·Basic Research·

A new method of duplication of the chronic intraocular hypertension model: underwater electrocoagulation

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Abstract

• AIM: To study the experimental method of inducing the chronic intraocular hypertension in rat eyes.

• METHODS: Twenty Wistar rats were randomly divided into ocular hypertension and sham control groups. Intraocular pressure (IOP) was raised by electrocoagulating at least 3 episcleral and limbal veins on the right eye of each rat in ocular hypertension group and its contralateral eye was used as control. At 1, 2, 3, 4 and 8 week after the electrocoagulation of the veins, IOP were measured.

• RESULTS: The treatment of electrocoagulation caused a significant IOP increase of the right eyes over the baseline, over the contralateral eyes, and over the sham control eyes (repeated measures ANOVA, $P<0.001$). At 1 week, IOP was 30.12± 5.18mmHg, and maintained the high IOP up to 8 weeks.

• CONCLUSION: The chronic intraocular hypertension model could be successfully created by electrocoagulating three or more episcleral and limbal veins.

• KEYWORDS: glaucoma; intraocular hypertension; animal model

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INTRODUCTION

Glaucoma is a leading cause for blindness throughout the world, and the precise mechanism of development is still unknown. Neither the mechanical stress theory, nor ischemia stress theory, nor the combination of both can satisfactorily explain its pathogenesis. Though intraocular pressure (IOP) elevation is not the only factor causing glaucomatous optic neuropathy, IOP elevation has been considered as a major effect. Experimental hyper IOP animal models have been used to study the pathophysiology and pharmacodynamics of glaucoma. To establish a stable and chronic experimental glaucoma model for further study glaucoma, we tried the electrocoagulation of episcleral and limbal veins.

MATERIALS AND METHODS

Materials Twenty Wistar rats (Experimental Animal Division of Kunming General Hospital of Chengdu Millitary District), both male and female, weighing between 200-300g, were used in this study. All the rats were numbered and divided randomly into 2 groups: vein electrocoagulating and sham groups (10 respectively). The right eye of each animal was experimental while the left eye served as control. Equipments: ZEISS ophthalmologic microscope, microscopic scissors, microscopic teeth forceps, underwater electric coagulator (BAUSCH&LOMB,USA), tono-pen XL tonometer (Mentor, Norwell, USA).

Methods All the Wistar rats were housed for one week before surgery in a standard animal room lit with fluorescent lights approximately 300lux under standard 12-hour light/12-hour dark cycle, and the room temperature was maintained at 24℃. Rats were under general anesthesia

Table 1 The intraocular pressure from vein electrocoagulation and sham groups before and after operation (mmHg)

Operation time	Model(n=10)		Sham(n=10)	
	Right eye	Left eye	Right eye	Left eye
Baseline	15.91 ± 3.22	15.33 ± 2.56	16.33 ± 2.55	15.68 ± 2.64
1 wk	30.35 ± 4.98	15.87 ± 3.31	16.12 ± 3.06	15.85 ± 3.41
2 wk	32.45 ± 5.68	16.41 ± 3.51	16.13 ± 3.73	15.84 ± 2.78
3 wk	31.45 ± 4.54	16.16 ± 3.31	16.68 ± 4.12	16.35 ± 3.14
4 wk	33.65 ± 5.74	15.88 ± 2.28	17.22 ± 2.72	16.62 ± 3.13
8 wk	34.47 ± 7.39	16.23 ± 1.79	17.01 ± 3.16	16.35 ± 3.09

intraperitoneally using 30g/L pentobarbital at 30mg/kg, The right eyes were received surgical intervention and incisions were placed along the limbus for around 270°, bulbar conjunctiva was dissected and episcleral veins were exposed. At least 3 episcleral veins and limbal veins were coagulated by direct application of the underwater electric coagulator. Blanching of the vessels was noted and used as the indication of successful coagulation. After the procedure, the bulbar conjunctiva was restored. For sham group, only the conjunctiva was sheared without electrocoagulation. All fellow eyes of normal group were not treated. Erythromycin ointment was applied to the right eyes, and rats were allowed to recover from anesthesia at room temperature. 2.5g/L chloramphenicol, t.i.d, was applied to the eyes and erythromycin ointment, q.d, was administered for five days. IOP measurements: bilateral IOPs of all the animals were recorded before ocular surgery and again recorded at 1,2,3,4 and 8 week post-surgery. IOPs were taken under general anesthesia(using 30g/L pentobarbital injected intraperitoneally at 25-30mg/kg along with the topical 5g/L dicaine application) using tono-pen XL tonometer.The mean of 3-5 valid readings was used for each eye. Statistical method: SPSS10.0 package was used. Repeated measures ANOVA were applied to detect the differences of IOPs. All results were considered statistically significant if P<0.05.

RESULTS

As presented in Table 1, IOP of right eyes in the coagulating group was elevated across all postoperative time points, which was significantly higher than that of the left eyes or than that of the sham control eyes (P<0.001). However, IOPs were not significantly defferent across the 5 time points during the 8 weeks observation(P>0.05). There were no statistical differences between IOPs of the right eyes and the IOPs of the left eyes in sham groups.

DISCUSSION

Investigators have used glaucoma animal models to study pathogenesis of glaucoma, effects of IOP lowering agents, and mechanisms of optic nerve damage. An ideal model should be easy to maintain in a laboratory environment, easy to handle, exhibiting a predicable onset and a clinical course, and pathology to mimic that in the human eye. Several methods have been reported to establish elevated IOP in animals, including anterior chamber and vitreous body perfusion, aqueous overload, intravenous injection of hypertonic solution and induction by other agents [1-5].

Although the morphology of anterior chamber angle in many species is apparently different from that of human, the tubules for aqueous humor flow are similar between human and animals in that the interior tubule wall is coated with continuous endothelial cells. So most commonly used glaucoma models are based on inferior mammalians. Several protocols have been reported to develop models of chronic glaucoma[6-9], but most of these methods, with strong irritant effects and injuries to the local as well as intraocular tissues, often result in abrupt of IOP elevation and intensified inflammatory response with a shortened clinical course. In the present study, underwater electric coagulator was applied to coagulate at least 3 groups of vortex vein and veins around the limbus in order to decrease the drainage of aqueous humor veins and elevate the IOP. This method, causing less inflammation and heat damage to sclera and intraocular tissues, due to the application of underwater electric coagulator which coagulates episcleral veins and limbus veins, and due to the localized heat conduction and subsequent lower temperature, is unique and advantageous to the study of pathogenesis of glaucoma[10,11], effects of IOP lowering agents and mechanisms of optic nerve damage. Our study showed that the IOP of right eye in the coagulating group increased significantly after surgery. The

IOP was 30.12 ±5.18mmHg at 1 week post surgical treatment, and kept steady in the following weeks. The right IOP was significantly higher than the left IOP within the coagulating group, and also significantly higher than IOPs in both sham and normal group at various times after operation (P <0.001). Less inflammation and longer clinical course with a steady IOP elevation was observed in the right eye of the coagulating group, and the findings were consistent with the laser cauterizers [7]. The model presented here may be very useful in glaucoma researches.

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