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The effect of absorbable gelatin sponge on experimental microvascular anastomoses

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Summary—Following a pilot study a controlled trial was carried out to assess the effect of absorbable gelatin sponge on reducing the bleeding from microvascular anastomoses of the femoral arteries and veins of rats. A model was used which approximated to the clinical situation and the bleeding time was significantly reduced in arteries and veins. There was no significant difference in patency rates or wound infection.

The success of a microvascular procedure is entirely dependent on the patency of the anastomosis. This is affected by:

(i) The anastomotic technique.
(ii) The diameter of the vessel.
(iii) The thrombogenic potential of the blood.

One problem which may develop after release of the vascular clamps is that leakage of blood between the sutures may continue, especially if anti-thrombotic agents have been used, *e.g.* heparin, or the more recently employed epoprostenol. To insert another suture into the anastomosis may be hazardous if the operative field is obscured with blood, and an incorrectly placed suture may narrow the vessel or cause intimal damage, predisposing to thrombosis.

If leakage continues unchecked, haematoma around the anastomosis or in the vessel wall may produce pressure on the vessel, cause intravascular thrombosis by the release of thrombogenic material and predispose to infection.

One solution could be to apply a haemostatic material to the anastomsis, but this must be entirely safe and not predispose to intravascular thrombosis.

Absorbable gelatin sponge (Sterispon®) is widely used in surgery for haemostasis in wounds, particularly for small vessel bleeding from raw areas. The aim of the present study was to investigate the properties of this material in preventing leakage from microvascular anastomoses, and to determine its effects on vascular patency.

Materials and methods

The study was performed on the femoral vessels of adult male rats and consisted of a pilot study followed by a controlled trial.

Pilot study

Ten rats were studied:

(i) To enable the operator to standardise his technique.
(ii) To determine the number of 10/0 nylon sutures per femoral artery or vein required to give a degree of anastomotic leakage which could be accurately observed without causing thrombosis or exsanguination.
(iii) To ascertain the amount and mode of application of the gelatin sponge which would be most likely to produce haemostasis.

The pilot study determined the following:

(i) Six evenly spaced arterial sutures and eight venous sutures produced a bleeding time of at least a minute in most circumstances.
(ii) The individual animals varied considerably in their bleeding and coagulation times and therefore it was decided that each animal should act as its own control.
(iii) Absorbable gelatin sponge applied to an anastomosis after release of the clamps tended to float away; therefore a standard piece of sponge 5 × 5 mm in area and 1 mm thick was

laid on the anastomosis immediately prior to release of the clamps.

(iv) Wrapping the sponge around the anastomosis appeared to constrict it: several such anastomoses occluded.

Experimental method

As a result of the pilot study, the following experimental method was established for the controlled trial:

After weighing the animal, anaesthesia was induced by ether and continued with oxygen, nitrous oxide and halothane. Systemic heparin was not used.

Each groin was shaved, and the relevant femoral vessel exposed using an operating microscope and microvascular instruments and techniques as described by O'Brien (1977). The diameter of the unclamped vessel was measured using a paper grid. The vessel was divided between microvascular clamps and the cut ends trimmed and irrigated with heparinised normal saline solution (1000 units per litre).

The vessel was re-anastomosed using 10/0 nylon (Ethilon) on a 75 micron 5.1 mm curved round-bodied needle. Six evenly-spaced sutures were used for an artery and eight for a vein. For anastomoses treated with absorbable gelatin sponge, a 5 × 5 mm piece of this material, 1 mm thick (Sterispon No. 3), was placed over the suture line. The clamps were then released (distal one first) and the time recorded until bleeding stopped, the area around the anastomosis being gently swabbed to keep the anastomosis in view.

After haemostasis, the skin was closed using continuous polyglycolic acid sutures, and the animals caged together after recovery.

It was decided to review the anastomoses of some of the animals within 2 weeks postoperatively to exclude the effect of recanalisation (Hayhurst and O'Brien, 1975), the remainder to be reviewed at 4 weeks to observe any delayed effects of use of the gelatin sponge.

At the appropriate time, the animal was re-anaesthetised as above and the patency of the anastomosis ascertained by the two-forceps technique described by O'Brien (1977). Any fibrosis or infection around the anastomosis was also noted. The animal was then sacrificed. Some of the anastomoses were examined histologically (Figs 1 and 2).

Controlled trial

Twenty-four adult male white or hooded rats, weight 280 to 475 gm (mean 385 gm) were studied. All the anastomoses were performed by the same operator. Each animal had an anastomosis performed on each side: one side was treated with absorbable gelatin sponge, the opposite side was a control. Arterial and venous anastomoses were not mixed in the same animal. Random grouping was made as shown in Table 1, in relation to the side treated, type of vessel and priority of the treated anastomosis, to avoid operator bias.

Eight of the animals had their anastomoses reviewed within 2 weeks postoperatively. The remaining 16 had their anastomoses reviewed 4 weeks postoperatively.

Differences in the bleeding times and patency rates between the two groups were compared using Student's t-test.

Results

The results are summarised in Table 2.

The two groups had comparable vessel diameters.

The mean bleeding time was greatly reduced in the group treated with absorbable gelatin sponge, by 30% in the case of arteries and by 59% in the case of veins.

However, there were some observed bleeding times in the treated group far in excess of the mean for the control group (maximum bleeding times in the treated group were 210 seconds for an artery and 105 seconds for a vein: control group ranges were 60–225 seconds for arteries and 0–110 seconds for veins). The difference in the bleeding times in the two groups is statistically significant ($p = < 0.05$).

Patency rates for arterial and venous anastomoses were reduced in the group treated with absorbable

Table 1 Plan of groupings of animals for controlled trial

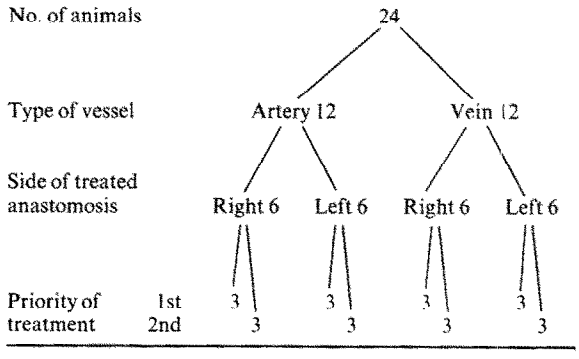

Fig. 1

Figure 1—Rat left femoral artery and vein (× 40) 7 days after arterial anastomosis (control). Note the sutures in the arterial wall(s) and thin fibrous tissue layer over the vessels (f).

Fig. 2

Figure 2—Rat right femoral artery and vein (× 60) 7 days after arterial anastomosis (same animal as Fig. 1). The vessels have been covered with a layer of absorbable gelatin sponge (ags) over which has developed a thin fibrous tissue layer (f).

Table 2 Table of results

	Treated group (absorbable gelatin sponge) 24		*Control group 24*
No. of anastomoses	(12 arteries, 12 veins)		(12 arteries, 12 veins)
Mean vessel diameter	Artery	0.9 mm	0.92 mm
	Vein	1.66 mm	1.64 mm
Mean bleeding time (range in brackets)	Artery	89.2 sec (0–210 sec)	127.5 sec (60–225 sec)
	Vein	32.1 sec (0–105 sec)	78.8 sec (0–110 sec)
Patency rate	Artery	9 (75%)	10 (83.3%)
	Vein	10 (83.3%)	12 (100%)
Infection		3 (all occluded)	3 (1 occluded)
Perivascular fibrosis (see text)	Mild	11 (none infected or occluded)	5 (none infected or occluded)
	Severe	3 (all occluded, 2 infected)	3 (1 occluded, 2 infected)

gelatin sponge but the numbers were too small to obtain any statistical significance. For occluded vessels, there was no relationship between bleeding time and subsequent occlusion.

The incidence of wound infection in each group was the same (three cases), and was associated with interference of the wound by the animal. In the treated group each infected case was associated with occlusion, but only one in the control group.

Perivascular fibrosis (*i.e.* macroscopically visible fibrous disposition around the anastomoses, but not distorting the vessels or hindering dissection) was present in 45.8% of the treated group and 20.8% of the control group, and not associated with vascular occlusion.

A more severe form of fibrosis, making dissection difficult, was present in three cases in each group, all being associated with occlusion in the treated group and one being occluded in the control group. Two out of the three cases in each group were infected.

There was no great difference in patency rates between animals which had their anastomoses examined within 2 weeks postoperatively (one treated artery, one treated vein and one control artery occluded) and those examined 4 weeks postoperatively (two treated arteries, one treated vein and one control artery occluded).

Discussion

The need for an effective haemostatic agent in microvascular surgery has long been appreciated and many experimental studies have been performed using the rat femoral vessel model.

McLean and Buncke (1973) claimed that the application of Saran wrap (plastic cling-film) around the anastomosis until haemostasis was complete improved patency rates.

The wrap-around principle was further modified by Nomoto *et al.* (1974) by applying silicone rubber cuffs to the anastomoses while clotting was inhibited by magnesium sulphate, to prevent leakage from the suture line. Patency rates of 100% were claimed.

Gelatin powder, oxidised cellulose, microfibrillar collagen and polytetrafluorethylene (PTFE) cuffs were compared by Achauer *et al.* (1982) but for the first three materials only three sutures per anastomosis were used and many animals exsanguinated on release of the clamps. Microfibrillar collagen did give some improved results; PTFE cuffs were more dramatic in reducing bleeding time but produced vessel wall distortion.

In a controlled trial, Colen and Mathes (1982) showed that topical microcrystalline collagen did not adversely affect patency rates. Absorbable gelatin sponge (Sterispon®) is readily available and has been effective in controlling capillary and venous oozing and, more recently, in therapeutic embolisation (Allison, 1979). It is completely absorbed by phagocytosis. It does not cause adhesion formation in the peritoneal cavity (Raftery, 1980) or synovium (Austin and Walker, 1979).

The present study was designed to approximate to the clinical situation of a leaking microvascular anastomosis. The results show that absorbable

gelatin sponge reduced the bleeding time in a series of experimentally produced leaking microvascular anastomoses. This reduction was more striking in veins. There was, however, wide variation between individual bleeding times in both treated and control groups. There was a small decrease in the patency rate for anastomoses treated with absorbable gelatin sponge. The numbers concerned are very small and these differences were not significant. Nevertheless, since the success or failure of a microvascular procedure is solely dependent on the patency of the anastomosis, these findings suggest that absorbable gelatin sponge should not be applied routinely to microvascular anastomoses.

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