

Multi-scale Transparent Arteriole and Capillary Vessel Models for Circulation Type Blood Vessel Simulator

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Abstract – We proposed a fabrication method for multiscale transparent arteriole and capillary vessel models and demonstrated the fabrication of microchannels with circular cross sections $\phi 10 - 500 \mu\text{m}$. First, we demonstrated fabrication of $\phi 10 - 500 \mu\text{m}$ arteriole and capillary vessel block models using photolithography. The circularity of fabricated $\phi 10, 50,$ and $500 \mu\text{m}$ microchannels were 84.0%, 61.5%, and 82.3%, respectively. Flow experiments demonstrated that these channels had no leakage. Next, we proposed a fabrication method for $\phi 100 - 500 \mu\text{m}$ arteriole membrane models, which connect larger membrane models and smaller block models. These models were prepared with grayscale lithography and a wax and PVA (polyvinyl alcohol) mixture material. The proposed approach overcame the brittleness of a previous sacrificial model fabricated by ink jet rapid prototyping. The membrane model had a circular cross section with a channel circularity of 90%. Finally, we succeeded in making transparent membranous and block arteriole model with which we can simulate blood circulation.

I. Introduction

Surgical simulators are used in practice and rehearsal for intravascular neurosurgery, and for development of new medical instruments, such as catheters. We developed tailor made three-dimensional (3D) elastic membranous blood vessel models with 3D wax models fabricated by ink jet rapid prototyping [1, 2]. Then, we developed a transparent surgical simulator by connecting the elastic membranous models. Blood vessels in a conventional surgical simulator are larger $500 \mu\text{m}$ in diameter. Smaller blood vessel models are needed to simulate a more realistic vessel environment, as shown in Fig. 1; however, these are very difficult to fabricate using ink jet rapid prototyping because of the brittleness of wax. Diseases exist in blood vessels that are narrower than $\phi 500 \mu\text{m}$, such as arteria basilaris; however, previous

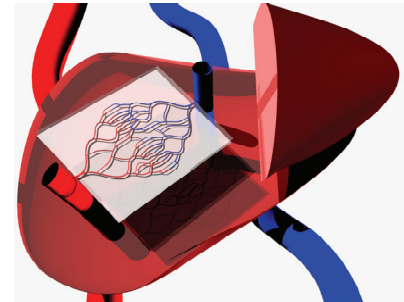
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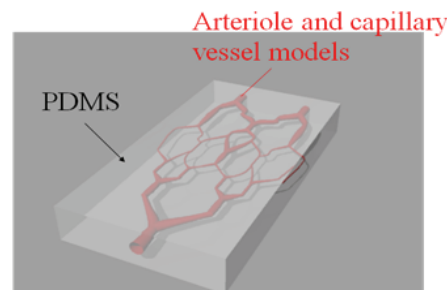
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(a) Concept of the circulation type blood vessel simulator



(b) Example of arteriole and capillary vessel model

Fig. 1. Blood vessel simulator with arteriole and capillary vessel model.

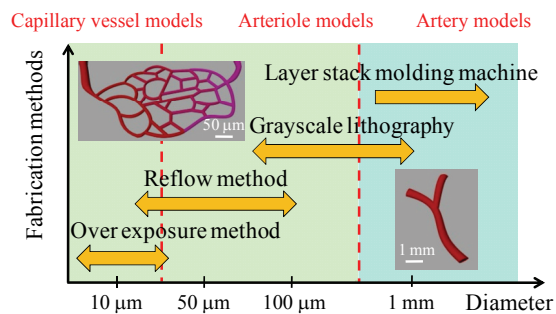


Fig. 2. Concept of fabrication methods for multiscale blood vessel models.

surgical simulators are not suitable for rehearsal and training for such diseases. This paper aims to fabricate multiscale transparent arteriole and capillary vessel models enabling easy simulation of blood circulation. For this purpose, we must fabricate microchannels with circular cross sections $\phi 10 - 500 \mu\text{m}$.

Many methods have been proposed for fabricating microchannels, such as machining, stereolithography, ink jet rapid prototyping, and photolithography. Machining is suitable for a straight channel up to around $10 \mu\text{m}$ in diameter, but not for complicated capillary vessel networks. Stereolithography is applicable for fabricating a mold; however, it is difficult to dissolve it to create a

hollow structure.

Ink jet rapid prototyping is beneficial for thicker channels, such as aortas, but not applicable for capillary vessel models. Photolithography is a fundamental technology for fabricating microchannels, and a high resolution around $1\ \mu\text{m}$ is easily attained. We have chosen photolithography for fabricating arteriole and capillary vessel models [3]. In general, it is quite complex to fabricate microchannels with a circular cross section. Microchannels were fabricated using semi-round photoresist patterns and light curable resin, but the cross sections of the fabricated channels were semicircular [4, 5]. These processes are not suitable for fabricating fine blood vessel models. Therefore, we proposed a new fabrication process for multiscale transparent arteriole and capillary vessel models.

II. Block models

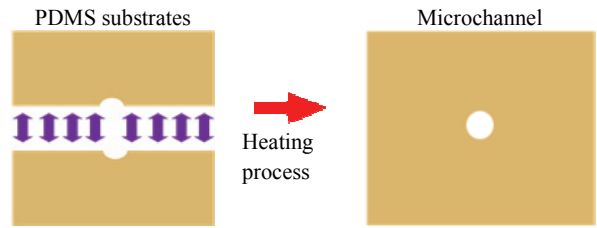
We proposed multiscale fabrication methods of blood vessel models (Fig. 2). The processes used were an over exposure, reflow, grayscale lithography, and ink jet rapid prototyping using a layer stack molding machine. Of these processes, photolithography processes, such as over exposure, reflow, and grayscale lithography are used for making fine structures. Because of the limits of fabrication accuracy, it is necessary to choose an appropriate method to fabricate a model with a targeted diameter. After exposure, fabricated photoresist patterns are transcribed onto poly(dimethylsiloxane) (PDMS). Arteriole and capillary vessel models with a circular cross section are completed by bonding two patterned PDMS substrates using plasma treatment and heating (Fig. 3). Here, we fabricated arteriole and capillary vessel block models with over exposure, reflow, and grayscale lithography [6–8].

Capillary vessel models of $10\ \mu\text{m}$ in diameter are fabricated by over exposure method, arteriole models of $20 - 100\ \mu\text{m}$ in diameter are fabricated by reflow method, and arteriole models of $100 - 500\ \mu\text{m}$ in diameter are fabricated by grayscale lithography.

We already succeeded in fabricating a capillary vessel block model. The diameter of the branched capillary vessel block model was about $10\ \mu\text{m}$ and its cross section was nearly circular. However, the channel cross section was slightly out of alignment (Fig. 4). To solve this problem, we fabricate both capillary vessel model patterns and alignment patterns for assembly using multi stage exposure photolithography, as shown in Fig. 5. The fabrication process flow is as follows:

A. Capillary vessel models with $\phi 10\ \mu\text{m}$ microchannels

1. Coating a substrate with $8\ \mu\text{m}$ thickness of photoresist PMER.
2. Exposing at hard contact points using both a mask and a resist sample until $1000\ \text{mJ}/\text{cm}^2$.
3. Developing for 7 min.
4. Hard baking at 145°C for 30 min.



Plasma treatment

Fig. 3. The concept of making a capillary vessel model. After plasma treatment, the two patterned PDMS substrates are aligned and bonded by heating and the capillary vessel model is completed.

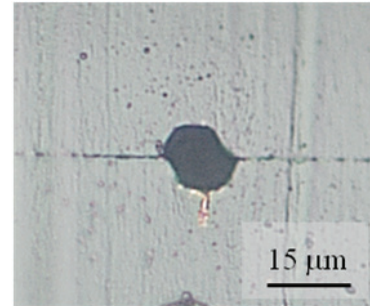


Fig. 4. Cross section of the capillary vessel model channel (before improvement).

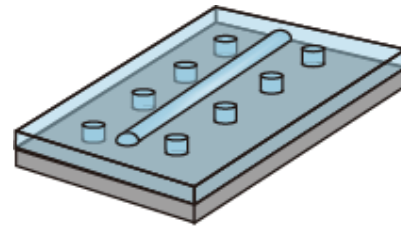


Fig. 5. Designed convex pattern for channel and patterns for alignment.

5. After plasma hydrophilic treatment, coating the substrate with $8\ \mu\text{m}$ thickness of photo resist PMER.
6. Exposing at hard contact points using both the mask and the resist sample until $1000\ \text{mJ}/\text{cm}^2$.
7. Developing for 7 min.
8. Hard baking at 145°C for 30 min.
9. After plasma hydrophilic treatment, coating the substrate with $8\ \mu\text{m}$ thickness of photoresist PMER.
10. Exposing at soft contact points using both the mask and the resist sample until $1500\ \text{mJ}/\text{cm}^2$.
11. Developing for 5 min.
12. Transcribing photoresist pattern on PDMS.

B. Reflow method

1. Coating posi-photoresist PMER on silicon.
2. Exposing under the best exposure conditions.
3. Developing at the best time assessed.
4. Heating the resist patterns from the bottom for 30 sec with a hot plate.
5. Transcribing these resist patterns onto PDMS.

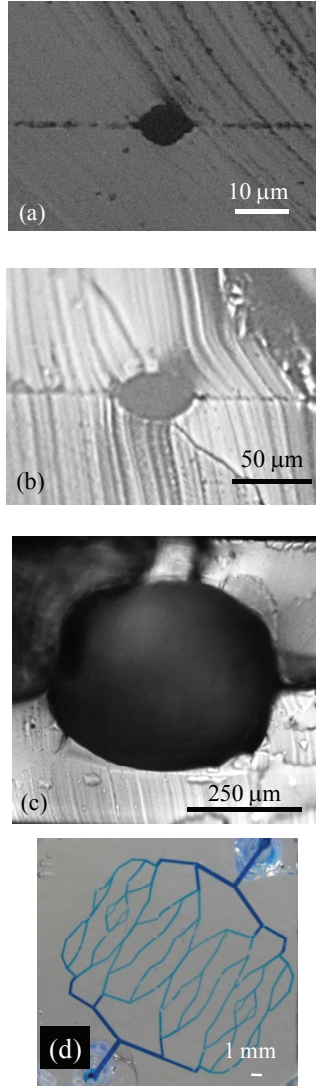


Fig. 6. Cross sections of fabricated model. (a) $\phi 10 \mu\text{m}$ microchannel; (b) $\phi 50 \mu\text{m}$ microchannel; (c) $\phi 500 \mu\text{m}$ microchannel; and (d) arteriole model.

C. Grayscale lithography

1. Coating nega-photoresist SU-8 on a glass substrate.
2. Exposing from bottom under the best exposure conditions.
3. Developing at the best time.
4. Transcribing the resist patterns onto PDMS.

After exposure and transcribing, each patterned surface of two patterned PDMS substrates are treated by plasma and heated at 120°C for 20 min. This creates arteriole and capillary vessel models with circular cross sections.

Figure 6 shows the cross sections of fabricated arteriole and capillary vessel models. We calculated the circularities of $\phi 10$, 50, and $500 \mu\text{m}$ microchannels by dividing the shortest axis by the longest axis and obtained 84.0%, 61.5%, and 82.3%, respectively. In addition, we

evaluated the alignment accuracy of the capillary vessel models. A model having no alignment patterns has an alignment error of $2.5 \mu\text{m}$, whereas, a model with alignment patterns has an alignment error of $0.6 \mu\text{m}$. The circularity of the capillary vessel model having no alignment patterns is 70% and for that with alignment patterns is 84%. Thus, alignment patterns reduced alignment error from $2.5 \mu\text{m}$ to $0.6 \mu\text{m}$ and improved circularity from 70% to 84%.

We performed experiment with flowing methylene blue solution in the channel of the arteriole model (**Fig. 6** (d)) to check for leakage; the channel had none.

III. Membrane models

Block models, however, cannot recreate the moderate compliance similar to that of real blood vessels. We believe that using arteriole and capillary vessel membrane models as surgical simulators can resolve this problem. Sacrificial models are needed to improve membrane reproduction. However, because of the brittleness of wax, we were unable to fabricate sacrificial models by the previous fabrication method of using a layer stack molding machine.

Here, we propose a novel method for fabricating transparent arteriole membranous models. We attempted to decrease the brittleness of wax by mixing it with PVA (polyvinyl alcohol). After fabricating sacrificial models using this mixture, arteriole membranous models were fabricated by dip coating. Creating membranous models smaller than $\phi 500 \mu\text{m}$ for surgical simulation (1) enables modeling of blood vessel diseases, such as arteria basilaris, that affects vessels with a diameter of $100 \mu\text{m}$, and (2) allows fabrication of high precision surgical simulators. This paper also reports on fabrication method, the fabricated arteriole membranous model, and the results of evaluation of the molding material (wax and PVA mixture).

A. Evaluation of wax + PVA mixture materials

For our fabrication process, a wax and PVA mixture was used to build the sacrificial models. These models were useful for fabricating membrane models with diameters less than $500 \mu\text{m}$. When using only wax for making the sacrificial models, a specific temperature had to be maintained to control the process. Furthermore, the fragility of wax proved unsuitable for our fabrication procedure. While, using PVA alone for fabricating sacrificial model, the model quickly deformed under its own weight when dip coated, because PVA has low bend strength. Therefore, we propose a mixture of wax and PVA for fabricating sacrificial models. We predict that the mixture will exhibit the properties of both wax and PVA. We evaluated the mixture properties, its melting time in different liquids (solubility) and the Young's modulus of each mixture using different mix ratios (wax:PVA = 1:0, 1:4, 2:3, 3:2, 4:1, 0:1).

Table 1. Experimental rigidity and extension

PVA:WAX ratio [%]	Young's modulus [MPa]	Extension [%]
1:0	9.57	75
4:1	9.69	75
3:2	8.78	75
2:3	15.7	44
1:4	22.8	4.0
0:1	20.6	6.0

a. Evaluation of mechanical properties (Young's modulus)

For evaluation experiments, a tensile tester was used to measure Young's modulus. Samples fabricated using the wax and PVA mixture were 50 mm × 10 mm × 2 mm (long × wide × thick) in size. We plotted the measurement results as a load–displacement curve. The tensile experiment in triplicate for each wax: PVA mix ratio was performed and Young's modulus was calculated using the curve data. Young's modulus for each mix ratio was then calculated by averaging the experimental results.

Table 1 shows Young's modulus for each mix ratio. Young's modulus using PVA alone was 9.57 MPa, whereas that using only wax was 20.6 MPa. We observed that it increased with the amount of wax. Extension is an expression of a material's brittleness and ductility. We observed an increased extension with greater amounts of PVA in the mix. Therefore, we were able to control the mechanical properties of the wax and PVA mixture by changing the mix ratio.

b. Evaluation of chemical properties (solubility)

The melting time of each mix ratio in different liquids was determined by measuring the mass changes. Experiments were performed to determine the solubility of each mix ratio, using an ultrasound bath set at 50°C and with DI water, acetone, and a mixture of DI water and acetone as solvents. DI water easily melted PVA and acetone easily melted wax. A 1:1 acetone to DI water mix to melt the wax and PVA combinations was decided to be used. **Figure 7** shows the experimental results examining the solubility of each sample in DI water, acetone, and the 1:1 DI water and acetone liquid. To evaluate our results, we examined the mass changes for wax in the samples, which occur due to heating and etching effects of the ultrasound bath. Because of the physical characteristics of DI water, the mix samples melted easily. Although acetone can melt wax easily, it could only melt a few mixtures in our experiments, possibly because the wax was coated with PVA and acetone could not reach the wax component. Thus, we assumed that the mixture of wax and PVA would melt easily in the DI water and acetone combination; this was verified by our experiment. Comparing these results, we observed that all mixtures of wax and PVA could be melted in the DI water and acetone combination, but the solution rate was not constant. PVA absorbed DI water and increased in mass at the beginning of these experiments. Our findings

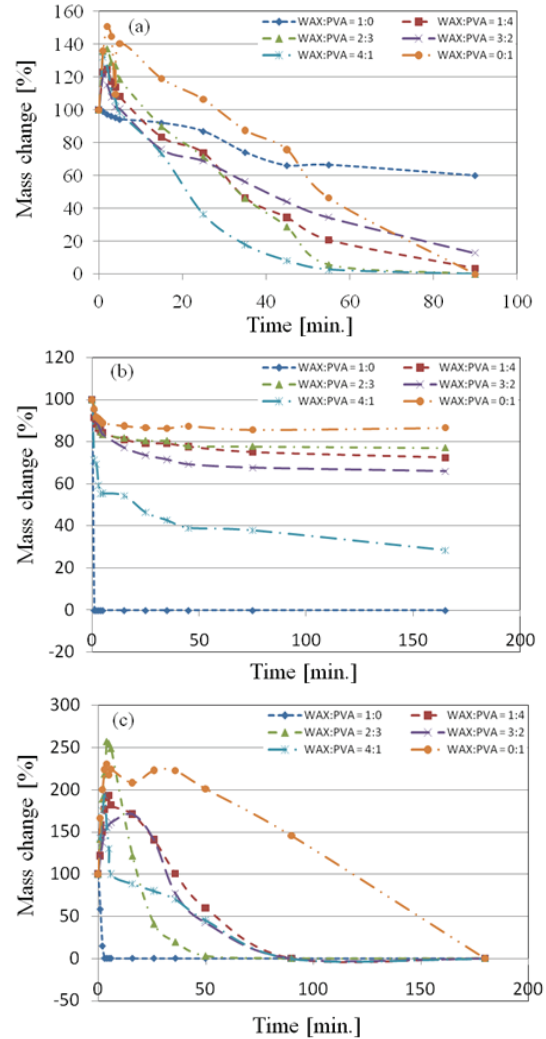


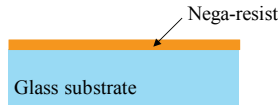
Fig. 7. Evaluation of chemical features. (a) DI water; (b) acetone; and (c) acetone + DI water.

indicate that a 1:1 combination can melt wax and PVA mixed materials.

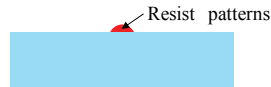
B. Fabrication of arteriole membrane models

Since wax is fragile, we opted to use a mixture of wax and PVA as a material for our sacrificial model. During the fabrication process of the membrane models, we created model patterns by grayscale lithography and transcribed the resist patterns onto PDMS (**Fig. 8**). We aligned two patterned PDMS substrates filled with the wax and PVA mixture on the patterned side, and dried them at room temperature. The fabricated sacrificial model (**Fig. 9**) was coated with PVA to smooth the surface and later dip coated with a transparent silicone resin to achieve a membranous structure. Then, the sacrificial model and PVA used for smoothing were dissolved by DI water and acetone leaving a synthetic transparent arteriole membrane, as illustrated in **Fig. 10**.

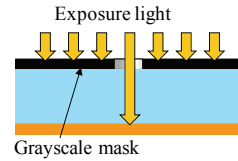
1. Coating a negative photoresist on the glass substrate



3. Development with best condition



2. Exposure with best condition from back of substrate



4. Transcribing the resist patterns onto PDMS

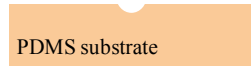
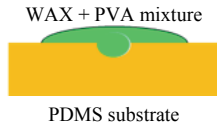


Fig. 8. Process chart of grayscale lithography.

1. Coating WAX + PVA mixture



3. Drying mixture



2. Splitting superfluous mixture



4. Aligning and bonding by WAX + PVA mixture



Fig. 9. Process chart for the sacrificial model.

Figure 11 shows a $\phi 500 \mu\text{m}$ arteriole membranous model with a circular cross section fabricated using this technique.

a. Fabrication process of sacrificial models

For fabricating arteriole membrane models, sacrificial models, fabricated using the wax and PVA mixture, were needed as molds for dip coating of PVA and silicone resins. Patterned PDMS substrates were fabricated by grayscale lithography with negative photoresist, and resist patterns were transcribed onto PDMS for molding the sacrificial models. The fabrication process was as follows:

- A) Grayscale lithography process (**Fig. 8**).
 1. Coating photoresist on the glass substrate.
 2. Exposing using the best exposure conditions from the back of the substrate.
 3. Developing with the best time assessed.
 4. Transcribing the resist patterns onto PDMS.
- B) Fabrication process used for the sacrificial models (**Fig. 9**).
 1. Coating the wax and PVA mixture on the patterned side of the PDMS substrate.

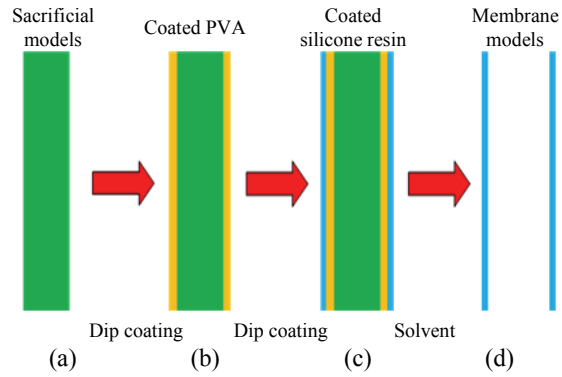


Fig. 10. Process chart for membrane model.

Arteriole membrane model

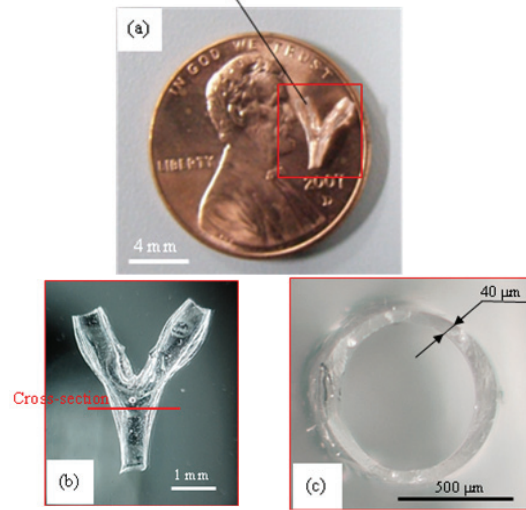


Fig. 11. $\phi 500 \mu\text{m}$ arteriole membrane model made using wax and PVA mixture material and grayscale lithography. (a) A fabricated arteriole membrane model on a penny; (b) A fabricated arteriole membrane model; (c) The cross section of a fabricated arteriole membrane model.

2. Splitting and removing the superfluous mixture.
3. Aligning and bonding two patterned PDMS substrates filled with the mixture on the patterned side.
4. After drying at room temperature, the PDMS substrates were removed to complete the sacrificial model.

The staking mixture was removed from the sacrificial model.

b. Fabrication process of membrane models

We fabricated arteriole membrane models using sacrificial models, which were coated with PVA to smooth the surface, and later dip coated with a transparent silicone resin to achieve a membranous structure. After dip coating, the sacrificial model and PVA for smoothing were dissolved in DI water and

acetone, resulting in a transparent arteriole membrane model. The fabrication process was as follows:

1. Dip coating of the sacrificial model with a 10 wt% PVA solution to smooth the surface.
2. After drying the PVA, the sacrificial model was dip coated with silicone resin.
3. The sacrificial models and PVA used for smoothing were dissolved in a DI water and acetone liquid mixture.
4. The membranous silicone structures were dried to obtain blood vessel membrane models.

We observed that the fabricated arteriole membrane model, shown in **Figure 11**, had a hollow structure and a circular cross section (**Fig. 11 (b)**). The circularity of the inside of the membrane model was calculated by dividing the shortest axis by the longest axis—the calculated circularity of this model was 90%.

IV. Transparent membranous and block models

Figure 12 shows the fabricated arteriole model; using this as an interface we can easily realize circulation type blood vessel simulator. In applications, this model will be used to evaluate drug delivery systems, diacrisis, and medical treatments by ultrasound.

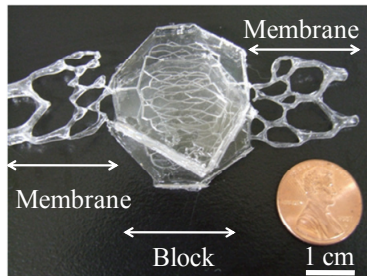


Fig. 12. Transparent membranous and block arteriole model.

V. Conclusions

We demonstrated a method for fabricating $\phi 10 - 500 \mu\text{m}$ transparent arteriole and capillary vessel models.

In fabricating block models, we improved the shape of the channel cross section with alignment patterns, which are very useful in assembling arteriole and capillary vessel models. We calculated the circularities of fabricated $\phi 10, 50,$ and $500 \mu\text{m}$ microchannels as 84.0%, 61.5%, and 82.3%, respectively. Flow experiments demonstrated that these channels had no leakage.

In fabricating membrane models, we proposed a fabrication method of $\phi 100 - 500 \mu\text{m}$ arteriole membrane models for surgical simulator and for connections. The arteriole membrane models were fabricated using grayscale lithography and a wax and PVA mixture material. We evaluated the mechanical and chemical properties of the mixture and observed that Young's modulus of the mixture material increased with the ratio

of wax. In addition, the mixture's extension increased with the ratio of PVA. DI water and acetone liquid mixture was the most effective for melting the wax and PVA mixture material. The proposed approach overcame the brittleness of the previous sacrificial models. The membrane model had a circular cross section with a channel circularity of 90%.

Finally, we succeeded in fabricating a transparent arteriole model. It is necessary to choose an appropriate exposure method. In the future, we plan to improve the accuracy of our model by (1) optimizing the fabrication conditions; (2) matching moderate compliance, which is similar to that of real blood vessels; (3) making complicated membrane models; and (4) testing the durability and reliability of these models in a surgical simulator. With this model, we plan to realize a circulation type blood vessel simulator.

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