International Journal of Science, Environment and Technology, Vol. 9, No 4, 2020, 666 – 670

ISSN 2278-3687 (O)
2277-663X (P)

DECELLULARIZED BOVINE OMENTUM IN COMBINATION WITH MITOMYCIN-C FOR SURGICAL MANAGEMENT OF CORNEAL ULCERATION IN A FRENCH BULLDOG

A.S. Thajunnisa[1], S. Anoop[2], L.M. Philip[3], K.M. Dileepkumar[4], V.N. Vasudevan[5], K.A. Lathief[6] and C.B. Devanand[7]

[1]MVSc Scholar, Department of Veterinary Surgery and Radiology, College of Veterinary and Animal Sciences, Mannuthy - 680651, Thrissur, Kerala, [2]Associate Professor, [3]Assistant Professor, [7]Professor and Head, Department of Veterinary Surgery and Radiology; [5]Associate Professor, Department of Livestock Products Technology, College of Veterinary and Animal Sciences, Kerala Veterinary and Animal Sciences University, Mannuthy-680651, Kerala; [4,6]Veterinary Surgeon, Animal Husbandry Department, Government of Kerala
E-mail: asthajunnisa@gmail.com

Abstract: An extensive full thickness corneal defect in a French Bulldog reconstructed using decellularized and gamma irradiated bovine omentum as an extra-cellular matrix scaffold could acheive enhanced corneal healing and excellent visual outcome. Intra-operative topical application of Mitomycin-C effectively controlled corneal fibrous metaplasia thus minimizing corneal scarring. The results of the present study gives promising outcome when omental graft combined with MMC is used for clinical application.

Keywords: ulcerative keratitis, corneal surface reconstruction, extra cellular matrix scaffold, bovine omentum, Mitomycin-C.

Introduction

Ulcerative corneal lesions are common and important ocular affections in dogs. Prompt and effective repair of large-diameter or full thickness corneal defects is desirable to avoid corneal perforation and restoration of vision. Several techniques for the surgical management of canine corneal ulcers have been developed through clinical trials including reconstruction with biomaterials. Decellularized and gamma irradiated bovine omentum is a relatively new biomaterial with preserved three diamentional network of collagen having the potential for recellularization (Porzionato *et al*., 2013). Single time intra-operative application of 0.02% Mitomycin-C (MMC) to the canine corneal keratocytes was found effective in controlling canine corneal fibrous metaplasia and thus reducing corneal scarring (Gupta *et al.*, 2011). Here, we report a case of full thickness corneal defect reconstructed with bovine omentum as an extra cellular matrix scaffold along with intra-operative topical application of MMC.

*Received July 19, 2020 * Published Aug 2, 2020 * www.ijset.net*

Case history and observations

A 10 month old female French Bulldog was presented to the Teaching Veterinary Clinical Complex, Mannuthy with the history of traumatic corneal injury in its left eye. A detailed clinical and ophthalmic examination was performed to evaluate the type and extent of the lesion and to assess the visual functions of the animal. Thorough ophthalmic examination of animal with bilateral evaluation of menace response, palpebral reflex, dazzle reflex and pupillary light reflex (PLR) revealed visual impairment. The dense and extensive corneal oedema negatively affected visualization of anterior chamber. Tear production measured using Schirmer Tear Test (STT) was 24 mm/min and the Fluorescent Dye Test (FDT) was positive on the day of presentation. The diameter of the lesion was measured as 9 mm and the area affected was calculated as 63.58 mm^2. The cornea was examined by direct and indirect ophthalmoscopy and slit-lamp biomicroscopy before and after fluorescein staining and diagnosed as a full thickness defect with prolapse of corneal endothelium (Fig.1).

Treatment and discussions

The animal was premediated with atropine sulphate (@ 0.045 mg/kg body weight) and xylazine hydrochloride (@ 1mg/kg body weight) administered intramuscularly and general anaesthesia was induced with ketamine hydrochloride (@ 5 mg/kg body weight) and midazolam (@ 0.1 mg/kg body weight) administered intramuscularly. Anaesthesia was further maintained with 2% of isoflurane after intubation. Disinfection of the peri-ocular region was done with 5% povidone iodine solution. The decellularized bovine omental transplant was prepared so that its size was consistently 1-2 mm larger than the area of lost corneal substance. The size-matched graft was hydrated in sterile normal saline for not less than 5 minutes before the procedure. Injection of Mitomycin-C (MMC) available as 2 mg per vial in powder form was reconstituted with 10 ml normal saline to make 0.02% (0.2 mg/ml) solution.

The superficial cornea was debrided to remove loose epithelium and tissue debris using a sterile cotton tip applicator. Corneal scissors was used to excise the non-viable epithelial tissue. Single time intra-operative application of 0.02 % MMC was carried out with a sterile cotton swab, retained for 2 minutes and thoroughly washed off with normal saline solution before placement of the graft. The size matched rehydrated transplant was sutured micro-surgically to the graft bed using absorbable monofilament suture material (Polycryl, Poly Glycolic Acid 910, size 10/0, Aurolab) after positioning the graft with simple interrupted sutures at four cardinal points of the corneal defect, and completed the suturing with 6 more

sutures in between (Fig.2). Liberal irrigation with normal saline was done throughout the procedure to avoid drying. Temporary tarsorrhaphy was performed and retained for a period of one week and an Elizabethan collar was advised to prevent self-mutilation. Systemic antibiotic therapy with Cefpodoxime @ 10 mg/kg once daily for 5 days and topical moxifloxacin (1drop TID) through the space at the medial canthus of eye were followed post-operatively.

Observations were serially recorded on day 7, 14, 21 and 60 post-operatively. FDT was negative on day 7 post-grafting indicating complete epithelalization of cornea. The diffuse corneal oedema present on the day of presentation and on the day of grafting were confined to the central cornea by day 7 and resolved completely by day 14 post-grafting. Neovascularization of cornea became intense by day 7 with multiple prominent blood vessels encroaching the central cornea (Fig.3). The blood vessels were less prominent on day 14 (Fig.4) and regressed by day 21 (Fig.5) and disappeared completely by day 60 leaving a ghost vessel at the center (Fig.6). Corneal scarring and pigmentation were completely absent resulting in a near normal clear cornea with positive visual function by day 60.

Simple uncomplicated ulcers typically epithelialize with medical therapy whereas immediate surgical interventions are indicated in full thickness defects. Several grafting techniques have been described with varying efficacy for the replacement of the lost corneal substance. This includes lamellar corneal graft, corneo-scleral transposition, penetrating keratolplasty, conjuctival grafts (Island, pedicle, bulbar, bridge, advancement or complete bulbar graft) (Hendrix, 2007) as well as synthetic grafts and biomaterial grafts (Goulle, 2012). Biomaterials used for corneal reconstruction included amniotic membrane (Barros, 1998), porcine small intestinal submucosa (Bussiers *et al.*, 2004 and Goulle, 2012), porcine urinary bladder submucosa (Chow and Westermeyer, 2016) and porcine cholecyst (Suhas, 2015).

Decellularized and gamma irradiated bovine omentum is a novel biological scaffold that can be used for corneal surface reconstruction. Preservation of three-dimentional architecture of connective, elastic and reticular fibers and glycosaminoglycans potentiates recellularization and expected to enhance corneal healing (Porzionato *et al*., 2013). Trans-differentiation of keratocytes to myofibrobasts is essential during corneal healing but lead to corneal fibrosis. The cytotoxic and anti-mitotic effects of mitomycin-C is beneficial in checking uncontrolled myofibroblast proliferation and thus corneal scarring (Netto *et al*. 2006). The results of the present study proves efficacy of decellularized bovine omentum as an extra cellular matrix scaffold to enhance corneal healing and MMC to control corneal fibrosis.

Acknowledgement

The authors are thankful to the Dean, College of Veterinary and Animal Sciences for extending all the facilities for the conduct of the study.

References

1] Barros, P.S.M. 1998. The use of xenologous amniotic membrane to repair canine corneal perforation created by penetrating keratectomy. *Vet. Ophthalmol*. 1: 119- 123.

2] Bussieres, M., Krohne, S. G., Stiles, J. and Townsend, W. M. 2004. The use of porcine small intestinal submucosa for the repair of full-thickness corneal defects in dogs, cats and horses. *Vet. Ophthalmol*. 7(5): 352-359.

3] Chow, D.W.Y. and Westermeyer, H.D. 2016. Retrospective evaluation of corneal reconstruction using ACell Vet alone in dogs and cats: 82 cases. *Vet. Ophthalmol*. **19 (5)**: 357-366.

4] Goulle, F. 2012. Use of porcine small intestinal submucosa for corneal reconstruction in dogs and cats: 106 cases. *J. Small Anim. Pract*. **53**: 34-43.

5] Gupta,R.,Yarnall, B.W., Giuliano, E.M., Kanwar, J.R., Dylan, G.B. and Mohan, R.R. 2011. Mitomycin-C, a promising agent for the treatment of canine corneal scarring. *Vet. Ohthalmol*. **14**(5): 304- 312.

6] Hendrix, D.V.H. 2007. Canine conjunctiva and nictitating membrane. In: Gelatt, K.N. (ed.). *Vet. Ophthalmol.* (4th Ed.). Wiley- Blackwell, pp. 662-689.

7] Netto, M.V., Mohan, R.R. and Sinha, S. 2006. Effect of prophylactic and therapeutic mitomycin-C on corneal apoptosis, cellular proliferation, haze and long term keratocyte density in rabbits. *J. of Refractive Surg*. **22**: 562 – 574.

8] Porzionato, A., Sfriso, M.M., Macchi, V., Rambaldo, A., Lago, G., Lancerotto, L., Vindigni, V. and De Caro, R. 2013. Decellularized omentum as novel biologic scaffold for reconstructive surgery and regenerative medicine..*Eur J.Histochem*; **57**(1): e4.

9] Suhas, K.P. 2015. Evaluation of porcine cholecyst derived collagen scaffold for the treatment of corneal injuries in dogs. *MVSc. Thesis*. Kerala Veterinary and Animal Sciences University, Pookode. 116p.

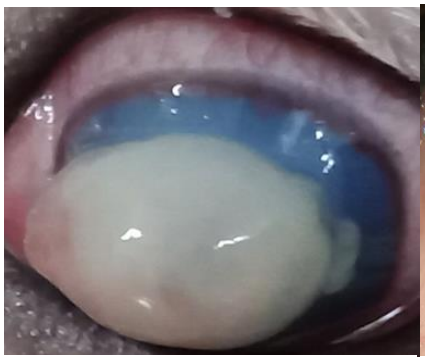

Fig1: Full thickness corneal defect with prolapsed edothelium

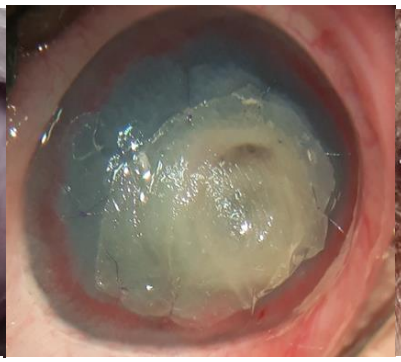

Fig 2: Corneal defect reconstructed with omental graft

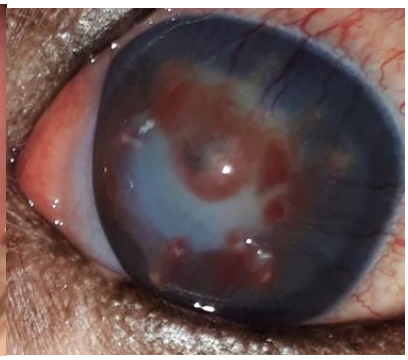

Fig 3: Day 7 post grafting with intense neovascularization

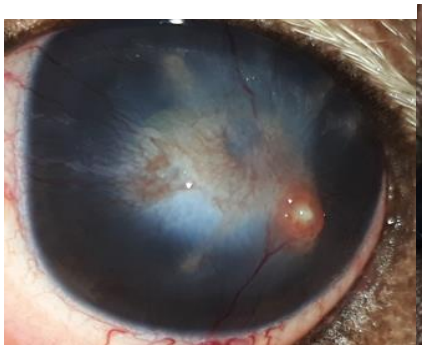

Fig 4: Day 14 post grafting with regressing vasculature and marked corneal fibrosis

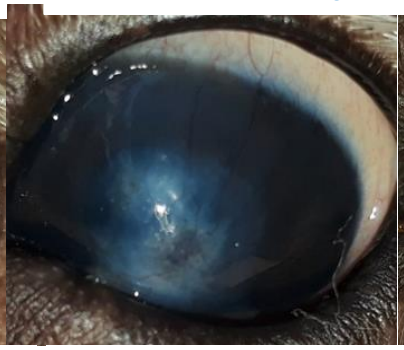

Fig 5: Day 21 post-grafting with faint corneal scar

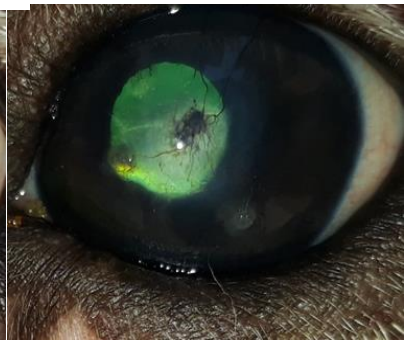

Fig 6: Day 60 post-grafting showing good corneal clarity and ghost vasculature