

Histology After Stenting of Human Saphenous Vein Bypass Grafts: Observations From Surgically Excised Grafts 3 to 320 Days After Stent Implantation

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Objectives. To gain insight into the mechanism of stenting in humans and its short- and long-term implications, we studied the vascular wall of saphenous vein aortocoronary bypass grafts after implantation of the Wallstent.

Background. The implantation of a stent in aortocoronary bypass grafts may provide an alternative solution for revascularization in patients who are poor candidates for reoperation. Because human histopathologic findings after stenting with the Wallstent have not previously been described in detail, we examined graft segments that were surgically retrieved from 10 patients (21 stents) at 3 days to 10 months after implantation of the stent.

Methods. The grafts were examined by a combination of the following techniques: light microscopy, immunocytochemistry and both scanning and transmission electron microscopy.

Results. Early observations revealed that large amounts of

platelets and leukocytes adhered to the stent wires during the first few days. At 3 months, the wires were embedded in a layered new intimal thickening, consisting of smooth muscle cells in a collagenous matrix. In addition, foam cells were abundant near the wires. Extracellular lipids and cholesterol crystals were found after 6 months. Smooth muscle cells and extracellular matrix formed the predominant component of restenosis. This new intimal thickening was lined with endothelium, in some cases showing defect intercellular junctions and abnormal adherence of leukocytes and platelets as late as 10 months after implantation.

Conclusions. This type of stent is potentially thrombogenic and seems to be associated with extracellular lipid accumulation in venous aortocoronary bypass grafts.

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Coronary artery bypass graft surgery using autologous veins is a common treatment for atherosclerotic coronary artery disease. However, 12% of patients with grafts require a second revascularization procedure after 5 years (1), that in the case of repeat bypass surgery, is associated with increased morbidity and mortality and inferior long-term patency with respect to results of the first procedure (2). Moreover, a significant number of patients are poor candidates for a repeat operation (3). Percutaneous transluminal coronary angioplasty may provide an alternative to surgery for many of these patients. Although the initial success rate of this procedure is high (90%), the restenosis rate in bypass grafts is also high. Only 41% of patients are alive and

event-free after 2 years (4). Furthermore, not all patients not eligible for surgical treatment, are candidates for balloon angioplasty (3).

The implantation of endovascular prostheses, also called stents, seems a promising new technique (5-8). The Thoraxcenter experience with the self-expanding Wallstent implanted in stenosed aortocoronary vein grafts now includes 69 patients with a total of 136 stents. Although the number of early complications in this high risk group of patients is considerable, the 34% rate of late restenosis compares favorably with that associated with angioplasty alone (8). From 10 patients with either early or late complications, we were able to obtain stented graft segments for histopathologic study that would provide a better understanding of the mechanism of stenting, as well as the short- and long-term effects of the procedure on diseased human vein grafts.

Methods

Patient characteristics. Between 1988 and 1990, a total of 136 stents (Wallstent) were implanted in aortocoronary vein grafts in 69 patients at the Thoraxcenter. All patients had severe symptoms and were poor candidates for repeat surgery or conventional balloon angioplasty for reasons such as

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Table 1. Characteristics of 10 Patients With Histopathologically Studied Stents

Pt No.	No. of Stents	Pre-PTCA	Stent Diameter (mm)	Duration of Implant (days)	Graft Age (yr)	Implant Reason	Retrieval Reason
1	3	Yes	4/4.5	3	10	UAP; primary; rescue	Bleeding
2	2/2	Yes	3.5/4	4/117	4	UAP; restenosis; elective	Occlusion
3	2	Yes	5.5/6	10	10	UAP; total occlusion; primary; elective	Occlusion
4	2	Yes	3.5	14	4	UAP; total occlusion (MI); primary; elective	Occlusion
5	1	?	?	85	8	Angina; rescue	Angina (other stenosis)
6	3	No	4	105	0.6	UAP; primary; elective	Restenosis
7	1/1	No/Yes	4.5/4.5	106/320	12	Primary/restenosis/both elective	Restenosis of primary lesion
8	2	Yes	5	184	6	Restenosis; elective	Restenosis
9	2	Yes	4	189	3	Primary stenosis	Restenosis
10	1	Yes	4.5	205	5	Restenosis; elective	Stenosis of stent ends

MI = myocardial infarction; Pt = patient; Pre-PTCA = angioplasty before stent implantation; UAP = unstable angina pectoris.

graft age (mean 83 months), lesion morphology or native coronary artery anatomy (3). Nine patients described here were from this group. One additional patient received a Gianturco-Roubin stent at the University of Michigan, Ann Arbor.

Graft segments were excised between 3 and 320 days after stent implantation during surgery for replacement of the stented bypass grafts (Table 1) and underwent pathologic analysis as approved by the Internal Research Board at the Thoraxcenter. Thrombosis within the stented segments or the need to interrupt aggressive anticoagulant therapy was an indication for surgical retrieval within the 1st 2 weeks. Restenosis within or adjacent to the stented segments was the indication for late reoperation.

Description of the stent. Wallstent endovascular prostheses (Schneider Europe AG) and the method of implantation have been described in detail (5). Briefly, the stent is a self-expanding, multifilament, woven mesh, constructed of 18 to 20 stainless steel wire filaments, each 70- to 90- μ m wide. It is constrained in an elongated fashion on a delivery catheter covered with a removable plastic sleeve. The stent mounted on the delivery system is brought into place by using a standard over the wire technique. By withdrawing the sleeve, the stent is released and anchors itself against the vessel wall in an attempt to regain its original diameter.

Histopathologic analysis. The main histologic features of the vein grafts are summarized in Table 2. The surgically retrieved specimens were processed for light microscopy (all cases), immunocytochemistry (three cases), electron microscopy (seven cases) or a combination of these techniques. The grafts were briefly rinsed in 0.96% saline solution. For light microscopy and immunocytochemistry, the material was fixed in a phosphate-buffered solution containing 4% paraformaldehyde for ≥ 48 h. For electron microscopy, the material was fixed in a phosphate-buffered solution containing 4% paraformaldehyde and 1% glutaraldehyde for ≥ 48 h. Glutaraldehyde makes the specimen suitable for

electron microscopy but renders it useless for immunocytochemical analysis.

After fixation, several 3- to 4-mm wide transverse sections were made, either of the whole vessel or, if electron microscopy was performed, from longitudinally cut vessels. The metal stent wires were removed by carefully pulling them out under a dissection microscope with a pair of fine tweezers. No fixed protocol was followed for studying the material.

Light microscopy. Paraffin-embedded tissue was stained with hematoxylin-azophloxine as a routine stain. Resorcin-fuchsin was used as an elastin stain; Goldner trichrome was used as a connective tissue stain. For Epon-embedded tissue, toluidine blue was used.

Immunocytochemical analysis. After rehydration, the sections were exposed to antibodies against smooth muscle cell-specific α -actin (mouse monoclonal antibody, Enzo Diagnostics), vimentin (rabbit polyclonal antibody, Eurodiagnostics) and desmin (mouse monoclonal antibody, Sanbio). Ulex europaeus (UEA-I, DAKO) was used as an endothelial cell marker. As a detecting reagent, horse radish peroxidase-labeled rabbit antimouse or goat antirabbit antibodies were used. Mayers hematoxylin was used as a counter stain.

Electron microscopy. After fixation, the material was rinsed and stored overnight in 0.1 mmol/liter of cacodylate buffer (pH 7.3). Postfixation was performed with a mixture of 50 mmol of $K_3[Fe(CN)_6]$ and 1% OsO_4 in 0.1 mmol/liter of cacodylate buffer (pH 7.2) for ≥ 4 to 6 h. After rinsing the specimen in 0.1 mmol/liter of cacodylate buffer, the material was dehydrated in a graded ethanol series.

For scanning electron microscopy, the specimen was critical point-dried in liquid carbon dioxide, mounted on a specimen table and sputter-coated with gold before examination in a Jeol-JSM-25 or 5200 scanning electron microscope.

For transmission electron microscopy, the material was

Table 2. Angiographic Data, Cholesterol Levels at the Time of Stent Implantation and Histologic Findings in the 10 Study Patients

Pt No.	QCA Pre-, Post-Stent and Follow-Up			Cholesterol Levels (mmol/liter)	Main Histologic Features
	Reference (mm)	Stent (mm)	MLD (mm)		
1	Rescue	No data	Rescue	4.8	Platelet and leukocyte adhesion to the stent
2	2.5 → 3.1 → 2.5 3.4 → 4.8 → 2.5	2.8 → 2.3	0.7 → 1.8 → 0.8 1.2 → 2.7 → 0.8	No data	Fibrous vein graft, thrombus
3	4.5 → 4.3 → 0 4.1 → 4.3 → 0	3.4 → 0 3.4 → 0	1.4 → 2.9 → 0 1.4 → 2.9 → 0	7.8; hypertriglyceridemia	Thrombus on atheromatous debris
4	0.0 → 2.7 → oc	2.6 → 0	0.0 → 2.4 → 0	4.5	Occlusive clot on atheromatous debris
5	No data	No data	No data	No data	Fibrous vein graft; limited new intimal thickening barely covering the stent; foam cells near stent wire
6	2.5 → 3.3 → 3.0 2.5 → 3.3 → 3.6 2.5 → 3.3 → 3.6	? → 2.7 ? → 2.7 ? → 2.7	1.2 → 2.4 → 2.2 1.2 → 2.4 → 1.5 1.2 → 2.4 → 1.5	5.1	Mostly limited new intimal thickening; fibrin remnants near stent wire
7	3.7 → 3.7 → 3.1 4.2 → 4.3 → 4.5	3.1 → 1.6 3.8 → 4.2	1.6 → 2.4 → 1.2 1.9 → 3.7 → 4.2	10.8	Vein graft with fibrous preexisting intimal thickening; new intimal thickening contains foam cells on top of stent wire; narrowing consists of smooth muscle cells and extracellular matrix
8	3.8 → 3.6 → 2.8 4.0 → 4.7 → 2.2	3.5 → 2.0 3.6 → 2.8	1.5 → 2.9 → 1.2 1.5 → 2.9 → 1.6	9.3	Fibrous vein graft; atheromatous plaque in new intimal thickening above the stent
9	2.8 → 2.8 → 3.1 2.7 → 3.2 → 3.2	2.6 → 2.7 3.2 → 2.4	0.8 → 2.1 → 1.2 0.8 → 2.7 → 0.4	No data	New intimal thickening consists of smooth muscle cells and extracellular matrix; fibrin near stent wires; recent thrombus
10	3.2 → 3.4 → 2.5	3.2 → 2.4	1.6 → 2.8 → 1.0	No data	Fibrous vein graft, atheromatous plaque in new intimal thickening above the stent; fibrin near stent wires

MLD = minimal lumen diameter; oc = occluded; Post = after; Pre = before; Reference = reference diameter calculated by the Quantitative Coronary Angiography (QCA) system; Stent = mean lumen diameter.

embedded in Epon. Ultrathin sections were cut with a diamond knife and collected on copper grids. Sections were stained with uranyl acetate and Reynold's lead citrate. Examination was performed in a Philips EM400.

Results

Histologic findings 3 to 14 days after stent implantation.

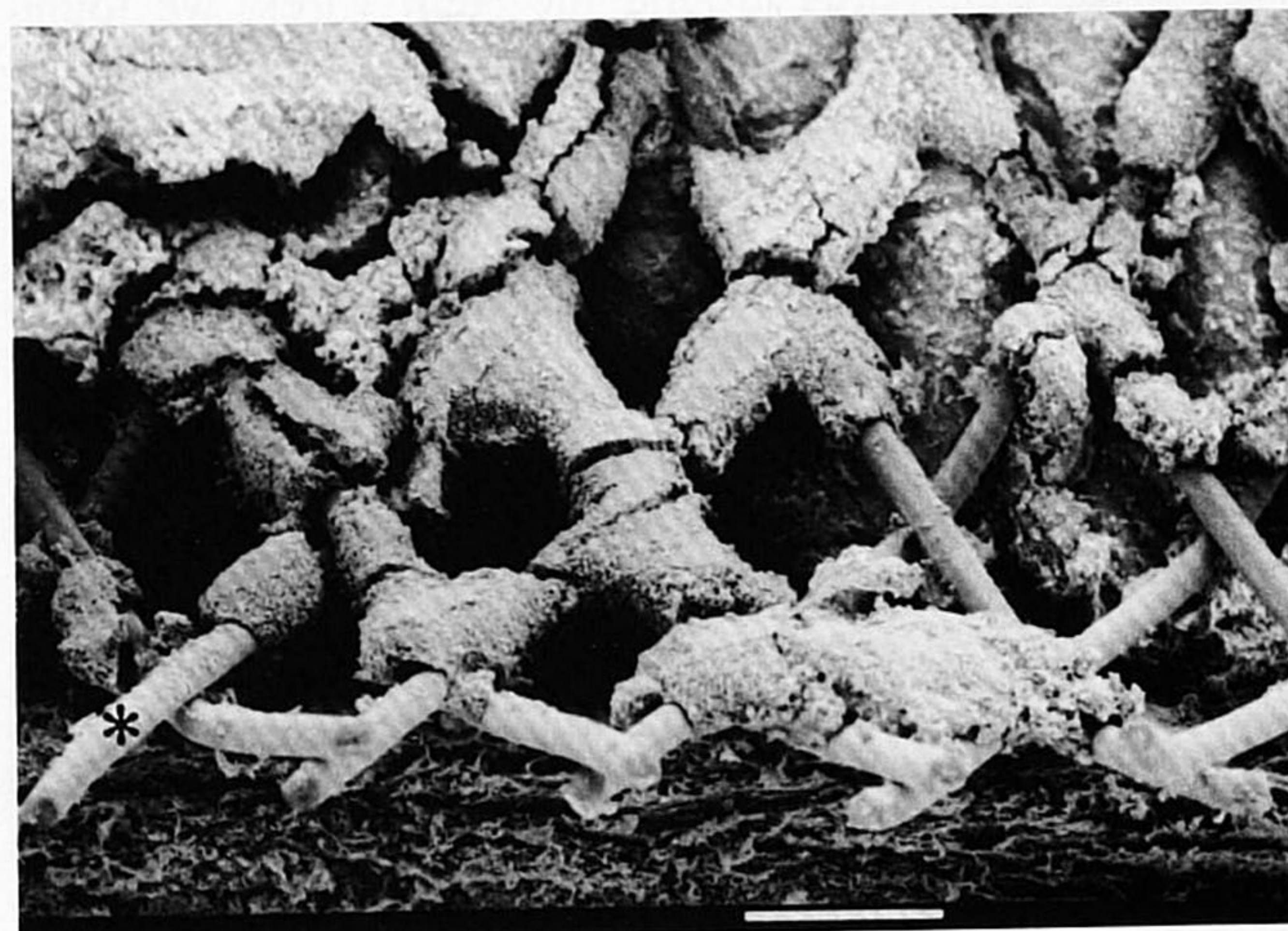
The earliest observation was 3 days after implantation of two stents <1 in. (<2.54 cm) apart, that were bridged several hours later by a third stent after ST segment elevation occurred and angiography revealed haziness (Tables 1 and 2, Fig. 1). Macroscopic examination showed no thrombotic mass, but scanning electron microscopy revealed significant amounts of leukocytes, platelets and fibrin adherent to the wires.

In Patients 2 to 4 (Tables 1 and 2), the grafts were resected because of acute thrombotic complications in the stented segments. Light microscopic analysis of the material from Patient 3 revealed that the proximal, nonstented graft segment (Fig. 2A) contained a preexistent circumferential layer resembling foam cells. However, the mechanical effect of stenting was clearly demonstrated in the adjacent stented segment (Fig. 2B), where the lumen diameter was enlarged. The stented segment further downstream (Fig. 2C) contained a classic atherosclerotic plaque with necrotic, lipid-rich cholesterol crystals, containing a core and a thin fibrous cap. This cap had been ruptured either during angioplasty or the

stenting procedure (although a mechanical artifact cannot be excluded). The stent wires, however, neatly tacked the intimal flap against the vessel wall.

Histologic findings 3 months after stent implantation. In Patient 5 (Tables 1 and 2), scanning electron microscopy of the stent revealed complete covering with polygonal endo-

Figure 1. Patient 1. Scanning electron microscopy 3 days after stent implantation. Large deposits of leukocytes and platelets are found adherent to the wire mesh, especially at branching points. In some places, the vessel wall protruded through the wires into the lumen. *Indicates the stent wire. Bar = 500 μ m.



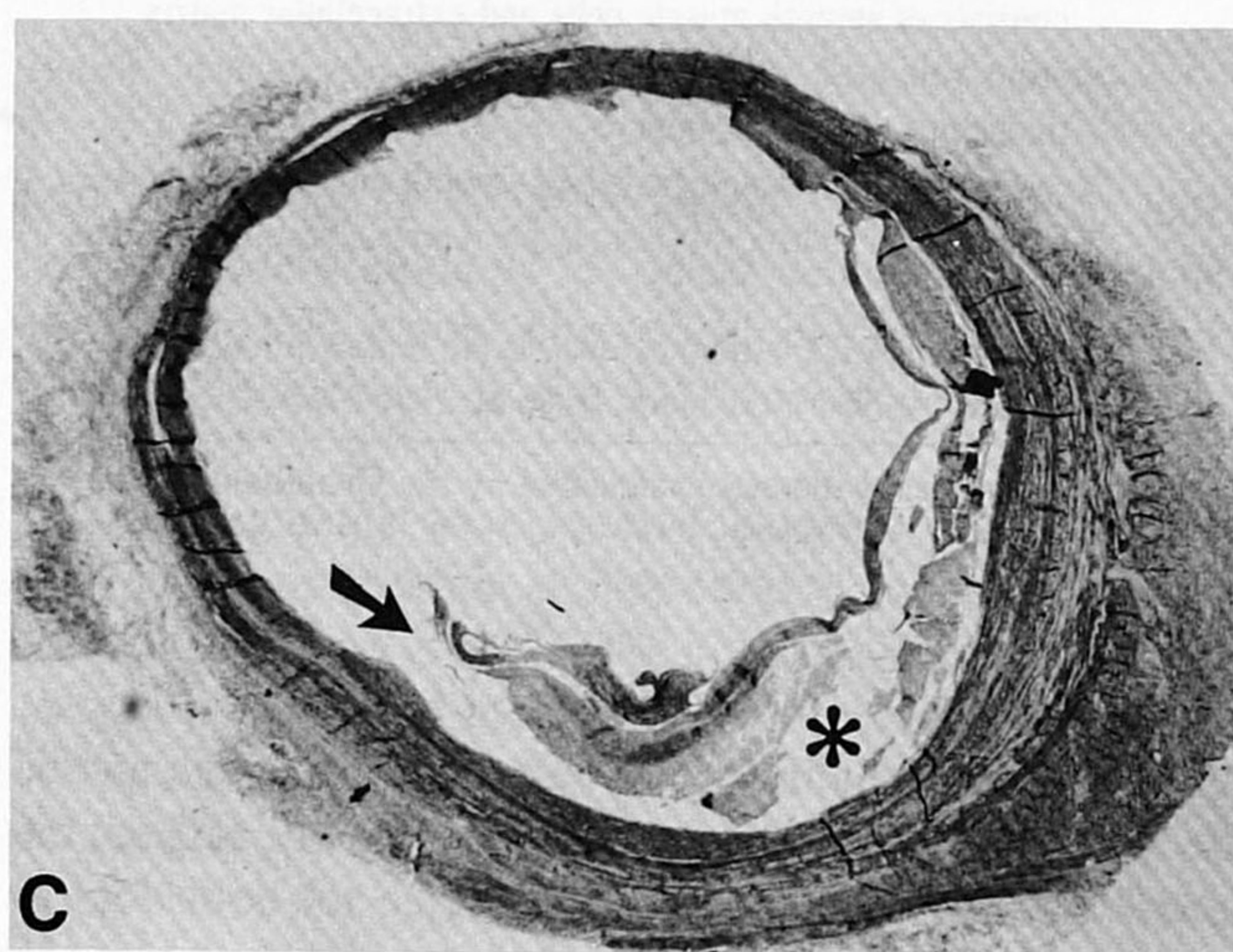
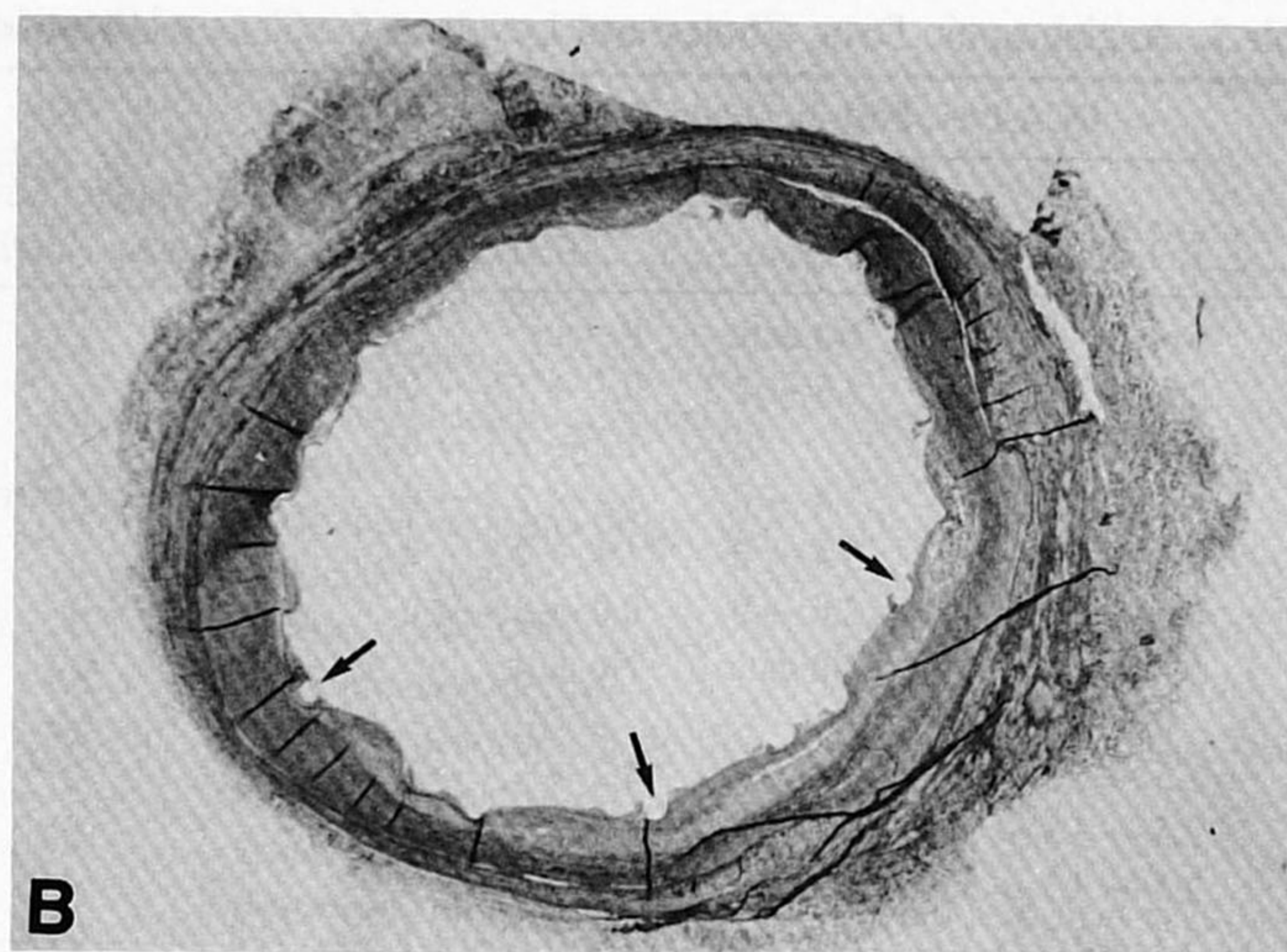
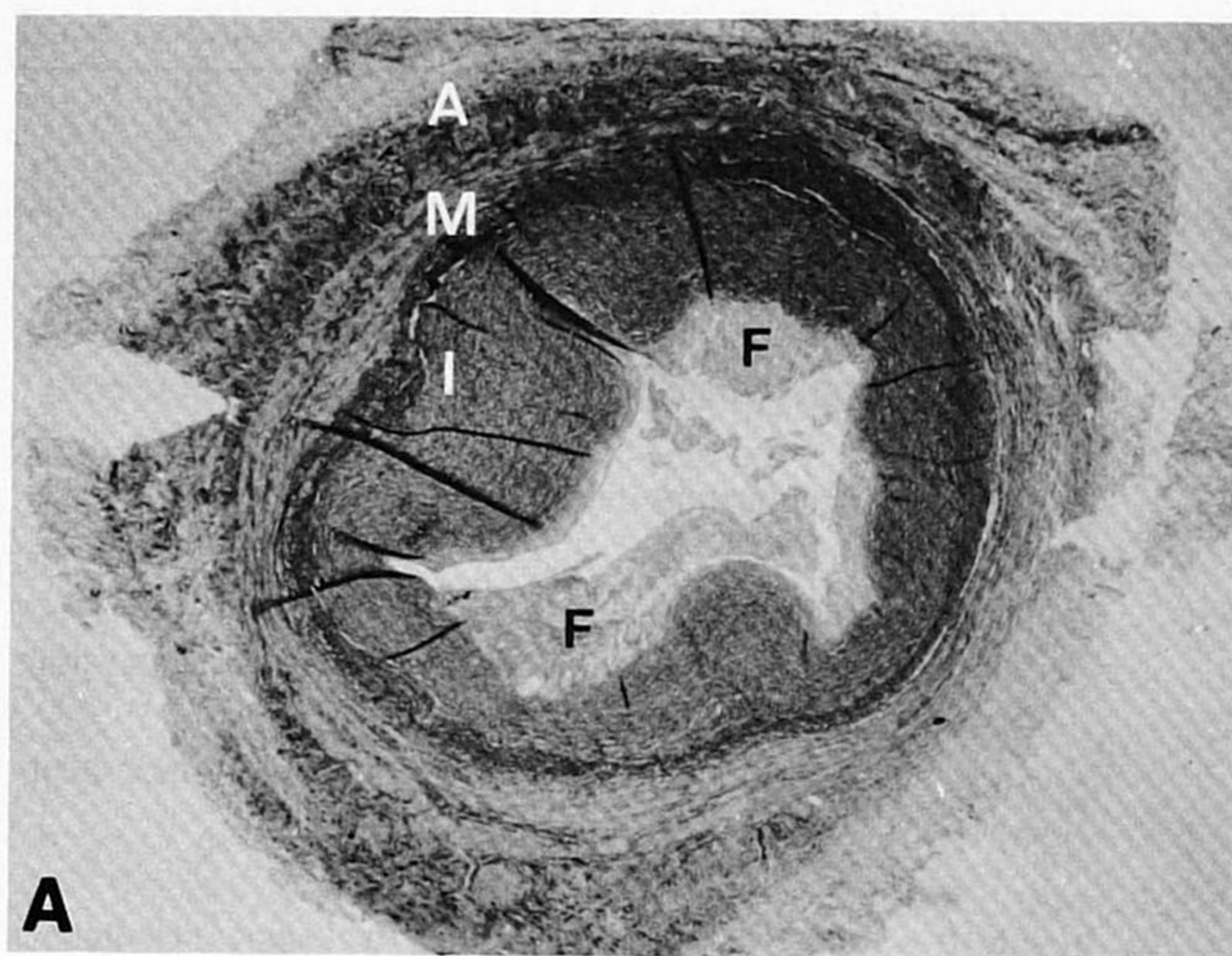


Figure 2. Patient 3. **A**, Light microscopy of the 10-year old graft adjacent to the stented segment, showing a superficial plaque consisting of foam cells (F) without the cover of a fibrous cap. The changes represent typical findings in old vein grafts. A = adventitia; I = intima; M = media. **B**, A few millimeters downstream from A. The dilating effect of angioplasty and stent implantation is obvious. Arrows point at some of the voids that remain after removal of the stent wires. **C**, In the middle of the stent at the site of a classic atheromatous plaque (*), the fibrous cap seems to have ruptured, causing a dissection (arrow) between the fibrous intimal plaque of the preexisting intimal thickening and the subjacent media. The stent wires neatly pushed the intimal flap against the wall, which came loose when the stent wires were removed. **A to C**, elastin stain, final magnification $\times 20$, reduced by 48%.

thelial-like cells and some single missing cells. Some areas exhibited loose junctions between cells, as well as a ruffled surface (Fig. 3A). Leukocytes were also found adhering to the intact endothelium, a feature considered abnormal for mature endothelium. Light microscopy revealed a new intimal thickening, consisting of several layers of smooth muscle cells within the stented grafts.

In a triangular area around the stent wires, we found various signs of the thrombotic event after stent implantation, consisting of fibrin (hematoxylin and azophloxin stain, not shown) in Patients 5 and 6 (Tables 1 and 2, Fig. 3B and C and 4) and abundant foam cells in Patient 7 (Fig. 3B and 5A). In this last patient (Case 7, Tables 1 and 2), the segment with the narrowest residual lumen contained a recent mural thrombus (Fig. 5B).

Histology 6 to 10 months after stent implantation. The second, nonrestenosed, stented graft segment in Patient 7 (Tables 1 and 2) resected 10 months after stent implantation was patent. Scanning electron microscopy revealed complete covering with endothelial cells, some with a protuberant appearance; leukocytes were also seen adhering in large numbers. Histopathology revealed a new intimal thickening

similar to the nonstenosed normal areas in the earlier cases. Foam cells were found scattered in this layer but were abundant around the stent wires.

Patient 8 (Tables 1 and 2) had two closely situated stents resected 6 months after implantation. Restenosis had developed in the proximal end of both stents. Although the endothelial lining seemed intact, transmission electron microscopy showed focal leukocyte infiltration. The new intimal thickening consisted of smooth muscle cell-like cells that exhibited an abundance of cell organelles and bundles of myofilaments in a collagenous matrix. In addition to foam cells around the stent wires, we also observed extracellular lipids and cholesterol crystals similar to a classic atherosclerotic plaque.

Histologic examination of the tissue within two restenosed stents in Patient 9 (Tables 1 and 2) revealed dendritic-like cells within an abundant collagen-rich matrix. Fibrin deposits were found near the stent wires and a small recent mural thrombus was found at the lumen surface.

In Patient 10 (Tables 1 and 2), the graft was resected for recurrence of angina pectoris 7 months after stenting. Two months earlier, angioplasty was performed for restenosis of

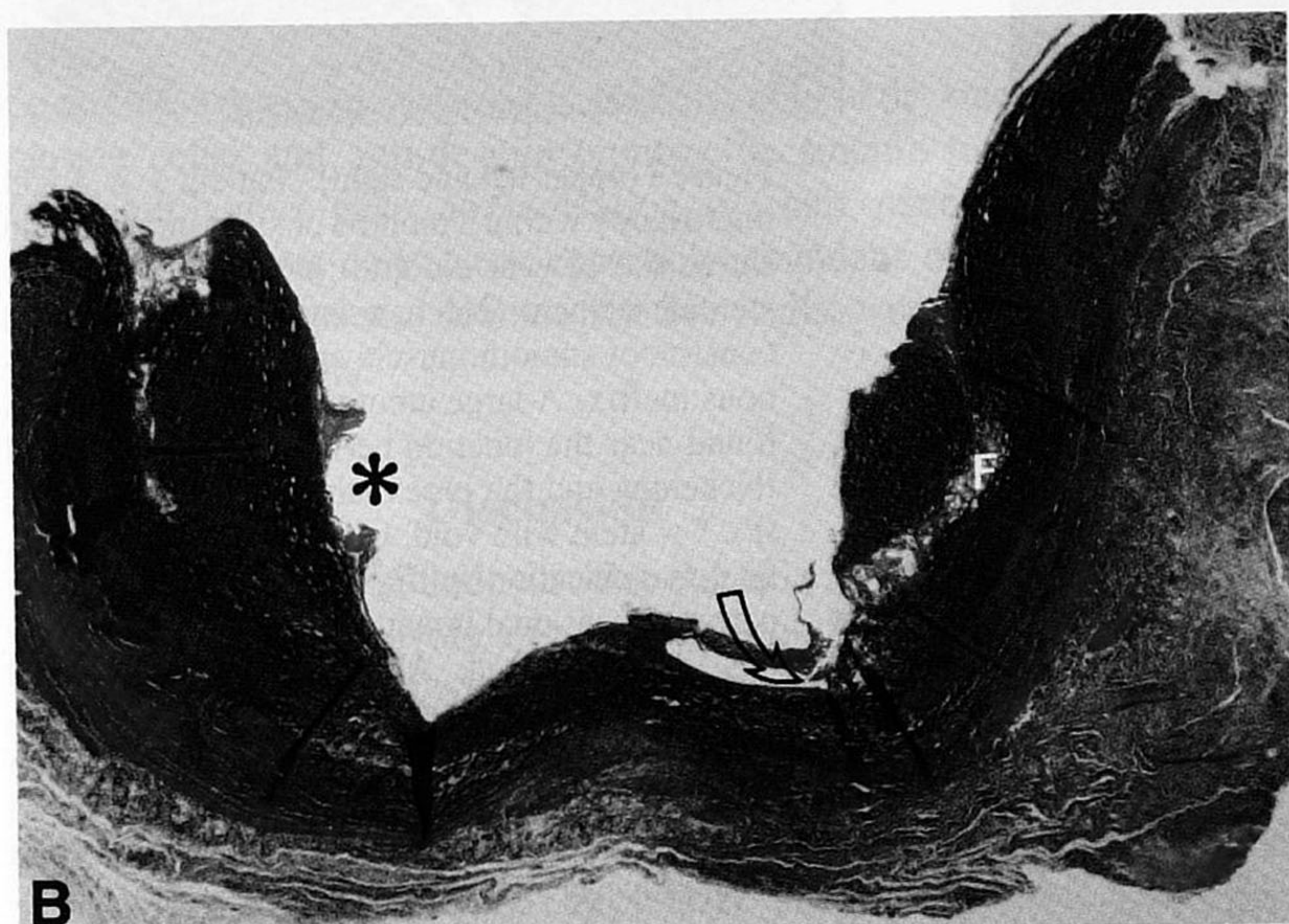
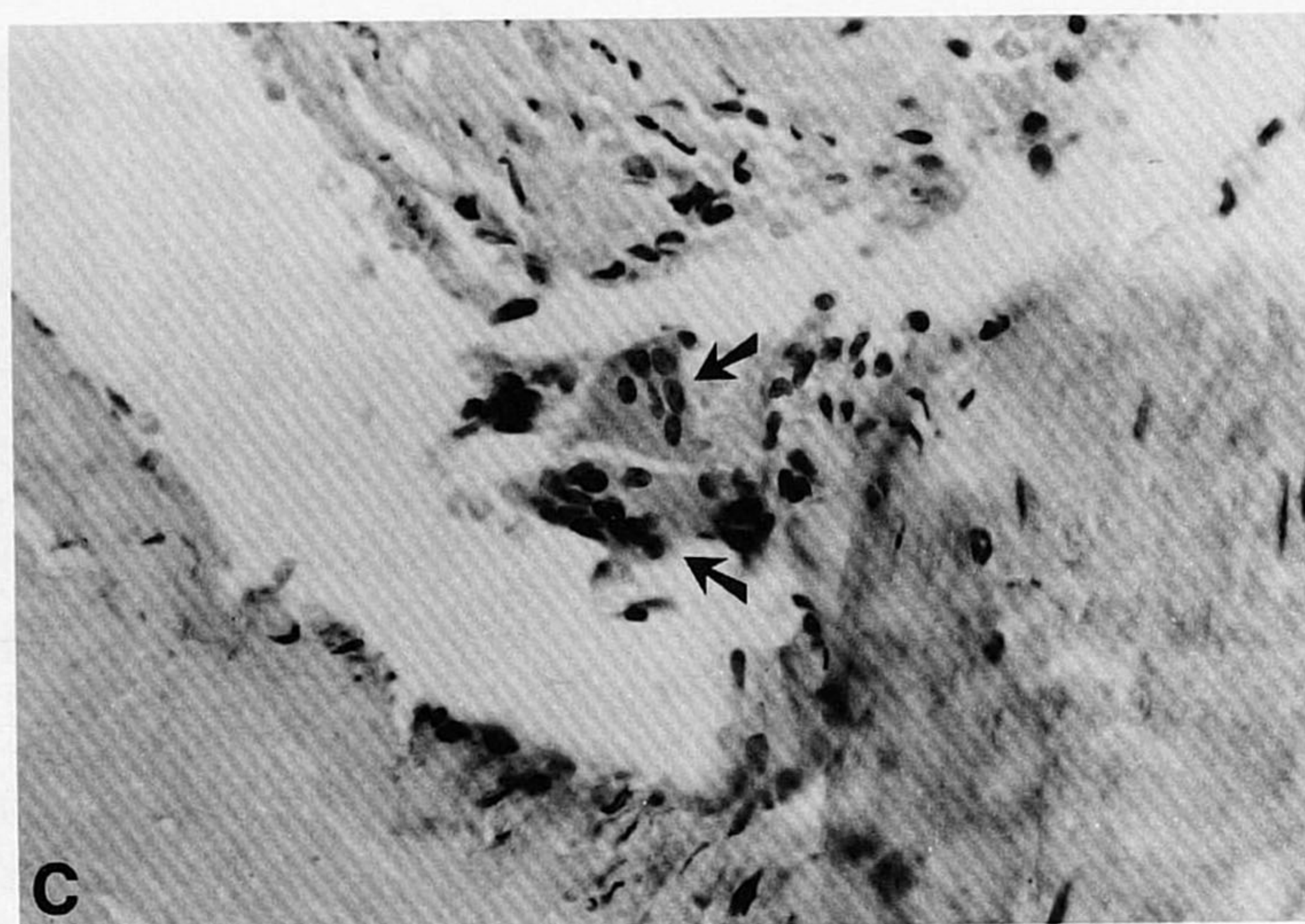
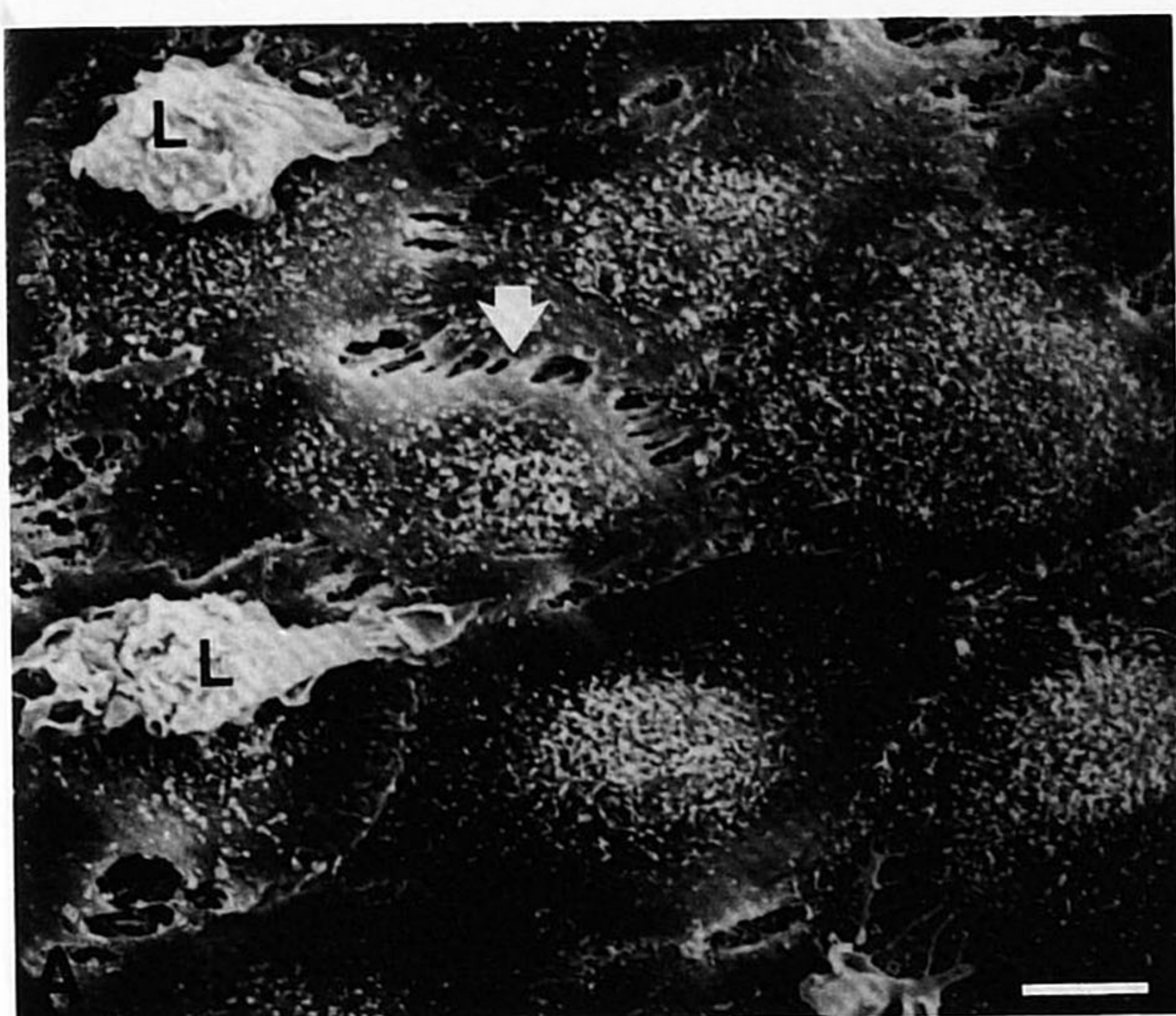
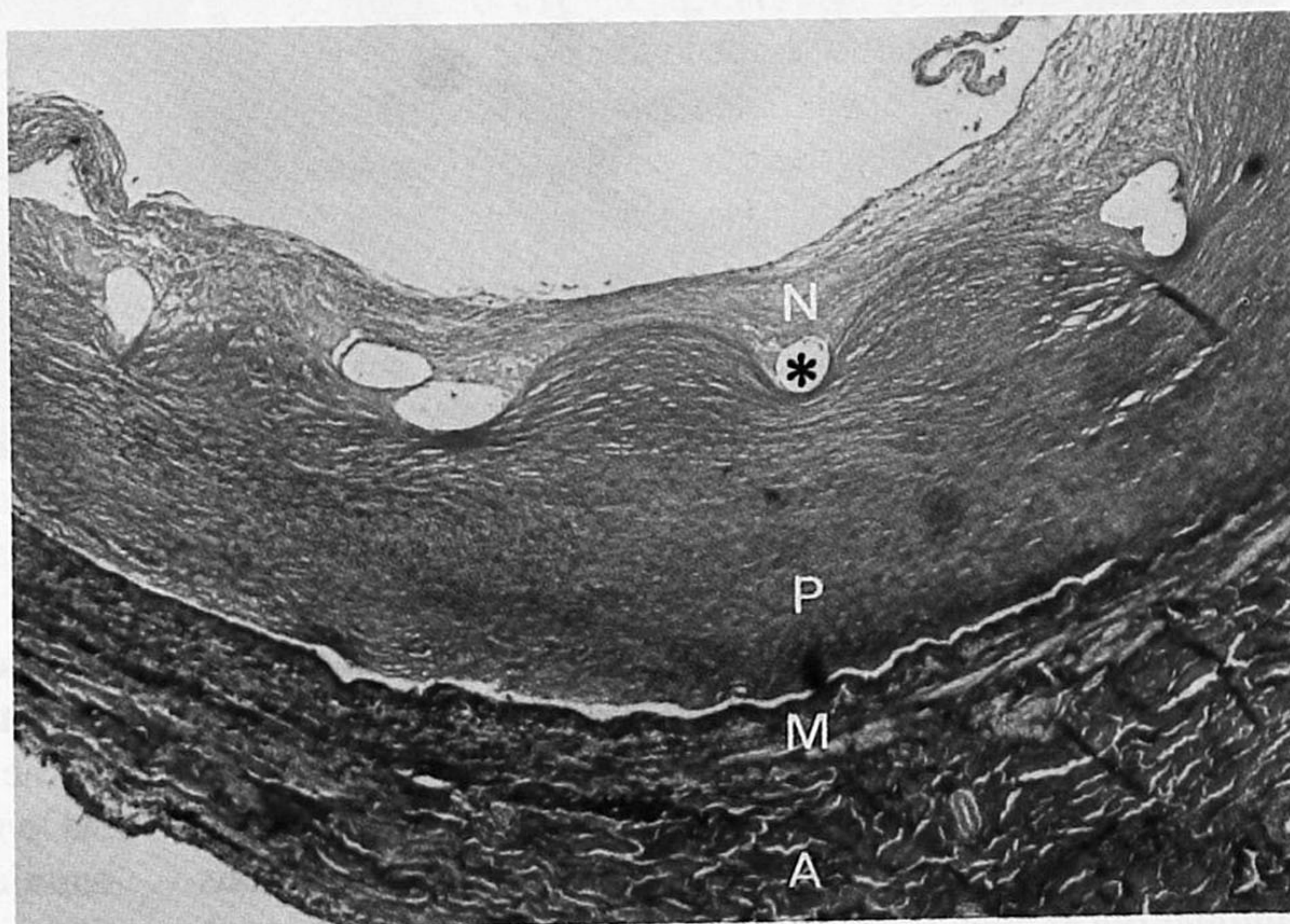


Figure 3. Patient 5. **A**, Scanning electron microscopy showing the endothelial lining, 3 months after stent implantation. Abnormal features seen include polygonal shape, loose intercellular junctions (arrow) and leukocyte adhesion (L). Bar = 5 μ m. **B**, Light microscopy from a section proximal in the stent, showing fibrin remnants (bright red) around a stent wire void (*), probably deposited shortly after stent implantation. The slit-like appearance of the stent wire void (arrow) indicates that the wire is located near the areas with foam cells (F). A multinucleated giant cell (arrow) is in close contact with the stent wire. Fibrin Lendrum stain, final magnification $\times 40$, reduced by 45%. **C**, Closer detail of **B**, showing the multinucleated giant cell (arrows). Hematoxylin-azophloxine stain, final magnification $\times 430$, reduced by 35%.

Figure 4. Patient 6. Light microscopy of a 6-month old graft obtained 3 months after stent implantation. Thrombus remnants (yellow areas) are still conspicuously present around the stent wire voids (*) and clearly demarcate the border between preexisting intimal thickening (P) and new intimal thickening (N). A = adventitia; M = media. Elastin stain, final magnification $\times 40$.



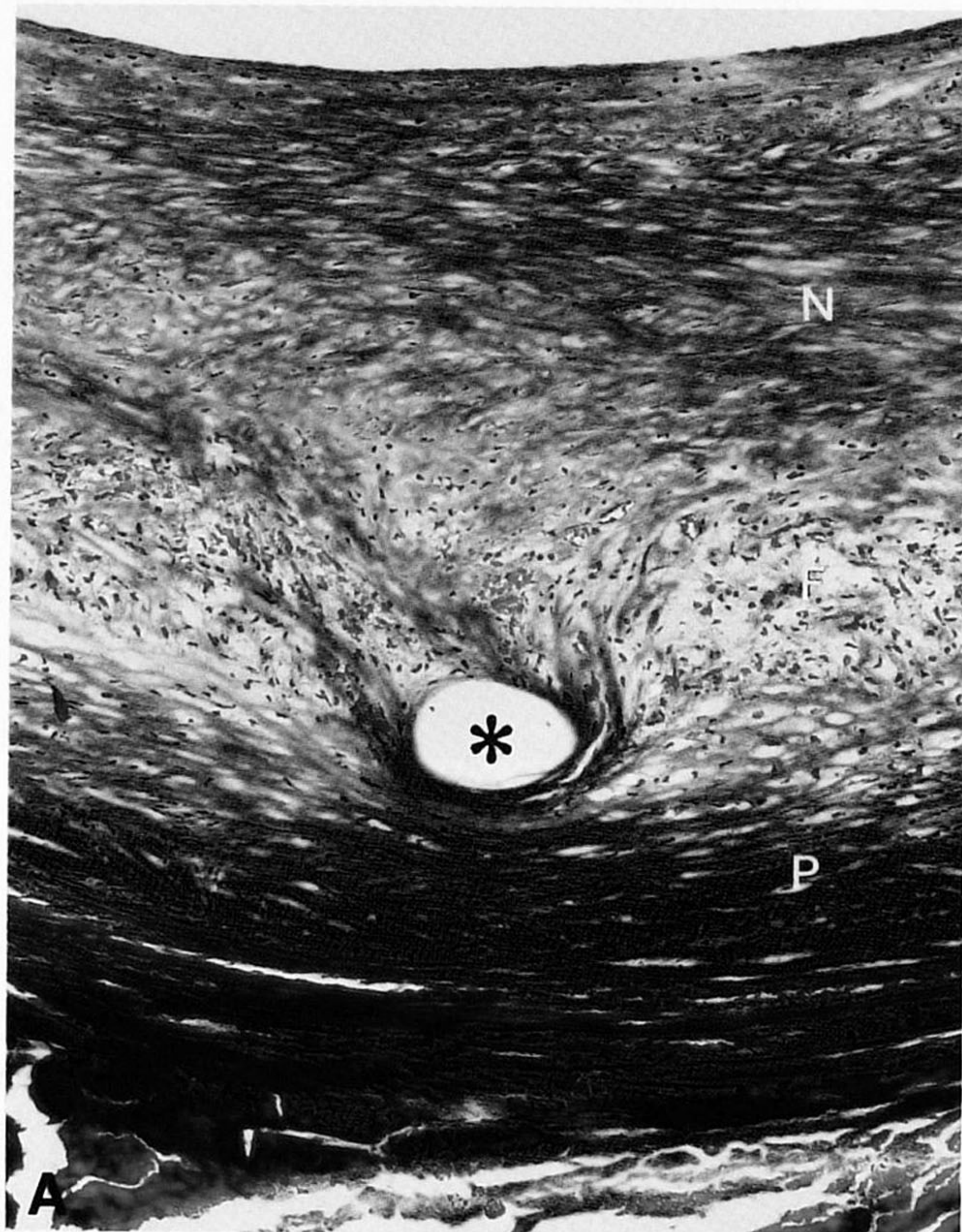


Figure 5 (upper left and right). Patient 7. **A**, Light microscopy within 3 months of stent implantation shows the 12-year old graft narrowed within the stented segment. The new intimal thickening (N) consists of smooth muscle cells within a collagenous matrix. A large amount of foam cells (F) is found near the junction between the new intimal thickening and the preexisting intimal thickening (P). * = stent wire void. Goldner trichrome stain, final magnification $\times 107$. **B**, At the site with the narrowest residual lumen, a recent mural thrombus (T) was found. Although the new intimal thickening nearest to the lumen is relatively acellular (arrows) near the thrombus, a region with increased cellularity and neovascularization is observed. Toluidin blue stain, final magnification $\times 40$, reduced by 35%.

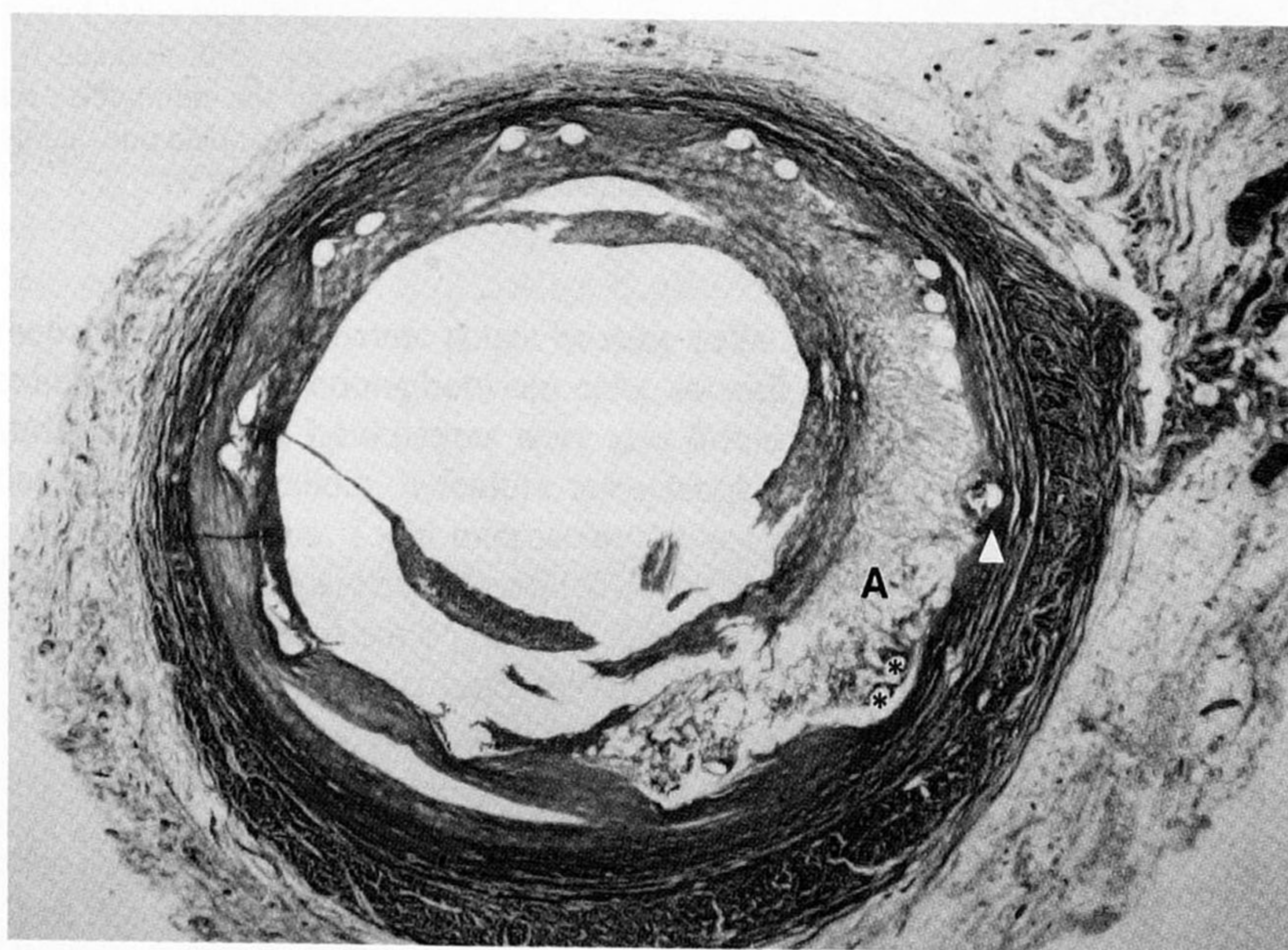
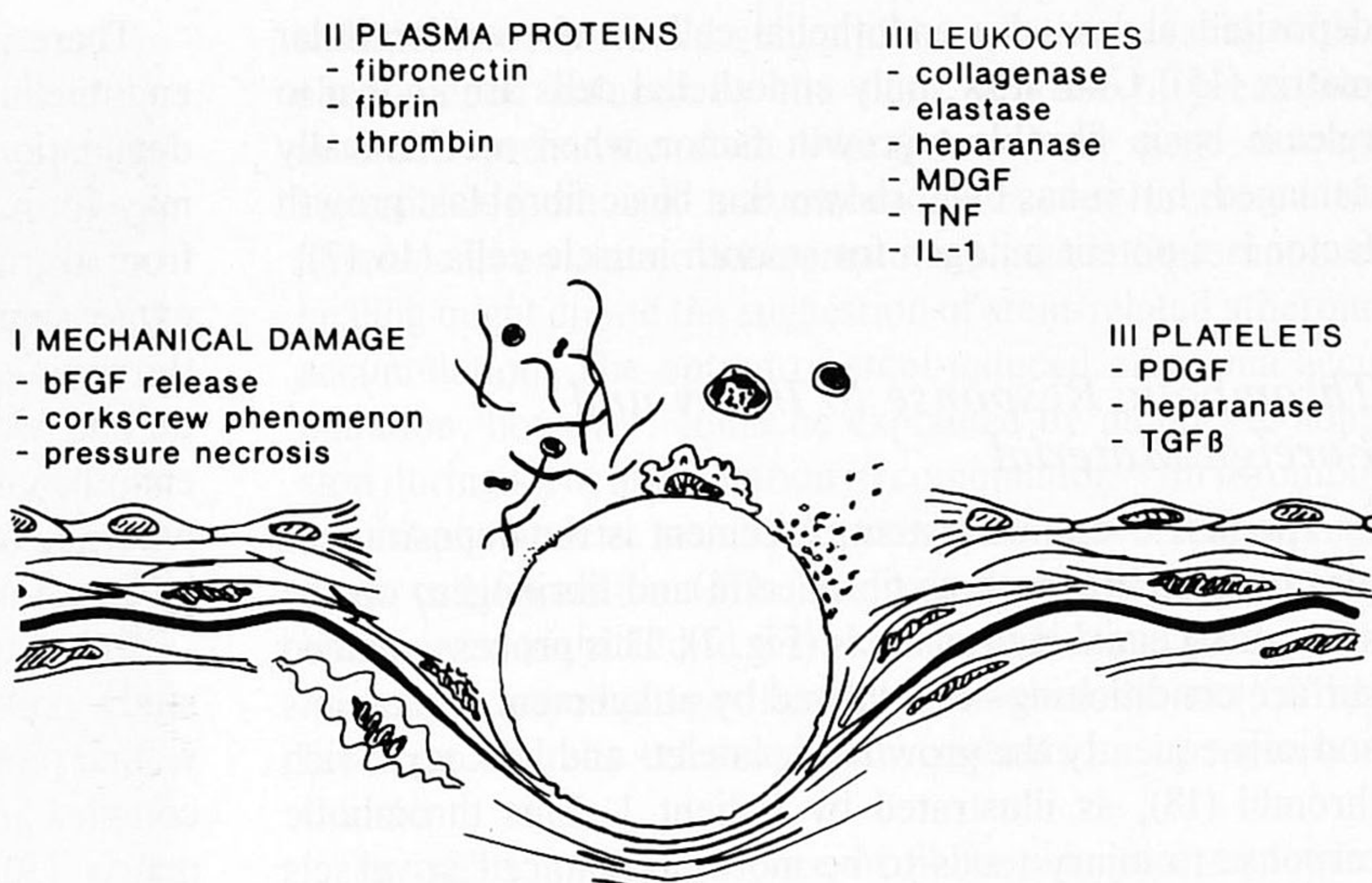


Figure 6 (lower left). Patient 10. Light microscopy 6 months after stent implantation. Proximal in the stented graft segment, an atheromatous plaque (A) is located on top of the stent wires (*). This micrograph, however, does not represent the minimal lumen diameter. Fibrin remnants (arrowhead) partly surround the wires. Goldner trichrome stain, final magnification $\times 21$, reduced by 35%.

the stent ends. The proximal stent segment located near the ostium of the graft was not excised. As in Patient 8, histopathologic study revealed a localized atherosclerotic plaque at the lumen side of the stent wires (Fig. 6). The distal stent end revealed a long dissection within the new fibrous intimal thickening.

Immunocytochemistry. Immunocytochemistry with positive staining for the lectin *Ulex Europaeus* confirmed that the new intimal thickening of three patients (Cases 6, 9 and 10) was covered by endothelium. Analysis with smooth muscle cell-specific anti-alpha-actin antibodies confirmed the presence of smooth muscle cells in the new intimal

Figure 7. Early events after stenting. The first event, mechanical (balloon- and stent-related) damage, is mainly inflicted during the implantation procedure and induces the release of basic fibroblast growth factor (bFGF) from damaged cells and matrix. As soon as blood flow is restored, plasma proteins adhere to the stent and damaged areas, followed by platelets and then leukocytes, an important source of growth factors, growth modulators, cytokines and enzymes. Surface irregularities to which macrophages often adhere can trap air nuclei, which are highly thrombogenic. IL-1 = interleukin-1; MDGF = macrophage-derived growth factor; PDGF = platelet-derived growth factor; TGF β = transforming growth factor β ; TNF = tumor necrosis factor.



thickening. Analysis with anti-vimentin (synthetic smooth muscle cells) and anti-desmin (contractile smooth muscle cells) antibodies gave an indication of their phenotype. Vimentin-positive cells were especially abundant near the stent wires, whereas desmin-positive cells were found mainly in the media.

Discussion

The implantation of endovascular stents in aortocoronary bypass grafts may provide an alternative solution for revascularization in patients who are poor candidates for reoperation (3). Our analysis of stented grafts resected because of stent failure or a nonstent-related complication revealed that large amounts of platelets and leukocytes adhere to the metal stent wires, during the first days after implantation and despite extensive anticoagulation, indicating that this type of stent is potentially thrombogenic. We also observed that the stent wires exert considerable pressure on the vessel wall and thereby might be able to rupture the sometimes thin fibrous cap covering atherosclerotic plaque, a possible mechanism of acute or subacute occlusion several days after an initially successful procedure.

Within 3 months, the stents were completely embedded in the vessel wall and were covered by polygonal bulging endothelium, showing leukocyte adhesion. Near the stent wires, large numbers of foam cells and fibrin deposits were found.

Between 6 and 10 months after stenting the endothelium still appeared abnormal. Large extracellular lipid deposits containing cholesterol crystals and a necrotic core were observed near the stent wires.

Tissue that narrowed the vessels always consisted of smooth muscle cells within an extensive extracellular matrix.

To provide a framework for a better understanding of the mechanism of stenting and its short- and long-term implications, we constructed a scheme (Fig. 7 and 8) describing the

sequence of events that we believe take place during and after stenting.

Mechanical Damage to the Vessel Wall

Acute injury (Fig. 7). Implantation of a stent is accompanied by damage to the endothelial lining and stretching of the vessel wall. The extent of acute injury depends on several factors, such as stent design, means of delivery, lesion morphology and additional balloon angioplasty. Stent size relative to the receiving vessel (9) determines the extent of stretching, whereas lesion morphology determines which part of the lesion or "normal" wall is stretched.

Chronic injury. The stent is also a source of constant injury. This is especially true of the self-expanding Wall-stent, which exerts an uninterrupted radial force on the vessel wall (10). Oversizing this stent by >0.7 mm relative to the reference diameter was found to be associated with restenosis in both native coronary arteries and venous bypass grafts (11).

We have also studied pig coronary arteries 1 and 4 weeks after implantation with the balloon-expandable Wiktor stent. When grading the damage according to Schwartz et al. (12), we found that significantly more damage was observed after 4 weeks. Movement of a rigid implant within the vessel wall could cause this late damage (13). An alternative explanation is local weakening of the media underneath the wire as a result of reparative processes, which could be aggravated by vasospasm.

Injury and growth factors. As shown by Fingerle et al. (14), the early proliferative response of smooth muscle cells to injury occurs even in the absence of platelets. Thus, growth factors other than those derived from platelets play a major role in the early proliferative response.

A possible incentive for cell proliferation after injury is the release of endogenous pools of growth factors from vascular cells, such as basic fibroblast growth factor (Fig. 7) from endothelial cells and smooth muscle cells, which are

deposited at least by endothelial cells in the extracellular matrix (15). Until now, only endothelial cells are known to release basic fibroblast growth factor when mechanically damaged, but it has been shown that basic fibroblast growth factor is a potent mitogen for smooth muscle cells (16,17).

Thrombotic Response to Injury and Foreign Material

The first event after stent placement is the deposition of plasma proteins (such as fibronectin and fibrinogen) on the stent surface and injured areas (Fig. 7). This process—called surface conditioning—is followed by attachment of platelets and subsequently the growth of platelet- and leukocyte-rich thrombi (18), as illustrated by Patient 1. This thrombotic response to injury tends to be more pronounced in vessels affected by intimal thickening or atherosclerosis (19).

The thrombotic response to the stent is further determined by three important surface variables: surface texture or roughness, surface charge or electrochemical potentials and surface chemistry. A rough surface may retain small air bubbles (air nuclei), which will turn a basically thromboresistant surface into a thrombogenic surface. Avoiding air nuclei (denucleation) is probably a more important factor in the biologic response than surface charge and chemistry. The latter two variables predominantly determine which proteins adsorb to the surface (18,20). However, it is unknown how plasma proteins influence tissue proliferation.

Reconstruction of the Vessel Wall

Migration and proliferation of smooth muscle cells. Within several days after stent implantation, the thrombus is colonized by smooth muscle cells. The source of these smooth muscle cells is not known. They might migrate from the media or plaque on which the stent was placed, especially at sites of internal elastic membrane rupture (Fig. 8), or through gaps in the internal elastic membrane widened by excessive stretch or enzymatic degradation by macrophages. An alternative origin of smooth muscle cells is the subendothelial space adjacent to the injured site. In normal porcine coronary arteries (21), thrombus is cleared away within a few weeks after stenting, but in a porcine vein graft model (22) and in the present study, thrombus remnants as a nidus for smooth muscle cell colonization remain associated with the stents for much longer.

Endothelial regeneration. In young pigs, the stent is completely endothelialized within 1 week (21). In humans, it is unknown how long it takes before the endothelium is regenerated, but at 3 months, the process seems complete. Up to 10 months after the intervention, however, endothelial cells revealed a polygonal shape, prominent bulging nuclei and leukocyte adhesion. These features are commonly observed on endothelium covering atherosclerotic plaque (23) and are perhaps a general feature of these grafts, extending well beyond the stented segments.

There is experimental and clinical evidence (24,25) that endothelium can be dysfunctional for weeks after balloon denudation. The prolonged presence of thrombus elements may form a constant source of thrombin that is protected from degradation (26,27) and cytokines. The latter inhibit the expression of the thrombin receptor thrombomodulin and thrombin-stimulated secretion of tissue plasminogen activator and protein C activation. This can turn the anticoagulant endothelial surface into a procoagulant surface (28,29). The presence of small thrombi as seen in Patient 7 (Fig. 5B) may be an example of this defect.

Reconstruction and growth factors. Although we did not study growth factors in our specimens, it is known that wound repair after injury is not a simple process but rather a complex interaction among cytokines, cells and extracellular matrix (30). Damaged endothelium and macrophages are known to release basic fibroblast growth factor (16,31). Macrophages and platelets can release the enzyme heparanase (32), which degrades (subendothelial) extracellular matrix, thereby releasing more basic fibroblast growth factor. This in turn induces a phenotypic change in smooth muscle cells that renders them more susceptible to mitogens and atherogenic stimuli (15). We hypothesize that constant stent-related injury may induce a proliferative response (33,34) by both injuring the extracellular matrix and damaging the cell and subsequently releasing intracellular growth factor pools.

From Reconstruction to Lumen Obstruction (Fig. 9)

Within approximately 3 months, the prosthesis is embedded in the vessel wall and covered by endothelium. Has an end stage now been reached? Some of our observations indicate that this question may not be a rhetorical one. All grafts we examined, both restenosed and nonrestenosed stented segments, revealed considerable adhesion of leukocytes to the endothelial layer. This suggests an ongoing atherosclerotic process, influenced perhaps by an immune-mediated hypersensitivity reaction (35) or chronic endothelial dysfunction (24,25), or both, which in Patient 7 may have caused repeated thrombus deposition and ultimately narrowing of the vessel (Fig. 5B).

It is likely that vascular injury of any cause will elicit basically the same healing process (36). Stent implantation, however, may be unique because a persistent foreign body is introduced in the vessel wall. The presence of multinucleated giant cells as found in Patient 5 shows that a "foreign body reaction" to the stent is indeed induced. Macrophage giant cells in vitro survive only a few days unless replenished by "fresh" monocytes. The presence of multinucleated giant cells long after implantation therefore implies continuous division of macrophages at the implant site or an influx of circulating monocytes, or both (13).

In almost every case, we found large accumulations of foam cells or extracellular lipids on top of the stent. This might indicate preexisting atherosclerotic changes, which

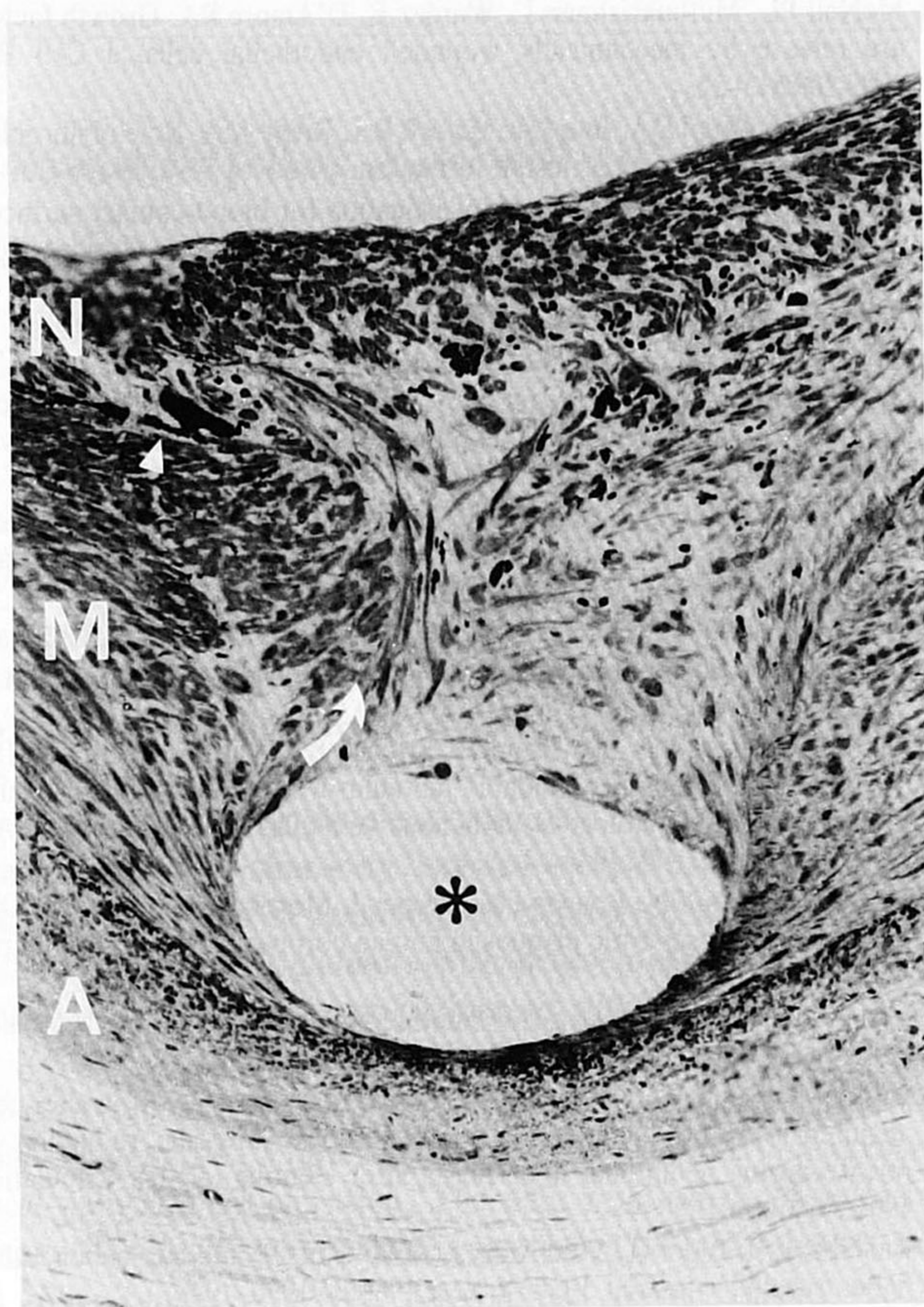


Figure 8. Medtronic Wiktor coronary stent 4 weeks after stenting of a porcine coronary artery. Geisers of smooth muscle cells seem to erupt from the media at the site of the stent wire (curved arrow). A = adventitia; M = media; N = new intimal thickening; * = stent wire void; arrowhead = lamina elastica interna.

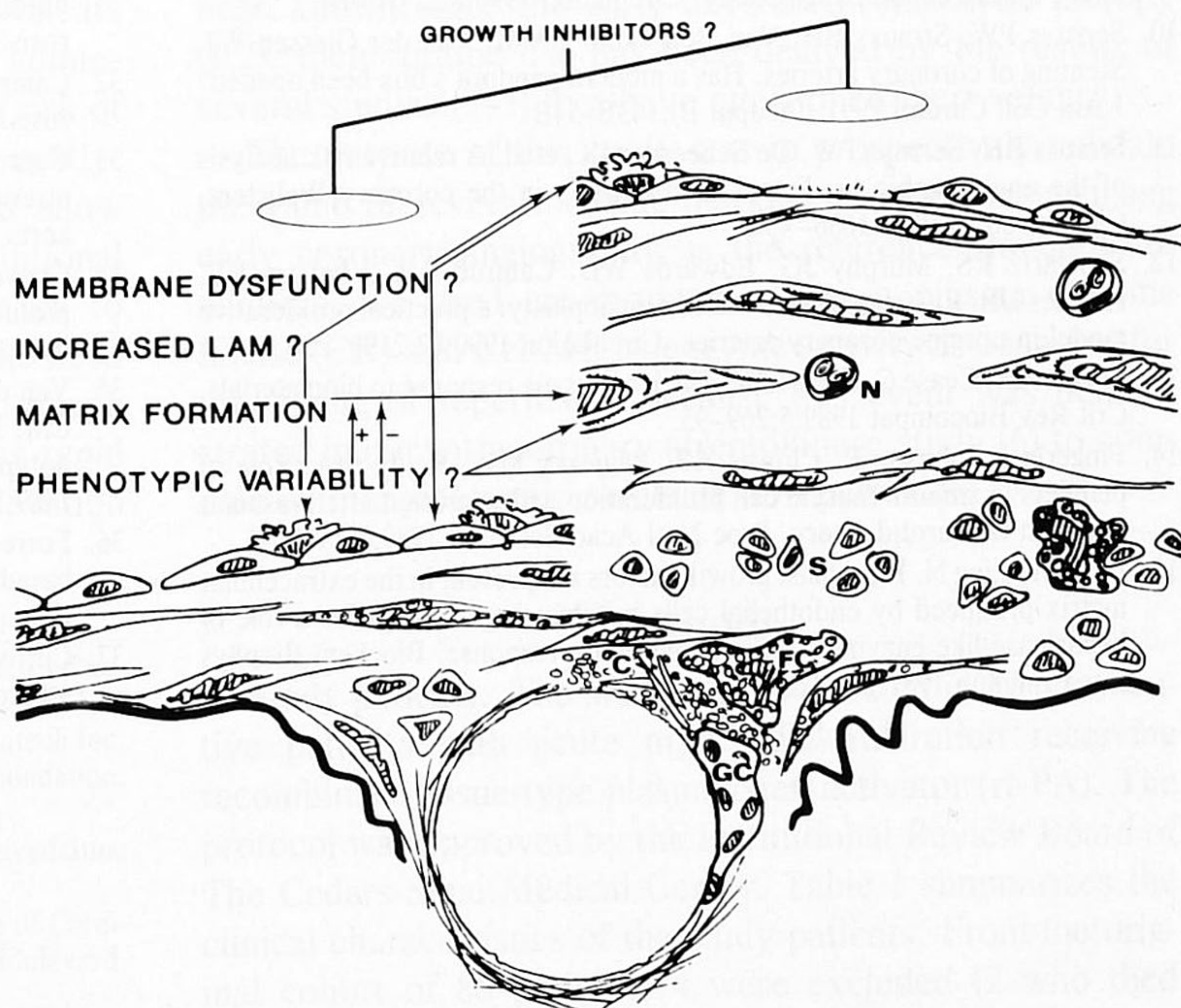
commonly occur in old vein grafts, or we are dealing with a phenomenon induced by the stent. In case of preexisting atherosclerosis, the stent wires may have gone through superficial foam cell accumulation or even lacerated the fibrous cap on classic atheromatous plaques. Superimposed healing might create the suggestion of stent-related atheroma accumulation. The option of stent-induced atheroma accumulation, however, could be explained by monocyte adhesion during stent implantation in combination with thrombin-activated platelets, which are potent inducers of macrophage foam cell formation (37). Late adhesion and migration of leukocytes might add to this. Extracellular lipid pools might arise from macrophages that have reached the end of their life span, thereby releasing their contents in the vicinity of the stent.

In addition, the presence of fibrin near the stent wires deep in the atheromatous plaque indicates that the stent was in contact with flowing blood for a considerable amount of time and points in the direction of delayed rupture of atheromatous plaques (that is, a preexisting lesion) or accelerated atheroma accumulation (that is, a new lesion).

Conclusions

Stenting of old stenotic bypass grafts may provide an attractive alternative to repeat bypass surgery or balloon angioplasty in selected cases. The main histologic findings in this study are that considerable acute thrombotic responses take place, even in a case considered to be without thrombotic complication. Later on, abnormalities after vessel wall reconstruction appear chronic in nature and consist of layers of smooth muscle cells within an extensive collagenous matrix, enhanced white blood cell adhesion to the endothe-

Figure 9. The chronic phase after stenting. During this phase, growth promoters and inhibitors may be out of balance, resulting in smooth muscle cell phenotypic variability, excessive matrix production and finally hyperplasia, whereas at the level of the endothelium, membrane dysfunction perhaps results in increased leukocyte adhesion, thrombogenicity and lipid infiltration. C = cholesterol crystals; FC = foam cells; GC = giant cells; LAM = leukocyte adhesion molecules; N = neocapillaries; S = smooth muscle cell in a longitudinal orientation.



lium and aggregates of foam cells and extracellular lipid accumulation. Although reconstruction of the vessel wall after arterial injury probably follows a uniform pattern, some of our observations may be unique to coronary stenting.

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