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ORIGINAL ARTICLE

Assessment of meibomian glands with topography in patients using unilateral antiglaucoma drops

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Abstract

Purpose: The study aims to evaluate the function and morphology of the meibomian glands and tear function tests in patients with unilateral glaucoma.

Methods: The files of 1100 glaucoma patients attending, Ophthalmology clinic from 2014 to 2018 were screened. In total, 38 eyes from 38 out of 84 patients using antiglaucomatous agents in one eye who abided by the criteria and accepted participation were included in the study. After general ophthalmologic examination including best corrected visual acuity, biomicroscopic and ophthalmoscopic examination, ocular surface disease index (OSDI) survey, tear osmolarity, noninvasive tear breakup time (NITBUT), meibography (MEBG) and lower lid tear meniscus height (TMH) measurement, followed by Schirmer test and tear breakup time (TBUT) were measured, respectively.

Results: With mean age of 68.6±12.8 years, 13 patients (34.3%) were female and 25 were male (65.7%). Mean duration of medication use was 37.97 months with mean OSDI score of 33.76±16.2 C4.10–77). The difference between NITBUT and atrophy percentage of meibomian glands in glaucomatous and control eyes was identified to be significant (NITBUT: 9.08±2.98; 12.01±4.30; p=0.001, MEBG 41.15%±14.04%, 28.33%±11.77%, p=0.001). A significant decrease was observed for TMH, TBUT and Schirmer test for eyes administered drops compared to control eyes (p=0.001; p=0.0001; p=0.009, respectively) and tear osmolarity was identified to be significantly high (p=0.0001).

Conclusion: In addition to the negative effects of topical antiglaucomatous drops on tear aqueous components, patients should be monitored for dry eye findings as closely as for intraocular pressure and popularizing the use of preservative-free medications is important in terms of patients' treatment compliance.

Keywords: Dry eye; glaucoma; meibography; preservative; tear breakup time; tear osmolarity.

Ocular surface disorders (OSD) are common findings in glaucoma patients using eye drops usually for a long period of time, making the adverse effects a major concern.[1,2] A variety of studies have shown that 25–59% of glaucoma cases receiving medical treatment have OSD and frequently attend with complaints like burning, irritation, allergy and redness and need to receive additional treatments and as a result of chronic topical medication use,

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the configuration and number of meibomian glands may reduce or morphological changes may be observed.[1–3] These disorders, may be reversible by discontinueing the drop, however irreversible disfunction may also be seen during antiglaucomatous treatment requiring chronic and frequently lifelong use and cause the patient to have permanent dry eye symptoms.[4]

Meibomian gland disease is a common problem where the obstruction of the gland ductus results in ocular surface problems as inflammation, tear film dysfunction, hyperemia and irritation, and with the lipid layer thinning, increase in tear film evaporation and hyperosmolarity.[5–7] Although, it is well established that the active agent as well as the preservative component in the antiglaucoma drugs may cause decrease in goblet cells and tear secretion, the direct and the indirect effects of the drops on the meibomian glands or ductus still remains controversial. Hyperkeratinization in the gland orifices and the stagnation of the sebum in the duct causes detoriation in the ducti as well as atrophy and loss of the gland due to the high pressure within the duct.[8]

Meibomian glands may be imaged with the use of invasive methods like meibography (MEBG), while in recent years, they may be imaged noninvasively with topography device.[1]

MEBG allows screening the morphology and the quantity of the meibomian glands and with the use of Sirius topography, (Sirius Scheimpflug Camera System, Costruzione Strumenti Oftalmici, Florence, Italy) noninvasive evaluation of the meibomian glands by infrared light and obtaining real time pictures to follow the patient by morphology as well as quantitively as area of lost glands. The studies in the literature evaluating the use of Sirius topography in dry eye patients, show the correlation of signs and symptoms with the area of meibomian gland (MG) loss.[9]

In our study, we aimed to compare dry eye symptoms, meibomian gland anatomy, number and morphology and tear function tests between the eye with glaucoma and the contralateral eye in patients receiving unilateral drop treatment for unilateral glaucoma.

Materials and Methods

This study is a comparative prospective study of patients using unilateral antiglaucomatous medication and received permission from Canakkale Onsekiz Mart University Faculty of Medicine Ethics Committee (Project number: 2011-KAEK-27/2017-E.67597). The ethical standards set forth in the Declaration of Helsinki were followed throughout the study. Within the scope of the study, files of 1100 patients monitored in the glaucoma unit between 2014 and 2018 were retrospectively screened. A total of 84 patients were identified to use antiglaucomatous drops unilaterally. Patients were called by telephone and invited to the clinic or patients informed about the study during check-ups in the glaucoma unit were included in the study based on the principle of volunteering. They were informed about tests and were included in the study after completing informed consent forms. Those with active ocular infection, allergy, eyelid deformity, blepharitis, using contact lenses or tear drops, patients with previous ocular and lacrimal surgery, patients with dermatologic diseases, patients using systemic diuretics, isotretinoin, anticholinergic or antipsychiatric drugs were not included in the study. From the total of 84 patients using antiglaucomatous drops in one eye, 38 eyes of 38 patients who abided by the criteria and accepted participation were included in the study. The contralateral eye not administered medication was accepted as control.

All patients had general ophthalmologic examination including best corrected visual acuity and biomicroscopic examination. Then, the patients had the noninvasive methods as a ocular surface disease index (OSDI) survey, tear osmolarity, noninvasive tear breakup time (NITBUT), MEBG and lower lid tear meniscus height (TMH) measurements followed by Schirmer test and tear breakup time (TBUT) measurements. Patients were first asked questions on the OSDI survey which is validated in Turkish, and OSDI score is calculated as reported by Versura et al.,[5] and a value for OSDI score between 0 and 100 was obtained with values above 13 accepted as being in favor of dry eye.[5,10] After the OSDI survey, 50-nL tears is obtained from the lower lid lateral tear meniscus and tear osmolarity was measured with a TearLab device TearLab Osmolarity System, TearLab Corporation, San Diego, CA, USA).[5] TMH was measured with noncontact measurements using an optic coherence tomography device anterior segment module (OCT-AS) (Cirrus HD-OKT, Carl Zeiss Meditec, Dublin, CA, USA) immediately after the patient blinked. The patient looked at a target in primary position, with vertical sections taken in the center of the lower lid and lower half of the cornea while the vertical TMH is measured vertically in micrometers (μm) from the point it touched the cornea (Fig. 1). Before each measurement, patients were asked to blink, and the mean of the three measurements was calculated. TBUT was first measured noninvasively by Sirius topography (Sirius Scheimpflug Camera

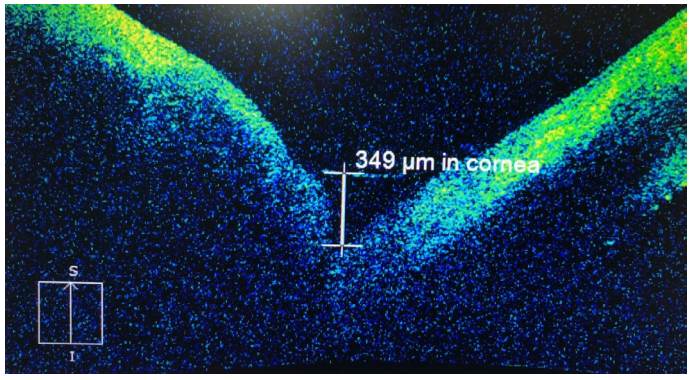

Fig. 1. Tear meniscus height measured by optical coherence tomography.

System, Costruzione Strumenti Oftalmici, Florence, Italy) device and recorded with a videokeratoscope. In this test intersecting lines are reflected on the surface of the eye and the time until the moment the lines break, gave the TBUT. If this value is below 10 s, it is assessed in favor of dry eye disease. The anatomy and configuration of the meibomian glands were assessed with a second measurement with the Sirius topography device. The meibomian glands are imaged with infrared light beaming onto the patient's lower eyelid. The device shows normal meibomian glands, channels and acini as hypoilluminescent grape-like clusters, with channels and underlying tarsus shown hyperilluminescent (Fig. 2). After taking the image, the whole tarsal area is marked with the program in the Sirius device and regions with meibomian glands are marked again. The proportion of the two areas to each other is recorded as a percentage as the meibomian gland atrophy area (Fig. 3). After assessment, the meibomian gland atrophy area proportion is classified from 0 to 3 as outlined by Arita et al.[11]

When measurement with noninvasive methods are completed, conventional tear function tests like Schirmer's test and TBUT measurements are followed. Reflex secretions were assessed with Schirmer I test. Filter paper with 5×35 cm size was placed in the intersection line of 1/3 center and 1/3 temporal of the lower lid conjunctival fornix. 5 min later the amount of wetting was measured. Results below 10 mm were assessed in favor of dry eye. TBUT was measured with the aid of fluorescein. After the patient blinked once, the cornea was observed with a biomicroscope under cobalt blue light. The time between the last blink and the first black point forming on the cornea which was stained yellow is recorded as TBUT. Duration below 10 s, is assessed in favor of dry eye.

Data obtained in this research was entered into the IBM-SPSS 20.0 statistical program and the Kolmogorov-Smirnov normal distribution test was applied to decide which data were suitable for parametric or nonparametric tests used for comparison of data. The results of this analysis determined some values had normal distribution ($p>0.05$), while some did not have normal distribution ($p<0.05$), so both parametric and nonparametric methods were used for comparison tests. Correlation analysis was completed with the Spearman ranked difference coefficient. Comparison analyses belonging to variables and comparison between patient and healthy eyes used the t-test, analysis of variance, Mann-Whitney U test and Kruskal Wallis test according to distribution of data. Descriptive statistics for the variables are given with arithmetic mean and standard de-

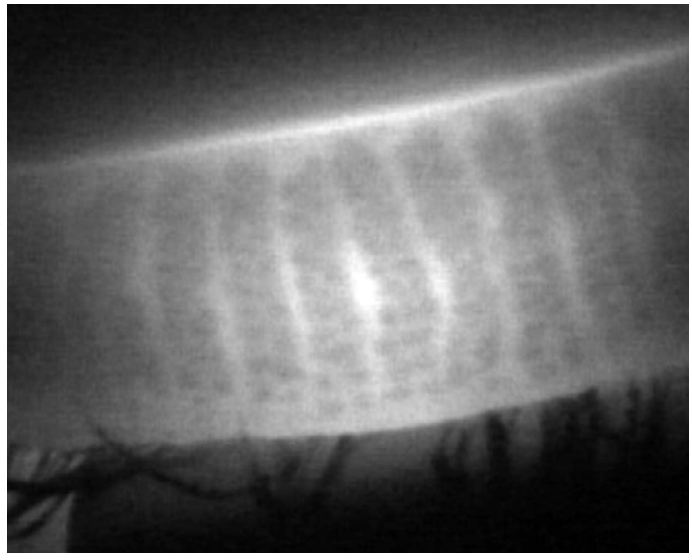

Fig. 2. Normal meibomian gland imaging by topography using infrared light.

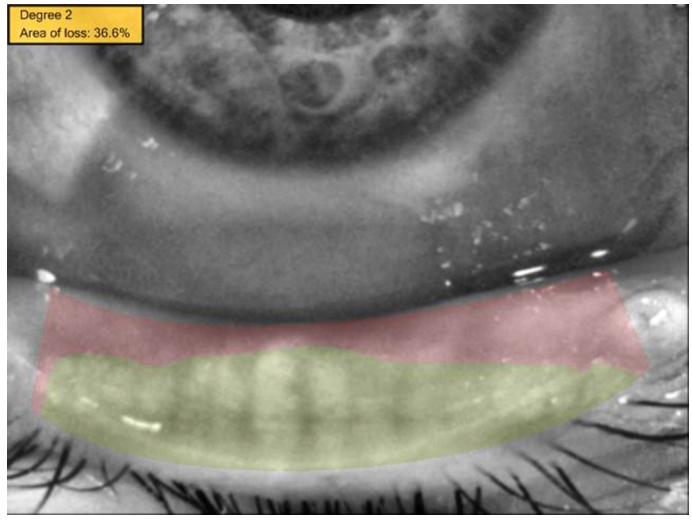

Fig. 3. The regions of the visible meibomian glands are marked with the program included in the software of the Sirius device and the proportion of this area to whole tarsal plate was recorded as percentage.

viation (±) of the variable set shown. A p≤0.05 is accepted to be significant.

Results

Our study included a total of 38 patients with unilateral glaucoma diagnosis, with 13 women (34.3%) and 25 men (65.7%) using drops in one eye and assessed a total of 76 eyes in 38 patients. The mean age of patients was 68.6±12.8 years, with mean age of female patients 66.1±1.6 years and mean age of male patients 69.2±5.0 years. Mean duration of medication use was 37.97 months (3–38 months) with 17 patients (44.7%) using single medication, 13 (34.2%) using two medications and 8 (21.1%) using three or more medications. The most frequently used active agents were timolol maleate, brimonidine tartarate, prostaglandin analogs, and carbonic anhydrase inhibitors, respectively. When medications were assessed according to preservative content, 41 (61.6%) drops contained benzalkonium chloride (BAC), 20 (29.8%) contained stabilized oxychloro complex (Purite) and 7 (9.1%) included polyquaternium-1 (Polyquad). Of patients, 21.1% (8 patients) used 1 drop per day, 23.7% (9 patients) used 2 drops per day and 55.6% (21 patients) used 3 or more drops per day.

Mean OSDI survey score were identified as 33.76±16.2 (range 4.1–77). Of patients, 5 had values <13, while 86.7% had OSDI score of 13 or above.

The results of mean tear osmolarity, mean TMH, mean NITBUT, mean MG atrophy area, Schirmer test and TBUT measurement results for the control group and study group are shown in Table 1. Significant differences were found between the results of two groups for tear osmolarity measurement (p=0.0001); TMH, (p=0.001); NITBUT (p=0.001); MG atrophy area percentage (p=0.0001); Schirmer test (p=0.009) and TBUT (p=0.0001).

Table 1. Comparison of tear function tests between glaucomatous and control eyes

	Glaucomatous eye	Control eye	p-value
NITBUT (s)	9.08±2.98	12.01±4.30	0.001
TBUT (s)	7.03±2.51	10.45±3.36	0.0001
Schirmer test (mm)	7.8±4.5	11.53±7.3	0.009
Tear osmolarity (mOsml)	305.08±11.5	294.89±9.11	0.0001
TMH (micron)	166±39.26	206.61±61.03	0.001
Atrophy area measured with MEBG (%)	41.15±14.04	28.33±11.77	0.0001

NITBUT: Noninvasive tear breakup time; TBUT: Tear breakup time; TMH: Tear meniscus height; MEBG: Meibography.

The MG atrophy percentage was staged from 0 to 3 for all eyes. In the glaucomatous eye group, stage 0 (no atrophic area) was not observed, with 19.4% observed to be in Stage 1, 81.6% observed to have Stage 2 and 3 atrophy. In the control eye group, the rate for stage 0 and stage 1 atrophy was 42%. The correlation between daily drop numbers and MG atrophy percentage measured with MEBG was examined. The atrophy area for patients using a single antiglaucomatous drop per day (8 patients) was 32.98%, while it was 39.49% for patients using two drops (9 patients) and 44.98% for patients using 3 or more drops per day (21 patients). As the number of drops increased, it appeared the gland atrophy area increased. The correlation between patient age with MG atrophy area in study and control eyes was evaluated, and for both eyes atrophy areas increased as age increased. A moderate positive significant correlation was identified between age and MEBG atrophy percentage (rho: 0.522, p<0.01; rho: 0.395, p<0.05, respectively).

Patients were divided into three groups according to duration of medication use. Group 1 included patients using drops for 12 months or less, Group 2 included those using drops for 13–24 months and Group 3 included those using drops for 25 months or longer. The mean percentage of MG atrophy areas according to groups are shown in Table 2. As the duration of drop use increased, an increase was observed in MG atrophy. A moderate positive correlation was identified between atrophy area identified with MEBG and drop use duration (rho: 0.348, p<0.05). Similarly, a moderate positive correlation was identified between drop use duration and atrophy stage identified with MEBG (rho: 0.323, p<0.05). When antiglaucomatous drops are evaluated according to their preservative contents as BAC-contained and preservatives other than BAC; the group using drops including only BAC (33 patients) had meibomian

Table 2. Mean of tear function test measurements according to duration of antiglaucomatous drop use

Drop use duration	≤12 months	13–24 months	≥25 months
OSDI	30.3±2.6	43.3±8.2	32.3±2.7
MEBG atrophy area (%)	31.3±7.95	35.25±6.24	44.7±2.18
TMH (micron)	157.3±26.8	184.3±18	163.7±5.97
NITBUT (s)	10.9±1.01	10.3±1.14	8.3±0.57
Tear osmolarity (mOsm/L)	306.8±2.3	302.5±6	305.3±2.35
Schirmer test (mm)	6.17±2.3	11.5±2.1	7.31±0.082
TBUT (s)	6.17±1.22	7.83±1.32	7.04±0.44

NITBUT: Noninvasive tear breakup time; TBUT: Tear breakup time; OSDI: Ocular surface diseases index; TMH: Tear meniscus height; MEBG: Meibography.

gland atrophy area of 41.44%, while this was identified as 39.28% for patients using medications containing preservatives other than BAC (polyquaternium-1 and/or stabilized oxychloro complex). BAC caused more atrophy but the difference was not significant.

When patients using a single drop containing BAC were compared with patients using 2 different drops containing BAC, the patient group with more exposure to BAC had higher MG atrophy percentage on MEBG, though there was no significant difference observed between the two groups ($p>0.05$). When the mean tear osmolarity is compared in the same groups, patients using 2 BAC medications were identified to have higher tear osmolarity with significant difference ($p=0.014$).

Discussion

Topical antiglaucomatous medications are the primary approach chosen for glaucoma treatment, with most patients using single or multiple antiglaucomatous drops until surgery, or if surgery is not required, for lifelong.[12] All topically used medications cause reactions on the ocular surface like punctate keratopathy, reduced sensitivity of the cornea or reduced tear synthesis.[1,11–13] In the literature, there are many studies about the main side effects of antiglaucomatous medications being due to preservative content.[14–18] Preservatives act as a surfactant to solubilize ionic components into otherwise immiscible solvents and are required for the active agent to pass the cornea, in addition to being required to protect the medication contained in multiple dose bottles, from microorganisms.[15,16] A study by Fechtner et al., with 630 glaucoma patients, identified that dry eye disease and glaucoma were common in the elderly and frequently were comorbid diseases.[2] Another study found dry eye disease was present in 8.4% of patients from 48 to 59 years, and in 19% of patients over the age of 80 and had age-linked prevalence.[19] A 2015 study by Saade et al., reported that as the number of antiglaucomatous medications and duration of use increased, the OSDI value increased.[20] In our study, similarly, the OSDI score was 33.76±16.2, and was assessed as being high. Ramli et al., showed the prevalence of ocular surface diseases varied from 37% to 91% in the glaucoma group regardless of preservative content of the eye drops.[21] The meibomian glands responsible for formation of the lipid components of the tear film layer are localized in the tarsal plate of the upper and lower eyelids.[22] Arita et al.,[1] showed that long-duration use of topical antiglaucomatous agents was associated with changes to the morphology and functions of the meibomian gland. Mathers et al.[23] and Jester et al.,[24] imaged the meibomian glands with the aid of infrared light in 1982 and 1994 and showed that on MEBG "normal" meibomian glands were grape-like clusters providing hypoillumination, with the ductus and underlying tarsus observed as hyperilluminant areas. In our study, the meibomian gland atrophy area assessed and staged with MEBG as described by Arita et al.,[11] was observed to be higher with a significant difference in eyes with drops administered compared to control eyes. Similarly, Portela et al.[25] and Arita et al.[26] reported significantly higher atrophy rates of meibomian glands in glaucoma patients compared to healthy volunteers as measured by MEBG.

Tear osmolarity is accepted as the gold standard for dry eye diagnosis and in 2009 the TearLab tear osmolarity measurement device (TearLab Corporation, San Diego, CA, USA) was permitted by the FDA (Food and Drug Administration). It provides convenience in diagnosis and monitoring of dry eye syndrome due to requiring a very small amount of tears, being noninvasive and numerically showing osmolarity values in a short duration like 5 s.[5,27] In a variety of studies, the threshold value for tear osmolarity in dry eyes was determined to be between 305 and 317 mOm/L.[5,28,29] Our mean tear osmolarity was 305.08±11.5 mOsm/L in the eye administered medication and 294.89±9.11 mOsm/L in the control eye. Prospective studies by Lee et al., examined tear osmolarity in 51 eyes using antiglaucomatous medication and 49 eyes in the control group. Similar to our study, they found the mean tear osmolarity in the group using medication was 307±9.3 mOsm/L and in the control group it was 301.4±7.7 mOsm/L.[30] Preservatives have a destructive effect on tear lipids causing increased evaporation of tears and elevation of tear osmolarity.[14,15] A study of 30 glaucoma patients identified tear osmolarity as 315 mOsm/L during latanoprost treatment including BAC, while 2 weeks after transition to tafluprost which does not contain preservatives, it was 308 mOsm/L and 12 weeks later it had decreased to 302 mOsm/L.[14] In our study, we identified that as the number of drops containing BAC increased, there was a significant increase in tear osmolarity. The mean Schirmer test in eyes included in our study was 7.8±4.5 mm in the glaucomatous eye and 11.53±7.3 mm in the control group, with a significant difference observed between both eyes. Another similar study identified that all 100 patients using antiglaucomatous agents had abnormal Schirmer test, with 62% of patients having mean measurement of 5 mm or less.[31] In our study, the mean TBUT in the control group was 10.45±3.6 s, while it was identified as 7.03±2.51 s in the eye using drops, with a significant difference between the

groups. Ramli et al., compared glaucoma patients using topical antiglaucomatous medications (n=105) with control group not using any topical medication (n=102) and observed abnormal TBUT in the patient group. The same study reported reduced TBUT was observed in patients using drops containing BAC compared to patients using preservative-free antiglaucomatous agents.[21] The mean TMH measured with the OCT-AS was 206.61±61.03 µm in control eyes and 166±39.26 µm in patient eyes. Mathers et al., compared 30 glaucoma patients with 27 cataract patients and observed that TMH in the glaucoma group was significantly low.[23] Preservatives in eye drops are used to prevent microbial growth within the bottle ensuring safe use of multiple dose containers, preventing biologic degradation and support preservation of the medication effect.[15] BAC is the most commonly-used preservative in topical glaucoma medications which adheres to cell membranes increasing membrane permeability and causing cell lysis, reduces stability of tear film on the ocular surface causing inflammatory cell infiltration and induces conjunctival and cornea epithelial cell abnormalities.[14,16,17] A study on New Zealand rabbits examined the negative effects on the ocular surface for drops containing high levels of BAC (0.03%) and containing Purite or lower levels of BAC (0.005%). It is reported that as the concentration of BAC increased, conjunctival cell infiltration and corneal damage increased.[17] Suzuki et al.[32] showed that lowering BAC concentration in tafluprost ophthalmic solution from 0.005–0.01% to 0.001–0.003% resulted in less corneal epithelial cell cytotoxicity as in preservative-free tafluprost. Gandolfi et al.[33] compared 2 separate patient groups of 371 glaucoma patients using BAC as preservative or travoprost containing polyquad and identified the side effects were mild and similar in both groups. In more recent times, Jayanthi et al., compared BAC containing travoprost with SofZia containing travoprost and reported significantly lower OSDI scores in patients using travoprost with SofZia preservative.[34] In our study, all drops containing BAC, polyquad and Purite caused atrophy of the meibomian glands; however, patients using more than one drop containing BAC were observed to have significantly more atrophy compared to patients using a single drop containing BAC.

Strong aspects of our study are that it was performed on both eyes of the same patient as eye using antiglaucomatous drop as study group and not using eye as control. This prevents the confounding effect of any systemic or environmental factors that may affect the meibomian glands such as age, gender, hormonal status, emotional stress, environmental temperature, humidity, body dehydration with low water intake, exposure to sun or visual screens as those would affect both eyes. Besides the prospective design and assessment of tear functions with objective and noninvasive methods along with conventional tests, photographically documentation of meibomian gland anatomy and configuration with topography for each patient and monitorization of patients for long durations are the other strong aspects.

Limitations of our study are that 46 of 84 patients using unilateral drops included in the scope of the study had begun treatment for dry eye findings at other clinics, who did not abide by study protocols or were lost follow-up are removed from the study and it was completed with lower numbers of patients than planned. For the same reason, our patient population had a distribution in favor of men in terms of gender distribution. Due to the low patient number and use of multiple combinations of the drugs by patients, it was not possible to compare subgroups according to active ingredient.

Conclusion

Topical antiglaucomatous drops, which require chronic use with multiple posology, cause dry eye complaints or increase the severity of dry eye disease. In addition to the negative effects on tear aqueous components of both the active ingredient and preservative in topical drops, it disrupts lipid secretions due to atrophy of the acini with chronic damage to the meibomian glands in the tarsus and may negatively affect compliance with treatment and follow-up among glaucoma patients.

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Ethics Committee Approval: This study was approved by Canakkale Onsekiz Mart University Faculty of Medicine Ethics Committee (date: 27.10.2017; number 18920478-050.01.04/E.127609).

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Authorship Contributions: Concept: A.A., A.T.Ç.; Design: A.A., A.T.Ç.; Supervision: A.T.Ç.; Resource: A.A., A.T.Ç.; Materials: A.A., A.T.Ç.; Data Collection and/or Processing: A.A., A.T.Ç.; Analysis and/or Interpretation: A.A., A.T.Ç.; Literature Search: A.A., A.T.Ç.; Writing: A.A., A.T.Ç.; Critical Reviews: A.T.Ç.

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