

Upper and Lower Tear Menisci in Sjögren's Syndrome Dry Eye

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PURPOSE. To measure the tear menisci in Sjögren's syndrome dry eye (SSDE) by optical coherence tomography (OCT) and to determine its relationships with the clinical tests.

METHODS. Twenty-six SSDE, 26 non-SSDE, and 26 control subjects completed the Ocular Surface Disease Index (OSDI) before OCT determination of upper tear meniscus volume (UTMV), lower tear meniscus volume (LTMV), and total tear meniscus volume (TTMV). These were followed by measurements of noninvasive tear breakup time (NITBUT), fluorescein tear breakup time (FTBUT), fluorescein staining, Schirmer test, and corneal confocal microscopy.

RESULTS. UTMV, LTMV, and TTMV were the lowest in SSDE among the three groups ($P < 0.05$). High sensitivity and specificity of UTMV (1.0; 0.96), LTMV (0.92; 0.92), and TTMV (0.96; 0.96) were found in the diagnosis of SSDE. For SSDE, the areas under the UTMV, LTMV, and TTMV receiver operating characteristic curves were larger than those in NITBUT, FTBUT, and Schirmer test ($P < 0.005$). In the SSDE group, NITBUT was correlated with UTMV ($R = 0.41$) and TTMV ($R = 0.39$) ($P < 0.05$). Fluorescein staining score was significantly correlated with UTMV ($R = -0.46$), LTMV ($R = -0.41$), and TTMV ($R = -0.53$) ($P < 0.05$). Superficial epithelial cell density was correlated with UTMV ($R = 0.18$), LTMV ($R = 0.51$), and TTMV ($R = 0.44$) ($P < 0.05$).

CONCLUSIONS. Tear menisci volumes estimated by OCT may have great potential in the diagnosis and monitoring of SSDE. They can also reflect ocular surface damage and tear film stability. (*Invest Ophthalmol Vis Sci.* 2011;52:9373-9378) DOI:10.1167/iops.11-7431

Sjögren's syndrome (SS) is a systemic autoimmune disease that targets mucosal tissues and their supporting secretory glands. It has a reported prevalence of 0.15% to 3.3%, depending on the diagnostic criteria used,¹⁻³ and 95% of the patients are women. Although most are perimenopausal and postmenopausal women, patients as young as 20 years of age have been reported.^{3,4} The lacrimal gland disease in SS includes multifo-

cal lobular lymphocytic infiltration, disorganization of glandular architecture with loss of secretory acini, and proliferation of ductal epithelia forming epithelial islands.⁴ These changes contribute to a profound decrease in the secretion of water and proteins and ultimately in various severe dry eye symptoms and signs and even blindness.^{5,6}

Tears are distributed in the upper tear meniscus (UTM), lower tear meniscus (LTM), precorneal tear film, and cul-de-sac.⁶ Both menisci account for 75% to 90% of the total,⁷ and they supply tears to the precorneal tear film. The LTM has been visualized and quantified using video interference meniscometry, photography, fluorescein-stained meniscometry, and optical coherence tomography (OCT) in healthy, elderly, and dry eye subjects.⁸⁻¹⁵ However, these methodologies have not been applied to the UTM, which contains an equivalent volume of tears and contributes significantly to tear dynamics.¹⁶ Furthermore, those methodologies cannot be used to estimate tear meniscus volume, a good indicator of the overall tear volume.¹⁰ Recent advances in OCT have enabled highly repeatable simultaneous measurement of upper, lower, and total meniscus volumes in normal eyes and in dry eyes.¹⁷⁻¹⁹

In our previous study using OCT, we found that the variables of tear menisci were significantly decreased in non-SS dry eye (non-SSDE) with aqueous tear deficiency.²⁰ We showed that the measurement of the tear meniscus dimensions by OCT was a good diagnostic method with high sensitivity and specificity for non-SSDE. SSDE also has a serious influence on a patient's quality of daily life. The clinical characteristics of SSDE and non-SSDE are modified by disease progression and evolve in predictably different ways.²¹ However, little is known about tear menisci in SSDE that might be valuable in diagnosis and in monitoring the development of the disease. The purpose of this study was to use real-time OCT to measure the volumes of the UTM and LTM in SSDE patients and to compare the results with non-SSDE patients and healthy control subjects. We then determined the relationships between tear menisci variables and noninvasive tear breakup time (NITBUT), fluorescein tear breakup time (FTBUT), fluorescein staining, Schirmer I test, and in vivo corneal confocal microscopy in SSDE.

SUBJECTS AND METHODS

Subjects

This research was conducted in strict accordance with the guidelines of the Wenzhou Medical College Review Board (Wenzhou, Zhejiang, China) and in accordance with the tenets of the Declaration of Helsinki. Informed consent was obtained from each subject after a full explanation of the procedures. Three groups of 26 subjects were enrolled in this study: 26 SSDE subjects (25 women, 1 man; age 42.3 ± 9.7 years [mean \pm SD]), 26 non-SSDE subjects (23 women, 3 men; 41.2 ± 10.0 years), and 26 non-dry eye controls (21 women, 5 men; 40.8 ± 9.3 years). All SS participants had been diagnosed with

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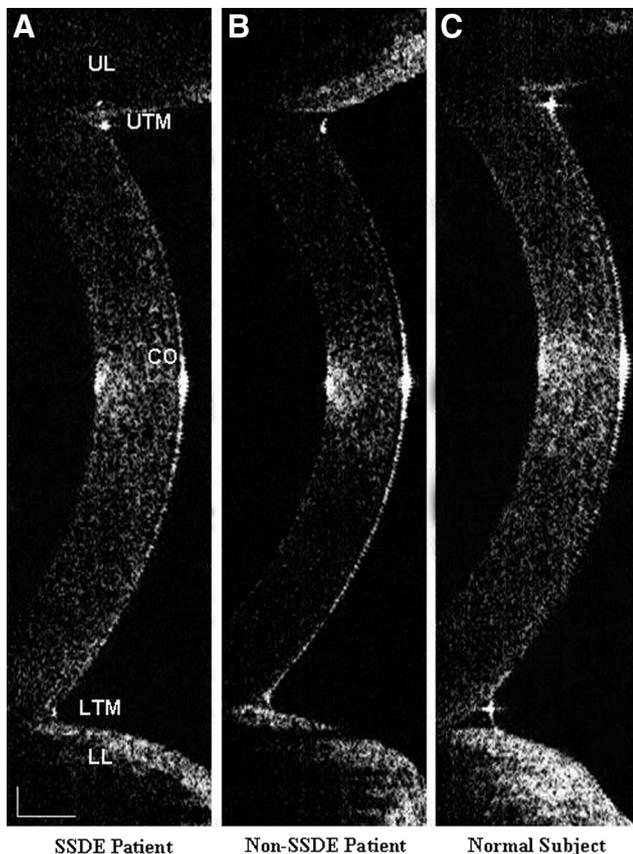


FIGURE 1. Upper and lower tear menisci in an SSDE patient, a non-SSDE patient, and a healthy subject. Real-time anterior segment OCT images of the cornea (CO) and the UTM and LTM around the upper and lower eyelids (UL and LL) were obtained immediately after a full blink in each (A) SSDE, (B) non-SSDE, and (C) healthy subject. Both UTM and LTM of SSDE patients (A) were much smaller than those of non-SSDE patients (B) and of healthy subjects (C). In addition, the tear menisci of non-SSDE patients (B) were significantly smaller than those of the healthy subjects. Scale bar, 500 μm .

primary or secondary SS at the Clinic of Wenzhou Eye Hospital using the American-European consensus criteria of 2002.²² Recruitment of these patients was achieved through telephone calls from the database of that clinic. No further preliminary screening was performed on this group because all had been confirmed as SS patients. Non-SSDE patients were also diagnosed at the Clinic of Wenzhou Eye Hospital. After excluding SS, all the subjects who reported dry eye symptoms with quantitative disturbance of the tear film (TBUT <5 seconds or Schirmer test <5 mm/s) and epithelial damage (fluorescein staining greater than 3 points) were diagnosed with non-SSDE.²³ Eyes with current punctal occlusion, conjunctivochalasis, corneal transplantation, anterior blepharitis, infectious conjunctivitis, contact lens wear, or other ocular diseases were excluded from this study.

Experimental Procedure

All enrolled subjects were tested in the following sequence: Ocular Surface Disease Index (OSDI), which is a psychometrically tested, valid, and reliable instrument for measuring the severity of dry eye²⁴; anterior segment imaging by OCT; NITBUT measured with a Keeler Tearscope (Keeler, Ltd., Windsor, United Kingdom)²⁵; slit-lamp microscope examination; FTBUT; fluorescein staining of the cornea in five regions, scored from 0 to 3 in each region²⁶; modified Schirmer I test with anesthetic; and confocal microscopy of cornea.

Measurement of the Tear Meniscus Volume by Anterior Segment OCT

A custom-built, high-speed, real-time anterior segment OCT was used in the present study to image the UTM and LTM, as previously reported.^{20,27} The light source was 1310-nm wavelength with a 60-nm bandwidth. It was affixed to a probe that was mounted on a standard slit lamp and connected to a telecentric light-delivery system. The widest scan width was 15 mm, with a 2-mm scan depth in air and 10- μm longitudinal resolution. The first clear image of the UTM and LTM after a blink and with the central specular hyperreflective reflex was selected to be processed. In addition, a digital camera was used to image the upper and lower eyelids between normal blinks. We used custom software to obtain the cross-sectional areas of both upper and lower menisci (UTMA and LTMA, respectively) and both eyelid lengths. The upper and lower tear meniscus volumes (UTMV and LTMV, respectively) were calculated based on the UTMA and LTMA, accounting for the curvature and lengths of the respective eyelids.^{18,28} The total tear meniscus volume (TTMV) was obtained as the sum of the UTMV and LTMV.

In Vivo Confocal Microscopy

Confocal microscopy was performed in the center of the cornea using a laser scanning confocal microscope (Heidelberg Retina Tomograph II with Rostock Cornea Module; Heidelberg Engineering, Dossenheim, Germany). The position of the fixation light was the same for all participants. Movement of the focal plane was performed manually. Cellular density was evaluated in the superficial, intermediate, and basal epithelial layers with a caliper tool (Analysis 3.1; Soft Imaging System, Munster, Germany). The area of the field was 400 \times 400 μm , and three images in each layer were chosen for analysis. Results were calculated as cells per square millimeter. Subbasal nerve tortuosity was classified in four grades according to the previous report.²⁹

Statistical Analysis

Descriptive statistics included means \pm standard deviations for all variables. Except for fluorescein staining scores in which the Mann-Whitney *U* test was used, ANOVA tests were performed to determine whether there were significant differences in each variable among the three groups. Tukey post hoc tests were performed to compare each variable between any two groups. $P < 0.05$ was considered significant for each comparison between any two groups. Tear meniscus variables UTMV, LTMV, and TTMV as diagnostic variables for SSDE were analyzed by receiver operating characteristic (ROC) curves to obtain the optimal cutoff values for sensitivity and specificity. The ROC curves were obtained by comparing the variables of SSDE and normal eyes. Pearson or Spearman's rank correlation was used to indicate the rela-

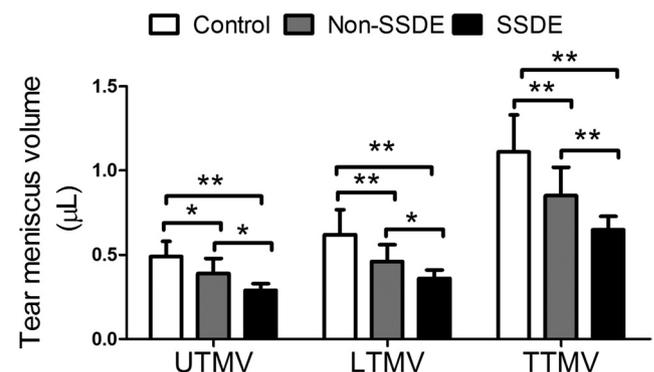


FIGURE 2. UTMV, LTMV, and TTMV in SSDE patients, non-SSDE patients, and healthy subjects. There were significant differences in UTMV, LTMV, and TTMV among SSDE patients, a non-SSDE patient, and a healthy subject, with the highest volumes in the healthy subjects and the lowest in the SSDE patients. * $P < 0.05$; ** $P < 0.01$.

tionships between tear meniscus and other variables in the SSDE group. Statistical analysis software (SPSS, version 13.0; SPSS, Inc., Chicago, IL) was used to analyze all data in this study.

RESULTS

In 8 of 26 SSDE patients, the tear meniscus could not be observed directly by slit-lamp microscope. In each of those eight patients, both UTM and LTM were clearly visualized by real-time OCT. There were significant differences in UTMV, LTMV, and TTMV among the SSDE, non-SSDE, and control groups. For all three volumes, the lowest was in the SSDE group and highest was in the

control group ($P < 0.05$; Figs. 1, 2). For ROC curves, larger areas under the curve indicated better sensitivity and specificity for diagnosis. For SSDE patients, the areas under the UTMV, LTMV, and TTMV ROC curves (Figs. 3A-C) were 0.984, 0.956, and 0.973, respectively. These values were larger than those for NITBUT (0.888; Fig. 3D), FTBUT (0.954; Fig. 3E), and Schirmer's test (0.737; Fig. 3F) ($P < 0.005$ for each comparison). When the cutoff value for abnormal UTMV was $0.37 \mu\text{L}$, good diagnostic accuracies were obtained with a sensitivity of 1.00 and a specificity of 0.96. For LTMV, when the cutoff value was $0.40 \mu\text{L}$, the sensitivity was 0.92, and the specificity was 0.92. For TTMV, when the cutoff value was $0.77 \mu\text{L}$, the sensitivity was 0.96, and the specificity was 0.96.

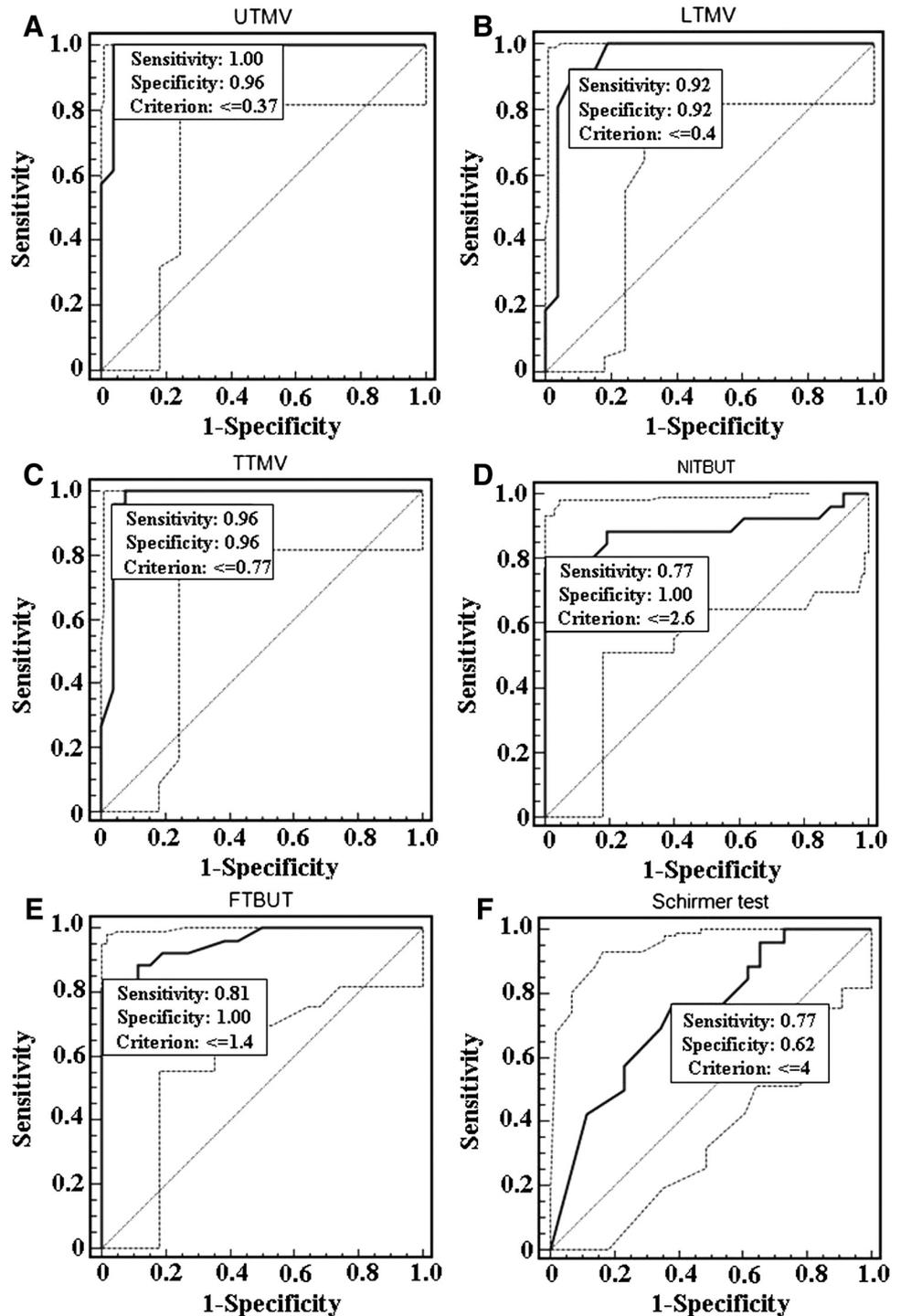


FIGURE 3. ROC curves, UTMV, LTMV, TTMV, NITBUT, FTBUT, and Schirmer test for the SSDE patients. For SSDE, the areas under the ROC curves for UTMV (A), LTMV (B), and TTMV (C) were 0.984, 0.956, and 0.973. These areas were larger than those for (D) NITBUT (0.888), (E) FTBUT (0.954), and (F) Schirmer test (0.737) ($P < 0.005$ for each comparison). Cutoff values for each abnormal variable are shown with the corresponding sensitivity and specificity.

TABLE 1. OSDI, NITBUT, FTBUT, Fluorescein Staining, and Schirmer I Test in the SSDE, Non-SSDE, and Control Groups

Group	OSDI	NITBUT (s)	FTBUT (s)	Fluorescein Staining	Schirmer I Test (mm/5 min)
SSDE	67.6 ± 20.7	2.3 ± 2.7	0.8 ± 1.0	7.7 ± 5.7	3.0 ± 4.1
Non-SSDE	31.7 ± 21.8	5.3 ± 3.0	2.3 ± 1.6	0.9 ± 2.1	6.2 ± 6.4
Control	2.0 ± 2.5	7.3 ± 5.4	4.2 ± 2.4	0.1 ± 0.6	10.2 ± 10.4

Brackets indicate significant differences: * $P < 0.05$.

FTBUT and NITBUT were significantly decreased in the SSDE group compared with the non-SSDE and control groups ($P < 0.05$ for each comparison; Table 1). Schirmer I test scores in the SSDE group were lower than in the control group ($P < 0.05$). The SSDE group also had significantly increased fluorescein staining and OSDI scores compared with the non-SSDE and the control groups ($P < 0.01$ for each comparison; Table 1). In the SSDE group, NITBUT was correlated with UTMV ($R = 0.41$) and TTMV ($R = 0.39$; $P < 0.05$). The fluorescein staining score was correlated with UTMV ($R = -0.46$; $P < 0.05$), LTMV ($R = -0.41$; $P < 0.05$), and TTMV ($R = -0.53$; $P < 0.01$). However, there were no significant correlations between the variables of the tear menisci and the Schirmer test, FTBUT, or OSDI in the SSDE group.

Consistent with previous reports,^{30,51} as shown by corneal confocal microscopy, the SSDE group had decreased cellular density in the superficial epithelial layers, increased subbasal nerve tortuosity, and decreased numbers of subbasal nerves compared with the non-SSDE and control groups ($P < 0.05$ each comparison; Fig. 4, Table 2). Superficial epithelial cell density was correlated with UTMV ($R = 0.18$; $P < 0.05$; Table 3), LTMV ($R = 0.51$; $P < 0.01$), and TTMV ($R = 0.44$; $P < 0.01$). There were no significant correlations between the variables of the tear menisci and cellular density in intermediate and basal epithelial layers, subbasal nerve tortuosity, or number of subbasal nerves ($P > 0.05$).

DISCUSSION

In the clinic, measurement by slit-lamp of LTM height has been the standard way of evaluating tear volume in the diagnosis and monitoring of dry eye.⁹ When the tear meniscus is too low, it cannot be directly observed by this technique. In this study, we found that the tear meniscus could not be observed directly by

slit-lamp microscopy in 8 of 26 SSDE patients; however, with real-time anterior segment OCT, all such low tear menisci were clearly visualized. We found that tear meniscus volumes were significantly lower in SSDE than in normal control and non-SSDE eyes. The traditional objective diagnosis of dry eye in the clinical setting relies on the Schirmer test, FTBUT, and vital staining of the ocular surface. These invasive tests may cause irritation and reflex tearing and can influence the results. In addition, these measurement methods are normally a one-time snapshot. However, the tear system is highly dynamic during the interblink period. These issues may lead to poor specificity and sensitivity of these tests. By contrast, the OCT used in this study can image both upper and lower tear menisci in a noninvasive and real-time way. We have previously shown the high sensitivity and specificity of the tear meniscus variables measured with this OCT device in diagnosing non-SSDE.^{19,32} In this study, the calculated values for UTMV, LTMV, and TTMV provided high sensitivity and specificity in the diagnosis of SSDE. In fact, these values were better than for the NITBUT, FTBUT, and Schirmer test. Therefore, we believe that the simultaneous evaluation of both tear menisci by our custom-built OCT instrument accurately reflects the deficiency of tear volume in SSDE. This could be very promising in the diagnosis and monitoring of SSDE.

Disagreement between the severity of dry eye symptoms and clinical signs has been demonstrated in many population-based studies.^{33,34} Schein et al.³³ found no association between the Schirmer test result or Rose Bengal staining score and dry eye symptoms in a study of 2240 patients. Another population-based study of 341 persons by Hay et al.³⁵ found only weak associations between self-reported symptoms of dry eyes and clinical tests. Damage in corneal nerves may lead to corneal mechanical hyperesthesia³⁶ or hypoesthesia,³⁷ which might explain the phenomenon of poor association between

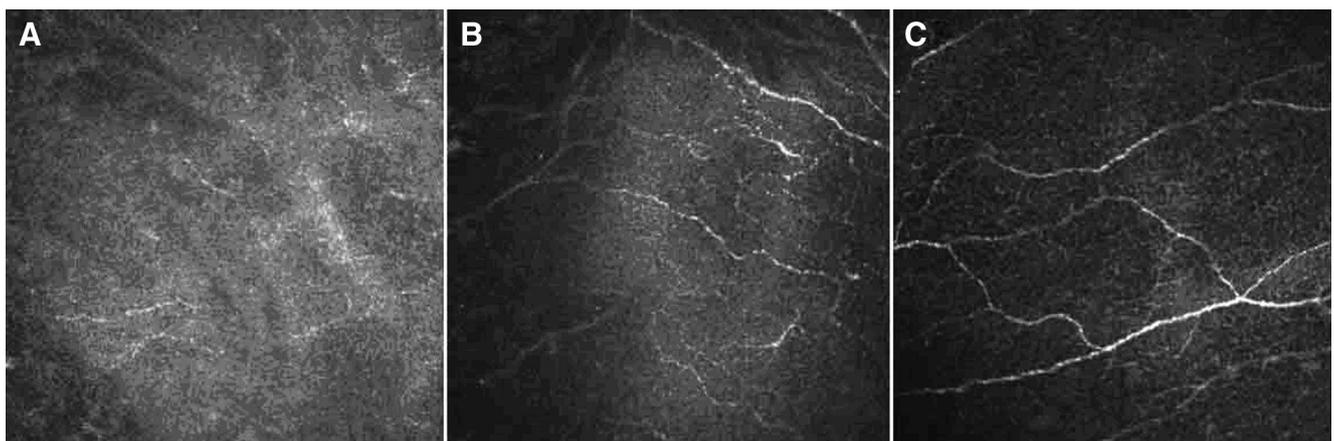


FIGURE 4. Subbasal nerves. The (A) SSDE group had greater subbasal nerve tortuosity and loss of nerve fibers than the (B) non-SSDE and (C) control groups.

TABLE 2. Epithelial Cell density and Subbasal Nerve in the SSDE, Non-SSDE, and Control Groups

Group	Superficial Epithelial Cell Density (cells/mm ²)	Intermediate Epithelial Cell Density (cells/mm ²)	Basal Epithelial Cell Density (cells/mm ²)	Subbasal Nerve Tortuosity	No. of Subbasal Nerves
SSDE	614 ± 264	3944 ± 1211	10007 ± 1870	3.5 ± 0.6	2.2 ± 1.9
Non-SSDE	* 948 ± 352	* 4620 ± 1466	9711 ± 2324	* 2.5 ± 0.6	* 7.7 ± 3.9
Control	1384 ± 394	5508 ± 1803	9690 ± 2097	0.7 ± 0.7	7.1 ± 3.6

Brackets indicate significant differences: * $P < 0.05$.

signs and symptoms in dry eye patients. In this study, we found no association between OSDI scores and variables of the tear menisci in SSDE patients. Consistent with these observations, we found that SSDE subjects had more severe pathologic corneal nerve alterations than did the non-SSDE patients and the control subjects.

NITBUT is a noninvasive method to observe the specular reflection of the tear film surface to estimate the stability of the tear film. Mainstone et al.⁸ reported that the LTM measured by photography was correlated with NITBUT. Wang et al.²⁵ also reported that LTM height and LTMA were correlated with NITBUT in healthy subjects. In this study, we found that UTMV and TTMV were significantly correlated with NITBUT, which means these meniscus volumes might be involved in the tear film instability in SSDE.

The association between FTBUT and the tear menisci is still controversial. Golding et al.³⁸ and Khurana et al.³⁹ suggested that the dimensions of tear menisci were correlated with FTBUT. In contrast, Savini et al.⁴⁰ found no relationship between LTM height measured with a commercial OCT and FTBUT. In the present study, we also found no significant correlations between the tear menisci variables and FTBUT. This was possibly due to reflexive tearing induced by the fluorescein itself.^{41,42} In addition, we found no significant correlation between the tear menisci and Schirmer test results, which has also been reported for healthy subjects.²⁵ The lack of correlation between the Schirmer test and tear meniscus measurements may be attributable to the use of topical anesthetic, which can affect tear secretion and drainage.⁴³ In addition, Schirmer I testing was performed after slit-lamp microscopy, FTBUT, and fluorescein staining examinations in this study. These invasive procedures before Schirmer I test may cause reflex tearing and affect the results of the Schirmer I test.

Punctate staining is an important sign of dry eye disease and ocular surface irritation. Punctate fluorescein-stained spots corresponded to damage in corneal superficial epithelial cells.

TABLE 3. Correlations among UTMV, LTMV, TTMV, and Other Tests in the SSDE

	UTMV	LTMV	TTMV
OSDI	0.06	-0.39	-0.21
NITBUT	0.41*	0.27	0.39*
FTBUT	0.38	0.19	0.33
Fluorescein staining	-0.46*	-0.41*	-0.53†
Schirmer I test	0.24	0.05	0.17
Superficial epithelial cell density	0.18*	0.51†	0.44†
Intermediate epithelial cell density	0.14	0.27	0.25
Basal epithelial cell density	0.03	0.27	0.17
Subbasal nerve tortuosity	-0.35	-0.27	-0.36
No. of subbasal nerves	-0.10	-0.27	-0.37

Correlations with R values > 0.40 (bold) were considered to be moderate to high.

* $P < 0.05$.

† $P < 0.01$.

Compromised tight junction integrity, increased superficial epithelial permeability, and cell death have been invoked as causes of fluorescein-stained spots.⁴⁴⁻⁴⁶ In this study, we found that UTMV, LTMV, and TTMV were significantly correlated with the fluorescein staining score and superficial epithelial cell density in the SSDE group. These observations, taken together, suggest that decreased tear menisci may be involved in the corneal superficial epithelial damage that can lead to irritation and symptoms in SSDE.

Subbasal sensory nerves in the cornea transmit afferent stimulation signals to the brainstem. After a series of interneurons, the efferent signal is transmitted through parasympathetic and sympathetic nerves to drive lacrimal tear production and secretion. Damage of corneal subbasal nerves can lead to decreased lacrimal tear production⁴⁷ and, consequently, lower the tear meniscus. However, we found no significant correlation between the variables of the tear menisci and subbasal nerve tortuosity or the number of subbasal nerves in this study. This may be because SS is a chronic autoimmune disorder characterized by infiltration of the lacrimal glands by mononuclear cells.^{48,49} It is likely that inflammation of the lacrimal gland rather than ocular surface nerve damage mainly contributes to the decreased tear production in SSDE.

In conclusion, tear meniscus volumes are lower in SSDE than in non-SSDE and healthy subjects. Tear meniscus volumes estimated by OCT are good candidates as diagnostic criteria. In addition, these volumes can serve as indicators of ocular surface damage and tear film stability. Therefore, we believe that measurement of tear meniscus volumes has great potential in monitoring the development of SSDE disease.

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