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Original Article

Evaluation of Corneal Alterations After Short-Term Silicone Hydrogel Contact Lens Use by Confocal Microscopy

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Abstract

Objectives: To evaluate the corneal subbasal nerve morphology, corneal sensitivity, and anterior segment alterations in short-term silicone hydrogel contact lens (SiHCL) users by confocal microscopy.

Materials and Methods: The study included 25 right eyes of 25 male volunteers aged 25-30 years who had never used SiHCLs before. ocular surface disease index (OSDI), tear break-up time, Schirmer test, tear meniscus area, strip meniscometry tube, corneal sensitivity, and corneal subbasal nerve morphology were evaluated before and after 1 month of CL use.

Results: OSDI was 10.6±1.1 before CL use and 17.2±1.2 after 1 month of CL use ($p<0.01$). Schirmer test distance was 16.3±2.3 mm before and 14.3±1.9 mm after 1 month of CL use ($p>0.05$). Tear film break-up time was 7.1±0.4 s before and 6.2±0.3 s after CL use ($p>0.05$). The tear meniscus area was 0.026±0.002 mm^2 before and 0.024±0.001 mm^2 after 1 month of CL use ($p>0.05$). Strip meniscometry tube results were 5.4±0.9 mm before and 4.9±0.8 mm after 1 month of CL use ($p>0.05$). Corneal sensitivity values were 3.2±0.4 mm before and 2.95±0.3 mm after 1 month of CL use ($p>0.05$). Dendritic cell density evaluated by confocal microscopy was 14.84±3.1 cells/mm^2 before and 32.57±4.2 cells/mm^2 after 1 month of CL use ($p<0.01$). Subbasal nerve tortuosity was 0.92±0.2 before and 1.03±0.2 after 1 month of CL use ($p>0.05$). Subbasal nerve density was measured as 4726±310 pixels/frame before and 4570±272 pixels/frame after 1 month of CL use ($p>0.05$).

Conclusion: After a month of SiHCL use, no significant changes were observed in tear secretion, corneal sensitivity, tear meniscus volume, subbasal corneal nerve density, reflectivity, or tortuosity, while a significant increase was found in OSDI and dendritic cell density.

Keywords: Confocal microscope, contact lens, corneal sensitivity, subbasal cell density, ocular surface disease index

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Introduction

With the rising myopia rates and improved comfort resulting from advances in contact lens (CL) technology, silicone hydrogel contact lenses (SiHCLs) are now widely used to eliminate refractive errors.[1] Although the use of CLs is advantageous for users in social terms, it is known to be associated with various complications, including ocular infections such as keratitis, which can cause permanent visual impairment.[2] Their frequent use in routine clinical practice has given rise to the need for further research on the effects of SiHCL use on the ocular surface, and the use of *in vivo* confocal microscopy (IVCM) in this context has steadily increased.

IVCM is a non-invasive imaging method that allows *in vivo* visualization of the ocular surface structure and is used both in the imaging of healthy corneas and in the diagnosis and follow-up of many corneal diseases.[3] Dendritic cells are potent antigen-presenting cells that are considered an immune activation marker and have an important role in the regulation of the immune response.[4,5] IVCM also enables in vivo imaging of corneal dendritic cells, which are frequently seen throughout the subbasal nerve plexus layer.[6,7] Previous studies using IVCM have demonstrated changes in dendritic cell density in the corneal morphology in conditions that cause ocular surface pathology, such as dry eye, allergy, keratitis, keratoconus, and corneal dystrophies.[8,9] Anterior segment inflammation results in activation of Langerhans cells in the cornea and conjunctiva as part of the immune response.[10,11] In addition, dry eye-related symptoms can cause many patients to shorten the duration of soft CL use or completely stop using CLs altogether.[12] Previous studies reported reduced corneal sensitivity associated with CL use.[13,14] However, corneal sensitivity was shown to return to normal after discontinuing CL use.[15,16] Traditional CLs with low oxygen permeability (Dk) create a hypoxic environment in the cornea, resulting in decreased sensory nerve function.[17] Other factors contributing to this loss of sensitivity include sensory adaptation and acidosis-suppressed sensory nerve function.[18,19] Most published studies were conducted with earlier generation CLs made of low-Dk materials. With new-generation SiHCLs, lens-induced hypoxia is considerably reduced or even eliminated. Changes in ocular surface sensitivity that may occur in this low hypoxic environment after short- and long-term SiHCL use are not fully known.[20]

The use of SiHCLs is especially common in young adults, and investigating ocular surface changes in SiHCL users will provide guidance in solving the complications and problems caused by SiHCLs. The aim of this study was to evaluate dry eye findings, corneal subbasal nerve morphology, corneal sensitivity, and anterior segment changes after the short-term (1 month) use of SiHCLs.

Materials and Methods

Participants and Contact Lenses

Approval for this prospective study was obtained from the Medical Research Ethics Committee of Muğla Sıtkı Koçman University Faculty of Medicine on March 31, 2021 (decision number 7/XII). The study adhered to the principles of the Declaration of Helsinki. All participants signed an informed consent form after being informed in detail about the nature and purpose of the study.

Before the study, all participants underwent a complete ophthalmological examination including refraction, visual acuity examination with Snellen chart, intraocular pressure measurement with applanation tonometry, biomicroscopy, and indirect ophthalmoscopy. Twenty-five right eyes of 25 patients between the ages of 18 and 30 years who presented to the ophthalmology outpatient clinic of Muğla Sıtkı Koçman University Medical Faculty to obtain CL and had no previous history of CL use were included in the study. Only men were included in the study to avoid hormonal factors such as pregnancy and menstrual cycle that may affect the measurements. For standardization, all participants used lotrafilcon B (Air Optix, Alcon Laboratories, Fort Worth, USA) silicone hydrogel monthly CLs (lens DK/t ratio 138 @ -3.00 diopters [D]). Eyes with spherical values between -1 and -6 D and no astigmatism were included in the study. Users were asked to wear the CLs for at least 8 hours a day and no fewer than 5 days a week. Exclusion criteria of the study were determined as severe dry eye signs or symptoms, history of systemic disease, previous ocular surface surgery, systemic or ocular surface disease that may affect the cornea, and history of medication use. The participants' ocular surface disease index (OSDI), tear break-up time, Schirmer test, tear meniscus area, strip meniscometry tube (SMTube), corneal sensitivity, and IVCM were evaluated before and after using the SiHCLs for 1 month.

Ocular Surface Disease Index

The OSDI is a 12-item questionnaire that assesses dry eye-induced ocular irritation symptoms and associated visual functions. All participants were asked these questions in face-to-face interviews before and after 1 month of SiHCL use, and their answers were recorded in the system. Each item in the OSDI questionnaire is scored between 0 and 4. As in the original questionnaire, OSDI scores were calculated by summing the points from all 12 items, multiplying by 25, and dividing by the number of items answered to transform the results into a 0-100 scoring system. OSDI scores of 0-12 were regarded as normal, 13-22 as mild, 23-32 as moderate, and 33-100 as severe ocular surface disease.[21]

Schirmer Test, Tear Break-Up Time, Tear Meniscus Area, and SM Tube

The Schirmer test was performed by placing a sterile Schirmer paper (Tearflo; Alcon Laboratories, Inc., Fort Worth, TX, USA) in the outer third of the lower eyelid, waiting for 5 minutes, and recording the amount of strip wetting. Evaluation of tear break-up time was performed by touching a sterile fluorescein strip to the conjunctiva, asking the patient to blink once and then keep their eyes open, and recording the time (in seconds) when the first dark spot appeared under the biomicroscope with cobalt blue light. Tear meniscus area was measured using anterior-segment optical coherence tomography (RTVue, Optovue Inc, USA) by delineating its borders and calculating the area in mm^2 (Figure 1a,b).[22] SMTube value is evaluated using meniscometry strips that measure the amount of tears accumulated in the tear meniscus. The strip was placed in the tear meniscus of the bottom eyelid without touching the ocular surface and the amount of wetting was measured in millimeters after 5 minutes using the scale on the strip.[22]

Corneal Sensitivity

To evaluate corneal sensitivity, we utilized a modified Cochet-Bonnet esthesiometer with 0.3 nylon filaments 0.09 mm in diameter (Unitika Ltd. Tokyo, Japan), as used previously in another study.[23] Seven filaments ranging in length from 0.5 cm to 4 cm were prepared. Starting from the longest (4 cm, lowest pressure), each length of nylon filament was used three times to stimulate the corneal nerves and elicit a corneal blinking response. The central cornea was touched with the filament held perpendicularly and avoiding the eyelashes and lid margins. The same procedure was repeated, reducing the length of the nylon filament by 0.5 cm each time, until a complete blink reflex was elicited. Three measurements were recorded for each eye and the average was used for analysis. The repeatability of this method for obtaining corneal sensitivity measurements was confirmed before performing any tests.

In Vivo Confocal Microscopy and Image Analysis

After all other assessments, topical anesthesia was provided with 0.5% proparacaine hydrochloride drops (Alcaine®, Alcon, Fort Worth, Texas, USA) and IVCM was performed with an HRT III (Heidelberg Engineering GmbH, Heidelberg, Germany) device using the Rostock cornea module. The internally mounted laser source in the HRT generates a 670-nm red diode laser. The high-resolution real-time images obtained in IVCM had a resolution of 1 µm/pixel and contained 384x384 pixels covering an area of 400x400 µm (horizontal x vertical). The images were recorded in a JPEG format with 8-bit data resolution and 128-bit binary floating-point format. Six to eight complete sequences were recorded, each containing 100 images from each cornea (each frame representing an area of 160.00 μm^2). For corneal morphological analysis, three non-overlapping representative images of each cornea were selected. Before IVCM was performed, a drop of Viscotears gel (Alcon Laboratories, Inc., Texas, USA) was placed in and on a TomoCap (Heidelberg Engineering) placed on the objective lens of the microscope. The tip of the TomoCap was brought in contact with the patient's cornea and the patient was asked to look at a fixed point while the scan was performed. Averages of the three best images obtained from the central cornea were obtained. Nerve number and density were measured using the NeuronJ add-on (National Institutes of Health, Bethesda, MD, USA) to the ImageJ software, which allows semi-automatic viewing of nerve fibers and enables their quantification. Distinguished by their bright cell bodies, dendritic cells in the subbasal nerve plexus were counted manually using the ImageJ software.[24] The degree of nerve tortuosity and reflectivity was assessed according to the scale (0-4) described by Oliveira-Soto and Efron.[25]

Figure 1. Representative image of tear meniscus area measurement using anterior-segment optical coherence tomography (OCT). Cross-sectional anterior-segment OCT image of tear meniscus before (a) and after (b) silicone hydrogel contact lens use. Patients with dry eye have significantly lower tear meniscus area than normal people. The black bar shows 250 µm

Statistical Analysis

Statistical analyses were performed using SPSS version 22.0 (IBM Corp., Armonk, NY, USA). Descriptive statistics were used to summarize the data. Normality of data distributions was tested all parameters using Shapiro-Wilk test. Normally distributed data were analyzed using one-way analysis of variance (ANOVA). If the result of this test was significant, post-hoc Tukey's test was used for pairwise comparisons. Results were evaluated within 95% confidence intervals and a p value less than 5% was considered statistically significant.

Results

In this study, the OSDI score was found to be 10.6±1.1 before and 17.2±1.2 after 1 month of SiHCL use ($p<0.01$) (Figure 2). Schirmer test results at these two time points were 16.3±2.3 mm and 14.3±1.9 mm ($p>0.05$) (Figure 3a) and tear film break-up times were 7.1±0.4 s and 6.2±0.3 s, respectively ($p>0.05$) (Figure 3b). Tear meniscus area was 0.026±0.002 mm^2 before and 0.024±0.001 mm^2 after 1 month of SiHCL use ($p>0.05$) (Figure 4a), while SMTube values were 5.4±0.9 mm and 4.9±0.8 mm, respectively ($p>0.05$) (Figure 4b). Corneal sensitivity test values before and after 1 month of SiHCL use were 3.2±0.4 mm and 2.95±0.3 mm, respectively ($p>0.05$) (Figure 4c). On IVCM, dendritic cell density was determined to be 14.84±3.1 cells/mm^2 before and 32.57±4.2 cells/mm^2 after 1 month of SiHCL use ($p<0.01$) (Figure 5a). Subbasal nerve tortuosity was evaluated as stage 0.92±0.2 before and stage 1.03±0.2 after 1 month of SiHCL use (Figure 5b). Subbasal nerve density at the two time points was 4,726±310 pixels/frame and 4,570±272 pixels/frame ($p>0.05$) (Figure 5c) and subbasal nerve reflectivity was grade 1.73±0.3 and grade 1.66±0.2, respectively ($p>0.05$) (Figure 5d). A representative IVCM image of dendritic cells is shown in

Figure 6. In brief, there were no statistically significant changes in tear secretion, corneal sensitivity, or tear meniscus volume after 1 month of SiHCL use compared to before SiHCL use in our study, whereas there was a statistically significant increase in OSDI. IVCM revealed no change in subbasal corneal nerve density, reflectivity, or tortuosity, while the increase in dendritic cell activation was found to be statistically significant.

Discussion

IVCM is a new, noninvasive imaging method that allows examination of the cornea at the cellular level and is frequently used both in healthy corneas and in the differential diagnosis and follow-up of many diseases. IVCM can provide high-resolution images of the corneal subbasal nerves and immune/inflammatory (dendritic) cells.[26] With the increasing clinical use of IVCM, many studies evaluating the subbasal nerves in the healthy and pathological cornea have been published.

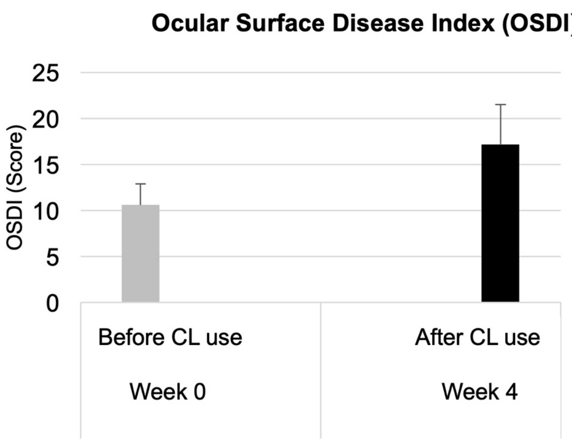

Figure 2. Ocular surface diseases index (OSDI) assessment. After short-term (1 month) use of silicone hydrogel contact lens, there was a statistically significant increase in OSDI compared to before use (*p<0.01)

In this study, we compared parameters such as OSDI score, tear break-up time, tear meniscus volume, Schirmer test results, corneal sensitivity, and corneal subbasal nerve morphology assessed before and after short-term (1 month) SiHCL use. Considering the previous literature, there is no study evaluating all of these parameters together in relation to short-term SiHCL use, thus the results of our study may be important. In our assessment after 1 month of SiHCL use, only the increases in dendritic cell activation and OSDI score were found to be statistically significant compared to before use. We observed no statistically significant changes in Schirmer's test, tear break-up time, corneal sensitivity, or corneal subbasal nerve density. Previous research on similar subjects includes studies demonstrating higher dendritic cell density in the central cornea in people using CLs. Zhivov et al.[27] observed a higher number of Langerhans cells in the corneal epithelium of CL users compared to non-users. These cells are thought to migrate centrally as a CL-induced inflammatory response.[28,29] This result is consistent with previous animal model studies demonstrating migration of Langerhans cells into the cornea in response to the mechanical effect of CL.[30,31] In their study, Zhang et al.[32] compared SiHCLs with low and high Dk value in a rat model and reported that the number of conjunctival dendritic cells increased more with the use of the low-Dk SiHCL. Similarly, two different studies led by Alzahrani et al.[33,34] showed that dendritic cell density was increased in people using soft CLs. These results may be an indicator of the inflammatory response in the cornea induced by the CL. Sindt et al.[35] proposed in their study that this dendritic cell activation could be due to CL-related ocular discomfort. Although we detected no statistically significant change in subbasal nerve density after short-term SiHCL use in this study, Liu et al.[36] reported lower nerve fiber density in CL users compared to non-users. However, they showed that there was no significant difference between CL users with and without dry eye. There are also studies in the literature showing increased dendritic cell activation in dry eye disease.[37] A report by Liu et al.[38] indicated that dendritic cell density gradually

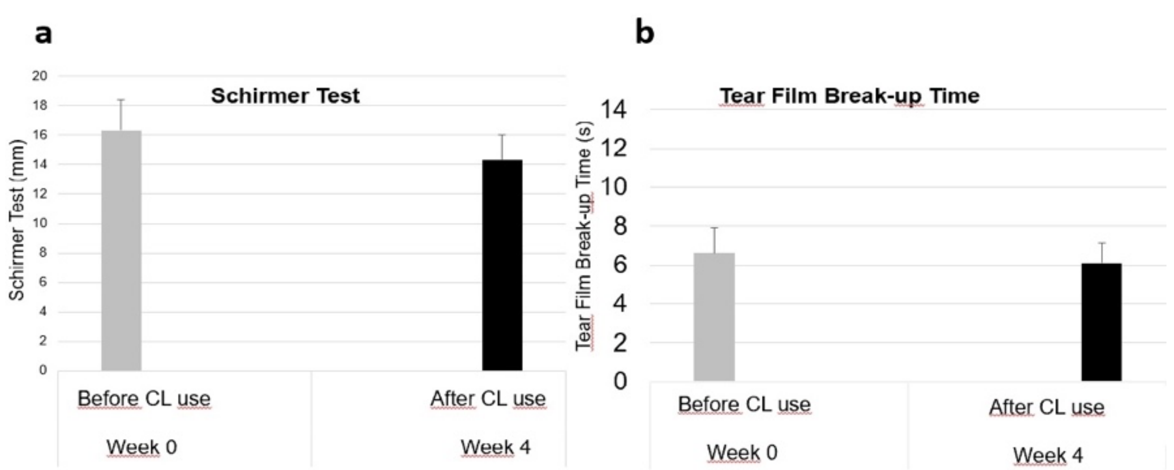

Figure 3. Comparison of Schirmer test and tear break-up time before and after silicone hydrogel contact lens (CL) use. Schirmer test (a) and tear break-up time (b) showed no statistically significant change after short-term use of a silicone hydrogel CL (p>0.05)

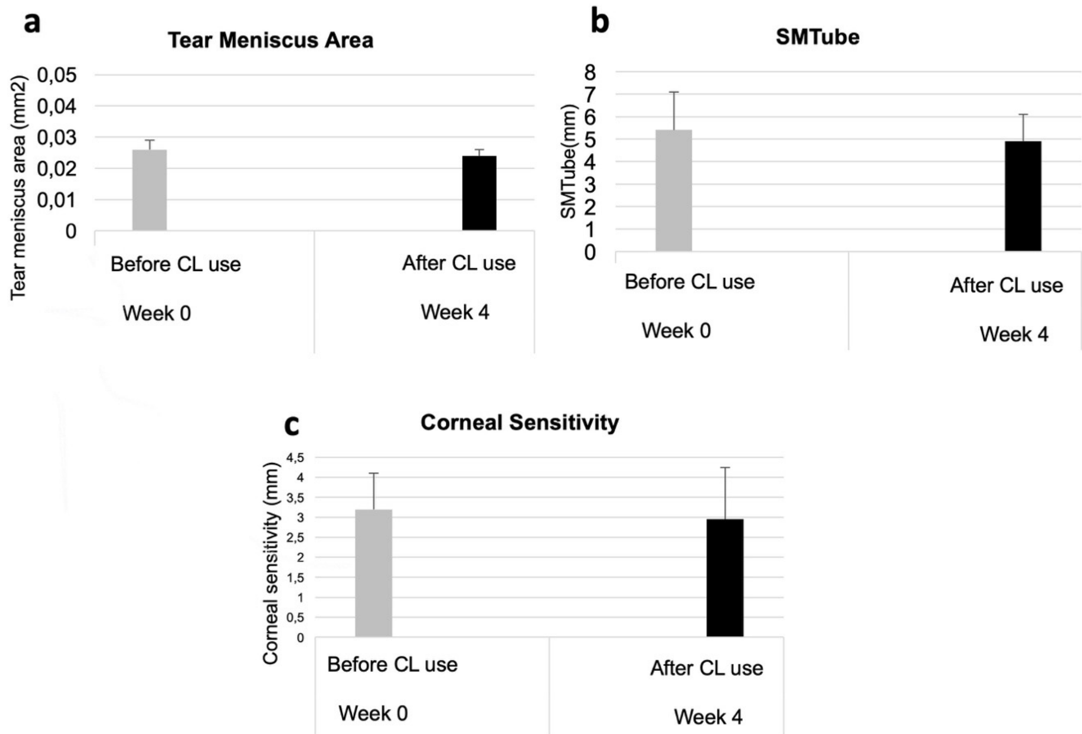

Figure 4. Comparison of tear meniscus area (a), strip meniscometry tube values (b), and corneal sensitivity (c) before and after silicone hydrogel contact lens use showed no statistically significant changes in any of the parameters (p>0.05)

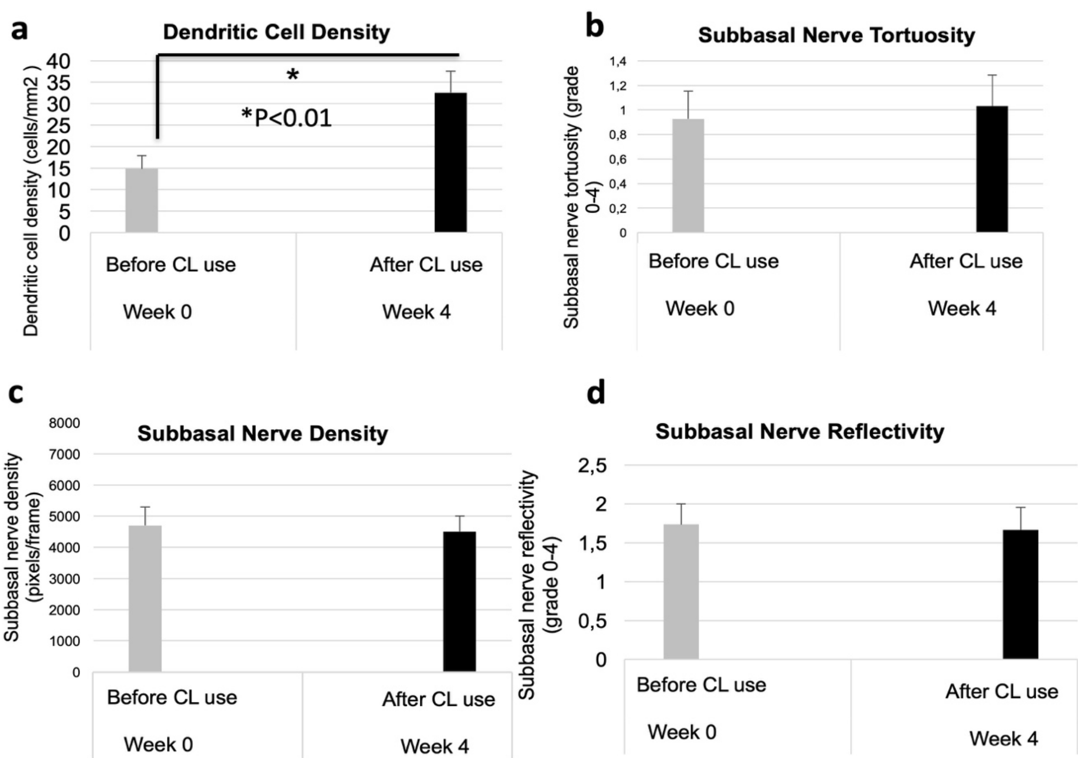

Figure 5. Before and after silicone hydrogel contact lens (CL) use. There was a statistically significant increase in dendritic cell density (a) (*p<0.01). There was no statistically significant change in subbasal nerve tortuosity (b), subbasal nerve density (c), or subbasal nerve reflectivity (d) (p>0.05)

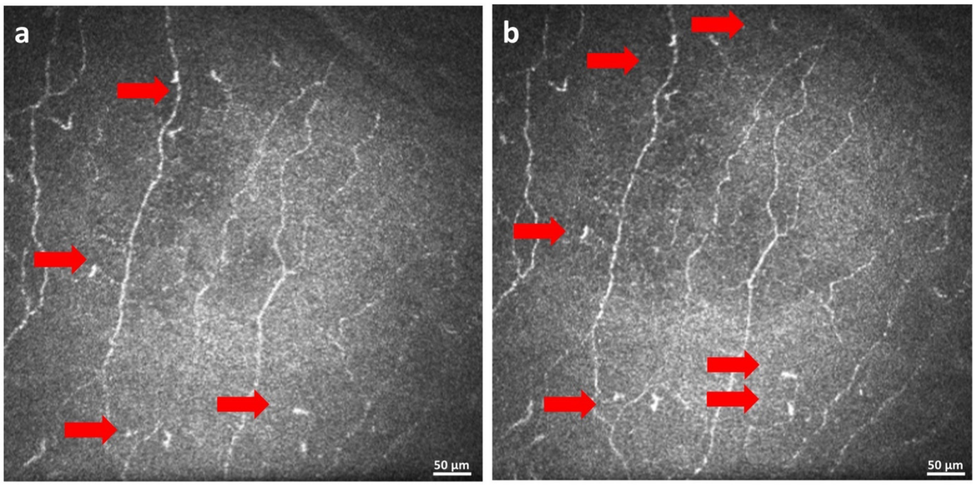

Figure 6. Representative in vivo confocal microscopy images showing dendritic cells (red arrows). Compared to before silicone hydrogel contact lens use (a), the same patient showed a marked and statistically significant increase in dendritic cell density after use (b). The white bar shows 50 μm

increased after starting CL use, reached a peak at week 4, and decreased after week 4. This suggests that ocular inflammation may be greatest at week 4. The decrease observed after week 4 may be associated with user adaptation and subsequent reduced CL-related discomfort.[39] Begley et al.[40] showed that most CL users had symptoms such as dry eye symptoms, itching, irritation, photosensitivity, pain, and blurred or variable vision. We believe the reason for the high OSDI scores after SiHCL use in our study was due to complaints that may occur with initial use, similar to the study by Begley et al.[40]

Previous studies have shown that the most important causes of CL-related dry eye are hypoxia, inflammation, infection, and mechanical effects.[41,42] CLs are foreign bodies that cause clinical or subclinical inflammation of the ocular surface.[43,44] This response may be due to mechanical irritation, hypoxia, and mediator release. Both the inflammation from CL-related dry eye and the inflammation induced by the CL itself may lead to increased dendritic cell density. At the same time, this situation may also be influenced by feelings of ocular discomfort caused by the CL, considering the increase in OSDI score. With long-term use, we believe this inflammation and dendritic cell activation will decrease due to adaptation.

Corneal sensitivity is very important to ensure the maintenance of a healthy ocular surface. The corneal epithelium becomes more susceptible to external factors if there is a decrease in the blink reflex or tear volume due for any reason. Excessive evaporation and cooling lead to increased tear osmolarity. The reduction in tear volume and this increase in osmolarity stress the ocular surface epithelium, resulting in local inflammation and peripheral nerve damage.[45] Local inflammation and nerve damage can cause short- and long-term genetic and molecular changes in primary sensory neurons.[46] Sensory nerve terminals are densely and superficially located between epithelial cells on the corneal surface. Therefore, corneal surface nerves are easily affected by environmental factors (air pollution, low humidity), trauma (cataract and refractive surgery), and ocular surface diseases (pterygium, conjunctivochalasis, keratoconus).[47,48] In this study, we observed no change in corneal sensitivity after short-term SiHCL use.

In addition to corneal subbasal nerve density, other morphological parameters most frequently evaluated include nerve tortuosity, reflectivity, and beading.[49,50] Our results showed no changes in these parameters, which may be because the mechanical exposure to the subbasal corneal nerves in the short 1-month period did not have any effect on neural degeneration and regeneration.

Study Limitations

Limitations of our study are that all participants were men, we used a single type of CL material, the study sample was small, and the results do not include longer term follow-up. Expanding the number of people in this study and creating a new group using a CL of a different material may allow a better comparison and lead to different results.

Conclusion

This study used objective procedures to investigate changes in ocular surface and corneal subbasal nerve structure variables related to the use of a SiHCL that is frequently used in daily practice. In conclusion, our study provides useful data on the anatomical effects of short-term SiHCL use on the ocular surface and corneal subbasal nerves, and may guide future studies designed to evaluate the long-term effects of CL use.

Ethics

Ethics Committee Approval: Ethics committee approval was obtained from Muğla Sıtkı Koçman University Faculty of Medicine Medical Research Ethics Committee with the decision number 7/XII on 03/2021.

Informed Consent: Obtained.

Peer-review: Externally and internally peer reviewed.

Authorship Contributions

Surgical and Medical Practices: C.Ş., C.K., Concept: C.Ş., A.K., Design: C.Ş., Data Collection or Processing: C.Ş., C.K., Analysis or Interpretation: C.Ş., A.K., Literature Search: C.Ş., C.K., Writing: C.Ş., C.K.

Conflict of Interest: No conflict of interest was declared by the authors.

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