Supplementary Online Content

Uchino Y, Uchino M, Yokoi N, et al. Alteration of tear mucin 5AC in office workers using visual display terminals: the Osaka Study. *JAMA Ophthalmol*. Published online June 5, 2014. doi:10.1001/jamaophthalmol.2014.1008.

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This supplementary material has been provided by the authors to give readers additional information about their work.

eMethods

Tear Function test

Tear stability and quantity were assessed by 2 different parameters: TBUT and Schirmer test value to determine TBUT, fluorescein vital staining was performed, and the subjects were requested to blink 3 times to ensure adequate mixing of the fluorescein dye with the tears. A stopwatch was used to measure the time interval between the last complete blink and appearance of the first corneal black spot, and the mean of 3 measurements was considered as TBUT. The Schirmer test I was performed without topical anesthesia after all the other examinations were complete. Schirmer strips (Whatman No. 41, Showa, Tokyo) were placed at the outer one-third of the temporal lower conjunctival fornix for 5 min, after which they were removed. Then the length of wetness on the strip (in millimeters) was recorded.

Ocular Surface Evaluation

Two microliters of a preservative-free combination of 1% lissamine green dye and 1% fluorescein dye was instilled into the conjunctival sac by a micropipette. To evaluate the fluorescein and lissamine green staining of the ocular surface, the eye was divided into 3 equal compartments representing the cornea, and the nasal and temporal conjunctiva. The maximum staining score for each area was 3 points; therefore, the overall maximum staining score was 9 points.

Sample Collection

Tears were collected from unanesthetized eyes by a micropipette after instillation of 50 μL saline in the cul-de-sac followed by movement of the eyes to mix the tear fluid content[14, 18]. Individual tear samples were centrifuged for 10 min at 14,000 rpm at 20 °C to remove any debris. The induction of reflex tearing may be a problem during collection of tears. Collecting undiluted tears involves touching the conjunctiva which may actually reflex tearing. We chose to instill saline on the ocular surface to minimize reflex tearing, and to increase the yield of protein during our collection as previously published[19]. Impression cytology specimens were obtained after the administration of topical anesthesia with 0.4% oxybuprocaine hydrochloride (Benoxil ophthalmic solution 0.4%; Santen Pharmaceutical Co., LTD., Osaka, Japan)[20]. Two separate strips of cellulose acetate filter paper (Millipore HAWP 304, Bedford, MA, USA) were applied on the temporal bulbar conjunctiva, pressed gently by a glass rod, and removed. The strip was then carefully transferred into a tube (Eppendorf, Fremont, CA, USA) containing RNA Stabilization Reagent (RNAlater™; Qiagen, Valencia, CA, USA)[21]. All samples were promptly frozen at −80 °C until analysis.

eTable 1. 12 Questions of Subjective Symptoms for Dry Eye Diagnosis

① ocular fatigue / eye strain	⑦ blurred vision
② uncomfortable sensation	⑧ discharge
③ dry sensation	⑨ foreign body sensation
④ heavy sensation	⑩ red eye
⑤ sensitivity to bright light	⑪ itching
⑥ pain	⑫ excess tearing

*Possible answers to each questions about symptoms included “Always,” “Often,” “Sometimes,” or “Never.”

eTable 2. New Criteria for Dry Eye Diagnosis in Japan

1. Subjective symptoms

*Clinical symptoms more than one of 12 questions (Table 2B) with “constantly” or “often” was considered as positive subjective symptoms.

2. Tear fluid abnormalities

① Schirmer I test ≦ 5 mm wetting (5 minutes)

② Tear breakup time (TBUT) ≦ 5 sec

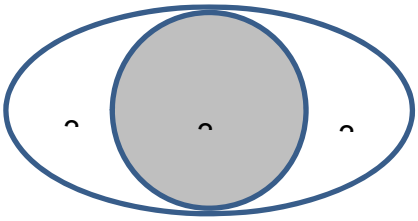
*Subject who meets either ① or ② is considered positive.

3. Keratoconjunctiva disorder

① Fluorescein stain

② Lissamine green stain

*Scoring 3 or higher out of 9 in either ① or ② is considered positive.



1. Subjective symptoms	○	×	○	○
2. Tear fluid abnormalities	○	○	×	○
3. Keratoconjunctiva disorder	○	○	○	×
Diagnosis	Definite Dry eye	Probable Dry Eye		

*Subjects with presence of 1or no positive criteria were diagnosed as normal

eTable 3. Visual Display Terminal Working Hours and Eye Strain

		Less than 6 hours	6 hours or longer	Total	Test
n		36	60	96	-
Eye strain (frequency)	Always / Often	12 (33%)	31 (52%)	43 (44%)	*p = 0.093
	Sometimes / no eye strain	24 (67%)	29 (48%)	53 (55%)	

* Fisher's exact test

