence of the graft to the vessel wall was categorized as densely adherent or easily separated after graft explantation. Traction applied to the graft-aortic anastomosis was equal to traction generated by suspending a standardized 2-kg weight. Infrarenal graft specimens were obtained at the time of revisional surgery by supraceliac aortic clamping, longitudinal aortotomy, and graft sampling before endograft revision.

RESULTS

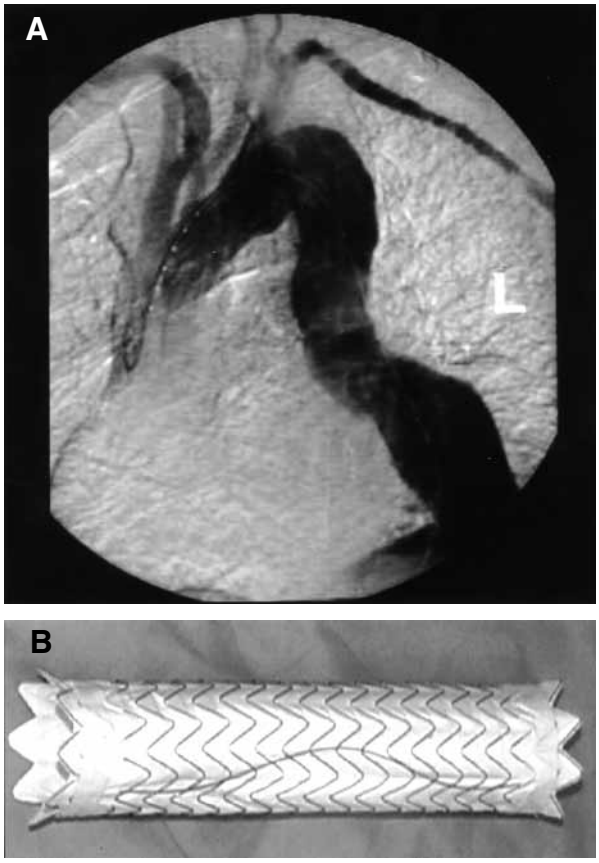
In eight patients, the entire graft was found through traction test to be firmly adherent to the arterial wall at the stent attachment sites. All endografts were loosely adherent to the thrombus within the aneurysmal sac. Inspection of the stent-to-artery interface of explanted endografts consistently demonstrated a translucent film of fragile thrombotic material extending across the artery-graft interface. The aortas from three patients were found to have grafts that were easily separated from the luminal surface at the sites of their stent anastomoses. Two of these patients had clinically apparent endoleaks (Table II). One endoleak was proximal, and the second was a distal endoleak. The apparent effect of both proximal and distal endoleaks was to increase the ease with which the endograft could be separated from the native aorta. Microscopic analysis of the stent-graft interface revealed compacted fibrin along this interface in the first of these patients who had a proximal endoleak and scant cellular or fibrous tissue along the interface in the second patient who had a distal endoleak.

The patient described in case #3 had recurrent infection 7 months after endovascular aortic aneurysm repair. Workup demonstrated a communication between the excluded aneurysm sac and the duodenum. It is unclear whether this communication developed after repair or was present before endovascular grafting. Graft excision and aortic ligation was the therapeutic intervention. Streptococcus grew in the cultures of the graft. The patient subsequently recovered. Gross analysis of the graft-artery interface revealed a stent densely adherent to the arterial wall. Microscopic analysis revealed compacted fibrin over the interface. It remains unclear whether the development of the aortoenteric fistula is a procedure-related problem.

Histopathologic analysis of specimens failed to demonstrate evidence of significant stent graft incorporation at anastomotic sites. Ingrowth of arterial wall cells was not seen. Specimens contained compacted fibrin and organized thrombus with an absence of myointimal cells. One specimen explanted after 6 months demonstrated a small amount of cellular material and fibrin, but was essentially free of organized tissue elements.

DISCUSSION

As the use of endovascular graft therapy for the treatment of aortic aneurysms continues to increase, the extent of healing necessary to achieve long-term fixation and suc-



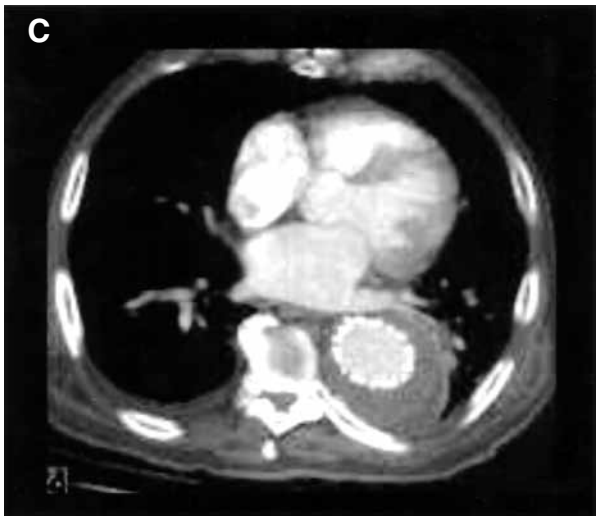


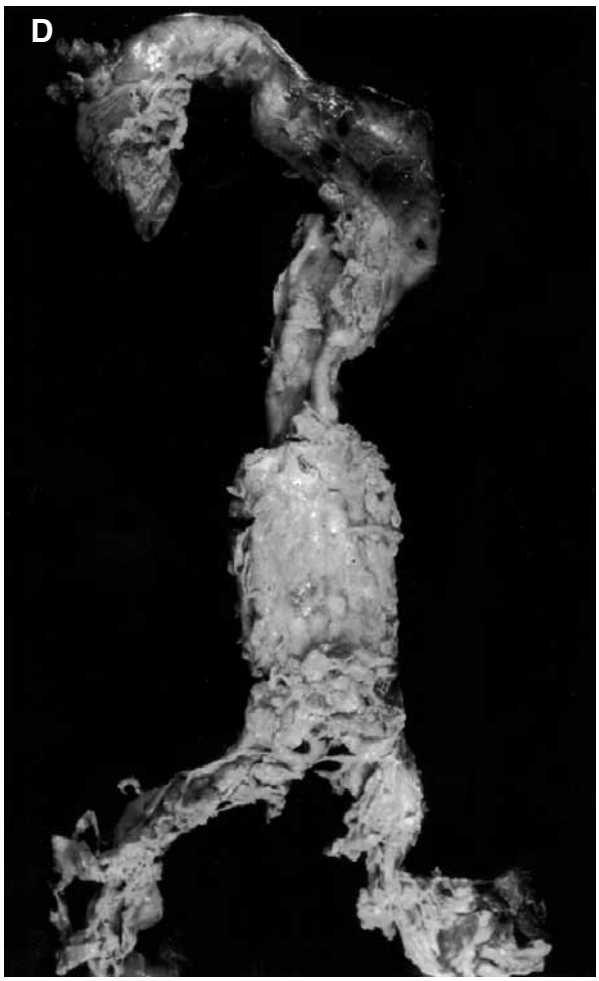
Fig 1. A, Preoperative aortic arch angiogram of an 83-year-old man with isolated descending thoracic aortic aneurysm. **B,** An Excluder device constructed from extended polytetrafluoroethylene (ePTFE) and nitinol metal wire. **C,** A computed tomography scan obtained postoperatively demonstrates successful aneurysm exclusion with contrast confined to endograft.

cess of endoluminally placed grafts will need to be determined. In general, the incorporation of a vascular prosthesis into surrounding tissue and the development of a stable neointima with endothelialization of the luminal surface are thought to be important events for complete graft healing, long-term function, and patency.[11] However, unlike the healing process observed in a variety of animal species, the extent of healing in humans after the placement of conventional grafts is incomplete.[1,5] The ability to fully endothelialize a graft has not been demonstrated, and tissue organization within grafts is generally limited to pannus outgrowth, a hyperplastic reaction from the ends of the host artery immediately adjacent to suture lines, which typically extends for a minimal distance. Most of the flow surface of a vascular graft placed in humans persists as an acellular fibrinous lining.[1] Although the healing of conventionally placed grafts has received considerable attention over the past 30 years, little is known about the healing of endoluminally placed prostheses.

Observations in animal models have revealed that healing occurs around endovascular grafts inserted to treat experimentally created aneurysms.[14,15] Certain animals demonstrate the ability to heal and endothelialize prosthetic grafts placed in extraluminal and endoluminal positions.[1,16,17] Furthermore, studies in animals have demonstrated enhanced graft healing, specifically, improved endothelialization and limited intimal hyperplasia of intraluminally placed grafts when compared with conventionally placed grafts in normal vessels.[5,18] The existence of smooth muscle cells, macrophages, and proliferating cells in a quiescent state is suggestive of complete healing.[5] Some authors have hypothesized that direct endothelial cell contact with the intraluminal graft may be a significant factor contributing to enhanced graft healing seen in experimental endografts.[5] Other theoretical advantages for the healing of endografts include improved hemodynamics related to an "in-line" graft configuration and less surrounding tissue trauma resulting from tissue dissection and the use of a sutured anastomosis.[5,8,9,18] Although the evidence regarding incorporation and endothelialization of intraluminally placed grafts in animals suggests that successful aneurysm exclusion and graft healing may be achieved, the relationship between the healing of endografts in animals and humans is limited.

Controversy exists regarding the effect of graft materials themselves on anastomotic healing.[19,20] It has been proposed that the healing of vascular anastomoses performed with materials such as polyurethane is superior to that achieved with polytetrafluoroethylene (PTFE).[19] The current study does not provide any support for the superiority of one graft material over another.

In our study of 11 explanted endovascular grafts that



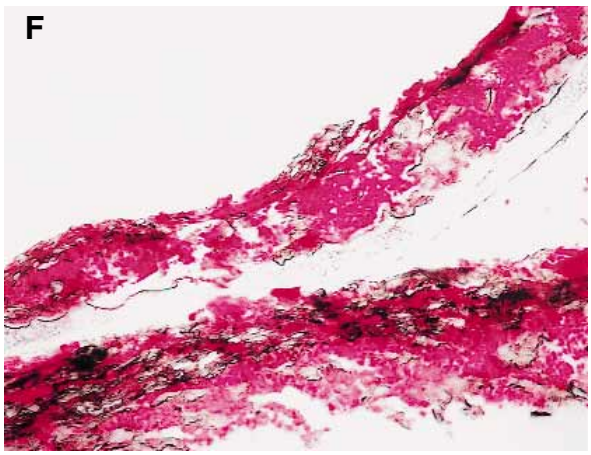


Fig 1, contd. D, The patient died 6 months later of myocardial infarction. During autopsy, intact aorta was removed en bloc. **E,** A window was created at site of proximal graft fixation. Note absence of intimal pannus across stent to artery interface at proximal attachment site. **F,** Light micrograph of endograft of cross section at level of proximal anastomosis. Two separate layers of ePTFE consistent with fabrication of the Excluder endograft can be identified. Compacted fibrin on luminal surface comprises the neointima.

had been used for the treatment of aortic aneurysms in humans, minimal tissue incorporation of the grafts was noted despite the apparent tight fixation of the grafts. Only eight of 11 grafts were found to be densely adherent to the arterial wall, but the characteristic pannus of endothelial and smooth muscle cells that is seen at the graft-artery interface of conventionally placed grafts was not identified in any specimen. Compacted fibrin was noted over the interface of four of eight studied specimens, whereas the remaining specimens were entirely devoid of any cellular or fibrous tissue. Overall, minimal graft incorporation was apparent in the grafts in this study. These findings strongly support the fact that the attachment system alone from an endovascular graft device is pivotal to graft fixation. It may not be accurate to assume that graft healing and incorporation will occur to provide long-term fixation.

The presence of a limited tissue-healing response at the proximal and distal graft-to-artery interface in these specimens is different from the healing response observed in animal studies. However, the time of explant of these specimens ranged from 4 days to 18 months, with half of the specimens explanted within 3 months of surgery. As a result of the relatively early explantation, the degree of anastomotic healing and graft incorporation may be diminished. Our previous studies in humans after endovascular therapy for aortoiliac occlusive disease demonstrated that a true neointima at the graft-artery interface was consistently present after 3 months, with some endothelialization evident by 6 weeks.[11] The specimens examined may

have been explanted too early to make significant conclusions regarding the lack of neointima formation. Future study is required to further substantiate our findings.

Furthermore, several patients underwent explantation of their grafts during surgery for the correction of endoleaks. Anastomotic healing was observed to be diminished in the cohort of patients exhibiting clinically significant endoleaks (patients 4, 5, and 6). Diminished anastomotic healing was also observed in the patient who experienced graft infection. This evidence would support the premise that graft healing mandates contact between the graft material and the arterial wall to achieve healing.[10] Contact between graft and artery after endovascular aortic aneurysm repair may be further inhibited by the presence of thrombus between the attachment system and the artery wall.

The study of endovascular graft healing in humans has been limited to the analysis of explanted endovascular grafts, of which there are a small number of reports. Our group previously reported histologic evidence of the healing of seven explanted endovascular grafts placed for the treatment of aortoiliac occlusive disease. In that report, we found that by 6 weeks, perianastomotic endothelialization was seen progressing into the graft 1 to 3 cm from the anastomosis. The degree of foreign body inflammatory reaction was based on the depth of placement of the graft within the arterial wall. Islands of FVIII–positive cells were identified as far as 8 cm from the graft-artery interface, supporting the possibility of endothelial cell seeding or ingrowth of endothelium through the interstices of the graft as has been reported in animal models.[21-24] The difference in the local environment between endografts used for aneurysm repair and those used in the presence of aortoiliac disease may account for differences observed in our studies.

Despite this encouraging evidence of healing in patients treated for aortoiliac occlusive disease, the same extent of healing has not been demonstrated in patients undergoing endovascular repair of aortic aneurysms. One investigative group reported on one explanted graft 67 days after implantation for the treatment of an iliac artery aneurysm.[12] The results were similar to those observed in this study. Encasement of the graft with dense collagenous tissue and an organizing hypocellular fibrinous intraluminal layer devoid of endothelial cells was observed. Although the graft seems well incorporated, the ultimate significance of the lack of cellular healing is uncertain.

A recent report of two explanted grafts used for the treatment of aortic aneurysms demonstrated the presence of neointima with smooth muscle cells and endothelium at the proximal, but not the distal, anastomosis after 42 and 21 days of implantation.[10] Another investigative group reported on a graft used for the treatment of an abdominal aortic aneurysm that was explanted after 7 months.[13] Whereas the presence of a neointima and some endothelialization at the proximal anastomosis was identified, the distal anastomosis was covered by a fibrinous membrane only. The aneurysm had been completely excluded before the patient's death of unrelated causes, and it is not possible to predict whether the extent of graft incorporation observed would have been sufficient for long-term aneurysm exclusion.

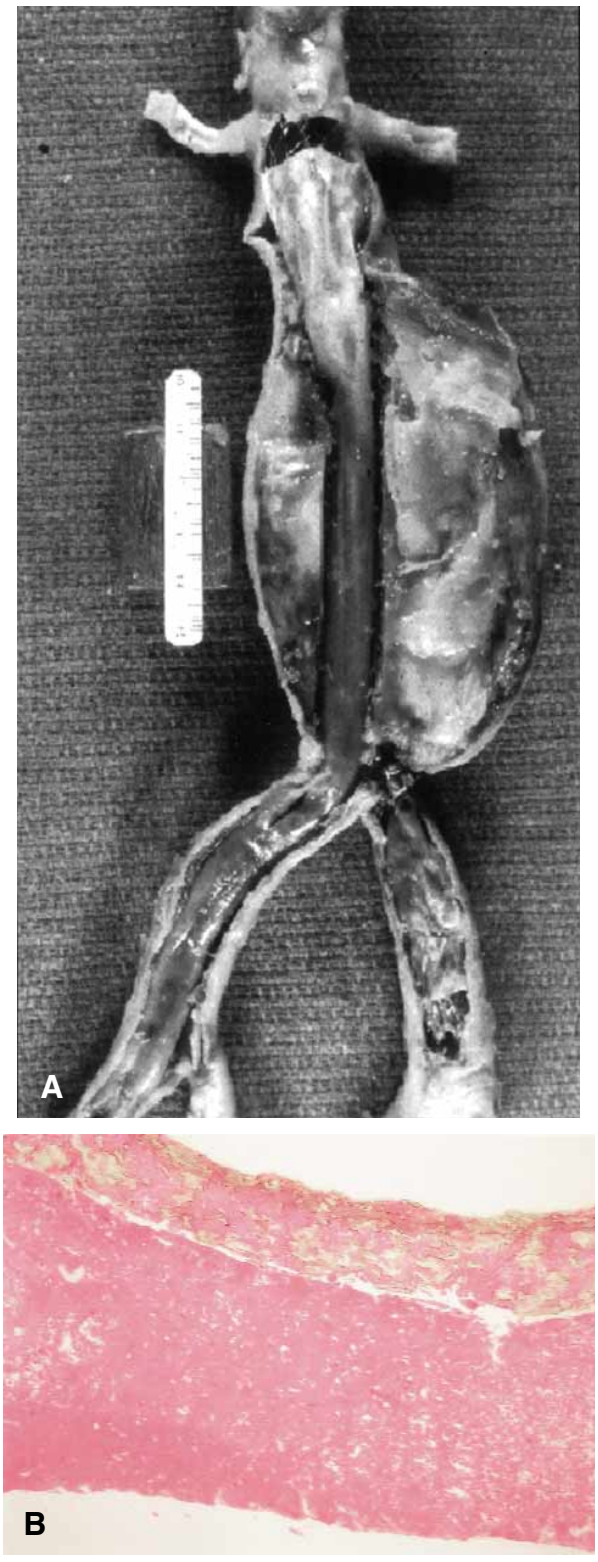


Fig 2. A, En bloc aorta specimen from an 81-year-old man after endovascular repair of infrarenal abdominal aortic aneurysm. Aorto-uni-iliac device is seen within explanted aorta after successful aneurysm exclusion. He died on fourth postoperative day of acute myocardial infarction. **B,** Light micrograph of section through mid-portion of graft contains thick uniform layer of fibrin.

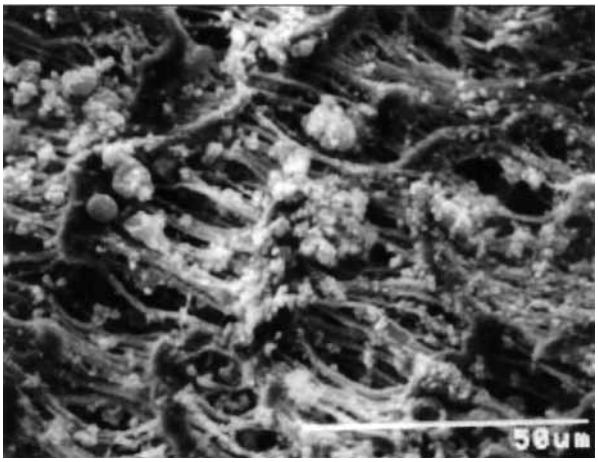


Fig 3. Electron microscopy performed on aortic endograft explanted 2 months after aortic endografting. Nodal pattern of ePTFE graft material is seen with adherent platelets and fibrin, but no organized cellular layer.

It seems likely that limited stent-artery incorporation occurs in patients after endovascular aortic aneurysm repair. These findings should be incorporated into new and future stent graft designs, in which the enhanced role of the attachment apparatus to long-term stent graft repair is recognized.

REFERENCES

1. Berger K, Sauvage LR, Rao AM, Wood SJ. Healing of arterial prostheses in man: its incompleteness. Ann Surg 1972;175:118-27.
2. Ohki T, Marin ML, Veith FJ, Yuan JG, Ohki M, Soundararajan K, et al. Anastomotic intimal hyperplasia: a comparison between conventional and endovascular stent graft techniques. J Surg Res 1997;69:255-67.
3. Edwards WH, Mulherin JL Jr. The role of graft material in femorotibial bypass grafts. Ann Surg 1980;191:721-6.
4. Echave V, Koornick AR, Haimov M, Jacobson JH II. Intimal hyperplasia as a complication of the use of the polytetrafluorethylene graft for femoral-popliteal bypass. Surgery 1979;86:791-8.
5. Ombrellaro MP, Stevens SL, Kerstetter K, Freeman MB, Goldman MH. Healing characteristics of intraarterial stented grafts: effect of intraluminal position on prosthetic graft healing. Surgery 1996;120:60-70.
6. Yee DC, Williams SK, Salzmann DL, Pond GD, Patula V, Berman SS, et al. Stent versus endovascular graft healing characteristics in the porcine iliac artery. J Vasc Interv Radiol 1998;9:609-17.
7. Chalmers RTA, Hoballah JJ, Sharp WJ, Kresowik TF, Corson JD. Effect of an endovascular stent on healing of an end-to-end polytetrafluorethylene-artery anastomosis in a canine model. Br J Surg 1994;81:1443-7.
8. Ombrellaro M, Stevens S, Schaeffer D, Freeman MB, Sciarrotta J, Carroll R, et al. The role of platelet-derived growth factor in intraluminal stented graft healing. J Am Coll Surg 1997;184:49-57.
9. Ombrellaro M, Stevens S, Sciarrotta J, Schaeffer DO, Freeman MB, Goldman MH. Effect of balloon-expandable and self-expanding stent fixation on endoluminal polytetrafluoroethylene graft healing. Am J Surg 1997;173:461-6.
10. Shin CK, Rodino W, Kirwin JD, Ramirez JA, Wisselink W, Papierman G, et al. Histology and electron microscopy of explanted bifurcated endovascular aortic grafts: evidence of early incorporation and healing. J Endovasc Surg 1999;6:246-50.
11. Marin ML, Veith FJ, Cynamon J, Sanchez LA, Bakal CW, Suggs WD, et al. Human transluminally placed endovascular stented grafts: Preliminary histopathologic analysis of healing grafts in aortoiliac and femoral artery occlusive disease. J Vasc Surg 1995;21:595-604.
12. White RA, Donayre CE, deVirgilio C, Weinstein E, Tio F, Kopchok G. Deployment technique and histopathological evaluation of an endoluminal vascular prosthesis used to repair an iliac artery aneurysm. J Endovasc Surg 1996;3:262-9.
13. McGahan TJ, Berry GA, McGahan SL, White GH, Yu W, May J. Results of autopsy 7 months after successful endoluminal treatment of an infrarenal abdominal aortic aneurysm. J Endovasc Surg 1995;2:348-55.
14. Laborde JC, Parodi JC, Clem MF, Tio FO, Barone HD, Rivera FJ, et al. Intraluminal bypass of abdominal aortic aneurysm: feasibility study. Radiology 1992;184:185-90.
15. Parodi JC, Palmaz JC, Barone HD. Transfemoral intraluminal graft implantation for abdominal aortic aneurysms. Ann Vasc Surg 1991;5:491-9.
16. Clowes AW, Kohler T. Graft endothelialization: the role of angiogenic mechanisms. J Vasc Surg 1991;13:734-6.
17. Golden MA, Hanson SR, Kirkman TR, Schneider PA, Clowes AW. Healing of polytetrafluoroethylene arterial grafts is influenced by graft porosity. J Vasc Surg 1990;11:838-45.
18. Ombrellaro M, Stevens S, Freeman M, Schaeffer D, Freeman MB, Goldman MH. Effect of intra-arterial environment on endothelialization and basement membrane organization in polytetrafluoroethylene grafts. Am J Surg 1977;174:29-32.
19. Marois Y, Paris E, Zhang Z, Doillon CJ, King MW, Guidoin RG. Vascugraft microporous polyesterurethane arterial prosthesis as a thoraco-abdominal bypass in dogs. Biomaterials 1996;13:1289-300.
20. Contreras MA, Quist WC, Logerfo FW. Effect of porosity on small-diameter vascular graft healing. Microsurgery 2000;20:15-21.
21. Clowes A, Kiekman T, Reidy M. Mechanisms of arterial graft healing: rapid transmural capillary ingrowth provides a source of intimal endothelium and smooth muscle in porous PTFE prosthesis. Am J Pathol 1986;123:220-30.
22. Harker LA, Slichter SJ, Sauvage LR. Platelet consumption by arterial prostheses: the effects of endothelialization and pharmacologic inhibition of platelet function. Ann Surg 1977;186:594-601.
23. Kusaba A, Fischer CR, Matulewski TJ, Matsumoto T. Experimental study of the influence of porosity on development of neointima in Gore-Tex grafts. Am Surg 1981:47:347-54.
24. Herring M, Baughman S, Glover J, Keseler K, Jesseph J, Campbell Jet al. Endothelial seeding of Dacron and polytetrafluoroethylene grafts: the cellular events of healing. Surgery 1985;96:745-54.

Submitted Mar 30, 2000; accepted Nov 22, 2000.