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Membrane Micro Emboss (MeME) Process for 3-D Membrane Microdevice

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1. Introduction

Recent advances in micro- and nanofabrication technologies have enabled the development of miniaturized accelerometers, gyroscopes, μTAS chips, etc. These microdevices are made of substrates having thicknesses relatively greater (~100μm) than the feature scale of the microfabricated components (1~10 μm, Fig.1a). Conversely, the microscale organelles or tissues of natural creatures are made of substrates, or membranes, that are relatively thin compared to their feature size. For example, a human blood capillary, which is 10~100 μm in diameter, has vessel walls with thicknesses of ~1 μm. To give another example, a cell with a diameter of ~10 μm is composed of lipid bilayer membranes with thicknesses of ~10 nm. This fundamental characteristic of the architecture of biological microstructures, which is totally different from that of artificial microdevices, makes life a highly adaptable system from both chemical and physical perspectives. The small thickness of the membrane enhances transport of heat and substances between the body and its surroundings, and it provides softness to the body, enabling passive and active morphological changes for adapting to the environment. These characteristics of biological microstructures should greatly encourage us to develop new types of MEMS and μTAS devices. However, in reality, little research has been conducted on the development of 3-D microdevices composed of thin membranes, which we call "3-D membrane microdevices" (Fig.1b).
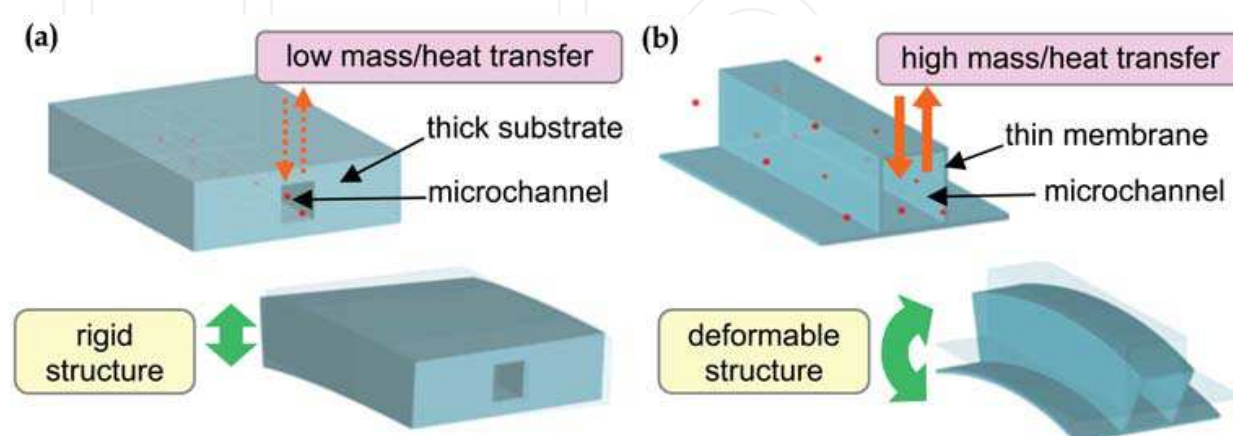

Fig. 1. Schematics of (a) conventional "bulk" microdevice and (b) "3-D membrane microdevice"

The purpose of this chapter is to introduce the concept of 3-D membrane microdevices and highlight some advances being made in our laboratory. The chapter starts with a section describing a novel microfabrication technique, namely, the membrane micro emboss (MeME) process, which was developed to realize 3-D membrane microstructures. In the following sections, several applications of 3-D membrane microdevices in µTAS and MEMS fields are presented. First, a microfluidic device composed of thin porous biodegradable membranes is described. This device was developed for tissue engineering purposes. Next, a novel micropneumatic actuator composed of folded 3-D membrane chambers is described. The actuator was intended for use as a microactive catheter for safer intravascular treatment. Finally, we conclude the chapter and present our perspectives on 3-D membrane microdevices.

2. Membrane Micro Emboss (MeME) process

Various micro fabrication processes can be used to fabricate MEMS or µTAS devices. However, few processes are useful for the fabrication of 3-D membrane microstructure, especially for polymer materials. Among conventional microfabrication processes, the chemical vapor deposition (CVD) process using parylene and the microthermoforming process can be employed. Although the CVD process using parylene is used to fabricate 3-D membrane microstructures (Zhenga et al., 2007; Liua et al., 2008), the limitations caused by the unavailability of suitable materials and low production rates present significant problems. The microthermoforming process (Truckenmüller et al., 2002; Giselbrecht et al., 2006) can be applied to a wide variety of thermoplastic materials and is suitable for mass production; however, it cannot be applied to highly porous membranes because the pressurized fluid leaks through the pores.
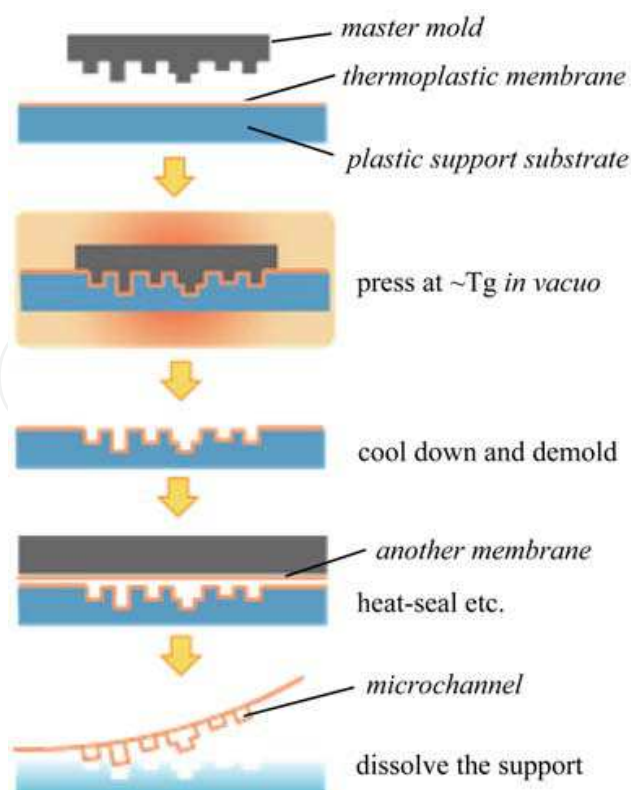

Fig. 2. Flowchart of the MeME process

The MeME process (Fig.2) was developed to realize 3-D membrane microstructures from a wide variety of materials including porous materials (Ikeuchi & Ikuta, 2005; Ikeuchi & Ikuta, 2006, a). This process needs a master mold, a thermoplastic polymer membrane, and a deformable plastic support substrate. First, the polymer membrane is set between the master mold and the support substrate. Then, this assemblage is heated to temperatures around the glass transition point (Tg) of the polymer membrane. Next, the master mold is pressurized against the membrane *in vacuo*. During pressurization, the membrane is deformed along with the support substrate to match the surface of the master mold. After cooling to the initial temperature, the master mold is separated from the deformed membrane. To fabricate sealed microchannels, another planar membrane is placed on the deformed membrane and sealed using heat-sealing, solvent gas bonding, or other sealing techniques. The fabrication of the membrane microfluidic device is completed by dissolving the support substrate in a selective solvent. The process is applicable to various materials, since it only requires the membrane material to be thermoplastic. When polylactic acid (PLA) (Tg, 57°C; thickness, 5 μm) was used as a membrane material and paraffin (melting point, 70°C) was used as a support substrate, the lateral and vertical resolutions of the process were at least 10 μm and 5 μm, respectively (Fig. 3) (Ikeuchi & Ikuta, 2006, a). The resolutions can be further improved by using thinner membranes and harder support substrates.

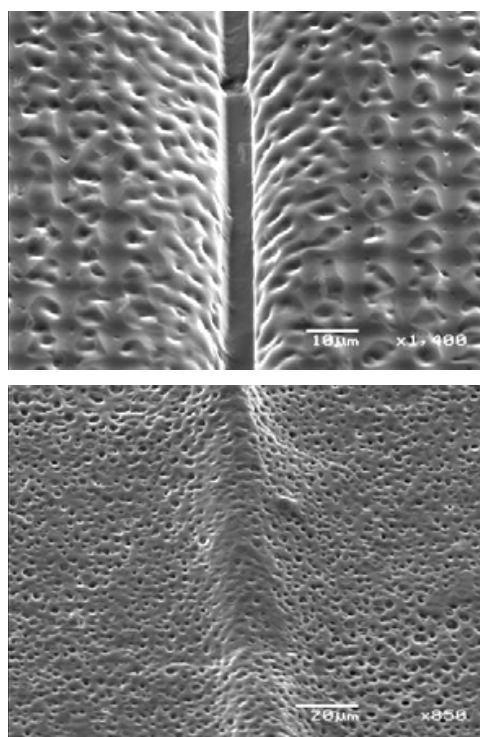In the following two sections, several applications of the MeME process are described.

Fig. 3. SEM images of the topside (upper) and backside (lower) of the deformed porous PLA membrane.

3. Membrane microfluidic device for tissue engineering

3.1 Background

Throughout the history of biology, cell culture has been carried out on planar glass or in polymer dishes. The cells cultured on a planar substrate proliferate laterally to form a thin

layer of cells. Biologists have studied cellular dynamics using these two-dimensional cellular constructs. In the natural environment, however, cells proliferate three-dimensionally, and thus, show behaviours and functions different from those of cells in 2-D *in vitro* cultures.

Recently, cell culture in 3-D conditions has attracted considerable attention for studying natural cell behaviours and, from a more practical perspective, for regenerating fully functional large tissues and organs for transplantation. Some biologists culture cells under 3-D conditions by using soft hydrogel materials (collagen, Matrigel™, etc.) or stacking cell sheets (Liu & Bhatia, 2002; Bryant & Anseth, 2002; Sekiya et al., 2006.). There is a big difference, however, between artificial 3-D conditions and *in vivo* conditions because of the thickness of the cultured cellular constructs. Thick tissues *in vivo* can survive on nutrients supplied from surrounding blood capillary networks. In contrast, we can stack only a few layers of cells *in vitro* due to limitations with regard to the diffusion distance of nutrients, which can be supplied only through the outer surface of the construct.

To solve this problem, King et al. (2004.) attempted to construct microfluidic chips made of biodegradable polymers. They fabricated microchannels in biodegradable polymer substrates using μTAS or lab-on-chip technologies, and they cultured cells on the chip by supplying the culture medium through microchannels (Fig. 4a). They were unable to culture thick tissues, however, because the cells cultured on the chip tended to be distributed at a low density with poor homogeneity. These problems arise due to the thickness of the chip. Cells seeded on the thick microchannel chips proliferate on the surface of the chip rather than growing within the chip substrate.

In this section, we describe the MeME process as applied to fabricate 3-D thin membrane microstructure, which solves the problems associated with conventional methods for tissue engineering.

3.2 Artificial capillary network chip

To realize both the nutrients supply and homogeneous cell distribution in 3-D constructs, we propose the artificial capillary network chip as a novel 3-D cell culture device (Fig. 4b). This chip has a microchannel network made of a thin biocompatible polymer membrane with penetrating micropores.

Cells seeded on this chip with soft hydrogel materials, or cells stacked on this chip as cell sheets, can maintain a thick 3-D construct because of nutrients supplied from the porous microchannel network. Unlike the thick conventional microchannel chip, the membrane composing the microchannel wall is thin enough for cells to distribute homogeneously in the 3-D constructs. Biodegradable polymers can be used, instead of conventional polymers, as the membrane material to regenerate tissues for transplantation. Larger tissues can be fabricated by stacking these chips (Fig. 4c).

A prototype of the chip with highly branched microchannels was fabricated from a porous PLA membrane. The porous PLA membrane was formed by spin-coating following phase separation technique (Ikeuchi & Ikuta, 2006, b). The diameter and density of the pores can be controlled independently by adjusting the water content and PLA content of the coating solution, respectively. Here, the pore diameter was adjusted to $\phi \sim 1$ μm to prevent the cells ($\phi \sim 5$ μm) from entering the microchannel, and the thickness was adjusted to 5 μm.

The master mold was made by microstereolithography developed in our laboratory (Ikuta & Hirowatari, 1993). The surface of the mold was coated with a fluorocarbon polymer for easy
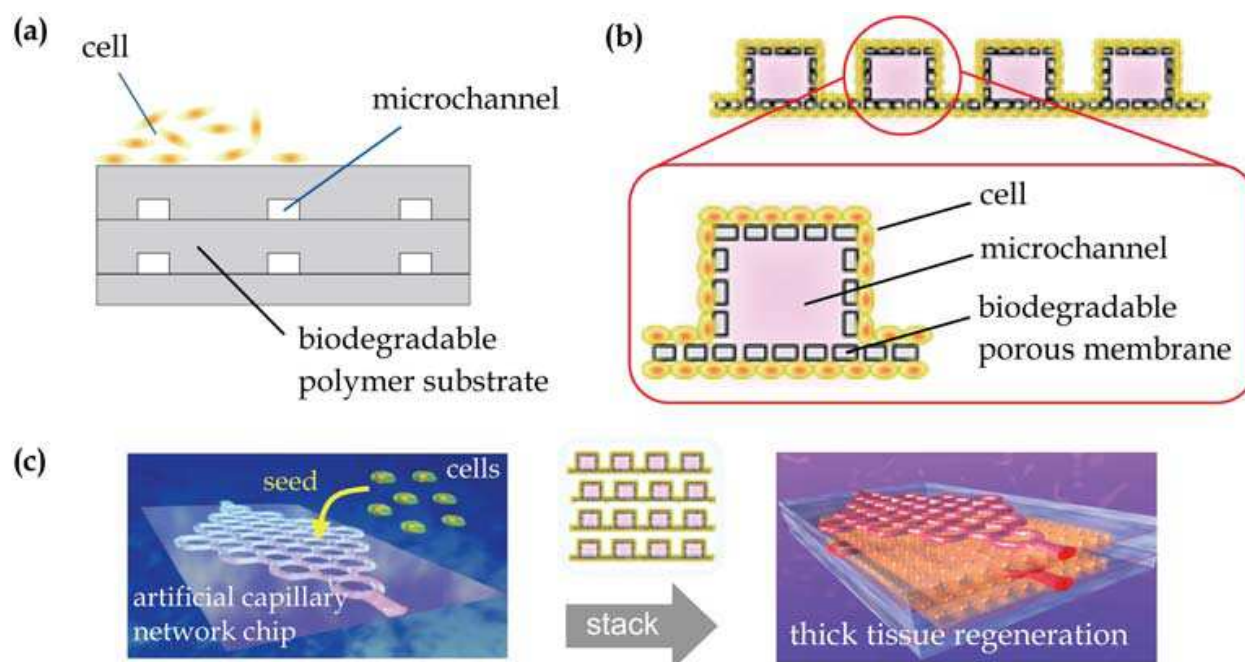
Fig. 4. (a) Schematic cross-section of the conventional microchannel chip for cell culture. (b) Schematic cross-section of artificial capillary network chip. (c) Conceptual scheme of *in vitro* 3-D thick tissue regeneration using artificial capillary network chip

removal of the mold. The master mold was pressurized onto the membrane at 0.5 µm/s for 500 s. at 55°C *in vacuo*. After cooling to 25°C, the mold was removed. The embossed membrane was heat-sealed with another membrane of the same material at 70°C for 30 s. A red solution was filled into the microchannels by capillary force. No leaking or blockage of the microchannel was observed (Fig. 5a). Figs. 5b and c show the topside and the backside of a microchannel in the prototype chip before sealing, respectively. Most of the micropores can be preserved on both sides of the microchannel wall even after the MeME process by fine tuning the process parameters (speed, temperature, and material of support substrate).

3.3 Validation of the chip
To check the size-selective permeability of the microchannel wall of the chip, a suspension of microbeads with diameters varying from φ100 nm to φ15 µm was poured on the chip (Fig. 6a). Beads smaller than φ1 µm penetrated the wall but larger beads were trapped on the wall (Fig. 6b). This result means that nutrients and gases flowing through the microchannels can diffuse out into the cellular constructs on the chip, while at the same time, the microchannel walls support the thick 3-D cellular constructs.

The biocompatibility of the chip was also tested by culturing human endothelial cells (HUVEC) using another prototype chip. Fig. 7a shows a fluorescent image of the cells on the chip after culturing for 120 h. The cells spread as usual and showed no damage. The time course of the cell density on the chip was also equivalent to that for conventional tissue culture polystyrene flasks (Fig. 7b). These results prove that the chip was biocompatible with HUVEC. The success of HUVEC culture on the microchannel offers interesting possibilities for co-culture with other parenchyma cells to fabricate functional tissues.
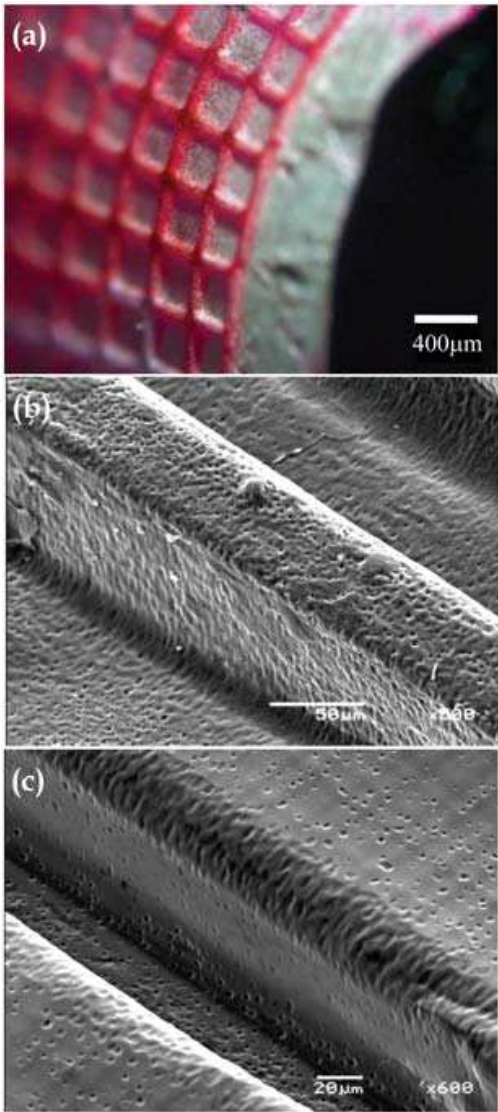
Fig. 5. A prototype chip made of a porous PLA membrane. (a) Optical microscopy image. (b, c) SEM images of the topside and the backside of the chip, respectively.
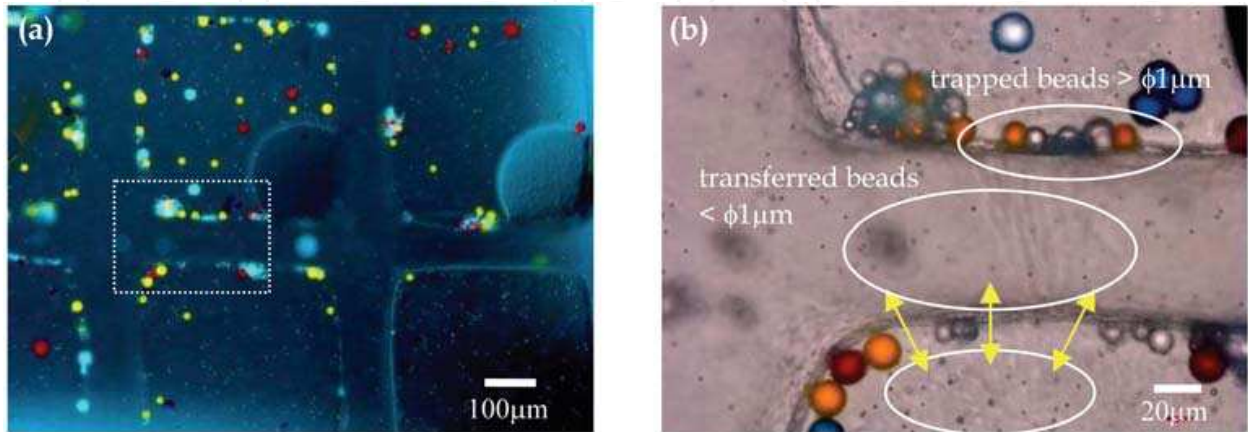

Fig. 6. (a) Fluorescent microscopy image of the chip after pouring a microbead suspension (b) Magnified view of the white-rectangle area in (a).
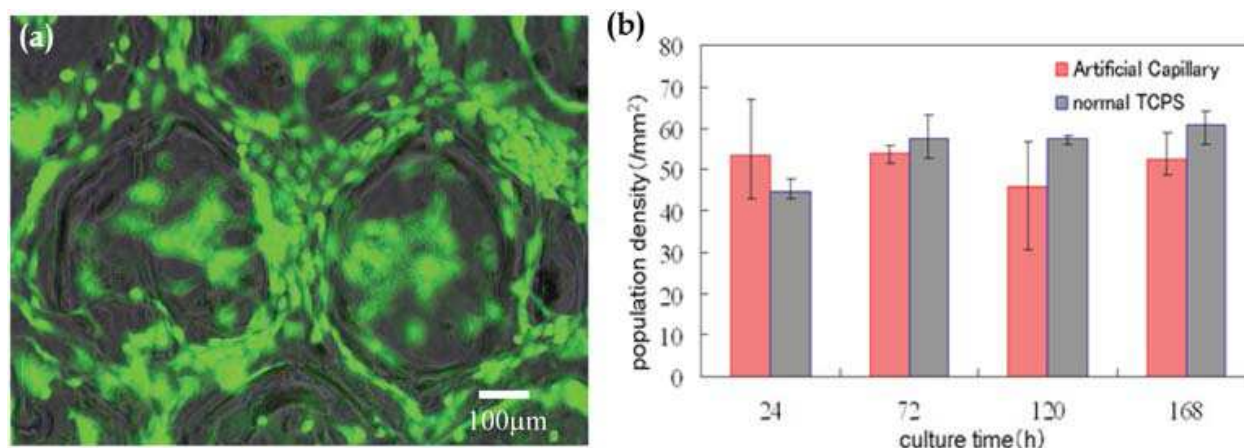
Fig. 7. HUVEC culture on the chip. (a) Fluorescent view of the cell on the chip after 120 h. (b) Transition of cell population density with culture time.

3.4 Summary

In this section, the artificial capillary network chip with a 3-D membrane microstructure was proposed and its development from the viewpoint of realizing thick 3-D tissue culture *in vitro* was described. Prototype chips were successfully fabricated using the MeME process, and their size-selective permeability and biocompatibility were verified. This chip could potentially become a key technology in the study of cellular dynamics under 3-D conditions; moreover, it could be used to regenerate large tissues or organs for transplantation in the near future.

4. Pressure-driven microactive catheter

4.1 Background

Recently, catheterization has been widely applied in intravascular surgery as an alternative to conventional surgical techniques, which are highly invasive. In catheterization, a thin flexible tube called a catheter is inserted into a blood vessel from the leg or arm. The catheter can be advanced into the patient's heart or brain for treatment or inspection. The operation leaves just a tiny puncture on the arm or leg where the catheter has been inserted, and therefore, causes less damage and fewer scars on the patient than conventional open surgery.

A major problem with catheterization, however, is the difficulty of manipulation in narrow and branched blood vessels. Since conventional catheters have no active bending capability at the tip, the doctor can control the direction of the tip only by pushing and rotating the catheter at the inlet which is far away from the tip. Thus, catheterization in narrow and complicated blood vessels is extremely difficult.

To solve this problem, several types of active catheters have been proposed (Mineta et al., 2002; Ikuta et al., 2003; Fang et al., 2007). They are classified into two types depending on the bending mechanism. The first type consists of electrically driven active catheters. These catheters have actuators that use shape memory alloys or polymer gels at the tip and can be bent from outside the body by applying a current to the actuators. Even though electrical actuators are suitable for miniaturization, the use of electricity inside the heart or brain

poses the risk of fatal damage due to microshock or heat in the case of an accident (Manecke et al., 2002; Bunch et al., 2005).

The second type consists of a pressure-driven active catheter, as proposed by Ikuta et al. (2003). It has a hollow bellows made of soft silicone rubber at the tip, and the tip can be bent by supplying saline water into the bellows through a tube connected to the bellows. Since no electricity is necessary for actuation, it is superior in safety compared to electrically driven active catheters. In addition, it can be applied to MRI monitoring, which is a fundamental tool in catheterization, because no metal parts are used in this catheter. In spite of its superiority, the minimum size of this type of catheter that can be attained with conventional injection molding processes using a pair of a male and a female mold is $\phi \sim 1$ mm, whereas the catheter must be smaller than $\phi \sim 300$ μm for complex intravascular surgery. This limitation arises due to the difficulties involved in 3-D fabrication of a pair of male and female molds with micrometer accuracy.

Although a pressure-driven balloon-type microactuator made from a polydimethylsiloxane (PDMS) molding technique was reported for use in MEMS applications (Konishi et al., 2006), it cannot be applied to catheterization due to the risk of damage to blood vessels caused by large expansion of the actuator during bending. In short, there is no process available to fabricate microscale pressure-driven active catheters.

In this section, we describe how the MeME process can be combined with an excimer laser ablation technique to realize a pressure-driven microactive catheter with a 3-D thin membrane microstructure.

4.2 Bellows composed of folded membrane microchambers

We designed a pressure-driven microactive catheter composed of hollow bellows; the catheter was made of a biocompatible polymer membrane (thickness, 5 μm), a motorized syringe, and a Teflon microtube (Fig. 8a). A pressure gauge was attached to the microtube at the base to monitor the pressure and provide the pressure value as feedback to the motorized syringe. The diameter of the catheter was set at ϕ 300 μm, because that is the minimum size used in clinical practice.

The bellows are composed of a series of folded microchambers and microchannels connecting the chambers. Since the bottom of each folded chamber is fixed to another membrane, only the upper part of the chamber can be expanded by increasing the inner pressure of the chamber (Fig. 8b). Thus, the bellows in their entirety can be bent in one direction by supplying saline water from the syringe through the microtube, because only one side of the bellows extends. Furthermore, the alternating arrangement of microchannels and microchambers prevents the bellows from expanding in diameter during bending, since the microchannels work as rigid frames to connect the topside and backside membranes of the bellows.

The catheter was fabricated using the membrane micro emboss following excimer laser ablation (MeME-X) process (Fig. 9) (Ikeuchi & Ikuta, 2008). In the MeME-X process, at first, the hollow microbellows were formed from PLA membranes (thickness, 5 μm) using the MeME process. By using excimer laser ablation (ArF, 193 nm), the outline of the bellows was cut out from the sealed membranes, and an opening was made at one end. After the bellows were connected to a microtube by an adhesive under an optical microscope, the
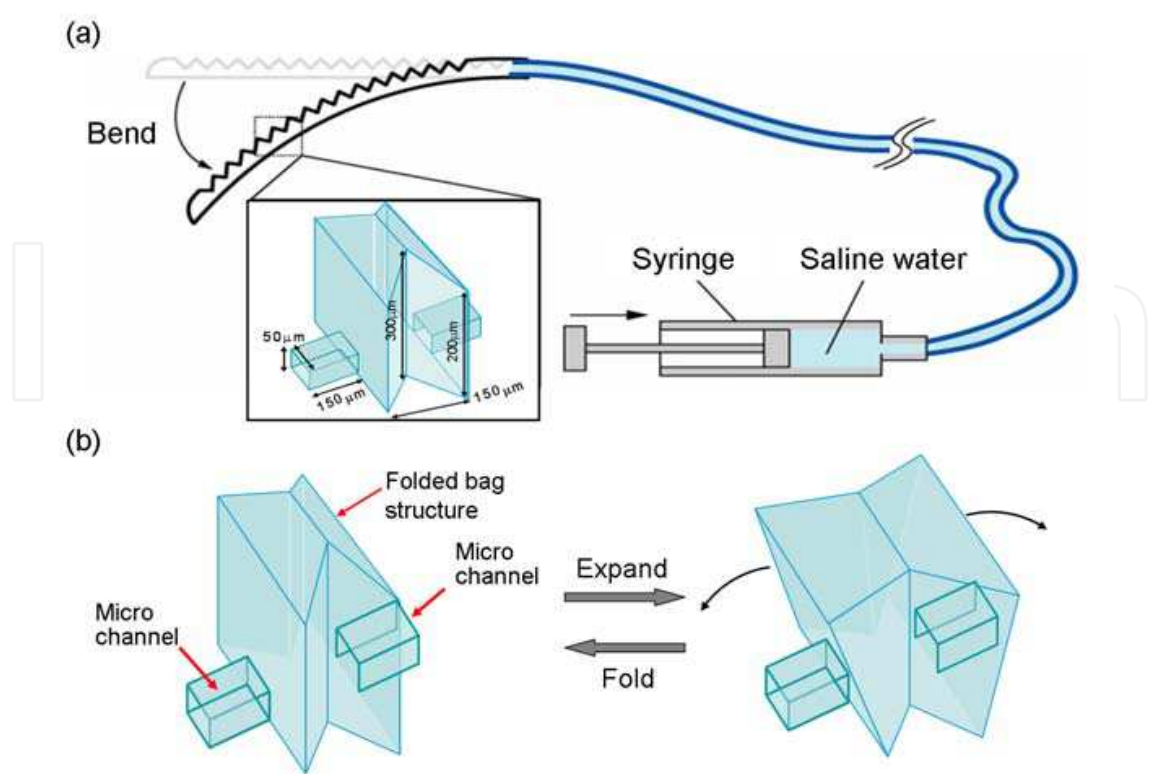
Fig. 8. (a) Schematic of the pressure-driven microactive catheter system with bendable bellows made of a thin membrane at the tip (b) Bending of the bellows through expansion of each folded microchamber
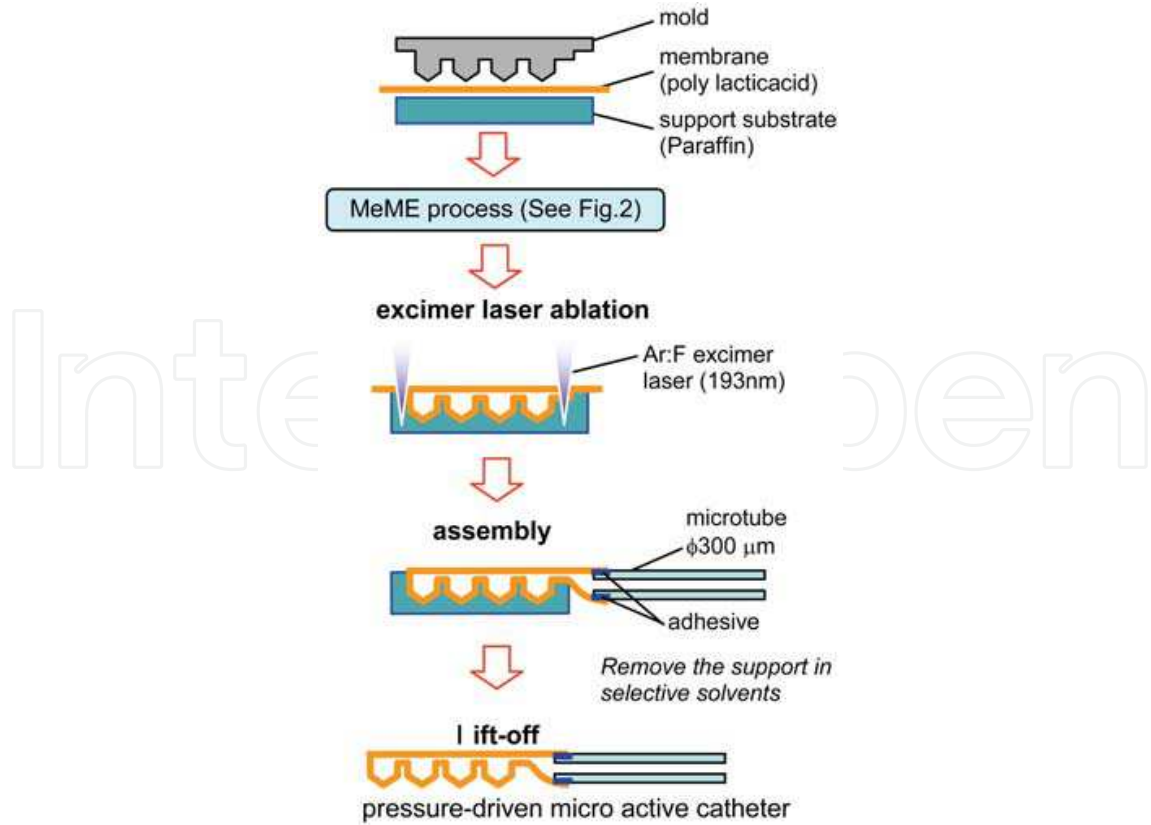

Fig. 9. Flowchart of the MeME-X process

support substrate was selectively dissolved by immersion in hexane. Finally, the catheter was successfully fabricated (Fig. 10a). The entire process was completed in 10–15min. To show the cross-section of the hollow bellows, the bellows were cut in the middle using the excimer laser. The bellows composed of a series of folded microchambers and microchannels were precisely fabricated on both the outside and the inside, and the thin membrane was uniformly deformed to yield a hollow microstructure (Fig. 10b).
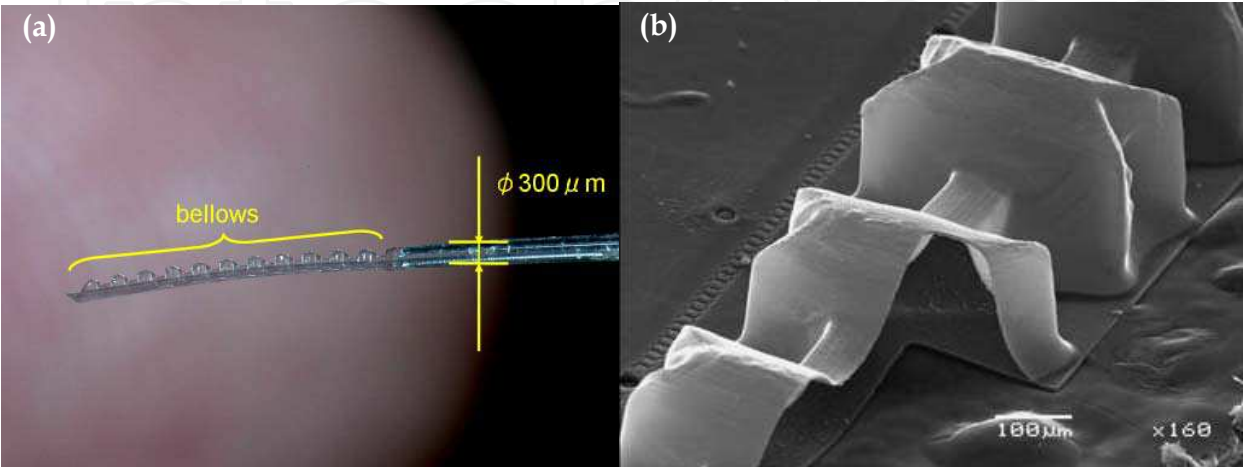

Fig. 10. (a) Completed pressure-driven micro active catheter ϕ 300 μm. (b) SEM image of the bellows cut at the middle to show the cross-section and the inner structure.

4.3 Validation of the catheter

The bellows were bent at an arbitrary angle between 0° and 180° through water pressure applied by a motorized syringe (Fig. 11a). The range of the bending angle is sufficient for intravascular operation, and it can be extended by increasing the folding angle of each microchamber of the bellows or by increasing the number of microchambers, if necessary.
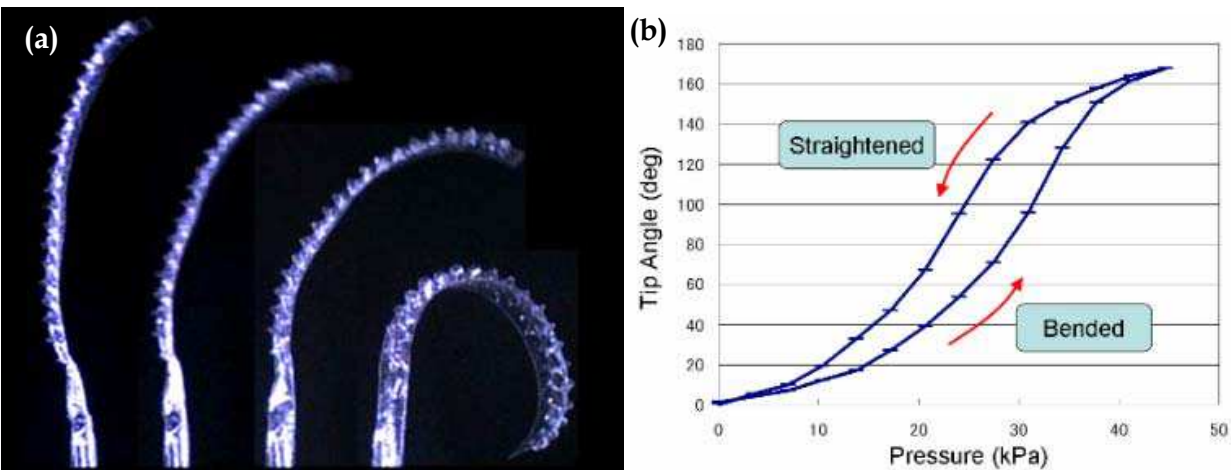

Fig. 11. (a) Bending demonstration of the pressure-driven micro active catheter from 0 to 180 degrees. (b) Relation between applied pressure (P) and bending angle (θ) of the tip

The hysteresis of the P-θ curve is apparently caused by the buckling behavior of the folded chambers and air trapped in the microtube (Fig. 11b). The buckling behavior can be improved by modifying the folding angle and pattern of the chambers, and the trapping of air in the system can be prevented by assembling the catheter *in vacuo*. Most importantly, little increase in the diameter of the bellows was observed during bending due to the microchannels inserted between the microchambers. This leads to a safer and smoother insertion of the catheter at bifurcations.

For *in vitro* demonstration of the active catheter, a small blood vessel model made of silicone was fabricated using the lost-wax method. The model consists of narrow blood vessels of φ 1 ~ 3 mm into which conventional active catheters could not be inserted. The pressure-driven microactive catheter was actuated and inserted into the narrow vessels (Fig. 12a). At the bifurcation, the catheter was bent slightly to the left from the straight position (Fig. 12b,c) by supplying saline water from the syringe, turned to the desired direction, and then successfully introduced into the target aneurysm (Fig.12d).
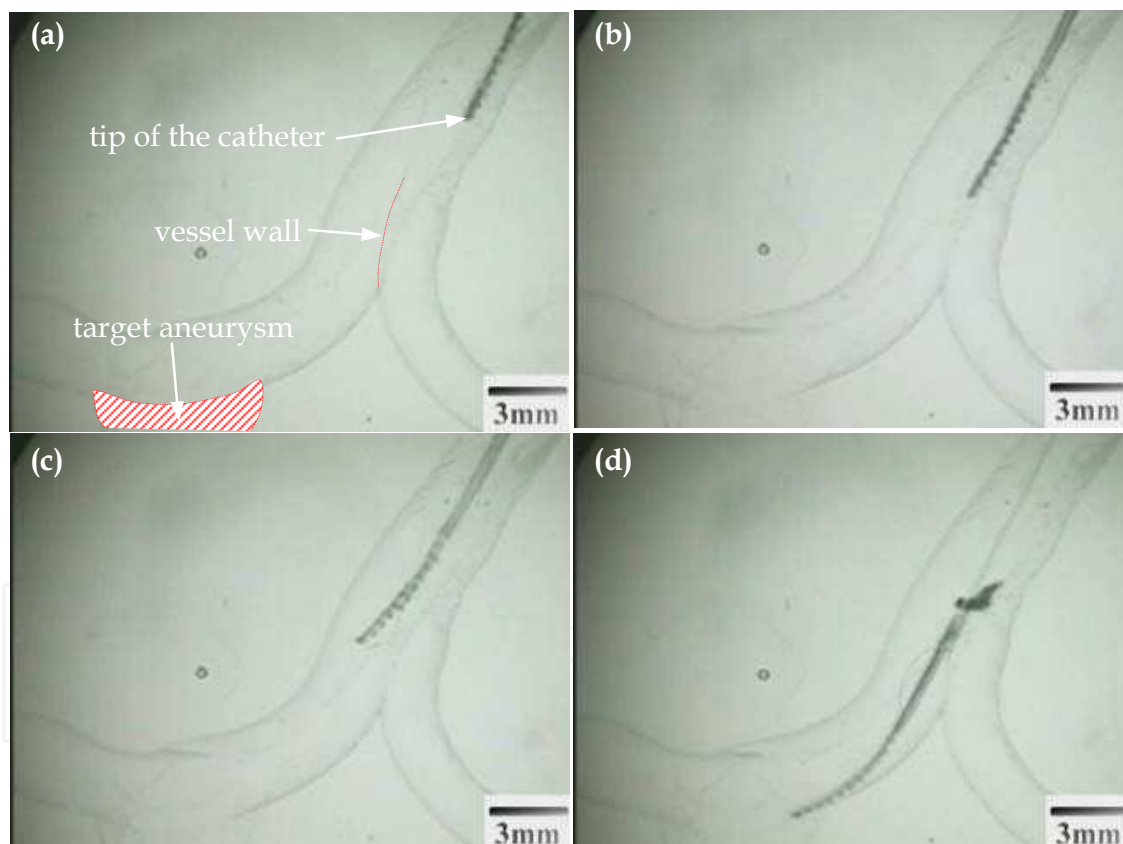

Fig. 12. Video frames showing insertion of the catheter into a 3-D vascular model

4.4 Summary

In this section, the pressure-driven microactive catheter was proposed and its development by the MeME-X process was described. The pressure-driven microactive catheter, with its extremely small size and high safety, should promote the application of catheterization in

complex intravascular surgery, which is at present not possible with conventional surgical tools. For further improvements, microchannels for drug delivery and/or blood sampling could be attached to the bellows. This can be achieved by simply adding microchannel templates on the master mold of the bellows. Furthermore, the nonelectrical actuation mechanism of this catheter, which has a 3-D membrane microstructure, can be widely extended to safe medical tools and microactuators in the microrobotics field.

5. Conclusions and perspectives

In this chapter, the concept of 3-D membrane microdevices was introduced and the development of the MeME process was described. To utilize its characteristics, the concept was applied to actual devices in two different fields. First, focusing on the efficient transfer of substances and heat in 3-D membrane microchannels, an artificial capillary network chip was developed for tissue engineering purposes. Second, utilizing the high elastic deformability of 3-D membrane microstructures, hollow bellows composed of folded microchambers and microchannels were developed to realize a pressure-driven microactive catheter.

Biological organisms are fundamentally characterized by a 3-D membrane microstructure. From intracellular organelles to vascular networks, from plant leaves to insect wings, the exquisite architectures prevalent in nature greatly inspires us to develop novel micro/nanodevices. The study of 3-D membrane microdevices has just emerged out of the proof-of-concept stage. To further expand the scope of applications of 3-D membrane microdevices, our laboratory is actively engaged in the exploration of a variety of materials applicable to the MeME process and improvement of the resolutions of the MeME process toward the nanometer scale. With its unique advantages, the 3-D membrane microdevice technology should contribute to drug delivery, tissue engineering, electric power generation, smart skin development and many other fields in the near future.

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Micro Electronic and Mechanical Systems

Edited by Kenichi Takahata

This book discusses key aspects of MEMS technology areas, organized in twenty-seven chapters that present the latest research developments in micro electronic and mechanical systems. The book addresses a wide range of fundamental and practical issues related to MEMS, advanced metal-oxide-semiconductor (MOS) and complementary MOS (CMOS) devices, SoC technology, integrated circuit testing and verification, and other important topics in the field. Several chapters cover state-of-the-art microfabrication techniques and materials as enabling technologies for the microsystems. Reliability issues concerning both electronic and mechanical aspects of these devices and systems are also addressed in various chapters.

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