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## Endothelin-like immunoreactivity in aqueous humor of patients with primary open-angle glaucoma and cataract

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**Abstract** ● Background: Experimental evidence suggests a role of endothelin-1 (ET) in the regulation of intraocular pressure (IOP).

● Method: Therefore, in patients undergoing cataract surgery, ET-like immunoreactivity (ETIR) was measured by radioimmunoassay in pooled samples of aqueous humor of eyes with primary open-angle glaucoma (POAG) and normotensive eyes with cataract only. ● Results: ETIR was significantly ( $P < 0.05$ ) higher in patients with cataract and POAG ( $20.5 \pm 1.8$  pg/ml,  $n = 12$ ; preoperative IOP  $21.4 \pm 1.1$  mmHg,  $n = 33$ ) than in patients with cataract only

( $15.8 \pm 1.6$  pg/ml,  $n = 15$ ; preoperative IOP  $16.0 \pm 0.6$  mmHg,  $n = 77$ ). ● Conclusion: This finding may indicate a role of ET in POAG or ocular antihypertensive treatment, and its relevance should be further investigated.

### Introduction

Endothelin-1 (ET), currently the most potent vasoconstrictive peptide, is probably involved in various vascular diseases and exhibits diverse activities in ocular tissues, including regulation of aqueous humor dynamics [1, 5]. We have shown ET-mediated contractions of isolated trabecular meshwork and ciliary muscle strips [3], and demonstrated for the first time that endothelin-like immunoreactivity (ETIR) in aqueous humor of human and bovine eyes is 2–3 times higher than the corresponding plasma level [4]. Furthermore, we have shown that human non-pigmented ciliary epithelial cells express a high potential for releasing ETIR [4]. Immunocytochemically, an intensive staining for ETIR was found in cultured human non-pigmented ciliary epithelial cells and in ciliary epithelial cells of donor eyes [4]. This suggests a role of ET in the regulation of aqueous humor secretion and outflow. Therefore, we measured ETIR in the aqueous humor of

patients undergoing cataract surgery and compared ETIR of patients having both cataract and primary open-angle glaucoma (POAG) with normotensive eyes with cataract only.

### Materials and methods

The study includes 110 eyes of 110 patients admitted for cataract surgery. 33 of these had POAG, and 77 age-matched normotensive eyes with cataract only served as controls. All eyes were free from ocular disease other than cataract or POAG. All pupils were dilated preoperatively by the same standard mydriatic protocol (three drops each of scopolamine 0.25% and a combination of tropicamide 0.5% and phenylephrine 2.5%). Before performing anterior capsulectomy, 100–300  $\mu$ l of aqueous humor was quickly withdrawn as part of the routine procedure and replaced by a viscoelastic substance. The samples were collected and stored at  $-20^{\circ}\text{C}$  before final measurement. As preliminary measurements and calculations indicated that 0.5–1.0 ml of aqueous humor was necessary for a single determination of ETIR, two to six samples, of the respective groups had to be pooled, depending on the volume that could be safely withdrawn

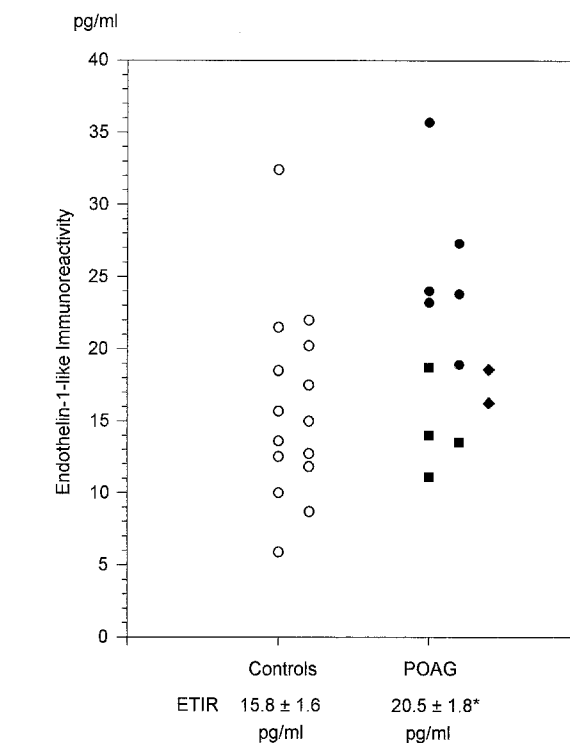
from the anterior chamber of the individual eyes. The samples were allocated according to the preoperatively applied antiglaucomatous medication (beta-blockers only, 6; beta-blockers in combination with other topical ocular antihypertensives, 2; various ocular antihypertensives without beta-blockers, 4) into a total of 27 pooled samples (12 POAG, 15 controls). ETIR was measured using a commercial radioimmunoassay kit (TTS, Wychen, Netherlands) as described in detail previously [4]. Data are given as mean values  $\pm$  SEM. Statistical significance was assessed by Student's *t*-test for unpaired observations.

## Results

Mean ETIR in the pooled samples (Fig. 1) of patients with POAG ( $20.5 \pm 1.8$  pg/ml) was significantly ( $P < 0.05$ ) higher than in normotensive eyes with cataract only ( $15.8 \pm 1.6$  pg/ml). Mean preoperative intraocular pressure (IOP) under the respective medication was  $21.4 \pm 1.1$  mmHg in the group with POAG and  $16.0 \pm 0.6$  mmHg in controls. Analysis of the subgroups indicates that eyes on topical antihypertensive treatment with beta-blockers tend to have higher ETIR than POAG eyes not on beta-blockers. However, this difference was not statistically significant, and further statistical analysis of subgroups was not feasible because of the small number of patients with POAG that were not on treatment with beta-blockers preoperatively.

## Discussion

The higher level of ETIR in patients with POAG and cataract than in patients with cataract only, as well as experimental evidence, suggests that ET might be involved in the pathogenesis or regulation of IOP in POAG. However, the level of the preoperative IOP or the chronic application of ocular antihypertensive medications such as beta-blockers may also influence the results. Furthermore,



**Fig. 1** Endothelin-like immunoreactivity in pooled samples of aqueous humor of patients with cataract only (*open circles*) and patients with POAG and cataract (*filled circles* beta-blockers only, *lozenges* beta-blockers in combination with other ocular antihypertensives, *squares* various ocular antihypertensives without beta-blockers). \* $P < 0.05$  compared with controls

even if it can be confirmed that POAG is associated with higher levels of ET, there is no indication from our data whether the increase in ETIR in aqueous humor of these patients is the cause of elevated IOP or just a coincidental event. However, it is interesting to note that plasma ETIR in normal-tension glaucoma is abnormal [2]. Therefore, the role of ET in POAG should be further evaluated.

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