A Nordic Research Team Addressing Global Challenges

How the work of researchers in Sweden can bring sight to those most in need around the world

The Editorial Board in *Oftalmolog* extends our congratulations to the authors of the article entitled "Bioengineered corneal tissue for minimally invasive vision restoration in advanced keratoconus in two clinical cohorts" published in *Nature Biotechnology* on August 11, 2022. The Editorial Team of *Nature Biotechnology* summarized this amazing achievement as follows: "This study is remarkably expansive, going from laboratory development of a collagen material for use as a synthetic cornea, to thorough safety testing in small and large animals, to clinical-grade manufacturing, to a pilot study in 20 patients with keratoconus in two countries. Encouraging clinical results after 2 years of follow-up suggest that the approach could benefit patients in low- to middle-income countries who do not have access to alternative therapies such as corneal transplantation."

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Bioengineered corneal tissue for minimally invasive vision restoration in advanced keratoconus in two clinical cohorts

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Visual impairment from corneal stromal disease affects millions worldwide. We describe a cell-free engineered corneal tissue, bioengineered porcine construct, double crosslinked (BPCDX) and a minimally invasive surgical method for its implantation. In a pilot feasibility study in India and Iran (clinicaltrials.gov no. NCT04653922), we implanted BPCDX in 20 advanced keratoconus subjects to reshape the native corneal stroma without removing existing tissue or using sutures. During 24 months of follow-up, no adverse event was observed. We document improvements in corneal thickness (mean increase of 209 ± 18 µm in India, 285 ± 99 µm in Iran), maximum keratometry (mean decrease of 13.9 ± 7.9 D in India and 11.2 ± 8.9 D in Iran) and visual acuity (to a mean contact-lens-corrected acuity of 20/26 in India and spectacle-corrected acuity of 20/58 in Iran). Fourteen of 14 initially blind subjects had a final mean best-corrected vision (spectacle or contact lens) of 20/36 and restored tolerance to contact lens wear. This work demonstrates restoration of vision using an approach that is potentially equally effective, safer, simpler and more broadly available than donor cornea transplantation.

Loss of corneal transparency and poor refractive function are among the leading causes of blindness globally[1–3]. Although corneal blindness can be treatable by transplantation, an estimated 12.7 million people await a donor cornea, with only one cornea available for every 70 needed[1]. With an incidence of over 1 million new cases of corneal blindness annually[3], the severe shortage of donor corneas presents an unequal burden of blindness heavily skewed towards low- and middle-income countries (LMICs) in Asia, Africa and the Middle East[4,5]. Over half of the world's population does not have access to corneal transplantation owing to a lack of infrastructure for tissue donation, harvesting, testing and eye banking in LMICs[4–7]. The access problem is complex, involving economic, cultural, technological, political and ethical barriers[4,8]. Additionally, infectious diseases and pandemics bring donor tissue procurement and use to a virtual standstill, necessitating further measures to ensure donor tissue safety[9–11].

For these reasons, intense research effort has focused on bioengineering tissue for corneal transplantation[12–16]. To date, however, no biotechnological advance has been able to address the burden of corneal blindness or improve access to transplantable corneal tissue. In many parts of the world including Europe and Australia, keratoconus—a corneal disease characterized by stromal thinning, weakening and scarring[17,18]—is the leading indication for corneal transplantation[19,20]. Keratoconus affects both men and women and all ethnic groups, with highest prevalence reported in China (0.9%, or 12.5 million)[21], India (2.3%, or 30 million)[22] and Iran (4% of the rural population, or 3.4 million)[23].

Keratoconus is progressive, but with a complex etiology that is not well understood. With proper screening and access to specialist care, keratoconus progression can be detected and halted in its early stages while vision is still good; however, if not addressed early and in LMICs where keratoconus is highly prevalent and access to healthcare is limited, the disease often progresses. In advanced stages, it requires transplantation to prevent blindness, using techniques such as penetrating keratoplasty (PK) or deep anterior lamellar keratoplasty (DALK)[20,24]. These techniques, however, are subject to the limited supply of donor corneas, risk of graft rejection, post-operative complications associated with sutures and wound healing, risk of corneal neovascularization and/or infection, high astigmatism after suture removal, need for long-term immunosuppression and necessity for long-term patient follow-up[25]. To partially address these issues, newer and less invasive techniques such as stromal lenticule addition keratoplasty[26] and Bowman layer transplantation[27] have been introduced. While promising and still developing, these techniques stabilize the condition but offer only marginal vision improvement[28,29], and rely on availability of donor corneas and tissue banking infrastructure and are thus inapplicable in many regions of the world.

To address these limitations, we bioengineered a cell-free implantable medical device as a substitute for human corneal stromal tissue. As a raw material we used natural type I collagen, the main protein in the human cornea[30]. For an abundant yet sustainable and cost-effective supply of collagen, we used medical-grade collagen sourced from porcine skin, a purified byproduct from the food industry already used in FDA-approved medical devices for

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> ***"Of the over 8000 papers published by* Nature Biotechnology, *this paper is in the top 10 of most widely disseminated papers"***
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Research briefing Check for updates

Accessible bioengineered corneal tissue to address a blinding disease globally

Porcine dermal collagen was chemically and photochemically bioengineered into an implantable tissue mimicking the human corneal extracellular matrix. The implant presents a simpler and safer method than donor cornea transplantation while delivering equivalent outcomes, and has restored vision to people with advanced keratoconus in resource-limited regions, where the burden of blindness is highest.

This is a summary of:
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The problem

Blindness due to disease-related loss of transparency or function of the cornea (the eye's outer layer) affects an estimated 12.7 million people globally, whose only option is awaiting a cornea transplant from a donor[1]. Keratoconus, a disease in which degradation of the corneal extracellular matrix induces tissue thinning and weakening, is a leading indication for corneal transplantation globally[2], with a prevalence that is highest in low- to middle-income countries (LMICs). However, owing to a severe shortage of donor tissue and poor access to tissue banking infrastructure, only 1 in 70 patients who need corneal transplantation receive it, with this inequity heavily skewed against LMICs[3]. Nevertheless, no adequate and widely accessible alternatives to donor tissue transplantation for late-stage blinding disease have been developed for over 110 years, since the first corneal transplant was performed[2]; as no technology has been able to match the visual outcomes attainable using donor tissue.

The solution

A potential solution to this problem is a bioengineered cornea. As the main protein of the human corneal extracellular matrix is collagen, we identified a sustainable source of purified type I porcine collagen as a by-product of the food industry. The collagen is extracted and purified from the overabundant leftover skin from food-grade pigs that are processed for meat products. We engineered this collagen into an implantable tissue by chemically and photochemically creating collagen–collagen bonds (crosslinks) to strengthen the implant while maintaining high transparency. For human implantation, the implant had to fulfill a range of stringent manufacturing, biocompatibility and toxicity standards, while also requiring robust sterilization, packaging and long-term storage to facilitate access and distribution. At the same time, we developed a minimally invasive technique to insert the implant within the existing cornea rather than replacing it. This surgical implantation technique was validated in a pig model using simple tools and in a single procedure, to avoid the need for suturing or long-term immunosuppression. This approach enabled us to obtain approvals to treat the first patients with keratoconus in India and Iran.

Crucially, after 24 months of storage, the sterilized and packaged bioengineered implant maintained its mechanical, optical and chemical properties, as assessed by stringent testing in vitro and in vivo. Packaged implants and technical details of surgical implantation were transferred to centers in India and Iran, where surgeons treated 20 individuals with advanced keratoconus on the waiting list for a corneal transplant, 14 of whom were legally blind. Surgeons found the implants simple to insert, and operations were completed without complications, with only a short 8-week period of postoperative immunosuppressant medication. Postoperatively, corneas were transparent and stable, while surprisingly, the corneal thickness and curvature reverted to a pre-disease state (Fig. 1). After 2 years of follow-up without adverse events, no patients were blind anymore, and vision in the entire group had improved to an equivalent degree as with standard transplantation with donor tissue. Three individuals in India who initially were blind achieved 20/20 vision.

The implications

This work demonstrates the possibility of manufacturing, storing (for up to 2 years before use) and distributing our implantable bioengineered tissue to regions with a high burden of corneal blindness. The proposed surgery is less invasive than traditional transplantation and could be implemented as a safer alternative, even with human donor tissue. Given the global shortage of donated corneas; the barriers to procuring, pathogen testing and storing donor tissue in certified tissue banks (where it needs to be used or discarded within 2 weeks); and the required long-term (at least 1 year) immunosuppression to avoid foreign tissue rejection, the proposed bioengineered alternative would be attractive, especially in resource-limited areas. Although we did not select patients for best visual outcome and focused only on keratoconus, the methods could be refined to optimize vision and to treat other types of visual impairment. The implant is cell-free, but, as we observed during this study and in our previous studies with similar materials, it can support the growth of corneal epithelial, endothelial[1] or stem cells, which should be evaluated as advanced therapy medicinal products in the future. We await longer-term clinical results and aim to conduct larger controlled trials for definitive evaluation of safety and efficacy before wider deployment of this technology.

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