Experimental and Clinical Experience with a Gelatin Impregnated Dacron Prosthesis

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Laboratory and clinical evaluation of a knitted Dacron graft impregnated with gelatin to confer zero porosity is described. Graft performance was tested by standard methods for biodegradation of the sealant and in vitro thrombogenicity. The gelatin sealant was removed after seven to nine days and there was no platelet adhesion to Gelseal compared with unsealed Dacron. Animal experiments revealed normal macroscopic appearances in the graft and histological disappearance of the gelatin impregnate between five and ten days, allowing a cellular response similar to unsealed Dacron. The first 100 patients to have Gelseal aortic bifurcation grafts implanted at Glasgow Royal Infirmary are described. The graft did not require precloeting. Blood transfusion was not necessary in 74% of patients. There is 100% patency at 21 months. A knitted Dacron graft sealed with gelatin is a safe, nonporous prosthesis at implantation. (Ann Vasc Surg., 1987, 1, 542-547).

KEY-WORDS: Knitted Dacron prosthesis. Gelatin impregnation.

Knitted Dacron has become the standard vascular prosthesis in the replacement of wide bore vessels [1, 2]. Ease of handling, sutureability and conformability are significant advantages and the healing properties of the graft result in a well-anchored pseudointima [3]. The price to be paid for these advantages is the need to «precloet» a knitted graft due to its inherent porosity. This excludes its use in situations of total heparinization such as cardiopulmonary bypass or in emergency aneurysm surgery where precloeting is impractical. A gelatin impregnation has been developed which renders a knitted graft impervious to blood while retaining its handling and healing qualities, thus dispensing with the necessity to precloet [4]. This paper reports the laboratory evaluation of such a graft and the clinical outcome of the first 100 aortic bifurcation grafts implanted in patients at Glasgow Royal Infirmary.

MATERIAL AND METHODS

In vitro Biodegradation

Three graft sealants were studied for the extent and rate of removal. These were Gelseal and other manufacturers' grafts sealed with gelatin and collagen.

The grafts were washed in phosphate buffered saline, dried, and weighed aseptically. After incubation in PBS for various times, the sealant loss

* Gelseal, Vascutek, Ltd., Glasgow, Scotland.
was calculated. A similar experiment was performed with the addition of 2 μg/ml of trypsin (concentration 1 in 250) to the PBS incubation to evaluate enzymatic degradation of the sealant.

**In vitro thrombogenicity**

Samples of treated and untreated Dacron graft were inserted into a Sixma Chamber primed with oxygenated Krebs-Ringer solution which was then replaced with citrated oxygenated blood at 37°C. The graft was exposed to a blood flow rate of 104 ml/min for three minutes. The blood had been collected from healthy nonsmoking, male volunteers who were not receiving drugs with known antiplatelet activity. The samples were removed, rinsed in saline, fixed in cacodylate buffered glutaraldehyde and submitted to examination by scanning electron microscopy (SEM).

**Animal experiments**

The infrarenal aortas of 30 mongrel dogs (weight 15 kg-20 kg) were bypassed with 50-60 mm lengths of crimped graft, of 6 mm internal diameter. Twenty-two Gelseal and eight preclotted Triaxial 
\* grafts were implanted. The operative procedure was standardized with 100 IU/kg of intravenous heparin administered immediately prior to clamping the aorta. End-to-end anastomoses were made with continuous 5/0 monofilament polypropylene sutures. The heparin was not reversed at the end of the procedure.

The grafts were removed after predetermined time periods. After injection of 100 IU/kg of heparin intravenously, the graft was removed, cut open longitudinally and rinsed gently with saline. The gross morphology of the luminal surface was examined and the graft fixed for histology in 10% buffered formalin. Pieces of graft with surrounding tissue were embedded in paraffin wax, sectioned and stained with Martius' Scarlet and Blue, which differentiated between gelatin, fibrin and collagen. The rate of disappearance of the impregnate could then be followed.

**Clinical details**

In the 19 month period from August, 1985 to February 1987, Gelseal aortic bifurcation grafts were implanted in 100 patients. There were 79 men (age range 42 years to 81 years; mean 62.3 years), and 21 women (age range 51 years to 75 years; mean 60.9 years).

Surgery was performed for aneurysms in 41 patients, two of whom had ruptured their aneurysm. Eighty-five patients presented with claudication (< 150 meters), while 20 had rest pain and 12 had gangrene. All surgery, apart from the cases of aneurysm rupture, was performed in a standard manner using antibiotic prophylaxis and 5 000 units of intravenous heparin given intravenously prior to aortic clamping. The anticoagulant was reversed with protamine in a titrated dose at the end of the procedure. Intravenous volume loading was carried out by the anesthetist before declamping.

Proximal anastomosis was end-to-end in 62 and end-to-side in 38 patients. We performed 40 iliac and 160 femoral anastomoses, 54 of which included an extended deep femoral angioplasty. A simultaneous femoropopliteal graft was performed in seven patients.

The surgeon was asked to complete a questionnaire about handling and sutureability as well as noting blood loss through the graft and total blood loss. All patients had routine preoperative and post-operative blood investigations. Twenty patients were randomized to receive either an untreated Dacron aortic bifurcation graft or a Gelseal graft. These ten Gelseal grafts form part of this study. Venous blood samples were taken preoperatively and at frequent intervals until five weeks after surgery. Plasma C-reactive protein and alpha 1 acid glycoprotein were measured as they are significant indicators of an inflammatory reaction [5], and platelet counts were made up to nine days after surgery. Follow-up of patients has been 100%, the shortest time following graft implantation being two months in four patients.

**RESULTS**

**In vitro biodegradation**

The Gelseal impregnation was removed faster than the other sealants tested. The other gelatin and collagen sealants were either not totally removed by hydrolysis or disappeared at a much lower rate by means of enzymatic degradation (Figs. 1, 2).

**In vitro thrombogenicity**

There was an obvious difference in platelet adhesion to the sealed and unsealed Dacron (Figs. 3, 4). Gelseal had virtually no platelets adhering to it, suggesting the gelatin/Dacron composite had a much lower thrombogenic potential than that of bare Dacron.

**Animal experiments**

Two grafts thrombosed, one in each group (Table I). No significant differences in the appearance of the removed grafts were seen. The shorter term grafts had variable areas of thrombus, not related to the type of graft. After three months, the

\* Vascutek Ltd., Glasgow, Scotland.
Table I. - Implanted at sacrifice.

<table>
<thead>
<tr>
<th>Time</th>
<th>Gelseal (n = 22)</th>
<th>Triaxial (n = 8)</th>
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<tr>
<td>5 hours</td>
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<td>6 months</td>
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<tr>
<td>Total</td>
<td>21</td>
<td>7</td>
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</tbody>
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Luminal surface showed a typical pseudointima and was essentially free of thrombus.

The section of Gelseal graft at time 0 (Fig. 5), i.e., nonimplanted, shows that the gelatin is impregnated rather than coated. Gelatin can be seen filling the spaces between fiber bundles and even between the individual fibers. At two days the gelatin can still be seen quite clearly. There is a definite boundary between the gelatin and the superficial fibrin matrix enmeshing the red blood cells, although the absorption of gelatin has already begun. The precipitated graft shows much more cellular penetration of the interstices after the same time, but the flow surface remains largely fibrin. With time, the gelatin impregnation is progressively absorbed. At five days there is cellular material throughout the thickness of the graft wall, the impregnation having been largely absorbed with only small areas persisting. There is very little

![Fig. 1. Degradation of sealant by hydrolysis.](image)

![Fig. 2. Degradation of sealant by trypsin.](image)
Fig. 3. — SEM of Gelseal after blood contact (x 1000).

Fig. 4. — SEM of Dacron after blood contact (x 1200).

Fig. 5. — Gelseal prior to implantation (MSB x 200).

Fig. 6. — Gelseal ten days post implant. Luminal surface is at the top (MSB x 200).

Fig. 7. — Dacron ten days post implant. Luminal surface is at the top (MSB x 200).

Fig. 8. — Gelseal three months post implant. Luminal surface is at the top (MSB x 800).
difference between both types of graft beyond five
days. At ten days after implantation the grafts show
early responses typical of a Dacron graft (Figs. 6,
7). Any gelatin remaining is confined to small
islands and at 21 days there is no difference
between the grafts.

The histology at three months shows a typical,
well-incorporated Dacron graft. The pseudointima
is thin and is not proliferating (Fig. 8).

Clinical results

There was no operative mortality. One patient
died of a myocardial infarct four months after sur-
gery. There were no discernible graft-related prob-
lems. Two patients required thrombectomy of a
graft limb-one which occluded six hours after sur-
gery due to atheromatous embolism and another
occluding one week after surgery due to an intimal
flap dissection in the deep femoral artery.

There were no postoperative wound infections and
two groin seromas while 14 patients had a postope-
rative chest infection. The mean postoperative
hospital stay was 10.5 days.

There was no measurable blood loss through the
graft although a few prostheses did « sweat » with
minimal blood staining. The details for blood loss
are shown in Table II. It is important to note that 74
(74%) of the patients did not require blood trans-
fusion during or after surgery.

Acute phase protein response

The peak mean value of plasma C-reactive pro-
tein for the sealed group was 226.3 ± 49.2 mg/l
and 221.3 ± 56.4 mg/l for the unsealed group. The
corresponding values for alpha 1 acid glycoprotein
were 1.49 ± 0.25 g/l and 1.44 ± 0.3 g/l. The plasma
C-reactive protein response returned more
quickly to normal, only two patients in each group
having a slightly elevated response after four
weeks, while the value for alpha 1 acid glycopro-
tein remained elevated at five weeks for the same
two patients. There was no significant difference
in the acute phase protein response of either group.
Similarly the platelet numbers showed the same
trend in each group, reaching their lowest value of
145000 by the third postoperative day and return-
ing to normal by the ninth day.

DISCUSSION

No artificial arterial prosthesis can be totally
non-thrombogenic or biocompatible. Current opin-
ion suggests that for the replacement of wide bore
and high flow velocity arteries, the optimal substi-
tute is a knitted graft with low internal and relati-
ively high external velour [2, 3]. A graft conform-
ing to these criteria, the Vascutek Triaxial, has
been extensively used for aortic surgery [3], with
excellent results. A major advance would be to
bestow zero porosity on such a graft without
detracting from the handling and healing proper-
ties of the knitted grafts and provide an « off the
shelf » prosthesis for most situations.

Gelatin, a mammalian protein with a proven
safe record during its use in plasma expanders [6]
and ease of availability due to its commercial pro-
duction, was chosen as the impregnate for a
Dacron/composite prosthesis based on the Vascu-
etek Triaxial. It is vital that the sealing protein is
non-thrombogenic and gelatin has been described
as blood compatible by the Cleveland Clinic Left
Ventricular Assist Device Group [6]. Tests of in
vitro thrombogenicity revealed virtually no plate-
let adhesion to Gelseal (Fig. 3). Further, the chem-
istry of gelatin allowed control of the rate of
removal of the graft impregnation by alteration of
formaldehyde cross linkage density. In vitro biode-
gradation experiments revealed a predictable rate
of gelatin removal which was faster and more
complete than with non-modified gelatin or colla-
gen (Figs. 1, 2).

This was confirmed in the histology of grafts
removed from the animal model which demon-
strated substantial gelatin removal by five days and
little or no gelatin remaining after ten days (Fig. 6).
The efficient disappearance of the gelatin impreg-
nate allows early cellular ingrowth and does not
significantly alter or delay normal healing, which
might be the case with sealants which cover the
Dacron for longer periods.

Our clinical experience with Gelseal was very
similar to our previous experience with Vascutek
Dacron [3]. There was 100% patency at 21
months, no false aneurysm formation or dilatation
problems. The postoperative morbidity was well
within expected norms for vascular surgery. The
subjective response of the surgeons (J.K.D.,
J.G.P.), was that the graft handled well and did not
leak although it did feel slightly stiffer than non-
treated Dacron. This can be overcome easily prior
to implantation by briefly immersing the graft in
saline which renders it more pliable. It held
braided Dacron and monofilament sutures easily
with no bleeding from stitch holes.

Perhaps the most striking feature of the graft
related to perioperative blood loss. There was no
measurable graft related blood loss and the average
loss for all patients, including the two with ruptured aneurysms, was 670 ml. The decision to transfuse was that of the anesthetist and 26 patients had blood replacement averaging 860 ml. The relatively small number of patients needing transfusion may have advantages in terms of cost saving by reducing the amount of blood used when inserting a knitted graft. It certainly has advantages to both patient and surgeon because it reduces the risk of transmitting blood borne viral disease. The acute phase protein response was studied since the introduction of protein to the graft may have produced an excessive acute or prolonged inflammatory response. This would have been reflected in an increase of serum acute phase proteins. There was no significant difference between the acute phase protein response in patients receiving Gelseal Triaxial or Triaxial grafts untreated with gelatin. The two patients in each group who maintained a slightly elevated plasma C reactive protein response and a prolonged alpha 1 acid glycoprotein response were identifiable as patients having other problems such as chest infection or superficial wound infection. A knitted Dacron graft impregnated with gelatin does not alter inflammatory response which occurs with an unsealed Dacron graft.

CONCLUSION

This study demonstrates that a Dacron composite graft impregnated with gelatin eliminated the need for preclotting and is safe in clinical use. The rapid removal of the gelatin impregnation does not delay normal healing.

REFERENCES


