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# Evaluation of tear meniscus height and lipid layer patterns in the tear film in domestic cats: An observational study

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**Abstract**

**Background:** Healthy vision in humans and animals requires a stable tear film. The environmental factor could affect the status of the tear film. Therefore, assessing the tear film in animals is essential to avoid visual system disturbance.

**Aim:** The current research used a noninvasive device to evaluate the tear meniscus height (TMH) and lipid layer pattern (LLP) in domestic cats. In addition, the scores were compared with those of humans with healthy eyes.

**Methods:** Fifty-four domestic cats (28 males and 26 females; mean ± SD = 13.9 ± 18.2 months) were randomly selected and included in the study. The cats were healthy, without any ocular disorders or diseases. Fifty-four healthy eye subjects (27 males and 27 females; mean ± SD = 25.6 ± 5.1 years) were randomly recruited and took part in the study for comparison. EASYTEAR View+ was used, for the first time, to assess the tear film parameters on the right eye of each subject. The examiner allowed a 5-minute gap between the tests. Each test was performed by the same examiner three times, followed by calculating the mean scores.

**Results:** Significant differences (Mann–Whitney *U* test) were found in the median scores of LLP ($p = 0.009$) between cats and subjects with healthy eyes. The median TMH score was higher in cats (0.18 mm) than in humans (0.14). However, no significant difference (Mann–Whitney *U* Test, $p = 0.210$) exists in the TMH scores between cats and humans. The LLP analysis indicated that a dense white–blue lipid layer (grade 4 or D; lipid layer thickness, LLT, = approximately 80 nm) was predominant in both cats ($N = 24$, 44.4%) and humans ($N = 29$, 53.7%). In comparison, variable colors lipid layer (grade 5 or E; LLT = 90–140 nm) was a minority in cats ($N = 5$, 9.3%) and common in humans ($N = 16$, 29.6%). The statistical analysis indicated medium correlations between cats' TMH and LLP scores ($r = 0.431$, $p < 0.01$) and between age and TMH scores in humans ($r = 0.440$, $p < 0.01$). In addition, it indicated a weak correlation ($r = 0.291$, $p < 0.05$) between the LLP scores in cats and humans.

**Conclusion:** Assessing animals' tear film is essential to avoid any ocular disorders. EASYTEAR View+ is efficiently used to evaluate domestic cats' TMH and LLP. Cats have thicker lipid layers and longer TMH comparable to those reported for humans with healthy eyes.

**Keywords:** Domestic cats, Tear film, Dry eye, Tear meniscus height, Lipid layer patterns.

## Introduction

The tear film provides nutrition, lubrication, smoothness, and protection against microbes (Georgiev *et al.*, 2017). Ocular surface disorders (e.g., dry eye) cause various problems and tear film instability. Dry eye due to a deficiency of tear production is expected in animals (Sebbag *et al.*, 2018). The Schirmer test is a simple method to measure tear production. However, animals such as cats do not tolerate the insertion of a paper strip into their fornix well for a long time. The measurement scores for the tear volume might not be accurate. The tear volume measured could be lower than the actual one due to stress that causes a temporary reduction in tear secretion during the test (Lim *et al.*, 2009). The lacrimal glands are antigenically affected by sympathetic nerves, leading to a disturbance in tear secretion (Dartt, 2009; Proctor and Carpenter, 2014). The sympathetic nerves also affect tear secretion in humans (Whitwell, 1961). A recent study indicated that the Schirmer test is reliable for measuring tear secretion in cats living in different environments without sympathetic stimulation (Sebbag *et al.*, 2020). Two measurements occurred 30 and 60 seconds after the insertion of the paper strip. No significant difference in the measurement scores between the left and right eyes was noticed (Sebbag *et al.*, 2020).

The tear ferning (TF) test assesses human tears' quality (Alanazi *et al.*, 2022; Fagehi *et al.*, 2022a). In addition,

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the test allows the evaluation of the quality of tears collected from cats (Veloso *et al.*, 2020). The median TF grade was 1 (0.5) using either grading scale (Veloso *et al.*, 2020). Based on the four and five grading scales (Rolando, 1984; Alanazi *et al.*, 2021), the data analysis showed no significant difference in TF grades between the right and left eyes. In addition, it showed no significant difference between the age of cats and the TF grads (Veloso *et al.*, 2020). Dry eye symptoms were reported in cats (Uhl *et al.*, 2018). However, the TF test showed that most cats (96.6%) showed normal and healthy tears based on the four-point grading scale. While using the five-point grading scale showed that all cats have normal eyes (Veloso *et al.*, 2020). The TF test is easy, safe, and simple, assessing the quality of cats' tears. However, the TF patterns depend on humidity, temperature, and tear composition (Norn, 1987). The TF requires at least 10 minutes for the tears to dry and inspect the fern using a light microscope. In addition, variation in the grading is possible among different examiners, suggesting that the scores depend on the observer's interpretation.

The osmolarity test showed no significant difference in the score recorded in normal-eye cats (328.5 ± 17.9 mOsms/l) and those having conjunctivitis (325.0 ± 24.8 mOsms/l) (Davis and Townsend, 2011). However, the tear breakup time (TBUT) was significantly higher in cats with conjunctivitis (Lim and Cullen, 2005; Lim *et al.*, 2009). The acceleration in TBUT is associated with a reduction in goblet cells responsible for mucin production and, therefore, an increase in TBUT (Lim *et al.*, 2009; Watanabe, 2022).

Evaluating the tear film in cats using other tests is still needed. The tests should be simple, noninvasive, fast, nonexpansive, and comfortable for the animals. Therefore, the current research reports the noninvasive assessment of tear meniscus height (TMH) and lipid layers patterns (LLPs) in cats using a single device for the first time.

## Materials and Methods

### *Cats and healthy eye subjects*

Fifty-four domestic cats (28 males and 26 females; mean ± SD = 13.9 ± 18.2 months) were randomly selected and included in the study. The cats were healthy, without any ocular disorders or diseases. In addition, 54 healthy eye subjects (27 males and 27 females; mean ± SD = 25.6 ± 5.1 years) were randomly recruited and took part in the study for comparison. EASYTEAR View+ was used to assess the tear film parameters on the right eye of each subject. The examiner allowed a 5-minute gap between the tests. Each test was performed by the same examiner three times, followed by calculating the mean scores. The participants completed the ocular surface disease index (OSDI), and subjects scoring above 13 were excluded (Schiffman *et al.*, 2000).

### *EASYTEAR view+*

EASYTEAR View+ (EASYTEAR S.R.L., Via Maioliche, Trento, Italy) was used to evaluate the tear film parameters in animals and humans. It assesses the TMH and visualizes the interference of the tear film with the lipid phase. It uses white, blue, and infrared LED lighting, minimizing film tear drying or alteration during measurements. A single examiner performed the TMH and LLP measurements in triplicate and then calculated average scores. Five minutes between consecutive tests were allowed as a gap. The examiner had difficulty measuring the noninvasive TBUT of cats with high consistency.

### *TMH test*

The TMH height (in millimeters) is the triangular cross-section between the cornea and the margin of the lower lid. A length lower than 0.2 mm is considered dry eyes (Masmali *et al.*, 2019).

### *Lipid layer pattern*

Grades 1 or A, 2 or B, 3 or C, 4 or D, and 5 or E were assigned to the LLP of domestic cats and humans (Garcia-Resua *et al.*, 2017; Fagehi *et al.*, 2022b). Grade E or 5 has the thickest (90–140 nm) lipid layer, and grade 1 or A has the thinnest (13–15 nm).

### *Statistical analysis*

SPSS (version 22; IBM Software, Armonk, NY) allowed the data analysis. The Kolmogorov–Smirnov test ($p < 0.05$) determined the non-normal distribution of the data. Therefore, the Mann–Whitney *U* test was employed to analyze the data in both groups. Spearman's correlation coefficient (*r*) tested the association between different parameters (Cohen, 1988). Meanwhile, the Wilcoxon signed-rank test was employed to investigate the significance of any differences between other parameters within the same group. The mean scores were calculated and represented as the median and interquartile range (IQR).

### *Ethical approval*

The individuals who took part in the present research were managed in accordance with the principles of the Declaration of Helsinki. Approval for the investigation was granted by the Institutional Review Board at King Saud University (E-21-6474).

## Results

Table 1 summarises the median scores for the TMH and LLP in the study (cats) and control (healthy-eye humans) groups. Significant differences (Mann–Whitney *U* test) were found in the median scores of LLP ($p = 0.009$) between cats and subjects with healthy eyes. The median TMH score was higher in cats (0.18 mm) than in humans (0.14 mm). However, no significant difference (Mann–Whitney *U* Test, $p = 0.210$) exists in the TMH scores between cats and humans. The side-by-side boxplots for the TMH and LLP in the study (cats) are shown in Figure 1. Meanwhile, those for the control (healthy-eye humans) groups are represented in Figure 2.

The LLP analysis indicated that a dense white–blue lipid layer (grade 4 or D; lipid layer thickness, LLT, = approximately 80 nm) was predominant in both

cats ($N$ = 24, 44.4%) and humans ($N$ = 29, 53.7%). In comparison, variable colors lipid layer (grade 5 or E; LLT = 90–140 nm) was a minority in cats ($N$ = 5, 9.3%) and common in humans ($N$ = 16, 29.6%). Figure 3 represents the LLP grade types for the cats and human groups.

The statistical analysis indicated medium correlations between cats' TMH and LLP scores ($r$ = 0.431, $p$ < 0.01) and between age and TMH scores in humans ($r$ = 0.440, $p$ < 0.01). In addition, it indicated a weak correlation ($r$ = 0.291, $p$ < 0.05) between the LLP scores in cats and humans.

## Discussion

Different tools, tests, and devices can assess cat tear film parameters. Various parameters need to be evaluated due to the intricate nature of the tear film. Using a single instrument to detect more than one tear film parameter is more convenient than using different ones. The current research is the first report to TMH and LLP on domestic cats using EASYTEAR view+. The results indicated that cats have TMH and LLP scores comparable to individuals with healthy eyes.

The use of EASYTEAR view+ showed significant differences in the TMH scores between subjects with healthy eyes and those for smokers ($p$ = 0.030). A similar observation has been made with subjects with a high body mass index (BMI; $p$ = 0.041) (Fagehi *et al.*, 2022b). In addition, it also showed significant ($p$ < 0.001) differences between subjects with healthy eyes, smokers, and subjects with a high BMI for the LLP grades (Fagehi *et al.*, 2022b). These results support the previous ones obtained using different tools and devices.

The quality of cats' tears ($N$ = 6) was assessed using the TF test based on a four-point scale (Rolando, 1984). Most cats (96.6%) had normal eyes (grades I and II). All cats had normal eyes (grades 0–2) (Veloso *et al.*, 2020) based on a five-point grading scale (Alanazi *et al.*, 2021). It should be noted that the TF test has been used in combination with other tests to assess tear film parameters (Masmali *et al.*, 2016; Alanazi *et al.*, 2019). The main proteins present in the tears of cats and humans are alike (Guedes *et al.*, 2021). The proteins are responsible for maintaining the homeostasis of cats' ocular surface and enhancing defense mechanisms (Veloso *et al.*, 2021).

Based on the Schirmer test (Sebbag *et al.*, 2020), no significant difference in tear production was found among cats examined at a private practice, clinic, and hospital. Another study suggested that the Schirmer tear test and strip meniscometry are viable options for evaluating cat tear production (Kovaļčuka *et al.*, 2021). In the strip meniscometry test, the variation in average measurements may be attributed to the inclusion of different cats that varied in age in each study and a

**Table 1.** Median (IQR) scores for the TMH and LLP in the study (cats) and control (healthy-eye humans) groups.

| Parameter | Cats ($N$ = 54) | Healthy-eye humans ($N$ = 54) | $p$-value* |
|---|---|---|---|
| TMH (mm) | 0.18 (0.10) | 0.14 (0.10) | 0.210 |
| LLP | 4 (1) | 4 (1) | 0.009 |

*Significant difference (Mann–Whitney $U$ test).

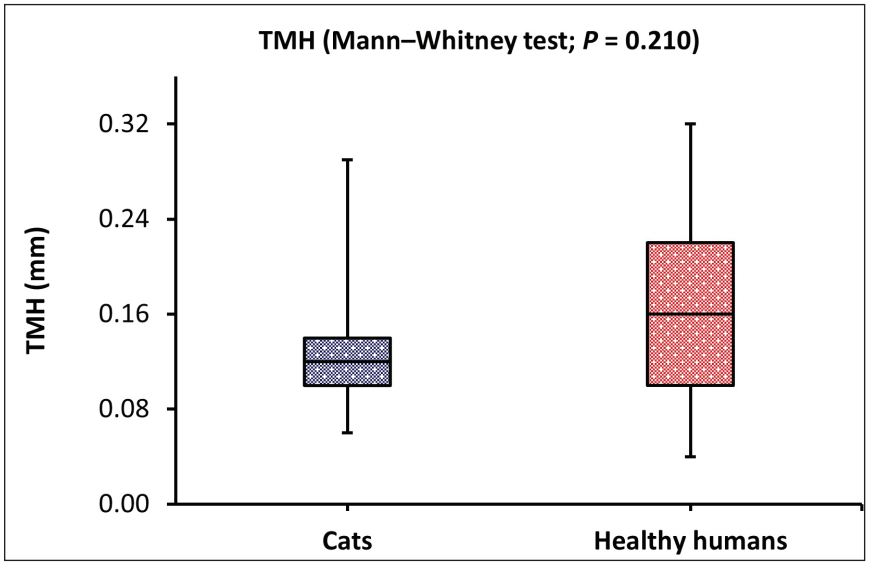


**Fig. 1.** Side-by-side boxplots of the TMH scores (mm) in cats and healthy-eye humans.

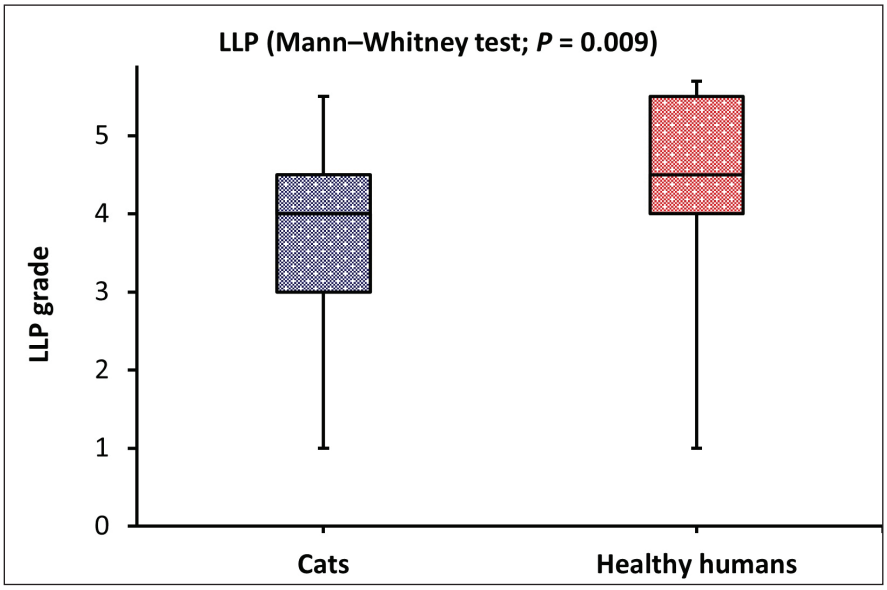


**Fig. 2.** Side-by-side boxplots of the LLP scores in cats and healthy-eye humans. There are five grades of the lipid layer. Grade 1 or A is gray and is 13–15 nm thick. Grade 2 or B is more compact and is 30–50 nm thick. Gray wave-like patterns characterize grade 3 or C and are 50–80 nm thick. Grade 4 or D is a dense white–blue layer that is approximately 80 nm in thickness. Variable colors characterize grade 5 or E and are 90–140 nm thick.

potential human factor (Rajaei *et al.*, 2018). Cats with a deficiency in tear production have displayed low scores in the Schirmer test (Sebbag *et al.*, 2020). Cats have a greater area of their corneas that requires lubrication compared to humans (Carrington and Woodward, 1986; Lim *et al.*, 2009; Salouti *et al.*, 2013). Ctas have extra secretory tissue that provides additional aqueous tears to supplement the lacrimal gland's primary tear production.

The Schirmer test score in cats was found to correlate with TBUT measurements (Grahn *et al.*, 2005). Possible causes for differences in average TBUT scores include the type of breed, age, size of the population, and the method of testing used. The osmolarity scores can differ between healthy cats and those with conjunctivitis (Davis and Townsend, 2011). Age does not impact tear production or blink rate. While breed does not affect blink rate, it does have a notable effect on Schirmer's test score (Oksa-Minaḷto *et al.*, 2023).

In summary, the measurements of tear film parameters (e.g., Schirmer, TBUT, and blink rate scores) in cats are varied using the same test. Therefore, using a single device to detect more than one tear film parameter in cats is noninvasive, convenient, and simple. This study offers a straightforward evaluation of cats' TMH and LLP.

The study's limitations include using a limited number of tests to assess the tear film parameters of domestic cats. Future research is needed to have different animals of different sizes that cover other locations in Saudi Arabia. In addition, evaluating the parameters that measure tear evaporation, stability, ferning, volume, and osmolarity can provide a better understanding of the status of the tear film in animals.

In conclusion, this is the first study to use a non-invasive device to assess tear film parameters in domestic cats. EASYTEAR View+ is efficiently used to evaluate domestic cats' TMH and LLP. Cats have thicker lipid layers and longer TMH comparable to those reported for humans with healthy eyes.

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***Conflict of interest***

The authors declare that there is no conflict of interest.

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***Author contributions***

Essam S. Almutleb: Software, validation, formal analysis, funding acquisition, and draft preparation. Gamal A. El-Hiti: Conceptualization, methodology, investigation, funding acquisition, project administration, supervision, draft preparation, review, and editing. Fahad B. Al-Okail: Investigation. Basal H. Altoaimi: Software, validation, formal analysis, and draft preparation. Meznah S. Almutairi: Software, validation, formal analysis, and draft preparation. Mashaaer A. Baashen: Software, validation, formal analysis, and draft preparation. Mohammed Althomali:

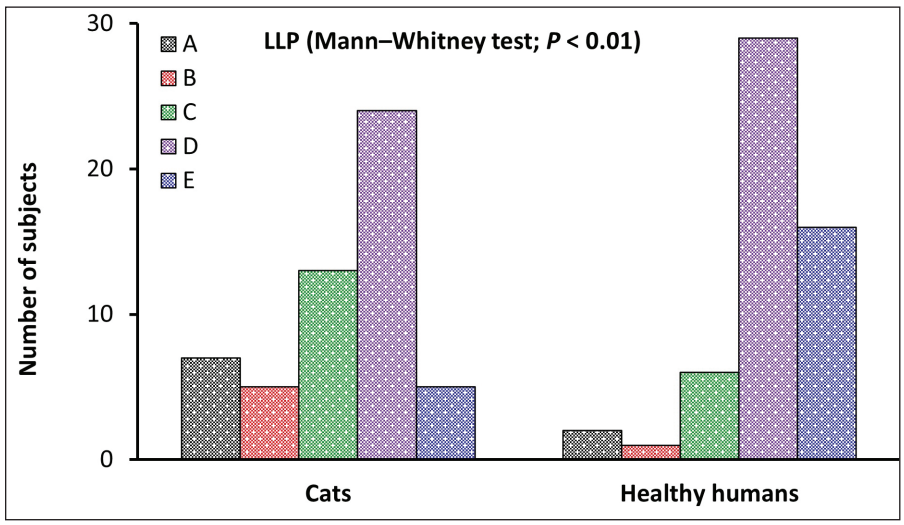


**Fig. 3.** LLP representation in the study (cats) and control (healthy-eye humans) groups. There are five grades of the lipid layer. Grade 1 or A is gray and is 13–15 nm thick. Grade 2 or B is more compact and is 30–50 nm thick. Gray wave-like patterns characterize grade 3 or C and are 50–80 nm thick. Grade 4 or D is a dense white–blue layer that is approximately 80 nm in thickness. Variable colors characterize grade 5 or E and are 90–140 nm thick.

Software, validation, and draft preparation. Saud A. Alanazi: Review and editing. Ali M. Masmali: Review and editing.

***Data availability***

All data supporting the findings of this study are available within the manuscript.

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