

HISTAMINE H3 RECEPTOR ANTAGONISM IS ENHANCED BY PHARMACOLOGICAL ASSOCIATION WITH A NITRIC OXIDE DONOR IN A NEW ZEALAND WHITE RABBIT MODEL OF GLAUCOMA

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Glaucoma is the second leading cause of blindness worldwide. It is characterized by progressive optic nerve head atrophy because of elevated intraocular pressure (IOP). IOP reduction is the only therapeutic approach demonstrated to preserve visual function in patients affected by glaucoma. The first line treatment consists of topical IOP-lowering drugs. A significant number of patients require more than one drug to reach a target IOP. Histamine H3R has been found in several ocular structures (Vila, 2012). We previously demonstrated that the topical treatment with H3R inverse agonists is effective in reducing IOP in rabbit models of glaucoma (Lanzi 2014, 2015). Nitric oxide (NO) donors are successfully associated with PGAs in the treatment of glaucoma (Galassi 2006).

Aim of this research was to evaluate the potential NO-donor boost effect on IOP reduction obtained by ciproxifan and to study the expression and localization of histamine receptors (HRs) in ciliary body, retina, optic nerve and trabecular meshwork (TM) cells derived from ciliary body, to outline the role of the histaminergic system in the eye.

Ocular hypertension was obtained by the injection of 50 μ l of hypertonic saline (5%) into the vitreous or by 100 μ l (0.1%) of Carbomer in the anterior chamber. IOP measurements were performed using applanation tonometry (Tono-Pen AVIAR, Reichert, USA) prior to saline or Carbomer injection (baseline), immediately before drug dosing (pre-treatment) and 1-4 hours after saline injection in the acute model and after 24 hours in chronic model. All the animals underwent Ecocolor Doppler evaluation before and after chronic drug treatment, Pourcelot Resistance Index (RI) was calculated. Furthermore we investigated the expression of HRs in ciliary bodies, retinae and optic nerve by Western blot analysis on homogenated tissue samples, immunofluorescence staining on polarized histological slides and in a primary TM cells culture.

IOP rose from 16.4 ± 3.2 mmHg at baseline to 39.4 ± 4.8 mmHg after hypertonic saline injection and from 14.2 ± 5.3 to 36.8 ± 5.6 four days after Carbomer injection. Ciproxifan and molsidomine (0.1, 0.3, 0.5, 1%) dose-dependently reduced IOP 60' after saline injection (0.5% p value <0.05, 1% p value <0.01). The combination that produced the higher IOP reduction was ciproxifan/molsidomine 0.5/0.5%. IOP and mean resistance index (RI) of retinic artery were significantly reduced by this combination in the chronic model setting (p value <0.01 at day 10, 12 and 13 for IOP; p value < 0.05 for RI). Western blot analysis demonstrated the presence of histamine H1 and H3 receptors in ciliary bodies, retinae and optic nerve. The immunofluorescence staining revealed H1, H3 and H4R localization in ciliary bodies, on the vasal endothelium and in retinae. Moreover, the expression of these receptors was detected in TM cells by immunostaining.

Histaminergic H3R antagonists represent an interesting new therapeutic option for the treatment of glaucoma, the association with a NO-donor showed a synergic effect, boosting the IOP lowering efficacy and ameliorating the vascular performance of the retinic artery.

Galassi (2006) Br J Ophthalmol. 90: 1414-9.

Lanzi (2014) Inflamm. Res, 63; S12.

Lanzi (2015) Inflamm. Res, 64; S33.

Vila (2012). J Comp Neurol.15; 528-43.