Original Article

Clinical Outcome of Amniotic Membrane Transplantation in Chemically Injured Eyes

Shafiul Ashraf Rubel[1], Natasha Kajmina[2], Tarzeen Khadiza Shuchi[3], Mahfuza Rahman[4], Farzana Hossain Tuli[5], Md. Abdul Quader[6]

Abstract

Purpose: To evaluate the clinical outcome of Amniotic Membrane Transplantation (AMT) in chemically injured eyes for ocular surface reconstruction. **Methods:** A prospective study of 30 consecutive chemically injured eyes was performed from December 2022 to May 2023 in National Institute of Ophthalmology & Hospital (NIO&H), Dhaka, Bangladesh. Twenty-seven patients (30 eyes having corrosive chemical injuries) were included in the study according to the inclusion & exclusion criteria. AMT was performed within 21 days of injury by single surgeon with modified surgical procedure. Post operative evaluation was done by Snellen chart, slit lamp biomicroscope and Schirmer-II test. **Results:** After 3 months of AMT, there was no pain or inflammation. Improvement of vision found in 20 (66.67%) of 30 eyes, 4 (13.33%) eyes of rest 10 eyes showed inaccurate Projection of Rays (PR) vision and rest 6 (20.0%) eyes of rest 10 eyes found deteriorated vision. Epithelialization occurred in all eyes. Twenty-four (80.0%) eyes had limbal stem cell deficiency and they experienced superficial corneal vascularization. Symblepharon was seen in 18 (60.0%) eyes, 6 (20.0%) of these 18 eyes developed marked conjunctival shortening with ankyloblepharon. None of the eyes showed perforation. **Conclusions:** AMT increases patient comfort and reduces inflammation. In mild burns, AMT alone restores corneal and conjunctival surfaces. In moderate to severe burns, it reduces conjunctival scarring sequelae, but does not prevent the sequelae of limbal stem cell deficiency that requires further limbal stem cell transplantation. In the acute stage, AMT has a protective role against the progressive melting and perforation.

Keywords: Amniotic Membrane Transplantation, Chemical injury, Limbal stem cell deficiency, Limbal stem cell transplantation.

(J.Natl.Inst.Ophthalmol.2023;6(1):21-26) *https://doi.org/10.3329/jnio.v6i1.69848*

1. Lieutenant Colonel, Classified Eye Specialist, CMH Dhaka and Fellow Cornea-Anterior Segment, NIO&H
2. Colonel, Classified Eye Specialist, CMH Dhaka
3. Assistant Professor, Department of Cornea, NIO&H
4. Major, Classified Eye Specialist, CMH Dhaka
5. Epidemiologist, NIO&H
6. Professor & Head of the department of Cornea, NIO&H

Address of correspondence:
Dr. Lieutenant Colonel Shafiul Ashraf Rubel
MBBS, FCPS (Ophthalmology)
Fellow Cornea-Anterior Segment
Classified Eye Specialist, CMH Dhaka
Dhaka Cantonment, Bangladesh
Email: shafiulrubel85@gmail.com

Received: 3 Sep. 2022 **Accepted: 12 Dec. 2022**

Introduction

Chemical injuries to the eyes are regarded as true ophthalmic emergencies, as they can cause permanent ocular and intraocular damage. Ocular exposure to the strong alkalis and acids results in damage to the ocular surface epithelium, cornea, anterior segment and limbal stem cells resulting in permanent unilateral or bilateral visual impairment and even blindness. Chronic inflammation hampers epithelialization, thus acceleration of corneal ulceration and melting with globe perforation takes place. The most devastating sequelae of chemical injuries are corneal melt, limbal stem cell deficiency and secondary glaucoma - tend to occur over the long term. Limbal stem cell destruction causes corneal conjunctivalization, vascularization, chronic inflammation, and recurrent or persistent epithelial defects. Conjunctival cells damage causes mucus deficiency and persistent subconjunctival inflammation resulting in severe

dry eye and fibrosis of subconjunctival tissue.[1] It also contributes to scarring sequelae like symblepharon, lid shortening and inflammatory granuloma in the chronic stage. In severe burns ischaemic changes result in anterior segment necrosis and sterile corneal ulceration at an early stage after the injury. In these cases the visual prognosis is poor and functional or physical loss of the eye is not uncommon.[2]

Medical and surgical management strategy for chemical injuries to the eyes has improved a lot over last decade. On the other hand, public health education level also improved, thus emphasis on eye protection in the workplace increased. Though, the final visual outcome is strongly related to the nature of the initial chemical exposure and its severity, the prognosis is highly influenced by the timing of appropriate treatment.[3]

Medical treatment for acute ocular burns includes topical and systemic ascorbate,[4] citrate,[5] tetracycline,[6] and steroids.[7] Application of glued on-hard contact lens,[8] tenoplasty,[9] use of tissue adhesives,[10] and large-diameter keratoplasty[11] are some of the surgical procedures tried in the management of severe ocular chemical burns.

Among various surgical procedures- AMT is a promising technique. Amniotic membrane is the innermost layer of the placenta and consists of three layers: a stromal matrix, a thick basement membrane (BM), and a single epithelial layer. Clinically, transplantation of amniotic membrane (AM) as a permanent surgical graft has been shown to promote epithelialization and reduce inflammation, scarring and neovascularization.[12] Several mechanisms of action are responsible for these attributes. Mimicry of the conjunctival BM components, reduction of proteolytic activity and increased goblet cell density are some of the actions of the amniotic basement membrane.[13] Furthermore, amniotic membrane appears to be an immune-privileged tissue and also to possess immune-regulatory factors, which make it ideal for reducing inflammation. Since only incomplete HLA antigens are found in amniotic membrane, it is immunologically inert and not rejected by the recipient.[14]

Reports regarding the role of AMT in acute ocular burns success is highly variable.[15] Meller et al[11] reported that AMT alone was sufficient to restore corneal and conjunctival surface in mild to moderate burns. In severe burns, it restored the conjunctival surface without symblepharon and reduced limbal stromal inflammation but did not prevent limbal stem cell deficiency that required further limbal stem cell transplantation. However, Dua et al[12] reported failure of AMT to restore ocular surface or preserve the integrity of eye in severe acute burns. A recent report by Kobayashi et al[16] also emphasized that immediate amniotic membrane transplantation is useful for mild to moderate acute chemical burns and preserves ocular surface integrity.

It is believed that when used at an early stage, AMT would promote healing of ocular surface by preventing leucocytic infiltration, decreasing the duration and severity of inflammation and protecting the proliferating epithelial stem cell.[17] In the current study, we report our experience with the use of amniotic membrane transplant for the treatment of ocular chemical injuries.

Materials and methods

In this prospective study, amniotic membrane transplantation was performed in 30 eyes of 27 consecutive patients having corrosive chemical injuries. The study was carried out in National Institute of Ophthalmology & Hospital (NIO&H), Dhaka, Bangladesh from December 2022 to May 2023 with the aim to evaluate VA, corneal epithelial healing, corneal vascularization and symblepharon formation variables after AMT in chemical injuries to the eyes.

Approximately 52.4% of patients achieve corneal vascularization after amniotic membrane transplant[14]. This study is designed to detect a 20% difference in spectacle independence. With 5% level of significance and a power of 80%, the required sample size was 24. Considering 10% non-response rate, the final sample size came to 27. Patients of both genders, between the age of 6 to 65 years of age with injured eyes by corrosive chemicals were included in this study.

Any associated ocular pathology, any history of previous ocular surface surgery, patients with

diabetes mellitus, with poor compliance, with corneal transplantation were excluded from the study. Preserved Amniotic Membrane, Biological Fibrin glue, were used in this study.

The severity was classified as grade II in 6 eyes, Grade III in 8 eyes and Grade IV in 16 eyes based on the criteria defined by the Roper-Hall.[18]

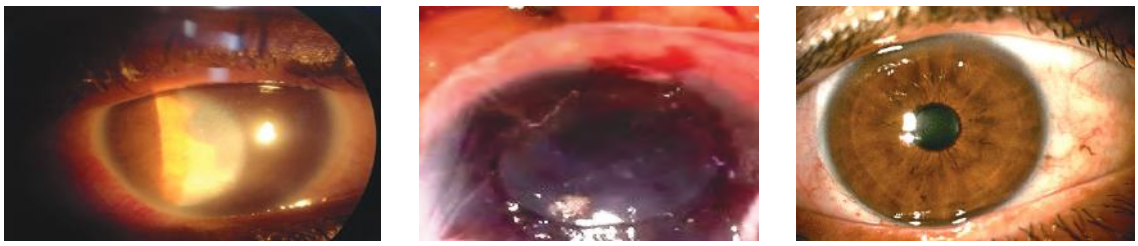

Fig 1: Acid burn of right eye of a 26 years old female. (a) Before AMT, (b) During AMT, (c) 3 months after AMT

All patients received a detailed ophthalmic examination, including visual acuity, slit lamp examination, tonometry, and fundus examination (wherever possible). The Schirmer test-II was performed to evaluate tear status. Cases with lid involvement were excluded from the study.

All the patients were initially treated with medical therapies consisting of copious saline irrigation, topical antibiotics, lubricants, cycloplegics, 10% citrate, 10% ascorbate, and systemic vitamin-C.

Surgical procedures: Preserved AMT was performed within 21 days (3-21days) after injury. These Amniotic membranes (tested free from hepatitis B, hepatitis C, syphilis and human immunodeficiency virus) were obtained from Gonoshastho Sangstha Hospital, Savar, Dhaka. Informed consent was obtained after explanation of the procedure from all the patients. In 12 eyes, amniotic membrane was used as a circular patch covering the cornea and limbus and sutured to the less damaged conjunctival surface; so that the damaged area was covered. In the remaining 18 eyes, amniotic membrane covered the whole ocular surface from lid margin to lid margin. Amniotic membrane was spread on the surface of eye stromal side down, a symblepharon ring of appropriate size was fitted over it. Edges of amniotic membrane after trimming were sutured to lid margins with 8/0 vicryl. Bandage contact lens was applied to all eyes after the surgery.

Postoperatively all patients were treated with Prednisolone acetate 1% eye drop, Moxifloxacin 0.5% eye drop, tear substitutes, topical cycloplegics, oral ciprofloxacin 500mg tablet, Ascorbic acid 250mg tablet. Drops were tapered and discontinued after 2–3 months.

Results

Table 1: Demographic characteristics of the patients (n=30)

Demographic characteristics	Number	Percentages
Gender	20	66.67
Male	10	33.33
Female		
Age (in years)	32.55 ± 13.62 (6.0-65.0)	

Majority of the patients (66.67%) were male while the mean age of the patients was 32.55 ± 13.62 years (table I).

Table II: Nature of injury

Nature of injury	Number	Percentages
Type		
Accidental	28	93.33
Homicidal	2	6.67
Causative agent		
Lime burn	20	66.67
Acid burn	10	33.33
Grading of injury		
Grade II	6	20.0
Grade III	8	26.67
Grade IV	16	53.33

Most of the patients (93.33%) had accidental injury and in majority (66.67%) cases, lime burn was the cause of injury. According to grade of injury, 8 (26.67%) patients had grade III and 16 (53.33%) patients had grade IV injuries (table II).

Table III: Clinical outcomes of AMT after chemical injuries to eyes

Clinical outcomes	After 1st week	After 1st month	After 3rd month
Improvement of visual acuity	6 (20.0%)	14 (46.67%)	20 (66.67%)
healing of conjunctival & corneal epithelial defect	18 (60.0%)	22 (73.33%)	30 (100.0%)
Tear film status >5mm wetting in Schirmer-II test	22 (73.33%)	26 (86.67.0%)	30 (100.0%)

AMT performed 3-21 days after initial injury. Amniotic membrane disintegrated on its own over a period of 7–20 days after transplantation. The final clinical outcomes of AMT after chemical injuries to eyes are shown in Table 3, Fig 2 and Fig 3.

Notably in our study, improvement in visual acuity was seen in 20 (66.67%) of 30 eyes. All eyes in Grade II (6 eyes) and Grade III (8 eyes) showed vision improvement. Whereas in Grade IV burns of 16 eyes only 6 eyes had vision improvement, in 4 eyes it remained same and in 6 eyes it deteriorated. 2 eyes became inaccurate Projection of Rays (PR) because of development of secondary glaucoma and uveitis.

Symblepharon was seen in 18 of 30 eyes. 2 eyes with Grade II burn developed minimal inferior forniceal shortening. 2 eyes in Grade III burns developed mild symblepharon. In all, 14 of 16 eyes with grade IV burns developed symblepharon. But symblepharon was mild in most of the cases except for 6 eyes that developed marked conjunctival shortening with ankyloblepharon. All these eyes had acid burns with 100% conjunctival involvement and total limbal ischemia. No eye experienced any corneal perforation.

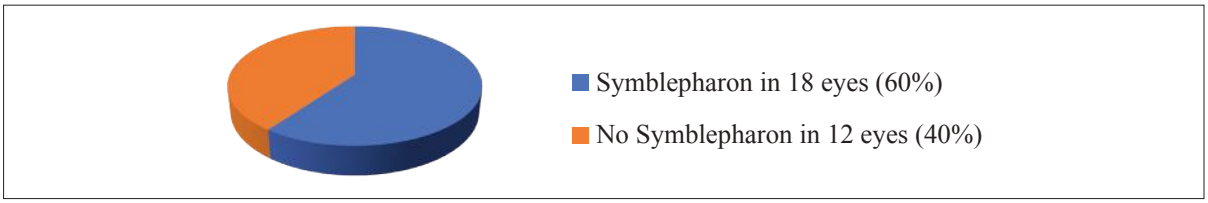

Fig 2: Symblepharon formation on injured eyes

Superficial corneal vascularization as an indicator of limbal stem cell deficiency was seen in 24 (80%) of 30 eyes. In Grade II burns 2 eyes had 2 clock hours, in grade III burns 4 eyes showed 3 clock hours of peripheral superficial corneal vascularization. 2 eyes of Grade III burns had 6 clock hour and all eyes of grade IV burns 8–12 clock hour of superficial corneal vascularization encroaching till the center. The severity of corneal vascularization was related to the severity of chemical burn.

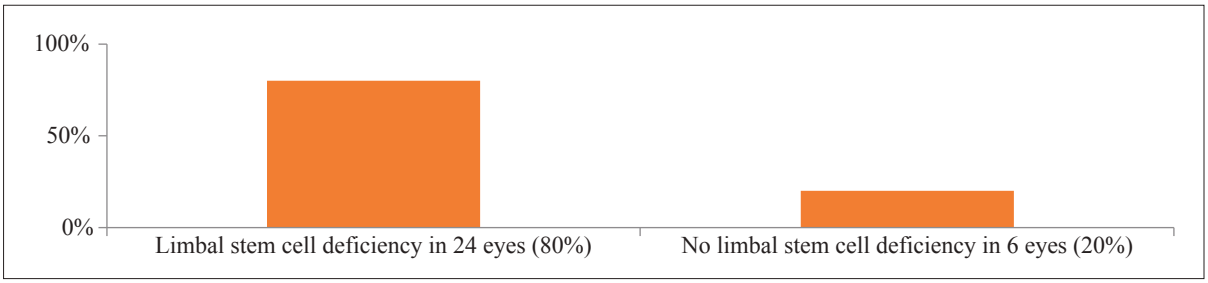

Fig 3: Corneal vascularization associated with Limbal Stem Cell Deficiency

Discussion

In our study, in Grade II burns, AMT restored the ocular surface rapidly as there was partial stem cell loss only. However, amniotic membrane by virtue of its anti-inflammatory and stem cell proliferating activity leads to a favorable outcome for long-term ocular surface restoration and visual recovery.

AMT was effective to a certain extent in Grade III burns also in our study. AMT promoted epithelialization and none of the eyes had persistent epithelial defects, and improvement in visual acuity was noted in 20 of 30 eyes. None of the eyes developed ulceration or perforation. Although AMT was not totally effective in preventing symblepharon and corneal vascularization, but their severity was mild to moderate.

Meller et al[11] treated 13 eyes of chemical burns with AMT within 2 weeks after the injury. A total of 7 eyes had grade II–III burns and 6 eyes had grade IV burns. Epithelial defects of all but two patients healed in 2–5 weeks. Only one patient developed a symblepharon. All eyes with grade IV burns experienced limbal stem cell deficiency. Out of five patients with total limbal ischaemia, three required limbal stem cell transplantation, one required large penetrating keratoplasty and conjunctival flap for a corneal perforation, and the last patient had a persistent epithelial defect at the end of 4 months after which he was lost to follow up. Thus, AMT alone could not maintain the ocular surface for burns with total limbal ischaemia.[11]

In our study, we found very limited utility of AMT in grade IV injury. Symblepharon and corneal vascularization were noted in all eyes. In all, 6 eyes developed ankyloblepharon and 4 eyes went into phthisis. It thereby suggested that in severe burns with extensive conjunctival damage it does not completely restore the conjunctival surface and with associated extensive limbal stem cell damage, it does not prevent the sequelae of limbal stem cell deficiency[19]. Although stable, a less inflamed external ocular surface was achieved in most of the cases and this may aid in obtaining more successful results when limbal stem cell transplantation is resorted to at a later date.

Similarly, Dua et al[12] reported that in extremely severe burns AMT does not establish the ocular surface or preserve the integrity of the globe. Kobayashi et al[16] also reported the usefulness of amniotic membrane patch. Burns were mild with

less than 1/3 limbal ischaemia in four patients and 1/2 limbal ischaemia in one patient. However, the results of their study cannot in any way suggest the usefulness of AMT in severe burns. A new modification in suture placement for amniotic membrane patch was also suggested.[12,16]

In our patients, instead of interrupted sutures we used running sutures to anchor the amniotic membrane to the lid margins and episcleral hitching was used. We also devised a new modification for amniotic membrane patching. After spreading the amniotic membrane on the ocular surface a symblepharon ring of appropriate size was fitted, and amniotic membrane edges after trimming were sutured to the lid margins. It has the benefit of not only keeping amniotic membrane taut but also there was no need for additional purse string sutures to be placed.

Conclusion

Grade II and III chemical burns to the eyes can be benefited from AMT in adjunct to medical therapy for faster recovery. But for Grade IV injury, AMT with other surgical procedures like tenonplasty, limbal stem cell transplantation or tectonic transplantation is important to obtain the ultimate purpose of maintaining the integrity of the eye for future visual rehabilitation.

Conflict of Interest

There was no conflict of any financial interest.

Acknowledgement

I would like to express my sincere gratitude to the Gonoshastho Sangstha Hospital, Savar, Dhaka, authority for their invaluable support during the course of my research.

References

1. Pfister RR, Paterson CA. Ascorbic acid in the treatment of alkali burns of the eye. Ophthalmol 2005; 87: 1050–1057.
2. Prabhasawat P, Tseng SC. Impression cytology study of epithelial phenotype of ocular surface reconstructed by preserved human amniotic membrane. Arch Ophthalmol 1997;115:1360–7.
3. Lee SB, Li DQ, Tan DT, et al. Suppression of TGF-beta signaling in both normal conjunctival fibroblasts and pterygial body fibroblasts by amniotic membrane. Curr Eye Res 2000;20:325–34.
4. Pfister RR, Haddox JL, Paterson CA. The efficacy of sodium citrate in the treatment of severe alkali burns of the eye is influenced by the route of administration. Cornea 2009; 1: 205–211.
5. Merle H, Gerard M, Schrage N, J Fr Ophthalmol 2008;31:723-34.
6. Malhotra R, Sheikh I, Dheansa B. The management of eyelid burns. Surv Ophthalmol 2009;54:356-71.
7. Vajpayee RB, Shekhar H, Sharma N, Jhanji V. Demographic and clinical profile of ocular chemical injuries in the paediatric age group. Ophthalmology 2014;121:377-80.
8. Kim JS, Kim JC, Na BK, et al. Amniotic membrane patching promotes healing and inhibits proteinase activity on wound healing following acute corneal alkali burn. Exp Eye Res 2000;70:329–37.
9. Pfister RR, Pfister DR. Alkali injuries of the eye (Chapter 99). In: Cornea. Krachmer JH, Mannis MJ, Holland EJ. 3rd ed. 2011. p. 1193-1202.
10. Eslani M, Baradaran Rafii A, Movahedan A, Djalilian AR. The ocular surface chemical burns. J Ophthalmol 2014; 2014:196-827.
11. Meller D, Tseng SC. Conjunctival epithelial cell differentiation on amniotic membrane. Invest Ophthalmol Vis Sci 1999;40:878–86.
12. Joseph A, Dua HS, King AJ. Failure of amniotic membrane transplantation in the treatment of acute ocular burns. Br J Ophthalmol 2001; 85: 1065–1069.
13. Arici C, Iskeleli G, Atalay E, Mangan MS, Kilic B. Molten Metal-related ocular thermal burn: Report on two cases. Arq Bras Oftalmol 2015;78:379-81.
14. Mark J, Mannis Edward J, Holland. Cornea Fundamentals, Diagnosis and Management, 4th Edition 2017;94:1106-1119.
15. Kobayashi A, Shirao Y, Yoshita T, Yagami K, Segawa Y, Kawasaki K et al. Temporary amniotic membrane patching for acute chemical burns. Eye 2003; 17: 140–158.
16. Parmar, et al.: Amniotic membrane tra nsplant in acute ocular surface burns Indian Journal of Ophthalmology. January 2021; Volume 69 Issue 1.
17. Fukuda K, Chikama T, Nakamura M, et al. Differential distribution of subchains of the basement membrane components type IV collagen and laminin among the amniotic membrane, cornea, and conjunctiva. Cornea 1999;18:73–9.
18. Roper-Hall MJ. Thermal and chemical burns. Trans Ophthalmol UK 1965; 85: 631–640.
19. Tseng SC, Li DQ, Ma X. Suppression of transforming growth factor-beta isoforms, TGF-beta receptor type II, and myofibroblast differentiation in cultured human corneal and limbal fibroblasts by amniotic membrane matrix. J Cell Physiol 1999;179:325–35.
20. Kubo M, Sonoda Y, Muramatsu R, et al. Immunogenicity of human amniotic membrane in experimental xenotransplantation. Invest Ophthalmol Vis Sci 2001;42:1539–46.