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REVIEW ARTICLE

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Treatment of lymphedema with lymphaticovenular anastomoses

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Abstract Although lymphedema in the extremities is a troublesome adverse effect following radical resection of various cancers, conventional therapies for lymphedema are not always satisfactory, and new breakthroughs are anticipated. With the introduction of supermicrosurgical techniques for the anastomosis of blood or lymphatic vessels less than 0.8 mm in diameter, we have developed a novel method of lymphaticovenular anastomosis for the treatment of primary as well as secondary lymphedema in the extremities. Here, we review the pathophysiological aspects of lymphedema, emphasizing the importance of smooth-muscle cell function in the affected lymphatic walls. We then describe the theoretical basis and detailed operative techniques of our lymphaticovenular anastomoses. Although technically demanding, especially for beginners, we believe that this method will become a new clinical standard for the treatment of lymphedema in the near future.

Key words Lymphedema · Lymphaticovenular anastomosis · Supermicrosurgery · Lymphangiogenesis

Introduction

Lymphedema in the extremities is one of the most troublesome and annoying adverse effects after the surgical treatment of various cancers. The importance of treatment for post-surgical lymphedema has not been fully appreciated among clinical oncologists, possibly because it is not life-threatening. However, it markedly perturbs patients' activities in daily life: working, housekeeping, and even walking. Non-surgical conservative therapies such as bandaging or massaging are usually recommended for patients, but the effects are quite limiting and time-consuming. Massive surgical treatments such as Thompson's method[1] are also obsolete, due to their extreme invasiveness and lack of satisfactory outcomes.

In the field of plastic and reconstructive surgery, a novel technique of supermicrosurgery (anastomosis of vessels or lymphatics with diameters of 0.5–0.8 mm) has radically changed the concept of microvascular anastomosis.[2] We believe that our strategy of supermicrosurgical lymphaticovenular anastomosis could be one of the most promising approaches for the treatment of lymphedema.[3–7] In this review, we first describe the pathogenesis of lymphedema, especially focusing on the vicious cycle of its progression and the functions of lymphatic smooth-muscle cells. Next, we describe our present techniques and recent refinements of lymphaticovenular anastomosis, together with some future perspectives for lymphedema therapies.

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Pathophysiology of lymphedema

Basic biology: lymphangiogenesis

Recent advances in the molecular understanding of vessel formation are quite outstanding. To begin with, we give a brief overview of basic science regarding lymphangiogenesis.

The mechanisms of lymphangiogenesis can be understood as analogous to those of angiogenesis. The vascular endothelial growth factor (VEGF) family and its receptors are well-known key players in angiogenesis, among which the VEGF-C, D/VEGF receptor (R)3 signal is found to be specific for lymphangiogenesis.[8] Indeed, mutations of the *VEGFR3* gene are found in the *Chy* mouse, an animal model of primary lymphedema,[9] and in human subjects with familial lymphedema.[10] Some malignant tumor cells express VEGF-C, and it is considered that tumor-derived VEGF-C induces lymphangiogenesis around the tumors and their

sentinel lymph nodes, thereby promoting lymphatic metastasis.[11,12]

What is the origin of the lymphatic vessels? As an analogy of the mechanisms of vasculogenesis and angiogenesis, there have been two theories regarding the development of lymphatic vessels. One is the de-novo formation of neolymphatics within the mesenchyme, and the other is a sprouting and transdifferentiation from preexisting vascular vessels. Recent analyses using mouse embryos seem to support the latter hypothesis: the first lymphatic vessels are formed in the cervical region by sprouting from the vein around embryonic day 10, and transdifferentiation of the lymphatic endothelium from the vascular endothelium is likely to be mediated by a transcription factor, Prox-1.[8,13]

Etiology and vicious cycle of lymphedema

Lymphedema is regarded as a low-output failure of the lymphatics that results in the stagnation of plasma protein molecules escaping the microcirculation, causing a high-protein edema.[14] Primary lymphedema includes Milroy's disease (congenital familial lymphedema), which was recently proven to be caused by a missense mutation of the *VEGFR3* gene, as referred to above.[10] Another type of primary lymphedema is often seen in young women, and precise diagnosis is often very difficult in such cases. Secondary lymphedema is caused by various factors. Most commonly, lymphedema is seen as a somewhat inevitable outcome following the surgical resection of cancers (such as those of the uterus, prostate, or breast) with lymph node resection and/or irradiation. Lymphedema due to filariasis is seen as endemic in tropical regions of Africa, South Asia, and South America. Other types of secondary lymphedema include obstruction of lymphatics due to trauma or tumors.

Although lymphedema is initially detected as a soft, pitting edema, the edema becomes more severe (non-pitting edema) as it progresses. This change, when prolonged, can be associated later with some irreversible structural changes, such as interstitial fibrosis or atrophy of smooth-muscle cells within the lymphatic walls.[15] It finally develops into so-called elephantiasis, an excess fibrosis and hardening of the skin. Campisi et al.[16] proposed a classification for the progress of lymphedema symptoms (stage I, initial and irregular edema; stage II, persistent edema; stage III, persistent and progressing edema with acute lymphangitis; stage IV, fibrolymphedema; and stage V, elephantiasis). Rare outcomes of lymphedema include lymphangiosarcoma (Stewart-Treves syndrome), the prognosis of which is extremely poor.

The progression of lymphedema can be modified in two aspects: one is the involvement of hydrostatic and osmotic changes, and the other is associated inflammation and infection, thereby forming a vicious cycle of progression. It is well known that the drainage of lymphatic fluid is determined by the interstitial pressure in the extracellular spaces which, in turn, is influenced by the contraction of the subjacent skeletal muscles, particularly during exercise, by arterial pulsation near lymphatics, and by the contraction of smooth muscle cells in the lymphatic trunk wall stimulated by the sympathetic nervous system.[17] In general, the formation of edema is described according to Starling's law:[18]

$$J = k(\Delta P - \Delta \pi) \quad (1)$$

where J is the amount of fluid transfer from the capillary vessels into the interstitial space, k, is the coefficient of vessel permeability, ΔP is the difference in hydrostatic pressure between the inside and outside of the vessels; and $\Delta \pi$ is the difference in osmotic pressure between the inside and outside of the vessels. All types of edema, including lymphedema, can be attributed to changes in the above-mentioned variables. For example, inflammatory or allergic edema is caused by a change in k. The edema seen in congestive heart failure is caused by a change in ΔP. The edema in hypoalbuminemia is caused by a change in $\Delta \pi$. In the pathogenesis of lymphedema, the primary causative change is insufficient lymphatic drainage, as mentioned before. This change, in turn, results in increasing osmotic pressure in the interstitial space, due to lymphostasis. When interstitial edema has progressed, the veins can be compressed within the tissue, resulting in high hydrostatic pressure, specifically in the veins. These factors positively affect the progression of lymphedema as a vicious cycle. Importantly, large veins are more sensitive to the external pressure than small venules, and this fact may be related to the efficacy of lymphaticovenular anastomosis, as described below.

Inflammation and infection are also very important in the establishment of the vicious cycle. Lymphatic fluid serves as a nutritionally ideal medium for bacterium. Therefore, slight skin damage, due to insect bites, dermatitis, or skin fungal infection can be a trigger for massive phlegmon in the extremity with lymphedema. Lymphedema is further worsened by such episodes, accelerating the progression of the vicious cycle, and tissue damage can be irreversible, with some structural changes. The aims for treatment of lymphedema thus should be to avoid progression by stopping these vicious cycles. In other words, a markedly favorable outcome can be anticipated even with small therapeutic interventions, if they can precisely and effectively block the vicious cycles.

Notably, we experienced patients with hemilateral lymphedema of the leg, in whom severe edema progressed bilaterally within a short time. Forty percent of cases of leg lymphedema are bilateral, and this suggests a potential risk of lymphedema in the unaffected side in unilateral cases.[19] Preventive intervention for the unaffected leg, including lymphaticovenular anastomosis, should be considered in unilateral cases.

Histological findings: roles of smooth muscles

In an experimental model, the latent phase of lymphedema is pathologically characterized by massive dilation of lymphatics and intracellular edema of the endothelium or other

components of the lymphatic walls. Endothelial vacuole formation and the deposition of protein-rich materials within the walls are also noted.[20]

Several irreversible pathological changes take place when lymphedema progresses. We carried out histological examination of specimens of lymphatic vessels taken from 18 patients who underwent lymphaticovenous anastomosis.[15] In the initial stage of lymphedema, destruction of the endothelial cells and smooth muscle cells was observed in the lymphatic trunks, especially at the proximal level. The destruction of smooth muscle cells resulted in a loss of the tunica media and dilatation of the thinned lymphatic wall. The lymphatics were then occluded by tissue organization, through which a few small recanalizations formed. In patients with lymphedema of longer duration, the lumen was filled with the internal endothelial layer, which was coated with a number of thick bundles of collagen fibrils, together with numerous smooth muscle cells in some patients. The smooth muscle cells provide a lymph pump, collecting fluid toward the proximal part; therefore, destruction and dysfunction of the smooth muscles can be regarded as a key step for the progressing pathological status of lymphedema.

Interestingly, in the upper extremities, even when the edema was of short duration, the lumens of lymphatic trunks in the upper arm had already become occluded, but those at the elbow joint were still open. In the lower extremities, in spite of the short duration of edema, endothelial cells and smooth muscle cells of the mid-calf lymphatic trunk had already disappeared. However, the smooth muscle cells of the trunk at the ankle level remained. In some patients, despite edema of long duration, the smooth muscle cells in the elbow and mid-calf still existed. Two clinically important conclusions can be drawn from these findings. (1) Destruction of the smooth muscle and occlusion of the vessels start at the proximal extremities, and progress toward the distal portion.[7,15] This means that a lymphaticovenular anastomosis should be performed in the distal extremities, where the pump function of the lymphatics may remain intact. (2) The timing of the occlusion and degeneration of the smooth muscle cells may not directly correspond to the duration of edema.[5] This means that a lymphaticovenular anastomosis can be worth trying even in patients with a duration of lymphedema of more than 20 years.

Surgical treatment of lymphedema: lymphaticovenular anastomosis with supermicrosurgery

Conventional therapies and their limitations

Conventionally, lymphedema was treated by non-surgical physiotherapy or by surgical excision and drainage. Földi[14] introduced the methodology of combined physiotherapy, including massage, intermittent pneumatic compression, and skin care. Although considerably effective in some patients, this type of approach requires a long time and extreme patience. In addition, we experienced several patients in whom progression of lymphedema could not be prevented by physiotherapy, but was satisfactorily reduced by lymphaticovenular anastomosis. We thus have the impression that conservative physiotherapies only are insufficient for obtaining an acceptable result: they should be regarded as an additional choice combined with the surgical treatment. As for classical examples of surgical treatment, Thompson[1] reported large denuded skin flaps introduced into the muscle belly, thereby facilitating lymph fluid drainage into the muscle. Kinmonth et al.[21] reported island ileum transfer to the inguinal lymph region, aiming at drainage via mesenteric lymph systems. Although these strategies were accepted worldwide and were dramatically effective in some patients, their effectiveness is never consistent. Considering their massive surgical invasiveness, these approaches can not be strongly recommended for patients with varying clinical status.

One of the breakthroughs in lymphedema surgery may be the microsurgical lymphaticovenous anastomosis reported by O'Brien et al. in 1977.[22] In their method, the proximal end of the transsected lymph vessel is microsurgically anastomosed to the main trunk of a large cutaneous vein (such as the saphenous vein) in end-to-side fashion. This anastomosis is expected to promote lymphatic drainage through the venous system. In 1990, O'Brien et al.[23] summarized the long-term operative results, showing that the procedure was less effective in the lower extremities than in the upper extremities; i.e., only one-third of operated patients with leg edema showed excess volume reduction. In some patients, venous backflow into the lymphatics was observed, and this was, possibly, related to the ineffectiveness of this method. They also stated that microlymphatic surgery was not applicable to primary lymphedema, in which the lymphatics are usually hypoplastic.

We consider that there are two serious shortcomings in the lymphaticovenous anastomosis reported by O'Brien et al.[22,23] (1) In limbs with lymphedema, large lymphatic trunks (over 0.8 mm in diameter) have disappeared,[5] and precise anastomosis between the remaining very small lymphatics and venules of less than 0.8-mm diameter can not be established with the conventional microsurgical technique. (2) Cutaneous venous pressure can be higher than lymphatic pressure, as indicated by Baumeister and Siuda.[24] As stated above, the pumping function of the lymphatic vessels is insufficient in lymphedema, due to degeneration of the smooth muscle cells. Under lymphaticovenous anastomosis, the clotting activity of the blood can lead to a high rate of thrombosis at the anastomosis. To overcome these shortcomings, we developed the novel surgical approach of supermicrosurgical lymphaticovenular anastomosis.

Supermicrosurgical lymphaticovenular anastomosis: principles

Recent super- (or supra) microsurgical techniques have made possible the precise anastomosis of vessels of 0.3- to

0.8-mm caliber size.[2] This technique involves a vascular anastomosis of less than 0.8 mm, with six to eight watertight stitches, using 11-0 or 12-0 nylon, and a 50- to 30-μm needle. We also use a fine needle holder (specially produced by MEDICON, Tuttlingen, Germany; Koshima ultramicro series) under high magnification (20 to 30 XX). By application of this technique for lymphedema surgery, conventional lymphaticovenous anastomosis was refined into a novel lymphaticovenular anastomosis: the lymphatics and the subdermal venular system, instead of large cutaneous veins, can be anastomosed with a high patency rate.

There are several advantages in choosing subdermal venules, rather than large cutaneous veins, for anastomosis. First, only lymphatics with small diameters remain available in lymphedematous extremities, as noted above. The subdermal venules are more preferable for anastomosis with such small lymphatics from the standpoint of diameter matching, where even end-to-end anastomosis is possible. Second, intravenous pressure may be lower in the tiny subdermal venules than in the large cutaneous veins, because the large veins are more sensitive to external pressure than the small venules, as stated above. Thus, tiny venules may have lower pressure than the affected lymphatics, with minimum backflow from the veins, reducing the chance of thrombosis.[4] Third, anastomosis with the small venules can be performed in the more distal extremities. As stated before, smooth muscle degeneration in the damaged lymphatics starts in the proximal portion.[15] Thus, a better drainage effect is anticipated in the distal lymphaticovenular anastomosis.

Operative techniques

Operative treatment was applied to patients with severe edema who showed little improvement with conservative treatment of at least half a year's duration. When using general anesthesia, surgery is started after the application of a tourniquet to the affected extremities. To facilitate preparation for lymphatics, a small amount of indigocarmine is occasionally injected intradermally a few centimeters distal to the skin incision. After short incisions are made on the lateral and medial aspects, the lymphatics and subdermal venules (each 0.5 mm in diameter) are explored, using a loupe or operating microscope.

The affected lymphatics are often dilated and sclerotic, and are weakly stained or unstained. Dye staining is not always a good indicator for finding lymphatics. When no suitable lymphatics are found at one site, other incisions are made in other portions. For the lower extremities, it is preferable to make the incisions on thin adiposal portions such as the medial aspect of the pretibial areas of the proximal and middle lower leg and ankle, because the lymphatics can be easily found with a short incision. The lymphatics, which are normally translucent, are sometimes white with a thick fibrotic wall, and can be detected by reflecting microscopic light between adiposal particles in the superficial or deep adiposal layer, and sometimes in the subdermal layer. For the upper extremities, the lateral and medial aspects of the forearm and snuff box are suitable for exploring both channels, as there are many small branches of the main cutaneous vein through these aspects. The subdermal venules, which exist anywhere beneath the dermis, can usually be located adjacent to the lymphatics.

Regarding the selection of these channels, lymphatics with strong drainage function, which are easily detected after transecting them under an operating microscope, should be selected to establish powerful lymph-venous shunting. Small subdermal venules of 0.5-mm caliber are most suitable for joining with the lymphatics, because larger cutaneous veins may have higher intravascular pressure than that of the lymphatics. Larger dilatated lymphatics (those over 0.5 mm), which are sometimes detected in either the superficial or deep adiposal layer, are anastomosed to the small venular branches of the main cutaneous vein. The distal side of the venous system must be transected to establish anterograde lymphatic flow, because venous valves exist in the venules. After the tourniquet is deflated, end-to-end lymphaticovenular anastomoses are carried out without vascular clamps. During anastomosis, lymphatic fluid coming back to the ends of the transected lymphatics expands both ends of the transected lymphatics and venules. After completion of the anastomosis, expansion of the venule is detected by filling of the lymphatic flow. Postoperatively, the limb is kept elevated at night; a low-pressure bandage is applied and a vascular dilating drug (prostaglandin E1) is given. Five or six hours after the surgery, erythema of the affected whole limb sometimes occurs in patients with establishment of reverse flow from the venous to the lymphatic system. The erythema usually disappears spontaneously within 5 days. Later, elastic stockings are used for at least half a year.

Recent refinements of our surgical treatment

Since 1994, we have carried out supermicrosurgical lymphaticovenular anastomosis in more than 80 patients. In the earlier patients, we made five to ten anastomoses per one extremity, exclusively under general anesthesia, considering that the detection of tiny lymphatics in the edematous legs is difficult under local anesthesia.[3] However, we gradually realized, through our experiences, that the majority of patients do not need a large number of anastomoses, and that two or three (only one in some cases!) anastomoses are enough to reduce the volume.[6] This fact reflects the aforementioned notion that the progression of lymphedema is due to a pathophysiological vicious cycle: even one anastomosis can occasionally break the chain reaction of progression, especially if done in an earlier stage. Therefore, we tried to detect lymphatics and perform a few anastomoses under local anesthesia, and found this method to be useful as minimally invasive surgery.[6] With local anesthesia, a tourniquet is not used. The duration of the operation is usually within 2 h when performed by experienced surgeons creating a few anastomoses, and the operator can make contact with the patient during the operation. This is advantageous, especially if an additional incision is needed when

lymphatics appropriate for anastomosis are not found with the original incisions. Postoperatively, the patients can be promptly encouraged to walk freely, to reduce the incidence of pulmonary infarction.

We are now also carrying out another type of trial: supermicrosurgical transfer of the lymphatic vessels.[7] The aim of this approach is to replace a malfunctioning lymphatic vessel with a fresh, intact one taken from a healthy portion of the body. This idea was originally derived from the idea of the lympholymphatic anastomosis reported by Baumeister and Siuda[24] and Baumeister et al.,[25] with a lymphatic graft of more than 30 cm in length obtained from the leg. However, our technique is fundamentally different from that of the Baumeister group,[24,25] because we transfer lymphatic vessels as long as 10 cm that are included within fatty tissues as a composite graft, taken from the inguinal or the axillar region. The blood circulation of this composite graft is maintained by arterial and venous anastomoses, and only the proximal end of the transferred lymphatic vessel is supermicrosurgically anastomosed with venules. A drainage effect is expected through the whole composite graft. Although the number of patients and the follow-up period are not enough at present for reaching a definitive conclusion, we have an impression that this strategy may be another option for treating lymphedema, especially in difficult cases.

Clinical results

Lymphaticovenular anastomosis is considerably effective, especially in patients with an earlier stage of lymphedema, when smooth muscle functions are well preserved. In patients with lymphedema of long duration, lymphaticovenular anastomosis often results in an acceptable outcome, because smooth muscle functions of the distal portion are often intact even in patients with a long history, of over 10 years. Even if the smooth muscle functions are damaged, lymph fluid can flow through the anastomoses by external pressure when a bandage is applied, and remarkable reduction of edema of phlegmon can be achieved. In this sense, we consider that supermicrosurgical lymphaticovenular anastomosis can be indicated for most patients with any type of lymphedema. (For representative results in two of our patients, see Figs. 1 and 2.) Combined with conservative pressure therapy, lymphaticovenular anastomoses show good results in most patients with upper-extremity lesions, and in about half of those with lower-extremity lesions.

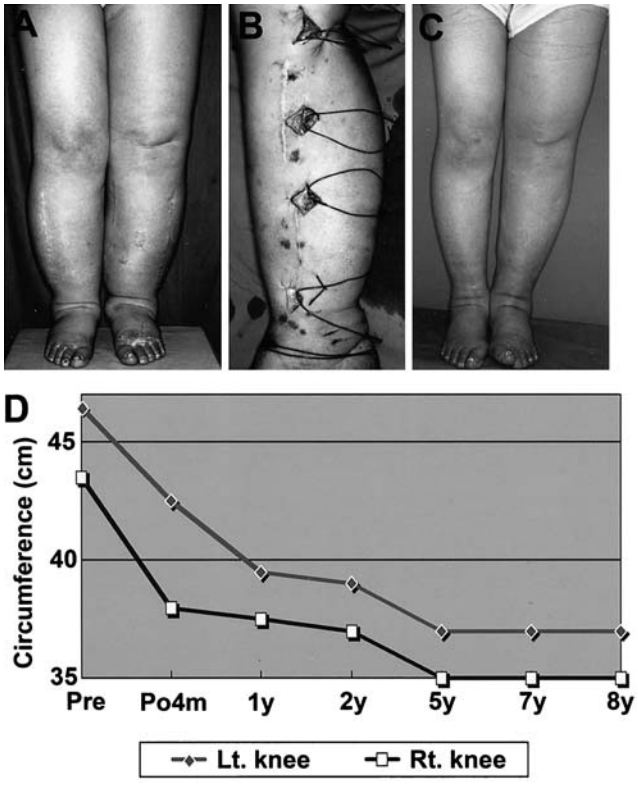

Fig. 1A–D. A 42-year-old woman with bilateral primary lymphedema. She had had lymphedema for 24 years, and Thompson's operation, performed 22 years before the present treatment, and other conservative therapies had been ineffective. **A** Preoperative (*pre*) view. **B** Five lymphaticovenular anastomoses in the right (*rt*) leg. **C** Nine years postoperatively. On the right side, a vascularized lymphatico-adipose composite graft was performed, with tissue taken from the groin region. **D** Postoperative (*po*) follow-up for 8 years. Edema was progressively reduced. Modified from Koshima et al. (2003),[5] with permission from the publisher

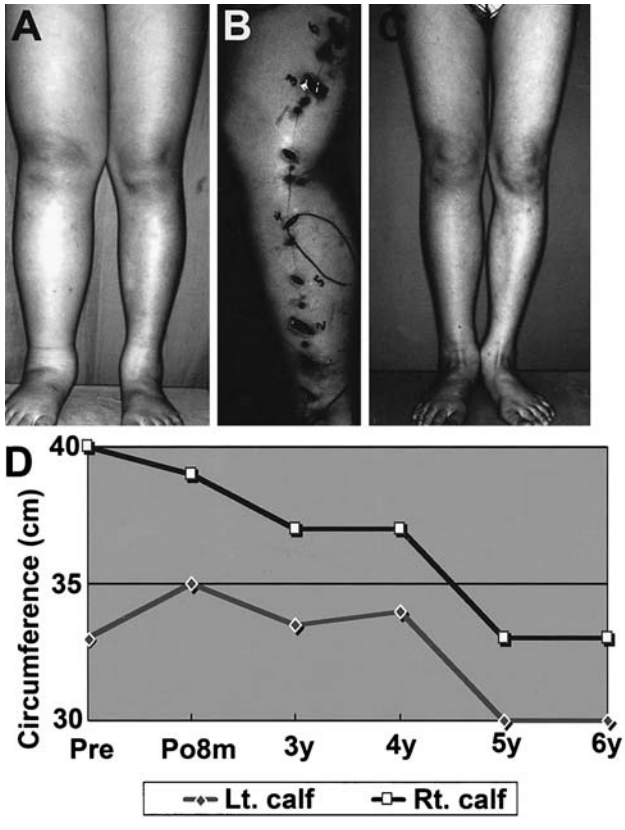

Fig. 2A–D. A 12-year-old girl with primary lymphedema in her right leg. Two years of conservative therapy was ineffective, **A** Preoperative view. **B** Five lymphaticovenular anastomoses were performed. **C** Eight years postoperatively. **D** Postoperative decrease in the circumference of the bilateral calves. Modified from Koshima et al. (2003),[5] with permission from the publisher

O'Brien et al.[23] mentioned that only one-third of their patients showed a decrease of more than 4 cm in circumference at the knee level with their conventional lymphaticovenous anastomoses. In our long-term results (published in 2003),[5] 62% of the patients showed a decrease of more than 4 cm (mean decrease, 4.7 cm in 4.6 ± 3.2 years' follow-up), possibly reflecting the superiority of our lymphaticovenular anastomosis. Notably, O'Brien et al.[23] mentioned failed anastomosis in 13% of their patients, due to sclerosis of lymphatics. We had no such cases, possibly because our anastomosis can be accomplished in the more distal portion where lymphatics are preserved intact. We also experienced good results in patients with primary lymphedema, which was regarded as a contraindication by O'Brien et al.[23]

From our experiences, lymph vessels had atrophied or disappeared in patients with poor results. Smooth muscle cells were diminished in number and size in the biopsy specimens from such patients.[15] In these patients, lymph flow was insufficient even if the anastomoses were accomplished, and additional anastomoses may be required later. Our recent approach of minimum invasive anastomosis performed under local anesthesia may be worthwhile to consider in this situation.

Future perspectives and concluding remarks

In this review, we described our sumpermicrosurgical approaches for lymphedema in detail. Although sumpermicrosurgical techniques are quite demanding, especially for beginners, this approach should be beneficial for patients, especially in terms of the minimum invasive surgery under local anesthesia. We believe that our strategy could become a new clinical standard in the quite near future.

A multidisciplinary approach may also become quite important in the treatment of lymphedema.[7] As stated above, prophylactic anastomosis in the normal leg is effective in patients with unilateral lymphedema, in whom the normal side has a risk of developing lymphedema later. This idea can be extrapolated to the idea of prophylactic anastomosis at the time of tumor resection and/or irradiation. Such prophylactic procedures will require close collaboration between plastic microsurgeons and clinical oncologists, such as breast surgeons, gynecologists, or urologists.

Finally, we mention the potential of molecular therapy for lymphedema. Various types of angiogenic therapies are now being investigated for limb ischemia, such as gene transfer of protein supplementation of VEGF or other angiogenic factors, or topical injections of various stem cells or vascular progenitor cells. These approaches could also be applicable for the treatment of lymphedema. Indeed, *VEGF-C* gene transfer has the effect of reducing lymphedema in the *Chy* mouse and other animal models.[9,26] If the progenitor cells for vascular and lymphatic vessels are really common, injections of such cells (transdifferentiation into lymphatic endothelium by Prox-1 may be necessary) may also be promising. Although only at an experimental stage at present, these lymphangiogenic approaches, combined with the supermicrosurgical technique, may open a new avenue toward the establishment of more thorough treatment of lymphedema.

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