Abstract

Amniotic membranes (AMs) and corneas are critical materials in ocular surface reconstruction. AM has specific structures (e.g., basement and two types of cells with stemness characteristics: amniotic epithelial cells and amniotic mesenchymal cells), which contribute to its attractive physical and biological properties that make it fundamental to clinical application. The corneal endothelial cell is a vital part of the cornea, which can influence postoperative vision directly. However, widespread use of fresh AM and cornea has been limited due to their short use span and safety concerns. To overcome these concerns, different preservation methods have been introduced. Cryopreservation is distinguished from many preservation methods for its attractive advantages of prolonged use span, optimally retained tissue structure, and minimized infection risk. This review will focus on recent advances of cryopreserved AM and cornea, including different cryopreservation methods and their indications in ophthalmology.

Keywords: amnion, cornea, cryopreservation, indication, ophthalmology

1. Cryopreservation of amniotic membranes

1.1 Introduction

Ever since Davis [1] first used human amniotic membrane (Figure 1) (AM) for skin transplantation, people have been exploring this remarkable biomaterial. AM is located in the innermost layer of the fetal membranes [2]. It is 0.02–0.05 mm thick, lightweight, elastic, almost transparent, and avascular membrane classically composed of three layers: the epithelium, the basement membrane and the stroma layer [2]. There are two types of cells with stemness properties in AM: amniotic epithelial cells (AECs) and amniotic mesenchymal cells (AMSCs) [3], which are responsible for its unique biological properties including anti-inflammatory, anti-scarring, anti-microbial, angio-modulating, immunomodulatory, and anti-cancer effects [4–10]. Due to these properties, AM has become an ideal material for ocular reconstruction including the treatment of persistent epithelial defects and non-healing corneal ulcers, corneal perforations and descemetoceles, bullous keratopathy, as well as corneal disorders with associated limbal stem cell deficiency, pterygium, conjunctival reconstruction, corneoscleral melts and perforations, and glaucoma surgeries. However, its use span is short and many viruses (such as HIV-1/2, hepatitis B, hepatitis C, human T-cell lymphotropic virus, syphilis, and cytomegalovirus)
can be in their “window period” and escape detection, further limiting the use of fresh AM. To overcome these concerns, different preservation methods have emerged, such as freezing, lyophilization, and cryopreservation. However, most methods result in the destruction of endogenous cells and cause varying degrees of extracellular matrix (ECM) damage, which can affect the functionality of AM and its clinical benefits for wound treatment [11, 12]. Cryopreservation was first introduced by Lee and Tseng and has been proven to achieve high success rate in AM transplantation, which has been distinguished from many methods for its attractive advantages of prolonging use span, optimally retaining tissue structure, and minimizing infection risk [13, 14].

In this part, we classify the cryopreservation methods applied to amnion by commonly used cryoprotectant and analyze the influence of cryopreservation on AM combined with specific clinical trials.

1.2 General cryopreservation techniques

The AM is normally washed using balanced saline solution containing antibiotics such as streptomycin, penicillin, neomycin, and amphotericin prior to storage. Pieces of AM resting on a carrier are placed in a vial containing cryoprotectant solution at a controlled cooling rate. Storage temperatures of $-80^\circ$C are often utilized, with the maximum storage times ranging between 1 and 2 years [1, 11, 12].

The main disadvantage of cryopreservation is the requirement of a deep-freezing facility, which is expensive, cumbersome, and frequently unavailable, especially in underdeveloped countries. In addition, maintaining stable storage temperatures during transportation is also relatively difficult.

1.3 Cryopreservation methods on AM

1.3.1 Glycerol-cryopreservation

Glycerol storage was first introduced in the Netherlands in 1984 to preserve donor skin for transplantation [13]. Positive results over subsequent decades have led to its clinical acceptance, including in the preservation of AM. Glycerol has led to higher cell viability and higher bFGF secretion for up to three months of AM storage [14]. After strict preservation and sterilization processes, pieces of AM resting on a carrier are placed in a vial containing storage solution. Tseng’s laboratory introduced a methodology of glycerol (86%) in Dulbecco’s Modified Eagle Medium at a ratio of 1:1 [15, 16]. The most common
A cryopreservation protocol reported in the literature involves the use of 50% glycerol and storage at −80°C [17–21]. Undiluted and 98% glycerol have also been reported to be clinically effective [15].

In 2011, Thomasen et al. [21] showed that long-term storage of 50% glycerol cryopreserved AM for durations up to 24 months at −80°C did not significantly impair the histology of AM. Wagner et al. [14] used 85% glycerol for cryopreserved AM, and their histological examinations had no significant alterations following cryopreservation, either for straight cryopreservation or with glycerol. They also demonstrated that neither tensile strength nor Young’s modulus was significantly influenced by the storage method. In addition, they also detected a significant increase in tensile strength over storage time, independent of the storage method.

Some groups have found that storage of AM in 50% glycerol at −80°C decellularizes the AM and results in low viability [17–20]. Interestingly, the results from Wagner et al. [14] research showed that epithelial cells were not significantly reduced during freezing in comparison to stromal cells, possibly indicating a higher sensitivity of stromal AM cells to freezing damage than epithelial cells (Figure 2). Through repeated measurement analysis, storage time showed a significant effect on cell viability. Prabhasawat et al. [22, 23] reported that the use of a highly concentrated glycerol solution abolishes AM cell viability. The possible toxic effect of glycerol is responsible for that.

To summarize, glycerol-cryopreserved AM retains the histological characteristics of fresh AM independent of an increase in glycerol concentration. Tensile strength and elasticity can also be better preserved, with tensile strength increasing with storage time. However, the cell viability of cryopreserved AM was significantly affected by storage time and glycerol concentration. In particular, the stromal cells were more sensitive. A previous study [24] showed that this method had little effect on the growth factors of AM. More research is needed to confirm the effect of glycerol cryopreservation on AMs.

![Figure 2. Pathways of cellular injury during freezing.](image-url)
1.3.2 DMSO-cryopreservation

DMSO has been used as an alternative for AM in glycerol-cryopreservation. An increasing concentration of DMSO is used instead of washing the AM with an antibiotic-saline solution after placenta collection [12]. Azuara-Blanco et al. [25] used 4%, 8%, and 10% DMSO, while Kubo et al. [26] used 0.5 M, 0.1 M, and 0.15 M DMSO for washing. AMs can be stored in 10% or 0.15 M DMSO at −80°C for several months without significant damage. In general, solutions containing DMSO are used less often for AM cryopreservation compared to glycerol, due to high toxicity [12]. However, AM storage solutions containing DMSO have been studied a lot regarding its ability to increase cell viability in AM under experimental conditions [2].

A cryopreservation method with DMSO from Duan-Arnold’s group [24] showed a retained cell viability of over 80%. Cryopreserved AM tested after three months of storage showed no changes in the tissue architecture and collagen IV, which exists in the basement membrane, compared with fresh AM. However, in 2015, Yazdanpanah et al. [8] showed that the viability of epithelial cells in fresh AMs was estimated at 97% after staining with trypan blue, decreasing to about 50% in DMSO cryopreserved tissues after six months. They evaluated the effects of cryopreservation on AM angiogenesis modulation activity compared to fresh tissue in an animal model, showing that cryopreserved AM has the same effect on angiogenesis as fresh AM. The epithelial surface of cryopreserved AM inhibited angiogenesis, and the mesenchymal surface augmented vessel sprouting and length. In 2013, Tehrani et al. [27] used 10% DMSO as a cryoprotectant to evaluate the antibacterial properties of AM after preservation in vitro. The results of this study showed that the antibacterial property of AM was maintained after cryopreservation, but was dependent on bacterial genus and strain.

To sum up, the literature we collected on DMSO-cryopreserved AM showed no significant differences in tissue integrity and biological properties (antibacterial and angiogenesis modulation) compared with fresh AM. However, although many research groups use DMSO as a cryoprotectant, the data related to cell viability vary. These conflicting results can be attributed to several factors, including differing cryopreservation procedures and storage times.

1.4 Controversy on cryopreserved AM

1.4.1 Variable cell viabilities

In 2000, Kruse et al. [18] believed that devitalized AM exhibited therapeutic effects, and their data showed that the preservation of viable cells in AM provided no additional benefits. This conclusion led to the development of cryopreservation methods including AM devitalization steps. One of them, known as the CRYOTEK® process, includes a freezing step before cryopreservation, resulting in devitalized tissue [28]. However, Yan et al. [29] demonstrated that the combination of exogenous cells and acellular AM resulted in faster wound closure compared with acellular AM alone. Duan-Arnold et al. [24] demonstrated that endogenous viable cells allow cryopreserved AM with higher angiogenic, anti-inflammatory, antioxidant, fibroblast, and keratinocyte chemo-attractive activities when compared with AM in devitalization. Before 2001, most studies reported that cell viability of 50% or less at cryopreserved post-thaw with cells failing to survive after 18 months of storage at −80°C [26, 30]. Since then, scientists have been attempting to improve the cryopreservation method, for improved cell viability retention. For example, the cryopreservation protocol invented by the group of Duan Arnold et al. [24]
can maintain 70% or greater cell viability after 24 months of storage at −80°C. AM storage solutions containing dimethyl sulfoxide (DMSO) have been studied, mostly under experimental conditions, and shown the ability to increase AM cell viability [2]. Although the survival of amniotic cells is related to storage time, different cryopreservation steps can also affect cell viability, thus exerting different clinical effects.

1.4.2 Storage temperature

The best storage temperature (−196°C or −80°C) is also a controversial issue for cryopreservation. AMs stored at −196°C have showed morphology similar to fresh AM in both preservation media, and AM stored at −80°C showed disruption of the stromal matrix [2, 31]. However, −80°C is still widely used by international scientists.

To sum up, cryopreservation protocols are not standardized. Preparation and sterilization before cryopreservation, as well as the selection of cryoprotectant during cryopreservation, will lead to high variability in cell viability [2, 32]. Different storage temperature and storage time also affects the structure and function of amniotic membrane. It is important to establish adaptable protocols for the clinical banking of AM that include verification of graft quality and viability before its release for transplantation, whether in the trial or clinical stage.

1.5 Commercially available cryopreserved AM

1.5.1 PROKERA®Slim (PKS)/PROKERA®(PK)

PROKERA®Slim (PKS) (Bio-Tissue, Inc., Miami, FL, USA) is a Class II medical device approved by the Food and Drug Administration in 2003 to be used as a temporary AM patch for delivering the biological actions of AM to a corneal surface without using sutures. It contains a piece of cryopreserved AM clipped into a concave poly-carbonate dual-ring system, like a symblepharon ring, that conforms to the corneal and limbal surface like a contact lens. The ring system has an inner diameter of 15 or 16 mm.

It has become the most common commercially available cryopreserved AM product in ophthalmology and is applied to various ocular surface and orbital disorders. It is a safe and effective method that makes AM transplantation sutureless and adhesiveless, contributing to healing and reconstruction of the ocular surface and orbit with minimal side effects [33]. However, PROKERA is not recommended for eyes with functioning blebs or glaucoma drainage implants because of the oppositional positioning of the retaining ring [34].

2. Cryopreservation of cornea

2.1 Introduction

Corneal disease is one of the world’s leading causes of blindness. Corneal scarring and haze due to various factors can affect vision, making corneal transplantation an important means of treatment for corneal diseases [35–37]. Advances in corneal preservation techniques have improved the survival rate of corneal grafts [38] and have largely contributed to the development of modern corneal transplant surgery [39]. With the flourishing of corneal preservation technology, breakthroughs have been made in preservation times and corneal activity. Nevertheless,
cryopreservation is the only current method that can virtually preserve tissue structure for a long time.

Meanwhile, the development of modern eye banks have been accompanied by the advancement of corneal preservation technology. The establishment of an eye bank provides favorable conditions for corneal transplantation [40, 41].

2.2 Corneal transplantation and preservation

The idea of replacing the turbid cornea with transparent tissue was first proposed by Pellier de Quengsy in 1789 [42, 43]. In 1824, Reisinger exploited animal corneas in surgery [44], which was named keratoplasty. Later in the nineteenth century, a large number of animal experiments helped doctors realize that inter-species transplantation was a necessary condition to avoid corneal opacity after transplantation [45–47]. In view of this, researchers began to experiment with human corneal transplantation. Early corneal transplantation relied on living donor tissues due to fears relating to transplanting dead tissue. The first successful full-thickness corneal transplantation (including all corneal layers) was completed in 1905 [48]. It was not until the 1930s that the cornea of the deceased donor was used and the entire eye was kept in a glass jug (wet room) for several days [49].

In 1912, Magitot reported that excised corneal grafts could be preserved in red blood cells at 5°C for eight days [50–52]. The grafts were successfully used for corneal lamellar transplantation [53]. At first, the freshness of the cornea was considered key to corneal transplantation [54, 55]. However, Ukrainian doctor Filatov systematically reported the application of corpus corneal tissue to clinical practice [56], which possessed an inter-generational meaning. It opened a new era of corneal preservation and transplantation [57–59]. These developments led to the establishment of the world's first ophthalmology bank in New York in the 1940s. The preservation technique of the original eye bank was very simple [60, 61]: eyeballs were kept in a small glass bottle in a humid and cool environment [62]. Immediate removal of the eyeball after donor death was the only way to ensure the quality of the corneal grafts [63–65]. Eye banks were established in major cities, such as London, to guarantee that eyeballs were promptly forwarded [66, 67]. In the early 1950s, the activity of CECs was first considered as an important factor affecting transplantation [68–70]. The emphasis on preservation techniques was transferred to maintain the activity and integrity of CECs [71, 72]. Since then, corneal preservation techniques have been increasingly successful, resulting in approximately 40,000 corneal transplants per year in the United States, 20,000 per year in Europe, and thousands per year in other countries, such as India.

2.3 Corneal preservation methods

Corneal preservation is divided into two categories according to the survival of CECs: inactive and active preservation [73–75]. The former method includes dry preservation and cryopreservation [76–78] and operates under the principle of removing corneal moisture while inhibiting enzyme activity and autolysis in cells for long-term preservation [79, 80]. Common preservatives are glycerin, molecular sieves, and silica gel [81–83], which can preserve intact lamellar collagen structure [84]. Active preservation comprises short-term (hours to two days), medium-long term (7 to 30 days) and long-term (months to years) preservation. In terms of storage conditions, it utilizes normal (34–37°C), low (usually 4°C) and deep low (subzero) temperatures [85–88].

Short-term corneal storage mainly refers to the preservation of wet rooms, the simplest and most convenient of all corneal storage technologies. For this reason,
it is still the basic technology for preserving cornea in the eye banks of developing countries. As for medium-term corneal preservation, corneal preservation solution is stored at 4°C for 4 to 14 days [89].

The prolongation of corneal preservation allows more preparation for patients and flexible adjusting of operation times, while also satisfying blood test and corneal transportation times. With the improvement of preservation techniques, the composition of the corneal preservation solution has been constantly changing. A certain concentration of chondroitin sulfate is added to modify M-K solution, which can alleviate the swelling state during preservation. Optisol corneal medium preservation solution was proposed by Lindstrom and has become the most common preservation solution in US eye banks, which is mainly a mixture of K-liquid and Dexsol solution [90]. Long-term corneal preservation refers to organ culture storage and cryopreservation. Organ culture is to simulate the presence of a normal human cornea environment with medium at 30–37°C [91].

At present, there are several corneal preservation methods applied in global eye banks, but none of those is perfect. Each preservation method has its own advantages and disadvantages, which differ from the preservation temperature, the composition of the preservation solution, and the penetrant preventing matrix edema.

2.4 Cryopreservation

After donor death, the sudden stop of the aqueous humor causes nutrient and oxygen shortages, leading to final depletion at room temperature, which can, in turn, lead to autolysis of the corneal cells and initial damage to the cornea [92]. During the period from donor death to corneal removal and storage, the donor’s corpse is exposed to room temperatures, necessitating minimal time delays to ensure that the initial donor cornea is healthy and intact along with functional endothelial cells.

The acceptable short storage time, as well as organ damage, poses a logistical challenge to organ storage and ultimately affects grafts and patient survival. Prolonged storage times can cause many transplantable organs, further exacerbating the growing imbalance between organ supply and demand. Organ cryopreservation is used to preserve long-lived cells and tissues. Theoretically, the storage of biological materials, including cells, tissues, and organs for transplantation at a low temperature (i.e., in liquid nitrogen at −160°C) is uncertain [93, 94]. Such a technique would have the potential to alter the way in which organs are recovered, distributed, and utilized for transplantation. However, ice is the biggest enemy in the cryopreservation of organs and tissues. Ice crystals, especially intracellular ice, can cause significant cellular damage and destroy the complex macroscopic tissues of intact organs. In this field, current developments are used to avoid the formation of ice, or mitigate it, during cryogenic storage. Any successful organ cryopreservation strategy requires a delicate balance between the relative needs of cryopreservation and toxicity in these situations.

2.5 Corneal cryopreservation technology

In 1954, Eastcott first adopted a cryopreserved human cornea for transplantation successfully [95, 96], pretreating the keratin tissue in glycerol before freezing it in a mixture of alcohol and carbon dioxide for cryopreservation of the full-thickness cornea [97]. This method generally removes the cornea under the protection of a cryogen to −80°C, and stores it in liquid nitrogen at −196°C. Therefore, the CECs are in a dormant state. The state can completely inhibit the metabolism
of cells, eliminate the toxic effects caused by the accumulation of metabolites, and avoid the need to change the liquid during organ culture. In addition, it also restrains microorganisms during cryopreservation, protecting the cornea from microbial invasion.

The components currently contained in corneal cryoprotectants include DMSO, propylene glycol, ChS, and sucrose. DMSO is a relatively stable protective agent to maintain the integrity of corneal cells, while sucrose molecules act as buffers in corneal protection, and ChS improves CEC activity in cryopreservation [98].

DMSO began to be treated as a tissue preservative to preserve cultured rabbit CECs by Smith [99]. Shortly thereafter, Mueller injected a preservation solution containing DMSO into the anterior chamber of an eyeball, placing the eyeball in a preservation solution containing glycerol. The cornea was removed before surgery for full-thickness transplantation [100, 101]. In 1965, Capella [102] used DMSO as an antifreeze to improve a cryopreservation method, ensuring corneal graft activity. According to another report [103], the clinical application of cryopreservation techniques has little differences in techniques. The corneal tissue must be preserved eight hours after death. By increasing the level of DMSO, it eventually reaches a concentration of 7.5%. The classic four-step cooling is to initiate a cooling rate at 1.5~2°C/min, drop the temperature to $-30^\circ$C, change to 5–7°C/min, and ultimately maintain $-80^\circ$C [104, 105].

It is still essential to further explore the rate of cooling to keep CEC activity and reduce cell loss [106, 107]. Temperature-controlled thawing before transplantation is a key step in protecting the corneal endothelium. At present, the prevalent view is that rapid rewarming could decrease the contact of cells with high concentrations of electrolytes and reduce cell damage [108]. The thawing process of the cryopreserved cornea must be strictly controlled, as the solute containing DMSO has endotoxicity once the temperature exceeds 37°C [109]. Cryopreservation would impair the morphology and function of the corneal endothelium. During the thawing process, an ascending solute concentration, the formation of crystals, changes in pH, and osmotic pressure will reduce the survival rate of CECs [110]. Glycerol, polyvinylpyrrolidone, and DMSO can all be used as cryopreservatives, but DMSO is currently the most widely used [111, 112].

2.6 Effect of corneal cryopreservation

The ultra-low temperature preservation method overcomes the drawbacks of most other corneal preservation methods, significantly prolonging corneal preservation time, reducing pollution, and avoiding the toxic effects of its own metabolic substances. Electron microscopy can observe changes in the subcellular morphology of CECs caused by cryopreservation, some of which are considered irreversible [113]. Studies have shown that, after cryopreservation, the barrier function of endothelial cells is impaired. Compared with wet room preservation and MK solution preservation, cryopreserved corneal grafts have been completely transparent for a long time after surgery. For one-year cryopreservation, 55% of endothelial cells were deactivated, while the rate of CECs preserved by MK solution was only 21–22% [114]. There are barely significant structural differences in microbiological, histological, and ultrastructural features when comparing long-term cryopreservation of tissue (>7 years) and short-term cryopreserved cat corneal sclera (<1 year) [115]. As such, tissues cryopreserved for up to 10 years could be used for tectonic support without structural or microbial barriers.

Under suitable conditions, no crystal solidification occurs during the freezing process, called vitrification [116]. Vitrification requires a high concentration of
cryoprotectant, yet theoretically, tissue could be stored in a very low temperature environment without forming intracellular or extracellular crystals, and corneal endothelium damage could be avoided significantly [117]. Glycerol, 1,2-propanetriol, and 2,3-butandiol are all considered as eligible cryopreservation agents for corneal vitrification [118, 119].

Studies have found that an effective concentration of a single cryopreservative is toxic to CECs, yet the mixture of preservatives or the addition of preservatives at low temperatures seems to reduce toxicity [120]. As a means of corneal preservation, further study is warranted to investigate whether vitrification would achieve good results. In 1981, Sperling used corneal grafts in a corneal preservation solution at the early stage and carried out a cryopreservation operation later. After rewarmed, the cornea was transferred to a preservation solution, identifying corneal activity. The following studies indicated that the corneal grafts maintained transparency 71% of the time after 1 year and 58% of the time after 12 years [121].

In our previous study, we performed lamellar keratoplasty combined with keratopigmentation in 22 corneal leukemia eyes using glycerol-cryopreserved corneal tissues, and no graft-rejection occurred during the 3 years of follow-up. Moreover, the outcome of a low graft rejection rate in glycerol-cryopreserved corneal tissues was also confirmed by our preceding study in treating Terrien marginal degeneration. In the subsequent study, for patients with refractive herpes simplex keratitis, 3 eyes of 27 eyes (11.1%) suffered allograft stromal rejection, all eyes reversed after prompt medication. Meanwhile, only 2 eyes (7.41%) exhibited refractive herpes simplex keratitis recurrence and the main site was located at the margin of the graft and the recipient bed. This result is consistent with the theory that grafts survive better when compared with reports clarifying that up to 33% of patients have suffered recurrence using fresh grafts. The recurrence rate in fresh grafts may be partially related to the long-term usage of topical steroid eye drops; however, it may be much more closely correlated with fewer keratocytes in the cryopreserved donor tissue to reactivate immune-inflammatory responses [122–124]. Based on the above information, glycerol-cryopreserved corneal tissues can be effectively and biosafely used with a low rejection and recurrence rate in corneal transplantation, especially in developing countries where good donor corneas are difficult to get.

3. Conclusion

The cryopreservation method can preserve the activity of the AM and cornea for extended periods up to several years, solving the problem of preservation time and activity deterioration. However, equipment complications, expensive technical support, and transport difficulties have become impediments to widespread use. The functional status of AM, endothelial cells, and corneal transparency have been of vital importance in the development of cryopreservation. As researchers become more aware of the function and properties of CECs, attempts to find a more conducive method and media for the preservation of AMs and corneas will continue.

Conflict of interest

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References

[1] Dua HS, Gomes JA, King AJ, Maharajan VS. The amniotic membrane in ophthalmology. Survey of Ophthalmology. 2004;49(1):51-77. DOI: 10.1016/j.survophthal.2003.10.004

[2] Hettiarachchi D, Dissanayake VHW, Goonasekera HWW. Optimizing amniotic membrane tissue banking protocols for ophthalmic use. Cell and Tissue Banking. 2016;17(3):387-397. DOI: 10.1007/s10561-016-9568-3

[3] Koike C, Zhou KX, Takeda Y, Fathy M, Okabe M, Yoshida T, et al. Characterization of amniotic stem cells. Cellular Reprogramming. 2014;16(4):298-305. DOI: 10.1089/cell.2013.0090

[4] Shimmura S, Shimazaki J, Ohashi Y, Tsubota K. Antiinflammatory effects of amniotic membrane transplantation in ocular surface disorders. Cornea. 2001;20(4):408-413. DOI: 10.1097/00003226-200105000-00015

[5] Sorensen OE, Cowland JB, Theilgaard-Monch K, Liu LD, Ganz T, Borregaard N. Wound healing and expression of antimicrobial peptides/polypeptides in human keratinocytes, a consequence of common growth factors. Journal of Immunology. 2003;170(11):5583-5589. DOI: 10.4049/jimmunol.170.11.5583

[6] Niknejad H, Paeini-Vayghan G, Tehrani FA, Khayat-Khoei M, Peirovi H. Side dependent effects of the human amnion on angiogenesis. Placenta. 2013;34(4):340-345. DOI: 10.1016/j.placenta.2013.02.001

[7] Hao YX, Ma DHK, Hwang DG, Kim WS, Zhang F. Identification of antiangiogenic and antiinflammatory proteins in human amniotic membrane. Cornea. 2000;19(3):348-352. DOI: 10.1097/00003226-200005000-00018

[8] Yazdanpanah G, Paeini-Vayghan G, Asadi S, Niknejad H. The effects of cryopreservation on angiogenesis modulation activity of human amniotic membrane. Cryobiology. 2015;71(3):413-418. DOI: 10.1016/j.cryobiol.2015.09.008

[9] Talmi YP, Sigler L, Inge E, Finkelstein Y, Zohar Y. Antibacterial properties of human amniotic membranes. Placenta. 1991;12(3):285-288. DOI: 10.1016/0143-4004(91)90010-D

[10] Seo JH, Kim YH, Kim JS. Properties of the amniotic membrane may be applicable in cancer therapy. Medical Hypotheses. 2008;70(4):812-814. DOI: 10.1016/j.mehy.2007.08.008

[11] Jirsova K, Jones GLA. Amniotic membrane in ophthalmology: Properties, preparation, storage and indications for grafting—A review. Cell and Tissue Banking. 2017;18(2):193-204. DOI: 10.1007/s10561-017-9618-5

[12] Riau AK, Beuerman RW, Lim LS, Mehta JS. Preservation, sterilization and de-epithelialization of human amniotic membrane for use in ocular surface reconstruction. Biomaterials. 2010;31(2):216-225. DOI: 10.1016/j.biomaterials.2009.09.034

[13] Hermans MHE. Clinical-experience with glycerol-preserved donor skin treatment in partial thickness burns. Burns. 1989;15(1):57-59. DOI: 10.1016/0305-4179(89)90074-0

[14] Wagner M, Walter P, Salla S, Johnen S, Plange N, Rutten S, et al. Cryopreservation of amniotic membrane with and without glycerol additive. Graefes Archive for Clinical and Experimental Ophthalmology. 2018;256(6):1117-1126. DOI: 10.1007/s00417-018-3973-1
[15] Lee SH, Tseng SCG. Amniotic membrane transplantation for persistent epithelial defects with ulceration. American Journal of Ophthalmology. 1997;123(3):303-312. DOI: 10.1016/S0002-9394(14)70125-4

[16] Malhotra C, Jain AK. Human amniotic membrane transplantation: Different modalities of its use in ophthalmology. World Journal of Transplantation. 2014;4(2):111-121. DOI: 10.5500/wjt.v4.i2.111

[17] Cooke M, Tan EK, Mandrycky C, He H, O’Connell J, Tseng SCG. Comparison of cryopreserved amniotic membrane and umbilical cord tissue with dehydrated amniotic membrane/chorion tissue. Journal of Wound Care. 2014;23(10):465. DOI: 10.12968/jowc.2014.23.10.465

[18] Kruse FE, Joussen AM, Rohrschneider K, You LT, Sinn B, Baumann J, et al. Cryopreserved human amniotic membrane for ocular surface reconstruction. Graefe’s Archive for Clinical and Experimental Ophthalmology. 2000;238(1):68-75. DOI: 10.1007/s004170050012

[19] Shortt AJ, Secker GA, Lomas RJ, Wilshaw SP, Kearney JN, Tuft SJ, et al. The effect of amniotic membrane preparation method on its ability to serve as a substrate for the ex-vivo expansion of limbal epithelial cells. Biomaterials. 2009;30(6):1056-1065. DOI: 10.1016/j.biomaterials.2008.10.048

[20] Niknejad H, Deihim T, Solati-Hashjin M, Peirovi H. The effects of preservation procedures on amniotic membrane’s ability to serve as a substrate for cultivation of endothelial cells. Cryobiology. 2011;63(3):145-151. DOI: 10.1016/j.cryobiol.2011.08.003

[21] Thomasen H, Pauklin M, Noelle B, Geerling G, Vetter J, Steven P, et al. The effect of long-term storage on the biological and histological properties of cryopreserved amniotic membrane. Current Eye Research. 2011;36(3):247-255. DOI: 10.3109/02713683.2010.542267

[22] Prabhasawat P, Kosrirukvongs P, Booranapong W, Vajaradul Y. Application of preserved human amniotic membrane for corneal surface reconstruction. Cell and Tissue Banking. 2000;1(3):213-222. DOI: 10.1023/A:1026542702099

[23] Prabhasawat P, Tseng SCG. Impression cytology study of epithelial phenotype of ocular surface reconstructed by preserved human amniotic membrane. Archives of Ophthalmology-Chic. 1997;115(11):1360-1367. DOI: 10.1001/archopht.1997.01100160530001

[24] Duan-Arnold Y, Gyurdieva A, Johnson A, Uveges TE, Jacobstein DA, Danilkovitch A. Retention of endogenous viable cells enhances the anti-inflammatory activity of cryopreserved amnion. Advances in Wound Care. 2015;4(9):523-533. DOI: 10.1089/wound.2015.0636

[25] Azaara-Blanco A, Pillai CT, Dua HS. Amniotic membrane transplantation for ocular surface reconstruction. British Journal of Ophthalmology. 1999;83(4):399-402. DOI: 10.1136/bjo.83.4.399

[26] Kubo M, Sonoda Y, Muramatsu R, Usui M. Immunogenicity of human amniotic membrane in experimental xenotransplantation. Investigative Ophthalmology & Visual Science. 2001;42(7):1539-1546

[27] Tehrani FA, Ahmadiani A, Niknejad H. The effects of preservation procedures on antibacterial property of amniotic membrane. Cryobiology. 2013;67(3):293-298. DOI: 10.1016/j.cryobiol.2013.08.010

[28] Amniox Medical, Inc. CRYOTEK process. Available from: www.amnioxmedical.com/CRYOTEK-process.html
[29] Yan GH, Sun HQ, Wang F, Wang JP, Wang FC, Zou ZM, et al. Topical application of hPDGF-A-modified porcine BMSC and keratinocytes loaded on acellular HAM promotes the healing of combined radiation-wound skin injury in minipigs. International Journal of Radiation Biology. 2011;87(6):591-600. DOI: 10.3109/09553002.2011.570854

[30] Rama P, Giannini R, Bruni A, Gatto C, Tiso R, Ponzin D. Further evaluation of amniotic membrane banking for transplantation in ocular surface diseases. Cell and Tissue Banking. 2001;2(3):155-163. DOI: 10.1023/A:1020158206073

[31] Tan EK, Cooke M, Mandrycky C, Mahabole M, He H, O’Connell J, et al. Structural and biological comparison of cryopreserved and fresh amniotic membrane tissues. Journal of Biomaterials and Tissue Engineering. 2014;4(5):379-388. DOI: 10.1166/jbt.2014.1180

[32] Paolin A, Trojan D, Leonardi A, Mellone S, Volpe A, Orlandi A, et al. Cytokine expression and ultrastructural alterations in fresh-frozen, freeze-dried and gamma-irradiated human amniotic membranes. Cell and Tissue Banking. 2016;17(3):399-406. DOI: 10.1007/s10561-016-9553-x

[33] Pachigolla G, Prasher P, Di Pascuale MA, McCulley JP, McHenry JG, Mootha VV. Evaluation of the role of ProKera in the management of ocular surface and orbital disorders. Eye & Contact Lens. 2009;35(4):172-175. DOI: 10.1097/ICL.0b013e3181a66a12

[34] Shupe AT, Cheng AMS. Case report: Use of Self-retained cryopreserved amniotic membrane in the treatment of acute chemical ocular burn. Optometry and Vision Science. 2017;94(11):1062-1065. DOI: 10.1097/OPX.0000000000001132

[35] Ayatollahi H, Nourani A, Khodaveisi T, Aghaei H, Mohammadpour M. Teleophthalmology in practice: Lessons learned from a pilot project. The Open Medical Informatics Journal. 2017;11:20-28. DOI: 10.2174/1874431107111010020

[36] Tseng SS, Yang WW, Hsiao JC. Bilateral corneal erosion due to retail purchase of unfitted prescription contact lenses: A case report. Cornea. 2008;27(10):1179-1181. DOI: 10.1097/ICO.0b013e318173140e

[37] Suh LH, Zhang C, Chuck RS, Stark WJ, Naylor S, Binley K, et al. Cryopreservation and lentiviral-mediated genetic modification of human primary cultured corneal endothelial cells. Investigative Ophthalmology & Visual Science. 2007;48(7):3056-3061. DOI: 10.1167/iovs.06-0771

[38] Chang KH, Tan HP, Kuo CC, Kuo DH, Shieh P, Chen FA, et al. Effect of nortriptyline on Ca^{2+} handling in SIRC rabbit corneal epithelial cells. The Chinese Journal of Physiology. 2010;53(3):178-184

[39] Yeh JH, Sun TK, Chou CT, Chen WC, Lee JK, Yeh HC, et al. Effect of sertraline on Ca^{2+} fluxes in rabbit corneal epithelial cells. The Chinese Journal of Physiology. 2015;58(2):85-94. DOI: 10.4077/CJP.2015.BAC255

[40] Horner IJ, Kraut ND, Hurst JJ, Rook AM, Collado CM, Atilla-Gokcumen GE, et al. Effects of polyhexamethylene biguanide and polyquaternium-1 on phospholipid bilayer structure and dynamics. The Journal of Physical Chemistry. B. 2015;119(33):10531-10542. DOI: 10.1021/acs.jpbc.5b07162

[41] Smith VA, Johnson T. Evaluation of an animal product-free variant of MegaCell MEM as a storage medium for corneas destined for transplantation.
[42] Pels E, Beele H, Claerhout I. Eye bank issues: II. Preservation techniques: warm versus cold storage. International Ophthalmology. 2008;28(3):155-163. DOI: 10.1007/s10792-007-9086-1

[43] Balarabe AH, Hassan R, Fatai OO. Eye health seeking habits and barriers to accessing curative services among blind beggars in an urban community in Northern Nigeria. Annals of African Medicine. 2014;13(4):184-188. DOI: 10.4103/1596-3519.142289

[44] Sanz-Marco E, Lopez-Prats MJ, Garcia-Delpech S, Udaondo P, Diaz-Llopis M. Fulminant bilateral Haemophilus influenzae keratitis in a patient with hypovitaminosis A treated with contaminated autologous serum. Clinical Ophthalmology. 2011;5:71-73. DOI: 10.2147/OPTH.S15847

[45] Saltzmann RM, Yep JM, Blomquist PH. Fusarium keratitis associated with ReNu with MoistureLoc sample kits. Eye & Contact Lens. 2008;34(6):337-339. DOI: 10.1097/ICL.0b013e3181c25bf

[46] Wilde C, Messina M, Moshiri T, Snape SE, Maharajan S. Interface Scopulariopsis gracilis fungal keratitis following Descemet’s stripping automated endothelial keratoplasty (DSAEK) with a contaminated graft. International Ophthalmology. 2018;38(5):2211-2217. DOI: 10.1007/s10792-017-0706-0

[47] Montes JA, Johnson D, Jorgensen J, McElmeel ML, Fulcher LC, Kiel JW. Potency and sterility of fortified tobramycin, fortified vancomycin, and moxifloxacin at 4, 24, and 35°C for 14 days. Cornea. 2016;35(1):122-126. DOI: 10.1097/ICO.0000000000000676

[48] Armitage WJ, Tullo AB, Larkin DF. The first successful full-thickness corneal transplant: A commentary on Eduard Zirm’s landmark paper of 1906. The British Journal of Ophthalmology. 2006;90(10):1222-1223. DOI: 10.1136/bjo.2006.101527

[49] Filatov VP. Perspective of optical transplantation of the cornea. Vestnik Oftalmologii. 1946;25(1):34-38

[50] John S, Sengupta S, Reddy SJ, Prabhu P, Kirubanandan K, Badrinath SS. The Sankara Nethralaya mobile teleophthalmology model for comprehensive eye care delivery in rural India. Telemedicine Journal and E-Health. 2012;18(5):382-387. DOI: 10.1089/tmj.2011.0190

[51] Fairbairn NG, Randolph MA, Redmond RW. The clinical applications of human amnion in plastic surgery. Journal of Plastic, Reconstructive & Aesthetic Surgery. 2014;67(5):662-675. DOI: 10.1016/j.bjps.2014.01.031

[52] Zimmerman AB, Richdale K, Mitchell GL, Kinoshita BT, Lam DY, Wagner H, et al. Water exposure is a common risk behavior among soft and gas-permeable contact lens wearers. Cornea. 2017;36(8):995-1001. DOI: 10.1097/ICO.0000000000001204

[53] Rambaud C, Tabary A, Contraires G, El Hassan F, Labalette P. Contextual study of Airsoft gun related ocular injuries. Journal Français d’Ophthalmologie. 2013;36(3):236-241. DOI: 10.1016/j.jfo.2012.01.012

[54] Meizner I. Three dimensional (3D) ultrasound in gynecology: A breakthrough? Harefuah. 2007;146(4):274-275, 317-8

[55] John T. Human amniotic membrane transplantation: Past, present, and future. Ophthalmology Clinics of North America. 2003;16(1):43-65 vi

[56] Greenwald R. The handling of corneal donor tissue before penetrating
Cryopreservation in Ophthalmology
DOI: http://dx.doi.org/10.5772/intechopen.91312

[57] Adeghate E, Donath T. Transplantation of tissue grafts into the anterior eye chamber: A method to study intrinsic neurons. Brain Research Brain Research Protocols. 2000;6(1-2):33-39

[58] Firdova M, Tkacova E, Jesenska Z, Minova M. Mycotic keratitis with uveitis cause by Scedosporium apiospermum. Ceska a Slovenska Oftalmologie: Casopis Ceske Oftalmologicke Spolecnosti a Slovenske Oftalmologicke Spolecnosti. 1997;53(4):248-251

[59] Wang B, Wang Y, Ding J. Repair of cornea-sclera defect by autogenous sclera graft from the same eye. Zhongguo xiu fu chong jian wai ke za zhi = Zhongguo xiufu chongjian waike zazhi [Chinese Journal of Reparative and Reconstructive Surgery]. 1997;11(4):206-207

[60] Xu JJ, Wang Y. The effects of amniotic membrane on corneal penetration of ofloxacin. [Zhonghua yan ke za zhi] Chinese Journal of Ophthalmology. 2006;42(7):624-627

[61] Socci RR, Tachado SD, Aronstam RS, Reinach PS. Characterization of the muscarinic receptor subtypes in the bovine corneal epithelial cells. Journal of Ocular Pharmacology and Therapeutics. 1996;12(3):259-269. DOI: 10.1089/jop.1996.12.259

[62] Yang H, Sun X, Wang Z, Ning G, Zhang F, Kong J, et al. EGF stimulates growth by enhancing capacitative calcium entry in corneal epithelial cells. The Journal of Membrane Biology. 2003;194(1):47-58. DOI: 10.1007/s00232-003-2025-9

[63] Wu X, Yang H, Iserovich P, Fischbarg J, Reinach PS. Regulatory volume decrease by SV40-transformed rabbit corneal epithelial cells requires ryanodine-sensitive Ca$^{2+}$-induced Ca$^{2+}$ release. The Journal of Membrane Biology. 1997;158(2):127-136. DOI: 10.1007/s002329900250

[64] Jeng BH, Meisler DM, Hollyfield JG, Connor JT, Aulak KS, Stuehr DJ. Nitric oxide generated by corneas in corneal storage media. Cornea. 2002;21(4):410-414. DOI: 10.1097/00003226-200205000-00015

[65] Meyer-Rochow VB, Mishra M. Structure and putative function of dark- and light-adapted as well as UV-exposed eyes of the food store pest Psyllipsocus ramburi Selys-longchamps (Insecta: Pscoptera: Psyllipsocidae). Journal of Insect Physiology. 2007;53(2):157-169. DOI: 10.1016/j.jinsphys.2006.11.002

[66] Lin MC, Jan CR. The anti-anginal drug fendiline elevates cytosolic Ca$^{2+}$ in rabbit corneal epithelial cells. Life Sciences. 2002;71(9):1071-1079. DOI: 10.1016/s0024-3205(02)01797-6

[67] Ma D, Mellon J, Niederkorn JY. Conditions affecting enhanced corneal allograft survival by oral immunization. Investigative Ophthalmology & Visual Science. 1998;39(10):1835-1846

[68] Tao W, Wu X, Liou GI, Abney TO, Reinach PS. Endothelin receptor-mediated Ca$^{2+}$ signaling and isoform expression in bovine corneal epithelial cells. Investigative Ophthalmology & Visual Science. 1997;38(1):130-141

[69] Lambiase A, Manni L, Bonini S, Rama P, Micera A, Aloe L. Nerve growth factor promotes corneal healing: Structural, biochemical, and molecular analyses of rat and human corneas. Investigative Ophthalmology & Visual Science. 2000;41(5):1063-1069

[70] Zhang F, Wen Q, Mergler S, Yang H, Wang Z, Bildin VN, et al. PKC isoform-specific enhancement of capacitative calcium entry in human corneal epithelial cells. Investigative
Cryopreservation - Current Advances and Evaluations

Ophthalmology & Visual Science. 2006;47(9):3989-4000. DOI: 10.1167/iovs.06-0253

[71] Xie Q, Zhang Y, Cai Sun X, Zhai C, Bonanno JA. Expression and functional evaluation of transient receptor potential channel 4 in bovine corneal endothelial cells. Experimental Eye Research. 2005;81(1):5-14. DOI: 10.1016/j.exer.2005.01.003

[72] Peterson RC, Wolffsohn JS. The effect of digital image resolution and compression on anterior eye imaging. The British Journal of Ophthalmology. 2005;89(7):828-830. DOI: 10.1136/bjo.2004.062240

[73] Xie Q, Zhang Y, Zhai C, Bonanno JA. Calcium influx factor from cytochrome P-450 metabolism and secretion-like coupling mechanisms for capacitative calcium entry in corneal endothelial cells. The Journal of Biological Chemistry. 2002;277(19):16559-16566. DOI: 10.1074/jbc.M109518200

[74] Yang H, Mergler S, Sun X, Wang Z, Lu L, Bonanno JA, et al. TRPC4 knockdown suppresses epidermal growth factor-induced store-operated channel activation and growth in human corneal epithelial cells. The Journal of Biological Chemistry. 2005;280(37):32230-32237. DOI: 10.1074/jbc.M504553200

[75] Petroll WM, Cavanagh HD, Lemp MA, Andrews PM, Jester JV. Digital image acquisition in vivo confocal microscopy. Journal of Microscopy. 1992;165(Pt 1):61-69. DOI: 10.1111/j.1365-2818.1992.tb04305.x

[76] Greenwald R. The handling of corneal donor tissue before penetrating keratoplasty. Journal of Ophthalmic Nursing & Technology. 1994;13(1):21-22

[77] Socci R, Chu A, Reinach P, Meszaros LG. In situ Ca\(^{2+}\)-induced Ca\(^{2+}\) release from a ryanodine-sensitive intracellular Ca\(^{2+}\) store in corneal epithelial cells. Comparative Biochemistry and Physiology B, Comparative Biochemistry. 1993;106(4):793-797. DOI: 10.1016/0305-0491(93)90032-z

[78] Johnstone EW, Williams KA, Lovric VA, Lubeck D, Barras CW, Coster DJ. Cryopreservation of rabbit and cat corneas at \(-18\) to \(-24\) degrees C. Cornea. 1992;11(3):211-220

[79] Nishida T, Ueda A, Otori T, Fujita H. Long-term storage of endocytosed latex beads in keratocytes in vivo. Cornea. 1991;10(6):532-535. DOI: 10.1097/00003226-199111000-00012

[80] Elias KA, Weiner RI, Mellon SH. Effect of extracellular matrix on prolactin secretion and mRNA accumulation in GH3 cells. DNA and Cell Biology. 1990;9(5):369-375. DOI: 10.1089/dna.1990.9.369

[81] Roth HW. Deformation of the central corneal surface following use of hard and soft contact lenses. Fortschritte der Ophthalmologie: Zeitschrift der Deutschen Ophthalmologischen Gesellschaft. 1989;86(3):185-188

[82] Savion N, Disatnik MH, Nevo Z. Murine macrophage heparanase: Inhibition and comparison with metastatic tumor cells. Journal of Cellular Physiology. 1987;130(1):77-84. DOI: 10.1002/jcp.1041300112

[83] Bethea CL, Kozak SL. Effect of extracellular matrix on PC 12 cell shape and dopamine processing. Molecular and Cellular Endocrinology. 1984;37(3):319-329. DOI: 10.1016/0303-7207(84)90102-3

[84] Bethea CL, Kozak SL. Further characterization of substratum influence on PC12 cell shape and
dopamine processing. Molecular and Cellular Endocrinology. 1985;42(1):59-72. DOI: 10.1016/0303-7207(85)90007-3

[85] Bethea CL. Glucocorticoid stimulation of dopamine production in PC12 cells on extracellular matrix and plastic. Molecular and Cellular Endocrinology. 1987;50(3):211-222. DOI: 10.1016/0303-7207(87)90019-0

[86] Rich LF. A technique for preparing corneal lamellar donor tissue using simplified keratomileusis. Ophthalmic Surgery. 1980;11(9):606-608

[87] Vlodavsky I, Folkman J, Sullivan R, Fridman R, Ishai-Michaeli R, Sasse J, et al. Endothelial cell-derived basic fibroblast growth factor: synthesis and deposition into subendothelial extracellular matrix. Proceedings of the National Academy of Sciences of the United States of America. 1987;84(8):2292-2296. DOI: 10.1073/pnas.84.8.2292

[88] Ma DH, Chen HC, Ma KS, Lai JY, Yang U, Yeh LK, et al. Preservation of human limbal epithelial progenitor cells on carbodiimide cross-linked amniotic membrane via integrin-linked kinase-mediated Wnt activation. Acta Biomaterialia. 2016;31:144-155. DOI: 10.1016/j.actbio.2015.11.042

[89] McCarey BE, Kaufman HE. Improved corneal storage. Investigative Ophthalmology. 1974;13(3):165-173

[90] Lindstrom RL, Kaufman HE, Skelnik DL, Laing RA, Lass JH, Musch DC, et al. Optisol corneal storage medium. American Journal of Ophthalmology. 1992;114(3):345-356. DOI: 10.1016/s0002-9394(14)71803-3

[91] Ehlers H, Ehlers N, Hjortdal JO. Corneal transplantation with donor tissue kept in organ culture for 7 weeks. Acta Ophthalmologica Scandinavica. 1999;77(3):277-278. DOI: 10.1034/j.1600-0420.1999.770306.x

[92] Basu PK, Hasany S. Autolysis of the cornea of stored human donor eyes. Canadian Journal of Ophthalmology: Journal canadien d’ophthalmologie. 1974;9(2):229-235

[93] Pegg DE. Principles of cryopreservation. Methods in Molecular Biology (Clifton, NJ). 2007;368:39-57. DOI: 10.1007/978-1-59745-362-2_3

[94] Engineering challenges in tissue preservation. Cell Preservation Technology. 2004;2(2):91-112. DOI: 10.1089/1538344044774101945

[95] Kim SY, Yang J, Lee YC. Safety of moxifloxacin and voriconazole in corneal storage media on porcine corneal endothelial cells. Journal of Ocular Pharmacology and Therapeutics: The Official Journal of the Association for Ocular Pharmacology and Therapeutics. 2010;26(4):315-318. DOI: 10.1089/jop.2009.0136

[96] Heindl LM, Riss S, Adler W, Bucher F, Hos D, Cursiefen C. Split cornea transplantation: Relationship between storage time of split donor tissue and outcome. Ophthalmology. 2013;120(5):899-907. DOI: 10.1016/j.ophtha.2012.11.012

[97] Lopez-Garcia JS, Garcia-Lozano I, Rivas L, Ramirez N, Mendez MT, Raposo R. Stability of growth factors in autologous serum eyedrops after long-term storage. Current Eye Research. 2016;41(3):292-298. DOI: 10.3109/02713683.2015.1016180

[98] Borderie VM, Lopez M, Lombet A, Carvajal-Gonzalez S, Cywiner C, Laroche L. Cryopreservation and culture of human corneal keratocytes. Investigative Ophthalmology & Visual Science. 1998;39(8):1511-1519

[99] Fischer KR, Opitz A, Boeck M, Geerling G. Stability of serum eye drops after storage of 6 months. Cornea.
Cryopreservation – Current Advances and Evaluations

2012;31(11):1313-1318. DOI: 10.1097/ICO.0b013e3182542085

[100] Lai JY, Wang PR, Luo LJ, Chen ST. Stabilization of collagen nanofibers with L-lysine improves the ability of carbodiimide cross-linked amniotic membranes to preserve limbal epithelial progenitor cells. International Journal of Nanomedicine. 2014;9:5117-5130. DOI: 10.2147/ijn.s69689

[101] Guindolet D, Crouzet E, He Z, Herbepin P, Jumelle C, Perrache C, et al. Storage of porcine cornea in an innovative bioreactor. Investigative Ophthalmology & Visual Science. 2017;58(13):5907-5917. DOI: 10.1167/iovs.17-22218

[102] Sabater AL, Guarnieri A, Espana EM, Li W, Prosper F, Moreno-Montanes J. Strategies of human corneal endothelial tissue regeneration. Regenerative Medicine. 2013;8(2):183-195. DOI: 10.2217/rme.13.11

[103] Patel D, Tandon R, Ganger A, Vij A, Lalwani S, Kumar A. Study of death to preservation time and its impact on utilisation of donor corneas. Tropical Doctor. 2017;47(4):365-370. DOI: 10.1177/0049475517713406

[104] Kim KY, Jung JW, Kim EK, Seo KY, Kim TI. Tectonic lamellar keratoplasty using cryopreserved cornea in a large descemetocele. Yonsei Medical Journal. 2016;57(1):269-271. DOI: 10.3349/ymj.2016.57.1.269

[105] Duncan K, Parker J, Hoover C, Jeng BH. The effect of light exposure on the efficacy and safety of amphotericin B in corneal storage media. JAMA Ophthalmology. 2016;134(4):432-436. DOI: 10.1001/jamaophthalmol.2016.0008

[106] Cunningham WJ, Moffatt SL, Brookes NH, Twohill HC, Pendergrast DG, Stewart JM, et al. The New Zealand National Eye Bank study: Trends in the acquisition and storage of corneal tissue over the decade 2000 to 2009. Cornea. 2012;31(5):538-545. DOI: 10.1097/ICO.0b013e318222c3f2

[107] Lee JK, Ryu YH, Ahn JJ, Kim MK, Lee TS, Kim JC. The effect of lyophilization on graft acceptance in experimental xenotransplantation using porcine cornea. Artificial Organs. 2010;34(1):37-45. DOI: 10.1111/j.1525-1594.2009.00789.x

[108] Mauger T, Quartetti E. The effects of the presence of the corneal epithelium and supplemental hydrocortisone on beta-glucuronidase levels with corneal preservation. Clinical Ophthalmology. 2011;5:415-417. DOI: 10.2147/opth.s17228

[109] Daoud YJ, Smith R, Smith T, Akpek EK, Ward DE, Stark WJ. The intraoperative impression and postoperative outcomes of gamma-irradiated corneas in corneal and glaucoma patch surgery. Cornea. 2011;30(12):1387-1391. DOI: 10.1097/ICO.0b013e31821c9c09

[110] Patel HY, Ormonde S, Brookes NH, Moffatt SL, Sherwin T, Pendergrast DG, et al. The New Zealand National Eye Bank: Survival and visual outcome 1 year after penetrating keratoplasty. Cornea. 2011;30(7):760-764. DOI: 10.1097/ICO.0b013e3182014668

[111] Feilmeier MR, Tabin GC, Williams L, Oliva M. The use of glycerol-preserved corneas in the developing world. Middle East African Journal of Ophthalmology. 2010;17(1):38-43. DOI: 10.4103/0974-9233.61215

[112] Yang JW, Lin HC, Hsiao CH, Chen PY. Therapeutic penetrating keratoplasty in severe infective keratitis using glycerol-preserved donor corneas. Cornea. 2012;31(10):1103-1106. DOI: 10.1097/ICO.0b013e31821c9ba2
[113] Fabiano A, Bizzarri R, Zambito Y. Thermosensitive hydrogel based on chitosan and its derivatives containing medicated nanoparticles for transcorneal administration of 5-fluourouracil. International Journal of Nanomedicine. 2017;12:633-643. DOI: 10.2147/ijn.s121642

[114] Gaum L, Reynolds I, Jones MN, Clarkson AJ, Gillan HL, Kaye SB. Tissue and corneal donation and transplantation in the UK. British Journal of Anaesthesia. 2012;108 (Suppl 1):i43-i47. DOI: 10.1093/bja/aer398

[115] Costa D, Leiva M, Naranjo C, Rios J, Pena MT. Cryopreservation (−20°C) of feline corneoscleral tissue: Histologic, microbiologic, and ultrastructural study. Veterinary Ophthalmology. 2016;19 (Suppl 1):97-104. DOI: 10.1111/vop.12393

[116] Hashimoto Y, Hattori S, Sasaki S, Honda T, Kimura T, Funamoto S, et al. Ultrastructural analysis of the decellularized cornea after interlamellar keratoplasty and microkeratome-assisted anterior lamellar keratoplasty in a rabbit model. Scientific Reports. 2016;6:27734. DOI: 10.1038/srep27734

[117] Tandon R, Singh A, Gupta N, Vanathi M, Gupta V. Upgradation and modernization of eye banking services: Integrating tradition with innovative policies and current best practices. Indian Journal of Ophthalmology. 2017;65(2):109-115. DOI: 10.4103/ijo.IJO_862_16

[118] Gupta N, Upadhyay P. Use of glycerol-preserved corneas for corneal transplants. Indian Journal of Ophthalmology. 2017;65(7):569-573. DOI: 10.4103/ijo.IJO_56_17

[119] Liu Z, Zhou Q, Zhu J, Xiao J, Wan P, Zhou C, et al. Using genipin-crosslinked acellular porcine corneal stroma for cosmetic corneal lens implants. Biomaterials. 2012;33(30):7336-7346. DOI: 10.1016/j.biomaterials.2012.06.080

[120] Miller TD, Maxwell AJ, Lindquist TD, Requard J 3rd. Validation of cooling effect of insulated containers for the shipment of corneal tissue and recommendations for transport. Cornea. 2013;32(1):63-69. DOI: 10.1097/ICO.0b013e3182685c8e

[121] Rijneveld WJ, Wolff R, Volker-Dieben HJ, Pels E. Validation of tissue quality parameters for donor corneas designated for emergency use in preservation of the globe. Cornea. 2010;29(2):128-132. DOI: 10.1097/ICO.0b013e3181ac07bc

[122] Bi YL, Bock F, Zhou Q, Cursiefen C. Central corneal epithelium self-healing after ring-shaped glycerin-cryopreserved lamellar keratoplasty in Terrien marginal degeneration. International Journal of Ophthalmology. 2013;6(2):251-252. DOI: 10.3980/j.issn.2222-3959.2013.02.27

[123] Liu X, Shen JH, Zhou Q, Liu ZX, Tang SF, Chen RR, et al. Personalised lamellar keratoplasty and keratopigmentation in Asian corneal leucoma patients. International Journal of Clinical and Experimental Medicine. 2015;8(6):9446-9453

[124] Liu X, Zhou Q, Huang X, Liu Z, Bi Y. Clinical evaluation of deep anterior lamellar keratoplasty using glycerol-cryopreserved corneal tissues for refractory herpetic stromal keratitis: An observational study. Medicine (Baltimore). 2016;95(39):e4892. DOI: 10.1097/MD.0000000000004892