

Department of Plastic and Reconstructive Surgery

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General Summary

Research in the Department of Plastic and Reconstructive Surgery is focused on 4 basic areas: 1) the causes and treatment of craniofacial anomalies, 2) the causes and treatment of hand and foot anomalies, 3) the mechanism of wound healing and grafting of skin and bone, and 4) microsurgical transplantation. The faculty of our department consists of surgeons representing virtually all areas of plastic surgery and clinicians from related disciplines. This diversity provides the stimulating atmosphere necessary for productive research. The participation of plastic surgery residents and postresidency fellows in research studies provides them with important experience and expands their understanding of anatomical and physiological factors involved in these special areas of surgery.

Research Activities

Gene analysis and staged surgical procedures in patients with syndromic craniosynostosis

Apert syndrome, or acrocephalosyndactyly I, is an autosomal dominant disease caused by allelic mutations of fibroblast growth factor receptor 2 (FGFR2). Two regions (Ser 252 Trp and Pro 253 Arg) of the FGFR2 gene are believed to be responsible for syndromic craniosynostosis. Four monoclonal antibodies that respond only to peptides derived from mice with a mutation of Pro 253 Arg have been successfully prepared.

Gene transfer into limb buds by means of electroporation

Electroporation was used to transfer genes into the extremities of cultured mammalian embryos. Std-ddy mice were anesthetized with ether on embryonic day 11. These embryos, together with the placenta and embryonic membranes, were dissected from the surrounding decidua. The yolk sac, amnion, and chorioallantoic placenta were preserved in Hanks Balanced solution. An injection of 0.1 μ l of pEGFP-N1 vector was performed into the yolk sac. The extremity was grasped with forceps-type electrodes and electroporated with 3 pulses of 30 to 50 V for 50 milliseconds. After the amnion was removed, the embryo was placed in a bottle filled with mouse serum solution. Ninety-five percent O₂ and 5% NO₂ were supplied to the bottle via a tube 4 times a day. The embryo was cultured at 37°C and rotated at 30 revolutions per minute for 24 hours. The placenta was removed, and the embryo was fixed in 4% paraformaldehyde. Frozen sections were prepared with liquid nitrogen and observed with fluorescent microscopy. The specimens demonstrated that green fluorescent protein was present throughout the body after 50-V electroporation but was distributed in a more restricted area after 30-V

or 40-V electroporation. The embryos that were electroporated with 30 V revealed gene transfer localized to the epidermis and dermis.

Distraction osteogenesis

The use of distraction osteogenesis in reconstruction continues to expand and evolve. The effects of the various rates and frequencies of distraction have been studied, and rates of 1 to 2 mm per day have been found to be adequate for the craniofacial skeleton. The division of daily distractions into smaller, more frequent distractions accelerates bone formation. We have developed a device with a built-in motor which can produce continuous distraction. Results of experiments using newly developed devices are being investigated.

Morphologic study of bone conduction mechanisms

Experiments of artificial bone osteoconductivity concern the extremities more often than the cranium. Therefore, we performed an experimental study of osteoconductivity of β -tricalcium phosphate (β -TCP) in a cranial bone defect. Bone regeneration was evaluated in full-thickness circular defects (10 mm in diameter) created bilaterally in the parietal bones of adult female Japanese white rabbits. The animals were divided into 3 groups. In group A, a β -TCP disk (9.5 mm in diameter, 2.0 mm in thickness) was inserted into the bone defect. In group B, granules of β -TCP (approximately 0.1 g) were inserted, and in group C, nothing was inserted. The periosteum was repaired, and care was taken to avoid damaging the dura. Bone regeneration was assessed with macroscopy, roentgenometry, intensity, and histological examination. The results showed that β -TCP has good biocompatibility with cranial bone.

Tissue engineering

Flaps lined with mucosa are in great demand for nasal, oral, tracheal, and urogenital reconstruction. Fascia lined by mucosal tissue have already been developed as a new reconstructive material. Sublingual mucosa was obtained from Japanese white rabbits, and separated mucosal cells were subcultured twice for 4 weeks. The cells were transplanted to the fascia of the femoral muscles in the same rabbits. The fascial tissue was removed together with the muscular tissue 1 week after transplantation. Specimens were stained with hematoxylin and eosin and immunohistochemically stained for cytokeratin, a specific marker of mucosal cells. The growth of mucosal tissue was confirmed with histological examination. Fasciomucosal complex tissue developed. Fascia proved to be a useful scaffold that cross-links the transplanted mucosa and muscle.

Hemodynamic analysis of capillary blood vessels in patients with diabetes

The recent increase in the number of patients with diabetes has led to an increase in the prevalence of diabetic foot gangrene. Amputation either below or above the knee should be avoided as long as possible through the use of both conservative and surgical treatments. However, other than the ankle-arm pressure index and the cardio-ankle vascular index, few effective methods have been reported for predicting diabetic foot

lesions. We found that hemodynamic analysis with video microscopy of blood flow through capillary vessels in the eponychium of the toes in patients with diabetes can indicate the stage of microangiopathy and might be used to predict diabetic foot lesions. The effectiveness of prophylactic treatment using HT_{2A} receptor antagonists will be investigated with this new device.

Functional analysis of desert hedgehog in patients with macrodactyly

Mou reported in 2008 that the expression of the protein desert hedgehog in the hypertrophic parts of the affected nerve was significantly greater in patients with macrodactyly than in patients with polydactyly. The purpose of the study is to detect the expression of mRNA of desert hedgehog and immunohistochemical reaction of desert hedgehog and Patched2 in the fatty tissues of patients with macrodactyly. Immunohistochemical reactions for desert hedgehog were observed in the epidermis and adipocytes of patients with macrodactyly, whereas mRNA reactions were detected in the nervous systems of both patients with macrodactyly and patients with polydactyly. Whether the up-regulation of desert hedgehog is due to the disease itself or is a consequence of surgery is unclear, and further investigation is planned.

Publications

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