HRQOL

HRQOL was assessed using the Japanese version of the Medical Outcome Study Short Form 8-item Health Survey (SF-8), which is the short version of the SF-36 implemented through a self-administered survey. The SF-8 consists of eight items that represent the eight domains of the SF-36: physical functioning, role physical, bodily pain, general health, vitality, social functioning, role emotional, and mental health [18, 19]. Using the differential weights assigned to each item, the SF-8 produces two summary scores: a physical health component summary (PCS) score and a mental health component summary (MCS) score. These scores were standardized based on the Japanese population in 2002. A score of 50 represented the mean for the Japanese general population, and a score of 10 was one standard deviation; lower scores indicate poorer HRQOL.

Statistical analysis

Differences in the baseline characteristics between men and women were examined using an unpaired t test and a χ^2 test. To examine the impact of the number of LBP days during the 1-month study period on HRQOL, we calculated the changes in PCS and MCS scores from the baseline (PCS_c and MCS_c, respectively) by subtracting the baseline SF-8 scores from the scores calculated after the 1-month study period. Negative values indicated a decline in HRQOL, while positive values indicated an increase.

We examined the relationship between the cumulative number of LBP days during the 1-month study period and differences in the changes in PCS and MCS scores between sexes using linear regression models. Before using linear regression models, we examined the linear relationship between PCS_c and MCS_c scores and the cumulative number of LBP days using a locally weighted scatter plot smoothing (LOWESS) curve, a technique for smoothing scattered values by fitting a weighted least-squares line into moving bands using nonparametric regression [20], and conducted linear regression analyses on the portion where linearity holds visually on the curve. We then examined the differences between men and women using the interaction terms of sex and cumulative number of LBP days in the regression models. The adjusted model included age, the number of baseline comorbidities (0, 1, ≥2), annual household income (<3,000,000 yen, 3,000,000–4,999,999 yen, 5,000,000–6,999,999 yen, 7,000,000–9,999,999 yen, 10,000,000–11,999,999 yen, and ≥12,000,000 yen), employment status (yes or no, with "yes" indicating a full-time or part-time job and "no" for any other response), and baseline PCS or MCS score as covariates [15, 16, 21]. We calculated individual comorbidities by counting the number of the following diseases that the person had: hypertension, diabetes mellitus, cerebrovascular disease, cardiovascular disease, lung disease, gastrointestinal disease, urinary disease, musculoskeletal disease, skin disease, mental disease, gynecological disease, cancer, and other diseases. All statistical analyses were conducted using Stata version 11.2 (Stata Corporation LP, College Station, TX, USA). An alpha level of 0.05 was set as the threshold to determine the statistical significance.

Results

Of the 2,358 subjects participating in the HDS, 8.0% ($n = 188$) were excluded due to missing annual household income data or an incomplete SF-8 questionnaire, leaving 2,170 subjects for the analysis. The excluded sample was, on average, older than the analysis sample (49.9 years old vs. 44.5 years old, $P < 0.01$) and contained a higher proportion of women (61.2% vs. 54.5%, $P = 0.08$).

Subject characteristics are described in Table 1. Women had lower scores than men for household income, employment status, baseline PCS, and baseline MCS. During the study period, 28.4% ($n = 617$) of the total sample experienced LBP at least once during the study period and were thus included in analysis. LBP was reported more frequently by women ($n = 373$, 31.5%) than by men ($n = 244$, 24.7%, $P < 0.01$). The average number of cumulative LBP days was 1.7 (SD: 4.8) for women and 1.3 (SD: 4.3) for men ($P = 0.04$). The average change in PCS and MCS scores was −1.1 (SD: 6.7) and 0.8 (SD: 6.7), respectively. Approximately one-third of the sample had more than one disease, and the most common baseline comorbidities were hypertension and musculoskeletal diseases.

The LOWESS curve between the cumulative number of LBP days and the changes in SF-8 revealed a change in the curve at approximately 15 days into the study period (Fig. 1). For men, the average change in PCS decreased linearly with the increasing cumulative number of LBP days. However, the scores in men experiencing LBP for approximately 15 days or more tended to increase as the number of days with LBP increased (Fig. 1a). For women, the average change in PCS was flat throughout the study period (Fig. 1b). In contrast, the MCS_c among men with less than 16 LBP days appeared to increase slightly as the number of LBP days increased, while the MCS_c among men with 16 or more cumulative LBP days decreased as the number of LBP days increased (Fig. 1c), no such relationship was observed in women (Fig. 1d). Because linearity was not visually confirmed in 58 subjects who had LBP for 16 or more days, these individuals were excluded from the analysis with regression models.

Table 1 Subject characteristics

	Total (n = 2,170)	Women (n = 1,183)	Men (n = 987)	P value
Age in years, mean (SD)	44.5 (15.2)	44.8 (15.5)	44.3 (14.7)	0.43
Cumulative days with LBP, mean (SD)	1.5 (4.6)	1.7 (4.8)	1 3 (4.3)	0.04
Comorbidities, number (%)				
0	1,518 (70.0)	806 (68.1)	712 (72.4)	0.11
1	442 (20.4)	252 (21.3)	190 (19.3)	
2+	210 (9.7)	125 (10.6)	85 (8.6)	
Annual household income, number (%)				
<3,000,000 yen	353 (16.3)	226 (19.1)	127 (12.9)	<0.01
3,000,000–4,999,999 yen	641 (29.5)	350 (29 6)	291 (29.5)	
5,000,000–6,999,999 yen	515 (23 7)	260 (22.0)	255 (25 8)	
7,000,000–9,999,999 yen	420 (19.4)	223 (18 9)	197 (20.0)	
10,000,000–11,999,999 yen	162 (7.5)	82 (6.9)	80 (8.1)	
≥12,000,000 yen	79 (3.6)	42 (3 6)	37 (3 8)	
Employment status, number (%)				
No	762 (35 1)	590 (49.9)	172 (17.4)	<0.01
Yes	1,408 (64.9)	593 (50.1)	815 (82.6)	
SF-8 (baseline), mean (SD)				
PCS	49.2 (6.5)	48.9 (6.6)	49.7 (6.4)	<0.01
MCS	48.0 (6.7)	47 7 (6.6)	48.4 (6 7)	0.01
PCS_c, mean (SD)	−1.1 (6.7)	−1.0 (6.6)	−1 3 (7.0)	0.32
MCS_c, mean (SD)	0.8 (6.7)	0.8 (6.6)	0.8 (6.8)	0.85

SD standard deviation, *PCS* physical component summary, *MCS* mental component summary, *LBP* low back pain

Table 2 shows the changes in PCS and MCS associated with a 1-day increase in the number of LBP days, as represented by the coefficients in the respective models with PCS_c and MCS_c in the unadjusted and adjusted models. In the unadjusted regression model for PCS_c, an increase in one LBP day was associated with a 0.51-point decrease in men's PCS scores ($P < 0.001$) versus a 0.04-point decrease in women's scores ($P = 0.70$), which is a statistically significant difference ($P < 0.001$). After adjusting for age, number of baseline comorbidities, annual household income, employment status, and baseline PCS score, an increase in one LBP day was associated with a 0.72-point decrease per day in men's scores ($P < 0.001$) versus a 0.29-point decrease in women's scores ($P < 0.001$), which is also statistically significant ($P < 0.001$).

In the unadjusted regression model for MCS_c, an increase in one LBP day was associated with a 0.27-point increase per day in MCS_c among men ($P = 0.008$) versus a 0.01-point decrease among women ($P = 0.87$), which is a statistically significant difference ($P = 0.03$). After adjusting for the above-mentioned covariates, an increase in one LBP day was associated with a 0.17-point increase per day in MCS_c among men ($P = 0.05$) versus a 0.02-point decrease among women ($P = 0.76$). However, this difference was not statistically significant ($P = 0.08$).

Discussion

In this study, we found that changes in HRQOL associated with an increased cumulative number of LBP days differed by gender. While PCS scores decreased more sharply in men than in women, the slope of change in MCS scores was not significantly different between both sexes after adjusting for age, the number of baseline comorbidities, annual household income, employment status, and baseline PCS/MCS scores. To our knowledge, this study is the first to prospectively examine the sex differences in the relationship between the number of LBP days and the change in HRQOL score. Our observation that men tend to suffer from more severe decreases in HRQOL than women with the same number of LBP days, which contrasts with previous findings, underscores the importance in accounting for the degree of LBP in examining sex difference in HRQOL.

Decreases in PCS scores noted in the present study were sharper in men than in women, indicating that men were more affected by LBP than women with respect to the physical domain of HRQOL. However, this finding appears to contradict previous work, such as those noted in the cross-sectional study by Salaffi et al. [15], which showed that women with LBP experienced worse physical health

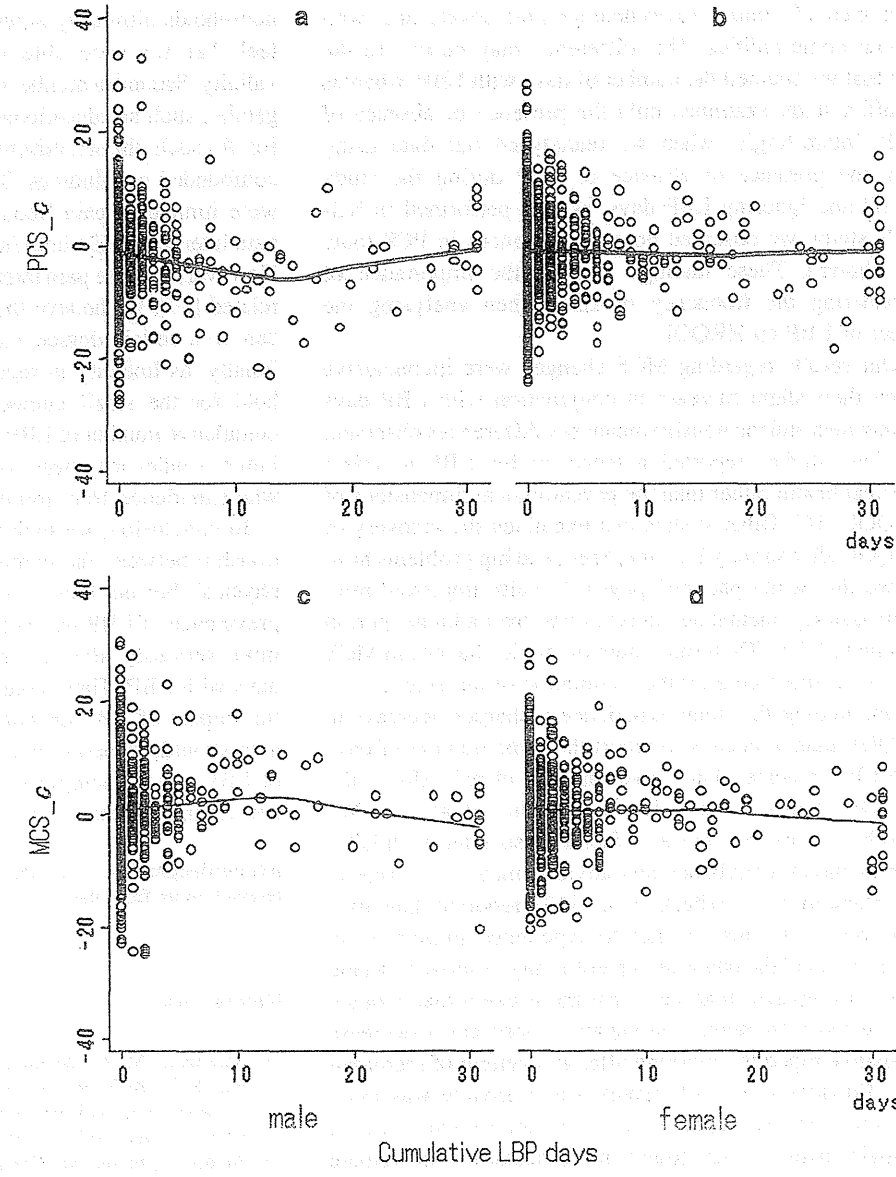

Fig. 1 Cumulative number of LBP days and differences in SF-8 summary score changes by gender. **a** Relationship between the cumulative number of LBP days and the change in PCS score in men. **b** Relationship between the cumulative number of LBP days and the change in PCS score in women. **c** Relationship between the cumulative number of LBP days and the change in MCS score in men. **d** Relationship between the cumulative number of LBP days and the change in MCS score in women

Table 2 Unadjusted and adjusted models of PCS_*c* and MCS_*c* from the cumulative number of LBP days in women and men

	Women			Men			Interaction
	Coefficient	95% CI		Coefficient	95% CI		
		Lower	Upper		Lower	Upper	
PCS							
Unadjusted	−0.04	−0.21	0.14	−0.51	−0.71	−0.31	<0.01
Adjusted	−0.29	−0.43	−0.14	−0.72	−0.89	−0.55	<0.01
MCS							
Unadjusted	−0.01	−0.19	0.16	0.27	0.07	0.47	0.03
Adjusted	−0.02	−0.16	0.12	0.17	0.004	0.33	0.08

PCS physical component summary, *MCS* mental component summary, *LBP* lower back pain, *CI* confidence interval

than men of similar socio-demographic levels and with similar comorbidities. This difference may be due to the fact that we counted the number of days with LBP, whereas Salaffi's study examined only the presence or absence of LBP. Interestingly, when we reanalyzed our data using only the presence or absence of LBP during the study period and ignoring LBP days, as was performed in Salaffi's study, we observed no sex differences in PCS (data not shown). These findings indicate the importance of considering the frequency of LBP when analyzing the effect of LBP on HRQOL.

Our results regarding MCS changes were inconclusive given their slight increase in conjunction with LBP days among men and the nonsignificant sex differences observed. Previous studies reported a tendency for LBP to affect physical health rather than the psychological dimensions of HRQOL [16]. Other studies that examined the recovery of HRQOL after surgery for back, knee, and hip problems have shown that while patients' physical health improved relatively quickly, mental health recovered over a longer period of time [22, 23]. Therefore, more dramatic changes in MCS may have appeared after the completion of our study.

Our finding that men experience a sharper decrease in HRQOL than women who report the same number of days with LBP indicates that a given amount of pain affects the HRQOL of men more seriously than that of women. We hypothesize that psychosocial factors associated with LBP, such as mood disturbance and anxiety, may play a role in this phenomenon. Affleck et al. [13] reported that men were more likely than women to experience an increase in negative mood the day after a painful day. Similarly, Keefe et al. [14] reported that men were more likely than women to experience an increase in negative mood and a decrease in positive mood the morning after an evening of increased pain. Edwards et al. [24] reported that anxiety was associated with increased pain severity and interference by pain in male patients but found no association in female patients. Robinson et al. [25] reported significantly stronger relationships between pain-related anxiety and LBP in men than in women. Taken together, these previous findings suggest that men experience more anxiety and greater mood disturbance than women for the same number of days of LBP, in turn contributing to the reduced physical HRQOL. Although mental health scores as assessed in the present study did not appear to capture this temporary psychological change, future studies may further explore the mechanisms behind this sex difference.

Several limitations of the present study warrant mention. First, our sample was selected from people registered with a market research company, and many households refused to participate in the initial random sampling. Therefore, our sample population may differ in certain respects from the general population. Nevertheless, because most resampled households ultimately agreed to participate in the study, we feel that we were able to sufficiently maintain internal validity. Second, a number of variables known to be related to gender, such as educational attainment, were not controlled for. As such, the sex differences in these variables may have confounded our findings. Third, our analysis used data that were limited to pain frequency and lacked information on pain intensity, while both factors can influence HRQOL [26]. However, because pain intensity and frequency are positively related [27], we believe that our analysis of pain frequency can, to a certain degree, capture the level of pain intensity. Finally, as linearity in regression analyses did not visually hold for the small number of subjects with 16 or more cumulative number of LBP days, we excluded these subjects. Future studies may need to include responses from subjects who experience 16 or more days of LBP.

In conclusion, we identified sex differences in the relationship between the cumulative number of LBP days and physical, but not mental, health status. Although a greater prevalence of LBP was noted in women, quality of life was more seriously affected in men for the same number of days with LBP. These results underscore the complexity of the impact of LBP on HRQOL, and future studies in this area should, in particular, take into consideration duration of LBP and not simply its presence or absence while also accounting for sex differences.

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References

1. Sprangers, M. A., de Regt, E. B., Andries, F, van Agt, H. M., Bijl, R. V., de Boer, J B., et al. (2000). Which chronic conditions are associated with better or poorer quality of life? *Journal of Clinical Epidemiology, 53*(9), 895–907
2. Alonso, J., Ferrer, M., Gandek, B., Ware, J. E., Jr., Aaronson, N K., Mosconi, P., et al. (2004). Health-related quality of life associated with chronic conditions in eight countries: results from the International Quality of Life Assessment (IQOLA) Project. *Quality of Life Research, 13*(2), 283–298.
3. Hart, L. G, Deyo, R. A., & Cherkin, D. C. (1995). Physician office visits for low back pain. *Spine, 20*(1), 11–19.
4. Walker, B. F., Muller, R., & Grant, W. D. (2003). Low back pain in Australian adults: The economic burden. *Asia-Pacific Journal of Public Health, 15*(2), 79–87
5. Maniadakis, N., & Gray, A. (2000). The economic burden of back pain in the UK. *Pain, 84*(1), 95–103.
6. Fanuele, J. C , Birkmeyer, N. J , Abdu, W. A., Tosteson, T. D., & Weinstein, J N. (2000). The impact of spinal problems on the health status of patients: Have we underestimated the effect? *Spine, 25*(12), 1509–1514.
7. Veerapen, K., Wigley, R. D , & Valkenburg, H. (2007). Musculoskeletal pain in Malaysia. A COPCORD survey. *Journal of Rheumatology, 34*(1), 207–213
8. Chenot, J. F., Becker, A., Leonhardt, C., Keller, S., Donner-Banzhoff, N., Hildebrandt, J , et al. (2008). Sex differences in

presentation, course, and management of low back pain in primary care. *Clinical Journal of Pain, 24*(7), 578–584.
9. Linton, S. J., Hellsing, A. L., & Halldén, K. (1998). A population-based study of spinal pain among 35–45-year-old individuals. Prevalence, sick leave, and health care use. *Spine, 23*(13), 1457–1463.
10. Müllersdorf, M., & Söderback, I. (2000). The actual state of the effects, treatment and incidence of disabling pain in a gender perspective—A Swedish study. *Disability and Rehabilitation, 22*(18), 840–854.
11. Wise, E. A., Price, D. D., Myers, C. D., Heft, M. W., & Robinson, M. E. (2002). Gender role expectations of pain: relationship to experimental pain perception. *Pain, 96*(3), 335–342.
12. Riley, J. L., 3rd, Robinson, M. E., Wise, E. A., Myers, C. D., & Fillingim, R. B. (1998). Sex differences in the perception of noxious experimental stimuli A meta-analysis. *Pain, 74*(2–3), 181–187.
13. Affleck, G., Tennen, H., Keefe, F. J., Lefebvre, J. C., Kashikar-Zuck, S., Wright, K., et al. (1999). Everyday life with osteoarthritis or rheumatoid arthritis. Independent effects of disease and gender on daily pain, mood, and coping. *Pain, 83*(3), 601–609.
14. Keefe, F J., Affleck, G., France, C. R., Emery, C. F , Waters, S , Caldwell, D. S., et al. (2004). Gender differences in pain, coping, and mood in individuals having osteoarthritic knee pain: A within-day analysis. *Pain, 110*(3), 571–577.
15. Salaffi, F., De Angelis, R., Stancati, A., Grassi, W., & MArche Pain Prevalence INvestigation Group (MAPPING) study (2005). Health-related quality of life in multiple musculoskeletal conditions. a cross-sectional population based epidemiological study. II. The MAPPING study *Clinical and Experimental Rheumatology, 23*(6), 829–839
16. Bingefors, K., & Isacson, D (2004) Epidemiology, co-morbidity, and impact on health-related quality of life of self-reported headache and musculoskeletal pain–a gender perspective. *European Journal of Pain, 8*(5), 435–450.
17. Fukui, T , Rhaman, M., Takahashi, O., Saito, M., Shinbo, T., Endo, H., et al (2005). The ecology of medical care in Japan. *JMAJ, 48*, 163–167.
18. Fukuhara, S , & Suzukamo, Y. (2004). *SF-8 Japanese version manual.* Kyoto: NPO Health Medicine Evaluation Research Organization.
19. Ware, J. E., Kosinski, M., Dewey, J. E., & Gandek, B. (2001). *How to score and interpret single-item health status measures: A manual for users of the SF-8 health survey.* Lincoln, (RI): QualityMetric Incorporated.
20. Diggle, P. J., Heagerty, P., Liang, K. Y., & Zeger, S. L. (2002). Exploring longitudinal data. In P. J Diggle, P. Heagerty, K. Y. Liang, & S. Zeger (Eds.), *Analysis of longitudinal data, 2nd edn* (pp. 33–53). Oxford. Oxford University Press.
21. Pereira, C. C., Palta, M., Mullahy, J., & Fryback, D. G. (2011) Race and preference-based health-related quality of life measures in the United States. *Quality of Life Research, 20*(6), 969–978.
22. Rampersaud, Y. R., Ravi, B , Lewis, S. J., Stas, V., Barron, R., Davey, R., et al. (2008). Assessment of health-related quality of life after surgical treatment of focal symptomatic spinal stenosis compared with osteoarthritis of the hip or knee. *The Spine Journal, 8*(2), 296–304.
23. Bachmeier, C. J., March, L. M., Cross, M. J., Lapsley, H. M , Tribe, K. L., Courtenay, B. G., et al. (2001). A comparison of outcomes in osteoarthritis patients undergoing total hip and knee replacement surgery. *Osteoarthritis Cartilage, 9*(2), 137–146.
24. Edwards, R., Augustson, E. M., & Fillingim, R. (2000). Sex-specific effects of pain-related anxiety on adjustment to chronic pain. *Clinical Journal of Pain, 16*(1), 46–53
25. Robinson, M. E., Dannecker, E. A., George, S. Z., Otis, J , Atchison, J W., & Fillingim, R. B (2005). Sex differences in the associations among psychological factors and pain report: A novel psychophysical study of patients with chronic low back pain. *J Pain, 6*(7), 463–470.
26. Skevington, S. M. (1998). Investigating the relationship between pain and discomfort and quality of life, using the WHOQOL. *Pain, 76*(3), 395–406.
27. Heymans, M. W., van Buuren, S., Knol, D L., Anema, J. R., van Mechelen, W , & de Vet, H C. (2010). The prognosis of chronic low back pain is determined by changes in pain and disability in the initial period. *The Spine Journal, 10*(10), 847–856.

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ORIGINAL ARTICLE

Trabecular Meshwork Length in Men and Women by Histological Assessment

Toshimitsu Kasuga[1,2], Yi-Chun Chen[1,3], Michele M. Bloomer[1], Kristin E. Hirabayashi[1], Yoshimune Hiratsuka[2], Akira Murakami[2], and Shan C. Lin[1]

[1]*Department of Ophthalmology, University of California, San Francisco, CA, USA,* [2]*Department of Ophthalmology, Juntendo University, Tokyo, Japan, and* [3]*Department of Ophthalmology, Cathay General Hospital, Taipei, Taiwan*

ABSTRACT

Purpose: To evaluate the length of the trabecular meshwork (TM) from the scleral spur (SS) to Schwalbe's line (SL) and assess the detectability of the SS in histopathology specimens.

Methods: This study included 158 angle images from 79 cross-sectional slides derived from eyes enucleated for melanoma. The slides were stained with hematoxylin-eosin (HE) or periodic acid schiff (PAS). Two ophthalmologists evaluated the TM length by using the slides stained with HE to assess the interobserver reproducibility. For intraobserver reproducibility, the first observer assessed 79 images in a different session. Also, 30 images that were randomly selected for PAS stain were evaluated to assess the agreement of the measurements between HE and PAS staining. Interclass correlation coefficients (ICC) were calculated to evaluate reproducibility of measurements. The images were also evaluated for detectability of the scleral spurs.

Results: Among the 79 included subjects, 40 were male and 39 were female. The average trabecular meshwork length was 694.9 ± 109.0 μm in the male group and 713.2 ± 106.9 μm in the female group ($p = 0.29$). Intraobserver and interobserver ICC were 0.89 and 0.62, respectively. ICC for agreement between HE and PAS was 0.89. Among the 158 angles graded, the first observer graded 40 images (25.3%) and the second observer graded 45 (28.5%) as difficult to identify the scleral spur.

Conclusions. There was no statistically significant difference between the average trabecular meshwork length in men and women. Among the angles evaluated, 25.3–28.5% were graded as difficult to identify the scleral spur.

KEYWORDS: Scleral spur, Schlemm's canal, Trabecular meshwork, Anterior chamber, Anatomy & histology, Angle closure glaucoma

INTRODUCTION

Primary angle closure glaucoma (PACG) is a frequent cause of blindness among glaucoma patients, particularly those of Asian descent. Angle parameters such as angle opening distance (AOD)[1] and angle recess area (ARA)[2] using ultrasound biomicroscopy (UBM) or anterior segment optical coherent tomography (AS-OCT) images are widely used in the clinical setting to evaluate for angle narrowing or closure. AOD was reported by Pavlin et al.[1] as the length of a line drawn perpendicular from the trabecular meshwork a given distance from the scleral spur to the anterior iris. Ishikawa et al.[2] defined ARA as the triangular area bordered by the anterior iris surface, the corneal endothelium, and the line drawn to the iris surface from a point 750 μm anterior to the scleral spur. More recently, AOD and ARA have been assessed at 250 μm, 500 μm, and 750 μm from the scleral spur. Most often, values at 500 μm and 750 μm are used as the primary outcome measures in studies of angle closure. Although these parameters have become widely and variably used, the distribution of the trabecular meshwork length has not been fully investigated from a histological perspective. Also, previous studies showed that PACG occurs more commonly in women than in men.[3–8] However, no studies have investigated the possibility of a difference in TM length between men and women. We speculate that women might

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Correspondence: Shan C. Lin, Department of Ophthalmology, San Francisco School of Medicine, University of California, Box 0730, 10 Koret Way, San Francisco, CA 94143-0730, USA. E-mail: lins@vision.ucsf.edu

have shorter TM length compared to men, thus partly accounting for their higher frequency of PACG.

The aim of this study was to evaluate the trabecular meshwork (TM) length from the scleral spur (SS) to Schwalbe's line (SL) in histological specimens. The results were compared between male and female groups, and inter- and intraobserver reproducibility for the measurements were evaluated.

There are previous reports that evaluated trabecular meshwork length using AS-OCT,[9,10] but to our best knowledge, there is no previous study to evaluate TM length from the scleral spur to Schwalbe's line by using histological specimens.

METHODS

Institutional approval was obtained from the University of California, San Francisco (UCSF) Committee on Human Research and the study adhered to the tenets of the Declaration of Helsinki. The pathological specimens were derived from eyes enucleated for posterior melanoma, which were available within our ocular pathology service. Subjects who had enucleation between 1990 and 2010 were evaluated and considered for enrollment into the study. The slides were made from the enucleated globe were cut through the center of the pupil to the optic nerve and stained with hematoxylin-eosin (HE) or periodic acid schiff (PAS). Images were captured with the Olympus microscope (BX43 with DP72 adaptor, Olympus, Tokyo, Japan) and measurements were performed with the DP2-BSW Version 2.2 software from Olympus.

Our inclusion criteria were (i) age 25 years or more, and (ii) availability of pathological specimen slides from the enucleated eyes of subjects with posterior ocular melanoma. Our exclusion criteria were (i) age less than 25 years, (ii) tumor cell invasion into the ciliary body, iris, or trabecular meshwork, (iii) history of any previous intraocular surgery, (iv) history of eye trauma, (v) history of primary or secondary glaucoma (either clinically or histologically evident), (vi) poor quality slides (such as collapse or folding of the anterior segment), or (vii) slides in which we were unable to appropriately identify SS or SL. Data on age, gender, race, axial length, and eye history were collected from pathological report.

TM length was defined as the length between the tip of the scleral spur to the end of Schwalbe's line (Figure 1). Each specimen contains two angles and both angle's TM length were evaluated. In the previous study utilizing AS-OCT, the scleral spur was defined as the point where there was a change in curvature of the inner surface of the angle wall, often appearing as an inward protrusion of the sclera.[11] In this study, we identified the tip of the scleral spur from the structural change in the transition of the sclera to the TM, specifically, where the dense connective tissue of sclera ends and the meshwork structure starts. SL was defined as the point where Descemet's membrane ends.

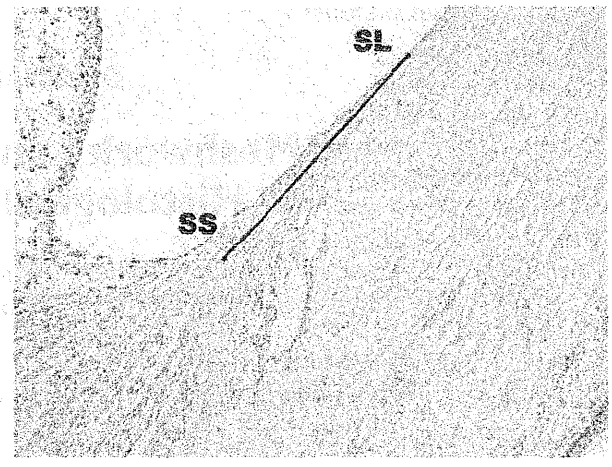

FIGURE 1 Example of trabecular meshwork(TM) length.

To assess the interobserver reproducibility, 158 images with HE staining from 79 subjects were evaluated by two ophthalmologists (TK and YC) masked to each other's results. To assess the intraobserver reproducibility, 79 images with HE stain were randomly chosen and measured again by a single ophthalmologist (TK) in a different session blinded to any previous results. Also, to assess the agreement between the measurements from HE and PAS staining, 30 specimens were selected to have both PAS and HE staining performed in order to compare TM length measurements. A single ophthalmologist (TK) graded both types of staining in blinded fashion. These PAS-stained slides were obtained from leveled sections of the same pathological specimens as HE-stained slides.

The images were also evaluated for detectability of the scleral spurs. Among the 158 angles with HE stain, two ophthalmologists (TK and YC) classified the images into two categories: (i) the scleral spur detectable, or (ii) the scleral spur was difficult to identify.

Statistical Analysis

Overall means and standard deviations were computed for all continuous variables; summary percentages for each category were computed for all discrete variables. Student's *t*-test was conducted to compare the ages, and axial length between male and female group.

Interclass correlation coefficient (ICC) was calculated to assess the inter- and intraobserver reproducibility, and the agreement between HE and PAS stain measurements. Bland-Altman plots[12] were also used to evaluate these agreements. Kappa statistics[6] were calculated to evaluate the agreement between two observers in classifying the image if the scleral spur as detectable or difficult to identify.

In accordance with Landis et al.,[13] the following interpretations for ICC and Kappa statistics were used: slight is <0.2, fair is 0.21–0.40, moderate is 0.41–0.60, substantial is 0.61–0.80, and almost perfect is 0.81–1.00. A p value <0.05 was considered to be statistically significant.

All analyses were performed with the R statistical package (R 2.13 for Macintosh, R Foundation for Statistical Computing, Vienna, Austria).

RESULTS

This study included 158 angle images from 79 cross-sectional slides derived from eyes enucleated for posterior ocular melanoma. Table 1 summarizes the patients' demographics and clinical data for the male and female groups. Of the subjects 40 were male and 39 were female. The mean age of the subjects was 61.8 ± 14.3 years for men and 61 8 ± 12.8 years for women (p = 0.99, Student's t-test). Mean axial length for men was 24.3 ± 1.9 mm and for women was 24.0 ± 1.9 mm (p = 0.36, Student's t-test). Among male subjects, 27 (67.5%) were White and 13 (32.5 %) were of undeclared ethnicity. Among female subjects, 25 (64.1%) were White, 1 (2.6%) was African American, 1 (2.6%) was Vietnamese and 12 (30.8%) were of undetermined ethnicity.

In male group, 19 specimens were right eyes and 21 were left eyes, while in the female group 19 samples were right eyes and 20 were left eyes.

Table 2 provides the results of our measurements. The average trabecular meshwork length measured by TK was 694.9 ± 109.0 µm in the male group and 713.2 ± 106.9 µm in the female group (p = 0.29, Student's t-test). TM length measured by CY was 732.0 ± 127.0 µm in male group and 747.6 ± 113.4 µm in the female group (p = 0.42, Student's t-test).

Figure 2 shows the Bland-Altman plots for the inter- and intrareproducibility and the agreement for the measurements between HE and PAS stain.

The average TM length of all the 158 angles by TK was 703.9 ± 108.0 µm and by YC was 739.7 ± 120.4 µm. ICC for interobserver reproducibility was 0.62 (95% CI: 0.52–0.71).

For the intraobserver reproducibility, ICC (TK as observer) was 0.88 (95% CI: 0.82–0.92) for the randomly selected 79 angles, and the average TM length for the first time and second time were 664.2 ± 101.0 µm and 685.4 ± 102.0 µm, respectively. Average TM length for selected 30 HE stain images was 701.5 ± 107.0 µm and for the PAS couterparts was 702.5 ± 98.6 µm. ICC for the agreement between HE and PAS specimens was 0.89 (95% CI: 0.76–0.95).

No statistically significant difference was found in mean age (p = 0.99), axial length (p = 0.36), and trabecular meshwork length between the male and female groups (p = 0.29) (Student's t-test).

Among the 158 angles graded, one observer (TK) graded 40 images (25.3%) and the other (YC) graded 44 (28.5%) as difficult to identify the scleral spur. Kappa statistics for the two observer's agreement was 0.37 and can be interpreted as fair agreement.

TABLE 1 Characteristics of the study groups.

Parameters	Male group (n = 40)	Female group (n = 39)	p value
Age (years)	61.8 ± 14.3	61.8 ± 12.8	0.99[a]
Axial length (µm)	24.3 ± 1.9	24.0 ± 1.9	0.36[a]
OD:OS	19:21	19:20	0.91[b]
Race, no. (%)			
White	27 (67.5)	25 (64.1)	
African American	0 (0)	1 (2.6)	
Vietnamese	0 (0)	1 (2.6)	
Unknown	13 (32.5)	12 (30.8)	

Data were expressed as mean value ± standard deviation.
[a]Student's t-test.[b]χ^2 test.

TABLE 2 TM length as assessed by two observers.

	Male group (n = 40)	Female group (n = 39)	p value[a]
First observer (TK), (µm)	694.9 ± 109.0	713.2 ± 106.9	0.29
Second observer (YC), (µm)	732.0 ± 127.0	747.6 ± 113.4	0.42
Difference between 2 observers, (µm)	62.6 ± 84.2	62.2 ± 85.5	0.98

Data were expressed as mean value ± standard deviation.
[a]p values are obtained by Student's t-test.

DISCUSSION

In this retrospective study, we evaluated the average length of the trabecular meshwork of human eyes using the histological images obtained from eyes enucleated for posterior ocular melanoma. We didn't find a statistically significant difference in the average TM length between male and female groups. Also, there were no statistically significant differences in ages, and axial length between these two groups. We originally hypothesized that higher frequency of PACG in women is partly due to their shorter trabecular meshwork compared to men; thus their angles were more easily occluded than men. However, our results did not show that the female group had shorter TM length. Previous studies[14,15] reported that women have shallower anterior chambers compared to men, which suggests that their increased prevalence of PACG may be more related to the anterior-to-posterior dimensions of the eye.

Our study is mainly based on the images with HE staining, since this stain is the most available among the study subjects. On the other hand, since PAS staining helps to visualize Descemet's membrane

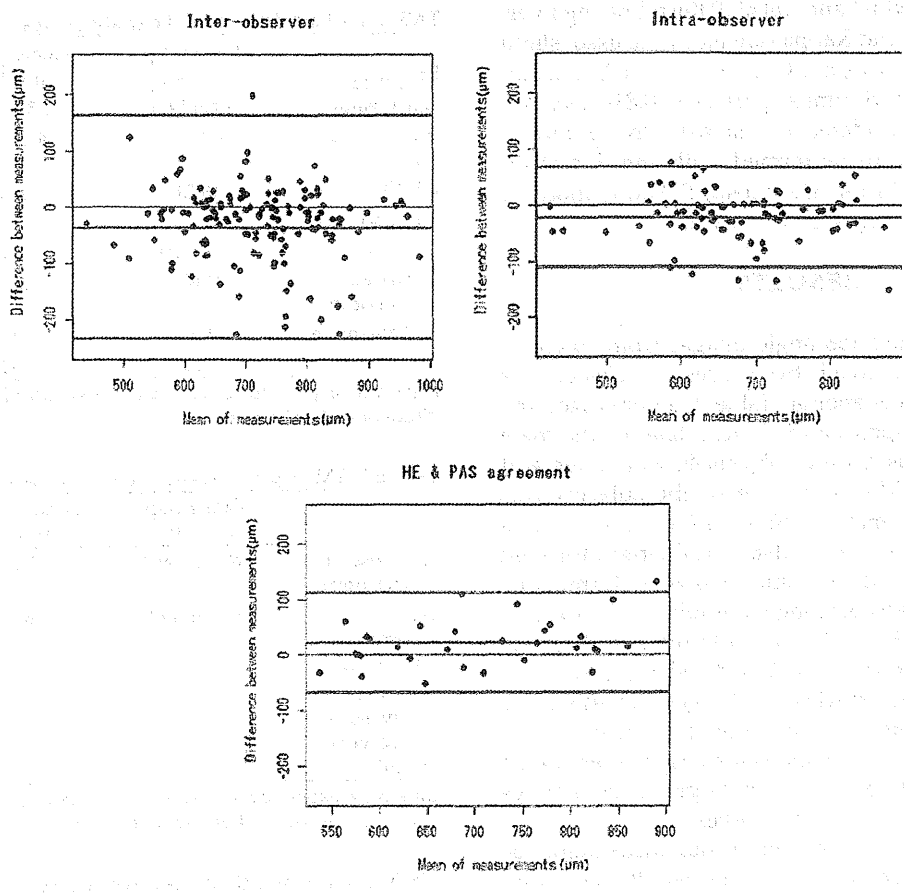

FIGURE 2 Bland-Altman plots of the trabecular meshwork length measurements for inter- and intraobserver reproducibility and HE and PAS stain agreement.

better than with HE staining, we speculated that the PAS stain could identify SL more precisely and thus affect our measurements. However, the agreement of the measurements between HE and PAS staining was classified as almost perfect (ICC:0.89) in this study. We concluded that our measurements were not have been substantially affected by the type of stain.

There are previous reports which have measured TM length using AS-OCT. Cheung et al.[9] reported that the average scleral spur-to-Schwalbe's line length measured by Fourier domain OCT (Cirrus OCT, Carl Zeiss Meditec, Dublin, California) among open-angle patients was 662 μm (n = 58) and closed-angle patients was 681 μm (n = 36). Usui et al.[10] reported the average trabecular meshwork length measured by anterior segment Fourier domain OCT (SS-1000, CASIA, Tomey Co., Nagoya, Japan) was 466.9 ± 60.7 μm (n = 84). Overall, our findings are the longest compared with these previous studies. In part, this may be related to differences in methodologies including the utilization of pathological specimens in our study. In this study, we used the point where the Descemet's membrane ends as Schwalbe's line. However, we've noted that there is often a "gap" between where the trabecular meshwork ends and Schwalbe's line begins. Spencer et al.[16] reported that there is a smooth area called "Zone S" between the apical portion of the trabecular meshwork and the end of the Descemet's membrane which is approximately 50–150 μm. We speculate that the Schwalbe's line found in AS-OCT images could include this "Zone S" and thus locate Schwalbe's line more peripherally, resulting in a shorter measurement. Also, since the majority of our study subjects were White, the results may not directly compare with the previous studies which mainly included Asian populations.

There are several studies that have evaluated the detectability of the scleral spur in the imaging obtained by AS-OCT. Sakata et al.[7] reported the detectability of the scleral spur in images obtained by Visante OCT (Carl Zeiss Meditec, Dublin, CA, USA) from normal populations in Singapore. They reported that the overall detectability of scleral spur in their study was 72% (n = 2008). Usui et al.[4] reported that 100% of scleral spurs were identified in images obtained by the SS-1000 OCT in their study which included 34 normal eyes and 26 eyes with a shallow anterior chamber (van Herick's grading 1 or 2). Cheung et al.[9] reported that 85% of scleral spurs could be identified (n = 117) by Cirrus OCT. Overall, the detectability of scleral spurs varies

between 72–100% in these studies using different OCT machines. We hypothesize that anatomical variation is also related to detectability of scleral spurs, and thus, have performed an assessment of the detectability of the scleral spur using HE slide specimens. Our finding that 69–72% of the images have detectable scleral spurs is close to the report by Sakata et al. The main reason for the difficulty of detecting the scleral spurs in the histological specimens were due to the density of the trabecular meshwork structure and the scleral spurs were almost same in some of the specimens. Although our results are obtained from examination of HE-stained specimens, there is a possibility that other types of stains could visualize the contrast between scleral spur and trabecular meshwork better and thus result in better detectability.

Narayanaswamy et al.[17] reported that among the parameters of AOD at 250, 500, and 750 μm, AOD750 is the most useful angle measurement for identifying individuals with gonioscopic narrow angles in gradable AS-OCT images. In their paper, narrow angle was defined as "the posterior pigmented trabecular meshwork was not visible for at least 180°." Our results showing that the average TM length in men and women is approximately 700 μm appear to support for their conclusions.

Our study has several limitations. First of all, due to the study's retrospective nature, some of the demographic data for the patients such as ethnicity were not available. Although the existence of posterior ocular melanomas should not be a confounding factor in TM measurement, there could still be selection bias since the specimens don't truly represent a normal population. Furthermore, a majority of our subjects were of White descent, so the results may not be applicable to other ethnicities. Finally, there may be artifactual shrinkage of the tissues related to processing, which may actually result in an underestimation of TM length.

In conclusion, we did not find that women have shorter trabecular meshwork distance in than men to possibly help explain the greater risk of angle closure in this population. However our results provide a reference for the average TM length in a small population of eyes.

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REFERENCES

[1] Pavlin CJ, Harasiewicz K, Foster FS. Ultrasound biomicroscopy of anterior segment structures in normal and glaucomatous eyes. *Am J Ophthalmol* 1992;113:381–389

[2] Ishikawa H, Liebmann JM, Ritch R. Quantitative assessment of the anterior segment using ultrasound biomicroscopy. *Curr Opin Ophthalmol* 2000;11.133–139.

[3] Holst JC. A statistical study of glaucoma. *Am J Ophthalmol* 1947;30:1267–1275.

[4] Posner A, Schlossman A. The clinical course of glaucoma; a review of 474 cases from private practice. *Am J Ophthalmol* 1948;31·915–934.

[5] Lemoine AN Jr. Glaucoma; a statistical review of 816 patients with 1,112 glaucomatous eyes. *Am J Ophthalmol* 1950;33:1353–1373.

[6] Smith R. The incidence of the primary glaucomas. *Trans Opthal Soc U K* 1958;78:245–57; discussion 257.

[7] Lowe RF. Comparative incidence of angle-closure glaucoma among different national groups in Victoria, Australia *Br J Ophthalmol* 1963;47:721–727

[8] Foster PJ, Oen FT, Machin D et al. The prevalence of glaucoma in Chinese residents of Singapore: a cross-sectional population survey of the Tanjong Pagar district. *Arch Ophthalmol* 2000;118:1105–1111.

[9] Cheung CY, Zheng C, Ho CL et al. Novel anterior-chamber angle measurements by high-definition optical coherence tomography using the Schwalbe line as the landmark. *Br J Ophthalmol* 2011;95:955–959.

[10] Usui T, Tomidokoro A, Mishima K et al. Identification of Schlemm's canal and its surrounding tissues by anterior segment fourier domain optical coherence tomography. *Invest Ophthalmol Vis Sci* 2011,52:6934–6939.

[11] Sakata LM, Lavanya R, Friedman DS et al. Assessment of the scleral spur in anterior segment optical coherence tomography images. *Arch Ophthalmol* 2008;126:181–185.

[12] Bland JM, Altman DG. Measurement error. *BMJ* 1996;313:744.

[13] Landis JR, Koch GG. The measurement of observer agreement for categorical data. *Biometrics* 1977;33:159–174.

[14] Fontana ST, Brubaker RF. Volume and depth of the anterior chamber in the normal aging human eye. *Arch Ophthalmol* 1980;98:1803–1808.

[15] Olurin O. Anterior chamber depths of Nigerians. *Ann Ophthalmol* 1977;9:315–326.

[16] Spencer WH, Alvarado J, Hayes TL. Scanning electron microscopy of human ocular tissues: trabecular meshwork. *Invest Ophthalmol* 1968;7:651–662.

[17] Narayanaswamy A, Sakata LM, He MG et al. Diagnostic performance of anterior chamber angle measurements for detecting eyes with narrow angles. an anterior segment OCT study. *Arch Ophthalmol* 2010;128:1321–1327.

Association of ARMS2 Genotype With Bilateral Involvement of Exudative Age-Related Macular Degeneration

HIROSHI TAMURA, AKITAKA TSUJIKAWA, KENJI YAMASHIRO, YUMIKO AKAGI-KURASHIGE, ISAO NAKATA, HIDEO NAKANISHI, HISAKO HAYASHI, SOTARO OOTO, ATSUSHI OTANI, AND NAGAHISA YOSHIMURA

- PURPOSE: To study the association of *ARMS2* A69S genotype with the development of exudative age-related macular degeneration (AMD) in the unaffected fellow eye and to estimate the duration until the development of AMD in the second eye.
- DESIGN: Retrospective cohort study.
- METHODS: We retrospectively reviewed 326 patients who had exudative AMD in at least 1 eye, genotyping of *ARMS2* A69S, and a minimum follow-up of 2 years. Survival analysis and Cox proportional hazard regression analysis were used to examine the association between candidate factors and the duration until the development of AMD in the second eye.
- RESULTS: One hundred nineteen patients (36.5%) had bilateral exudative AMD at the initial visit. A risk allele of *ARMS2* A69S was more frequently seen in patients with bilateral AMD ($P = .0270$) than in those with unilateral AMD. Of the 207 unilateral AMD patients, 23 (11.1%) had AMD in the fellow eye after a mean duration of 56.3 ± 40.4 months. Fellow-eye involvement was associated with *ARMS2* A69S genotype (hazard ratio [HR], 2.673; $P = .0013$), age (HR, 1.102; $P = .0005$), and smoking history (HR, 0.680; $P = .3663$). As HRs indicate, correlation of genotype (2.673) was as high as that of 10-year aging ($1.102^{10} = 2.641$). Survival analysis revealed that patients with risk homozygous (TT) genotype had second-eye involvement significantly earlier than those with other genotypes ($P = .0028$). When the observation duration reached 120 months, second-eye involvement had developed in 50%, 6.6%, and 11.2% of the TT, GT, and GG cohorts, respectively.
- CONCLUSION: *ARMS2* A69S genotype is associated with second-eye involvement of exudative AMD and with the period between first- and second-eye involvements. (Am J Ophthalmol 2012;xx:xxx. © 2012 by Elsevier Inc. All rights reserved.)

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From the Department of Ophthalmology and Visual Sciences, Kyoto University Graduate School of Medicine, Kyoto, Japan.

Inquiries to Hiroshi Tamura, Department of Ophthalmology and Visual Sciences, Kyoto University Graduate School of Medicine, 54 Kawahara, Shogoin, Sakyo, Kyoto 606-8507, Japan, e-mail: htamura@kuhp.kyoto-u.ac.jp

EXUDATIVE AGE-RELATED MACULAR DEGENERATION (AMD) is one of the most common vision-threatening eye diseases currently seen in developed countries. Although its exact pathogenesis remains unknown, authors of population-based studies have reported various factors associated with the development of exudative AMD, including age, cataract, sunbathing, sex, history of smoking, hypertension, and soft drusen.[1,2] In the clinical setting, some patients with unilateral exudative AMD maintain good visual function in the fellow eye for a long time, while others have development of exudative AMD in the fellow eye. When visual disturbance attributable to AMD is seen in 1 eye, the impairment of quality of life (QOL) may be limited, but the involvement of exudative AMD in the second eye, when accompanied by a visual disturbance, often causes a severe decrease in QOL. The rate of bilateral involvement of exudative AMD in whites has been reported to vary from 6% to 9% annually.[2–4] In Japanese patients, the rate is relatively low, with a cumulative incidence of only 11% to 12% over 5 years having been documented.[5–8]

Recently, many genetic factors have been reported in the development of exudative AMD, including *ARMS2/HTRA1*, *CFH*, and C2/CFB.[9–14] Although *CFH* is the most prevalent susceptibility gene in whites, *ARMS2/HTRA1* is the most prevalent gene associated with AMD in Asians.[15–17] Andreoli and associates have shown that *ARMS2/HTRA1* is associated with phenotypic attributes of AMD, while *CFH* is not.[18] A higher risk for bilateral advanced disease has been shown in several articles,[13,14] and a higher risk of *ARMS2/HTRA1* for exudative disease than for atrophy has also been described.[19] An increasing number of reports have shown that *ARMS2* A69S is strongly associated with exudative AMD as well as with typical AMD and polypoidal choroidal vasculopathy (PCV). In addition, *HTRA1* polymorphism has been significantly associated with bilateral involvement of exudative AMD,[20] and Sakurada and associates recently reported a significant association between *ARMS2* A69S polymorphism and bilaterality of PCV.[21] Accordingly, it might follow that patients with unilateral exudative AMD have a higher risk for the development of exudative AMD in the fellow eye if they have a risk allele of

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ARMS2 A69S. It would be a great help for both physicians and patients to be better able to estimate the risk of fellow-eye involvement by exudative AMD in order to determine visit frequency and treatment strategy. However, limited information is available about genetic risk factors for fellow-eye involvement of exudative AMD. In the study described herein, we assessed the association of the genotype of *ARMS2* A69S and fellow-eye involvement by exudative AMD. In addition, survival analysis was conducted to estimate the elapsed time from the initial visit for first-eye involvement until second-eye involvement, depending on the particular genotype of *ARMS2* A69S.

PATIENTS AND METHODS

FOR THIS OBSERVATIONAL CASE STUDY, WE REVIEWED retrospectively the medical records of 326 patients with exudative AMD who visited the Macular Service of the Department of Ophthalmology at Kyoto University Hospital between May 1, 2004 and April 30, 2007. Inclusion criteria of this study were 1) exudative AMD in at least 1 eye, 2) initial comprehensive ophthalmic examination of both eyes, and 3) minimum follow-up of 2 years after the initial presentation. The diagnosis of exudative AMD was based primarily on indirect ophthalmoscopy and fluorescein angiography, according to the definition of the International Classification System for Age-Related Maculopathy,[22] but we also used indocyanine angiography and optical coherence tomography (OCT) to make the diagnosis. The current study of AMD included patients with PCV and retinal angiomatous proliferation (RAP). However, patients with other macular abnormalities (ie, pathologic myopia, idiopathic choroidal neovascularization [CNV], presumed ocular histoplasmosis, angioid streaks, and other secondary CNV) were excluded from the study. If detailed examination of either eye was difficult because of ocular disease other than AMD, the patient was also excluded from the study.

Baseline characteristics of the patients were obtained from their medical charts, including age, sex, presence of hypertension and diabetes, and history of smoking. Each patient's smoking status was categorized into never smoker, former smoker, and current smoker, according to the classification by Nakanishi and associates.[23] At the initial visit, each patient underwent a comprehensive ophthalmic examination, including determination of best-corrected visual acuity (VA), intraocular pressure measurement, indirect ophthalmoscopy, slit-lamp biomicroscopy with a contact lens, and OCT examination. After fundus photographs were taken, fluorescein angiography and indocyanine green angiography were performed on each patient, using a confocal laser scanning system (HRA-2; Heidelberg Engineering, Dossenheim, Germany). At each scheduled follow-up visit, each patient underwent a complete ophthalmic examination, including VA measurement, slit-lamp biomicroscopy, indirect fundus ophthalmoscopy, and OCT examination. Fluorescein and indocyanine green angiography was performed if necessary.

TABLE 1. General and Fundus Characteristics in Eyes With Unilateral or Bilateral Exudative Age-Related Macular Degeneration at Initial Presentation.

	Unilateral n = 207	Bilateral n = 119	*P* Value
Sex, n (%)			.1987
Female	68 (32.9)	31 (26.1)	
Male	139 (67.1)	88 (73.9)	
Age (y; mean ± standard deviation)	70.1 ± 7.9	74.0 ± 7.7	<.0001
Smoking, n (%)			.0076
None	97 (46.9)	40 (33.6)	
Former	51 (24.6)	49 (41.2)	
Current	42 (20.3)	28 (23.5)	
Diabetes mellitus, n (%)	20 (9.7)	8 (6.7)	.4798
Hypertension, n (%)	49 (23.7)	24 (20.2)	.4650
Genotype of *ARMS2* A69S (GG/TG/TT)			.0270
GG	43 (20.8)	22 (18.5)	
TG	88 (42.5)	33 (27.7)	
TT	76 (36.7)	64 (53.8)	
Polypoidal lesion in either eye, n (%)	144 (69.6)	65 (54.6)	.0068

TABLE 2. General and Fundus Characteristics in Patients With a New Development of Age-Related Macular Degeneration in the Fellow Eye

	Fellow-Eye Involvement (+) n = 23	Fellow-Eye Involvement (−) n = 184	*P* Value
Sex			.6192
Female	6 (26.1)	62 (33.7)	
Male	17 (73.9)	122 (66.3)	
Age (y; mean ± standard deviation)	69.8 ± 7.9	72.4 ± 7.7	.0110
Smoking (none/former/current)			.0619
None	13 (56.5)	84 (45.7)	
Former	10 (43.5)	51 (27.7)	
Current	0 (0)	42 (22.8)	
Diabetes mellitus	3 (13.0)	17 (9.2)	.8353
Hypertension	6 (26.1)	43 (23.4)	.9769
Polypoidal lesion in the first eye	15 (65.2)	130 (70.7)	.7679
ARMS2 A69S genotype (GG/TG/TT)			.0054
GG	3 (13.0)	40 (21.7)	
TG	4 (17.4)	84 (45.7)	
TT	16 (69.6)	60 (32.6)	

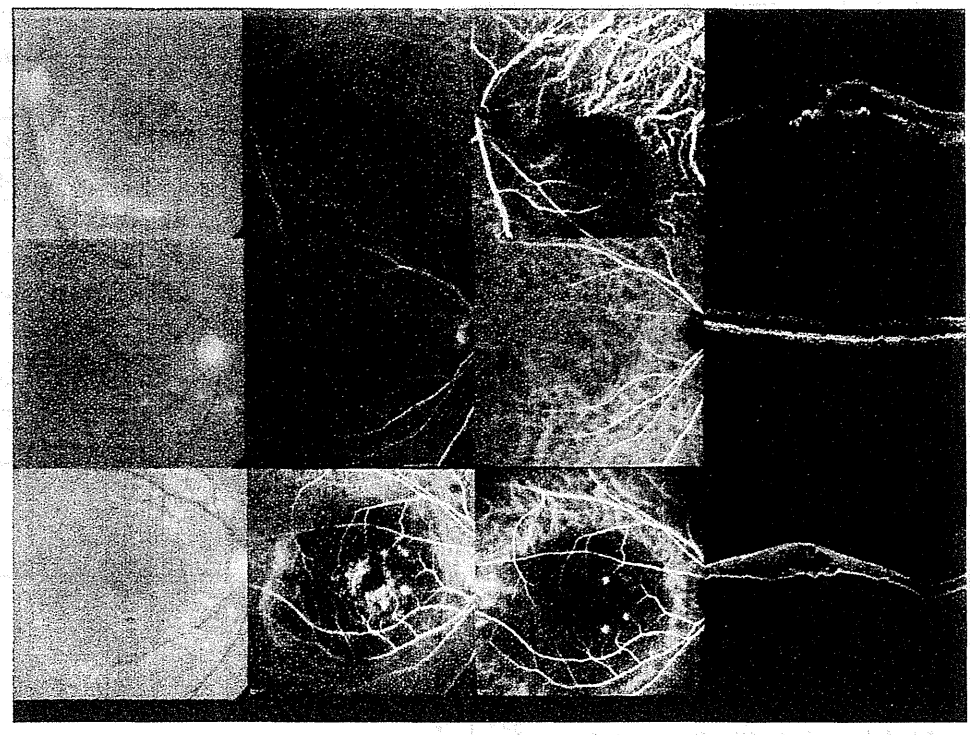

FIGURE 1. Development of exudative age-related macular degeneration in the fellow eye. An 83-year-old woman was referred to our clinic with a 6-month history of metamorphopsia and visual acuity loss in the left eye. At the initial visit, her visual acuity was 20/2000 OS. (Top left) Initial fundus photograph of the left eye shows a grayish lesion with subretinal hemorrhage and hard exudate. (Top, second from left) Fluorescein angiography (FA) shows minimally classic choroidal neovascularization (CNV). (Top, second from right) Indocyanine green angiography (IA) shows blocked fluorescence. (Top right) A sectional image with optical coherence tomography (OCT) shows a pigment epithelial detachment and cystoid macular edema. (Middle left) Initial fundus photograph of the right eye shows only soft drusen in the macular area. No CNV was seen, even by (Middle, second from left) FA, (Middle, second from right) IA, or (Middle right) OCT. Her visual acuity was 20/30 in this eye. (Bottom left) Thirty months after the initial visit, fundus photograph of the right eye shows a grayish exudate and subretinal hemorrhage with a large pigment epithelial detachment. Visual acuity had decreased to 20/130 OD. (Bottom, second from left) FA shows minimally classic CNV corresponding to the lesion seen on fundus photograph. (Bottom, second from right) IA shows retinal angiomatous proliferation. (Bottom right) A sectional image with OCT shows a large pigment epithelial detachment with cystoid macular edema. The genotype of ARMS2 A69S was identified as TT. She had no smoking history and had no known systemic disease.

Preparation of genomic DNA was carried out from peripheral blood using a DNA extraction kit (QuickGene-610L, Fujifilm, Minato, Tokyo, Japan). *CFH* Y402H rs1061170, I62V rs 800292, and *ARMS2* A69S rs10490924 were genotyped via the Taqman SNP assay with the ABI PRISM 7700 system (Applied Biosystems, Foster City, California, USA).

All values are presented as mean ± standard deviation. Statistical analysis among genotypes was performed using χ^2 test for trend or its exact counterpart. In the current study, the date of occurrence of exudative AMD in the second eye was regarded as the date when the physicians documented AMD newly developing in the fellow eye. A Cox proportional hazard regression analysis was conducted to analyze the association between genotype, smoking history, or age with involvement of this fellow eye. In the current study, survival analysis, with the AMD-free period in the better eye after initial visit, was conducted using Kaplan-Meier methods to analyze the relationship between genotype and second-eye involvement. Of the 207 patients, 29 (14%) were lost to follow-up. A difference was considered statistically significant when the *P* value was less than .05.

RESULTS

IN THE CURRENT STUDY, WE EXAMINED 326 PATIENTS (227 male and 99 female) with exudative AMD. The patients ranged in age from 50 to 90 years (71.6 ± 8.0 years) and all were Japanese. Of the 326 patients, 119 (36.5%) were diagnosed as having bilateral exudative AMD at the initial visit. Table 1 shows the general and ocular characteristics of patients with either unilateral or bilateral AMD at the initial visit. There was no significant difference in sex distribution or in coexisting diabetes mellitus or hypertension between patients with unilateral AMD and those with bilateral AMD ($P = .1987$, $P = .4798$, and $P = .4650$). The mean age of patients with bilateral AMD was signif-

TABLE 3. Cox Proportional Hazard Regression Analysis of Relationship Between Genotype, Smoking History, or Age and Duration From Initial Visit to Second-Eye Involvement of Age-Related Macular Degeneration

Variables	Fellow-Eye Involvement (+)	Fellow-Eye Involvement (−)	Hazard Ratio	95% CI	*P* Value
Genotype			2.673	1.443–5.489	.0013
GG	3	40			
TG	4	84			
TT	16	60			
Smoking	(Never & Former) vs Current		0.680	0.286–1.573	.3663
Never & former	23	135			
Current	0	42			
Age (y), mean ± standard deviation	69.8 ± 7.9	72.4 ± 7.7	1.102	1.043–1.169	0005

CI = confidence interval; GG = non-risk homozygous; TG = heterozygous; TT = risk homozygous.

icantly higher than that of patients with unilateral AMD ($P < .0001$), and the proportion of current smokers among bilateral AMD patients was significantly greater than in unilateral AMD patients ($P = .0076$). A risk allele of *ARMS2* A69S was associated significantly with bilaterality of AMD ($P = .027$). In addition, polypoidal lesions were more commonly seen in patients with unilateral AMD than in those with bilateral AMD ($P = .0068$) at the initial visit.

To determine those factors associated with fellow-eye involvement, we further examined 207 patients (139 male and 68 female) with unilateral AMD at the initial visit (Table 2). The mean follow-up duration was 56.0 ± 30.2 months (range, 24–182 months). In 23 of these 207 patients (11.1%), exudative AMD developed in the fellow eye during the follow-up period (Fig. 1). The mean elapsed time from the initial visit until the development of exudative AMD in the fellow eye was 56.3 ± 40.4 months (range, 2–149 months). Table 3 shows general and ocular characteristics of patients with and without fellow-eye involvement. There was no significant difference in sex distribution, smoking, coexisting diabetes mellitus or hypertension, or detection of polypoidal lesion in the first eye between the 2 groups ($P = .6192$, $P = .8353$, $P = .9769$, and $P = .7679$, respectively). The mean age of the fellow eye (−) group was higher than that in the fellow-eye involvement (+) group ($P = .0110$). Regarding the distribution of *ARMS2* A69S genotypes, the GG, TG, and TT genotypes were seen in 3, 4, and 16 patients with fellow-eye involvement, respectively, while seen in 40, 84, 60 patients without fellow-eye involvement, respectively. The risk allele of *ARMS2* A69S was significantly associated with fellow-eye involvement ($P = .0054$). In contrast, no association was observed with *CFH* Y402H rs1061170 or I62V rs 800292 in the current study.

Fellow-eye involvement was associated with *ARMS2* A69S genotype (hazard ratio, 2.673; 95% CI, 1.443–5.489; $P = .0013$), age (hazard ratio, 1.102; 95% CI, 1.043–1.169; $P = .0005$), and smoking history (hazard ratio, 0.680; 95% CI, 0.286–1.573; $P = .3663$), in decreasing order (Table 3). As hazard ratios indicate, correlation of genotype (2.673) was as high as that seen with 10 years of aging ($1.102^{10} = 2.641$).

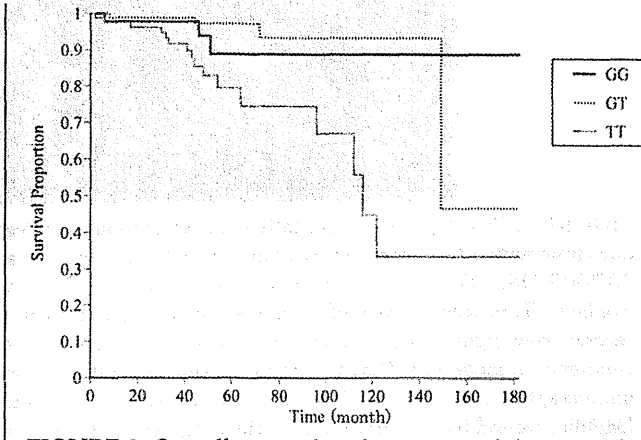

FIGURE 2. Overall survival analysis curve of the period free from second-eye involvement by age-related macular degeneration among patients with discrete genotypes of *ARMS2* A69S. Patients with the risk homozygous genotype (TT) experienced second-eye involvement in a significantly shorter period of time than did those with other genotypes ($P = .0028$). At 120 months after the initial visit, 50% of TT patients presented with second-eye involvement, while only 6.6% of GT patients and 11.2% of GG patients had second-eye involvement.

Survival analysis for the AMD-free duration in the second eye revealed that the risk homozygous TT genotype caused second-eye involvement significantly earlier than other genotypes ($P = .0028$). The median survival time was 120 months for the TT cohort, was 150 months for the TG cohort, and was not determined for the GG cohort. When the observation duration reached 120 months, second-eye involvement was seen in 50% of the TT cohort, compared with 6.6% of the GT cohort and 11.2% of the GG cohort (Figure 2).

DISCUSSION

TO DATE, VARIOUS RISK FACTORS FOR AMD HAVE BEEN seen in cohort studies, including the Age-Related Eye Disease Study (AREDS), the Beaver Dam Eye Study, the Rotterdam Study, and the Blue Mountains Eye Study.[1,2] From these reports, it is generally recognized that smoking and age are common risk factors for any type of AMD.[1] The AREDS recommended supplementation, a combination of zinc and antioxidants (β-carotene, vitamin C, and vitamin E); this produced a 25% reduction in the incidence of advanced AMD over 5 years and a 19% reduction in severe vision loss in those deemed to be at high risk of having an advanced form of the disease.[2] However, dietary supplementation cannot completely prevent AMD or its fellow-eye involvement. Furthermore, the response to this AREDS supplementation is reported to be related to genotypes.[24]

Both in whites and in Asians, *CFH* and *ARMS2/HTRA1* genes seem to be the major susceptibility genes for AMD.[9,10,13,14] Although in whites, *CFH* is the most significantly associated gene, followed by *ARMS/HTRA1*, AMD in Asian patients showed a stronger association with *AMRS2/HTRA1* than with *CFH*.[17,25] A phenotypic study for AMD revealed that *ARMS2/HTRA1* is associated with visual acuity, RPE hyperpigmentation, drusen size, and CNV size, while *CFH* is not associated—at least in the Japanese population.[25] We have also demonstrated that, unlike *CFH*, *ARMS2/HTRA1* is associated with CNV size in both AMD and PCV,[16] and is also significantly associated with bilaterality of these conditions.[13,20,21] Furthermore, recent reports have shown that the *ARMS2/HTRA1* genotype affects visual prognosis of AMD and PCV—even after photodynamic therapy.[26–28]

In the current study, a risk allele (T) of *ARMS2* A69S was more frequently seen in patients having bilateral AMD at the initial presentation than in those having unilateral presentation. However, even in patients with unilateral AMD at the initial visit, the *ARMS2* A69S risk allele is associated with a higher risk for the development of exudative AMD in the fellow eye. As far as our literature survey could ascertain, there have been no reports on the relationship between *ARMS2* and the AMD-free period in the second eye after the initial presentation. Survival analysis revealed that patients with the TT homozygous genotype presented with second-eye involvement significantly earlier than did patients with other genotypes. When the observation duration reached 120 months, second-eye involvement was evident in 50% of the TT cohort.

The current study also showed that patients with other genotypes of *ARMS2* A69S had a lower risk for bilateral AMD. Patients that do not have risk homozygous *ARMS2* A69S are estimated to have about a 10% risk of having fellow-eye involvement by AMD in 10 years, which may be of help to physicians who are determining the endpoint of treatment of the first eye with advanced AMD, especially when visual function is poor. If visual disturbance is limited to 1 eye because of AMD and other ocular diseases, the quality of life may be not impaired greatly, but once the second eye is also involved and the visual function of both eyes is impaired, QOL will be significantly damaged.[29] These academic discussions have been applied already to clinical practice, as is clear in the assessment for amblyopia screening in Health Technology Assessment.[30]

Smoking status and age at the initial visit are also risk factors for bilateral AMD. In the EUREYE study, patients with bilateral AMD tended to have a heavier smoking history than did those with unilateral involvement.[31] On the other hand, Sakurada and associates did not report any association of smoking history with bilateral development of PCV.[21] In the current study, smoking status had a significant association if bilateral AMD was diagnosed at the initial visit, but had no significant association with second-eye involvement by AMD or with the duration until second-eye involvement. Of smokers at the initial visit, a considerable proportion stopped smoking after being informed that smoking is the major risk factor for AMD. Thus smoking status at the initial visit may not be the best explanatory variable for the second-eye involvement model. There remains conflicting evidence about the relationship between smoking and second-eye involvement by AMD, and the influence of smoking seems to require more investigation with a larger body of data, although in the current study, aging was correlated significantly with second-eye involvement by AMD, which is consistent with previous findings.[32] As the hazard ratios indicate that the correlation of genotype to second-eye involvement (2.673) was as high as that of 10 years of aging ($1.102^{10} = 2.641$), the genotype of *ARMS2* A69S has as strong an association with second-eye involvement as 10 years of aging.

The current study has several limitations that need to be pointed out. First, this investigation was conducted as a retrospective study of relatively small size. Second, elderly patients (over 80 years of age at the initial visit) were included in the current study, and it might be inappropriate to include such elderly patients for estimation of the future occurrence of AMD in the second eye. Third, exudative AMD includes subgroups such as PCV and RAP. It has been reported that typical AMD and PCV have a similar probability of involvement of the fellow eye in unilaterally affected Japanese patients, even though PCV and RAP have different clinical presentations. Finally, dietary supplementation was not considered in the current study, and it is possible that such supplements may contribute to the avoidance of second-eye affection.

In the current research, we reconfirmed the association of *ARMS2* A69S genotype with second-eye involvement of

AMD and found an association with elapsed time until second-eye involvement. However, future research involving more candidate genes and other possible factors may reveal more precisely the future risks of fellow-eye involvement by AMD.

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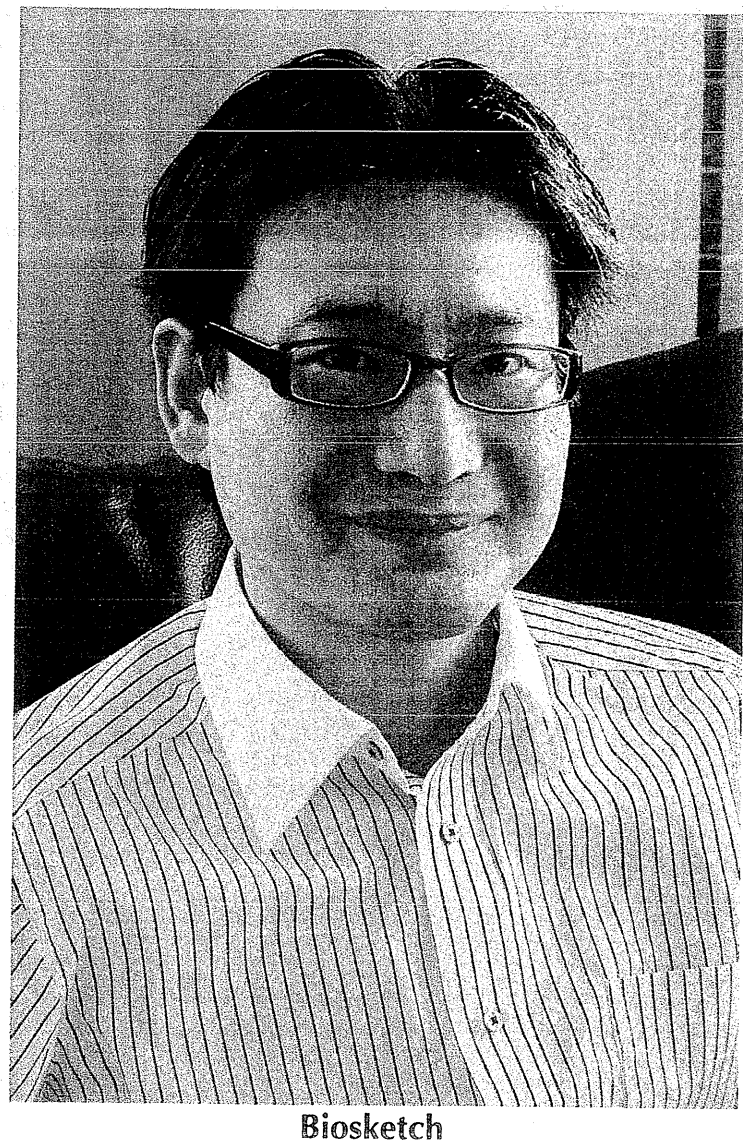
REFERENCES

1. Smith W, Assink J, Klein R, et al. Risk factors for age-related macular degeneration: pooled findings from three continents. *Ophthalmology* 2001;108(4):697–704.
2. AREDS Study Group. A randomized, placebo-controlled, clinical trial of high-dose supplementation with vitamins C and E, beta carotene, and zinc for age-related macular degeneration and vision loss: AREDS report no. 8 *Arch Ophthalmol* 2001;119(10):1417–1436.
3 Macular Photocoagulation Study Group. Five-year follow-up of fellow eyes of patients with age-related macular degeneration and unilateral extrafoveal choroidal neovascularization. *Arch Ophthalmol* 1993;111(9):1189–1199
4. Sandberg MA, Weiner A, Miller S, Gaudio AR. High-risk characteristics of fellow eyes of patients with unilateral neovascular age-related macular degeneration. *Ophthalmology* 1998;105(3):441–447.
5. Uyama M, Takahashi K, Ida N, et al. The second eye of Japanese patients with unilateral exudative age related macular degeneration. *Br J Ophthalmol* 2000;84(9):1018–1023.
6 Yuzawa M, Hagita K, Egawa T, et al. Macular lesions predisposing to senile disciform macular degeneration *Jpn J Ophthalmol* 1991;35(1):87–95.
7. Ueta T, Iriyama A, Francis J, et al. Development of typical age-related macular degeneration and polypoidal choroidal vasculopathy in fellow eyes of Japanese patients with exudative age-related macular degeneration. *Am J Ophthalmol* 2008;146(1):96–101.
8. Mori K, Horie-Inoue K, Gehlbach PL, et al. Phenotype and genotype characteristics of age-related macular degeneration in a Japanese population. *Ophthalmology* 2010;117(5):928–938.
9. Edwards AO, Ritter R 3rd, Abel KJ, et al. Complement factor H polymorphism and age-related macular degeneration. *Science* 2005;308(5720):421–424.
10. Klein RJ, Zeiss C, Chew EY, et al. Complement factor H polymorphism in age-related macular degeneration. *Science* 2005;308(5720):385–389.
11. Magnusson KP, Duan S, Sigurdsson H, et al. CFH Y402H confers similar risk of soft drusen and both forms of advanced AMD. *PLoS Med* 2006;3(1):e5.
12. Rivera A, Fisher SA, Fritsche LG, et al. Hypothetical LOC387715 is a second major susceptibility gene for age-related macular degeneration, contributing independently of complement factor H to disease risk. *Hum Mol Genet* 2005;14(21):3227–3236.
13. Seddon JM, Francis PJ, George S, et al. Association of CFH Y402H and LOC387715 A69S with progression of age-related macular degeneration. *JAMA* 2007;297(16):1793–1800.
14. Seddon JM, Reynolds R, Maller J, et al. Prediction model for prevalence and incidence of advanced age-related macular degeneration based on genetic, demographic, and environmental variables. *Invest Ophthalmol Vis Sci* 2009;50(5):2044–2053.
15. Gotoh N, Yamada R, Hiratani H, et al. No association between complement factor H gene polymorphism and exudative age-related macular degeneration in Japanese. *Hum Genet* 2006;120(1):139–143.
16. Gotoh N, Yamada R, Nakanishi H, et al. Correlation between CFH Y402H and HTRA1 rs11200638 genotype to typical exudative age-related macular degeneration and polypoidal choroidal vasculopathy phenotype in the Japanese population. *Clin Experiment Ophthalmol* 2008;36(5):437–442.
17. Hayashi H, Yamashiro K, Gotoh N, et al. CFH and ARMS2 variations in age-related macular degeneration, polypoidal choroidal vasculopathy, and retinal angiomatous proliferation. *Invest Ophthalmol Vis Sci* 2010;51(11):5914–5919.
18. Andreoli MT, Morrison MA, Kim BJ, et al. Comprehensive analysis of complement factor H and LOC387715/ARMS2/HTRA1 variants with respect to phenotype in advanced age-related macular degeneration. *Am J Ophthalmol* 2009;148(6):869–874.
19. Sobrin L, Reynolds R, Yu Y, et al. ARMS2/HTRA1 locus can confer differential susceptibility to the advanced subtypes of age-related macular degeneration. *Am J Ophthalmol* 2011;151(2):345–352.e3.
20. Chen H, Yang Z, Gibbs D, et al. Association of HTRA1 polymorphism and bilaterality in advanced age-related macular degeneration. *Vision Res* 2008;48(5):690–694.
21. Sakurada Y, Kubota T, Imasawa M, et al. Role of complement factor H I62V and age-related maculopathy susceptibility 2 A69S variants in the clinical expression of polypoidal choroidal vasculopathy. *Ophthalmology* 2011;118(7):1402–1407.
22. Bird AC, Bressler NM, Bressler SB, et al. An international classification and grading system for age-related maculopathy

and age-related macular degeneration. The International ARM Epidemiological Study Group. *Surv Ophthalmol* 1995;39(5):367–374.
23. Nakanishi H, Yamashiro K, Yamada R, et al. Joint effect of cigarette smoking and CFH and LOC387715/HTRA1 polymorphisms on polypoidal choroidal vasculopathy. *Invest Ophthalmol Vis Sci* 2010;51(12):6183–6187.
24. Klein ML, Francis PJ, Rosner B, et al. CFH and LOC387715/ARMS2 genotypes and treatment with antioxidants and zinc for age-related macular degeneration. *Ophthalmology* 2008; 115(6):1019–1025.
25. Goto A, Akahori M, Okamoto H, et al. Genetic analysis of typical wet-type age-related macular degeneration and polypoidal choroidal vasculopathy in Japanese population. *J Ocul Biol Dis Infor* 2009;2(4):164–175.
26. Sakurada Y, Kubota T, Imasawa M, et al. Association of LOC387715 A69S genotype with visual prognosis after photodynamic therapy for polypoidal choroidal vasculopathy. *Retina* 2010;30(10):1616–1621.
27. Bessho H, Honda S, Kondo N, Negi A. The association of age-related maculopathy susceptibility 2 polymorphisms with phenotype in typical neovascular age-related macular degeneration and polypoidal choroidal vasculopathy. *Mol Vis* 2011;17:977–982.
28. Tsuchihashi T, Mori K, Horie-Inoue K, et al. Complement factor H and high-temperature requirement A-1 genotypes and treatment response of age-related macular degeneration. *Ophthalmology* 2011;118(1):93–100.
29. Brown GC. Vision and quality-of-life. *Trans Am Ophthalmol Soc* 1999;97:473–511.
30. Carlton J, Karnon J, Czoski-Murray C, et al. The clinical effectiveness and cost-effectiveness of screening programmes for amblyopia and strabismus in children up to the age of 4-5 years: a systematic review and economic evaluation. *Health Technol Assess* 2008;12(25):iii, xi-194.
31. Chakravarthy U, Augood C, Bentham GC, et al. Cigarette smoking and age-related macular degeneration in the EUREYE Study. *Ophthalmology* 2007;114(6):1157–1163.
32. Zanke B, Hawken S, Carter R, Chow D. A genetic approach to stratification of risk for age-related macular degeneration. *Can J Ophthalmol* 2010;45(1):22–27.



Biosketch

Hiroshi Tamura, MD, PhD, is a graduate of the Kyoto University Graduate School of Medicine, Kyoto, Japan. He completed an ophthalmology residency and fellowship at Kobe City General Hospital and Kyoto University Hospital in Japan. Following the fellowship, he has worked at Kyoto University Hospital as assistant professor. He also participated in 2011 Summer Institute of Epidemiology and Biostatistics at the Johns Hopkins School of Public Health, Baltimore, Maryland, and 2011 Program in Clinical Effectiveness at the Harvard School of Public Health, Boston, Massachusetts.

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CLINICAL INVESTIGATION

Visual prognosis of eyes with submacular hemorrhage associated with exudative age-related macular degeneration

Naoko Ueda-Arakawa · Akitaka Tsujikawa · Kenji Yamashiro · Sotaro Ooto · Hiroshi Tamura · Nagahisa Yoshimura

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Abstract
Purpose To study the retinal structural changes associated with submacular hemorrhage due to exudative age-related macular degeneration (AMD) and their relationships with visual prognosis.
Methods We retrospectively reviewed the medical records of 31 consecutive patients (31 eyes) with visual impairment due to an acute submacular hemorrhage associated with typical AMD (10 eyes) or polypoidal choroidal vasculopathy (21 eyes).
Results Optical coherence tomography (OCT) revealed that submacular hemorrhage exhibited intense hyperreflectivity beneath the neurosensory retina and often seemed to infiltrate it. In the OCT sections, mild to moderate amorphous hyperreflectivity and/or hyperreflective dots were observed within the neurosensory retina, resulting in the loss of the junctions between the inner (IS) and outer (OS) segments of the photoreceptors. Of the 31 eyes, the foveal IS/OS line could be seen incompletely in 12 eyes and was totally absent in 16 eyes. The initial integrity of the foveal photoreceptor layer was correlated with the final visual acuity; the initial detection of the IS/OS just beneath the fovea was correlated with good final visual acuity ($r = 0.375$, $p = 0.038$).
Conclusion As a hallmark of integrity of the foveal photoreceptor layer, the initial detection of the IS/OS just beneath the fovea may predict good visual outcomes.

N. Ueda-Arakawa · A. Tsujikawa (✉) · K. Yamashiro · S. Ooto · H. Tamura · N. Yoshimura
Department of Ophthalmology and Visual Sciences, Kyoto University Graduate School of Medicine, 54 Shogoin-Kawahara-cho, Sakyo-ku, Kyoto 606-8507, Japan
e-mail: tujikawa@kuhp.kyoto-u.ac.jp

Keywords Age-related macular degeneration · Optical coherence tomography · Polypoidal choroidal vasculopathy · Submacular hemorrhage

Introduction

Submacular hemorrhage is a vision-threatening complication associated with exudative age-related macular degeneration (AMD) [1]. It can cause a sudden loss of central vision, often resulting in permanent visual loss [2]. The natural visual prognosis of submacular hemorrhage is extremely poor [3–6]. In a retrospective study by Bennett et al. [3], the mean initial visual acuity (VA) in eyes with submacular hemorrhage (20/860) improved to no better than 20/480 after a mean follow-up of three years. Numerous surgical procedures and several modifications are described, aimed at improving the poor visual prognosis [3–14]. For acute large submacular hemorrhage, surgical drainage or pneumatic displacement either with or without tissue plasminogen activator is thought to improve the visual prognosis [8, 11–14]. However, the effectiveness of subretinal clot removal is limited [7, 8].

The exact mechanism as to why a thick submacular hemorrhage can cause sudden visual loss remains unclear, even when the sensory retina within the macular area can be clearly seen on fundus examination [2]. Histologic reports show that submacular hemorrhage can cause substantial damage to the outer retina [16, 17]. In experimental studies with an animal model, Glatt and Machemer [18] report that the photoreceptors overlying areas of the hemorrhage appear to have degenerated and exhibit pyknosis within 24 h. Subsequent reports suggest other mechanisms of damage to the photoreceptor layer, including clot

retraction [19], iron toxicity [20, 21], and blockage of nutrient diffusion [2].

Optical coherence tomography (OCT) enables the detection of changes in the retinal architecture and quantitative measurement of retinal thickness [22–25]. Recent advances in OCT technology contributed to a more detailed understanding of the pathomorphology of many macular diseases [26–28]. However, there is still little information available on morphologic changes in the neurosensory retina associated with the submacular hemorrhage [15]. In addition, various factors are thought to be associated with visual prognosis in eyes with a submacular hemorrhage, including hemorrhage size [4, 6], elevation of the overlying retina [3, 4, 6], and the etiology of the original disease [3, 5]. However, quantitative evaluations are limited. In the present study, we studied OCT sections of eyes with submacular hemorrhage associated with exudative AMD. This was done to elucidate the structural changes in the overlying neurosensory retina in order to measure each characteristic manifestation quantitatively and to study the associations between the structural changes and visual function. Based on the results of these evaluations, we can now prognosticate the visual outcome of eyes with acute submacular hemorrhage.

Subjects and methods

For this observational case study, we retrospectively reviewed the medical records of 31 consecutive patients (31 eyes) with visual impairment due to an acute thick submacular hemorrhage associated with exudative AMD; each of these patients had visited the Macula Service of the Department of Ophthalmology at Kyoto University Hospital between April 2007 and March 2010. Of the 31 eyes, 10 had typical AMD and 21 had polypoidal choroidal vasculopathy (PCV). All 31 eyes included in this study exhibited a submacular hemorrhage just beneath the center of the fovea. Eyes with either a small subretinal hemorrhage with <1 disc area or an old yellowish discolored submacular hemorrhage were excluded. Each patient underwent angiography with a confocal laser scanning system (HRA-2, Heidelberg Engineering, Heidelberg, Germany). In the current study, the presence of choroidal neovascularization (CNV) was confirmed with fluorescein and indocyanine green angiography, which were performed either at the initial visit or after the resolution of the submacular hemorrhage. The diagnosis of PCV was based on indocyanine green angiography, which shows a branching vascular network that terminates in polypoidal swelling. Eyes with other macular abnormalities (e.g., pathologic myopia, idiopathic CNV, presumed ocular histoplasmosis, angioid streaks, other secondary CNV, or retinal arterial macroaneurysm) were excluded. Of the 31 eyes of our patients, 21 were treated with intravitreal injections of antivascular endothelial growth factor agents, 2 by photodynamic therapy, and 1 by pneumatic displacement of the submacular hemorrhage. This study was approved by the Institutional Review Board of the Kyoto University Graduate School of Medicine and adhered to the tenets of the Declaration of Helsinki.

At the initial examination after the submacular hemorrhage had occurred, each patient underwent a comprehensive ophthalmologic examination, including measurement of best-corrected VA, determination of intraocular pressure, indirect ophthalmoscopy, slit-lamp biomicroscopy with a non-contact lens, and examinations with the Spectralis HRA+OCT (Heidelberg Engineering). Using these initial OCT images, we measured the thickness of the neurosensory retina, the thickness of the submacular hemorrhage, and the total foveal thickness (Fig. 1). When the retinal pigment epithelium (RPE) was not visible under the fovea because of an overlying thick submacular hemorrhage, the OCT measurements were made from a presumed RPE line obtained from a clearly detectable RPE line in a more peripheral retinal area not covered by the hemorrhage. Furthermore, to assess the integrity of the outer foveal photoreceptor layer, we examined the junction between the inner and outer segments of the photoreceptor (IS/OS) line and the external limiting membrane (ELM) line in the fovea. The status of the IS/OS and ELM lines under the fovea was defined as complete, incomplete, or absent. To assess the density of the submacular hemorrhage, the detection of the underlying RPE line was also determined as complete, incomplete, or absent.

At the final examination, best-corrected VA was measured and indirect ophthalmoscopy, slit-lamp biomicroscopy with a noncontact lens, and examinations with the Spectralis HRA+OCT were performed, and all eyes showed complete resolution of the submacular hemorrhage beneath the fovea. After the submacular hemorrhage was resolved, the patients often exhibited a subfoveal mass, which appeared as a subretinal deposit or fibrosis on the fundus photographs. Since it was often difficult to determine its border with respect to the RPE, we performed three measurements in the fovea using the OCT images obtained at the final examination, including the thickness of the neurosensory retina, thickness of the subretinal mass and Bruch's membrane, and the total thickness (Fig. 1). The thickness of the neurosensory retina was defined as the distance between the vitreoretinal interface and the inner border of hyperreflectivity of either the RPE or the subretinal mass. The thickness of the subretinal mass and Bruch's membrane was defined as the distance between the outer border of the neurosensory retina and the fine straight line of the elastic fiber layer of the Bruch's membrane. Total thickness was defined as the distance between the