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THE SIGNIFICANCE OF INTERFEROMETRY IN PATIENTS WITH EVAPORATE TYPES OF DRY EYE DISEASE AS A RESULT OF MEIBOMIAN GLAND DYSFUNCTION

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Abstract

Interferometry is a modern method for assessing the thickness of the lipid layer of the tear film, which through its analysis may lead to the diagnosis of the evaporative type of dry eye disease in patients suffering from Meibomian Gland Dysfunction. The aim of this study was to determine the significance of the interferometry and the relationship of interferometric values with clinical findings and dry eye tests in patients with Meibomian Gland Dysfunction.

Material and methods: The study included 50 subjects suffering from Meibomian Gland Dysfunction in whom an examination of the eyelids of both eyes was performed, the clinical stage of the disease was determined, TBUT, Schirmer II test, meniscometry, Oxford score and interferometry was performed.

Results: Statistical analysis of 100 eyes showed a significant correlation of interferometric values with the clinical grade ($r = -0.9574$, $p < 0.001$), with TBUT ($r = 0.7858$ $p < 0.001$) and with Oxford score ($r = -0.7496$, $p < 0.001$), while Schirmer II test and meniscometry showed a weak, statistically insignificant correlation with interferometric values.

Conclusion: The thickness of the lipid layer of the tears has an important role in the diagnosis and grading of evaporative type of dry eye disease in patients suffering from Meibomian Gland Dysfunction, which will mean easier and faster diagnosis, monitoring and treatment of these patients**.**

Keywords: meibomian gland dysfunction, interferometry, lipid layer thickness, TBUT, Schirmer II test, meniscometry, Oxford score.

Introduction

The tear film plays a major role in maintaining ocular surface homeostasis and tear film instability is a major feature of dry eye disease [1].

Meibum, an oily secretion of the meibomian glands, is the primary component of the lipid layer of the tear film. Disruption of meibomian gland function leads to tear film degradation [2].

According to the International Workshop on Meibomian Gland Dysfunction (MGD), MGD is a chronic, diffuse abnormality of the meibomian glands, characterized by terminal duct obstruction and/or qualitative/quantitative changes in gland secretion that can result in tear film alterations, ocular irritation, clinically detectable inflammation, and ocular surface disease, including dry eye [3]. MGD is the leading cause of evaporative dry eye disease [4].

Advances in noninvasive technology in ophthalmology allow visualization of meibomian gland morphology in vivo,[5] as well as assessment of tear film thickness.

Technological advances in noncontact interferometry for determining tear film thickness have enabled quantification of the lipid layer of the tear film and assessment of tear film stability [1].

The normal function of the tear film is crucial for maintaining the health of the ocular surface, which in turn is necessary for maintaining good visual acuity.

The impact of MGD and the subsequent low quality of the lipid layer of the tears leading to disruption of the ocular surface has been described in the literature, and studies indicate that as many as 63% of patients undergoing cataract surgery suffer from MGD. At the same time, patients diagnosed with MGD had worse postoperative visual acuity, and satisfaction in these patients was lower postoperatively compared to subjects without MGD [6].

For the correct diagnosis and classification of the disease of the evaporative type of dry eye, it is necessary to perform numerous dry eye tests and a clinical examination of the eyelids with an accurate assessment of the expressivity of the meibomian glands, which is a complex procedure that also requires a lot of time for the ophthalmologist in his clinical practice.

Nowadays, the literature increasingly discusses new modern tools such as interferometry, which measures the thickness of the lipid layer of the tear film, which could allow us to reach a diagnosis and classification of the disease more simply and quickly [7].

The aim of this study was to determine the significance of interferometric values of the thickness of the lipid layer of the tear film and to find the relationship with the clinical findings and dry eye tests in patients with MGD.

Material and Methods

The study was designed as a cross-sectional cohort study and included 50 subjects suffering from MGD, who presented for examination at the University Clinic for Eye Diseases in Skopje. The study was conducted over a 6-month period, and patients who met the following criteria were included:

1) At least one symptom of dry eye
2) At least one sign of abnormality on the lid margin
3) Poor meibum expressibility

Exclusion criteria were: subjects under 18 years of age, subjects who had undergone ocular surgery less than 3 months ago, patients with dry eye with exclusively aqueous component deficiency, patients who had undergone eyelid surgery, patients with eyelid abnormalities, patients with acute ocular surface infection or ocular allergy, patients with Punctal plugs, patients with nasolacrimal duct stenosis, patients using regular topical ocular therapy (antibiotic and antiglaucoma), patients with autoimmune diseases, patients with Stevens–Johnson syndrome, patients with rosacea and seborrheic dermatitis.

After signing the informed consent, an examination of the eyelids of both eyes was performed, the clinical degree of the disease was determined (from grade 1 to grade 4) and the following dry eye tests were performed: TBUT, Schirmer II test, meniscometry and Oxford score. Then, we approached the new modern method: interferometry (LacryDiag, Quantel medical), a method with which we determined the thickness of the lipid layer of the tears in each eye.

After the interferometry was performed, the thickness of the lipid layer in each eye was analyzed. The values obtained in our subjects (according to the classification of the device itself) were: <15nm, 15nm, 30nm, 30-80nm and 80nm.

Results

The study included 50 subjects, whose both eyes were analyzed, with a diagnosis of meibomian gland dysfunction. This cohort of analyzed eyes included 35% eyes with clinical grade 2 of the disease, followed by 25% eyes with clinical grade 3, 23% eyes with clinical grade 4 and 17% eyes with clinical grade 1.

According to the results of the TBUT test, the tear film breakup time ranged from 1 to 9 seconds, with an average of 4.28 ± 2.1 seconds. In 50% of eyes, this time was shorter than 4 seconds (median = 4 IQR 2.5-6). Significantly worsened condition, with a time shorter than 5 seconds, was measured in 45% of eyes.

The value of the aqueous component of the tear film determined by the Schirmer II test ranged from 6-35 mm, with an average of 18.87 ± 8.7 mm, with a median of less than 16 mm in 50% of eyes. Reduced values of the aqueous component were recorded in 6% of patients.

The average height of the tear film determined by the TMH test was 0.25 ± 0.07 mm, the lowest height was 0.1 mm, the highest 0.53 mm; in 50% of eyes the height of the tear film was less than 0.21 mm (median 0.21 IQR 0.2-0.27), and it was within the reference values in 38% of eyes.

The Oxford score (0-4), had a mean value of 1.87 ± 1.3, in 50% of eyes it was greater than 2 (median 2 IQR 1-3), the most frequently recorded score was 1 (31% of eyes).

Interferometric values of the lipid layer thickness most frequently presented a result of 30 (33% of eyes). These results are shown in Table 1.

Table 1. Characteristics of the analyzed eyes.

variable		n
Clinical grade	1	17 (17%)
	2	35 (35%)
	3	25 (25%)
	4	23 (23%)
TBUT (s)	mean ± SD (min- max) median (IQR)	4.28 ± 2.1 (1 – 9) 4 (2.5 – 6)
	>5	55 (55%)
	5-10	45 (45%)
Schirmer II (mm)	mean ± SD (min- max) median (IQR)	17.87 ± 8.7 (6 – 35) 16(10 – 23)
	>6	94 (94%)
	<6	6 (6%)
TMH (mm)	mean ± SD (min- max) median (IQR)	0.25 ± 0.07 (0.1 – 0.53) 0.21(0.2 – 0.27)
	0.2-0.3 мм	38 (38%)
	<0.2 мм	48 (48%)
	>0.2 мм	14 (14%)
Oxford score	mean ± SD (min- max) median (IQR)	1.87 ± 1.3 (0 – 4) 2(1 – 3)
	0	16 (16%)
	1	31 (31%)
	2	17 (17%)
	3	22 (22%)
	4	14 (14%)
Interferometry	<15	23 (23%)
	15	25 (25%)
	30	33 (33%)
	30 – 80	16 (16%)
	80	3 (3%)

Table 2 shows the distribution of eyes according to clinical disease grade and interferometry. All eyes with tear film thickness <15nm had clinical grade 4, all eyes with tear film thickness 80nm had clinical grade 1, eyes with tear film thickness 15nm most often had clinical grade 3 (88%); eyes with tear film thickness 30nm most often had clinical grade 2 (90.91%), eyes with tear film thickness 30-80nm most often had clinical grade 1 (87.5%).

The differences in clinical disease grade according to interferometry were statistically significant ($p<0.0001$).

Table 2. Clinical grade depending on interferometry/thickness of the lipid layer of the tear film.

Clinical grade	**interferometry/thickness of the lipid layer of the tear film**					
	n	<15 n(%)	15 n(%)	30 n(%)	30 – 80 n(%)	80 n(%)
1	17	0	0	0	14 (87.5)	3 (100)
2	35	0	3 (12)	30 (90.91)	2 (12.5)	0
3	25	0	22 (88)	3 (9.09)	0	0
4	23	23 (100)	0	0	0	0
total	100	23	25	33	16	3

p(Fisher's exact test)

Table 3 shows the distribution of eyes in terms of TBUT and interferometric values of lipid layer thickness. TBUT less than 5s was observed in all eyes with lipid layer thickness <15nm, 80% of eyes with lipid layer thickness 15 nm, 30.3% of eyes with lipid layer thickness 30 nm and 12.5% of eyes with lipid layer thickness 30-80 nm. TBUT from 5s to 10s was observed in all eyes with lipid layer thickness 80 nm, followed by 87.5% of eyes with lipid layer thickness 30-80 nm, 69.7% of eyes with lipid layer thickness 30 nm and 20% of eyes with lipid layer thickness 15 nm.

The differences in the distribution of eyes with TBUT less than 5 and from 5 to 10 in terms of lipid layer thickness were statistically significant (p<0.0001).

Table 3. TBUT depending on interferometry/lipid layer thickness.

TBUT (s)	**lipid layer thickness**						p-level
	n	<15 n(%)	15 n(%)	30 n(%)	30 – 80 n(%)	80 n(%)	
<5	55	23 (100)	20 (80)	10 (30.3)	2 (12.5)	0	p<0.0001
5-10	45	0	5 (20)	23 (69.7)	14 (87.5)	3 (100)	
total	100	23	25	33	16	3	

p(Fisher's exact test)

Table 4. Elaborates the relationship between lipid layer thickness and the Schirmer II test. With the Schirmer II test, less than 6 mm was measured in 8% of eyes with a lipid layer thickness of 15 nm and 12.12% of eyes with a lipid layer thickness of 30 nm.

All eyes with lipid layer thickness <15 nm, 30-80 nm and 80 nm had normal Schirmer test results. The described differences in the distribution of Schirmer test results depending on the lipid layer thickness were statistically insignificant (p=0.351).

Table 4. Schirmer depending on interferometry/lipid layer thickness.

Schirmer II (s)	**lipid layer thickness**						p-level
	n	<15 n(%)	15 n(%)	30 n(%)	30 – 80 n(%)	80 n(%)	
>6	94	23 (100)	23 (92)	29 (87.88)	16 (100)	3 (100)	p=0.351
<6	6	0	2 (8)	4 (12.12)	0	0	
total	100	23	25	33	16	3	

p(Fisher’s exact test)

TMH values lower than the reference values were recorded in 39.13% of eyes with lipid layer thickness <15 nm, in 56% of eyes with lipid layer thickness of 15 nm, in 42.42% of eyes with lipid layer thickness of 30 nm, in 56.25% of eyes with lipid layer thickness of 30-80 nm and in 66.67% of eyes with lipid layer thickness of 80 nm.

In 21.74% of eyes with lipid layer thickness <15, 8% of eyes with lipid layer thickness of 15 nm, 15.15% of eyes with lipid layer thickness of 30 nm and in 12.5% of eyes with lipid layer thickness of 30-80 nm the TMH test values were above the reference values.

The differences in the distribution of TMH test results depending on interferometry were statistically insignificant (p=0.9). These results are shown in Table 5.

Table 5. TMH depending on interferometry/lipid layer thickness.

TMH (s)	**lipid layer thickness**						p-level
	n	<15 n(%)	15 n(%)	30 n(%)	30 – 80 n(%)	80 n(%)	
<0.2	48	9 (39.13)	14 (56)	14 (42.42)	9 (56.25)	2 (66.67)	p=0.9
0.2 – 0.3	38	9 (39.13)	9 (36)	14 (42.42)	5 (31.25)	1 (33.33)	
>3	14	5 (21.74)	2 (8)	5 (15.15)	2 (12.5)	0	
total	100	23	25	33	16	3	

p(Fisher's exact test)

Table 6. presents the relationship between the Oxford score and the thickness of the lipid layer. Oxford score 0 was present in 81.25% of eyes with a lipid layer thickness of 30-80nm and all eyes with a lipid layer thickness of 80nm; score 1 was most often registered in the group of eyes with a lipid layer thickness of 30 nm (60.61%); score 2 was most often present in eyes with a lipid layer thickness of 15 nm (28%); scores 3 and 4 were most often presented in eyes with a lipid layer thickness of <15 nm (39.13% and 43.48%, respectively).

In the group of eyes with lipid layer thickness <15 nm, the majority had an Oxford score of 4, in the group of eyes with lipid layer thickness 15 nm, Oxford scores 1, 2 and 3 were respectively 28% of eyes, in the group of eyes with lipid layer thickness 30 nm the most frequently registered Oxford score was 1 (60.61%), in the groups of eyes with lipid layer thickness 30-80 nm and 80 nm score 1 dominated (81.25% and 100%, respectively).

The differences in the distribution of eyes with Oxford scores 0, 1, 2, 3 and 4 depending on the interferometrically measured tear film thickness were statistically significant.

Table 6. Oxford score depending on interferometry/lipid layer thickness.

oxford score	**lipid layer thickness**						p-level
	n	<15 n(%)	15 n(%)	30 n(%)	30 – 80 n(%)	80 n(%)	
0	16	0	0	0	13 (81.25)	3 (100)	p<0.0001
1	31	2 (8.7)	7 (28)	20 (60.61)	2 (12.5)	0	
2	17	2 (8.7)	7 (28)	8 (24.24)	0	0	

3	22	9 (39.13)	7 (28)	5 (15.15)	1 (6.25)	0	
4	14	10 (43.48)	4 (16)	0	0	0	
total	100	23	25	33	16	3	

p(Fisher's exact test)

Lipid layer thickness negatively correlated significantly with the clinical grade of the disease (r=-0.9574, p<0.0001), and with the Oxford score (r=-0.7496, p<0.0001), and positively correlated significantly with the TBUT test (r=0.7858, p<0.0001).

Lipid layer thickness non-significantly correlated with the Schirmer II test (p= 0.377) and TMH (p=0.62). Lipid layer thickness values decreased with increasing clinical grade of the disease. These results are shown in Table 7.

Table 7. Correlation of interferometry with clinical grade, TBUT, Schirmer II, TMH and Oxford score.

Correlations		
interferometry	Spearman R	p-level
clinical grade	-0.9574	<0.0001
TBUT	0.7858	<0.0001
Schirmer II	-0.0893	0.377
TMH	-0.045	0.62
Oxford score	-0.7496	<0.0001

**sig p<0.01

The statistical processing of the data obtained from the research was done in the statistical program Statistical Package for the Social Sciences programme (SPSS Inc, Chicago, Illinos) version 25.0. Shapiro Wilk's W test was used to test the distribution of the data. Qualitative variables are shown with absolute and relative numbers.

Quantitative variables are shown with average, minimum and maximum values, standard variation, median and interquartile rank. The correlation between interferometry with clinical grade, TMH, TBUT and Scirmer II was analyzed with Spearman's rank correlation coefficient. Differences in the distribution of the results of clinical grade, TMH, TBUT, Schimer test and Oxford score in relation to interferometry were tested with Fisher exact test.

Statistical significance was defined at the level of p<0.05.

Discussion

According to the latest 2025 Tear Film and Ocular Surface Society Dry Eye Workshop (TFOS DEWS III) Diagnostic Methodology Report, Meibomian Gland Dysfunction (MGD) is the most important factor in the development of dry eye disease [8].

The diagnosis of MGD is primarily clinical, through the analysis of changes in the eyelid margin, as well as a combination of dry eye tests that indicate a disorder of glandular secretion.[9] Making the diagnosis and classifying it requires a lot of time from the ophthalmologist, given the numerous tests that he has to perform. Studies are increasingly talking about determining the grade of the disease, so that appropriate medical treatment can be carried out for each patient individually [10].

The eyelid margin examination includes a detailed analysis of any abnormalities of the lid margin such as the appearance of telangiectasias, "plugging" of the gland openings, and irregularity and thickening of the lid margin, [11] and the examination continues with an assessment of the ocular surface epithelium using a slit-lamp bimicroscope, to finally assess the expressivity and quality of the meibum [9] (given that qualitative and quantitative changes in the secretion of the meibomian glands have been identified as the most prominent aspects of MGD) [12].

According to the International Workshop on Meibomian Gland Dysfunction [7], the subjects included in our study were classified into 4 groups (4 grades) after the scores obtained through the lid margin examination, expressivity and quality of the meibum.

This study evaluated the thickness of the lipid layer of the tear film (assessed using interferometry) in relation to the severity of the clinical grade of the disease, TBUT test, Schirmer II test, tear meniscus height and Oxford score in subjects suffering from MGD disease.

Interferometry, which assesses the thickness of the tear film, is a modern imaging tool in ophthalmology that is increasingly being used in clinical practice. Interferometry is an optical technique that, using the surface patterns of the so-called fringe pattern (light stripes), dynamics and reflection, is used to quantify the lipid layer of the precorneal tear film [13,14].

In our study, most of the subjects had clinical grade 2 disease, the mean value of the TBUT test was 4.28 ± 2.1 seconds, the mean value of the Schirmer II test was 18.87 ± 8.7 mm, the mean value of the TMH was 0.25 ± 0.07 mm, the mean value of the Oxford score was 1.87 ± 1.3, and the most common value of the lipid layer thickness was 30 nm.

Statistical analysis in our study showed that the thickness of the lipid layer negatively correlated significantly with the clinical grade of the disease ($r=-0.9574$, $p<0.0001$), which means that the thickness of the lipid layer was lower in patients with a higher clinical grade of MGD disease.

Correlation of the clinical finding with the thickness of the lipid layer was also found by Arita R et al. in their study. In addition to the correlation made, they also explain the importance of measuring the thickness of the lipid layer in the central parts of the cornea, given that the lipid layer spreads from the lower parts of the cornea upwards, and the breakdown of the tear film, in patients with dry eye, is believed to occur first in the central parts.

According to them, studies that use interferometers that have the ability to measure the thickness of the lipid layer only in the lower parts do not show the real picture. That is why it is important to measure the thickness of the lipid layer of the tear film with devices that have the ability to visualize the entire cornea [1], like the device that was used in our study. Lee et al. in their study, analyzing the condition of the glands according to the meibographic finding, divide MGD into obstructive, hyposecretory, normal and hypersecretory. In their study, subjects with hyposecretory and obstructive types were also associated with reduced values of lipid layer thickness [15].

In our study, patients with seborrheic dermatitis and rosacea were excluded by exclusion criteria, so the subjects included in our study belonged to the hyposecretory or obstructive type of gland secretion, which confirms our findings with the aforementioned study.

Similar correlations were also found in the studies of Kim HM et al. and Kim WJ et al. [16,17].
Further statistical analysis in our study showed that lipid layer thickness negatively significantly correlated with the Oxford, and positively significantly correlated with the TBUT test.

Such significant correlations as in our study have been described in the literature, [18,19] indicating that reduced lipid layer thickness leads to instability and faster tear film breakdown, as well as disruption of the ocular surface.

In our study, lipid layer thickness insignificantly correlated with the Schirmer II test and with the height of the tear meniscus.

In the report of the II Tear Film and Dry Eye Society Workshop (TFOS DEWS II) from 2017, in addition to establishing the positions on the classification of dry eye disease, it also addresses the large number of questions arising from the interpretation of the original classification scheme. One of these issues is precisely the distinction between the two primary dry eye conditions and the coexistence of deficiencies in the quantitative and qualitative characteristics of the tear film in a dry eye patient.

They believe that once the patient enters the "Circulus vitiosus", regardless of the initial cause, changes in the tear film follow that cause instability, hyperosmolarity and inflammation, which in turn stimulate further adverse changes, leading to difficulty in detecting the underlying or basic etiology of the dry eye disease and whether it is a lack of the aqueous layer or an evaporative type of dry eye [20].
So according to their explanations, initially there is a compensatory increase in the aqueous component in the tear film, so that in the later stages of the disease and the development of that "Circulus vitiosus", a state follows where the lack of the aqueous and lipid components of the tear film are intertwined.

Their recommendation is to try to remove the perception of exclusivity in the classification itself and that it is better to see the disease as an extension of one condition, rather than two independent conditions. In doing so, the elements of each of these categories should be taken into account in diagnosis and treatment and the disease should be classified according to the predominant component, despite the overlapping signs that exist [21].

The above explains our insignificant correlation of the thickness of the lipid layer with the Schirmer II test and with the height of the tear film [20].

Conclusion

Interferometry, which measures the thickness of the lipid layer of tears, is an important modern tool in ophthalmology that can evaluate the lipid layer of the tear film in vivo, and at the same time its visualization can significantly help us towards an easier and more effective diagnosis of the disease of meibomian gland dysfunction and evaporative dry eye disease.

List of abbreviations:
MGD - Meibomian Gland Dysfunction
TBUT - Tear Break-Up Time
TFOS DEWS -Tear Film and Ocular Surface Society Dry Eye Workshop
TMH - Tear Meniscus Height

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