Stem-cell transplantation techniques bring new era in keratoplasty for corneal chemical burns

Charalambs S Siganos

ADVANCES in the understanding of corneal physiology, and improved surgical techniques have made it possible to restore vision to eyes that just a few years ago would have been considered lost causes, said Charalambs S Siganos MD, Dept of Ophthalmology, Heraklion University Hospital, University of Crete, Heraklion, Crete.

One particular indication that has greatly benefitted from these advances has been chemical injuries to the cornea. Centres from around the world now regularly report spectacular results with techniques involving autologous or allogeneic stem cell transplants in eyes that were damaged almost beyond recognition by thermal and chemical injuries, Dr Siganos told a Cornea Surgery Meeting of the ESCRs.

The structural alterations that can result from such injuries include limbal stem cell deficiency, conjunctivalisation of cornea, symblephara, lid deformities, and corneal and scleral melts. Prior to the development of stem cell techniques, keratoplasty often failed in such cases because of poor re-epithelialisation of the cornea, he said.

Surgical innovation preceded scientific understanding

Dr Siganos noted that the use of stem cell transplantation actually pre-dated the realisation that the limbus contains the stem cells that regenerate the corneal epithelium. As far back as 1994, Dr Jose Barraquer reported at the World Cornea Congress that transplantation of epithelial limbus and conjunctivo-corneal tissue from the fellow eye improved the outcome of keratoplasty procedures in eyes that had sustained thermal injuries (Holland E & Schwartz G. Cornea;23(5):425-31).

Over the following decades the keratoplasty techniques involving limbal and conjunctival tissues became more refined and in the late 1970s and early 1980s research confirmed that the limbus is the source of stem cells for the corneal epithelial regeneration. In 1989 Kenyon and Tseng described their limbal autograft transplantation procedure for ocular surface diseases, which was to be the prototype for the techniques that followed.

The techniques that have evolved include both autologous and allogeneic approaches. They range from the simple transplant of limbal tissue from the donor cornea to the affected eye to the ex-vivo expansion and transference of limbal stem cells from a donor or recipient’s healthy eye to the damaged eye.

In most of the published studies using the techniques, the use of amniotic membrane sutured over the transplant has been shown to improve the results by promoting re-epithelialisation and inhibiting fibrovascular proliferation, Dr Siganos said.

Limbal autografts may be indicated in patients in whom stem cell deficiency only affects one eye. The procedure involves the transplantation of a small amount of limbal tissue from the healthy to the unhealthy eye. Where the injury is superficial, the procedure is often enough to restore good vision. However, when the stroma is also badly damaged the procedure can be combined with a subsequent keratoplasty procedure.

As an example of the kind of results that can be achieved with limbal autografts, Dr Siganos described the case of a patient who had a third-degree chemical injury in one eye. Two months after the injury the patient had a non-healing epithelial defect, fibrovascular proliferation, conjunctivalisation of the cornea, symblepharon and limitation of ocular motility, with visual acuity of counting fingers.

Dr Siganos commenced the limbal autograft procedure by first performing a 360 degree peritomy and then a superficial keratectomy to remove all fibrovascular tissue on the cornea. He then removed the fibrovascular tissue under the conjunctiva, including the symblepharon.

The next step was to harvest the limbal tissue from the healthy eye. Using a 15 degree knife, he removed three clock hour hours of tissue from the upper limbus and two clock hour hours from the lower limbus and included an amount of conjunctival tissue, as required by the case.

He then sutured the amniotic membrane on the periphery of the sclera under the conjunctiva and on top of the graft at the 12 and 6 o’clock positions. At the end of surgery he removed the central part of the amniotic membrane so that there would be a direct epithelialisation from the limbal graft onto the corneal surface.

At three years’ follow-up, the patient’s cornea had maintained the restored epithelial integrity and the regression of the neovascularisation. In addition, ocular motility was fully recovered and there were no symblephara. Visual acuity was 0.8.

Another case he described was a 13-year-old with a third-degree chemical injury. Since this case also had a deep stromal opacity, Dr Siganos first performed a limbal autograft combined with AM transplantation, and then six months later performed a penetrating keratoplasty. The patient achieved a visual acuity of 0.6 until five years postoperatively, when unfortunately he lost the eye due to ocular trauma.

When both eyes have injuries causing limbal stem cell sufficiency, keratolimbal allografts can often produce good results, but they involve a higher risk of rejection than conventional keratoplasty and therefore require greater use of immunosuppressive therapy postoperatively, Dr Siganos noted.

This procedure involves first preparing the recipient cornea in a way similar to that used in the limbal autografts with peritomy and superficial keratectomy. The surgeon then trephines 8.0mm of the donor cornea and dissects the posterior part of the cornea to retain the limbal rim.

The limbal ring is then sutured into place and a patch of amniotic membrane is sutured on top of it to promote epithelialisation. When the patient also requires a penetrating keratoplasty, the corneal button and limbal ring are sutured into place with eight 10-0 nylon sutures each, and the amniotic patch is placed as usual.

“You can get good results with keratolimbal allografts, but in general the results of allografts are not as good as autografts because of rejection problems. Alternatively you can use pieces of limbal tissue only and not the whole ring and in my hands this is better as far as rejection is concerned,” Dr Siganos said.

One of the most exciting new techniques involves the ex-vivo expansion of epithelial stem cells harvested from a small limbal biopsy. The cells are cultured and then transferred to the recipient’s eye on a mesh with a 1.5x1.5mm square cut in the middle. The stem cells become attached to the amniotic membrane after a further 10-14 days. The expanded corneal epithelial cell populations are then placed on the centre of a patch of amniotic membrane, which has been applied to a circular stainless steel mesh with a 1.5x1.5mm square cut in the middle. The stem cells become attached to the amniotic tissue after a further 10-14 days in culture medium. Prior to transferring the cells on their amniotic membrane carrier to the corneal surface, the recipient is prepared as are those undergoing keratolimbal grafts. The amniotic membrane with a limbal sheet is then sutured to the patient’s eye. The amniotic membrane will dissolve in weeks leaving only the remaining cells.

“Eyes with chemical corneal injury are challenging cases, yet very rewarding once treated. Remarkable progress has been made over the last few years. Many eyes considered ‘lost’ in the past have been saved and the results of penetrating keratoplasty have been improved. However, the fight is still on against dryness, lid problems, and rejection,” Dr Siganos concluded.

csiganos@med.uoc.gr