Abstract: Meibomian glands secrete the oily layer of the tear film, which prevents excessive evaporation of tear fluid. Dysfunction of meibomian glands is not only one of the causes of evaporative dry eye but also one of the main causes of entire dry eye. To understand the pathophysiology of meibomian gland dysfunction, it is important to evaluate both the morphology and function of the meibomian gland. We previously reported that meibography enabled visualization of the morphology of the meibomian gland. Meanwhile, tear interferometry was introduced as an evaluation method for the function of the meibomian gland. We combined observations of the oily layer and the aqueous layer of the tear film and found that a tear film compensatory system may work toward maintenance of tear film homeostasis. In this review, we describe both morphological evaluation systems for the meibomian gland, including noninvasive meibography, and functional evaluation systems, including tear interferometry. We further describe the morphological changes of the meibomian glands in various ocular surface diseases. Finally, we demonstrate the concept of a tear film compensatory system and propose a method for tear film component-oriented diagnosis.

Key Words: meibomian gland, meibomian gland dysfunction, noninvasive meibography, tear interferometry, dry eye, tear film

The meibomian gland was named after the German physician Heinrich Meibom, who discovered the existence of secretory glands inside the tarsal plates in the 17th century. Meibomian glands secrete oily components called meibum that form the oily layer of the tear film. The oily layer of the tear film prevents evaporation of the tear fluid, and thus meibomian gland dysfunction (MGD) mainly causes evaporative dry eye. The number of patients with evaporative dry eye is currently larger than the number with aqueous-deficient dry eye. The International Workshop on Meibomian Gland Dysfunction defines MGD as a chronic diffuse abnormality of the meibomian glands, commonly characterized by terminal duct obstruction or qualitative/quantitative changes in glandular secretion. The oily layer of the tear film in patients with MGD is unstable, resulting in ocular discomfort, ocular surface inflammatory reactions, and ocular surface diseases. Therefore, adequate evaluation of the morphology and function of meibomian glands is an important factor in diagnosing MGD.

MORPHOLOGY OF THE MEIBOMIAN GLAND VISUALIZED BY NONINVASIVE MEIBOGRAPHY

Meibography was introduced as a method for visualization of the meibomian gland. However, a previous observation system involving meibography had several disadvantages, such as pain, heat sensation, and limited visualization area, thus making it inadequate for regular clinical examinations. We previously described a noninvasive meibography system as a novel method for visualization of the meibomian gland. This noninvasive meibography system enabled us to observe the entire meibomian gland, from the nasal part to the temporal part.

FIGURE 1. Representative images of meibomian glands in the upper and lower eyelids of a normal subject obtained by noninvasive meibography. Reprinted with permission from the Association for Research in Vision and Ophthalmology from Efron et al. Copyright © 2017 Wolters Kluwer Health, Inc. All rights reserved.
of the upper and lower eyelids within 1 minute.12 A slit-lamp microscope equipped with a charge-coupled device camera and infrared-pass filter visualized the meibomian gland as a bright area (Fig. 1).12 This observation system clearly revealed dropout, shortening, dilation, or distortion of meibomian glands related to ocular surface diseases.12–19

Currently, 4 types of noninvasive meibography systems are commercially available (Table 1): slit-lamp–equipped type (BG-4M/DC4 BG-5 attached to a slit-lamp microscope; TOPCON, Tokyo, Japan),12 mobile type (Meibom Pen; Japan Focus Corporation, Tokyo, Japan),20 topography–equipped type (Keratograph 5M; OCULUS Optikgeräte GmbH, Wetzlar, Germany),21 and fundus camera–equipped type (Eye Top Topographer, Sirius Scheimpflug Camera, and Cobra Fundus Camera; CSO, Florence, Italy, and bon Optic Vertriebs GmbH, Lübeck, Germany).

We previously developed the meiboscore, a semiqualitative index obtained by noninvasive meibography.12 The meiboscore is defined as follows: partial or complete loss of meibomian glands is scored for each eyelid from grade 0 (no loss) to grade 3 (lost area constituting more than two-thirds of the total gland area). The meiboscore of each eyelid is then summed, giving a total meiboscore for the bilateral upper and lower eyelids ranging from 0 to 6.12 (Fig. 2). We found that a diagnostic cutoff value could be defined based on a comparative study of normal and MGD subjects, revealing the sensitivity and specificity of the meiboscore for the diagnosis of MGD.14 Other groups have reported several types of evaluation scales for the morphology of the meibomian gland.11,22 The subjective intrarater and interrater agreements of the scales were also evaluated.22 These qualitative evaluations of the meibomian gland were applied to diagnosis and evaluation of treatment for MGD. However, objective approaches to meibomian gland evaluation are much more appropriate for slight changes in the morphology of meibomian glands.23–26

### TABLE 1. Types of Noninvasive Meibography Systems

<table>
<thead>
<tr>
<th>Light source</th>
<th>Other functions</th>
<th>Mobile type</th>
<th>Topography attached type</th>
<th>Fundus camera attached type</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wavelength 890 nm</td>
<td>All functions with a slit lamp</td>
<td>Wavelength 940 nm</td>
<td>Diffuser</td>
<td>Infrared illumination</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Blue filter</td>
<td>Fundus camera</td>
</tr>
<tr>
<td>Product name</td>
<td>Company (City, Country)</td>
<td>Mobile type</td>
<td>Topography attached type</td>
<td>Fundus camera attached type</td>
</tr>
<tr>
<td>SL-D701 BG-4M/DC4 BG-5</td>
<td>TOPCON, (Tokyo, Japan)</td>
<td>Meibom Pen</td>
<td>Keratograph 5M</td>
<td>Eye Top Topographer, Sirius Scheimpflug Camera, and Cobra Fundus Camera</td>
</tr>
<tr>
<td>Meibom Pen</td>
<td>Japan Focus Corporation (Tokyo, Japan)</td>
<td>OCULUS Optikgeräte GmbH (Wetzlar, Germany)</td>
<td>Fluorescence imaging</td>
<td>CSO (Florence, Italy), bon Optic Vertriebs GmbH (Lübeck, Germany)</td>
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</tbody>
</table>

LIPCOF, lid parallel conjunctival folds; NIKBUT, noninvasive keratograph breakup time.

FIGURE 2. Grading of partial or complete loss of meibomian glands from grade 0 to grade 3 as the meiboscore. Reprinted with permission from Elsevier from Arita et al.12 Copyright Elsevier, Philadelphia, PA. All permission requests for this image should be made to the copyright holder.
TABLE 2. Previous Reports Related to Morphological Changes Evaluated by Noninvasive Meibography

<table>
<thead>
<tr>
<th>Number of Subjects</th>
<th>Findings</th>
<th>Reference</th>
</tr>
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<tbody>
<tr>
<td>Aging</td>
<td>Morphology of meibomian glands changed with age</td>
<td>Arita et al12 (Fig. 2)</td>
</tr>
<tr>
<td></td>
<td>Number of gland dropout was correlated with age</td>
<td>Ban et al25</td>
</tr>
<tr>
<td>Contact lenses</td>
<td>Meibomian gland loss increased with age</td>
<td>Yeotik et al28</td>
</tr>
<tr>
<td></td>
<td>Morphological changes in contact lens wearers were significantly worse than those in noncontact lens wearers</td>
<td>Arita et al13 (Fig. 3)</td>
</tr>
<tr>
<td></td>
<td>A best-fitting multivariate regression model revealed that the lost area of meibomian glands was higher in contact lens wearers (odds ratio, 2.45)</td>
<td>Pucker et al29</td>
</tr>
<tr>
<td></td>
<td>Changes in meibomian gland morphology and function were associated with contact lens wear</td>
<td>Alghamdi et al30</td>
</tr>
<tr>
<td></td>
<td>Long-term contact lens wear was associated with meibomian gland loss</td>
<td>Tang et al31</td>
</tr>
<tr>
<td>Allergy</td>
<td>Meibomian gland duct distortion was significantly higher in patients with allergic conjunctivitis</td>
<td>Arita et al38 (Fig. 4)</td>
</tr>
<tr>
<td></td>
<td>57 of the 58 subjects developed papillary hypertrophy and meibomian gland duct distortion</td>
<td>Na et al32</td>
</tr>
<tr>
<td>MGD</td>
<td>ROC curves were generated and AUC values were calculated to differentiate eyes with MGD from healthy eyes. Diagnosis with any 2 of ocular symptom score, lid margin abnormality score, and the meiboscore had sensitivity of 84.9% and specificity of 96.7%</td>
<td>Arita et al34 (Fig. 5)</td>
</tr>
<tr>
<td></td>
<td>Meibomian gland loss and the meibum showed a significant positive correlation</td>
<td>Eom et al35</td>
</tr>
<tr>
<td></td>
<td>Area of the chalazion showed partial or complete loss of meibomian glands</td>
<td>Srinivasan et al36</td>
</tr>
<tr>
<td></td>
<td>Area of the chalazion showed low reflectivity corresponding to lipid granules</td>
<td>Nemoto et al37</td>
</tr>
<tr>
<td></td>
<td>Long-term application of glaucoma eye drops affected the morphology and function of meibomian glands</td>
<td>Arita et al38</td>
</tr>
<tr>
<td></td>
<td>Blebs could give rise to meibomian gland loss, particularly when the blebs were avascular</td>
<td>Sagara et al39</td>
</tr>
<tr>
<td></td>
<td>The meiboscore was significantly higher in patients with phlyctenular keratitis</td>
<td>Suzuki et al40</td>
</tr>
<tr>
<td></td>
<td>Morphology of meibomian glands was detected in a patient with marginal staphylolococal keratitis</td>
<td>Koh et al41</td>
</tr>
<tr>
<td></td>
<td>The meiboscore was significantly higher in patients with ocular rosacea</td>
<td>Palamar et al42</td>
</tr>
<tr>
<td></td>
<td>Reduced meibomian gland loss and density were significantly detected in patients with rosacea</td>
<td>Machalinska et al43</td>
</tr>
<tr>
<td></td>
<td>Meibomian gland loss was significantly increased in patients with GVHD</td>
<td>Engel et al44</td>
</tr>
<tr>
<td></td>
<td>The meiboscore was significantly higher in patients with granular corneal dystrophy type 2</td>
<td>Sakimoto45</td>
</tr>
<tr>
<td></td>
<td>The meiboscore was significantly higher in patients who had undergone radiotherapy</td>
<td>Ito et al46</td>
</tr>
</tbody>
</table>

AUC, area under the curve; GVHD, graft-versus-host disease; ROC, receiver-operating characteristic.
Noninvasive meibography allowed us to evaluate various morphological changes in meibomian glands related to ocular surface diseases (Table 2). In normal populations, morphological changes of the meibomian glands increased with age. Contact lens wear was associated with decreased numbers of functional meibomian glands (Fig. 3). Allergic conjunctivitis induced duct distortion of meibomian glands (Fig. 4). In patients with MGD (see Fig. 5 for representative images), the meiboscore was useful for diagnosing MGD. Moreover, the meiboscore was correlated with the function of meibomian glands. The meiboscore was significantly higher in patients with phlyctenular keratitis, rosacea, graft-versus-host disease, and granular corneal dystrophy type 2, and in patients who underwent radiotherapy.

**FUNCTIONAL EVALUATION OF THE MEIBOMIAN GLAND BY TEAR INTERFEROMETRY**

The oily layer, which is located at the superficial surface of the tear film, prevents evaporation of the tear fluid. The distributed oily layer provides an interferometric fringe of the tear film. Tear interferometry can not only visualize the interferometric pattern but also measure the thickness of the oily layer of the tear film based on the interferometric pattern. Currently, Tearscope, Tearscope Plus (Keeler, Windsor, United Kingdom), DR-1/DR-1α (Kowa, Aichi, Japan), and LipiView (TearScience, Morrisville, NC) are commercially available. The reliability of interferometric evaluation was investigated, revealing that MGD was detectable with sensitivity of 65.8%, specificity of 63.4%, and lipid layer thickness (LLT) of 75 nm as a cutoff value based on LipiView observation. However, these values for the diagnosis of dry eye were not consistent. The number of meibum-secreting meibomian glands was significantly related to the LLT, based on observation of 110 patients with dry eye, indicating that a thinner LLT may induce MGD. It was reported that the LLT measured by LipiView was negatively correlated with the remaining meibomian gland area evaluated by meibography, with LLT being significantly larger in control patients than in patients with MGD ($P = 0.028$) in a detailed quantitative analysis. An algorithm to determine LLT based on interferometric fringes obtained by the DR-1 system was also developed. These series obtained with the DR-1 system revealed that the lipid layer condition of the tear film was related to the entire tear film condition and the pattern of blinking. Tear interferometry is currently established as a daily clinical examination to visualize the lipid layer condition of the tear film.

Based on 138 tear interferometric patterns in normal subjects, we found that the interferometric patterns could be classified as follows: pearl-like appearance (monotonous gray interferometric fringe), Jupiter-like appearance (multicolored...
interferometric fringe), and crystal-like appearance (grayish amorphous interferometric fringe) (Fig. 6). We further found that the combination of the interferometric fringe pattern and noninvasive breakup time could indicate the dry eye subtypes.

COMPENSATION THEORY FOR TEAR FILM HOMEOSTASIS

The tear film, lacrimal glands, corneoconjunctival epithelium, and meibomian glands coordinately maintain the homeostasis of the ocular surface. The maintained ocular surface is critical for visual function. Neural connections, systemic hormones, antiinflammatory factors, immunoregulation, osmolarity, tear proteins, microbes, and blinking all affect homeostasis of the ocular surface.

The balance of the tear film components is important for stability of the tear film, and thus a tear film compensatory mechanism regulates the changes in the components of the tear film. We previously reported that tear fluid volume in patients with MGD was positively correlated with the meiboscore revealing damage to the meibomian glands, indicating that increased tear fluid may compensate for decreased function of the lipid layer. Furthermore, we demonstrated that the LLT was increased in subjects with decreased lacrimation, indicating that the increased lipid layer compensated for the decreased function of the aqueous layer (Fig. 7). This compensatory system is changeable,
similar to the tear film conditions, and thus the aqueous layer or lipid layer may be secreted as a reaction to changes in the tear film conditions. Our observations did not evaluate the mucin condition directly. It can be speculated that adaptation of mucin production may require a longer frame.

Our quantitative evaluation of LLT and tear film kinetics by LipiView demonstrated the importance of the tear film compensatory system for maintenance of the tear film balance, and its failure may result in the abnormalities of the tear film observed in patients with dry eye.51 Our results indicated that interferometric fringe patterns and tear film stability related to the balance between the aqueous layer and the lipid layer further indicated the subtypes of dry eye diseases.51

The mechanism for the reaction of the tear film kinetics to changes in the tear film has not been well investigated. The LLT and aqueous volume were both low in patients with combined aqueous-deficient dry eye and MGD. The compensatory system was not functionally active in these patients. Further investigations are required to reveal the detailed mechanism of the tear film compensatory system.

CONCLUSIONS

Morphological evaluation by noninvasive meibography provides much clinical information for the diagnosis of evaporative dry eye. Furthermore, evaluation of the lipid layer of the tear film by interferometry enables monitoring of the function of meibomian glands. Thus, combined evaluation of the morphology and the function of meibomian glands can provide important insights into dry eye pathophysiology.

REFERENCES


