Experimental Models of Bifurcation and Terminal Aneurysms: Construction Techniques in Swine

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Summary: We report our preliminary experience in the surgical construction of five experimental bifurcation and terminal aneurysm models in swine. We used unilateral neck vessels to construct models in which the relative directions and sizes of the parent and daughter arteries could be varied by surgery, resulting in aneurysms with high morphologic similarity to human intracranial lesions. Steps in the construction of each model are detailed.

Index terms: Aneurysm; Interventional neuroradiology, models; Animal studies

Intracranial saccular aneurysms are of considerable importance in cerebrovascular pathology, being the main cause of nontraumatic subarachnoid hemorrhage (1). Further insight into the behavior of these lesions and their clinical management depends to some extent on the availability of research models that can faithfully reproduce their gross morphology and pathophysiologic characteristics in experimental settings.

Naturally occurring saccular aneurysms and, hence, potential natural models are extremely rare in animals (2). Therefore, the need has arisen in the last few decades to create various mathematical, computer, in vitro, and in vivo models to achieve a degree of reproducibility in scientific experiments regarding these lesions. The most common of these models are those surgically constructed in laboratory animals. To date, in vivo construction of lateral aneurysms has received the most attention despite the overwhelming preponderance of bifurcation and terminal (simulating basilar tip and internal carotid termination type) aneurysms in humans. Of the several methods described in the literature for creating bifurcation aneurysms, the most commonly used entails bilateral neck dissection and manipulation of both common carotid arteries (3). This may require prolonged surgery and results in aneurysms situated at bifurcations with similar-sized daughter and parent arteries. This configuration is infrequently found in human intracranial aneurysms. Furthermore, two previous attempts at vein pouch terminal aneurysm construction (4, 5) have reported either high animal mortality or less than adequate configuration of the resultant model.

In this article, we describe our initial experience in the surgical construction of several bifurcation aneurysm and terminal aneurysm models, conceived with a view to: 1) shortening and simplifying surgical procedures by using vessels on one side of the neck only; and 2) reproducing the gross morphologic characteristics of human saccular aneurysms as accurately as possible, especially regarding the relative directions and sizes of the aneurysm sac and the parent and daughter arteries.

Materials and Methods

All animal experimentation was conducted in accordance with policies set by the local university chancellor's animal research committee and National Institutes of Health guidelines. Five new types of aneurysms (three bifurcation aneurysms, and two terminal aneurysms) were constructed in the necks of adult Red Duroc swine. These animals weighed 30 to 40 kg, were of mixed sex, and were maintained on a standard laboratory diet. After an overnight fast, each swine was premedicated with intramuscular 20

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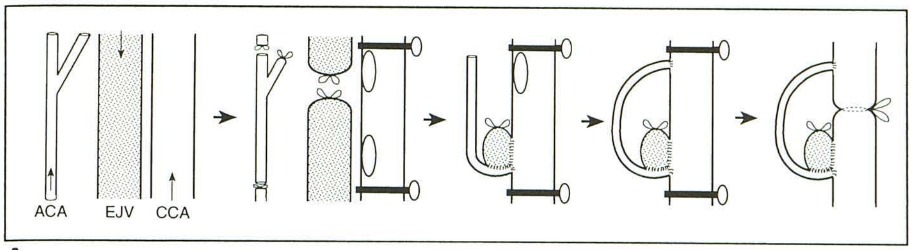

A

B

Fig. 1. Bifurcation aneurysm 1.

A, Schematic diagram outlining steps in surgical construction.

B, Common carotid arteriogram demonstrating new model. Stenosis in carotid artery is caused by external partial ligature.

mg/kg of ketamine and 2 mg/kg of xylazine. General anesthesia was maintained with mechanical ventilation and inhalation of 1% to 2% halothane after endotracheal intubation.

One side of the neck was shaved and scrubbed with povidone-iodine solution then draped in a sterile fashion. Under sterile conditions, a 10-cm incision was made in the neck parallel to the sternocleidomastoid muscle. Reflecting sternocleidomastoid muscle medially, a short segment (3 cm) of the external jugular vein, free of tributaries, was dissected and isolated. If tributaries were encountered, those smaller than 1 mm were coagulated, and those larger than 1 mm were double-ligated and divided. A 2-cm segment of the vein was isolated at both ends with a ligature and then divided to form an open-ended vein segment, to be used as the venous graft. The adventitia was carefully removed and the lumen cleaned before placing in heparinized saline. Attention was next directed to dissection of the common carotid artery and/or the more laterally situated ascending cervical artery. Self-retaining retractors were used to facilitate exposure. Segments of these arteries were isolated and cleaned of adventitia. Vasoconstriction caused by handling of the arteries was relieved by topical application of papaverine hydrochloride. The remaining steps in the surgical technique differed according to the type of aneurysm model being constructed.

Bifurcation Aneurysm Model 1

A 5-cm segment of ascending cervical artery was double-ligated at each end, transected, and removed. Side branches of this artery were ligated or coagulated. Temporary clamping of the common carotid artery allowed the fashioning of two small elliptical arteriotomies, about 3 cm apart. After cleaning with heparinized saline, one end of the ascending cervical artery segment was partially end-to-side anastomosed with the common carotid artery at the caudad arteriotomy site. The previously prepared vein graft was sutured to the V-shaped notch formed by this anastomosis. The open end of the vein graft was then tied to form a vein pouch. The free end of the ascending cervical segment was looped over the vein pouch and anastomosed to the cephalad common carotid artery arteriotomy. Clamp removal reinstated flow in the common carotid artery and the ascending cervical artery bypass loop and converted the vein pouch into a vein aneurysm sac. Initially we found that the presence of high common carotid artery flow at the site of the cephalad anastomosis resulted in poor filling and emptying of the bypass loop. Consequently, an incompletely tied ligature was placed on the common carotid artery between the two anastomoses to allow generous flow diversion into the bypass loop and aneurysm sac (Fig 1A).

Bifurcation Aneurysm Model 2

A clamp was placed on the ascending cervical artery well above the bifurcation of the thyrocervical trunk. Close to this bifurcation, the ascending cervical artery was double-ligated and transected to leave a free open-ended arterial pedicle. Next, a small elliptical arteriotomy was fashioned on the common carotid artery after temporary clamping. The free end of ascending cervical artery was partially end-to-side anastomosed to the common carotid artery at the arteriotomy. The previously prepared vein graft was sutured to the V-shaped notch formed by the anastomosis. The open end of the vein graft was tied to form a vein pouch. Clamp removal resulted in filling of the aneurysm sac and flow resumption in both the common carotid artery and the ascending cervical artery (Fig 2A).

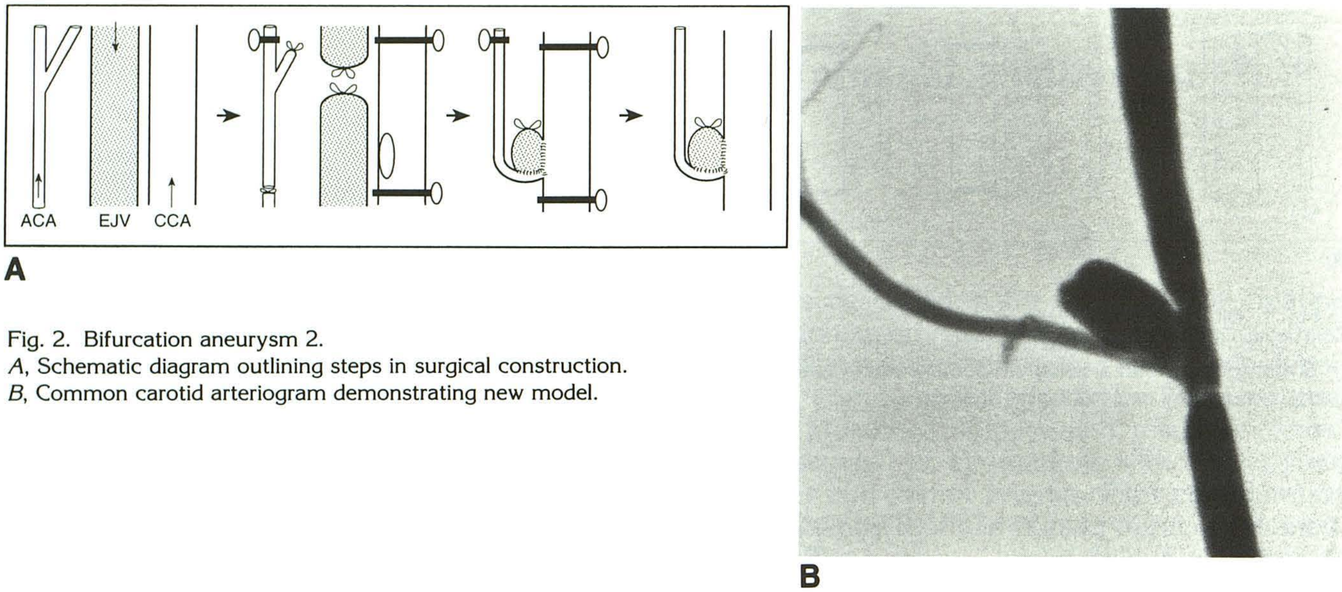

Fig. 2. Bifurcation aneurysm 2.
A, Schematic diagram outlining steps in surgical construction.
B, Common carotid arteriogram demonstrating new model.

Bifurcation Aneurysm Model 3

The ascending cervical artery was traced in a cephalad direction until its first large muscular branch was encountered, approximately 5 cm from the bifurcation of the thyrocervical trunk. Three small clamps were placed to isolate the arteries at this bifurcation. A small arteriotomy was fashioned in its fork. The previously prepared vein graft (in this case from the smaller anterior facial vein) was anastomosed to the arteriotomy, and its open end was tied to form a vein pouch. Clamp removal resulted in filling of the aneurysm sac and flow resumption in the ascending cervical artery and its branch (Fig 3A).

Terminal Aneurysm Model 1

For this model, the preliminary preparation of the external jugular vein involved isolation of a short 2-cm segment for the usual vein aneurysm pouch and a longer 5-cm segment for use as a bypass loop (as in bifurcation aneurysm model 1). A venous anastomotic bypass loop was constructed, as described above. The aneurysm sac was similarly placed in the notch of the caudad anastomosis. Both arteriotomies were placed as far apart as possible to allow the vein loop to be taut and in close contact with the vein pouch before clamp removal. Upon clamp removal, the marked expansion of the vein loop as it filled with blood caused displacement of the aneurysm sac toward the axis of the afferent common carotid artery (parent artery). This also compressed the (efferent) common carotid artery segment between the two arteriotomies away from the parent artery axis, also obviating the need for placement of an incomplete ligature as in bifurcation aneurysm model 1 (Fig 4A).

Terminal Aneurysm Model 2

A 5-cm segment of the common carotid artery was transected and removed after double ligation at its cephalad end and clamping at its caudad end. Two small elliptical arteriotomies were made opposite each other, half the distance along the length of this free segment of artery. Next, this arterial graft was placed perpendicular to the clamped common carotid artery, and an end-to-side anastomosis was fashioned to one arteriotomy. The external jugular vein aneurysm graft was also anastomosed end-to-side to the opposing arteriotomy, to form an aneurysm at a T junction. Both free ends of the common carotid artery graft were now anastomosed end-to-side to the internal jugular vein on one side and the external jugular vein on the other (at previously formed venotomies). This created a carotid-bijugular fistula, with rapid flow across the T junction (carrying its aneurysm) on clamp removal. Flow in the external jugular vein may be directed orthogradely or retrogradely, according to whether the vein graft was removed above or below the fistula site, respectively. A smaller vein graft may be obtained from the anterior facial vein, leaving an intact external jugular vein with fistulous flow in both directions (Fig 5A).

Subcutaneous tissues and skin were sutured in layers. During the procedure, the swine received 0.9 to 1.2×10^6 U of penicillin G intramuscularly. No systemic heparin was administered. Immediately after surgery, and within the next 4 days, transfemoral rapid-sequence (up to 30 frames per second) arteriography was performed under general anesthesia and the aneurysms demonstrated in multiple planes.

Results

Eleven swine were used in this preliminary study, all tolerating the surgery well, with no neurologic side effects. Successful aneurysm models were constructed in 10 of 11 animals. One bifurcation aneurysm (a model 3 type: entirely on the ascending cervical artery) throm-

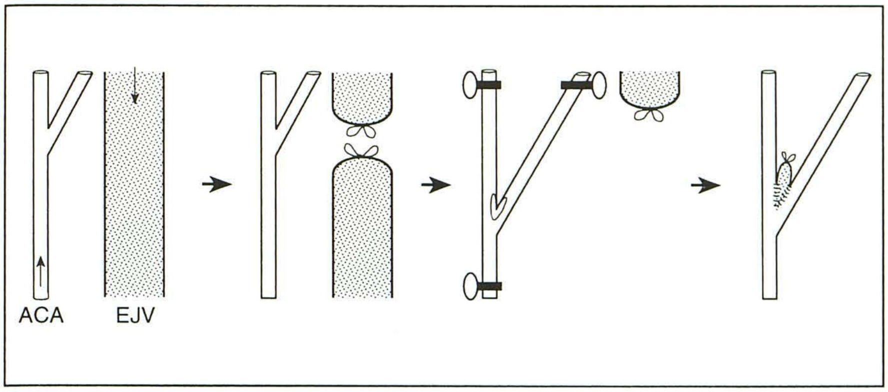

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Fig. 3. Bifurcation aneurysm 3.
A, Schematic diagram outlining steps in surgical construction.
B, Ascending cervical arteriogram demonstrating new model.

bosed immediately after surgery, because of the large size of the aneurysm sac relative to the ascending cervical artery.

Each of the successfully constructed models had morphologic features that accurately reflected those found in human intracranial aneurysms. Using an arterial bypass loop (bifurcation aneurysm model 1), we could create a bifurcation aneurysm with different-sized daughter arteries, as is found in most intracranial lesions (Fig 1B). By anastomosing the ascending cervical artery to the common carotid artery it was possible to create a bifurcation aneurysm at a junction where a daughter artery was almost at a right angle to the parent artery, simulating an internal carotid-posterior communicating artery aneurysm (Fig 2B). Placement of an aneurysm at a natural bifurcation of the ascending cervical artery (ie, with small but different-sized parent and daughter arteries) simulated distally situated aneurysms, such as on the cerebral arteries (Fig 3B). Regarding the terminal aneurysm models, the vein bypass loop (model 1) (Figs 4B–4F) and the T-shaped bifistula (model 2) (Fig 5B) methods produced daughter arteries of dissimilar and similar sizes, respectively. The former simulated aneurysms at the termination of the internal carotid artery (where the anterior cerebral artery is smaller than the middle cerebral artery), and the latter simulated basilar tip aneurysms.

Preliminary experience with flow pattern analysis of these new models was possible using rapid-sequence angiography. Clear visualization of inflow and outflow zones and intraaneurysmal vortices was possible except in the small aneurysms situated entirely on the ascending cervical artery. Early follow-up angiography revealed some size increase in several of the aneurysms, although this was not quantified. Our new aneurysms were used within 4 days of creation for training in endovascular therapeutic techniques. Consequently, no long-term follow-up investigations were conducted in this preliminary study.

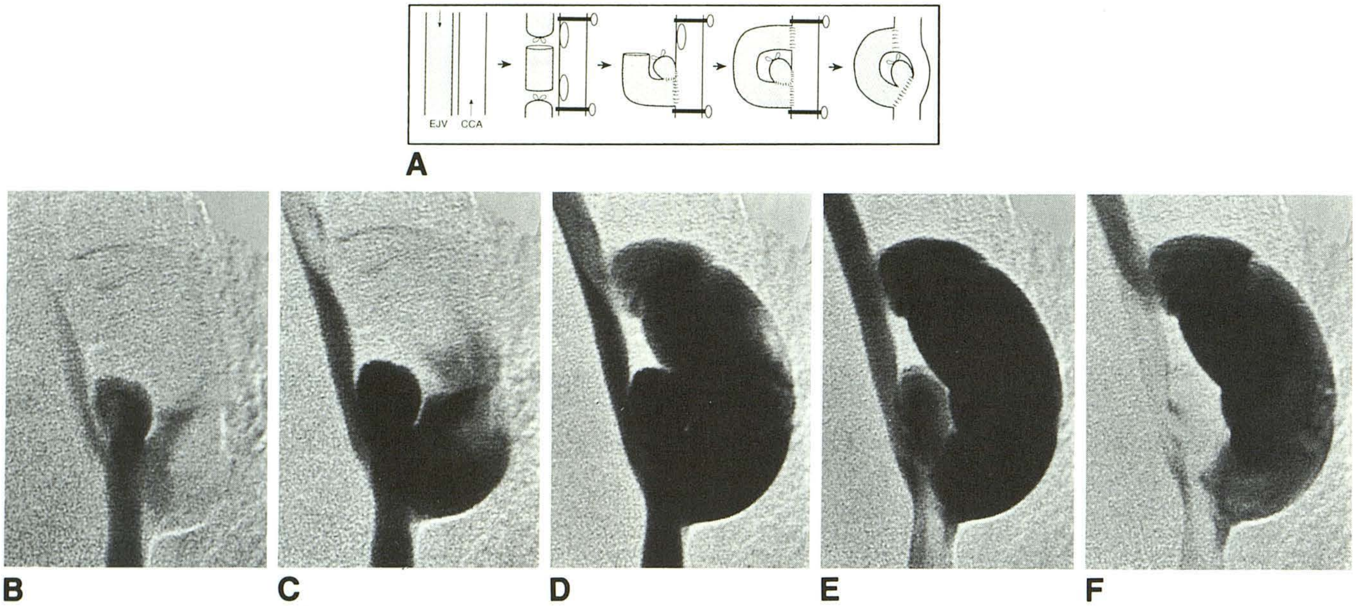


Fig. 4. Terminal aneurysm 1.
A, Schematic diagram outlining steps in surgical construction.
B–F, Selected sequential images from a common carotid arteriogram demonstrating new model.

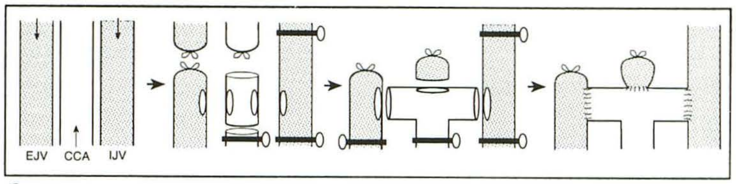
Discussion

Of all the methods used for in vivo creation of experimental saccular aneurysms, those using vascular surgery with vein pouch-to-artery grafts seem to produce the most realistic models. Although these aneurysms differ from human saccular aneurysms in their histology and reluctance to rupture (6), they seem to provide reasonable structural and dynamic replicas (7).

Models of lateral aneurysms have been constructed and used more commonly in the past despite the overwhelming preponderance of bifurcation aneurysms in humans. Until recently, bifurcation aneurysm models have been difficult to produce because of high animal morbidity and aneurysm thrombosis. Past examples of these models include the placement of a venous pouch or patch at the carotid bifurcation in rats (8), at the carotid-superior thyroid artery bifurcation in dogs (9), and at a lingual-basilar artery anastomosis in dogs (8). Currently, however, the surgical technique most commonly used for construction of experimental bifurcation aneurysms is that of Forrest and O'Reilly (3). In this technique, both sides of the animal's neck are dissected and the left common carotid artery is lifted over or under the trachea to the right side of the neck. The left common carotid artery is partially end-to-side anastomosed with the right common carotid artery, and a vein pouch is sutured to the V-shaped notch formed by the anastomosis. This results in aneurysms situated at bifurcations with similar-sized parent and daughter arteries, a less frequent finding in the human counterparts.

Previous in vitro investigations of basic flow patterns in glass aneurysm models have demonstrated that intraaneurysmal flow characteristics depend on the geometry of the bifurcation and the flow ratio between the branches (10). Therefore, in vivo models with more realistic asymmetrically sized daughter arteries, and in which the bifurcation angle can be varied at surgery, would theoretically represent an improvement on previous models in terms of accurate replication of the dynamic features in human bifurcation aneurysms. This may be important for more precise and detailed basic pathophysiologic/natural history investigations of aneurysms, the results of which also may be more faithfully extrapolated to the study of aneurysms after treatment by endosaccular coil placement.

In this article we present our preliminary experience in construction of models that require surgical dissection on one side of the neck only, and whose aneurysms simulate closely the morphologic features of intracranial lesions. All new bifurcation aneurysm models were easy to construct, requiring no more surgical skill than necessary for construction of previously reported models. In bifurcation aneurysm model 1, transection of the ascending cervical artery for use in a bypass loop should have no detrimental effect on richly supplied cervical structures. The placement of a partial ligature on the common carotid



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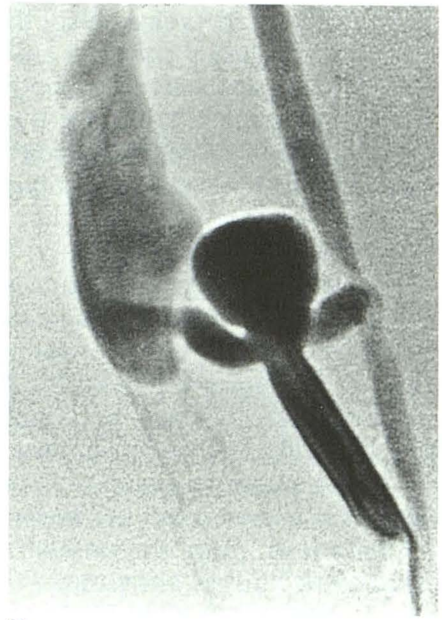

B

Fig. 5. Terminal aneurysm 2.
A, Schematic diagram outlining steps in surgical construction.
B, Common carotid arteriogram demonstrating new model.

artery was necessary to divert flow into the ascending cervical artery loop and aneurysm. The tightness of the ligature provides a way of adjusting the amount of flow entering each daughter artery of the bifurcation. This may be useful for future intraaneurysmal hemodynamic studies. In model 2, varying the length of the ascending cervical artery pedicle and the position of the arteriotomy on the common carotid artery provides a means of altering the angle of bifurcation. A perpendicular approach of the ascending cervical artery relative to the common carotid artery produces a bifurcation simulating the internal carotid-posterior communicating artery junction. Model 3 produced aneurysms that were too small (approximately 3 to 4 mm in diameter) for appreciation of flow characteristics. These aneurysms may be more appropriate for training in endovascular occlusion techniques because a superselective route is necessary for their approach.

Terminal aneurysms may be classified as straight, in which the aneurysm is in the axis of the afferent parent artery and in the same plane as the daughter arteries, or angled, in which the aneurysm remains in the axis of the parent artery but rises at an angle relative to the plane of both daughter arteries (11). Past attempts at in vivo simulation of these aneurysms are sparse. Stehbens sutured a vein pouch into the fork of the aortic bifurcation in rabbits (4). Four of 13 animals died or developed paraplegia within 24 hours. More recently, terminal aneurysms were constructed by Strother et al by end-to-end anastomosis of the right and left common carotid arteries in the dog (5). The proximal segment of the right common carotid artery was then anastomosed end-to-side into the undersurface of the U formed by this linkage, and a vein pouch was attached immediately above this anastomosis. Their technique, however, resulted in significant angulation between the aneurysm sac and its afferent parent artery. This did not occur in the two models we report. Model 1 (vein bypass loop) has the drawback of relying on expansion of the vein loop segment to deviate the aneurysm into the axis of the parent artery. This also compresses the common carotid artery between the two arteriotomies (with similar effect to the partial ligature in bifurcation aneurysm model 1) to allow adequate filling and emptying of the bypass loop. If this aneurysm deviation is inadequate, then the model assumes the morphology of a bifurcation aneurysm. On the other hand, growth of the aneurysm may result in progressive stenosis and eventual occlusion of the (common carotid) daughter artery. Furthermore, the angle of bifurcation in this model is always acute and is difficult to fashion. Model 2 (T-shaped bifistula) required more surgical anastomoses, but its appearance reflected accurately that of basilar tip aneurysms. The angle of bifurcation also can be changed to an acute symmetrical one (thus simulating acute angle splaying of both posterior cerebral arteries at the basilar tip, or the much rarer arterial fenestration anomaly associated with a proximal aneurysm [12]) by placement of more cephalad venotomies on both jugular veins. Also, the diameter of the daughter arteries may be made smaller than that of the parent artery by using a segment of the ascending cervical artery for the horizontal limb of the T-shaped anastomosis.

The swine offers several advantages for the purpose of constructing saccular aneurysm models. Animals in the 30- to 40-kg range have neck vessels that are large enough to manipulate surgically with ease. Thus, it was possible to use the ascending cervical artery in anastomoses with other vessels, obviating the need for the contralateral common carotid artery and limiting animal morbidity by unilateral neck surgery only. The similarity of the swine coagulation system to that of humans (13) allows more pertinent extrapolation of results from pathophysiologic and therapeutic studies to human intracranial aneurysms. The swine is also appropriate from an economic and ethical point of view (14).

In conclusion, we have demonstrated the feasibility of creating bifurcation and terminal saccular aneurysm models in swine. These models are easy to construct and are more similar to intracranial lesions than previously reported models. Our initial experience suggests that the arterial bypass loop method (bifurcation aneurysm model 1), the ascending cervical artery to common carotid artery anastomosis method (bifurcation aneurysm model 2), and the T-shaped bifistula method (terminal aneurysm model 2) are technically the most dependable and yield the most representative aneurysms. In particular, our bifistula terminal aneurysm model seems to be the first reliable in vivo facsimile of basilar tip aneurysms. The availability of these models bodes well for future accurate hemodynamic and natural history studies. They also serve as excellent replicas of human aneurysms for testing new endovascular occlusive therapies and for training and gaining experience in embolization techniques.

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