Introduction

Human hand transplantation became a reality on 23 September 1998 when the first successful case was performed, with long-term transplant survival and promising functional recovery [1]. Twenty-four hand transplantations have been performed until the writing of this book (12 unilateral and 6 bilateral). However, there remains considerable debate over the ethical consequences of hand allotransplantation, and many questions must be answered before it becomes a routine procedure.

A functional primate model for hand transplantation does not exist, and the rat hind-limb allograft is the most widely used animal model for human hand transplantation research. Although hind-limb reimplantation on rats was first described in 1977 by Shapiro and Cerra [2], a precise description of entire operation has not been published. There are many research groups around the world performing limb transplantation on rats. Each group, however, uses its own surgical technique and type of anaesthesia. Surgery duration, animal survival rate and functional results vary, which makes it difficult to compare studies. However, we are convinced that optimal hind-limb transplantation with good functional outcome and safe, simple and easily controlled anaesthesia will make research more transparent.

In this section, we describe in detail a simple, quick and reliable surgical technique with excellent functional results. Using this technique, an experienced microsurgeon is able to perform hind-limb transplantation in a rat within 2 h. Further, we introduce safe, simple and easily controlled anaesthesia and an early postoperative management programme.

Hind-Limb Transplantation in Rats

Animals

The Animal Research Application Form for study must be approved by the institution's Animal Ethics Committee. All procedures using experimental animals must be carried out according to the Health and Medical Research Council's code of practice for the care and use of animals for scientific purposes. Ideally, animals are male rats between 10 and 16 weeks old and weighing between 250 and 400 g. Such animals are large enough for comfortable surgery and are young enough for long-term survive after transplantation, if necessary. For transplantation research, usually strong rejection is advisable. Therefore, in our research, we almost always use two inbred strains (Brown Norway and Lewis) because of their strong antigenic mismatch [3].

Anatomical Minimum

Rat hind-limb muscles can be divided into thigh, leg and foot muscles. Muscles of the thigh form
four groups: (1) anterior femoral muscles (thigh extensors), supplied by the posterior section of the femoral nerve; (2) medial muscles (adductors), supplied by the obturator nerve (n. obturatorius); (3) gluteal muscles, supplied by gluteal nerves; (4) posterior muscles (hamstrings), supplied by the sciatic nerve. Leg muscles are all supplied by branches of the sciatic nerve and are composed of three groups: (1) anterior (dorsal foot flexors), supplied by the peroneal nerve; (2) posterior muscles (plantar foot flexors), supplied by the tibial nerve; (3) the lateral group, supplied by the peroneal nerve (n. peroneus). Foot muscles are supplied by the ischiadic nerve (n. ischiadicus) and are divided into: (1) dorsal muscles (toe extensors), supplied by n. peroneus; (2) plantar muscles (flexors), supplied by the branches of the tibial nerve.

The saphenous nerve (n. saphenus) supplies sensory function to the medial surface of the lower leg and dorsal foot skin in the region of the first metatarsal. The skin of the dorsal distal third of the leg is innervated by the sural nerve. The lateral side of the lower leg is supplied by the peroneal nerve, and this nerve also supplies the dorsal area of the foot (except for the part supplied by the saphenous nerve). Terminal branches of the tibial nerve (n. tibialis) innervate the plantar area of the foot and toes. The femoral nerve is formed from 2–4 lumbar nerves and appears between the psoas minor muscle (m. psoas minor) and iliacus muscle (m. iliacus) and runs under the inguinal ligament together with external iliac vessels. Before entering the thigh, it divides into anterior and posterior sections. The anterior section innervates m. iliacus and pectineus muscle (m. pectineus) while the posterior division supplies quadriceps femoris muscle (m. quadriceps femoris). The third branch from the femoral nerve (n. femoralis) is the sensory saphenous nerve (n. saphenous) [4].

**Surgical Procedure**

Choosing the side of the animal on which to perform the surgery is the first step. For the right-handed surgeon, right hind-limb transplantation is more convenient. However, surgery time for left hind limb was found to take on average only 7 min longer. Moreover, from an ethical point of view, it is most desirable to use both hind limbs from one donor.

The surgical procedure begins on the donor. A circumferential skin incision of the donor hind limb is made at mid-thigh level. The inguinal fat flap with pedicle superficial epigastric artery (a. epigastrica superficialis) and superficial epigastric vein (v. epigastrica superficialis) and sensory nerve branch from n. saphenus is sharply dissected and after isolation of the pedicle flap is reflected distally. This flap can be retained in place, or after ligation of the pedicle, it can be removed (Fig. 1).

The saphenous nerve is prepared and transected proximally at the level of its branching from n. femoralis. The femoral artery and vein are then identified and skeletonised, and after ligation of all branches at mid-thigh level with 9-0 nylon, are ligated at the level of the inguinal ligament with a 6-0 nylon suture. The femoral artery is clamped distally from the ligature with a single microvascular clamp, transected closely distal to the ligature and cannulated using a 24-gauge intravenous (i.v.) polyurethane catheter. The artery clamp is removed, the femoral vein transected near the ligature and perfusion washout is performed with 4°C cold heparinised solution (1,500 UI heparin in 500 ml 10% dextan 40 i.v. infusion BP in 0.9% sodium chloride i.v. infusion). This perfusion is conducted by gravity at a height of 135 cm and continued until outflow from the femoral vein becomes clear. This procedure takes from 5 to 10 min, with volume range between 4 and 6 ml [5]. After perfusion, the catheter is gently removed, and the artery and vein are clamped with microsurgical single clamps. The thigh muscles are sharply cut approximately 1 cm distally from the level of the femoral nerve branching into the muscular branches and n. saphenus. This muscle dissection must be performed very carefully, as it is close to the branching of the femoral artery and vein into the saphenous and popliteal vessels. During muscle dissection, the sciatic nerve is solicitously protected when it emerges between thigh adductors and quadrate muscle (m. quadratus femoris) on the one side and