Ingrowth of aorta wall into stent grafts impregnated with basic fibroblast growth factor: A porcine in vivo study of blood vessel prosthesis healing

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Objective: **Endovascular aneurysm repair is an alternative treatment of abdominal aortic aneurysm. The procedure is less invasive, and morbidity and most probably mortality are reduced. However, some problems, such as endoleakage, are yet to be resolved. Endoleakage can occur after graft migration, as a result of insufficient fixation of the stent graft. One cause is deficient healing between the aortic neck and the stent graft. We hypothesize that better healing, achieved by induction of vascular cell ingrowth into the graft material, results in better graft fixation. Previously we demonstrated ingrowth of neointima into the graft material if the stent graft is impregnated with a coat of basic fibroblast growth factor (bFGF), heparin, and collagen. In this study we evaluated healing with bFGF-heparin-collagen–coated stent grafts in vivo.**

Methods: **In 4 pigs, 32 endovascular stent grafts, manufactured from standard Dacron and Gianturco Z-stents, were placed in the aorta. The stent grafts were impregnated with either bFGF-heparin containing collagen (n = 16) or control collagen (n = 16). After 4 and 8 weeks animals were killed, and ingrowth and healing of the stent grafts were macroscopically and electron microscopically evaluated.**

Results: **After 8 weeks all bFGF-impregnated stent grafts demonstrated ingrowth of tissue and healing between the graft and the aorta, whereas the control nonimpregnated stent grafts showed no ingrowth. Microscopic evaluation demonstrated α-smooth muscle actin–positive cells, most probably smooth muscle cells or myofibroblasts, growing from the vascular wall through the graft material.**

Conclusion: **A Dacron prosthesis impregnated with collagen, heparin, and bFGF induced graft healing in an in vivo pig model, in contrast to nonimpregnated stent grafts. This in vivo study confirms our previous findings in vitro. These results indicate that healing between Dacron and the aorta can be achieved, and suggest that type I endoleakage may be resolved by inducing healing between the aortic wall and the prosthesis with graft material containing growth factor. (J Vasc Surg 2004;39:850-8.)**

Endovascular aortic aneurysm repair (EVAR) has been in development for approximately 10 years. After an experimental period, EVAR has now been accepted by many as an alternative to conventional surgery. After use of EVAR for several years, endoleakage early and late after the operation is still a problem. Endoleakage is defined as leakage at the proximal stent of the endovascular prosthesis or as backflow into the aneurysm through one of the lumbar arteries.[1] It is the result of an insufficient seal between the endografts and the vessel wall. One reason for this problem is impaired healing of the endograft to the vessel wall. This hypothesis has been demonstrated by others.[2-4] However, it is also known from experimental work performed in the 1960s and from clinical observations that Dacron graft material and the (aortic) vessel wall never heals adequately.[5] The risk of endoleakage is that the aneurysm sac can be pressurized. Consequently, the risks for expansion of the aneurysm and even of rupture remain. Endoleakage is sometimes caused by stent-graft migration,[1,6-9] and the phenomenon contributes to the high proportion of patients, as many as 27%, with various types of endoleakage.[8,10-13]

We hypothesized that improved healing and ingrowth of vascular cells into the prosthesis material would improve fixation of the endograft. To induce healing between the prosthesis and the aortic wall, an impregnated vascular prosthesis would be used, capable of releasing a growth factor that stimulates cells in the aortic wall. This may result in ingrowth of cells in the prosthesis and in stronger fixation between the graft and the aorta. Previously, in in vitro experiments with an aorta organ culture model of human and pig aorta, we demonstrated that ingrowth of vascular cells into graft material could be achieved with use of basic fibroblast growth factor (bFGF).[14] A neointima was induced in approximately 4 weeks. Furthermore, we demonstrated that impregnating the Dacron prosthetic material with a collagen coating containing heparin and bFGF resulted in slowly controlled release of bFGF over 5 weeks

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and ingrowth of neointima smooth muscle cells or myofibroblasts into the porous prosthetic material, which resulted in healing and fixation of Dacron to the aortic wall.

Although we suggested that bFGF impregnation of vascular prosthesis material is a successful approach to stimulate graft healing in vitro, in vivo application of such an approach may be met with several complications. Little is known, for example, on the effect of variables in vivo, such as blood flow, blood pressure, and vessel wall stress, on ingrowth in vivo and on the washout effect of this growth factor from the blood flow. Therefore we here address the question of whether induction of neointima is possible in vivo, and consequently whether cellular ingrowth and graft healing in vivo occur.

In this study we tested the healing of bFGF-impregnated collagen-coated stent grafts after endovascular delivery in the aorta in a porcine animal model in vivo. In addition, we studied the ingrowth of vascular cells from the aorta into the endovascular stent grafts.

MATERIAL AND METHODS

Study design. The effect of impregnation of collagen-coated stent grafts with bFGF was compared with that with nonimpregnated control stent grafts. Thirty-two stent grafts, 16 impregnated with collagen and bFGF and 16 controls, were implanted in the aortas of four pigs. Before the operation the length and diameter of the aortas were measured at various levels with digital subtraction angiography. On the basis of these measurements, each stent was custom made to guarantee oversizing of 10% of the stent graft with respect to the site of deployment in the porcine thoracic or abdominal aorta. All stents were made for a specific location in the aorta, as determined with the angiographic information. All stent sizes were unique. Because small changes may have occurred, the oversizing, as measured after hand-sewing, varied between 10% and 15%. Two animals each were killed at 4 and 8 weeks. Healing of the stent graft was subsequently analyzed macroscopically, microscopically, and with scanning and electron microscopy.

Stent grafts. The stent grafts were Gianturco Z-stents made of stainless steel (Cook, Denmark) and standard Dacron fabric (Vascutek; Renfrewshire, England), which we impregnated with collagen. We chose to test standard graft material first, because it is likely that this material, rather than material with greater pore size, is used for applications in human beings. The self-expanding stents were 4 cm in diameter and 2.5 cm long. The diameter of the stent graft was determined according to that of the Dacron fabric tube, which varied from 0.9 to 1.8 cm, depending on the aortic diameter. Pre-procedural angiographs showed average diameters of 16 mm for the thoracic aorta and 10 mm for the abdominal aorta.

Vascular prosthetic material was standard woven Dacron fabric used for reconstructive surgery. Under sterile conditions the Dacron vascular prosthetic material was impregnated with collagen, as described.[14-16] Type I insoluble collagen, derived from bovine Achilles tendon (Sigma, St Louis, Mo), was swollen in acetic acid solution. The mixture was dispersed with crushed ice in a blender, then homogenized (Ultra-Turrax T25; IKA Labortechnik, Staufen, Germany). The resulting slurry was filtered through a series of filters (Cellector Screen; Bellco, Feltham, England) with pore size decreasing from 140 μm to 10 μm, and mounted in 47-mm diameter disk filter holders (Swinnex; Millipore, Etten-Leur, The Netherlands). After de-aeration at a pressure of 0.06 mbar, the resulting suspension was plastered as a film on small pieces of Dacron in sterile Petri dishes and dried at room temperature in a sterile flow cabin. The collagen-impregnated Dacron was cross-linked with *N*-(-3-dimethylaminopropyl)-*N*′-ethylcarbodiimide hydrochloride (EDC) and *N*-hydroxysuccinimide. The cross-linking was carried out in 2-morpholinoethane sulfonic acid buffer, to minimize hydrolysis of EDC. The cross-linking was completed, and stopped by washing the impregnated Dacron with 0.1M of Na_2HPO_4 solution.

After repeated washing with sterile water the Dacron, impregnated with cross-linked collagen and heparin (heparin sodium salt; Bufa Chemie, Castricum, The Netherlands) was bound to the prosthetic material, as described.[14,15]

After this procedure the collagen-heparin–impregnated Dacron fabric was washed with a solution of Na_2HPO_4, NaCl, and, finally, distilled water. The binding of bFGF was carried out after incubating the collagen-heparin–impregnated Dacron fabric in phosphate-buffered saline solution (PBS). After blotting dry, the films were incubated with 500 ng of bFGF in PBS containing 1 mg/mL of bovine serum albumin at room temperature. Previous experiments in vitro have shown that the bFGF released at this point was 5 to 8 ng per 24 hours, for 28 days.[14]

After impregnation of the Dacron fabric with either "plain" collagen or bFGF-coated collagen, a stent graft was constructed. Dacron tubes 2.5 cm long and varying in diameter from 0.9 to 1.8 cm were hand-sewn with 6-0 vascular sutures (Gore-Tex) to the Gianturco stents. Various calipers were used to guarantee that the diameter of the stent graft was oversized by 10% compared with the aortic diameter at the intended deployment site.

Release of bFGF from the collagen-impregnated Dacron fabric was analyzed after manipulation of the coated stent grafts before and after the stent grafts were pushed through the sheath of the delivery system. The release of bFGF was evaluated with a test in which every 24 hours a sample was obtained from the culture media in which the collagen-heparin-bFGF–impregnated Dacron fabric was placed. Every 24 hours the medium was replaced after the sample was taken. The concentration of bFGF in the samples was determined with a sandwich enzyme-linked immunosorbent assay (Quantikine bFGF ELISA; R&D Systems, Abingdon, England).

Animals. All animal experiments and animal care were performed according to the directives of the Ethical Committee for Animal Experimentation at Leiden University Medical Center.

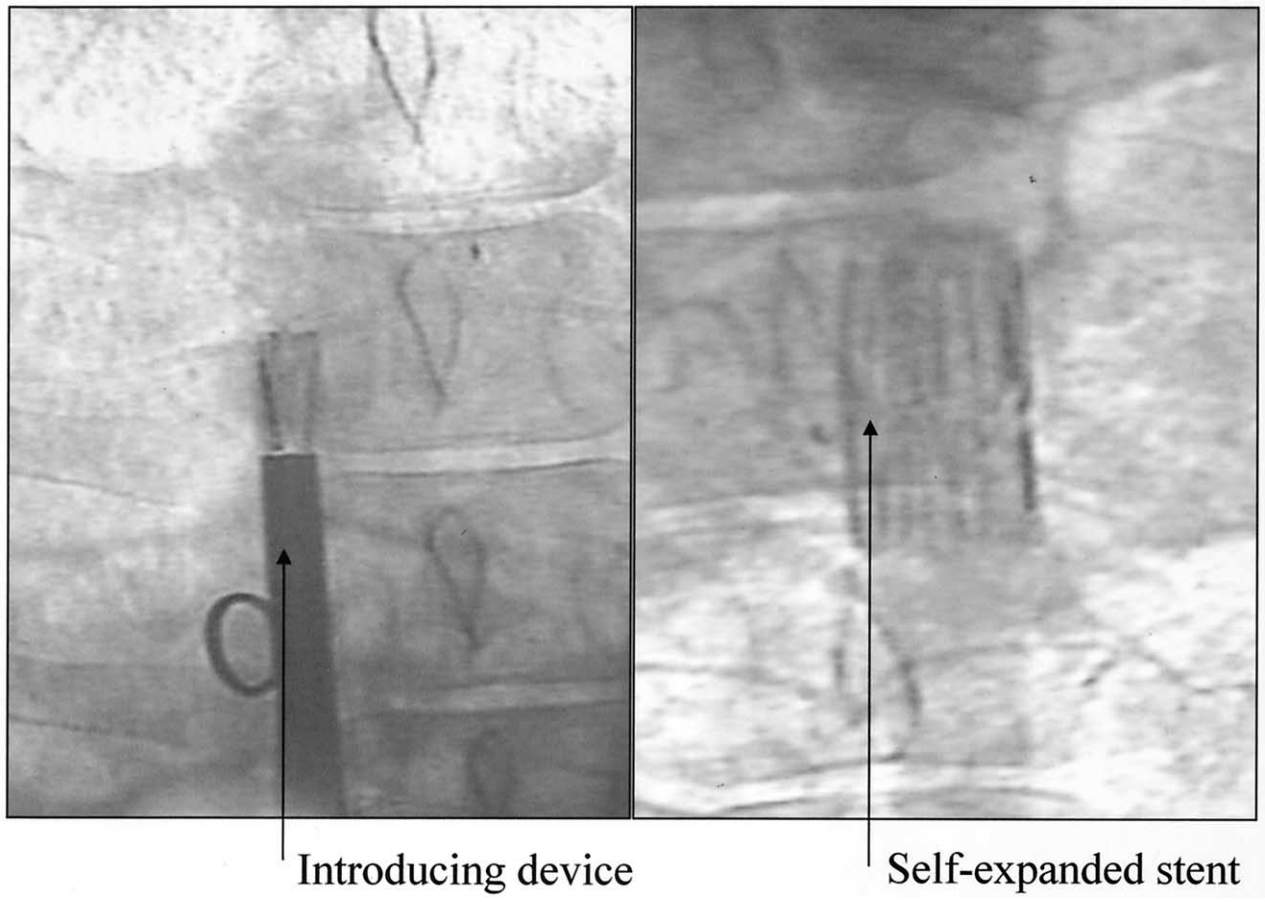

Fig 1. X-ray control of placement of endovascular stent grafts.

Average age of the four female pigs was 10.1 ± 1.5 months. Identification was accomplished with ear labels. The average weight of the animals at the time of the procedure was 58.7 ± 2.3 kg. The average weight of the pigs killed at 4 weeks was 62.1 ± 1.6 kg, and of those killed at 8 weeks was 68.3 ± 1.7 kg. All pigs received oral anticoagulation therapy with warfarin sodium (Coumadin) after the operation, to prevent thrombosis and guarantee patency of the stent graft. Every week blood was drawn. The average international normalized ratio for all pigs was 2.3 ± 0.2. For each intervention the pigs were anesthetized. After premedication with azoperone (2 mg/kg) and atropine (0.05 mg/kg), the pigs were anesthetized with thiopental (10 mg/kg IV), an endotracheal tube was placed, and the lungs were ventilated with NO_2 60%/O_2 40%/isoflurane 0.5% to 3%. Muscle relaxation was obtained with pancuronium bromide (Pavulon; 4-6 mg IV).

Operative technique. The procedure was performed in the animal laboratory under fluoroscopy. Through a midline incision the abdominal aorta was dissected, and a small arteriotomy was made for introduction of the delivery system, which was a 20F endovascular sheath (Cook). Through the 20F sheath, eight stent grafts were introduced and deployed in the aorta in sequence from the thoracic level to just proximal to the aortic bifurcation. To prevent the effect of bFGF upstream in the aorta on more downstream located stent grafts, the stent grafts were deployed in a specific sequence; the four control stent grafts were inserted in the thoracic aorta and subsequently more downstream, and the bFGF-impregnated stent grafts were inserted in the aorta below the diaphragm. Thus each pig could act as its own control. The location, deployment, and apposition were visualized with fluoroscopy before the operation was completed (Fig 1). Subsequently all incisions were closed.

Harvesting of the stent grafts and aorta. The pigs were killed after 4 (n = 2) and 8 (n = 2) weeks. After anesthetizing a pig, a midline incision was made and the thoracic and abdominal aorta was dissected. The pig was euthanized with potassium chloride, and subsequently the aorta was removed completely. After retrieving the aorta, all stent grafts were identified and dissected for macroscopic and microscopic analysis, including standard microscopy and scanning, and transmission electron microscopy. The samples of aortic wall and Dacron graft material for analysis were cut from the stent grafts between the Z-stent. Triangular samples were obtained without any metal from the

stents, which would have made manufacture of the slides for microscopic evaluation impossible.

Histologic analysis. Paraffin-embedded tissue was sectioned at 6 μm. After dehydration, sections were used either for histochemistry or immunohistochemistry. For measurement of intima thickness, sections were stained with hematoxylin, eosin, and saphran. Sections used for immunohistochemistry were incubated with 0.3% H_2O_2 and 0.1% sodium azide in PBS for 15 minutes, to eliminate endogenous peroxide activity, and were rinsed with PBS. Sections were incubated overnight at 4°C with monoclonal antibodies. To identify smooth muscle cells a monoclonal antibody was used that recognizes smooth muscle α-actin (Sigma) at a dilution of 1:750. Bound monoclonal antibodies were detected with horseradish peroxidase–conjugated rabbit anti-mouse antibodies (1:300; DAKO, Glostrup, Denmark). All antibodies were diluted in PBS with 0.1% bovine serum albumin. The sections were exposed for 4 minutes to 0.04% diaminobenzidine tetrahydrochloride in 0.05 mol/L of Trismaleate buffer (pH 7.6) with 0.006% H_2O_2. Sections were briefly counterstained with hematoxylin.

Image analysis. To determine the amount of neointima formation, morphometry was performed with image analysis (QWin; Leica Imaging Systems, Cambridge, England) in histologic sections of the vessel segments. The neointima area is given in square millimeters per microscopic field.

Electron microscopy. Scanning electron microscopy was performed with segments of the harvested stents, which were fixed in 0.05 mol/L of phosphate- buffered (pH 7.0) 2% glutaraldehyde for 16 to 20 hours, and rinsed in the same buffer. Further fixation in 1% OsO_4 in 0.1M sodiumcacodylate (pH 7.4) for 2 hours was followed by dehydration through 70% to absolute acetone. Dehydrated sections were critical point dried, sputter-coated with gold, and examined with a Jeol 6400 scanning electron microscope.

Transmission electron microscopy was used for proper identification of the ingrowing vascular cells. Segments of the isolated stents with aorta were fixed in a 0.1M sodiumcacodylate (pH 7.4) buffered mixture of 2% paraformaldehyde and 1% glutaraldehyde for 2 hours. Further fixation in 1% OsO_4 in 0.1M sodiumcacodylate (pH 7.4) for 2 to 24 hours (until stained black) was followed by dehydration through 70% to absolute alcohol and embedding in Epon. Ultra-thin (70-nm) sections were examined with a Jeol 100 CX or Jeol 1010 electron microscope, with lead citrate and uranyl acetate used for contrast.

RESULTS

Preparation of stent grafts. The effect of manipulation of the coated stent grafts on bFGF release from the coating was analyzed by measuring the release of bFGF from a coating before and after the stent graft was pushed through the sheath of the delivery system. This manipulation caused only a marginal, nonsignificant decrease in the amount of bFGF on the stent graft (6.2 ± 1.1 ng/24 hr vs 5.9 ± 0.9 ng/24 hr). These results were similar to those observed previously in our in vitro study of bFGF release.[14]

Macroscopy. Macroscopic analysis revealed adhesion of the prosthesis material to the aorta wall with bFGF-impregnated Dacron stent grafts, whereas no adherence between the stent graft and the aorta was observed in the control stent grafts. Furthermore, it could clearly be observed macroscopically that with the bFGF-impregnated collagen-coated stent grafts both the metal parts of the stent and the Dacron graft material at the luminal side were completely covered with a white shiny tissue (Fig 2, *A*). There was no space between the Dacron and the aorta, indicating that the Dacron stent graft had completely healed to the vessel wall. During dissection of the stent graft to obtain samples for microscopy, the Dacron proved to be firmly attached to the aorta. On the other hand, the control stent grafts were not covered with any tissue on the luminal side, and there appeared to be a space between the Dacron and aortic wall, which was filled with loose thrombus (Fig 2, *B*). Moreover, during careful dissection for microscopy the Dacron could easily be detached from the aorta, indicating the absence of any healing.

Incorporation of the stents was macroscopically observed after 4 weeks in the bFGF-impregnated stent grafts. No discernible difference between the results obtained at 4 and 8 weeks was observed. At the site of the bFGF-impregnated collagen-coated stent graft no stenosis due to excessive neointima was observed at either 4 or 8 weeks.

Histologic analysis. Since none of the histologic analyses revealed any differences regarding the amount of cells and the type of cells growing into the stents between the 4-week and 8-week groups for both bFGF-impregnated collagen-coated and control stent grafts, the results are not presented separately. Histologic analysis demonstrated that when the animals were killed, at 4 and 8 weeks after the initial operation, the bFGF-impregnated Dacron stent grafts were fully incorporated into the vessel wall, whereas the control grafts were not fixed. Detailed analysis revealed that in the coated stent grafts the prosthetic graft material was not only covered with tissue, but cells grew completely throughout the Dacron fabric (Fig 3, *A*). By 4 weeks almost all the collagen coating in the bFGF-impregnated stent grafts had disappeared; the control grafts showed only an inflammatory reaction (Fig 3, *B*). A large number of cells on the luminal side of the aorta and within and through the Dacron graft were observed. These cells proved to be α-smooth muscle actin-positive cells, most probably smooth muscle cells or myofibroblasts. Further evaluation of this neointima with elastin and HES staining showed that elastin and collagen were absent in the neointima. In addition, at histologic analysis no capillaries were found in the neointima. Endothelial cells could not be identified histologically on the luminal side of the stent graft. However, it should be acknowledged that endothelial cells could have been lost at dissection or processing of the samples.

At image analysis, neointima development and ingrowth was quantified. After 4 weeks, there appeared to be a significant increase in the neointima in the coated group

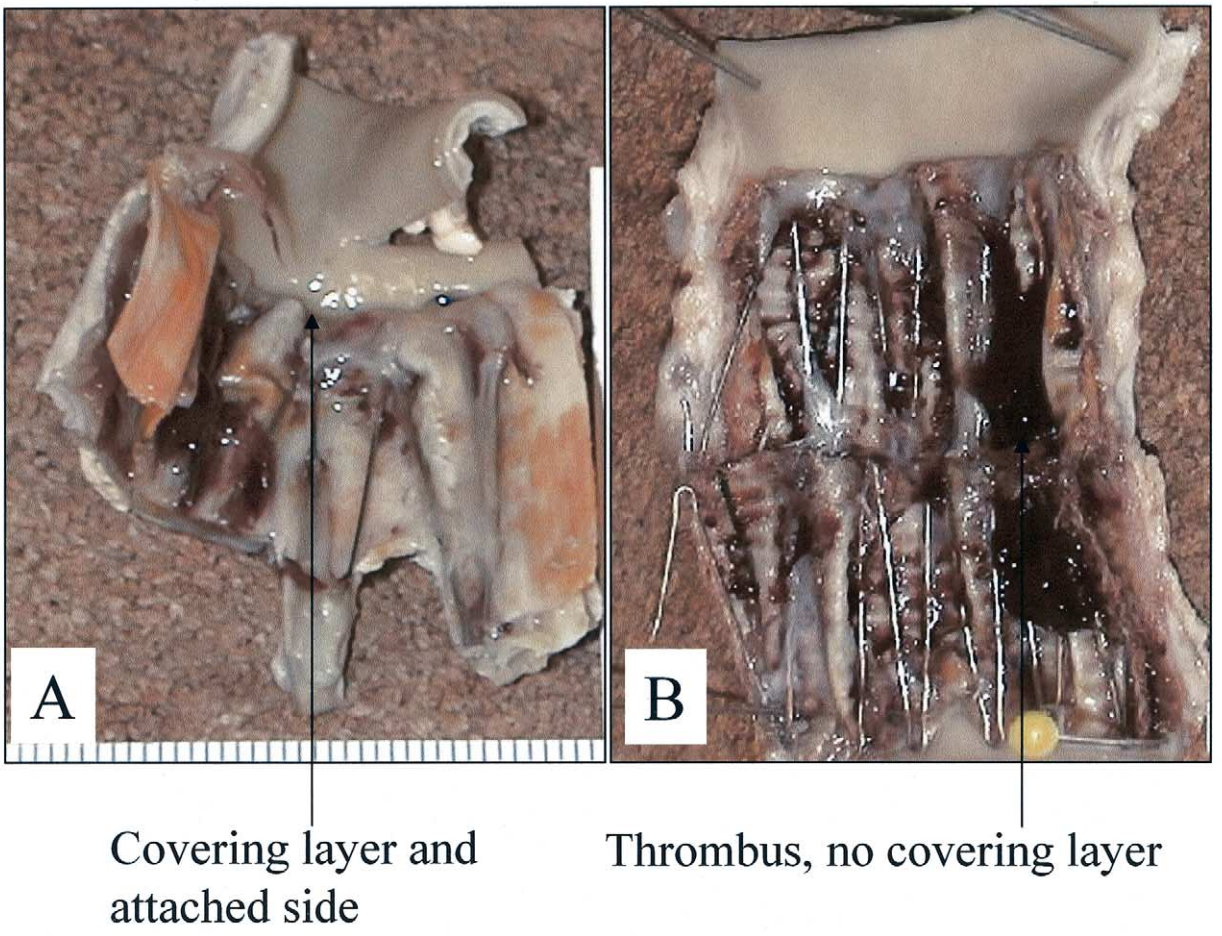

Fig 2. Macroscopic difference between stents impregnated or not impregnated with basic fibroblast growth factor. **A,** Impregnated stent demonstrates a tissue layer on the luminal side and across the proximal landing zone. **B,** Control stent exhibits thrombus.

compared with the control group. The neointima in the bFGF-impregnated stent group was 0.34 ± 0.021 mm^2 per cross section, whereas in the control group the neointima was 0.11 ± 0.015 mm^2 ($P < .05$; Fig 4). Thereafter, the additional increase in the amount of intimal hyperplasia up to 8 weeks appeared to be minimal. This slight increase in neointima from 4 to 8 weeks was not statistically significant.

The control stent grafts were only covered with a layer of thrombus material, both between the vessel wall and the stent graft, but also on the luminal side of the stent graft. When samples were taken, there appeared to be no adherence between the stent graft and the aortic wall. Histologic image analysis demonstrated only erythrocytes between the aortic wall and the stent graft. No cells were observed growing through the Dacron fabric. There were no signs of ingrowth, healing, or fixation.

Scanning electron microscopy. Scanning electron microscopy revealed a pattern similar to the macroscopic appearance. bFGF-impregnated stent grafts demonstrated a fully incorporated stent graft, covered with smooth muscle cells or myofibroblasts, whereas in the control stent grafts no ingrowth was observed (Fig 5, *A* and *B*). SEM analysis showed no differences between 4 and 8 weeks in the Bfgf-impregnated stent grafts. In the control group only red blood cells and fibrin covered the stent graft. Both the Dacron and metal stents were visible.

Transmission electron microscopy. Transmission electron microscopy analysis of the bFGF-impregnated stent grafts demonstrated, on the luminal side, endothelial cells, identified by morphologic features. In the same layer, the neointima, there was an abundance of proliferating cells with a long, stretched appearance, which were identified as young myofibroblasts rather than smooth muscle cells, although the morphologic differences between these two cell types are minimal. All the cells and tissues were completely grown into the Dacron (Fig 6).

DISCUSSION

In a previous in vitro study we demonstrated that bFGF coating of vascular Dacron graft material induced ingrowth

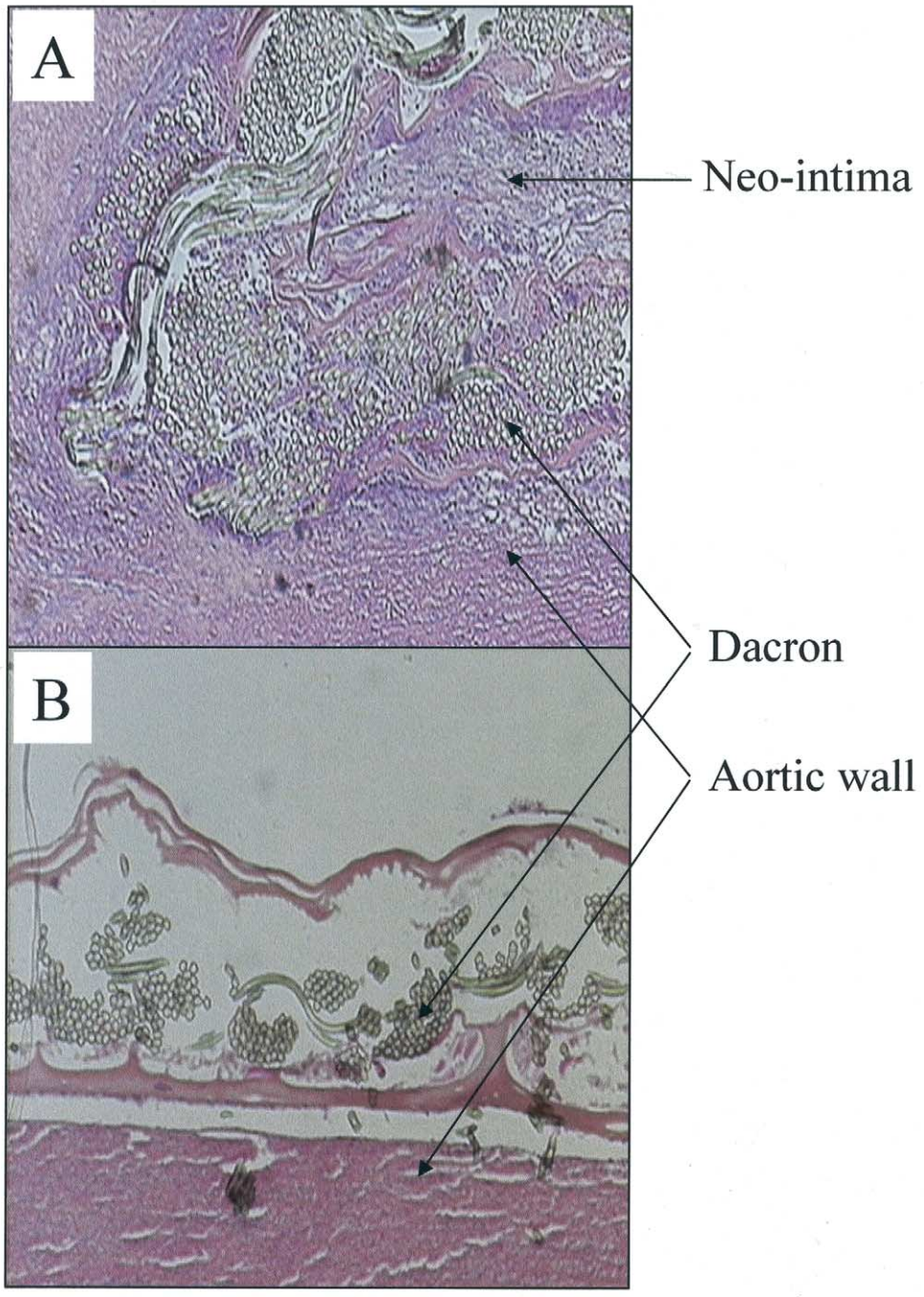

Fig 3. Macroscopic analysis of stents. **A,** Stent impregnated with basic fibroblast growth factor clearly demonstrates neointima growing through the Dacron fabric. **B,** Control stent demonstrates some thrombus, and no attachment to the aorta.

of vascular cells from the aorta.[14] The main finding of this study is that ingrowth and healing of endovascular stent grafts in vivo in pigs can be accomplished by impregnating the grafts with collagen to which bFGF is bound and slowly released over several weeks after implantation.

The main objective of our manipulation of stent grafts was to improve the fixation, now primarily based on stents, into a more permanent fixation, independent from stents. However, the ingrowth or healing does not mean fixation. We have not yet quantified the mechanical quality of our

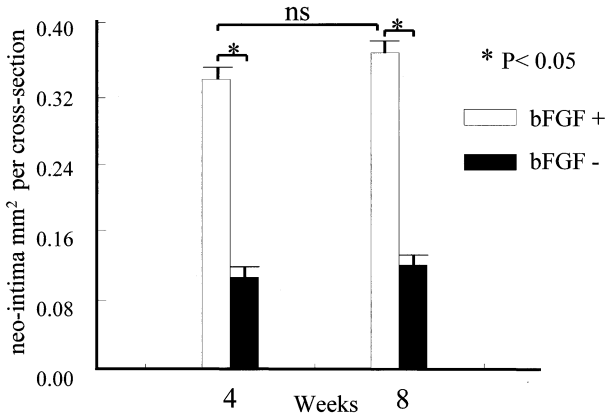

Fig 4. Measurement of the neointima at 4 and 8 weeks after stent placement. There is no difference in neointima induction in stents impregnated with basic fibroblast growth factor (bFGF) at 4 and 8 weeks. Control stents did not induce any significant neointima.

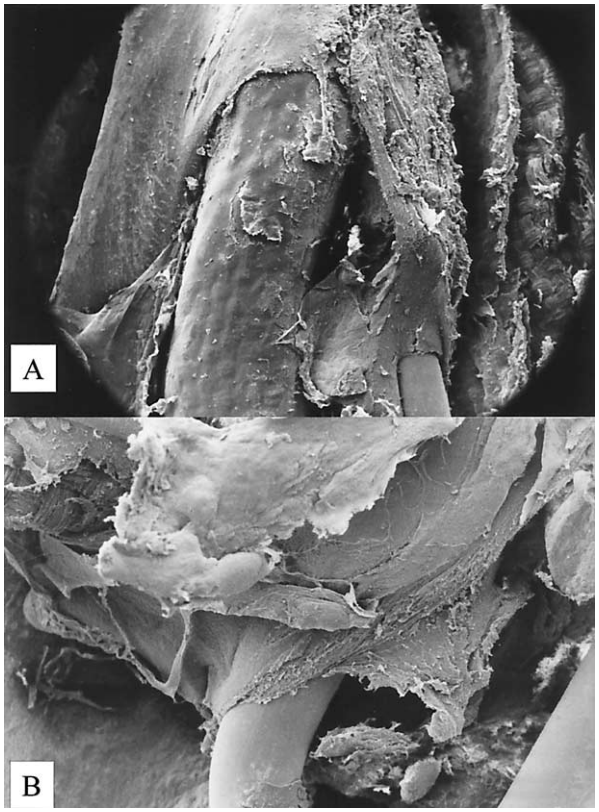

Fig 5. Scanning electron microscopy of stent grafts. Stent impregnated with basic fibroblast growth factor **A,** demonstrates a tissue layer, whereas the control stent **B** does not.

biologic fixation. However, we found that the bFGF-impregnated stent grafts were tightly fixed to the aortic wall, and it appeared to be impossible to separate these from the aortic wall manually, whereas the control stents were never attached and could easily be separated from the aorta. This mechanical difference between the two groups can be explained from the histologic findings. The bFGF-impregnated stent grafts demonstrated a neointima without thrombus, whereas in the controls there was only thrombus between the stent graft and the vessel wall. This suggests that formation of the neointima starts directly after implantation of the stent graft. This could lead to direct sealing of the proximal landing zone of the stent graft, and would therefore not lead to any thrombus formation between the vascular wall and the stent graft, or the thrombus could have functioned as a matrix for the neointima to grow in the stents.

The findings were assessed both macroscopically and microscopically. Macroscopically, all bFGF-impregnated stent grafts demonstrated ingrowth of cells from the vessel wall, whereas the control stent grafts did not demonstrate any ingrowth. This effect was already present at 4 weeks after implantation in the pigs, and did not quantitatively or qualitatively change after another 4 weeks. Microscopically, this effect was confirmed. There was abundant evidence of ingrowth of neointima cells growing between the aortic wall and the bFGF-impregnated stent graft, and growing through and on the luminal side of the Dacron fabric. These findings were also confirmed at scanning and transmission electronic microscopy. The cells growing through the Dacron were identified as stretched, young cells that were α-actin-positive cells. These morphologic characteristics suggest that these cells were most probably myofibroblasts originating from the media of the wall of the aorta. However, the type and origin of these cells are difficult to determine exactly,[17] and we identified these cells at transmission and scanning electron microscopy as smooth muscle–like cells or young smooth muscle cells.

Although the graft material was incorporated, there was no evidence of covering with endothelial cells on the luminal side in sections analyzed at light microscopy with von Willebrand staining. However, transmission electron microscopy demonstrated endothelial-like cells on the surface. Therefore we conclude that healing of the stent grafts occurred with covering of the stent with endothelial-like cells. We believe we could not demonstrate endothelial cells at immunohistologic light microscopy because of the fixation process. Endothelial cells can be easily removed during handling of the incorporated stent grafts.[18]

Although the bFGF-impregnated grafts were placed downstream from the control grafts, we anticipated the possibility that the release of bFGF from downstream grafts could have had an effect on healing of the downstream grafts by concentration of bFGF in circulating blood. We found no evidence for any such possible effect. First, the half-life of bFGF in circulating blood is short, between 40 and 60 minutes, which results, in theory, in a systemic bFGF concentration that is not detectable. Second, we also measured the bFGF systemic concentration as a control of the stent design, and no bFGF was detected at any of time interval. Finally, by placing the two stent designs in one animal we were able to eliminate any interspecies variation.

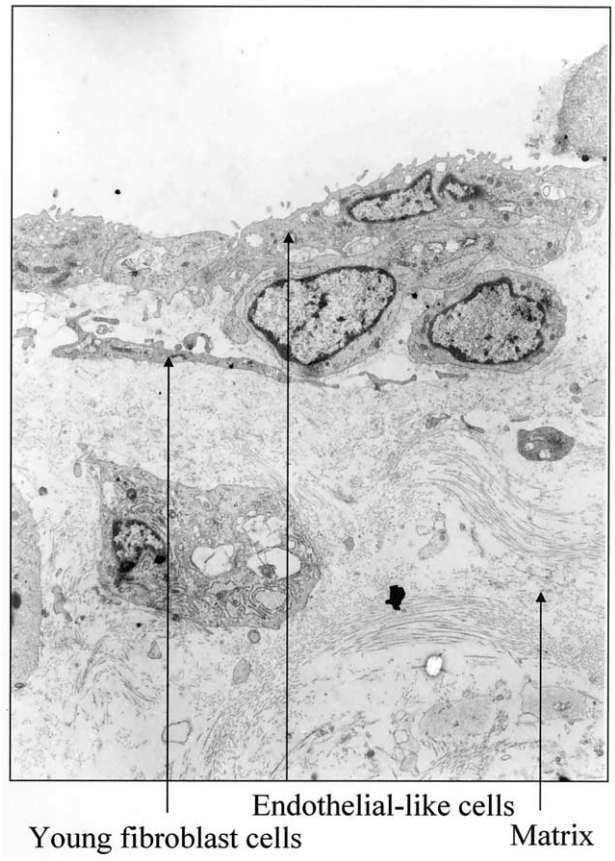

Fig 6. Transmission electron microscopy of stent impregnated with basic fibroblast growth factor clearly shows that the Dacron fabric is covered with tissue.

It has long been known that vascular prostheses are well incorporated in animals that are young and healthy.[19] For this reason we chose the period of 4 to 8 weeks for our experiments, to prevent the natural healing in the pig model. The pig vascular model is well-known for its capability of incorporating and endothelializing conventional vascular prostheses after a few months. To prevent any natural ingrowth or healing by the pigs, we chose the relatively short periods of 4 and 8 weeks before the animals were killed. These relatively short periods prevented the possibility of studying other facets of stent grafts procedures, for example, migration. Migration will be evaluated in our next studies. The observation that the control endovascular stents grafts did not show any incorporation in the pig aorta, except for a minor inflammatory reaction, even after 8 weeks, supports our conclusion that the effects can be exclusively attributed to the growth factor.

It is important for optimal fixation of the stent graft that both the stent and the Dacron fabric are well-incorporated in the vessel wall. It is universally accepted that incorporation of metal stents in the vessel wall is superior to that of graft material. But it is doubtful that fixation of stents is sufficient to prevent migration if the aorta dilates. Mangell et al[20] studied the fixation of endografts in pigs. They implanted Gianturco self-expandable stents and Palmaz balloon-expandable stents. In their study the pigs were killed after 18 weeks, enabling the weight to increase from a mean of 21 kg to 95 kg. These authors found that self-expandable stents continued to show good apposition to the vessel wall, whereas most of the balloon-expandable stents were partially detached from the aortic wall, indicating insufficient incorporation under circumstances of dilation of the aorta as a result of growth of the animals.[20]

It is also well known that incorporation of Dacron graft material at the level of the suture line almost never occurs in the older patient with atherosclerosis who undergoes vascular reconstruction to treat aneurysmal or occlusive disease. Since the beginning of vascular surgery it has been well demonstrated that the integrity of an anastomosis between an artery and a vascular prosthesis depends on the suture line as long as the prosthesis remains in patient.[5] This may be one of the causes of false aneurysm formation, which may occur in as many as 20% of those patients still alive after long-term follow-up of 15 years.[21,22] It has also been well demonstrated that healing of endovascular grafts is absent in human beings. Malina et al[2] retrieved 23 Dacron stent grafts from patients treated for an aortic aneurysm 9 months (range, 1-31 months) after insertion.

Between the graft and the aortic wall a space filled with poorly organized blood components persisted up to 2-1/2 years after implantation of the stent graft. No firm incorporation of the grafts was observed, and only a friable neointimal layer covered parts of the luminal aspect of the grafts. They concluded that endovascular healing provides poor fixation of Dacron stent-grafts in human beings.[2] This absence of healing may be responsible for part of the migration of endografts used to treat aneurysms. Recently, one of the pioneers of vascular surgery, very familiar with the absence of healing with conventional vascular prosthesis as established in the 1960s, warned against making the same underestimation of the problem of inadequate healing and its consequences as was made in the past.[4] The results from studies obtained in animals cannot be directly extrapolated to human beings. However, previously we demonstrated that bFGF can induce ingrowth of aortic cells into Dacron graft material in human atherosclerotic aorta, suggesting that this effect may also be achieved in appropriately impregnated endografts.

We used bFGF because of its capacity to induce intimal hyperplasia, which implies the risk for inducing stenosis in the treated vessel segments. However, no stenosis was found in the pig aortas after 8 weeks. Although stenosis is not likely over the long term, because collagen impregnation as well as bFGF release disappears after 4 weeks, studies should be performed to address this issue over a prolonged period.

In conclusion, we believe that successful induction of ingrowth of vascular cells into stent grafts and the subsequent healing of endovascular stent grafts may help reduce migration of the stent grafts. The incidence of endoleak at the proximal and distal landing zones of endovascular stent grafts can be reduced. On the other hand, cellular ingrowth is not equivalent to the quality of fixation and migration, and further experiments are required to prove the clinical effects. This might represent a step forward in the clinical treatment of human abdominal or thoracic aneurysms with EVAR, because known complications such as endoleak, stent graft migration, and risk for subsequent rupture of the aneurysm may be reduced or even prevented.[21]

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