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Research Article

EFFECTIVENESS AND SAFETY OF AMNIOTIC MEMBRANE GRAFTING FOR CORNEAL SURFACE DISORDER: A RANDOMIZED CLINICAL STUDY

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ABSTRACT

Objective: The aim of the study was to evaluate the effectiveness and safety of freeze-dried amniotic membrane grafting (AMG) and compare it against traditional medical therapy for the management of corneal surface disorders.

Methods: A randomized clinical trial was conducted on 60 patients with corneal surface disorders who were randomized equally either into the AMG group (n=30) or medical management group (n=30). Patients in both groups were followed up for 8 weeks after receiving group-specified intervention or till complete resolution (whichever was earlier). Treatment outcome, healing time, improved vision, decreased pain, recurrence rate, and corneal clarity were the main outcome metrics.

Results: The most common corneal disorder was persistent epithelial defect (38.33%), closely followed by impending perforated corneal ulcer (31.66%). Majority of the patients (36.66%) had corneal involvement of >75%, while 31.66% had involvement ranging from 50 to 75%. The success rate of AMG (96.7%) was significantly higher than that of medical management (76.7%) in the treatment of corneal surface disorders (p=0.022). During the follow-up period, the incidence of allergic reactions was slightly lower in the AMG group (6.7%) compared to the medical management group (10.0%). Overall, the incidence of side effects and other complications was low in both the AMG group and the medical management group. The severity of side effects was comparable in both the study groups (p=0.886). The recurrence rate of corneal surface disorders in the in the AMG group (6.7%) was statistically lower than that in the medical management group (26.7%) (p=0.037).

Conclusion: The findings of the present study supported the use of AMG as an effective and safe alternative to medical management for the treatment of conditions affecting the corneal surface.

Keywords: Amniotic membrane grafting, Corneal surface disorders, Healing time, Visual acuity, Recurrence rate, Corneal clarity, Safety.

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INTRODUCTION

Corneal surface disorders, such as corneal ulcers, epithelial abnormalities, and recurring erosions, cause significant patient discomfort and visual impairment [1]. The goal of treating corneal surface disorders is to reduce patient pain and corneal opacity to improve vision. Traditional therapies are less likely to succeed in chronic instances due to the avascular nature of the cornea, which also limits the disease's ability to spread [2]. In such cases, the purpose of treatment is to alleviate the symptoms, while safeguarding vision of the patients. Although corneal grafting is a therapy for blindness, it has limitations such as donor availability and tissue rejection [3].

Human amniotic membrane transplantation (AMT) is an evolving technique for ocular surface restoration and protection [4]. It not only promoted epithelialization but also reduces inflammation, vascularization, scarring, discomfort, tissue adhesion, and apoptosis while preserving the normal epithelial phenotype [5]. The amniotic membrane functions by increasing the life span of epithelial progenitor cells and retaining their ability to replicate [6]. When handled and stored appropriately, the amniotic membrane can be employed as a graft to repair damaged ocular surface stromal matrix and eliminate unwanted inflammatory responses [7]. The use of AMT in ophthalmology is increasing, and it has emerged as a rewarding procedure in reconstructive surgery. With its ophthalmic uses, AMT has considerably improved the management of devastating ocular surface disorders [8].

The present study was undertaken to evaluate the effectiveness and safety of freeze-dried amniotic membrane grafting (AMG) and compare it against traditional medical therapy for the management of corneal surface disorders.

Aim and objectives

Aim

The aim of the study was to evaluate and compare the effectiveness and safety of AMG and medical management in the patients with corneal surface disorders.

Objectives

The objectives of the study are as follows:

i. To compare the incidence of side effects between the AMG group and the medical management group.
ii. To compare the recurrence rate of corneal surface disorders between the AMG group and the medical management group.

METHODS

The present study was a randomized clinical trial conducted in the Department of Ophthalmology, in a tertiary care hospital of North India. The study included a total of 60 consenting patients with corneal surface disorders, who were randomly assigned to either the study group (AMG) or the control group (medical therapy). Randomization of patients into either group was performed using computer-generated random number tables. All participants were provided with written informed consent before enrollment. The ethical approval was taken from the Institutional Ethics Committee (IEC), before the study. The

identity of patients and their data were kept confidential. The study was conducted in accordance with the principles outlined in the Declaration of Helsinki and other relevant ethical guidelines.

Inclusion criteria

The following criteria were included in the study:

i. Patients diagnosed with corneal surface disorders, such as corneal ulcers, epithelial defects, or recurrent erosions
ii. Age between 18 and 80 years
iii. Patients who give informed consent to participate in the study
iv. Patients with stable corneal surface disease and no active ocular infection.

Exclusion criteria

The following criteria were excluded from the study:

i. Patients with history of previous corneal surgery or AMG
ii. Patients with immunocompromised conditions that may affect the healing process
iii. Pregnant or lactating females.

Procedure

Group A: AMG

AMG was used to treat the corneal surface problem in patients in Group A. A dehydrated AMG was applied over the damaged cornea as part of the surgery. The AMGs came from a reputed tissue bank and were handled with sterility during storage. Patients received the usual post-operative treatment after the surgery, which included topical antibacterial and lubricating eye drops.

Group B: control group (medical therapy)

Patients in Group B received routine care for conditions affecting the corneal surface, which frequently included topical antibacterial eye drops, lubricating eye drops, and/or bandage contact lenses. The treating ophthalmologist chose a specific treatment plan based on the individual patient's condition and clinical judgment.

Outcome measures

Patients in both groups were followed up for 8 weeks after receiving group-specified intervention or till complete resolution (whichever was earlier). Treatment outcome, healing time, improved vision, decreased pain, recurrence rate, and corneal clarity were the main outcome metrics.

Complete resolution of corneal lesion with healed epithelium, minimum to absent infiltration, was regarded as successful treatment. Persistence of any of aforementioned signs or excessive thinning of cornea leading to desmatocele formation was labeled as treatment failure.

The length of time between the start of treatment and the cornea's full re-epithelialization was referred to as the healing time. Using the logarithm of the minimum angle of resolution scale, visual acuity improvement was measured. Patients were asked to rate their level of pain using a numeric rating scale, which ranged from 0 (no pain) to 10 (the worst agony possible). By examining the likelihood of corneal surface disorder recurrence during follow-up visits, the recurrence rate was calculated. A grading system was used to assess the clarity of the cornea, with higher scores indicating superior clarity.

Data collection and statistical analysis

Baseline demographic information, including age, gender, and type of corneal surface disorder, was recorded for all participants. Treatment outcome, healing time, visual acuity, pain scores, recurrence rate, and corneal clarity were assessed at predetermined intervals during follow-up visits. Adverse events, such as allergic reactions or infections, were also documented.

The collected data were organized and tabulated in Microsoft Excel 2016 (Microsoft Office 2016 package) and statistical analysis was done using the Statistical Package for the Social Sciences version 23.0 (IBM Corp., Illinois, Chicago). The data were analyzed by appropriate statistical tools and represented by various tables, graphs, diagrams, etc. Descriptive statistics were used to summarize the demographic characteristics of the participants. Continuous variables were presented as mean values with standard deviations, and categorical variables were reported as frequencies and percentages. The independent t-test was used to compare continuous variables between the two groups, depending on the normality of the data distribution. Chi-square test or Fisher's exact test was employed to compare categorical variables. Statistical significance was set at $p<0.05$.

RESULTS

The study included 60 patients with ocular surface abnormalities, 30 of whom were divided into the AMG group and the medical management group. The two groups' demographics, including the distribution of age and gender, were comparable (Table 1).

The most common corneal disorder was persistent epithelial defect (38.33%) closely followed by impending perforated corneal ulcer (31.66%) (Table 2). The distribution of corneal involvement by site in Group A (the AMG group) and Group B (the medical management group) is shown in Table 3. The peripheral, paracentral, and central areas of the cornea are all affected, and their distribution in the two study groups was comparable. Majority of the patients (36.66%) had corneal involvement of >75%, while 31.66% had involvement ranging from 50% to 75% (Table 4).

The pre-operative severity of symptoms in patients with corneal surface abnormalities is shown in Table 5. The symptoms (pain, photophobia, foreign body sensation, watering, redness, discharge, and impaired visual acuity) have been grouped into three severity levels: Mild, moderate, and severe.

Table 1: Demographic characteristics of the study population

Characteristics	Amniotic membrane grafting group (n=30)	Medical management group (n=30)	p-value
Age (years)	52.4±8.6	50.9±9.2	0.769
Gender (male/female)	14/16	16/14	0.606

Table 2: Distribution of case studied

Corneal disorder	No. of patients	Percentage
Persistent epithelial defect	23	38.33
Impending perforated corneal ulcer	19	31.66
Perforated corneal ulcer	13	21.66
Symblepharon due to chemical injury	5	3.33

Table 3: Site of corneal involvement

Location	Group A (amniotic membrane grafting group)	Group B (Medical management group)	p-value
Central	16	18	0.453
Paracentral	12	8	
Peripheral	2	4	
Total	30	30	

Table 4: Extent of corneal involvement

Extent	Number of patients	Percentage
<25%	5	8.33
25–50%	14	23.33
50–75%	19	31.66
>75%	22	36.66

In terms of final treatment outcome, 96.7% of the patients receiving AMG and 76.7% of the patients receiving medical management had treatment success. The success rate of AMG was significantly higher than that of medical management in the treatment of corneal surface disorders (p=0.022) (Table 6).

During the follow-up period, side effects were monitored and recorded for both groups. The incidence of allergic reactions was slightly lower in the AMG group (6.7%) compared to the medical management group (10.0%). Similarly, the incidence of infection was observed in 3.3% of patients in the AMG group and 6.7% in the medical management group. The incidence of side effects was comparable in both the study groups (p>0.05) (Table 7). In addition to the specific side effects, other complications such as delayed healing, foreign body sensation, and corneal haze were also assessed but were found to have negligible occurrences in both groups. Overall, the incidence of side effects and other complications was low in both the AMG group and the medical management group.

According to the grading system, majority of side effects reported in both groups were mild in nature, with only a small percentage classified as moderate. In the AMG group, 13.3% of patients experienced mild side effects, while 3.3% experienced moderate side effects. Furthermore, in the medical management group, 16.7% of patients reported mild side effects, while 3.3% reported moderate side effects. No severe side effects were reported in any of the study groups. The severity of side effects was comparable in both the study groups (p=0.886) (Table 8).

Table 5: Pre-operative severity of symptoms

Symptoms	Mild (%)	Moderate (%)	Severe (%)
Pain	10 (16)	20 (33.33)	30 (50)
Photophobia	8 (13.33)	22 (36.66)	30 (50)
F.B. Sensation	10 (16)	15 (25)	35 (58.33)
Watering	10 (16)	15 (25)	25 (58.33)
Redness	8 (13.33)	25 (41.66)	27 (45)
Discharge	25 (41.66)	15 (25)	20 (33.33)
DOV	5 (8.33)	20 (33.33)	35 (58.33)

Table 6: Treatment outcome

Treatment outcome	Amniotic membrane grafting group (n=30) (%)	Medical management group (n=30) (%)	p-value
Success	29 (96.7)	23 (76.7)	0.022
Failure	1 (3.3)	7 (23.3)	

Table 7: Incidence of side effects

Side effects	Amniotic membrane grafting Group (n=30) (%)	Medical management group (n=30) (%)	p-value
Allergic reactions	2 (6.7)	3 (10.0)	0.933
Infection	1 (3.3)	2 (6.7)	
Epithelial ingrowth	0 (0.0)	1 (3.3)	
Other complications	1 (3.3)	0 (0.0)	

Table 8: Severity grading of side effects

Side effects	Amniotic membrane grafting Group (n=30) (%)	Medical management group (n=30) (%)	p-value
Mild	4 (13.3)	5 (16.7)	0.886
Moderate	1 (3.3)	1 (3.3)	

During the follow-up period, the recurrence of corneal surface disorders was monitored and recorded for both groups. The recurrence rate of corneal surface disorders in the medical management group was higher (26.7%) than that in the AMG group (6.7%); and this difference was statistically significant (p=0.037) (Table 9).

DISCUSSION

The history of occurrence of corneal of corneal disorders dates back to times immemorial and so is the history of their management. The conventional management of corneal disorders is based on the principle of natural wound healing supported by local application of drugs and rest to the eye. However, corneal wound healing defers from other tissues primarily due to its avascularity [9].

Many methods have been tried for the management of corneal disorders not responding to conventional management. These methods include conjunctival flap, human AMG, therapeutic penetrating keratoplasty, vasculoepithelioplasty, and stem cell transplantation [10]. The use of freeze-dried AMG for the management of corneal surface disorders has attracted attention of ophthalmologists in the recent past [11,12].

The present study is the first of its kind to compare the effectiveness and safety of AMG transplantation against conventional medical management among patients with corneal surface disorders. Careful history-taking revealed that the development of corneal surface disorders among most participants was attributed to fall of dust particle in eyes and injury vegetative in origin due to their rural residence and positive history of working in open fields. Furthermore, the presenting clinical picture was almost similar in all the cases. Most of the patients presented with symptomatology that included pain, photophobia, foreign body sensation watering, discharge, redness, and diminution of vision. Mechanical and chemical impact of toxins on exposed nerve terminals cause pain, foreign body sensation, and reflex hyperlacrimation leading to watering of eyes. Corneal haze causes blurry vision [13]. However, the severity of symptoms varied from patient to patient ranging from mild to severe depending upon the extent of corneal involvement. In the majority of cases, symptoms were severe.

The present study observed a 96.7% treatment success rate of AMG transplantation as compared to 76.7% success rate of medical management for the treatment of corneal surface disorders which were statistically significant. Although an exhaustive review of the literature revealed no comparable studies, Meller *et al.* [14]. hypothesized that a healthy corneal epithelium is a key element in ocular surface stability. Simple epithelial defects typically recover without any issues. Corneal ulceration, descemetocele, and even perforation can swiftly develop if they are left untreated or treated inappropriately. In patients with persistent epithelial defects, both those with and those without corneal ulcers, amniotic membrane has been employed clinically as a replacement for the basement membrane [15,16]. Seitz were able to establish epithelial closure with no recurrence in 65% of cases after a median follow-up period of 18 months [15]. Following procedures such as tarsorrhaphy or injecting botox into the upper eyelid might considerably increase the success rate of AMT [15,16].

Our study observed that the incidence and severity of side effects such as allergic reactions, infection, epithelial ingrowth, and complications (delayed healing, foreign body sensation, or corneal haze) were low and comparable in both the study groups. These findings suggest that side

Table 9: Recurrence rates of corneal surface disorders

Recurrence	Amniotic membrane grafting Group (n=30) (%)	Medical management group (n=30) (%)	p-value
Yes	2 (6.7)	8 (26.7)	0.038
No	28 (93.3)	22 (73.3)	

effects and other concerns related to the management of ocular surface diseases are similar between AMG and medical management in terms of safety profiles. In a study by Thatte [17], patients with various corneal ulcers, chemical burns, and bullous keratopathy experienced significant symptomatic improvement (reduced pain and redness) after receiving AMT. Patients with corneal ulcers also saw enhanced epithelialization and stromal repair. According to Lee and Tseng [18], AMT can enhance visual acuity by restoring the corneal surface as well as increasing corneal transparency. However, a study by Eslami *et al.* [19] found that patients using AMT for the treatment of pterygium had a greater post-operative inflammation rate (28.6%).

In the present study, freeze-dried amnion membrane is processed and preserved (at-800c) form of human amniotic membrane, measuring 5×5 cm in size and 0.02–0.05 mm in thickness. A single layer of cuboidal epithelium, a substantial foundation membrane, and an avascular stromal matrix make up the amniotic membrane, which is loosely linked to the chorion. Amniotic membrane from one placenta can be used in 20–30 transplants of the eye. Amniotic membrane can also be simply kept. According to Shimazaki *et al.* [20], the amniotic membrane's epithelium can endure cryopreservation for up to 70 days. The stromal side of the membrane has a special matrix component that inhibits TGF-B signaling, as well as normal human corneal and limbal fibroblast proliferation and myofibroblast development, as well as normal conjunctival fibroblasts. This process explains how AMT lessens corneal haze after phototherapeutic keratectomy and photorefractive keratectomy, prevents recurrent scarring after pterygium excision, and minimizes scar formation during conjunctival surface reconstruction. In this context, systemic immunosuppressive medications are not necessary in AMT because the amnion's epithelium does not express HLA class I or class II antigens [21].

From our study, the recurrence rate of corneal surface disorders in the AMG group (6.7%) was significantly lower than that in the medical management group (26.7%). These findings suggest that AMG is more effective in reducing the recurrence of corneal surface disorders compared to medical management. The lower recurrence rate in the AMG group highlights the potential of this intervention as a valuable treatment option for patients with corneal surface disorders.

Despite all of the advantages of AMG, it must be kept in mind that the graft is a biological-derived material and with certain drawback in practical application. There is always a danger of infectious disease transmission when transplanting human organs and tissues. Therefore, the use of AM must follow the same safety guidelines and procedures as organ transplantation. Effective screening must be done on potential donors to identify any risk factors that would make them ineligible for donation. It is recommended to do a review of pertinent medical records to check for risk factors for and clinical evidence of HIV, hepatitis B, hepatitis C, CMV, syphilis, and other potential illnesses. There also remains a chance that the donor is in the "window period" of infection. The AM can be stored at −80°C until samples are proven to be free of infectious illnesses [22].

CONCLUSION

The findings of the present study support the use of AMG as an effective and safe alternative to medical management along for the treatment of conditions affecting the corneal surface. However, further investigations with larger sample size are warranted to validate our study observations and evaluate the specific nature and severity of the observed side effects and complications. Nevertheless, AMG stimulates the healing process, encourages re-epithelialization, and improves symptoms. In addition, it preserves corneal moisture and offers barrier defense. There is no need for intraocular intervention or immunosuppression, and the process is simple and repeatable. AMG is a useful substitution for standard care and is successful in treating recurrent illnesses and corneal perforations.

AUTHORS' CONTRIBUTIONS

All authors contributed equally to the conceptualization, supervision, analysis, and writing of the manuscript.

CONFLICTS OF INTEREST

The authors have no conflicts of interest to declare.

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