

Establishing a Training Model for Side-to-Side Anastomosis using Rat Femoral Vessels: Immediate and Delayed Patency

Abstract

Introduction: Side-to-side anastomoses are a relatively newer and more difficult technique used in neurovascular surgery for complex aneurysms/lesions. In view of the complex surgical technique and the infrequent clinical opportunities to acquire skills relevant to this surgery, laboratory training assumes great importance. The authors studied the feasibility of establishing a training model for performing side-to-side anastomosis using rat femoral vessels and compared the immediate and delayed patency rates in this animal model. **Materials and Methods:** After Institutional Animal Ethics Committee clearance, 16 Sprague-Dawley rats were used in this prospective study between August 2013 and March 2015. Using the standard groin incision, the femoral vessels were dissected. After applying temporary clamps, opening of approximately 3–4 mm (at least double the diameter of the wider vessel) of length was made on the facing surfaces of both vessels. 10-0 nylon was used for anastomosis. The clamps were released, and the anastomoses patency confirmed. Leg movements following the anastomoses were studied. The animals were subsequently sacrificed to assess delayed patency. **Results:** The immediate patency rate was 87.5% (14/16). The delayed patency rate (mean follow-up 209 days) was 53.8% (7/13). Three rats died during the follow-up period. The mean clamp duration and suturing time was 57.25 and 41.50 min in the first eight cases and 57.50 and 36.75 min in the last eight experiments, respectively. **Conclusion:** This animal model was found to be extremely useful for training in the difficult art of side-to-side anastomoses. The need of the hour is to establish well-planned training programs in centers with animal research facilities and use such models. This will promote younger colleagues and trainees to take up and perfect this difficult art. The present work could be used as a baseline study in this direction.

Keywords: Anastomoses, rat femoral vessels, side-to-side, training

Introduction

Microvascular anastomoses remain a critical part of the neurosurgical skill for treating complex aneurysm and very rarely atherosclerotic occlusive cerebrovascular disease. Alexis Carrel about 100 years back, described the technique of vascular anastomosis, giving birth to vascular surgery.^[1] In view of the complex surgical technique and the infrequent clinical opportunities in this era of minimally invasive and endovascular neurosurgery, laboratory training assumes greater importance to acquire skills relevant to this surgery. Several methods of microvascular anastomosis training with gauze,^[2] silicon tube,^[3] cryopreserved rat vessels,^[4] and chicken wings^[5] have been described previously. Several training models were developed for laboratory training in plastic and reconstructive surgery in the

20th century.^[6-11] Subsequently, training scenarios were adopted for the education of neurosurgeons.^[5,12] As compared to other types of anastomoses (end-to-end, end-to-side), side-to-side anastomosis requires a higher level of dexterity.^[13-15]

The purpose of this study was to train in side-to-side anastomoses using rat femoral vessels and compare the results (patency rates, clamp, and suturing duration) with end-to-side anastomoses.

Materials and Methods

The study design was prospective, and its duration was from August 2013 to March 2015. After obtaining clearance from the Institutional Ethics Committee, 16 Sprague-Dawley rats were used. All the procedures were carried out under general anesthesia (GA) by the first author under X16 magnification with an operating microscope (the first author has a 5-year

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experience of practicing microvascular anastomoses). Using standard groin incision, the femoral artery and vein were identified and skeletonized adjacent to each other [Figure 1]. Before the division of the vessel, the adventitia of the proposed division site was completely removed. Topical antispasmodic solution (few drops of 2% lidocaine) was applied to relieve vasospasm. Temporary clamps were applied. Arteriotomy and venotomy of approximately 4–5 mm (at least double the diameter of the wider vessel) were made using micro scissors. Irrigation of the lumens of these vessels with heparinized saline (100 units/ml) was done until all blood elements were visibly cleared. Colored background was used to identify the edges clearly. 10-0 nylon was used for suturing. After taking an apex suture, the back wall was sutured first [Figure 2]. Inserting the back wall sutures is the most demanding part because these sutures have to be put in an “inverted” fashion, i.e., the knot should be outside the lumen. This is followed by the second apex suture [Figure 3]. The front wall is then sutured using interrupted sutures in a regular manner. A small piece of femoral fat was put over the anastomoses. The clamps were subsequently released. Hemostasis was achieved by gentle packing using fat, gelfoam, and wet cotton balls. Vessels were checked for a change of color and pulsatility to

determine immediate patency. In a successful anastomoses, the femoral vein was found to be stretched, pulsating and red in color [Figure 4].^[14-16] The total clamp duration and time taken for suturing was recorded. The skin was subsequently sutured. The rat was allowed to go back to his regular activities. Its movements especially those of hind limbs were observed. Under GA, at a later date, the wound was reexplored and delayed patency was checked by cutting the vein proximal and distal to the anastomoses. Color and pulsatility of both vessels were also noticed. The animals were subsequently sacrificed.

End-to-side anastomoses was performed by the same author in rat femoral vessels in a separate study. It was done using the epigastric vein to femoral artery model in 23 rats and femoral vein to femoral artery model in 3 rats in the previous study.

Results

The immediate patency rate was 87.5% (14/16). Duration of vessel clamping did not defer in between the first and last eight cases. However, the suturing time came down by approximately 5 min in the last eight cases ($P = 0.130$, Mann–Whitney U-test). The rats were observed, and limb movements were studied till they were sacrificed at a later date. At a mean duration of 7 months (range 83–437 days), the animals were reexplored. Three animals died during the observation period. The hind limb movements were not

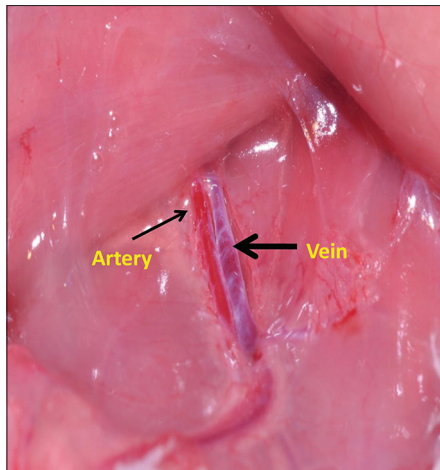


Figure 1: Femoral vessels identified and skeletonized adjacent to each other following a standard groin incision

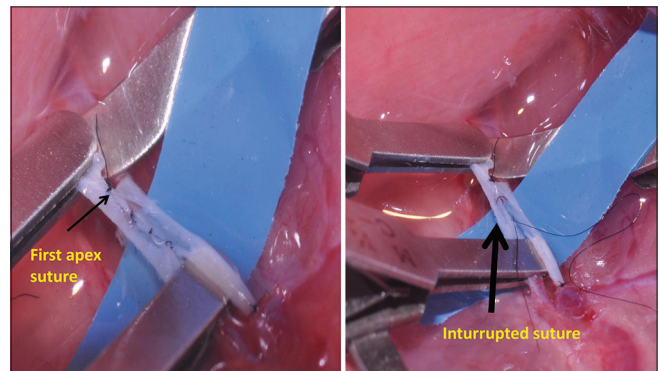


Figure 2: Apex and back wall suturing

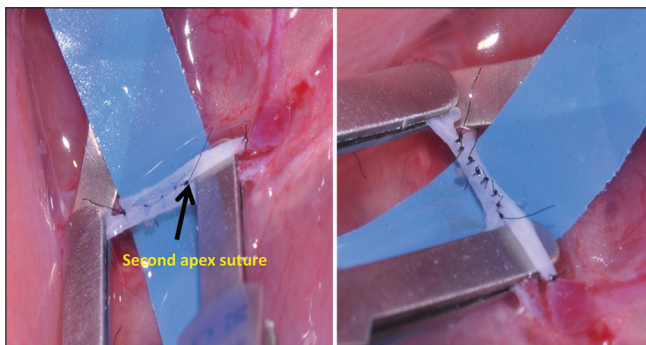


Figure 3: Second apex and front wall suturing

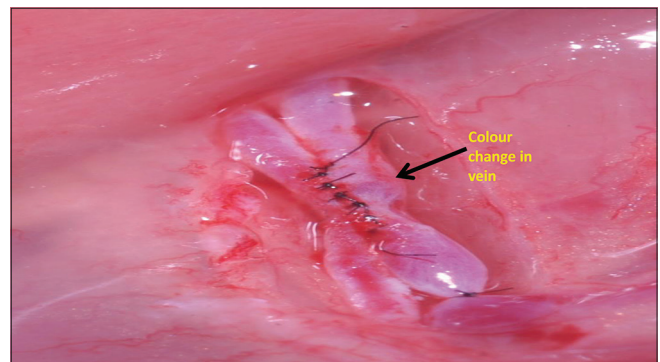


Figure 4: End result following successful anastomosis

affected by the surgery. Delayed patency rate was found to be 53.8% (7/13) [Table 1].

Table 2 shows the comparison of our results of the side-to-side anastomoses with the end-to-side anastomoses group. The table clearly shows that the clamp duration ($P < 0.001$; Mann–Whitney U-test) and suturing time ($P < 0.001$; Mann–Whitney U-test) are significantly less and patency rates (immediate patency $P = 0.042$; delayed patency $P = 0.022$ Fisher exact test) are significantly more in the side-to-side category [Table 2].

Discussion

There are very limited clinical scenarios in neurosurgery where side-to-side vascular anastomoses are indicated, as it requires both the vessels to be parallel and adjacent to each other. There are four areas of brain where this requirement is fulfilled: B/L anterior cerebral arteries over the genu and rostrum of the corpus callosum (A2 and A3 segments); the middle cerebral artery branches, including the anterior temporal artery, when they pass through the Sylvian fissure; the posterior cerebral artery and superior cerebellar artery as both pass through the ambient cistern; and the posteroinferior cerebellar arteries as they course around the posterior medulla and tonsils in the cisterna magna.^[17]

There are very few standardized rat models described in the literature for side-to-side microvascular anastomoses. Matsumura *et al.* previously described two studies of side-to-side anastomoses in a rat model between the internal and external carotid arteries and between the femoral artery and vein.^[14,15] The rat model for femoral vessels anastomoses was described on seven rats with both immediate and delayed patency rate of 100% after 7 days.^[15] In comparison, our model has significant differences. First of all, we included more number of subjects ($n = 16$) in our study and checked delayed patency after a mean duration of 7 months as compared to 7 days described by them. Important parameters such as clamp and suturing durations

were also not described in their study. Unlike continuous suturing of the back wall by Matsumura *et al.*, we used intermittent stitches with 10-0 nylon for anastomoses. Another important difference is that we sacrificed the rat at a later date and checked delayed patency by cutting the vessel whereas empty and refill test was employed by Matsumura to check the delayed patency. According to literature the best method to check delayed patency is by a direct section of the vessel proximal and distal to anastomoses site.^[16] Despite these differences, these are the only studies which provide objective data in the form of immediate and delayed patency rates. Such data can be extremely useful in establishing the pass criteria for a training program to assess skill development of neurosurgery trainees.

Although traditionally side-to-side anastomoses are described as more complex compared to other types (end-to-end, end-to-side);^[17] this has not been proven till now in any experimental or clinical study, to the best of our knowledge. Our results showed a significant decrease in clamp duration and suturing time with better patency rates in the side-to-side as compared to end-to-side category [Table 2]. These findings are in stark contrast with the available literature.^[17] This should prompt further studies in this direction.

Training in different models helps one to master the skill to perform anastomoses. Sigounas *et al.*^[18] (Department of Vascular Surgery) demonstrated significant improvement in the surgical technique after practicing simulation for vascular anastomosis technique. Nossek and Ram^[19] emphasized the need for improving microsurgical skills by augmented training through a surgical skills laboratory to address concerns of decreased operating room exposure. Liu^[20] and Kanazawa and Teramoto.^[21] too described different training models for microvascular anastomosis of submillimeter vessel in rats.

However, there are very few training models described for microvascular anastomoses in neurosurgery. The authors

Table 1: Clamp and suturing times, immediate patency rates (side-to-side $n=16$)

	Clamp time	Suturing time	Patency (immediate)	Patency (Delayed)
First eight	57.3±6.7 min	41.5±5.5 min	87.5%	37.5% ($n=8$)
Last eight	57.5±8.3 min	36.8±4.7 min	87.5%	60% ($n=5$)
	$P=0.959$ Mann Whitney U test	$P=0.130$ Mann Whitney U test	$P=1$ Fisher Exact test	$P=0.592$ Fisher Exact Test

Table 2: Comparison of side to side with end-to-side group

	Side to side ($n=16$)	End to side ($n=26$)	P
Clamping time (Mean)	57.4±7.3 minutes	77.8±16.7 minutes	$P<0.001$ Mann Whitney U test
Suturing time (Mean)	39.1±5.5 minutes	57.6±14.8 minutes	$P<0.001$ Mann Whitney U test
Immediate patency (%)	87.5%	53.8%	$P=0.042$ Fisher Exact Test
Delayed patency (%)	53.8%	15.4%	$P=0.022$ Fisher Exact Test

of this article believe that objective quantification methods are the need of the day to accelerate the acquisition of skill necessary for a difficult procedure like microvascular anastomosis. Although cost and ethical issues along with the need of facilities to house rats continue to be the limitations; using rats as a training model has its own advantages. The rat femoral vessels diameters are from 0.6 to 1 mm to the artery and 1–2 mm to the vein.^[22] They mimic human cerebral vessels in terms of texture and haptic qualities and pulsation and coagulability in physiological conditions.^[23] Moreover due to consistent size and anatomy of the femoral vessels; dissection is easier and faster. These vessels are also useful to learn adventitial stripping and gentle tissue handling which are hardly reproduced in nonanimal models. Another advantage is the possibility to evaluate flow after the completion of the anastomosis.

We feel that the femoral vessels of Sprague-Dawley rat are a valid training model. We found that after the clamps are released following the completion of the procedure, vessels are seen to expand; hence, it is easy to assess the immediate patency rate. Furthermore, thin and inelastic venous wall in rats makes precise passing of needle difficult allowing better skills to acquire. However, the scenario in a real operation is, of course, different in view of increased depth of working field, narrow side spaces, and susceptible brain to clamping.

There are certain drawbacks of this model. First of all, it is an artery to vein model in contrast to the traditional artery to artery model. However, veins are more difficult to handle. Furthermore, there is no depth training.

Yasargil in his illustrious career has often stressed on the absolute necessity of laboratory training in microtechniques before their application to humans in the operating room.^[24] This holds true more so in this era of minimally invasive neurosurgery where chances to refine and practice surgical skills are few and far between.

Conclusion

Our model of using rat femoral vessels for training of side-to-side vascular anastomoses was found to be extremely useful in providing adequate exposure in a simulated environment to neurosurgery trainees to acquire the skills for this complex procedure. We demonstrated the relative patency rates in both types suggesting that side-to-side anastomoses may not be as difficult as is suggested. The need of the hour is to establish well planned training programs in centers with animal research facilities and use such models. This will promote younger colleagues and trainees to take up and perfect this difficult art. The present work could be used as a baseline study in this direction.

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Nil.

Conflicts of interest

There are no conflicts of interest.

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