

Tear Interferometric Patterns Reflect Clinical Tear Dynamics in Dry Eye Patients

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PURPOSE. We investigated whether the tear interferometric pattern was able to identify differences in tear film kinetics among clinical subtypes of dry eye.

METHODS. A total of 138 eyes of 76 subjects (38 men and 38 women; mean age \pm SD, 61.6 \pm 16.2 years) with or without dry eye who visited Itoh Clinic from May to August 2015 were enrolled in a cross-sectional study. Clinical diagnosis of dry eye subtype was based on tear film parameters. The pattern of tear film kinetics determined by interferometry was classified as 0 (monotonous gray or multicolor interferometric fringe with a noninvasive breakup time [NIBUT] of ≥ 5 seconds), 1 (multicolor interferometric fringe with a NIBUT of < 5 seconds), or 2 (grayish amorphous interferometric fringe with a NIBUT of < 5 seconds), and reliability of classification was evaluated. Lipid layer thickness (LLT) for the tear film was also determined by interferometry.

RESULTS. Interrater κ values for evaluation of interferometric patterns ranged from 0.57 to 0.94 for both physicians and nonphysicians with reference to a dry eye expert, the latter of whom showed an intrarater reliability of 0.90. The distribution of eyes among interferometric patterns 0, 1, and 2 coincided well with the clinical subgroups of normal tear condition, non-Sjögren syndrome aqueous-deficient dry eye, and meibomian gland dysfunction, respectively. A multicolor interferometric fringe was essentially observed only at an LLT of > 70 nm.

CONCLUSIONS. Tear interferometry was able to reliably distinguish clinical subtypes of dry eye by reflecting the balance between the lipid and aqueous layers of the tear film.

Keywords: tear interferometry, dry eye, meibomian gland

The tear film consists of aqueous, mucin, and lipid layers with different levels of complexity.¹ The quality and quantity of each layer, as well as their interactive relations, are important for homeostasis of the tear film. Impairment of the quality or changes in the quantity of tear film components can result in dry eye disease, which is categorized as either aqueous-deficient dry eye (ADDE) or evaporative dry eye (EDE).^{2,3} Dysfunction or hypofunction of lacrimal glands and the consequent reduced volume of the aqueous layer of the tear film thus underlie ADDE. Similarly, dysfunction or hypofunction of meibomian glands (which produce the lipid components of the tear film) or reduced production of mucin proteins by conjunctival goblet cells increases the evaporation of tear fluid and results in EDE.

Interferometry has been applied as a noninvasive method for visualization of the lucent lipid layer at the surface of the tear film.⁴ Tear interference images associated with surface phenomena of the tear film have thus been obtained⁵⁻⁸ based on a principle first described by Newton.⁹ This approach has been adopted to study tear dynamics in individuals with Sjögren syndrome¹⁰ or dry eye,¹¹ including EDE,¹² as well as in contact

lens wearers.^{6,13} It has also been used to measure the thickness of the lipid layer of the tear film in normal subjects,¹⁴ in individuals with ADDE,¹⁵ in office workers with dry eye,¹⁶ in patients with Sjögren syndrome or non-Sjögren dry eye,¹⁷ and in those with meibomian gland dysfunction (MGD).¹⁸ Tear interferometry thus provides information on both the quality and quantity of the lipid layer, yielding insight into the function of the entire tear film.

The balance of tear film components is important for tear film stability, and a compensatory system is thought to operate in response to changes in these components.^{5,11,17,19-21} We have now evaluated the overall aqueous and lipid kinetics of the tear film, as well as measured the thickness of the lipid layer by tear interferometry, revealing the balance between the lipid and aqueous layers of the tear film,^{11,15,22,23} in individuals with dry eye disease. Our results indicate that the interferometric color and fringe patterns associated with tear film stability reflect the balance between the aqueous and lipid layers of the tear film, and they suggest that these patterns are able to identify subtypes of dry eye.



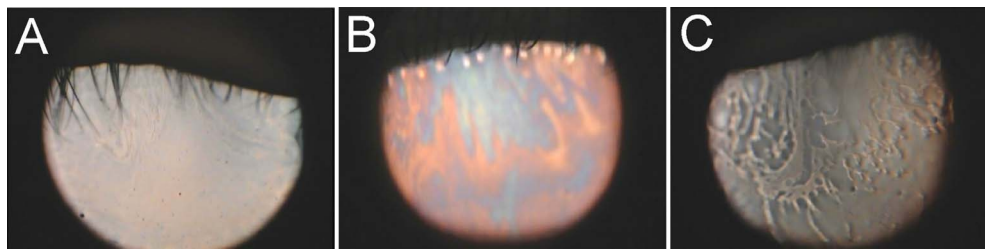


FIGURE 1. Representative interferometric fringe images. (A) Pearl-like appearance (monotonous gray interferometric fringe). (B) Jupiter-like appearance (multicolored interferometric fringe). (C) Crystal-like appearance (grayish amorphous interferometric fringe).

METHODS

Study Design

This cross-sectional study was approved prospectively by the Institutional Review Board of Itoh Clinic and adhered to the tenets of the Declaration of Helsinki. Written informed consent was obtained from all subjects before examination. Patients who visited Itoh Clinic from May to August 2015 were candidates for enrollment in the study. Exclusion criteria included acute infections of the eyelids, ocular allergies, obvious eyelid or ocular surface disorders, contact lens wear, continuous use of eyedrops with the exception of those for dry eye treatment, a history of eye surgery, and systemic or ocular diseases (other than those to be studied) that might interfere with tear film production or function. Individuals who wore eye makeup were also excluded. Individuals with ocular symptoms were enrolled as dry eye subjects and classified as having non-Sjögren ADDE (non-SS-ADDE), MGD, or both non-SS-ADDE and MGD as appropriate (total of 84 eyes of 47 patients; 19 males and 28 females; mean age \pm SD, 61.4 ± 13.6 years). Meibomian gland dysfunction was evaluated as a surrogate for EDE, given that it is a major cause of EDE.²⁴ Non-SS-ADDE was diagnosed on the basis of criteria proposed by the Dry Eye Research Group in Japan²⁵: (1) the presence of dry eye symptoms, (2) abnormal tear production as determined by Schirmer's test (≤ 5 mm for 5 minutes) or abnormal tear film stability as determined by breakup time (BUT, ≤ 5 seconds), and/or (3) the presence of conjunctival and corneal epithelial damage as evidenced by a fluorescein staining score of ≥ 3 as determined by the van Bijsterveld method.²⁶ Meibomian gland dysfunction was also diagnosed on the basis of previously described criteria²⁷: (1) at least one symptom from among ocular fatigue, discharge, foreign body sensation, dryness, uncomfortable sensation, sticky sensation, pain, epiphora, itching, redness, heavy sensation, glare, excessive blinking, burning sensation, and ocular discomfort on arising; (2) at least one lid margin abnormality; and (3) poor meibum expression. Individuals without any ocular conditions other than refractive error or cataract were recruited as control subjects (total of 54 eyes of 29 subjects; 19 males and 10 females; mean age \pm SD, 67.2 ± 13.4 years). Those individuals who used eyedrops were instructed not to apply them for at least 4 hours before examinations.

Examinations

Examinations were performed sequentially as follows: (1) tear interference and noninvasive BUT (NIBUT) were determined by tear interferometry (DR-1 α ; Kowa, Nagoya, Japan).^{11,15} The subject was asked to blink naturally twice and then to keep both eyes open as long as possible. Examination of the second eye was performed at least 1 minute after that of the first. (2) Lipid layer thickness (LLT) of the tear film was also measured by tear interferometry (LipiView; TearScience, Morrisville, NC, USA).²⁸ The subject was asked to blink naturally during the examination.

(3) Abnormalities of the upper and lower lid margins were evaluated by slitlamp microscopy and scored from 0 to 4 according to the number present.²⁹ (4) Fluorescein staining of the ocular surface was divided into three zones (nasal conjunctival, corneal, and temporal conjunctival areas) for determination of the superficial punctate keratopathy (SPK) score. The staining score ranged from 0 to 3 for each zone, yielding a total score of 0 to 9 for the ocular surface.²⁶ (5) Tear film BUT was measured after instillation of 1 μ L of a preservative-free solution of 1% fluorescein into the conjunctival sac with the use of a micropipette. The subjects were asked to blink several times, and BUT was determined three times with a stopwatch. The mean of the three values was calculated. (6) The upper and lower eyelids were evaluated with the use of a noninvasive meibography system (SL-D701 DC-4 BG-5; Topcon, Tokyo, Japan).³⁰ Partial or complete loss of meibomian glands was scored as the meiboscore for each eyelid as previously described.³⁰ The meiboscores for the upper and lower eyelids of each eye were summed to obtain an overall score of 0–6. (7) A Schirmer strip (Whatman no. 41; Showa, Tokyo, Japan) was inserted over the lower lid margin (midway between the middle and outer thirds) for 5 minutes without topical anesthesia. Subjects were asked to close their eyes during the measurement. (8) Digital pressure was applied to the upper tarsus, and the degree of ease with which meibomian secretion (meibum) was induced was evaluated semiquantitatively.¹⁹ All examinations were completed on the same day in the morning, usually within a total time of 15 minutes. Both room temperature ($24.5 \pm 3.7^\circ\text{C}$) and humidity ($40.5 \pm 8.5\%$) were relatively constant.

Repeatability of LLT and NIBUT Measurements

The repeatability of LLT measurement with LipiView and NIBUT measurement with DR-1 α was assessed by having one examiner (R.A.) repeat the measurements three times in five eyes of five normal subjects selected randomly. The intra-examiner repeatability for each eye was assessed as the coefficient of variation (SD for the three measurements/mean of the three measurements), and the mean \pm SD for the coefficient of variation for the five eyes was then calculated.

Interferometric Pattern Classification

One dry eye expert (R.A.) developed a classification system for tear lipid interferometric images based on viewing videos obtained from the 138 eyes of the study subjects. The pattern of tear film kinetics and NIBUT were determined with the DR-1 α tear interferometer. The interferometric images were classified into three types: pearl-like appearance (monotonous gray interferometric fringe), Jupiter-like appearance (multicolored interferometric fringe), or crystal-like appearance (grayish amorphous interferometric fringe) (Fig. 1). The time from the last blink to the appearance of a break or discontinuity in the surface of the tear film visualized by interferometry without fluorescein staining was measured as the NIBUT by reviewing

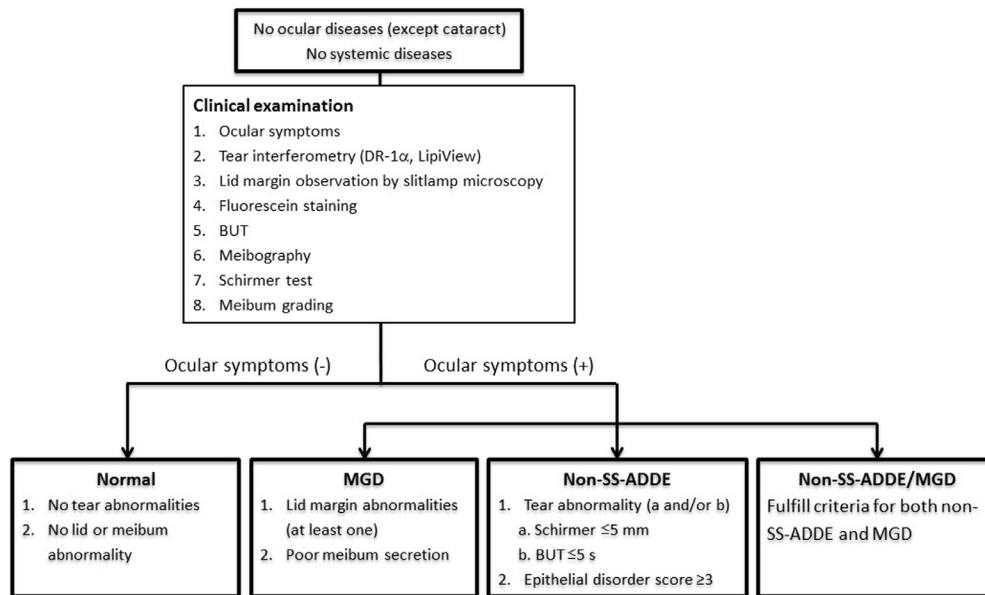


FIGURE 2. Flowchart showing participant progress through the study.

interferometric movies. Receiver operating characteristic (ROC) curves for NIBUT in normal subjects and in patients with non-SS-ADDE or MGD yielded a cutoff value of 4 seconds (area under the curve = 0.9116, 95% confidence interval [CI] = 0.8377–0.9537), and we therefore defined a NIBUT of <5 seconds to be shortened. Interferometric images were combined with NIBUT to yield the following interferometric patterns: class 0 (pearl- or Jupiter-like appearance, NIBUT of ≥ 5 seconds), class 1 (Jupiter-like appearance, NIBUT of <5 seconds), and class 2 (crystal-like appearance, NIBUT of <5 seconds; Supplementary Videos S1–S3).

Validation Test

A validation test was performed to evaluate the robustness of the proposed interferometric pattern classification. Videos for evaluation included tear lipid interferometry of the central cornea (8.0 mm wide \times 7.2 mm long) performed with DR-1 α for the 138 eyes of the 76 subjects. Each movie had a randomly assigned number for analysis. Each rater categorized each image according to the interferometric pattern classification. The test was performed by each rater at a separate site. After the test, the assigned numbers of the classified images were recorded for statistical analysis.

Interrater Reliability

An introduction to and explanation of the interferometric pattern classification were given before the test by a dry eye expert (R.A.). Consistency between the results for R.A. and those for each of two groups of raters including two Japanese and two Korean physicians (S.E., N.M., K.Y.S., and J.L.C.) and two Japanese nonphysicians (T.F. and Yasuko Fukuda, Itoh Clinic) was evaluated as interrater reliability.

Intrater Reliability

The dry eye expert (R.A.) who participated in determination of interrater reliability performed the test a second time after an interval of >2 weeks. Consistency between the results of the first and second tests was evaluated as intrater reliability.

Statistical Analysis

All statistical analysis was performed with Stata MP software, release 14.1 (StataCorp LP, College Station, TX, USA). Pairwise κ values were determined for assessment of interrater and intrater reliability. In addition, Fleiss's κ value was computed for the seven evaluators as a group, for the five physicians as a group compared with the two nonphysicians as a second group, and for the three Japanese physicians as a group compared with the two Korean physicians as a second group. For comparison of groups of raters, the difference in Fleiss's κ between groups was bootstrapped for 1000 replications, and the asymptotic SE of the difference was computed. Each test of the difference in κ between groups was performed by calculating the 95% CI of the difference with the use of the bootstrapped SE. If the 95% CI excluded the value of zero, then the difference was declared statistically significant at the 5% level of type I error ($\alpha = 0.05$). Fleiss's κ values of <0.01, 0.01–0.20, 0.21–0.40, 0.41–0.60, 0.61–0.80, and 0.81–1.00 were considered as poor, slight, fair, moderate, substantial, and almost perfect agreement, respectively.³¹ The Kruskal-Wallis test was applied for comparison of parameters among three groups. Differences in parameters between two groups were evaluated with the Steel-Dwass test. A *P* value of <0.05 was considered statistically significant.

RESULTS

The study subjects with dry eye were classified into groups of non-SS-ADDE, MGD, and non-SS-ADDE/MGD comprising 23 eyes of 14 patients (4 males and 10 females; mean age \pm SD, 66.2 \pm 15.9 years), 51 eyes of 27 patients (12 males and 15 females; 60.4 \pm 12.2 years), and 10 eyes of 6 patients (3 males and 3 females; 55.2 \pm 12.5 years), respectively (Fig. 2).

Repeatability of LLT and NIBUT Measurements

The intraexaminer coefficient of variation for measurement of LLT or NIBUT was 0.98 \pm 0.32% and 4.72 \pm 1.76%, respectively.

TABLE 1. Tear Film Parameters According to Tear Interferometric Pattern for All Study Subjects

Parameter	Interferometric Pattern Classification				P
	0	1	2	Total	
LLT, nm	100.0 ± 32.3	114.3 ± 28.5	45.8 ± 17.5	81.6 ± 39.6	<0.0001
Lid margin abnormalities, 0-4	0.2 ± 0.5	1.1 ± 0.6	1.9 ± 0.9	1.1 ± 1.0	<0.0001
NIBUT, s	10.4 ± 3.4	2.5 ± 1.3	2.2 ± 1.2	5.4 ± 4.6	<0.0001
BUT, s	5.9 ± 3.5	3.0 ± 1.8	3.1 ± 1.7	4.1 ± 2.9	<0.0001
SPK score, 0-9	0.1 ± 0.3	0.9 ± 0.9	0.7 ± 0.5	0.5 ± 0.7	<0.0001
Meibum grade, 0-3	0.0 ± 0.0	0.1 ± 0.3	1.6 ± 0.8	0.7 ± 0.9	<0.0001
Meiboscore, 0-6	2.7 ± 1.5	3.0 ± 1.9	3.7 ± 1.3	3.2 ± 1.6	0.0054
Schirmer value, mm	8.7 ± 5.4	3.9 ± 3.8	10.6 ± 7.8	8.4 ± 6.7	<0.0001
LLT/Schirmer value	17.7 ± 18.9	59.2 ± 47.6	8.4 ± 9.9	23.3 ± 32.6	<0.0001

Data are means ± SD. P values for comparisons of tear film parameters among the three interferometric patterns were determined with the Kruskal-Wallis test. For calculation of the LLT/Schirmer test value ratio, a Schirmer test value of 0 was assigned a value of 0.9 mm.

Validation Test for Tear Interferometric Pattern Classification

The κ value for evaluation of intrarater reliability for classification of tear interferometric patterns by a dry eye expert (R.A.) who performed the test twice was 0.90. Evaluation of pairwise interrater reliability for performance of the validation test yielded a range of κ values from 0.57 to 0.94. Fleiss's κ values for all seven evaluators, the five physicians, the two nonphysicians, the three Japanese physicians, and the two Korean physicians were 0.79, 0.75, 0.87, 0.75, and 0.75, respectively. The 95% CI for the difference between Japanese physicians and Korean physicians ranged from -0.1164 to +0.1164, which does not exclude zero, and the difference is therefore not statistically significant (the difference is zero, and the 95% CI is distributed symmetrically around zero).

Relation of Tear Film Parameters and Clinical Subtypes of Dry Eye to Tear Interferometric Patterns

Tear film parameters for eyes of both normal subjects and dry eye patients classified according to the three types of interferometric pattern (0, 1, and 2) are shown in Table 1, and the statistical significance of differences in these parameters between pairs of the three groups are shown in Table 2. The LLT in group 2 was significantly smaller than that in the other two groups. Lid margin abnormality score was significantly higher in group 2 than in group 1, as well as in group 1 than in group 0. Both NIBUT and BUT were significantly longer and SPK score was significantly smaller in group 0 than in

groups 1 and 2. Meibum grade was significantly worse in group 2 than in the other two groups. Meiboscore was significantly higher in group 2 than in group 0. The Schirmer test value was significantly lower in group 1 than in the other two groups. The LLT/Schirmer test value ratio differed significantly for each pairwise comparison, with the distribution of this parameter among each group of eyes of both normal subjects and dry eye patients being shown in Figure 3. We also examined the relation between tear interferometric patterns and clinical classification of the study subjects (Table 3). The interferometric pattern classification of 0, 1, and 2 coincided well with the clinical classification of normal, non-SS-ADDE, and MGD, respectively.

Relation Between LLT and Schirmer Test Value According to Tear Interferometric Pattern

The relation between LLT and tear secretion as measured by the Schirmer test was examined for each tear interferometric pattern (Fig. 4A) and for each clinical diagnosis (Fig. 4B). Interferometric pattern 1 was apparent only in eyes with an LLT of >74 nm. Eyes with interferometric pattern 1 tended to

TABLE 2. P Values (Steel-Dwass Test) for Pairwise Comparisons of Tear Film Parameters Among Tear Interferometric Patterns for All Study Subjects

Parameter	Pattern Classification		
	0 vs. 1	0 vs. 2	1 vs. 2
LLT	0.1097	<0.0001	<0.0001
Lid margin abnormalities	<0.0001	<0.0001	<0.0001
NIBUT	<0.0001	<0.0001	0.5684
BUT	0.0001	<0.0001	0.9966
SPK score	<0.0001	<0.0001	0.5630
Meibum grade	0.0235	<0.0001	<0.0001
Meiboscore	0.8522	0.0028	0.1957
Schirmer value	<0.0001	0.5386	<0.0001
LLT/Schirmer value	<0.0001	<0.0001	<0.0001

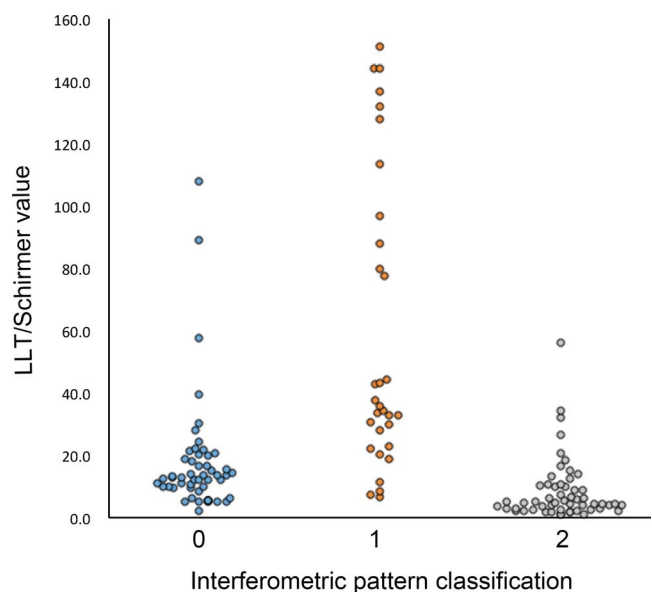


FIGURE 3. Distribution of the LLT/Schirmer test value ratio for all study subjects according to interferometric pattern classification. If the Schirmer test value was 0 mm, we used a value of 0.9 mm to calculate the ratio.

TABLE 3. Comparison of Tear Interferometric Pattern Classification With Clinical Diagnosis for the Study Subjects

Pattern	Normal	Non-		Total
		SS-ADDE	MGD	
0	47	3	3	53
1	5	20	2	30
2	2	0	46	55
Total	54	23	51	138

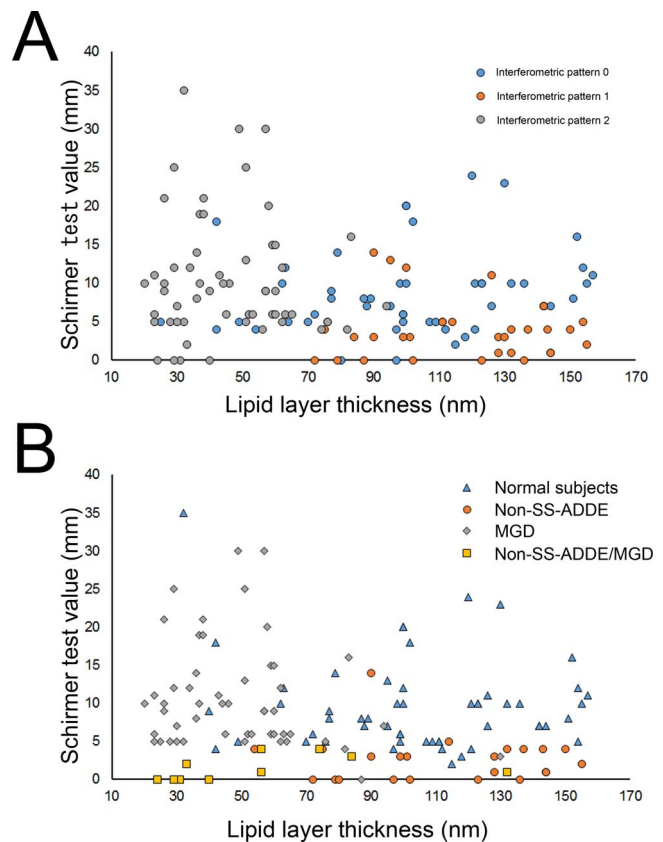
be distributed in the area of the plot corresponding to a normal or high LLT and a low Schirmer test value, whereas those with interferometric pattern 2 tended to be distributed in the area corresponding to a low LLT and a normal or high Schirmer test value. The distributions of normal, non-SS-ADDE, and MGD subjects were similar to those of interferometric patterns 0, 1, and 2, respectively. Non-SS-ADDE/MGD subjects were localized to the area corresponding to a low LLT and a low Schirmer test value.

DISCUSSION

We classified tear interferometric kinetics into three patterns and examined the relation of these patterns to subtypes of dry eye defined on the basis of tear film parameters. Our results have shown that the interferometric pattern classification largely coincides with the subtypes of dry eye. Interferometric patterns 1 and 2 were associated with a normal or high and a low LLT, respectively. The ratio of LLT to the Schirmer test value as a measure of tear secretion differed significantly for pairwise comparisons among all three interferometric patterns. We therefore suggest that our classification of tear interferometric kinetics reflects the balance of tear film components.

Tear interferometry is a noninvasive technique for assessment of the precorneal tear film that allows observation of the surface interference fringe of the lipid layer of the tear film,^{6,8,12,32,33} as well as semiquantitative^{5,14,34,35} or quantitative²⁸ evaluation of LLT. In addition to observing the interferometric fringe, we evaluated the stability of the tear film by determining NIBUT. We combined evaluation of the interferometric fringe with measurement of NIBUT to classify interferometric kinetics into three patterns. The tear interferometric fringe pattern was previously related to the severity of dry eye,¹¹ whereas tear dynamics visualized by interferometry were related to the volume of the aqueous layer of the tear film,^{15,23} suggesting that the interferometric fringe pattern indicates the condition of both the lipid and aqueous layers. Given that our classification is based not only on the interferometric fringe pattern but also on tear film stability, it potentially reflects the relation between tear dynamics and the condition of the lipid layer.

The three interferometric patterns of our classification system were found to be highly related to the clinical condition of the tear film based on measurement of tear film parameters. In general, tear film stability is disrupted in subjects with any tear film abnormalities, giving rise to a shortened tear film BUT. The presence of a shortened NIBUT was incorporated into our classification scheme to distinguish patterns 1 and 2 from pattern 0, reflecting abnormal and normal conditions of the tear film, respectively. The three patterns were also based on the interferometric fringe, with patterns 0, 1, and 2 corresponding to a pearl-like, Jupiter-like, or crystal-like fringe appearance. The Jupiter-like appearance was associated with a thickened lipid layer of the tear film (approximately >70 nm), consistent with previous findings.¹⁴ The crystal-like appear-

**FIGURE 4.** Relation between Schirmer test value and LLT for all study subjects according to interferometric pattern classification (A) or dry eye subtype (B).

ance, reflecting grayish amorphous interferometric fringe, was associated with a reduced LLT. Interferometric pattern 1 (Jupiter-like appearance with a shortened NIBUT) thus corresponds to an unstable tear film with a thick lipid layer, indicative of loss of the aqueous layer, or ADDE. On the other hand, interferometric pattern 2 (crystal-like appearance with a shortened NIBUT) corresponds to an unstable tear film with a thin lipid layer, indicative of a tear film with an insufficient lipid layer but with sufficient tear fluid, or EDE. Indeed, we found that most cases of non-SS-ADDE and of MGD showed interferometric patterns of classes 1 and 2, respectively. Furthermore, patients with both non-SS-ADDE and MGD showed either pattern 1 or pattern 2, suggesting that the interferometric pattern of such individuals may be influenced by the tear condition of the moment (reduced aqueous or lipid volume). Our interferometric pattern classification may thus be applicable to identifying the current tear condition of these patients.

A previous study found that tear film evaporation does not necessarily correlate with LLT.³⁶ Other studies showed that NIBUT or BUT, each of which reflects tear film stability, was not significantly correlated with LLT.^{23,28,37–39} In the present study, we measured both LLT and tear secretion as indicated by Schirmer's test. We found that many cases categorized as interferometric pattern 1 showed an increased LLT, reduced tear secretion, and shortened NIBUT. Furthermore, these cases tended to correspond to a clinical diagnosis of non-SS-ADDE. The shortened NIBUT and increased LLT in these subjects indicate that an increase in LLT does not necessarily improve tear film stability. Rather, the balance between the aqueous and lipid layers is likely to be a key determinant of tear film stability.

Validation analysis for interferometric pattern classification revealed κ values for intra- and interrater reliability ranging from 0.57 to 0.94, with the physician raters including a dry eye specialist and general ophthalmologists. Furthermore, Japanese and Korean physicians showed similar validation results, with no significant difference being detected in their performance. Similar results were obtained for comparison between physicians and nonphysicians. The moderate to almost perfect κ values obtained reflect the easily discriminated classification criteria, and they suggest that our classification system is applicable to international evaluation of interferometry data.

Our interferometric pattern classification has limitations with regard to evaluation of tear film abnormalities, however. In subjects with an obviously reduced volume of tear fluid or with severe corneal epithelial disorders, such as Sjögren syndrome, Stevens-Johnson syndrome, or graft-versus-host disease, an interferometric fringe is not observed because of the inadequate tear film. The interferometric pattern classification is thus not applicable to such patients. The dynamics of the lipid layer in individuals with these diseases remain to be investigated.

We also here propose the LLT/Schirmer test value ratio as an index of tear film balance. Although the Schirmer test value is thought to fluctuate, the test is still a standard procedure for the evaluation of tear fluid secretion.⁴⁰ Loss of balance between LLT and tear fluid volume reflects an imbalanced condition of the tear film. The LLT/Schirmer test value ratio differed significantly for all pairwise comparisons among the three interferometric patterns, suggesting that imbalance in the tear film condition is reflected in the classification of interferometric kinetics. The balance of tear film components may thus be critical for maintenance of the tear film, with loss of such balance giving rise to dry eye disease.

Subjects with interferometric pattern 1 tended to have a normal or high LLT and a low Schirmer test value, whereas those with interferometric pattern 2 tended to have a low LLT and a normal or high Schirmer test value. We have previously shown that tear fluid secretion is increased in individuals with MGD, suggesting that such secretion increases to compensate for the reduced production of the lipid layer.²¹ We have now found that LLT tended to be increased in subjects with reduced tear fluid secretion, suggesting that production of the lipid layer increases to compensate for the reduced volume of tear fluid. Given that the condition of the tear film is thought to be dynamic, such compensation would also be expected to be dynamic. Secretion of both tear fluid and lipid components of the tear film may be able to adapt rapidly to changing conditions, whereas adaptation of mucin production may occur over a longer time frame. The mechanisms responsible for sensing and responding to changes in the condition of the tear film remain to be determined. Subjects classified as non-SS-ADDE/MGD tended to have both a low LLT and a low Schirmer test value. Such individuals appear to lack the ability to increase secretion of both aqueous and lipid components of the tear film and thus possess defective compensatory systems.

Blinking may represent an important reflex response to dynamic changes in the tear film condition. In the present study, we evaluated tear interferometric patterns after each subject was asked to blink naturally twice to maintain the tear film condition. Our data therefore reveal the compensatory systems of the tear film without the influence of blinking.

In summary, we proposed a classification of tear interferometric kinetics that is closely related to the condition of the tear film. Evaluation of patients based on our tear interferometric patterns is expected to be widely applicable for characterization of tear film kinetics and dry eye pathophysiology, and it may provide a strategy for dry eye management.

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References

1. Wolff E. The muco-cutaneous junction of the lid margin and the distribution of the tear fluid. *Trans Ophthalmol Soc UK*. 1946;66:291-308.
2. Mishima S, Maurice DM. The oily layer of the tear film and evaporation from the corneal surface. *Exp Eye Res*. 1961;1:39-45.
3. Lemp MA. Report of the National Eye Institute/Industry workshop on Clinical Trials in Dry Eyes. *CLAO J*. 1995;21:221-232.
4. McDonald JE. Surface phenomena of tear films. *Trans Am Ophthalmol Soc*. 1968;66:905-939.
5. Norn MS. Semiquantitative interference study of fatty layer of precorneal film. *Acta Ophthalmol*. 1979;57:766-774.
6. Guillon JP. Non-invasive Tearscope Plus routine for contact lens fitting. *Contact Lens Anterior Eye*. 1998;21(suppl 1):S31-S40.
7. Olsen T. Reflectometry of the precorneal film. *Acta Ophthalmol*. 1985;63:432-438.
8. Doane MG. An instrument for in vivo tear film interferometry. *Optometry Vis Sci*. 1989;66:383-388.
9. Newton I, ed. *Opticks: Or a Treatise of the Reflections, Refractions, Inflections and Colors of Light*. 2nd ed. London, Printed for William and John Innys at the West End of St. Paul's. 1718:168-206.
10. Danjo Y, Hamano T. Observation of precorneal tear film in patients with Sjogren's syndrome. *Acta Ophthalmol Scand*. 1995;73:501-505.
11. Yokoi N, Takehisa Y, Kinoshita S. Correlation of tear lipid layer interference patterns with the diagnosis and severity of dry eye. *Am J Ophthalmol*. 1996;122:818-824.
12. Goto E, Tseng SC. Differentiation of lipid tear deficiency dry eye by kinetic analysis of tear interference images. *Arch Ophthalmol*. 2003;121:173-180.
13. Thai LC, Tomlinson A, Doane MG. Effect of contact lens materials on tear physiology. *Optometry Vis Sci*. 2004;81:194-204.
14. Korb DR, Baron DF, Herman JP, et al. Tear film lipid layer thickness as a function of blinking. *Cornea*. 1994;13:354-359.
15. Goto E, Tseng SC. Kinetic analysis of tear interference images in aqueous tear deficiency dry eye before and after punctal occlusion. *Invest Ophthalmol Vis Sci*. 2003;44:1897-1905.
16. Goto E, Dogru M, Fukagawa K, et al. Successful tear lipid layer treatment for refractory dry eye in office workers by low-dose lipid application on the full-length eyelid margin. *Am J Ophthalmol*. 2006;142:264-270.
17. Goto E, Matsumoto Y, Kamoi M, et al. Tear evaporation rates in Sjogren syndrome and non-Sjogren dry eye patients. *Am J Ophthalmol*. 2007;144:81-85.
18. Olson MC, Korb DR, Greiner JV. Increase in tear film lipid layer thickness following treatment with warm compresses in patients with meibomian gland dysfunction. *Eye Contact Lens*. 2003;29:96-99.
19. Shimazaki J, Sakata M, Tsubota K. Ocular surface changes and discomfort in patients with meibomian gland dysfunction. *Arch Ophthalmol*. 1995;113:1266-1270.
20. Yokoi N, Mossa F, Tiffany JM, Bron AJ. Assessment of meibomian gland function in dry eye using meibometry. *Arch Ophthalmol*. 1999;117:723-729.

21. Arita R, Morishige N, Koh S, et al. Increased tear fluid production as a compensatory response to meibomian gland loss: a multicenter cross-sectional study. *Ophthalmology*. 2015;122:925-933.
22. King-Smith PE, Fink BA, Nichols JJ, Nichols KK, Braun RJ, McFadden GB. The contribution of lipid layer movement to tear film thinning and breakup. *Invest Ophthalmol Vis Sci*. 2009;50:2747-2756.
23. King-Smith PE, Hinel EA, Nichols JJ. Application of a novel interferometric method to investigate the relation between lipid layer thickness and tear film thinning. *Invest Ophthalmol Vis Sci*. 2010;51:2418-2423.
24. Lemp MA, Crews LA, Bron AJ, Foulks GN, Sullivan BD. Distribution of aqueous-deficient and evaporative dry eye in a clinic-based patient cohort: a retrospective study. *Cornea*. 2012;31:472-478.
25. Shimazaki J. Definition and diagnosis of dry eye. *Atarashii Ganka (J Eye)*. 2006;24:181-184.
26. van Bijsterveld OP. Diagnostic tests in the Sicca syndrome. *Arch Ophthalmol*. 1969;82:10-14.
27. Amano S, Arita R, Kinoshita S, et al.; for the Japanese Dry Eye Society MGD Working Group. Definition and diagnostic criteria for meibomian gland dysfunction. *Atarashii Ganka (J Eye)*. 2010;27:627-631.
28. Finis D, Pischel N, Schrader S, Geerling G. Evaluation of lipid layer thickness measurement of the tear film as a diagnostic tool for Meibomian gland dysfunction. *Cornea*. 2013;32:1549-1553.
29. Arita R, Itoh K, Maeda S, et al. Proposed diagnostic criteria for obstructive meibomian gland dysfunction. *Ophthalmology*. 2009;116:2058-2063.
30. Arita R, Itoh K, Inoue K, Amano S. Noncontact infrared meibography to document age-related changes of the meibomian glands in a normal population. *Ophthalmology*. 2008;115:911-915.
31. Landis JR, Koch GG. The measurement of observer agreement for categorical data. *Biometrics*. 1977;33:159-174.
32. King-Smith PE, Fink BA, Fogt N, Nichols KK, Hill RM, Wilson GS. The thickness of the human precorneal tear film: evidence from reflection spectra. *Invest Ophthalmol Vis Sci*. 2000;41:3348-3359.
33. Hamano H. The change of precorneal tear film by the application of contact lenses. *Contact Intraocular Lens Med J*. 1981;7:205-209.
34. Goto E, Dogru M, Kojima T, Tsubota K. Computer-synthesis of an interference color chart of human tear lipid layer, by a colorimetric approach. *Invest Ophthalmol Vis Sci*. 2003;44:4693-4697.
35. Khamene A, Negahdaripour S, Tseng SC. A spectral-discrimination method for tear-film lipid-layer thickness estimation from fringe pattern images. *IEEE Trans Biomed Eng*. 2000;47:249-258.
36. King-Smith PE, Reuter KS, Braun RJ, Nichols JJ, Nichols KK. Tear film breakup and structure studied by simultaneous video recording of fluorescence and tear film lipid layer images. *Invest Ophthalmol Vis Sci*. 2013;54:4900-4909.
37. Suzuki S, Goto E, Dogru M, et al. Tear film lipid layer alterations in allergic conjunctivitis. *Cornea*. 2006;25:277-280.
38. Giraldez MJ, Naroo SA, Resua CG. A preliminary investigation into the relationship between ocular surface temperature and lipid layer thickness. *Contact Lens Anterior Eye*. 2009;32:177-180.
39. Fenner BJ, Tong L. More to stable tears than thickness of the tear film lipid layer. *Invest Ophthalmol Vis Sci*. 2015;56:1601.
40. Research in dry eye: report of the Research Subcommittee of the International Dry Eye WorkShop (2007). *Ocular Surf*. 2007;5:179-193.