The Relationship between *Demodex* and Ocular Discomfort

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Purpose. To determine the correlative relationship between the prevalence of *Demodex* in eyelashes and the severity of ocular discomfort, by investigating the demographic epidemiology associated with *Demodex*.

Methods. One hundred seventy patients underwent epilation of four eyelashes of each eye, and the number of *Demodex* was counted. The patients answered questionnaires about ocular surface discomfort and underwent ophthalmic examinations, including slit lamp, tear film breakup time (BUT), Schirmer test, and microbial culturing. The correlative relationship between the number of *Demodex* and these variable data was analyzed.

Results. *Demodex* was found in 120 (70%) of the 170 tested patients. Of 1360 eyelashes, 740 (54%) had *Demodex*. There was no significant difference in the prevalence of *Demodex* between males and females ($P = 0.35$). The number of *Demodex* showed significant positive correlations with increased age, ocular discomfort, and 1/BUT ($P < 0.001$), but not with the Schirmer scores. The number of *Demodex* was significantly higher in patients with conjunctival papillary hypertrophy than in those without ($P = 0.003$). The presence or distribution of bacteria on eyelashes was similar between eyelids with and without *Demodex*. However, methicillin-resistant *Staphylococcus aureus* (MRSA) was detected more often on eyelids with *Demodex*, but this difference was not statistically significant.

Conclusions. There is a strong correlation between the number of *Demodex* and the severity of ocular discomfort, suggesting that *Demodex* plays a pathogenic role in the ocular discomfort linked with aging. (*Invest Ophthalmol Vis Sci.* 2010;51: 2906–2911) DOI:10.1167/iovs.09-4850

The ectopic parasite *Demodex* is the most common parasite in humans. It inhabits the eyelids, cilia, meibomian glands, face, and external otic tract. These obligate mites are transparent, elongated in shape, and divided into head-neck and body-tail parts, with eight short legs attached to the anterior body segment.[1,2] There are many species of *Demodex*, but only *D. folliculorum* and *D. brevis* are found on the human body.[3] *D. folliculorum*, 0.35 to 0.4 mm in length, lives in the lash follicles, and *D. brevis*, 0.15 to 0.2 mm in length, lives deep in the meibomian glands and the sebaceous glands of the lash. They eat skin cells, hormones, and oils that accumulate within the hair follicle.[4–6]

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According to a literature review in dermatology, *Demodex* colonizes normal human skin everywhere; on average, the *Demodex* population is approximately ≤5 per square centimeter of skin in the adult population. They are not usually the cause of any dermatologic problems, but when the parasites penetrate the dermis, they can cause dermatologic diseases, such as acne, rosacea, and folliculitis, when the population increases.[7–12]

In ophthalmology, *Demodex* is thought to be an etiologic factor in chronic blepharitis, conjunctival inflammation, and meibomian gland dysfunction.[4,5,13,14] Furthermore, *Demodex* has also been reported to cause unusual ocular manifestations such as superficial corneal neovascularization, marginal corneal infiltration, phlyctenule-like lesions, superficial corneal opacity, and nodular corneal scars, especially in patients with ocular rosacea.[15] These studies were based on the clinical improvement noted after substantial reduction of *Demodex* counts in lids scrubbed with tea tree oil.[1,16,17]

However, *Demodex* can be found in asymptomatic individuals, and its pathogenic role has long been debated. Many studies have reported that *Demodex* plays a pathogenic role in causing blepharitis, pityriasis folliculorum, papulopustular rosacea, and folliculitis.[17–20] However, many other studies have advocated that *Demodex* is a nonpathogenic parasite and demodicosis is found in persons who are immunosuppressed.[21–24] The basis of this argument is that the pathogenicity of *Demodex* has not been demonstrated. Because it is a host-specific obligate parasite that cannot be grown in vitro, *Demodex* is very difficult to study and inducing an experimental infestation is difficult.[7]

In an attempt to understand how *Demodex* plays a role in causing ocular discomfort in general Asian patient populations, we investigated the prevalence of *Demodex* and relevant demographic information in Yongsan-Gu, Seoul, Republic of Korea, an area with a moderate socioeconomic level, and evaluated the correlation with other clinical parameters including ocular discomfort and microbial isolation from the eyelid.

Methods

Patients

One hundred seventy of the patients who visited in our clinic (Yongsan Hospital of Chung-Ang University) for ophthalmic examinations between March 1 and September 30, 2007, were included in the study. The patients had made appointments for a regular check-up, prescriptions for contact lenses, examination of diabetic or hypertensive retinopathy, or examination of glaucoma. Patients were excluded from participating if they met any of the following criteria: history of an ocular burn, clinical evidence of goblet cell deficiency of the ocular surface, or obstruction of the canaliculus or nasolacrimal duct. Informed consent was obtained from all participants for the examination after the possible consequences of the study were explained. The study was approved by the Chung-Ang University Yongsan Hospital Institutional Review Board, and all the methods described adhered to the principles of the Declaration of Helsinki.

TABLE 1. Incidence of *Demodex* According to Age Group

Age (y)	Patients (*n*)	Sum of *Demodex* Count	Average Number of *Demodex*/Patient
<30	37	21	0.56 ± 1.14
30–49	44	169	3.84 ± 3.82
50–69	51	247	4.84 ± 4.17
>70	38	303	7.97 ± 5.60

Examinations

All patients underwent complete ophthalmic examination under a slit lamp biomicroscope. Examinations for the detection of ocular manifestations were performed on the eyelids (erythema, telangiectasia, and meibomian gland secretion), conjunctiva (injection and papillary hypertrophy), and cornea (erosion, opacity, and other corneal abnormalities). The tear film break-up time (BUT) and the Schirmer test were also performed. We measured the amount of tears with the Schirmer test, using topical anesthesia to avoid reflex tearing by ocular irritation due to corneal erosion or physical contact with the Schirmer strip. The Schirmer test was performed 5 minutes after the application of the anesthetic. The patients also underwent epilation of eight eyelashes (two eyelashes in both the upper and lower eyelids) and the number of *Demodex* was counted with an optical microscope. We tried to epilate lashes with cylindrical dandruff around the root of the lash as deeply as possible.[3] Last, the patients answered a questionnaire about ocular discomfort.

Questionnaire

The questionnaire was administered by an ophthalmologist who had no information about the presence of ocular manifestation. The questionnaire was divided into a patient background section and an ocular symptoms section. The background section included age, sex, existence of ocular discomfort, duration of ocular discomfort, ophthalmic surgical history, contact lens history, systemic disease (hypertension and diabetes), and systemic medications.

The questions pertaining to ocular symptoms contained three categories and 15 items. We modified the ocular surface disease index (OSDI)[25] by addition of three questions about chronic blepharitis (questions numbered 6 to 8 in the following items).

The patients' answers were based on their experience during the past week, and severity was graded on a scale of 0 to 4: 0, none of the time; 1, some of the time; 2, one-half of the time; 3, most of the time; and 4, all the time. The answers to the items were converted into a numerical score using the equation, $A \times 25/B$, where A is the sum of scores for all questions answered, and B, is the total number of questions answered. The questionnaires were assessed on a scale of 0 to 100, with higher scores representing greater ocular disability.

The first category of the questionnaire pertained to ocular symptoms: (1) Are the eyes sensitive to light? (2) Do the eyes feel gritty? (3) Are the eyes painful or sore? (4) Is there blurred vision? (5) Is the vision poor? (6) Do the eyes feel itchy? (7) Are the eyelids injected in the morning? and (8) Is there a discharge that makes opening the eyes in the morning difficult? The second category was about problems with the eyes limiting performance in any of the following activities: (9) Reading? (10) Driving at night? (11) Working with a computer or bank machine? and (12) Watching TV? The third category was about discomfort in the eyes in the following situations: (13) Windy conditions? (14) Areas with low humidity (beside a heater)? and (15) Areas that are air conditioned?

Examination for Bacteria on Eyelid

To analyze the characteristics of the bacterial distribution on the eyelids relative to the presence of *Demodex*, we performed bacterial cultures in 144 eyes randomly sampled (84 eyelids with *Demodex* and 60 eyes without *Demodex* as a control group). After squeezing the meibomian gland, we scraped the discharge of the meibomian gland with a sterile cotton tip, not touching the eyelid skin. The collection was inoculated into a blood agar plate, chocolate agar plate, Sabouraud dextrose agar plate, and thioglycollate broth. To test for the virulence of *Staphylococcus aureus*, we performed an antibiotic sensitivity test (Vitek II system; BioMerieux, Durham, NC).

Statistical Analysis

All data are expressed as the mean ± SD. The data between groups were assessed with Student's *t*-test, and the data between variables were assessed with the Pearson correlation test. To decrease age bias, we used multiple regression analysis and analyzed the different age groups separately. All results were considered statistically significant when $P < 0.05$ (SPSS, ver. 16.0; SPSS, Inc., Chicago, IL).

RESULTS

Prevalence of *Demodex*

One hundred seventy patients (64 males and 106 females, with a mean age of 50.8 ± 19.3 years; range, 15–87) were enrolled in the study. *Demodex* was found in 120 (70%) of 170 patients and 740 (54%) of 1360 eyelashes. The mean total *Demodex* count per patient was 4.4 ± 4.7. *Demodex* was found in 48 (75%) of 64 males and 72 (68%) of 106 females. The mean total *Demodex* count per male was 4.3 ± 4.4 and that per female was 4.4 ± 4.9. There was no statistically significant difference in the prevalence of *Demodex* between the males and females ($P = 0.35$). There was no statistically significant relationship between the prevalence of *Demodex* and systemic diseases ($P > 0.5$).

The prevalence of *Demodex* according to age is shown in Table 1. The average number of *Demodex* per patient of each group increased with age. The total number of *Demodex* per patient had a significant positive correlation with increased age in all patients (Fig. 1; $P < 0.001$, correlation coefficient = 0.544). The mean age of patients with *Demodex* was 56.8 ± 16.5 years, and the mean age of patients without *Demodex* was 36.5 ± 18.1 years. The difference between the two groups was statistically significant ($P < 0.001$).

Relationship with Ocular Manifestations

The number of *Demodex* significantly increased when the ocular surface disease index score was high (Fig. 2; $P < 0.001$,

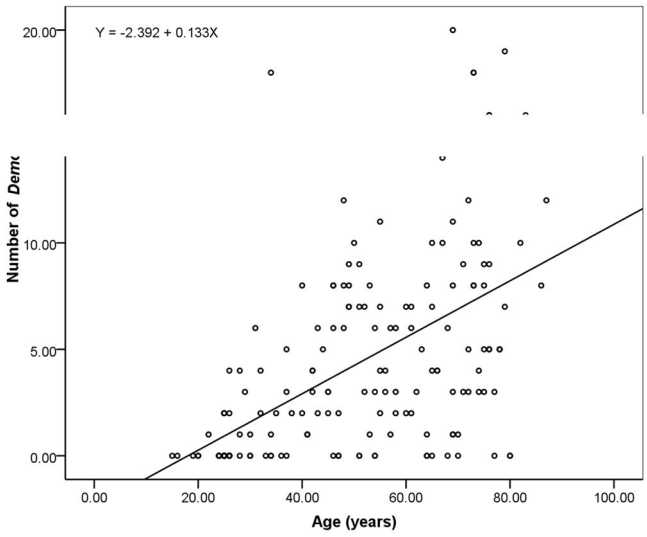

FIGURE 1. The number of *Demodex* showed a significant positive correlation with increasing age.

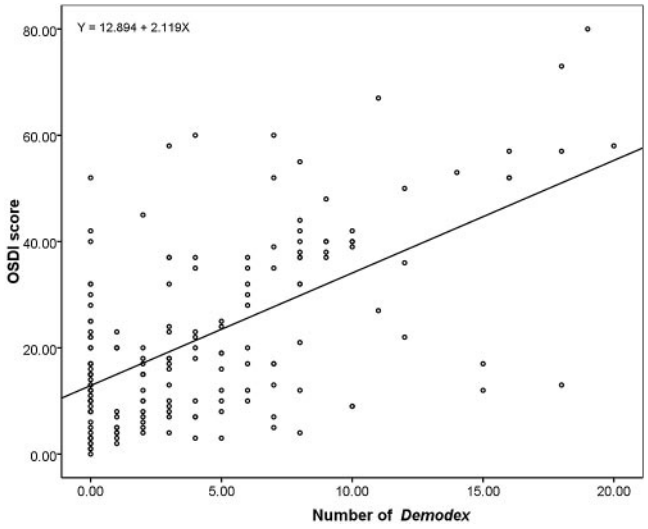

FIGURE 2. The relationship of *Demodex* count and ocular discomfort showed that the OSDI increased significantly when the number of *Demodex* was high.

correlation coefficient = 0.597). In multiple regression analysis, the *Demodex* count still correlated significantly with ocular discomfort ($P < 0.001$). The regression equation using factors of ocular surface discomfort is: OSDI = 27.081 − 0.096 (age) + 1.956 (number of *Demodex*) − 1.746 (average time of BUT) − 0.072 (Schirmer score) (Table 2). The mean score of ocular discomfort with *Demodex* was 25.0 ± 17.8 and without *Demodex* was 15.1 ± 11.3. The difference between the groups was statistically significant ($P < 0.001$).

The patients were divided into four groups according to age (<30, 30–40, 50–60, and >70 years) to decrease the bias accompanying the aging process. With the exception of those patients <30 years of age, the number of *Demodex* significantly increased in those with more severe ocular symptoms (Table 3).

The relationship of *Demodex* and BUT showed that the number of *Demodex* was significantly higher when the BUT was shorter (Fig. 3; $P < 0.001$, correlation coefficient = −0.522). The average of the BUT of the four groups according to age (<30, 30–40, 50–60, and >70 years) was 4.62 ± 2.10, 4.59 ± 1.85, 4.44 ± 1.95, and 4.36 ± 1.97 seconds, respectively. The number of *Demodex* significantly increased when the BUT decreased in three of the subgroups (30–40, 50–60, and >70 years; $P < 0.001$), but not in the <30-years group ($P = 0.102$). The mean time of BUT with *Demodex* was 4.1 ± 1.7 seconds and without *Demodex* was 5.6 ± 2.2 seconds, which was also significantly different ($P = 0.003$).

TABLE 3. The Relationship between *Demodex* and the OSDI According to Age Group

Age (y)	*P**	Correlation Coefficient	Average OSDI
<30	0.434	−0.133	19.43 ± 11.18
30–49	<0.001	0.515	19.61 ± 16.38
50–69	<0.001	0.715	23.17 ± 16.82
>70	<0.001	0.724	26.21 ± 20.70

* Pearson correlation test.

There was no statistically significant correlation between the amount of tears (Schirmer test) and the *Demodex* count (Fig. 4, $P = 0.898$).

There was a statistically significant correlation between the presence of *Demodex* and conjunctival papillary hypertrophy. The mean total *Demodex* count in patients with papillary hypertrophy was significantly higher than in those without papillary hypertrophy ($P = 0.006$). The total number of patients with conjunctival papillary hypertrophy was 68, and the average count of *Demodex* in them was 6.0 ± 5.2. The total number of patients without conjunctival papillary hypertrophy was 102 and the average count of *Demodex* in them was 3.3 ± 4.0. Conjunctival papillary hypertrophy was observed in 14 (28%) of 50 patients without *Demodex* and 54 (45%) of 120 patients with *Demodex*. There was a statistically significant difference in the papillary reaction between both groups ($P = 0.003$).

Characteristics of Bacterial Distribution

There was no statistically significant difference in the presence or type of bacteria in the two groups: patients with and those without *Demodex.* The ratio of isolating coagulase negative *Staphylococcus*, *Corynebacterium diphtheriae*, and *S. aureus* was not different between the group with *Demodex* and the group without ($P = 0.440$). Although the ratio of methicillin-resistant *S. aureus* (MRSA) was 100% (8/8) in eyes with *Demodex* and 75% (3/4) in eyes without it (Table 4), the difference was not statistically significant ($P = 0.333$, Fisher's exact test).

DISCUSSION

Early reports suggested that the incidence of *Demodex* is higher in patients with blepharitis than in those with no ophthalmic diseases.[7,26–28] Also, studies have reported that *Demodex* infestation may be related to corneal and conjunctival pathologic features, and the severity of disease decreases after *Demodex* is treated.[15,17] These reports implied that *Demodex*

TABLE 2. Result of Multiple Regression Analysis

	Unstandardized Coefficients		Standardized Coefficients		
	β	SE	β	T	P
Constant	27.081	4.067		6.659	0.000
Age, y	−0.096	0.065	−0.110	−1.462	0.146
Demodex, n	1.956	0.311	0.551	6.290	0.000
BUT, s	−1.746	0.634	−0.203	−2.756	0.007
Schirmer, mm	−0.072	0.152	−0.029	−0.475	0.635

Relationship between ocular surface discomfort and four risk factors (age, number of *Demodex*, BUT, and Schirmer test). Dependent variable: OSDI.

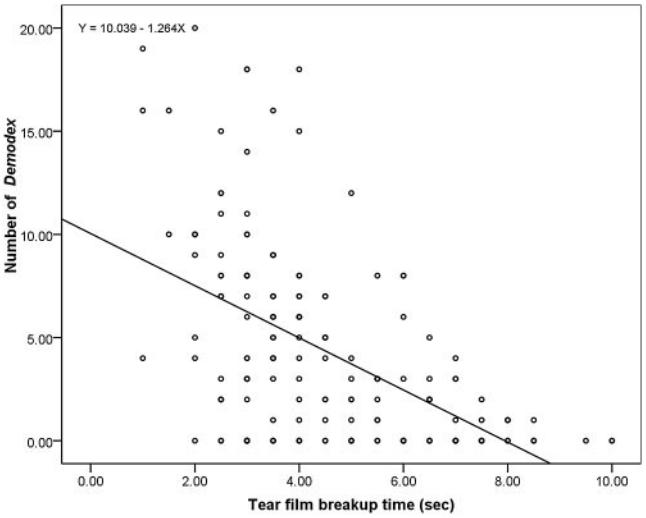

FIGURE 3. The relation of *Demodex* and tear film BUT showed that the number of *Demodex* was significantly higher when the tear film BUT was shorter.

is a pathogen, but the exact mechanism of disease or prevalence have not been revealed.

The number of *Demodex* increased in our subjects in proportion to age. There may be several explanations for this finding. Since it is a mite living in symbiosis, there may be a relationship between the number of *Demodex* and the age of the patient. On the other hand, poor sanitary conditions with increasing age may be associated with the increase in *Demodex*. Based on published papers and the authors' experience, the latter seems to be more probable than the former. To further support this reasoning, we found in our study that old patients with good eyelid hygiene had fewer *Demodex* relative to their age, while young patients with poor eyelid hygiene had a greater count relative to their age. Thus, we concluded that the poorer the eyelid hygiene, the greater the number of *Demodex*.

Lacey et al.[29] reported that the eye surface is protected by a bony protrusion, which is why the eyelid is not cleaned by cleansing the face. Westerners have sunken eyes, while Asians have protruding eyes. Unlike the eyes of Westerners, the eyes of Asians may not be protected by bony protrusions such as the brows, nose, and cheeks. Also, the eyelashes of Westerners are longer and thicker than those of Asians. Because of these differences, the eyelashes of Asians can be easily cleaned by washing the face without cleansing the lashes separately. These features may influence the relationship between hygiene of the eyelids and the number of *Demodex* in the eyelashes of Asians.

Some researchers have insisted that there is no relationship between age and the number of *Demodex*.[24] However, they based their research mostly on the relationship between blepharitis and *Demodex*. Indeed, there may have been a negligible relationship between age and eyelid hygiene among patients with *Demodex*; therefore, the number of *Demodex* may have appeared to be unrelated to age. If they had conducted research on general patients with or without blepharitis, as in the present study, they might have been able to determine the differences in eyelid hygiene according to age and to conclude that the prevalence of *Demodex* increases with age.

There was no relationship between *Demodex* and the sex of the subject. Türk et al.[26] reported a higher detection rate of *Demodex* in male patients, whereas Forton et al.[7] reported a higher detection rate in female patients. To the contrary, Kemal et al.[24] reported that there is no sex-related difference in the detection rate of *Demodex*, as was also found in the present study. It can thus be inferred that *Demodex* has no relationship to sex hormones.

There were no relationships in our study between *Demodex* and systemic diseases such as diabetes and hypertension. Forton et al.[24] also reported that 96% of patients in whom *Demodex* was detected were healthy. Yet, reports have described higher detection ratios in patients with diabetes or in those with low immunity.[21–23] These findings may be secondary to poor sanitary conditions, rather than to systemic diseases.

In our study, an increase in *Demodex* caused an increase in subjective symptoms of the ocular surface. Considering that one of the typical characteristics of aging is decreased tear secretion, which may lead to increased ocular discomfort, this finding might be related to aging. However, in a multiple regression analysis, *Demodex* was found to be significantly related to ocular surface discomfort and the aging factor did not correlate significantly with ocular surface discomfort (Table 2). In an additional analysis of the four age groups, all groups, except the group <30 years of age, showed a significant relationship between these two factors (Table 3). Therefore, it can be concluded that even when age-related changes are taken into consideration, an increase in *Demodex* causes changes and a subsequent increase in ocular surface discomfort. In the group of <30 years of age, which showed different results in the analysis, we conclude that the reverse relationship was not established. That is, ocular surface discomfort is not necessarily evidence of an increase in *Demodex*. The increase in ocular surface discomfort in the <30-years age group may have resulted from double eyelid surgery, refractive surgery, and the use of contact lenses.

There are positive correlations between *Demodex* and conjunctival papillary reactions. Those groups with conjunctival papillary hypertrophy more often have allergies that may be caused by *Demodex*. Currently, allergies to mites are identified by skin testing. However, these tests are actually intended, not for *D. folliculorum* or *D. brevis*, but for house dust mites. We found that the result of skin tests for house dust mites had no relationship to *Demodex*. It is not known whether *Demodex* or their excretions and secretions cause allergic reactions. This possibility can be explored in the future when there is a test method available to identify allergies to *D. folliculorum* or *D. brevis*.

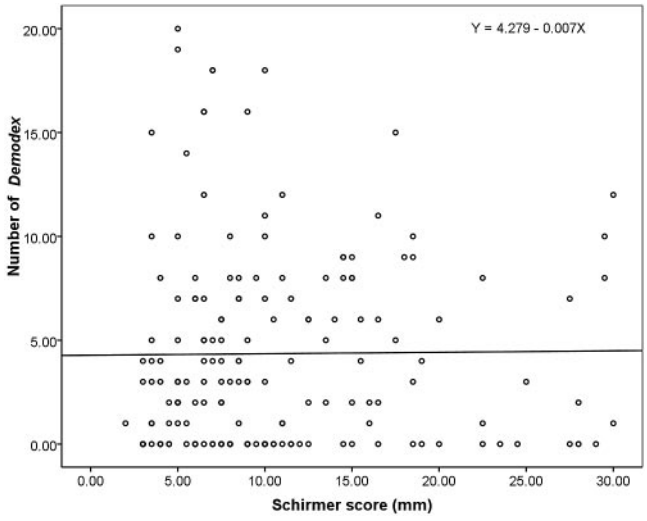

FIGURE 4. The relation between the Schirmer test and the *Demodex* count. There was no statistically significant correlation.

Table 4. Comparisons of Pathogens According to Existence of *Demodex* in Eyelashes

	Eyes with *Demodex* (n = 84)	Eyes without *Demodex* (n = 60)
Growth	72 (85.7)	50 (83.3)
Two species co-infection	31 (36.9)	17 (28.3)
Three species co-infection	1 (1.2)	1 (1.7)
Gram positive cocci		
Coagulase negative staphylococcus	59 (56.2)	42 (80.86)
Staphylococcus aureus	8 (7.6)	4 (5.7)
MRSA/*S. aureus*	8/8 (100)	3/4 (75)
Gram positive rod		
Corynebacterium diphtheria	33 (31.4)	20 (28.9)
Paenibacillus polymyxa	1 (0.95)	0
Gram negative cocci		
Neisseria species	0	1 (1.45)
Gram negative rod		
Enterobacter aerogenes	0	1 (1.45)
Serratia marcescens	0	1 (1.45)
Achromobacter xylosoxidans	2 (1.9)	0
Citrobacter	1 (0.95)	0
Fungus		
Candida parapsilosis	1 (0.95)	0

Data are n (%) with bacterial growth.

An increasing number of *Demodex* reduced the BUT, but did not affect the results of the Schirmer test. These results are in agreement with those in previous studies showing that *Demodex* may cause damage to the meibomian glands, leading to an abnormal lipid tear film, and the lipid tear film is stabilized by treating ocular demodicosis.[15–17] We can infer the effect of *Demodex* on tears from this evidence. The BUT is mainly dependent on the lipid components of the meibomian gland, whereas the Schirmer test is dependent on the tear output of the lacrimal gland. In another of our studies, we also found that inflammatory markers of tears were increased in eyes with *Demodex* (data not shown). Therefore, we can conclude that *Demodex* affects the meibomian glands to cause instability of the tear film, but does not affect the lacrimal glands.

No correlation was found between *Demodex* and the distribution of bacteria. In comparing eyelids in which *Demodex* was detected and those in which *Demodex* was not found, we did not find any difference in the bacterial detection ratio, superinfection, and distribution of any detected bacteria. These results are contrary to those in a study in which the investigators reported that secretion of *Demodex* functioned as a vector for bacteria.[30] The results of this study indicate that ocular surface diseases related to *Demodex*, which are hard to treat, appear to be caused not by changes in adherent bacteria, but by *Demodex* itself. However, the relationship between *Demodex* and MRSA requires further study. Although there was little statistical significance, the *S. aureus* strains isolated from the eyelids with *Demodex* infestation were all (100%) MRSA-positive, whereas 75% of the *S. aureus* strains isolated from eyelids without *Demodex* infestation were MRSA-positive. A study involving a larger patient population is needed for further clarification of this difference.

This study focused on the relationship between *Demodex* and ocular discomfort in the general Asian population. The results showed that the increase in *Demodex* was relevant to age. The effects of *Demodex* on the ocular surface include inflammation of the meibomian gland and conjunctival allergic reactions, whereas the sex of the host, tear secretion, and the prevalence or type of bacteria had no relationship to *Demodex*. The severity of ocular surface discomfort showed a strong positive correlation with the number of *Demodex*, irrespective of age. These results support that *Demodex* plays an independent pathogenic role in ocular surface conditions. Also, the findings suggest that the treatment of *Demodex* on eyelids can help to improve ocular discomfort. Therefore, we believe that lid hygiene, examination for *Demodex* on the eyelashes, and the treatment of *Demodex* are important and necessary in patients with ocular discomfort, especially elderly patients.

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