Research Paper

Volume : 5 | Issue : 4 | April 2016 • ISSN No 2277 - 8179 | IF : 3.508 | IC Value : 69.48

Fresh Human Amniotic Membrane –A Versatile Treatment for Ocular Surface Disease

Medical science

KEYWORDS : - ocular surface disorders, fresh amniotic membrane, graft

Dr. Amit S.Nene	- Department of Ophthalmology, Assam Medical College & Hospital, Dibrugarh, Assam. Pin code- 786002, India.
Dr. (Mrs.) B.S. Puzari	- Department of Ophthalmology, Assam Medical College & Hospital, Dibrugarh, Assam. Pin code- 786002, India.
Dr. P. Bharali	- Department of Ophthalmology, Assam Medical College & Hospital, Dibrugarh, Assam. Pin code- 786002, India.

ABSTRACT

Background – Chronic ocular surface disorders, which can result in severe functional impairment, have been viewed for decades as untreatable diseases. Amniotic membrane transplantation (AMT) is indicated for numerous ocular surface disorders, the spectrum of which has been expanding recently. The management of patients with severe ocular surface disease has always been a challenge for ophthalmologists. AMT offers the advantage over conventional corneal transplantation – relative ease of surgery and avoidance of allograft rejection & has become attractive alternate for countries where there is shortage of corneal tissue. Even if corneal transplantation is required it can be performed after AMT where ocular surface is not inflamed.

Aim – To evaluate the role of amniotic membrane as a graft material in the management of ocular surface disorders.

Design – Prospective analytical study.

Methods - The study included 45 patients; 24 (53.3%) cases of pterygium, 11 (24.5%) Chemical injury, 5 (11.2%) Symblepharon, 2(4.4%) thermal injury, 2 (4.4%) persistent corneal epithelial defect and 1(2.2%) SJS. They were treated with fresh amniotic membrane graft and improvement in the signs and symptoms were evaluated. The overall success rate was 84.4% in achieving goal of symptomatic relief, improved epithelialization, stromal healing and vision.

Conclusion - AMT offers the advantage over conventional corneal transplantation – relative ease of surgery and avoidance of allograft rejection & has become attractive alternate for countries where there is shortage of corneal tissue. Even if corneal transplantation is required it can be performed after AMT where ocular surface is not inflamed.

Introduction

Amniotic membrane transplantation (AMT) is indicated for numerous ocular surface disorders, the spectrum of which has been expanding recently. The management of patients with severe ocular surface disease has always been a challenge for ophthalmologists.[1] The advent of amniotic membrane (AM) and limbal stem cell grafts have transformed the treatment of diseases resulting in ocular surface failure.[2] AMT has been used for a variety of ocular surface problems including persistent corneal epithelial defects, for pterygium surgery, conjunctival defects after removal of surface tumors, LSCD as in acute and chronic chemical injuries, Steven Johnson syndrome and shield ulcer of vernal keratoconjunctivitis.

De Roth (1940) was the first to describe live placental membranes to repair conjunctival defects.[3] Inclusion of the highly immunogenic chorion resulted in a low success rate. Sorsby et al. (1946) used chemically processed dry amniotic membrane (AM) termed amnioplastin as a temporary patch for the treatment of ocular burns. However, the need for repetitive applications made the technique rather unpopular until its revival in 1995, by Kim and Tseng. When appropriately processed and preserved, amniotic membrane can be used as a graft to replace the damaged ocular surface stromal matrix or as a patch to prevent unwanted inflammatory insults from gaining access to the damaged ocular surface. The objective of the study is to evaluate the role of amniotic membrane as a graft material in the management of Ocular surface disorders.

II. Methods

The study is a prospective, analytical & interventional case series of all patients who presented with features of ocular surface disorders to the Department of Ophthalmology, Assam Medical College & Hospital, Dibrugarh, Assam. All patients presenting to Out-patient Department in this manner between 1st July 2013 to 30th June 2014 were considered to meet the inclusion criteria. The patients with persistent corneal epithelial defects, bullous keratopathy, pterygium, chemical injury, thermal burns, neurotropic corneal ulcers, mechanical trauma & Stevens – Johnson's syndrome are included in the study. Patients not giving consent for the procedure & with infected corneal ulcer are excluded.

A total of 45 patients satisfying the above criteria underwent detailed pre-operative examination which included best corrected visual acuity, extraocular movements, slit lamp evaluation & posterior chamber evaluation wherever possible. In this study we used fresh amniotic membrane which was obtained from patients undergoing elective caesarean section in Assam Medical College & Hospital in Department of Obstetrics & Gynecology. The amniotic membrane were harvested from consenting seronegative (Negative for Hepatitis B and C virus, Syphilis and HIV) maternal donors. Under sterile conditions, the placenta was thoroughly washed with balanced salt solution to remove clots and debris. The AM was separated from the chorion by blunt dissection and thoroughly rinsed in Normal saline. The membrane was then bathed with balanced salt solution containing 50 µ/ml streptomycin, 100 µg/ml neomycin, and 2.5 µg/ml amphotericin B.[4,5,6,7,8] After appropriate anesthesia to the eye, the devitalized tissues on the cornea or conjunctiva were debrided using a micro sponge and fine forceps & the amniotic membrane was then secured to host tissue using 10-0 nylon sutures on the cornea and 8-0 vicryl used to suture onto sclera whenever required.

Post-operative evaluation was done after 1st post-operative day, 1st week, 2nd week, 4th week, 2nd month & 6th month after operation. The amniotic membrane graft was assessed by looking at acceptance of graft by host & graft shrinkage. Improvement was graded as minimal, moderate & marked based on symptoms of photophobia, watering, visual acuity, irritation, congestion, pain and signs like decrease in congestion, size of lesion, depth of lesion, adhesions between the surfaces if any.

The study was approved by the Institutional Review Board (IRB) and Ethics Committee at Assam Medical College and Hospital, Dibrugarh. Informed consent was obtained from all subjects and the study was conducted as per the tenets of declaration of Helsinki.

III. Results

In this study we have considered each ocular surface disease (OCD) of the eye as a 'single' case, while OCD of both eyes is counted as two cases.

Age and sex distribution

Out of 45 cases included in this study, 19 (42.2%) were male, and 26 (57.8%) were female. Average age of presentation was 37.4 years.

Frequency distribution – Based on diagnosis

Pterygium accounted for maximum of 24 cases (53.3%) while chemical injury 11(24.5%), Symblepharon 5(11.2%), PED 2(4.4%), Thermal burns 2(4.4%) & Mucocutaneous disorders 1(2.2%) constituted the rest.

Outcome of different ocular surface disorders

Amniotic membrane transplantation was successful in 38 (84.4%) cases while it was a failure in 7 (15.6%) cases. (Table 1)

Table 1.Diagnosis outcome – cross tabulation

Cause of ocular surface disease	Outcome		Total
	Success	Failure	
Chemical injury	9(81.8%)	2(18.2%)	11
Pterygium	22(91.67%)	2(8.33%)	24
Symblepharon	4(80%)	1(20%)	5
Persistent corneal epithelial defects	1(50%)	1(50%)	2
Thermal burns	2(100%)	0	2
Stevens – Johnson's syndrome & other mucocutaneous disorders	0	1(100%)	1

Chemical burns

Eleven patients presented in early stages after chemical injury to our hospital ranging from few hours to 1 week & in all these cases surgery was done within two weeks period depending on requirements of each case. Most of the cases in our series were caused due to alkali injuries-8(72.73%) while 3(27.27%) due to acid injuries. They were further classified according to Dua's classification. 100% of acid injury cases were of Grade III according to Dua's classification. In case of alkali injuries 50% were of Grade V, 37.5% of Grade IV & 12.5% of Grade III.

Visual acuity (VA) at presentation ranged between PL+ve to CF at 2 meters (M). Poor vision in acute cases was due to epithelial defect & stromal edema. When compared to VA at presentation there was a definite improvement in VA at final follow up. Only 18% had poor vision (PL+) when compared to 63% at presentation. Whereas no patient had VA better than CF 2 M at presentation, about 54 % had VA of 6/18 or better at 6 months follow up. The overlay technique of AMT was used in all the patients. AM disintegrated on its own in all the patients at an average of 2 to 3 weeks.

Primary progressive pterygium & recurrent pterygium

21 cases of primary progressive pterygium were treated with excision and AMT. The mean healing period was around 4-6 weeks & there were no post-operative complications .The graft was taken up well in all the cases (100%); however recurrences were observed in 2(9.52%) cases at 6 months follow up. Vascularization was seen in 3 (14.29%) cases while there was complete re-epithelialization at final follow up.

In the present study we had 3 eyes of recurrent pterygium. They underwent pterygium excision with AMT & conjunctival autograft. All the cases (100%) accepted the graft well with no recurrence on 6 months follow up.

Symblepharon

In case of traumatic symblepharon, release of symblepharon and AMT was done which showed complete acceptance of graft with no recurrence. In cases of symblepharon due to chemical injury etiology, release of symblepharon and AMT was done, however in subsequent follow ups there was recurrence of symblepharon amounting to failure. Successful fornicial reconstruction and symblepharon release was done in two cases of chronic cicatricial conjunctivitis with restoration of ocular motility .There were no signs of recurrence.

Persistent epithelial defect

Two cases of persistent epithelial defect was treated with amniotic membrane and were followed up for 24 weeks along with lubrication therapy. No recurrence of the defect was observed in the case secondary to alkali burn, however, second case secondary to Mooren's ulcer showed superficial vascularization with recurrence of epithelial defects amounting to failure of the graft.

Thermal burns

Two cases of thermal injury were included in this study. Conjunctival adhesions were broken down using a glass rod. Debridement of devitalized tissue was done with AMT. It was successful in terms of symptomatic relief, re-epithelialization, decreased inflammation and decreased vascularity. (Table 2)

Table 2. Outcome of thermal burns

Sr. no	Cause	Treatment	Visual acuity				Outcome
			Day1	1st week	1st month	6th month	
1	Firework injury	AMT	CFCF	6/24	6/12	6/9	Success
2	Welding arc injury	AMT	CFCF	6/36	6/12	6/6	Success

Mucocutaneous disorder

One case of Ocular cicatricial pemphigoid was treated with AMT which was successful in terms of decreased inflammation and decreased vascularity at 2 months follow up. But there was recurrence of symblepharon at 6 months follow up.

Complications

They were reported in 9(20%) cases. Early disintegration of the membrane was seen in 3 (6.67%) cases; in 2 cases following grade 5 chemical injury and one case of pterygium. Loose sutures were seen in 3 (6.67%) cases of pterygium which were resutured on the 1st postoperative day. 2(4.44%) cases had hemorrhage under the membrane which disappeared on 1st week follow up.one (2.22%) case of PED following mooren's ulcer showed incomplete epithelialization which resulted in graft failure.

Volume : 5 | Issue : 4 | April 2016 • ISSN No 2277 - 8179 | IF : 3.508 | IC Value : 69.48

Table 3. Post-operative complications of amniotic membrane grafting

Sr.no	Complications	No. of cases affected
1.	loose/broken sutures	3
2.	Haemorrhage under the membrane	2
3.	Infection	0
4.	early disintegration of the membrane	3
5.	Incomplete epithelialization	1

IV.Discussion

This study was an effort to treat cases with the use of amniotic membrane grafting in a setting where there was ample supply of fresh amniotic membrane but which lacked proper harvesting and storage facilities in an endeavour to introduce an old technique with relatively newer interests. With the objective of determining the efficacy of AMT in ocular surface disorders, the above study was conducted and the following outcomes do support the use of fresh AM as the graft material.

Results of this present study are comparable with Ricardo J R et .al (2009) , who studied ten eyes with chemical ocular burn & Jose Alvaro Perira Gomes et al. (2003), who performed AMT for 20 patients with limbal stem cell deficiency secondary to ocular chemical injury.[9,10] Significant improvement was observed in all cases after surgery, except for 2 eyes that maintained pre-operative visual acuity .Failure in these eyes is mainly due to deficient limbal stem cells of >1/3 of limbus. It may be related to the elaboration of collagenases as a result of inflammation that can cause early degradation of the amniotic membrane before the epithelialisation is complete. AMT seems to be an effective adjuvant for ocular surface reconstruction in chemical burns. It promotes epithelisation and suppress inflammation and thereby prepares the eye for future penetrating keratoplasty, with less chances of corneal graft failure due to reduced inflammation.

Multiple surgical approaches have been used to treat pterygium. Although conjunctival autograft is considered to be the most efficient, AMT appears to be a reasonable option in cases with diffuse conjunctival involvement and patients in whom bulbar conjunctiva must be preserved for a prospective glaucoma filtering procedure. Amniotic membrane transplantation after pterygium excision is a safe and effective procedure with no significant postoperative complication. In present study, graft was taken up well in 100% cases while superficial vascularisation was seen in 14.29% cases at final follow up. There is a very good correlation in the success rate of present study (90.48%) and the study of Pinnita et al. (89.1%), demonstrating the efficacy of AMT in primary progressive & recurrent pterygium. [11]

There was complete epithelialization after release & covering the resultant bare area with amniotic membrane in all cases of symblepharon. However recurrence was noted in one case, which was secondary to alkali burn grade V. Amniotic membrane is a very useful adjunct for fornicial reconstruction after symblepharon release. Severe grades of chemical burn result in increased chances of recurrence due to exaggerated cicatrisation. According to Panda et al. AMT was helpful in symblepharon with a success rate of 80%, smooth ocular surface and free movements.[12] The results are comparable with our present study.

In patients presenting with persistent epithelial defects (PED), the results showed no recurrence of epithelial defects arising secondary to alkali burn. However PED's secondary to Mooren's ulcer showed recurrence in late stages. Patient with OCP showed good results after AMT in terms of decreased inflammation, vascularity and increased patient comfort initially. However recurrences in these cases occurred due to continuous inflammatory response in the body. In a study, Barabino et al. (2003) demonstrated reduced conjunctival inflammation with complete epithelialisation. However at 6 month follow up there was recurrence of symblepharon formation with conjunctival inflammation.[13] AMT can be a first-step procedure for ocular surface reconstruction in OCP, but its effectiveness deteriorates slightly over time.

AMT is not a surgery that will work in every ocular surface disease. The success of AMT can be limited by other factors that definitely affect the host wound healing response. Hence, several limiting factors have been recognized, and they include inflammation that has passed the acute stage and has become relentless, and ischemia where no viable cells can reside to flourish. Furthermore, if the host cells are intrinsically abnormal, for example during the loss of epithelial stem cells in the setting of LSCD or squamous metaplasia, AMT alone cannot achieve successful reconstruction, without transplanting healthy epithelial or mesenchymal progenitors. Fresh AM is important for ocular surface reconstruction in hospitals where there is no banked frozen tissue and in that where cost and availability may prohibit the use of the preserved tissue.

V. Acknowledgement

1. Financial assistance was provided by DBT under BT/Med/15/Vision-NER/2011 for this research work.

2. References

3. Sangwan VS, Tseng SCG. New Perspectives in ocular surface disorders. An integrated approach for diagnosis and management. Indian J Ophthalmol 2001; 49:153-68.
4. Role of amniotic membrane transplantation in ocular surface disorders-Arya SK, Bhala S, Malik A, Sood S Department of Ophthalmology, Government Medical College and Hospital, Chandigarh, India. Arya S K et al Nep J Oph 2010;2 (4) :145-153.
5. De Roth. Plastic repair of conjunctival defects with fetal membrane. Arch Ophthalmol.1940; 23:522-525.
6. Kim JC, Tseng SC. Transplantation of preserved human amniotic membrane for surface reconstruction in severely damaged rabbit corneas. Cornea 1995;14:473-484.
7. Honavar SG, Bansal AK, Sangwan VS, Rao GN. Amniotic membrane transplantation for ocular surface reconstruction in Steven Johnson syndrome. Ophthalmology. 2000;107:975-979.
8. Method of Obtaining and Preparation of Fresh Human Amniotic Membrane for Clinical Use; Mohammad Ashraf Ganatra, Khalid M. Durrani (Department of Plastic, Reconstructive Surgery and Bums, Dow Medical College and Civil Hospital, Karachi.) June, 1996 JPMA.
9. Amniotic Membrane Transplantation; Baradaran-Rafii et al ;Iranian journal of ophthalmic research 2007; vol. 2, no. 1.
10. Salman AG (2010) Value of Fresh Amniotic Membrane Graft in Management of Resistant Non Infected Corneal Ulcer. J Clinic Experiment Ophthalmol 1:108. doi:10.4172/2155-9570.1000108.
11. Ricardo J R et .al Amniotic membrane transplantation for severe acute cases of chemical ocular burn and stevens-johnson syndrome. Arq bras oftalmol. 2009 mar-apr;72(2):215 20.
12. . Gomes JAP, Santos MS, Cunha MC, et al. Amniotic membrane transplantation for partial and total limbal stem cell deficiency secondary to chemical burn. Ophthalmology.2003; 110:466-473.
13. Prabhasawat Pinnita. Comparison of conjunctival autografts, Amniotic membrane grafts, and primary closure for pterygium excision.Ophthalmology1997; 104:9.
14. Panda A. Amniotic membrane in ophthalmology AIOC proceedings. 2002.
15. Barabino S, Rolando M, Bentivoglio G, et al. Role of amniotic membrane transplant for conjunctival reconstruction in ocular cicatricial pemphigoid. Ophthalmology. 2003;110:474–480.