

# Porcine pericardium as glaucoma implant tube coverage - an experimental study

*Uso de pericárdio suíno no recobrimento de implantes de drenagem de glaucoma - um estudo experimental*

Luciano Moreira Pinto<sup>1</sup>  
 Caio Vinicius Saito Regatieri<sup>2</sup>  
 Ivan Maynard Tavares<sup>3</sup>  
 Moacyr Pezati Rigueiro<sup>4</sup>

## ABSTRACT

**Purpose:** To evaluate the inflammatory response associated with the use of processed porcine pericardium and glycerin-preserved homologous sclera as silicone glaucoma drainage device coverage in a rabbit experimental model. **Methods:** Eight New Zealand white rabbits' eyes received an equal-sized glycerin-preserved homologous scleral patch or a double-layered processed porcine pericardium that was sutured to the bare sclera covering a silicone tube. Conjunctival hyperemia was graded using the double-blind method during the immediate postoperative period and during the first, third, and seventh postoperative weeks. After the seventh week, the enucleated eyes were histopathologically examined. They were also evaluated for signs of patch graft melting, tube erosion and chemosis. **Results:** There was no occurrence of graft melting or tube exposure, although porcine pericardium was associated with greater inflammation through clinical observation. Light microscopy revealed marked inflammation surrounding the porcine pericardium with foreign body granuloma formation. On the other hand, the sclera group presented milder inflammation with foreign body granulomas only around the sutures. **Conclusions:** Porcine pericardium is associated with significant inflammation when used as tube coverage in a rabbit model, at both histopathologic and clinical levels, when compared to glycerin-preserved homologous sclera during the seven-week follow-up period.

**Keywords:** Transplantation, heterologous; Pericardium; Glaucoma drainage implants; Glaucoma; Swine

## INTRODUCTION

Glaucoma drainage devices (GDD) are a useful modality for treating complicated glaucomas and also in cases where other treatments have failed. These devices can be inserted into the anterior chamber through the limbal tissues or via pars plana into the vitreous cavity. Erosion of both the conjunctiva and sclera above a GDD occasionally occur. In order to avoid this sort of complication, insertion into the anterior chamber under a partially thick scleral flap was initially tried. However, there was a propensity to intraocular tube rotation and consequent corneal touch. Later, erosion of the scleral flap and conjunctiva, exposing the tube, as well as internal erosion were observed<sup>(1-3)</sup>. To decrease the risk of late exposure through the conjunctiva, free hand scleral grafts of glycerin-preserved donor sclera patches were used to cover the anterior tube of the drainage devices<sup>(4)</sup>. Donor sclera has been used quite extensively for this

Trabalho realizado no Departamento de Oftalmologia da Universidade Federal de São Paulo - UNIFESP - São Paulo (SP) - Brasil.

<sup>1</sup> Oftalmologista do Departamento de Oftalmologia da Universidade Federal de São Paulo - UNIFESP - São Paulo (SP) - Brasil.

<sup>2</sup> Residente do Departamento de Oftalmologia da UNIFESP - São Paulo (SP) - Brasil.

<sup>3</sup> Professor Afiliado do Departamento de Oftalmologia e Chefe do Setor de Glaucoma da UNIFESP - São Paulo (SP) - Brasil.

<sup>4</sup> Professor Adjunto do Departamento de Patologia da UNIFESP - São Paulo (SP) - Brasil.

**Endereço para correspondência:** Luciano Moreira Pinto, Rua Botucatu, 822 - São Paulo (SP) CEP 04023-062 E-mail: [luciano@oftalmo.epm.br](mailto:luciano@oftalmo.epm.br)

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purpose even though its availability is somewhat limited in some regions. Dependence on eye bank supplies precludes its use in emergency cases and increases its cost where a local eye bank is not available.

Another issue is that sclera variability in thickness and handling characteristics can cause surface problems, such as delamination, when a thick graft is sutured to the sclera close to the limbus, or tube erosion when thin scleral grafts are used. Evidences of HIV in donor sclera were found after treatment with heat, alcohol and formalin<sup>(5)</sup>. Because many eye banks preserve scleral grafts in glycerin or alcohol and neither of these solutions were sterile, infectious disease transmission such as human immunodeficiency virus (HIV) remains a possibility despite donor screening<sup>(6)</sup>. Immunologic reaction in the recipient leading to graft melting months to years after the procedure has also been described<sup>(7)</sup>.

Alternative materials have been suggested, such as, dehydrated human dura mater<sup>(7)</sup>, clear corneal graft<sup>(8)</sup>, fascia lata<sup>(9)</sup>, and human pericardium<sup>(10)</sup>. Bovine pericardial patch grafts have also been used in ophthalmologic surgery with varying results. A recent prospective study showed that bovine pericardium is a safe wrapping material for hydroxyapatite orbital implants in patients undergoing enucleation during uveal melanoma<sup>(11)</sup>. Bidar et al.<sup>(12)</sup> reported 18 cases of conjunctival defects in patients who had undergone orbital implantation of hydroxyapatite implants wrapped in bovine pericardium. The transmission of the agent responsible for transmissible spongiform encephalopathy (TSE) is an issue when using bovine pericardium<sup>(13)</sup> or donor sclera<sup>(14)</sup>. However, most of the iatrogenic transmission cases described were due to human cadaveric growth hormone injections or dura mater transplantation<sup>(15)</sup>. Dura mater also carries the risk of prionic disease transmission, even after inactivation procedures<sup>(16)</sup>. No cases of TSE transmission have been described to date when using porcine pericardium, although there was a recent report of TSE in a porcine dura mater recipient<sup>(17)</sup>.

Porcine and bovine pericardium have been used for cardiac valve replacement for over 30 years<sup>(18-19)</sup> with a low incidence of valve-related complications and high durability<sup>(20)</sup>. To the best of our knowledge, there is no similar study in the literature evaluating porcine pericardium as silicone tube coverage or its use in any ophthalmologic surgery. The aim of this study was to evaluate the inflammatory response associated with the use of processed porcine pericardium and glycerin-preserved homologous sclera as tube shunt coverage in a rabbit experimental model.

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## METHODS

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All experimental procedures employing animals were carried out according to the Association for Research in Vision and Ophthalmology (ARVO) and Use of Animals in Research. The protocol was reviewed and approved by the Animal Care Committee of the Federal University of São

Paulo. Only one eye from each animal was used in the experiment, avoiding bilateral visual disability, in accordance to The Guide to the Care and Use of Laboratory Animals of the Canadian Council on Animal Care, which is endorsed by ARVO.

The experiment was carried out using eight 1-year-old healthy New Zealand albino rabbits (1-2 kg), with no preexisting ocular inflammation. Each rabbit received a 7.0 x 4.0 mm graft of either glycerin-preserved homologous rabbit sclera (Sclera group) or double-layered porcine pericardium manufactured at Braile Biomedica Ltd, São José do Rio Preto, São Paulo, Brazil (Pericardium group). Graft material was preoperatively washed five times in a balanced saline solution and rinsed for twenty minutes in a gentamycin solution.

After anesthesia with intramuscular ketamine hydrochloride (56 mg/kg) and xylazine hydrochloride (6 mg/kg), the left eye was draped with towels and exposed with a lid speculum. Following the application of local anesthesia with 0.5% proxymetacaine hydrochloride (Alcon, Brazil) eye drops, a fornix-based conjunctival dissection was performed in the superotemporal quadrant. Clear cornea traction with a 7-0 silk suture was used to improve surgical exposure. A 6 mm silicone-rubber tube was sutured to the bare sclera with 10-0 mononylon, and then covered with sclera or double-layered pericardium sutured to the sclera with two interrupted 10-0 nylon sutures. The conjunctiva was sutured to the limbus with two buried 10-0 nylon sutures. Postoperatively, a combination of ciprofloxacin and dexamethasone eye drops was instilled in the inferior fornix.

A clinical examination and photographic records were made using a surgical microscope and a digital camera, respectively, in the immediate postoperative period, and consequently in the first, third, and seventh postoperative weeks. The presence of conjunctival hyperemia, subconjunctival hemorrhage, discharge, suture exposure, patch graft thinning and tube exposure through the graft and conjunctiva were assessed under observation with a microscope. The grading of conjunctival hyperemia was performed by a third observer, applying the double-blind method, using the photographic records. Hyperemia grades vary from 0, indicating absence of hyperemia to 4, indicating intense hyperemia. To compare the groups with regard to conjunctival hyperemia, a nonparametric analysis of ordered categorical data from repeated measurements was used<sup>(21)</sup>.

The animals were euthanized at the seventh postoperative week by a lethal dose of intracardiac KCl 20%, after a large dose of ketamine and xylazine, consistent with the recommendations of the American Veterinary Medical Association (AVMA) Guidelines on Euthanasia. The eyes were enucleated, and the section containing the graft was dissected from each eye. The silicone tube was removed, and the specimens fixed in 10% buffered formalin were sent for histopathologic analysis. Sections of 5 µm were cut and stained with hematoxylin-eosin.

**RESULTS**

There was no occurrence of graft melting, tube exposure, infection or severe inflammation. Mild mucoid discharge occurred in association with suture exposure in one case in the Sclera group. The conjunctival hyperemia was similar in both groups, in the immediate postoperative period ( $p=1.000$ ) and in the first postoperative week ( $p=0.821$ ), but was less severe for the Sclera group in the third and seventh postoperative weeks ( $p=0.001$  and  $0.034$ ; respectively) (Table 1 and Figures 1 and 2).

Light microscopy performed in the seventh postoperative week revealed marked inflammatory reaction surrounding the porcine pericardium with foreign body granuloma formation (Figure 3A). The scleral patch elicited a much milder inflammatory reaction with foreign body granulomas only around the sutures (Figure 3B). Both groups showed epithelial inclusion and initial cyst formation around the graft (Figures 3C and 3D).

**DISCUSSION**

Numerous materials have been used for tube coverage, e.g., autologous graft such as sclera, dura mater and pericardium. Smith et al.<sup>(22)</sup> showed that no material was more prone to melting than another, studying 64 eyes that underwent tube shunt surgery using processed pericardium (23 eyes), processed dura mater (18 eyes), and donor sclera (23 eyes). However sixteen eyes experienced patch thinning without conjunctival defect: six eyes of the sclera group, four eyes of the dura group, and six of the pericardium group. The authors reported no statistical difference in rate of thinning between groups, although the pericardium group had a shorter follow-up (32.6 months). A similar study evaluated 44 eyes that underwent GDD surgery with the use of human processed pericardium through a mean follow-up of  $10.2 \pm 4.0$  months. Five cases of asymptomatic thinning of the pericardial patch graft were reported, with no evidence of conjunctival breakdown or tube extrusion<sup>(10)</sup>.

Despite those two favorable reports, Lama et al.<sup>(23)</sup>

reported two cases of tube erosion in patients without predisposing ocular and systemic factors, four and five months after the surgery, and seven other cases of graft thinning. Yalvac et al.<sup>(24)</sup> described a double-layered donor pericardium sandwich technique as an alternative surgical approach for a patient with glaucoma and severe scleral melting.

Ainsworth et al.<sup>(25)</sup> reported three cases of tube exposure secondary to necrosis of the overlying bovine pericardial

Group	Animal	Immediate PO	1 <sup>st</sup> PO week	3 <sup>rd</sup> PO week	7 <sup>th</sup> PO week
Sclera	Rabbit 1	0	0	0	1
	Rabbit 2	0	2	0	0
	Rabbit 3	0	1	0	0
	Rabbit 4	1	2	0	0
Pericardium	Rabbit 5	0	1	2	1
	Rabbit 6	0	1	1	0
	Rabbit 7	0	2	2	1
	Rabbit 8	1	1	2	2

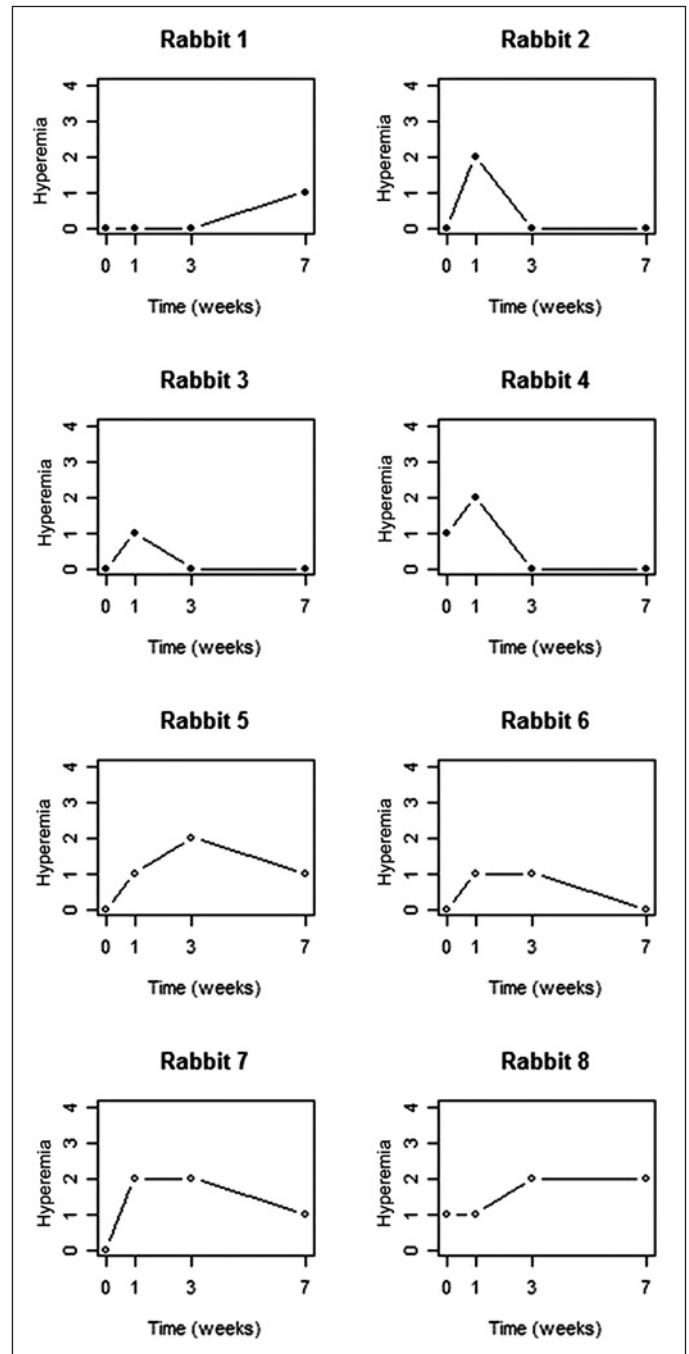


Figure 1 - Conjunctival hyperemia grading by the 7-week postoperative (PO) period (one eye of each animal)



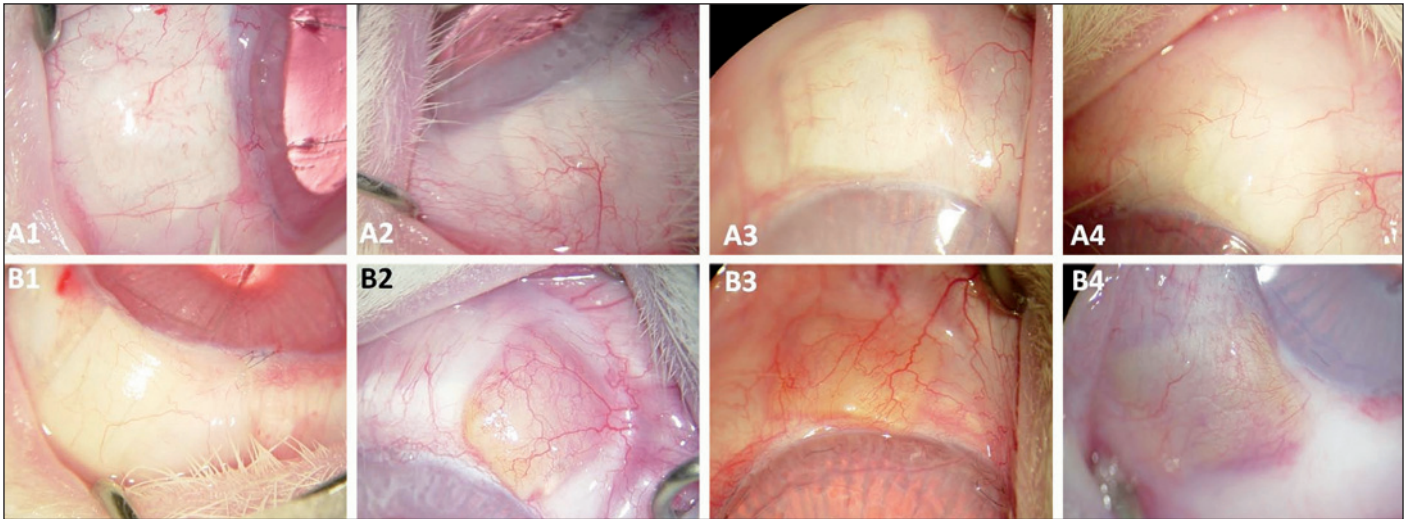


Figure 2 - Clinical aspects of both scleral (A) and pericardium (B) patches in the immediate postoperative period (A1 and B1) and in the first (A2 and B2), third (A3 and B3) and seventh (A4 and B4) postoperative weeks

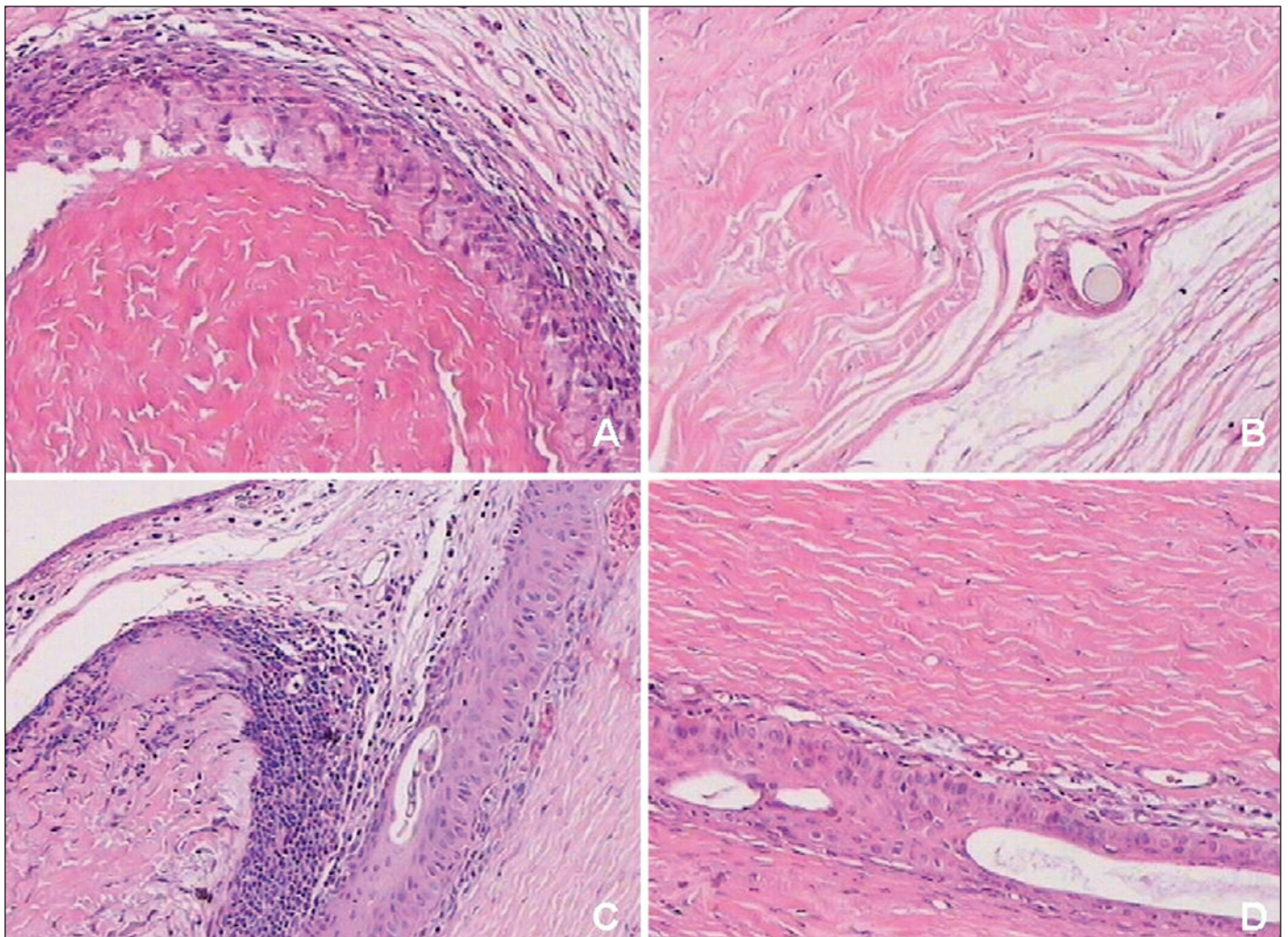


Figure 3 - Histologic sections stained with hematoxylin-eosin. (A) Pericardium patch with intense foreign body granulomatous inflammatory reaction (200x). (B) Scleral patch with foreign body granuloma around the suture (150x). (C) Epithelial inclusion with initial cyst formation around pericardium patch (150x). (D) Epithelial inclusion with initial cyst formation around scleral patch (150x).



patch and conjunctiva that were covered with amniotic membrane. This type of melting of pericardial patch grafts has been previously described<sup>(26)</sup>. In these reports a single piece of pericardium was used. Attempting to avoid this kind of complication, in the present study a double-layered pericardium was used and no cases of tube extrusion were found during the seven-week period of follow-up.

Animal studies on white rabbits, comparing bovine pericardium and homologous sclera as wrapping materials for hydroxyapatite orbital implants<sup>(27)</sup>, as well as a lower eyelid spacer graft<sup>(28)</sup>, showed that both materials were well tolerated at the clinical level, although histopathologically bovine pericardium elicited a more intense inflammatory response. The response consisted of both chronic and granulomatous inflammatory reactions, with the presence of lymphocytes, macrophages, multinucleated giant cells and eosinophils encompassing the circumference of the implant<sup>(28)</sup>. In the present study, the use of porcine pericardium resulted in a similar inflammatory pattern, with a preponderance of lymphocytes and macrophages, as well as foreign body granuloma formation. However, there was also an intense inflammation at the clinical level. Some inflammation could occur in part due to the porcine pericardium graft being preserved in paraformaldehyde, even though the tissue was washed five times with balanced saline solution before insertion.

Advantages of porcine pericardium as patch graft include its proven good performance in a critical site such as the heart<sup>(19-21)</sup>; avoidance of the potential transmission of undetected human infectious agents, such as HIV<sup>(5-6)</sup>, that can potentially occur with homologous materials, as well as prionic diseases<sup>(14)</sup>. Other advantages over sclera include uniform quality, easier handling, and commercial availability without dependence on an eye bank with potentially lower costs.

## CONCLUSIONS

Porcine pericardium is associated with significant inflammation when used as silicone glaucoma drainage device coverage in a rabbit model although no cases of graft melting and tube exposure were found. The inflammation was marked at both histopathologic and clinical levels. Further studies, with a longer follow-up, are necessary to evaluate the long-term importance of this inflammatory response.

## RESUMO

**Objetivo:** Avaliar a resposta inflamatória associada com o uso de pericárdio suíno processado e esclera homóloga preservada em glicerina no recobrimento de tubo de drenagem de silicone utilizado em cirurgia de glaucoma em modelo experimental. **Métodos:** Oito olhos de 8 coelhos albinos da raça Nova Zelândia receberam enxertos do mesmo tamanho de esclera homóloga, preservada em glicerina ou pericárdio suíno

processado, suturados à esclera recobrimdo um tubo de silicone. Os olhos foram avaliados quanto à hiperemia conjuntival de modo mascarado na primeira, terceira e sétima semanas de pós-operatório. Após a sétima semana os olhos foram enucleados e submetidos a estudo histopatológico. Os olhos também foram avaliados quanto à presença de afinamento do enxerto, exposição do tubo e quemose. **Resultados:** Não ocorreram casos de afinamento do enxerto ou exposição do tubo, porém o pericárdio suíno esteve associado clinicamente a uma maior inflamação. Achados à microscopia óptica incluíram intensa reação inflamatória em torno do enxerto de pericárdio com formação de granuloma do tipo corpo estranho. Por outro lado, no grupo que recebeu esclera, ocorreu formação de granulomas apenas em torno dos fios de sutura. **Conclusão:** O pericárdio suíno esteve associado a maior inflamação tanto à histopatologia quanto clinicamente, quando usado no recobrimento de tubo de silicone, em comparação à esclera homóloga preservada em glicerina, durante o período de acompanhamento de sete semanas.

**Descritores:** Transplante heterólogo; Pericárdio; Implantes para drenagem de glaucoma; Glaucoma; Suínos

## REFERENCES

- Minckler DS, Heuer DK, Hasty B, Baerveldt G, Cutting RC, Barlow WE. Clinical experience with the single-plate Molteno implant in complicated glaucomas. *Ophthalmology*. 1988;95(9):1181-8.
- Melamed S, Fiore PM. Molteno implant surgery in refractory glaucoma. *Surv Ophthalmol*. 1990;34(6):441-8. Review.
- Lotufo DG. Postoperative complications and visual loss following Molteno implantation. *Ophthalmic Surg*. 1991;22(11):650-6.
- Freedman J. Scleral patch grafts with Molteno setons. *Ophthalmic Surg*. 1987;18(7):532-4.
- Seiff SR, Chang JS Jr, Hurt MH, Khayam-Bashi H. Polymerase chain reaction identification of human immunodeficiency virus-1 in preserved human sclera. *Am J Ophthalmol*. 1994;118(4):528-30.
- Lusky M, Weinreb RN. Preservation of scleral grafts to avoid HIV infection. *J Glaucoma*. 1992;1(4):221.
- Brandt JD. Patch grafts of dehydrated cadaveric dura mater for tube-shunt glaucoma surgery. *Arch Ophthalmol*. 1993;111(10):1436-9.
- Rojanapongpun P, Ritch R. Clear corneal graft overlying the seton tube to facilitate laser suture lysis. *Am J Ophthalmol*. 1996;122(3):424-5.
- Tanji TM, Lundy DC, Minckler DS, Heuer DK, Varma R. Fascia lata patch graft in glaucoma tube surgery. *Ophthalmology*. 1996;103(8):1309-12.
- Raviv T, Greenfield DS, Liebmann JM, Sidoti PA, Ishikawa H, Ritch R. Pericardial patch grafts in glaucoma implant surgery. *J Glaucoma*. 1998;7(1):27-32.
- Gupta M, Lyon F, Singh AD, Rundle PA, Rennie IG. Bovine pericardium (Tutopatch) wrap for hydroxyapatite implants. *Eye*. 2007;21(4):476-9.
- Bidar M, Hawes MJ, Dresner SC, Epstein G, Lucarelli MJ, Glover T, et al. Conjunctival melting syndrome associated with the use of bovine pericardium wrapping material. *Ophthalm Plast Reconstr Surg*. 2003;19(4):257-61.
- Medicinal and other products and human and animal transmissible spongiform encephalopathies: memorandum from a WHO meeting. *Bull World Health Organ*. 1997;75(6):505-13.
- Mehta JS, Franks WA. The sclera, the prion, and the ophthalmologist. *Br J Ophthalmol*. 2002; 86(5): 587-92. Comment in: *Br J Ophthalmol*. 2002;86(12):1459.
- Panà A, Jung M. Prion diseases and iatrogenic infections I. A review. *Ig Sanita Pubbl*. 2005;61(4):325-77.
- Pauli G. Tissue safety in view of CJD and variant CJD. *Cell Tissue Bank*. 2005;6(3):191-200.

17. Heath CA, Barker RA, Esmonde TF, Harvey P, Roberts R, Trend P, et al. Dura mater-associated Creutzfeldt-Jakob disease: experience from surveillance in the UK. *J Neurol Neurosurg Psychiatry*. 2006;77(7):880-2.
18. Ionescu MI, Tandon AP, Mary DA, Abid A. Heart valve replacement with the Ionescu-Shiley pericardial xenograft. *J Thorac Cardiovasc Surg*. 1977;73(1):31-42.
19. Ionescu MI, Smith DR, Hasan SS, Chidambaram M, Tandon AP. Clinical durability of the pericardial xenograft valve: ten years experience with mitral replacement. *Ann Thorac Surg*. 1982;34(3):266-77.
20. Mykén PS. Seventeen-year experience with the St. Jude medical biocor porcine bioprosthesis. *J Heart Valve Dis*. 2005;14(4):486-92.
21. Brunner E, Langer F. Nonparametric analysis of ordered categorical data in designs with longitudinal observations and small sample sizes. *Biomet J*. 2000;42(6):663-75.
22. Smith MF, Doyle JW, Ticerney JW Jr. A comparison of glaucoma drainage implant tube coverage. *J Glaucoma*. 2002;11(2):143-7.
23. Lama PJ, Fechtner RD. Tube erosion following insertion of a glaucoma drainage device with a pericardial patch graft. *Arch Ophthalmol*. 1999;117(9):1243-4.
24. Yalvac IS, Duman S, Izgi B. Double-layer pericardium sandwich technique of ahmed glaucoma valve implantation in patient with anterior necrotizing Scleritis. *Tech Ophthalmol*. 2005;3(2):86-9.
25. Ainsworth G, Rotchford A, Dua HS, King AJ. A novel use of amniotic membrane in the management of tube exposure following glaucoma tube shunt surgery. *Br J Ophthalmol*. 2006;90(4):417-9.
26. King AJ, Azuara-Blanco A. Pericardial patch melting following glaucoma implant insertion. *Eye*. 2001;15(Pt 2):236-7.
27. DeBacker CM, Dutton JJ, Proia AD, Holck DE, Stone T. Bovine pericardium versus homologous sclera as wrapping materials for hydroxyapatite ocular implants: an animal study. *Ophthal Plast Reconstr Surg*. 1999;15(5):312-6.
28. DeBacker CM, Dutton JJ, Proia AD, Stone T, Holck DE. A comparative study of bovine pericardium (periguard) and homologous sclera as lower eyelid spacer graft analogs in New Zealand white rabbits. *Ophthal Plast Reconstr Surg*. 2000;16(2):156-61.

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