学 位 論 文 の 要 旨

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論 文 内 容 の 要 旨
INTRODUCTION

Glaucoma is one of the major causes of severe visual loss and blindness in the world. Elevated intraocular pressure (IOP) is the primary risk factor for open-angle glaucoma (OAG), including primary open-angle glaucoma (POAG) and glaucoma secondary to pseudoexfoliation syndrome (EXG). In these OAGs, the increased IOP is explained by reduced aqueous humor outflow from the anterior chamber (AC) into Schlemm's canal (SC), which passes through the trabecular meshwork (TM)/Schlemm's canal endothelium (SCE) complex.

In POAG, increased TM resistance is due to changes in the extracellular matrix (ECM), while in EXG, the accumulation of abnormal fibrillar materials in the TM leads to IOP elevation. Although factors like immune responses and oxidative stress are implicated, the precise mechanisms of ECM changes in OAG remain unclear.

In POAG, it has been previously reported that the cross-sectional area of SC was significantly smaller compared to normal eyes, and among elderly patients with POAG, the SC size was remarkably smaller in eyes with a family history of glaucoma, while a significant reduction of SCEs was observed in eyes without a family history. These findings suggest that not only the size of SC but also the presence of SCEs themselves are determinants of IOP.

In previous studies, immunostaining for thrombomodulin (TBM) has been used for histologic examination of SCE. However, very few studies have conducted a morphologic comparison of SC and SCE between POAG and EXG using anti-TBM antibody staining. In this study, we established a database using trabeculectomy specimens collected in a consecutive series of patients with both POAG and EXG. Background factors were standardized using propensity scores, allowing for a comprehensive comparison of various parameters between the two disease types.

MATERIALS AND METHODS

The subjects were 152 POAG patients comprising a total of 182 eyes and 116 EXG patients comprising a total of 138 eyes who underwent trabeculectomy at the Japanese Red Cross Medical Center between January 1997 and June 2018. All trabeculectomies were performed by one of the authors (TH). For all patients, information regarding age, sex, maximum preoperative IOP, and preoperative medication score (1 point for each component of topical medication or 1 point for each tablet of oral acetazolamide) was obtained.

Trabeculectomy-excised tissues were fixed in a mixture of 2.5% or 5% formalin and 1% glutaraldehyde, and they were divided into three to five blocks. All divided specimens were embedded in paraffin and cut into sections with a thickness of 3 μm. These paraffin sections underwent hematoxylin-eosin (HE) staining and immunohistochemical staining with anti-TBM antibodies. The stained sections were then photographed and digitally imaged using an optical microscope system. On the obtained photographs, SC morphology parameters were estimated using ImageJ software on a Windows 10 computer. Four primary parameters consisted of the lengths of TBM-positive and opened (POSC) or closed (PCSC) SC, as well as the lengths of TBM-negative and opened (NOSC) or closed (NCSC) SC were measured. After obtaining these parameters, additional parameters, including the total SC length (TSC), TBM positive/negative SC lengths (PSC/NSC), and opened/closed SC lengths (OSC/ CSC), were calculated. All the images were evaluated independently by two examiners (i.e., AT and TH), and the four primary parameters were determined by consensus between the examiners. Out of the sections analyzed, 26 eyes in the POAG group and 5 eyes in the EXG group were excluded from the study due to tissue destruction during excision of the TM.

To compare the measured parameters between the two groups, an equal number of eyes were selected from each group using propensity score matching with the nearest available matching method. Propensity score matching was conducted based on two covariates (i.e., age and gender; $n = 87$ in each group) or on four covariates (i.e., age, gender, maximum preoperative IOP, and medication score; $n = 64$ in each group). After selecting the cases, the measurement parameters were compared between two groups by unpaired t-test for continuous variables and by Fisher's exact probability test for categorical variables. The possible association between the measured parameters and background parameters was assessed by multiple regression analyses. A P value less than 0.05 was considered statistically significant. All statistical analyses were performed using the JMP Pro statistical software version 16.00. The study protocol was approved by the Research Ethics Committee of Shimane University Faculty of Medicine.

RESULTS AND DISCUSSION

When matched for age and gender, 87 cases each were selected in each group. In these cases, compared with POAG, the maximum preoperative IOP ($P < 0.0001$) and medication score ($P = 0.0027$) were significantly higher in the EXG group. The TSC was no significant difference between the POAG (287.1 μm) and EXG (300.5 μm) groups ($P = 0.16$), while the PSC ($P =$

0.025) was significantly shorter and, vice versa, NSC ($P = 0.0010$) and NOSC ($P < 0.0001$) were significantly longer in the EXG group than the POAG group. This suggests a reduced presence of TBM-positive SCE in EXG, despite the maintenance of the SC lumen, leading to the hypothesis that the loss of SCE is more pronounced in EXG compared to POAG.

To assess the potential effects of background factors on the morphology parameters, multiple regression analyses were performed, including age, gender, maximum preoperative IOP, medication score, and disease type as dependent variables in these 174 cases. The results showed that older age was associated with longer NSC ($P = 0.0053$) and NCSC ($P = 0.033$), while it was associated with shorter PSC ($P < 0.0001$), OSC ($P = 0.031$), and POSC ($P = 0.0001$). Female gender was linked to shorter POSC ($P = 0.030$), and a higher medication score was associated with longer CSC ($P = 0.011$) and NCSC ($P = 0.011$). Even after adjusting for differences in background factors by this multivariate analysis, once again, EXG rather than POAG was associated with longer NOSC ($P = 0.0012$).

Multivariate analysis confirmed that the maximum preoperative IOP and medication scores were influential factors contributing to the reduction of SCE and narrowing of the SC. Therefore, propensity score matching was performed again, including the maximum preoperative IOP and medication scores in addition to age and gender. This time 64 cases each were selected in each group. After matching on four factors, there were no significant differences in any of the background factors. The results indicated that NOSC ($P = 0.0049$) was significantly longer in EXG than POAG. These findings supported the hypothesis that EXG is associated with a more pronounced loss of SCE.

In the multivariate analysis, a decrease in PSC, OSC, and POSC, along with an increase in NSC and NCSC, was observed with aging. These results suggest that aging is associated with both a reduction in SCE and a decrease in SC size. The decline of SCE with aging has been reported in the past. In this study, a correlation between female gender and the shortening of POSC was also observed. However, the reasons for this relationship remain unclear and require further research. Additionally, it was found that a higher medication score was associated with longer CSC and NCSC. Since a high medication score typically indicates high IOP, this correlation may reflect the influence of preoperative IOP. However, the impact of the use of eye drops, such as prostaglandins, which are thought to cause atrophy of SC, cannot be ruled out. The duration of eye drop usage was unclear in this study, so future studies should consider including information about the specific formulations of eye drops used.

CONCLUSION

In this study, even after adjusting for background factors using propensity score matching, it was shown that NOSC was longer in EXG group compared to POAG group. This suggests that while the lumen structure is maintained, SCE are undergoing dropout. SCE loss may contribute more critically to IOP elevation in EXG than in POAG.