Surgical construction of a novel simulated carotid siphon in dogs

Laboratory investigation

HUÁ-QIÁO TÁN, M.D., PH.D., MÍNG-HUÁ LI, M.D., PH.D., YUE-QÍ ZHÚ, M.D., PH.D., CHÚN FÁNG, M.D., CHÚN-GÉNG WÚ, M.D., PH.D., YÍNG-SHÉNG CHÉNG, M.D., PH.D., JUE WÁNG, M.D., JIÁN XÍÉ, M.D., PH.D., AND HÉ ZHANG, M.D., PH.D.

Department of Radiology, Shanghai Sixth People’s Hospital, Shanghai Jiao Tong University, Shanghai, China

Object. The development and preclinical assessment of new endovascular devices necessitate readily available and reproducible animal models. The purpose of this study was to develop an in vivo carotid siphon model for testing the properties of covered stents specially designed for the intracranial vasculature.

Methods. Six carotid siphon–shaped devices were created. Six dogs underwent surgery to expose and isolate both common carotid arteries (CCAs). The right CCA origin was ligated and incised distal to the ligation point after temporary constriction of the distal right CCA. The distal left CCA was ligated and incised proximal to the ligation point after the left CCA origin was temporarily clamped. The proximal isolated left CCA was passed through the shaped device and then Anastomosed end-to-end to the distal isolated right CCA. Finally, the shaped device was fixed and embedded in the neck. Intraarterial digital subtraction angiography was performed at 7 days, 2 weeks, and 1 month postprocedure. All models underwent endovascular interventional simulation. The carotid siphon models were evaluated.

Results. The animals tolerated the surgical procedure well. The mean time for surgical construction of the model was 90 minutes. The morphology and endovascular manipulation of the siphon models were similar to those in humans. Stenosis of anastomotic stoma occurred in 2 models, and mural thrombosis of anastomotic stoma occurred in 1 model; however, all models were patent at postprocedural follow-up angiography.


Key Words • animal model • cerebrovascular disease • dog • endovascular therapy • intracranial vascular navigation

Endovascular covered stents have been sporadically used for the treatment of neurovascular diseases including carotid cavernous fistulas, aneurysms, and iatrogenic or traumatic pseudoaneurysms in the intracranial portion of the ICA.¹²,¹¹,¹⁶ This seems to suggest that endovascular treatment with covered stents represents a promising alternative to the established neurosurgical or endovascular options. However, owing to the lack of covered stents specially designed for intracranial use, the available covered stents have been typically coronary stent grafts, which lacked longitudinal flexibility and were difficult to navigate through the tortuous carotid siphon.¹¹,¹⁶ Thus, the use of covered stents for the treatment of neurovascular diseases was limited to selected patients. Recently, some covered stents specially designed for intracranial use have been reported.¹² However, there are currently no suitable animal models resembling the human carotid siphon to test the flexibility of such a covered stent; therefore, the biological response to the covered stent implantation remains unknown. The purpose of the present study was to develop in vivo carotid siphon model to test the properties of covered stents specially designed for intracranial vasculature.

Methods

Creation of the Carotid Siphon–Shaped Device

Six carotid siphon–shaped devices were created using stereolithographic biomodeling and the lost–wax technique.¹⁹ Briefly, 3D angiographic data sets were obtained
from 6 individuals with the aid of a single C-arm angiography system (Axiom Artis dTA, Siemens Medical Solution) with a rotational angle of 200°. Data were obtained every 1.5° at 26.6 frames per second. The imaging data were then transferred to the postprocessing workstation (Leonardo, Siemens Medical Solution). After defining the volume of interest, the slices were calculated as follows: matrix 512 × 512, slice thickness 0.13 mm, and voxel size ranging from 0.068 to 0.17 mm³.

The data were transmitted to the National Die & Mold CAD Engineering Research Center at the Shanghai Jiao Tong University, via an optical disc, where these files were converted to STL (StereoLithography Format) by using appropriate software (Mimics version 10.01, Materialise). The STL file was processed to remove the vascular structure with the exception of the carotid siphon and then transferred to a 3D printer via compact disk. The 3D printer (Modemaker II, Solidscape) formed wax copies of the carotid siphon. The resolution of the build layer was 0.076 mm.

Subsequently, these wax copies were manually coated with 4–6 layers of the previously prepared prosthetic silicone elastomer (silastic Biomedical grade ETR Q7–4780, Dow Corning Co.), and the wax was removed thermally and chemically, leaving a replica of the carotid siphon (Fig. 1). These carotid siphon replicas were used for the carotid siphon–shaped devices. The lumen diameter of the shaped devices ranged from 4.8 to 5.0 mm, and the wall thickness ranged from 1.0 to 1.2 mm. The configuration of the shaped devices remained unchanged with aid of a 5-mm-diameter and 20-mm-long dilation catheter positioned in the shaped devices at the midportion of the angular point of the curve and inflated at 6 atm (Fig. 2). Before surgical construction of the in vivo carotid siphon model, the shaped devices were measured using high-resolution radiography before and after balloon inflation at 6 atm to detect material inhomogeneities.

**Surgical Construction of the Model**

The model was surgically created in 8-month-old dogs of both sexes weighing 20–25 kg. The protocol was approved by the animal research committee of our institution and was conducted in accordance with the guidelines of the International Council on Animal Care. All animals were maintained on a standard laboratory diet, and their physical conditions were monitored daily. After an overnight fast, anesthesia was induced with thiopental 20 mg/kg followed by endotracheal intubation and maintained with 1.5–2.0% isoflurane before the surgical procedure.

Under sterile conditions, a 10-cm midline incision of the anteroinferior neck was performed. Both CCAs were exposed and isolated, and each was ≈ 8–10 cm in length. The right CCA was ligated and incised distal to the ligation...
point after the distal right CCA was temporarily closed with an atraumatic hemostatic clamp. The distal isolated lumen of the CCA was flushed with heparinized saline by using a syringe with a blunt tip. The distal left CCA was ligated and incised proximal to the ligation point after the proximal left CCA origin was temporarily clamped. The proximally isolated lumen of the CCA was also flushed with heparinized saline by using a syringe with a blunt tip. The edge of the incision was trimmed regularly. Then, the proximal isolated left CCA was passed through the carotid siphon–shaped device. The distal segment of the divided right CCA was swung to the left and was anastomosed end-to-end to the proximal isolated left CCA using continuous 6-0 Prolene sutures in front of the trachea (Fig. 3). The bilateral clamps were loosened, and the extended CCAs were allowed to refill. Finally, the carotid siphon–shaped device was fixed with a suture and embedded in the soft tissue of the left neck. A layered wound closure was performed.

Assessment of the Carotid Siphon Model

After induction of general anesthesia, selective carotid angiography and 3D rotation angiography were performed at 7 days, 2 weeks, and 1 month postprocedure in all dogs by using the same imaging parameters as for the individuals. According to 3D angiography (frontal and lateral projections of the carotid siphon), the morphology of the carotid siphon models was visually evaluated and compared with that of the original individual by 2 independent neuroradiologists (M.H.L. and C.F.). The patency of the carotid siphon models and the stenosis of the anastomotic stoma were evaluated.

To evaluate whether the model provided a practical environment encountered in clinical practice, during 1-month follow-up angiography all dogs underwent endovascular interventional simulation through the femoral artery approach after induction of general anesthesia. Under fluoroscopic guidance with the aid of road mapping, the balloon-expandable covered stent (Willis, MicroPort Medical [Shanghai] Co.) with a diameter of 4.5 mm and length of 13 mm was navigated and deployed at the midpoint of the angular point of the curve in the models. The subjective realism of the procedure was evaluated compared with that of clinical manipulation, and the changes of the morphology and position of the model were observed. While the dogs were still in a state of general anesthesia, they were killed by a barbiturate overdose.
Results

All dogs tolerated the general anesthesia and surgical construction. The average time for surgical construction of the model was 90 minutes. No dog died during surgery. After the initial procedure, no neurological deficits secondary to the procedure were noted in any dog. Evaluation of the models by 2 neurointerventionists revealed good acceptance of the morphological characteristics that simulated the original carotid siphon geometry. Representative images are shown in Fig. 4A and B. Stenosis of the anastomotic stoma occurred in 2 siphon models, and thrombosis of the anastomotic stoma was found in 1 siphon model. However, all models remained patent on postprocedural angiography at 7 days, 2 weeks, and 1 month. The carotid siphon geometry of the models was not found to have significant change over time due to movement of the animal's neck. At the time of testing, the model geometry remained unchanged and the model position remained at the level of immobility encountered in the human carotid siphon (Fig. 4C and D). Endovascular manipulation within the siphon model was subjectively regarded as realistic, and the resistance to navigation of the covered stent within siphon model was similar to that encountered in clinical practice.

Discussion

In vitro, cadaveric, and in vivo models have been used to evaluate endovascular devices. In vitro models are easy to handle and are highly reproducible. They permit testing at reduced cost and without ethical considerations. However, in vitro models lack the vessel viscoelastic properties and biological responses. Their use is confined primarily to structural, mechanical, and hemodynamic investigation. Cadaveric models are also suitable for testing of endovascular device, but they, too, lack the physiological responses, such as vasospasm, thrombus, and embolic complications. In vivo models are invaluable for evaluating new materials and devices. Elements unique to in vivo models include pulsatile blood flow and vascular responses. Thus, in vivo models not only assess the physical properties of endovascular devices, but they can also be used to test the biological responses to endovascular devices. The experimental primate siphon anatomy resembles that of humans, but such models are difficult to justify from an ethical and economic standpoint. Therefore, in the present study, we have established an appropriate experimental model of the carotid siphon.

For our study, the dog was selected to create a siphon model for the following reasons: 1) Adult dogs have a long CCA (10–12 cm). Therefore, the extended CCA can provide sufficient length to construct a carotid siphon model. 2) The caliber of the canine CCA is comparable to that of a human ICA, with a constant diameter of 4–5 mm from the proximal end to the distal end. Thus, the caliber of canine vessels permits the replication of conditions similar to those encountered during intracranial stent implantation in humans. 3) The blood supply of the central nervous system seems to be obtained almost entirely through the vertebrobasilar system. Moreover, there exist anastomoses between each network of the ICA, the external carotid artery, and the vertebral arteries, and these anastomoses are generally permanently functional in dogs. Therefore, this anatomical characteristic helps prevent death when the bilateral CCAs are temporarily occluded and may permit long-term follow-up to observe the biological responses following implantation of endovascular devices. 4) Canine vessels behave more like human vessels than do those of pigs with respect to vasospasm, recoil, neointimal proliferation, and thrombotic potential. Furthermore, the number of cells, their composition, and the accompanying proteoglycan matrices are essentially the same as in humans. Consequently, these attributes indicate that the canine model may accurately predict the human biological response to endovascular device implantation.

An in vivo carotid siphon model has been developed in pigs and has been used to test neurovascular devices. This model has been demonstrated to closely mimic the human carotid siphon and is believed to be more physiologically relevant than previous models, such as in vitro and cadaveric models. However, several limitations have been noted. First, the dimensions of the arteries are slightly larger than those in humans, and the S-shaped curves are not as pronounced as they are in many patients. Second, the artificial siphon is not encased in an osseous canal,
Surgical construction of a carotid siphon model which could add an additional element of difficulty to the implantation of the endovascular device. Third, this model does not reconstruct the level of immobility encountered in the human carotid siphon. Fourth, the geometric configuration of the carotid siphon was reproduced by fixing the extended CCA with a suture, so it provided inferior reproducibility. In addition, the structure and histological composition of the vascular wall in swine differ from those in the human carotid siphon, and the porcine vessels have a greater propensity to spasm.

When compared with the existing in vivo carotid siphon model, our model offers several advantages. The shaped devices were established using stereolithographic modeling and the lost-wax technique, based on data sets from rotational cerebral angiography in actual individuals. This modeling technique allows one to create highly complex tubular in vitro models of the cerebral vasculature with high accuracy. Thus, the shaped devices were anatomically replicated from the human carotid siphon, which allows the spatial course of the encased CCA to conform to the characteristics of the human carotid siphon. In addition, this technique also allowed for the production of an unlimited number of shaped devices reproducing the carotid siphon with great accuracy. When these shaped devices were used to encase the extended CCA, the superior reproducibility of the in vivo models could be provided. Moreover, our model simulated the surrounding structure of the human carotid siphon because the firmness of the shaped devices approximates the ICA canal at the base of skull, which has been demonstrated on the in vitro test for material properties and the following-up angiography after implanted in the animal neck. Additionally, the shaped devices are embedded and fixed in the soft tissues of the neck, which reproduce the level of immobility encountered in the human carotid siphon.

Our model is also ideal for practical reasons. The model creation procedure is quite simple and easy to perform. It does not involve any microanastomosis, so the procedure can be performed quite easily without extensive surgical experience. Investigators can adopt the model in a short period of time and reliably achieve reproducible results. The total procedure time is ~90 minutes. After the procedure, there is no vascular occlusion resulting from acute thrombosis of the anastomotic stoma. Although, in our study 1 dog was found to have mural thrombosis of anastomotic stoma on angiography at 1 week, this case remained patent on follow-up angiography.

There are several limitations of our study. First, the CCA was inevitably stretched to some extent when it was passed through the shaped devices due to a 3D tubular structure and higher frictional coefficient of the interior surface, which may induce vasospasm. Second, although the silicone elastomer produced the shaped device has a good biocompatibility, this material is still a foreign body to the dog, which could stimulate adventitial hyperplasia and induce infrequent and moderate granulomatous reaction, leading to stenosis of the siphon portion. Third, unlike the intracranial ICA which lacks a thick adventitia and is composed of a higher percentage of smooth-muscle cells, the canine CCA with adventitia has a lower percentage of smooth-muscle cells, which may result in an underestimation of the biological response to the endovascular device. In addition, our model did not reproduce the branches of the human carotid siphon. Nevertheless, the intent of our model is not to investigate the influence of device implantation on the function of side branches or perforating branches of the carotid siphon.

In the present study, the developed carotid siphon model accurately simulated the spatial structure of the carotid siphon in humans. Therefore, this model may be used to test the physical properties of stents, such as flexibility and apposition to the carotid siphon, and also to study the biological responses to endovascular device implantation. In addition, based on these findings, a carotid siphon model of vascular diseases could be established which may facilitate the study of the genesis of neurovascular diseases, and may assist in the development of novel endovascular devices.

Conclusions

We have demonstrated the feasibility of the surgical construction of a carotid siphon model in dogs by elongating the CCA and with the aid of a shaped device. This model simulates the spatial structure of the carotid siphon in humans, which is highly reproducible and reliable, and can be used for testing neurovascular devices and developing disease models with a carotid siphon.

Disclosure

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Address correspondence to: Ming-Hua Li, M.D., Ph.D., No. 600, Yi Shan Road, Shanghai, China. email: Shliminghua@yahoo.com.cn.