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Trisacryl Gelatin Microspheres for Therapeutic Embolization, II: Preliminary Clinical Evaluation in Tumors and Arteriovenous Malformations

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PURPOSE: To evaluate an embolic agent that is precisely calibrated, perfectly spherical in shape, and soft but nonresorbable for use in the embolization of vascular disease of the head, neck, and spine in humans. **METHODS:** We used supple, hydrophilic, and calibrated trisacryl gelatin microspheres 200, 400, 600, 800 and 1000 μm in diameter for superselective embolization in 105 patients (27 tumors, 14 facial arteriovenous malformations [AVMs], 37 spinal cord AVMs, 21 cerebral AVMs, and 6 miscellaneous diseases). We used particles in 200 to 600 μm in diameter for tumors and for facial AVMs, particles 400 to 600 μm in diameter for spinal cord AVMs, and particles over 1000 μm in diameter for cerebral AVMs. **RESULTS:** Delivery of the embolic material was easy: microspheres did not aggregate and catheters did not become obstructed by particles. It was possible to control the embolization through precise accounting of the amount of microspheres and matching of the particle size to the size of the pathologic vascular network. **CONCLUSION:** The microspheres are easy to use and allow precise control of the embolization procedure. Their physical characteristics make them a safe embolic agent.

Index terms: Arteriovenous malformations, embolization; Interventional materials, particles and microspheres

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Embolization microspheres are occlusive agents that are easier to use than nonspherical embolic agents (1, 2). Microspheres studied up to now have been either resorbable or nonresorbable. Resorbable microspheres are made of gelatin (3), albumin (4), polysaccharides (dextran) (1, 2), starch (5), ethyl cellulose (6, 7), and poly(D,L lactide/glycolide) copolymer (8, 9). Nonresorbable microspheres have been studied less extensively. These are made of hard material such as glass (10), wax (11–13), silicone (14–16), or polystyrene (2, 17), and,

more rarely, supple material such as polymethyl methacrylate (18) or poly(2-hydroxyethyl methacrylate) (19).

In a preliminary study, we performed experimental embolizations in animals by using supple nonresorbable hydrophilic microspheres made of trisacryl gelatin for which cell cultures had previously proved their biocompatibility. These microspheres were shown to be effective as vascular occlusive agents (20).

In the present study we conducted a clinical evaluation of these trisacryl gelatin microspheres in patients who had tumors or facial, spinal cord, or cerebral arteriovenous malformations (AVMs).

Materials and Methods

Microsphere Preparation

Trisacryl gelatin microspheres were prepared by reverse emulsion (N.E. Brown et al, French patent no. 7723223; 1977) (21, 22). A mixture of *N*-acryloyl-2-amino-2-hydroxymethyl-propane-1,3 diol and *N,N'*-methyl-

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TABLE 1: Patients embolized with microspheres

Disease	No. of Patients	No. of Embolizations
Intracranial tumors	6	13
Extracranial tumors	18	18
Spinal tumors	3	3
Facial AVMs	14	14
Spinal cord AVMs	37	51
Cerebral AVMs	21	23
Miscellaneous	6	6
Total	105	128

Note.—AVMs indicates arteriovenous malformations.

ene-bis-acrylamide and gelatin was emulsified in paraffin oil. After polymerization, the microspheres were wet-sieved with standardized square-mesh sieves (Bioblock, Illkirch, France); calibration was controlled by means of measurement performed under a microscope. The microspheres were evenly distributed into sterile, apyrogenic, 10-mL glass flasks and suspended in sterile water used for injection. The flasks were stopped, capped, and sterilized. Bacteriologic control was done by direct culture of the nutritive environment for aerobic and anaerobic germs and yeasts. The check for pyrogenic substances was performed with the limulus amoebocyte chromogenic test on the suspension liquid containing the microspheres (23).

Several sizes of microspheres were prepared for this clinical evaluation: 200 ± 50 , 400 ± 50 , 600 ± 100 , 800 ± 100 , and $1000 \pm 100 \mu\text{m}$. During embolization, microspheres of a particular size were chosen according to the size of the vascular network. The injected volume varied according to the type of disease, from a few spheres for spinal cord AVMs to several hundred spheres in cases of tumors.

Before injection, the contents of the flasks were poured into a stainless steel cup and mixed with contrast medium (half and half or one third/two thirds) and the desired quantity of microspheres was taken from the cup after gentle shaking by hand. The syringe was attached directly to the catheter, and injection was monitored under fluoroscopic control.

Patients

All patients ($n = 105$) were treated between 1987 and 1993. Diseases included AVMs (principally of the spinal cord) and hypervascular tumors (Table 1). In cases of facial AVMs, embolization was performed as a presurgical or an analgesic intervention. In cases of spinal cord AVMs, embolization was in all cases performed as a treatment to improve clinical signs and symptoms or to prevent recurrent hemorrhage in patients who were not candidates for surgery. In cases of cerebral AVMs, embolization was performed either presurgically or as a symptomatic intervention intended to correct progressive neurologic deficits when acrylic glue could not be used without risk. In cases of hypervascular tumors, embolization was performed as

presurgical therapy to limit blood loss or, in some cases, as an analgesic therapy (spinal metastasis).

For all patients, radiologic evaluation consisted of computed tomography (CT), magnetic resonance (MR) imaging, and global and selective angiography before embolization, superselective angiography during embolization, and global and selective angiography control after embolization.

Catheters

Embolizations were performed with different types of catheters: 3.6F catheter, $0.6 \times 1 \mu\text{m}$ Pursil catheter, Magic STD (all from Balt, Montmorency, France), and Tracker 25 and Tracker 18/12 (Target Therapeutics Inc, San Jose, Calif).

Embolization Techniques

The extent of devascularization varied according to the disease. For tumors, devascularization was performed in one session in the majority of cases, or in several stages for intracerebral tumors. In cases of spinal cord AVMs, devascularization was performed after superselective catheterization. In cases of facial AVMs, devascularization was intended to be as total as possible with the complete angiographic disappearance of the AVM. In cases of cerebral AVMs, embolization was aimed at obtaining a devascularization of the catheterized pedicle or, failing that, a reduction in arteriovenous shunting.

All patients were followed up clinically after embolization to monitor any onset of pain, fever, deficit, or change in clinical state.

Histology

For 22 patients (16 tumors and 6 facial AVMs), embolization was followed by surgery with histologic verification of the pathologic process (Table 2). For the 16 tumors, we compared the results obtained in an initial group ($n = 8$) treated with microspheres that ranged narrowly in size (200 ± 50 , 400 ± 50 , 600 ± 100 , 800 ± 100 , and $1000 \pm 100 \mu\text{m}$) with results obtained in a second group ($n = 8$) embolized with microspheres that ranged widely in size ($500 \mu\text{m} \pm 300$), made possible by use of a special sieving technique. We evaluated three criteria: 1) vascularization of tumors before and after embolization, 2) surgical benefit (feasibility of surgery, volume of compensated blood loss during surgery), and 3) histologic data (presence and size of necrosis patches, and number of microspheres on the histologic slide of the tumor).

Results

Conditions and Ease of Use

Because of their suppleness, the microspheres could be injected into catheters that

TABLE 2: Types of tumors embolized that were confirmed by histologic findings

Tumor	No. of Tumors Embolized	No. of Embolizations with Histologic Confirmation
Meningioma	4	3
Nasopharyngeal fibroma	8	5
Spinal metastasis (kidney carcinoma)	3	1
Infratemporal fossa fibrosarcoma	1	1
Hemangioblastoma	2	2
Glomus tumor	6	1
Temporoauricular sarcoma	1	1
Neurofibroma	1	1
Hemangiopericytoma	1	1
Total	27	16

had internal distal diameters that were less than the diameters of the particles. No aggregation of particles or any obstruction of catheters by particles was noted when using the microspheres. Of a total of 128 procedures, no proximal vascular occlusion or any backward surge of microspheres causing the involuntary occlusion of healthy branches was noted.

Angiographic and Clinical Results

The degree of angiographic devascularization by embolization varied among spinal cord AVMs, cerebral AVMs, facial AVMs, and hypervascular tumors (Table 3).

In cases of spinal cord AVMs ($n = 37$), embolization produced the following results: total devascularization in 21% of cases ($n = 8$) (Fig 1), significant reduction of the flow through the malformation (greater than 50% devascularization) in 45% of cases ($n = 17$), and partial devascularization (less than 50% devascularization) in 27% of cases ($n = 10$). In all cases the degree of devascularization was dependent on

the maintenance of sufficient flow in healthy branches (anterior spinal artery, posterior spinal artery). Devascularization was most often obtained by injecting a few spheres in a controlled flow (in general from between 2 to 10 spheres) under fluoroscopic monitoring. In 5% of cases ($n = 2$), no devascularization was noted; this occurred in arteriovenous fistulas whose shunts were too big to be occluded by the biggest microspheres at our disposal. The diameter of the microsphere used most often was $600 \mu\text{m} \pm 50$. In no case was there any involuntary obstruction of normal medullary arterial afferents, and there were no clinical complications.

In cases of cerebral AVMs ($n = 21$), embolization produced the following results: a global slowing of the flow through the malformation or selective devascularization in four (19%) of 21 cases and ineffective embolization in 17 (81%) of 21 cases. The number of microspheres used was generally on the order of a few hundred. The most commonly used microsphere had a diameter of about $1000 \mu\text{m}$. There were no

TABLE 3: Disease type and effective size of microspheres

Type of Disease	Efficient Size Range of Microspheres, μm	No. of Microspheres Used	Angiographic Result
Tumors ($n = 27$)	200–400	100–300	Total occlusion of pedicle
Cerebral AVMs ($n = 21$)	400–600	100–500	Flow reduction
Spinal AVMs ($n = 37$)	800–1000	10–50	Partial devascularization
Facial AVMs ($n = 14$)	400–600	50–100	Six total devascularizations (43%) Eight partial devascularizations (57%)

Note.—AVMs indicates arteriovenous malformations.



Fig 1. Spinal cord arteriovenous malformation (AVM) in a 32-year-old man with a dramatic paraparesis due to Cobb syndrome.

The spinal cord malformation (arrow in A) was fed by the anterior spinal artery (B). Embolization with 600- μ m-diameter microspheres allowed a satisfactory occlusion of the afferences to the spinal cord AVM, with preservation of the descending branch of the anterior spinal artery (long arrow in B). After embolization, paraparesis totally disappeared. Note the reappearance of the ascending branch of the anterior spinal artery (short arrow in B).

complications. Clinically, improvement occurred in 52% of cases ($n = 11$), even in those instances in which embolization had not led to modification of the angiographic aspect of the AVM. Beneficial clinical results lasted on average 6 months (range, 24 hours to 3 years).

In cases of facial AVMs ($n = 14$), total angiographic devascularization was obtained in 6 patients, all of whom underwent a second operation (Fig 2). Gross total excision was achieved in these cases. In 8 other patients, with extended and complex AVMs, only partial devascularization was obtained by embolization. The diameter of the microspheres used was generally between 200 and 400 μ m. There were no complications. The patients suffered only a local temporary inflammatory syndrome involving pain, heat, swelling, and redness, probably due to secondary thrombosis of the malformation.

In cases of hypervascular tumors, devascularization was always performed progressively and checked by fluoroscopic monitoring. The diameter of the microspheres used varied from 200 to 800 μ m. In the majority of cases, 600 μ m was effective. The number of microspheres used was generally a few hundred. All catheterized and embolized pedicles were totally devascularized. However, complete devascularization of the tumor could be obtained in only 20% of cases, owing to vascular supply to the tumor from perforating arteries arising from noncatheterizable or nonembolizable arteries (Fig 3).

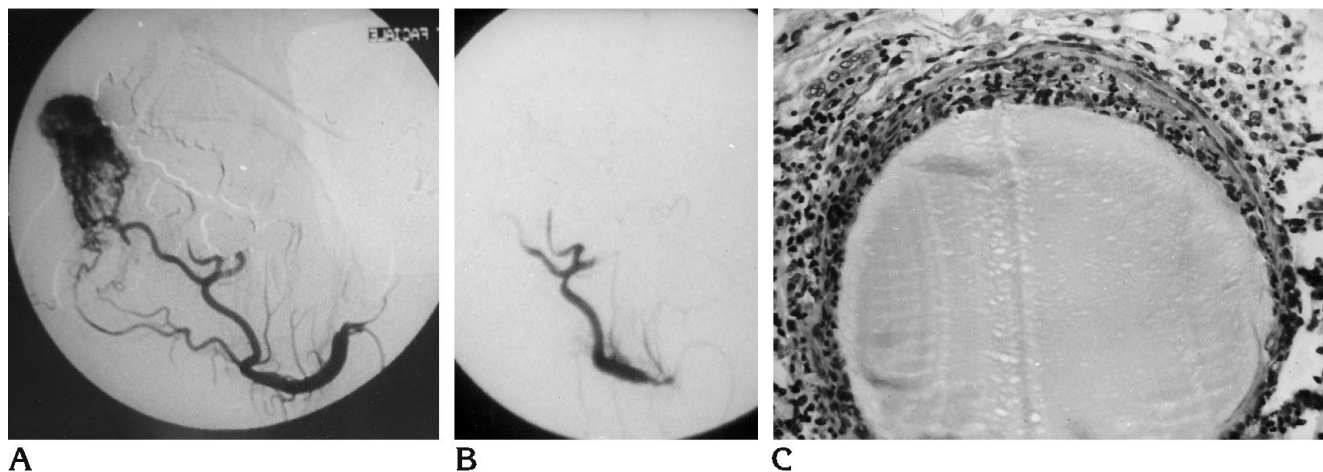


Fig 2. Facial (inferior lip) arteriovenous malformation (AVM) in a 25-year-old man.

A, The malformation is fed by the facial artery.

B, Embolization with calibrated microspheres (200 μ m) allowed a progressive and complete devascularization. Surgery was simple, without significant bleeding.

C, Histologic study confirmed the occlusion of the vascular network and showed a moderate inflammatory reaction.

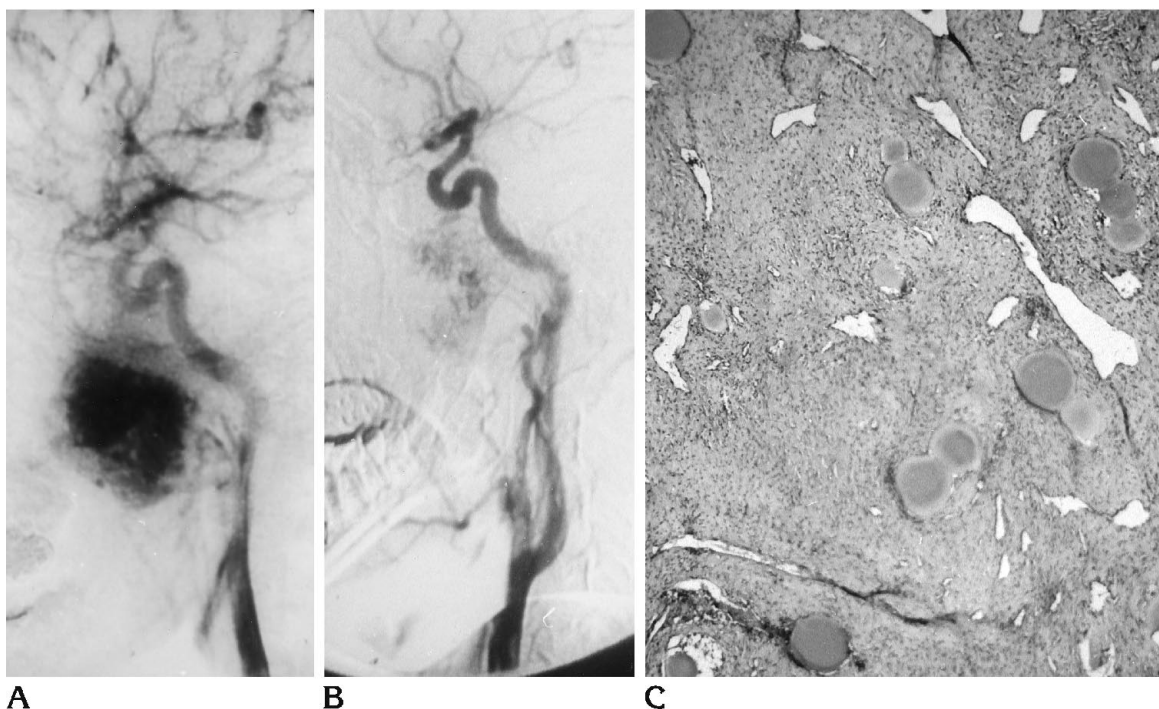


Fig 3. Nasopharyngeal fibroma in a 16-year-old boy.

A, The hypervascular tumor is fed mainly by the external carotid artery, but is partially fed by the internal carotid artery.

B, Devascularization, after embolization with 400- and 600- μ m-diameter microspheres, was almost total. The tumor was fed only by transosseous arteries from the internal carotid artery. Surgery was simple.

C, Histologic study shows a homogeneous distribution of microspheres in the tumor.

Most of these cases were nasopharyngeal fibromas. Embolization was tolerated without immediate complication; however, early in our study two cases of massive meningioma, vascularized by branches of internal and external carotid arteries, were extensively embolized (subtotal devascularization) in one session, producing tumoral necrosis leading to cerebral edema and aggravation of preexisting neurologic symptoms. These meningiomas had been embolized with small-diameter microspheres (200 μ m). The disorders of consciousness linked to the edema and cerebral compression receded in these two cases and the patients were able to undergo surgery satisfactorily after a suitable interval. Surgery was extremely rapid in both cases. Neither blood transfusions nor blood derivatives were required.

For 16 of the 19 hypervascular tumors removed surgically after embolization, angiographic devascularization seemed to be sufficient, whether performed with microspheres of widely ranging sizes (± 300 μ m) or with those of narrowly ranging sizes (± 50 μ m to ± 100 μ m). No difference in angiographic devascularization was noted among the groups. The

amount of replacement fluids required during surgery was, on average, 3 L (0.5 to 8.5 L), and duration of surgery was, on average, 7.5 hours.

Histologic Effects

For all histologic samples from facial AVMs ($n = 6$) and hypervascular tumors ($n = 16$) we observed similar phenomena related to the microspheres: moderate inflammatory reaction around the microspheres, absence of microsphere degradation, and presence of a few endothelial cells on the microspheres' most external layer. In no case were cellular alterations indicative of material toxicity (cellular deterioration or death) close to the microspheres. These histologic data related to toxicity were independent from data related to the necrosis induced by embolization.

For the 16 tumors surgically removed after embolization, more particles in the tumor were found when embolization had been performed with microspheres that ranged narrowly in size rather than widely. This difference is statistically significant ($P < .02$, Yates's corrected χ^2). Intratumoral necrosis was bigger and more fre-

TABLE 4: Results of correlation (angiography, surgery, and histology) according to the type of microspheres used for embolization

	Large Size Range, $\pm 300 \mu\text{m}$ (8 cases)	Narrow Size Range, $\pm 50 \text{ to } \pm 100 \mu\text{m}$ (8 cases)
Satisfactory devascularization	8	8
Surgical benefit	8	8
Histology		
No. of intratumoral microspheres		
<5	6 (75%)	1 (12.5%)
>20	2 (25%)	7 (87.5%)
Necrosis		
Yes	0 (0%)	6 (75%)
No	8 (100%)	2 (25%)

quent when embolization was performed with a narrow size range of microspheres (Table 4). This difference was equally significant ($P < .02$, Yates's corrected χ^2).

In the first two cases of meningioma mentioned above, in which total occlusion was obtained with small-diameter microspheres, extensive necrosis was found histologically.

Discussion

Choice of Microsphere Diameter

In the present study we noted that the operator would often use bigger and bigger microspheres successively in the same embolization session, and that there was a difference between the diameter initially requested and the diameter that finally permitted good vascular occlusion. This discrepancy was $400 \mu\text{m}$ on average. For example, embolization that started with microspheres of $200 \text{ to } 400 \mu\text{m}$ diameter often ended with microspheres of $600 \text{ to } 800 \mu\text{m}$ diameter to obtain a satisfactory occlusion. This discrepancy between the diameter requested at the start of the procedure and the effective final diameter was possibly due to the user's initially choosing the diameter of the microsphere as if choosing the caliber of polyvinyl alcohol particles. The latter have a much wider size distribution owing to the irregularities of particle shape that allow formation of particle agglomerates. Hence, the larger particles and agglomerates occlude vessels that have a larger diameter than that of the expected particle size. Inversely, when using calibrated microspheres, there is a much more accurate correspondence between the diameters of the particles and those of the

occluded vessels, an advantage when it is necessary to target occlusion.

Interest of Calibration and Distality

Embolization is difficult to target with irregularly shaped particles, which are also difficult to calibrate by sieving (24). Hence, there is a risk of involuntary embolization of small normal arteries by small particles (25–27) as well as a risk of catheter occlusion (28) or of too proximal occlusion of vessels (2) by large-sized particles or agglomerates.

Optimal Size Related to Disease

It is only with calibrated microspheres that we can accurately approach the angioarchitecture and size of vessels within the pathologic network of AVMs and tumors. In our study, the use of calibrated microspheres allowed us to show that the effective diameter had to be adapted to the disease (Table 4). In cases of AVM, the effective diameter was in the region of $200 \text{ to } 400 \mu\text{m}$ for facial AVMs; it was larger for spinal cord AVMs ($500 \text{ to } 700 \mu\text{m}$). In cases of cerebral AVMs, the optimal size seemed to be larger than $1000 \mu\text{m}$.

In cases of hypervascular tumors, embolization is useful because it facilitates or enables surgery by limiting blood loss (29, 30). Tissue effects of embolization are linked directly to particle size, as previously suggested (31). Our results agree with published data (1, 5, 9); the smaller the particles' diameter, the easier it is to obtain necrosis of the tumor. In certain cases, this tumoral necrosis can be dangerous, notably in compressive intracerebral tumors such as meningioma, especially if they are vascularized by the external and internal carotid arteries (32, 33). In such cases, it is justified to perform tumoral devascularization in several steps in order to avoid acute, massive necrosis. Such a controlled devascularization is made easier when using calibrated microspheres, the volume of devascularized tumor being controlled by the number of injected microspheres.

Size and Safety

With calibrated microspheres, safety can be maintained even when embolization is difficult to perform. This is the case for spinal cord AVMs, in which embolization particles must be

perfectly calibrated (34). Spinal cord AVMs are normally fed by dilated central arteries stemming from a dilated anterior spinal artery. The ascending branch of the anterior spinal artery and its descending portion beyond the AVM have normal diameters and will be seen poorly or not at all as a result of the preferential flow toward the AVM (35). It is necessary to use microspheres with a diameter that is at once equal to the shunts, smaller than the anterior spinal artery feeding the AVM, and larger than the descending branch of the anterior spinal artery (Fig. 1).

In cases of tumors in which it is necessary to avoid damaging normal small branches fed by vessels going to the tumor (dangerous anastomosis) (29), it suffices to choose calibrated microspheres large enough to prevent passage through healthy branches.

Conclusion

The microspheres made for this study are well calibrated, thus allowing good control of occlusion sites and of the correspondence between the size of the particles and that of the AVMs and vascular tumor networks.

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