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Morphology and Function of Meibomian Glands and Other Tear Film Parameters in Junior High School Students

Takanori Mizoguchi, MD,† Reiko Arita, MD, PhD,†‡ Shima Fukuoka, MD,†§ and Naoyuki Morishige, MD, PhD†¶*

Purpose: We measured tear film parameters, including the morphology and function of meibomian glands, in junior high school students at 15 years of age.

Methods: A total of 111 eyes of 111 students (56 males and 55 females) were enrolled in the study. The ocular symptom score (0–14), after-school study time, lipid layer thickness (LLT) of the tear film, partial blink rate, lid margin abnormalities (0–4), tear film breakup time, corneal and conjunctival epithelial damage (fluorescein staining score, 0–9), meiboscore as determined by noncontact meibography (0–6), Schirmer test value, and meibum grade (0–3) were determined. The relationships between parameters were evaluated with the Spearman correlation coefficient (ρ).

Results: The meiboscore was 2.8 ± 1.2, and the meibum grade was 1.8 ± 1.2. The meiboscore significantly correlated with the meibum grade (ρ = 0.272, $P = 0.004$), Schirmer test value (ρ = −0.220, $P = 0.021$), and LLT (ρ = −0.264, $P = 0.005$). The breakup time significantly correlated with LLT (ρ = 0.261, $P = 0.006$), meibum grade (ρ = −0.338, $P < 0.001$), and fluorescein staining score (ρ = −0.214, $P = 0.025$). The partial blink rate significantly correlated with the Schirmer test value (ρ = −0.240, $P = 0.011$). The meiboscore ($P < 0.001$) and meibum grade ($P = 0.032$) were significantly greater in males than in females.

Conclusions: The morphology and function of meibomian glands are altered even at 15 years of age, with the changes being more prominent in males than in females.

Key Words: meibomian gland, tear film, meibography, adolescence

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From the *Mizoguchi Eye Clinic, Sasebo, Nagasaki, Japan; †Lid and Meibomian Gland Working Group (LIME), Tokyo, Japan; ‡Itoh Clinic, Saitama, Japan; §Omiya Hamada Eye Clinic, Saitama, Japan; and ¶Division of Cornea and Ocular Surface, Ohshima Eye Hospital, Fukuoka, Japan.

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Reprints: Reiko Arita, MD, PhD, Department of Ophthalmology, Itoh Clinic, 626-11 Minami-Nakano, Minumaku, Saitama 337-0042, Japan (e-mail: ritoh@za2.so-net.ne.jp).

Dry eye disease is one of the most common conditions of the anterior segment worldwide, with a prevalence of 3.9% to 50% among various populations.[1–6] Meibomian gland dysfunction (MGD), a chronic abnormality of meibomian glands characterized by terminal duct obstruction or qualitative or quantitative changes in glandular secretion,[7] is one of the most important underlying causes of dry eye disease.[8] The prevalence of MGD increases with age[9] and was previously found to be significantly increased in persons older than 55,[10] 60,[11,12] 65,[13] or 70 years[14] compared with younger individuals. We have previously found no abnormalities in meibomian gland morphology in infants and pre–high school children.[11,15] However, the functional and morphological properties of meibomian glands in adolescents have not previously been described. Adolescents experience pronounced changes in the levels of various hormones and might therefore be expected to manifest changes in meibomian glands and tear film parameters compared with younger individuals. We have now measured meibomian gland and other tear film–related parameters in junior high school students at 15 years of age.

SUBJECTS AND METHODS

This prospective cross-sectional study recruited ninth grade students (15 years of age during the school year) at a junior high school in Sasebo City, Nagasaki Prefecture, Japan, as subjects, with written informed consent being obtained from the parents for participation of the subjects. Exclusion criteria were eye diseases of the anterior segment, such as allergic conjunctivitis and atopic dermatitis, other systemic diseases that might be expected to affect the anterior segment, a history of ocular surgery, use of oil-based cosmetics, and regular wear of contact lenses. We examined the right eye of each subject unless it was affected by the excluding conditions, in which case the left eye was examined. A total of 111 eyes of 111 subjects (56 males and 55 females) were enrolled in the study. The study was performed in accordance with the Declaration of Helsinki and was approved by the ethics committee of Mizoguchi Eye Clinic. All participants were instructed not to apply any type of eye drops for at least 2 hours before examinations.

Ophthalmologic examinations were performed according to a previously described procedure.[16] All subjects were questioned regarding the presence of 14 ocular symptoms or signs[16] (ocular fatigue, discharge, foreign body sensation, dryness, uncomfortable sensation, sticky sensation, pain,

epiphora, itching, redness, heavy sensation, glare, excessive blinking, and history of chalazion or hordeolum). The symptoms or signs were scored from 0 to 14 according to the number present. The subjects were also asked about the total number of hours spent studying after school (including those spent on a computer or smartphone). The lipid layer thickness (LLT) of the tear film and the partial blink rate were measured using the LipiView instrument (TearScience, Morrisville, NC). Room temperature (25.2 ± 2.1°C) and humidity (41.2% ± 5.8%) were maintained relatively constant for all examinations. LLT was measured once under natural blinking conditions. Four lid margin abnormalities (irregular lid margin, vascular engorgement, plugged meibomian gland orifices, and anterior or posterior displacement relative to the mucocutaneous junction) were scored from 0 to 4 according to the number present in the examined eye.[16] The tear film breakup time (BUT) was measured after instillation of 1 μL of preservative-free 1% fluorescein dye in the conjunctival sac with a micropipette. The subjects were asked to blink several times, after which BUT was measured 3 times consecutively with a stopwatch, and the average of the 3 measurements was calculated.[17] The fluorescein staining score of the ocular surface was determined for nasal conjunctival, corneal, and temporal conjunctival areas, with each area being scored from 0 to 3 points to yield a total score of 0 to 9 points.[18] Meibomian glands of the upper and lower eyelids were evaluated using a noncontact meibography system (Topcon, Tokyo, Japan), and the meiboscore (0–3) for each eyelid was summed to give a total score of 0 to 6. Tear production was measured by the Schirmer method without topical anesthesia. Finally, digital pressure was applied to the central 5 meibomian glands of the upper eyelid, and the degree of ease with which meibum secretion was induced and meibum quality were evaluated semiquantitatively as follows: 0, clear meibum easily expressed; 1, cloudy meibum expressed with mild pressure; 2, cloudy meibum expressed with more than moderate pressure; and 3, meibum not expressed even with strong pressure.[19]

Data are presented as mean ± SD. The relationships between measured parameters were assessed by calculation of the Spearman correlation coefficient. The association of fluorescein staining of the ocular surface with lid margin abnormalities was evaluated with the Fisher exact test. Differences were assessed with the Mann–Whitney *U* test. All statistical analyses were performed using JMP Pro version 11 software (SAS, Cary, NC). $P < 0.05$ was considered statistically significant.

RESULTS

The characteristics of the subjects are presented in Table 1 and Figure 1. The relationship between the presence or absence of lid margin abnormalities and fluorescein staining of the ocular surface is presented in Table 2. Fluorescein staining was more frequent in eyes with vascularity of lid margins than in those without vascularity ($P = 0.032$). No significant difference was detected in the frequency of fluorescein staining between eyes with or without plugging ($P = 0.5$). The correlations between measured parameters are presented in Table 3. Significant correlations were detected between the meibum grade of the upper eyelid and tear film BUT ($\rho = -0.338$, $P < 0.001$), between LLT and the meiboscore ($\rho = -0.264$, $P = 0.005$), between the meiboscore and meibum grade ($\rho = 0.272$, $P = 0.004$), between the partial blink rate and Schirmer test value ($\rho = -0.240$, $P = 0.011$), and between BUT and LLT ($\rho = 0.261$, $P = 0.006$). Of note, significant correlations were not detected between lid margin abnormalities and either the meiboscore or meibum grade.

TABLE 1. Characteristics of the Study Population (111 Eyes of 111 Subjects)

Parameter	Mean ± SD	Range
Ocular symptom score (0–14)	3.7 ± 2.3	0–13
Study time, h	3.5 ± 1.3	1–11
LLT, nm	68.7 ± 23.1	29–134
Partial blink rate (0–1)	0.8 ± 0.3	0–1
Lid margin abnormalities (0–4)	0.1 ± 0.3	0–2
BUT, s	8.6 ± 7.2	2.0–33.6
Fluorescein staining score (0–9)	1.1 ± 1.4	0–6
Meiboscore (0–6)	2.8 ± 1.2	0–5
Schirmer test value, mm	20.2 ± 11.5	0–35
Meibum grade (0–3)	1.8 ± 1.2	0–3

Sex differences in the test values are presented in Table 4. Significant sex differences were apparent in the meiboscore (males, 3.3 ± 1.1; females, 2.4 ± 1.2; $P < 0.001$) and meibum grade (males, 2.0 ± 1.2; females, 1.6 ± 1.2; $P = 0.032$).

DISCUSSION

In this study, we evaluated ocular symptoms and signs as well as meibomian glands and other tear film–related parameters in 15-year-old students. We found that the condition of the lipid layer of the tear film, as reflected by parameters including the meiboscore (2.8 ± 1.2) and meibum grade (1.8 ± 1.2), seemed to be worse than that described in previous studies,[10,11] with the meiboscore for teenagers having previously been found to range from 0 to 1[11] and meibum secretion abnormality having been detected in only a small proportion of younger individuals.[10] The reason for these differences between our results and those of the previous studies is unclear. The meiboscore in this study did not correlate with lid margin abnormalities. Our findings suggest that the mechanism of meibomian gland changes in adolescents might differ from that in older populations.

We found that changes to the meibomian gland morphology and meibum quality were significantly greater in males than in females at 15 years of age. These differences might be due to sex hormones, such as androgens and estrogens, the production of which changes markedly around this age. Both androgens and estrogens were previously shown to affect the condition of meibomian glands,[20–41] which suggests that the imbalance in these sex hormones might contribute to the sex differences in meibomian glands of high school students. In particular, the production of

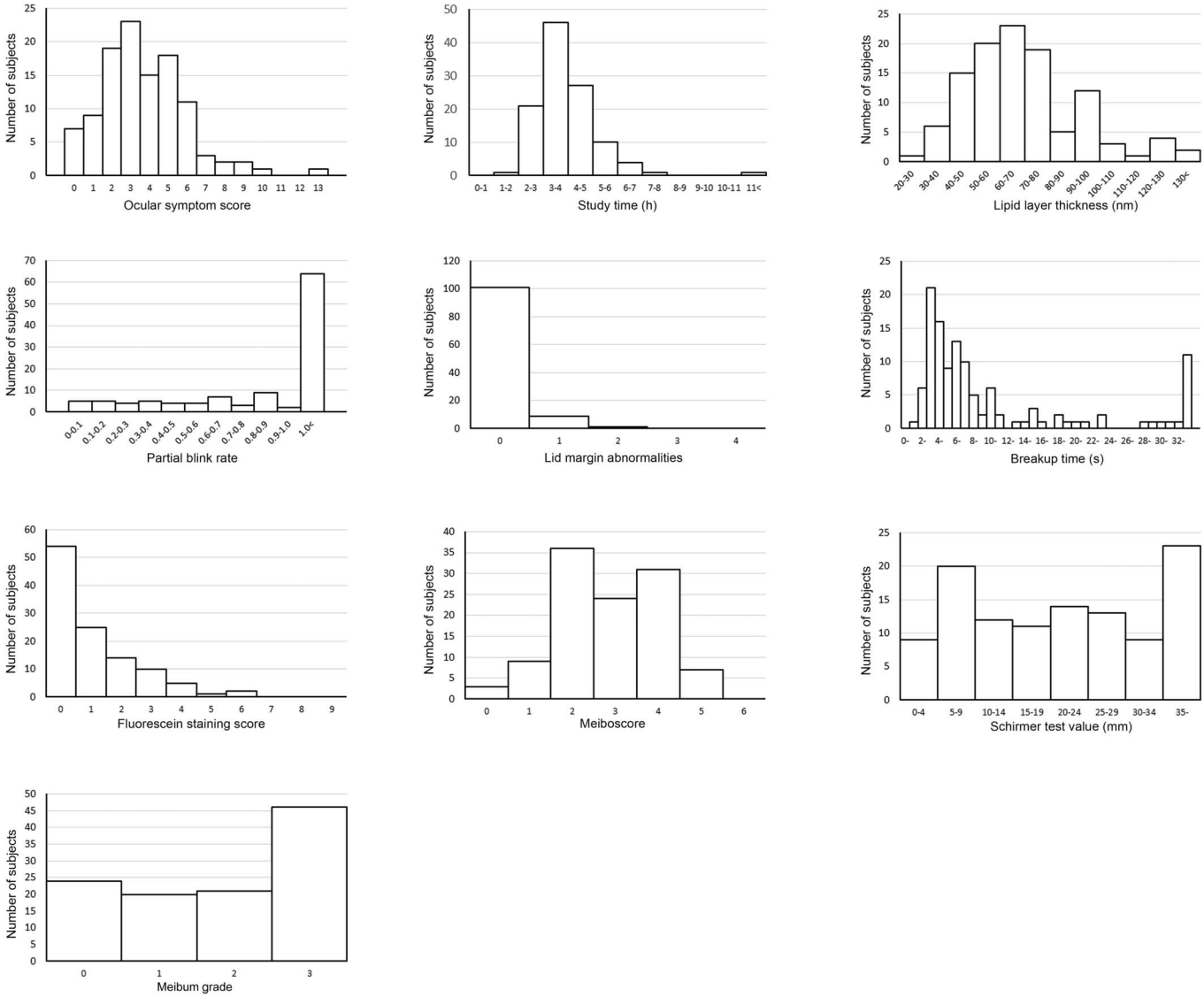

FIGURE 1. Distribution of the measured parameters among the study subjects.

testosterone increases in males between 12 and 18 years of age, and this hormone influences the secretion pattern of sebaceous glands throughout the body.[42] Testosterone levels in postmenopausal women were recently shown to be related to the severity of MGD.[43]

We found that lid margin abnormalities were correlated with the fluorescein staining score but not with the meiboscore or meibum grade. Lid margin abnormalities and the meiboscore are related in aged individuals.[7,11,16] In patients with MGD, lid margin abnormalities include vascularity and plugging and result in a change in the meibomian gland structure,[44,45] with lid margin abnormalities and the meiboscore thus being related. In the young population of this study, an increased meiboscore did not seem to be due to

TABLE 2. Association of Lid Margin Abnormalities With Fluorescein Staining of the Ocular Surface

	Vascularity	Irregularity	Plugging	MCJ
Lid margin abnormality	9/111 (8%)	0/111 (0%)	2/111 (2%)	0/111 (0%)
Presence of staining with lid margin abnormality	8/9 (89%)	0/0 (0%)	2/2 (100%)	0/0 (0%)
Presence of staining without lid margin abnormality	49/102 (48%)	57/111 (57%)	55/109 (50%)	57/111 (57%)
P	0.032*	0	0.5	0

*$P < 0.05$ (Fisher exact test).
MCJ, mucocutaneous junction.

TABLE 3. Spearman Rank Correlation Coefficient (ρ) and *P* Values for the Relationships Between Pairs of Characteristics

	Ocular Symptom Score	Study Time	LLT	Partial Blink Rate	Lid Margin Abnormalities	BUT	Fluorescein Staining Score	Meiboscore	Schirmer Test Value
Study time									
ρ	0.142								
P	0.14								
LLT									
ρ	0.062	0.080							
P	0.52	0.41							
Partial blink rate									
ρ	0.000	0.056	0.129						
P	1	0.56	0.18						
Lid margin abnormalities									
ρ	–0.017	–0.035	0.159	0.048					
P	0.86	0.71	0.095	0.62					
BUT									
ρ	–0.053	–0.029	0.261	–0.006	0.021				
P	0.58	0.76	0.006*	0.95	0.83				
Fluorescein staining score									
ρ	0.042	0.030	0.089	–0.005	0.192	–0.214			
P	0.66	0.76	0.35	0.96	0.043*	0.025*			
Meiboscore									
ρ	0.002	0.001	–0.264	–0.013	0.051	–0.135	0.118		
P	0.99	0.99	0.005*	0.89	0.6	0.16	0.22		
Schirmer test value									
ρ	0.078	0.089	–0.105	–0.240	–0.117	–0.005	–0.140	–0.220	
P	0.42	0.35	0.27	0.011*	0.22	0.96	0.14	0.021*	
Meibum grade									
ρ	0.017	0.180	–0.119	–0.001	–0.018	–0.338	0.054	0.272	–0.021
P	0.86	0.059	0.21	0.99	0.85	<0.001*	0.57	0.004*	0.83

*$P < 0.05$.

alteration of the lid margin, which indicates that the mechanism underlying the change in meibomian gland morphology may be different from that operative in older individuals.

TABLE 4. Characteristics of the Study Population According to Sex (56 Males and 55 Females)

Parameter	Males	Females	*P*
Ocular symptom score (0–14)	3.3 ± 1.9	4.1 ± 2.6	0.27
Study time, h	3.5 ± 1.1	3.4 ± 1.5	0.33
LLT, nm	66.3 ± 20.5	71.3 ± 25.3	0.37
Partial blink rate (0–1)	0.8 ± 0.3	0.8 ± 0.3	0.63
Lid margin abnormalities (0–4)	0.09 ± 0.35	0.11 ± 0.31	0.52
BUT, s	9.4 ± 8.0	7.8 ± 6.2	0.4
Fluorescein staining score (0–9)	0.8 ± 1.1	1.3 ± 1.6	0.11
Meiboscore (0–6)	3.3 ± 1.1	2.4 ± 1.2	<0.001*
Schirmer test value, mm	20.5 ± 11.5	19.9 ± 11.5	0.76
Meibum grade (0–3)	2.0 ± 1.2	1.6 ± 1.2	0.032*

*$P < 0.05$.
Data are mean ± SD, and *P* values were determined with the Mann–Whitney *U* test.

Adolescent males and females often have acne. In the United States and European countries, acne is treated by systemic therapy, such as that with isotretinoin (Accutane). Accutane therapy affects sebaceous glands throughout the body and might therefore be expected to influence meibomian glands.[46,47] Accutane is not approved in Japan, however, and so none of the subjects in this study received such treatment. We also excluded subjects who were being treated with antibiotics. The possible effect of Accutane therapy on evaluation of meibomian glands should be considered in countries in which such treatment is administered.

There are several limitations to our study. First, given the study design, we did not include individuals of a different age group as control subjects. Second, we enrolled preparatory school students, whose lifestyle, including time devoted to study, eating habits, and sleep time, possibly differs from that of attendees of other types of schools. Third, we did not perform a systemic examination of the subjects such as by performing blood tests or by measuring the heart rate or blood pressure.

In conclusion, we have measured meibomian gland and other tear film–related parameters in apparently healthy individuals at 15 years of age. Our results have revealed

changes in meibomian glands of this cohort and sex differences that might be due to differences in systemic sex hormone levels. Our data provide a basis for further investigations into meibomian gland physiology.

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