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Research Article

Risk Factors Associated with Prolapsed Nictitating Membrane Gland in Cats with Conjunctivitis: Analysis of 65 Cases

İrem ERGİN [1] Aytaç AKÇAY [2] Sümeyye SAİNKAPLAN [3] Oytun Okan ŞENEL [1(*)]

[1] Ankara University, Faculty of Veterinary Medicine, Surgery Department, TR-06070 Ankara - TÜRKİYE
[2] Ankara University, Faculty of Veterinary Medicine, Biostatistics Department, TR-06070 Ankara - TÜRKİYE
[3] Ankara University, Graduate School of Health Sciences, TR-06070 Ankara - TÜRKİYE

(*) **Corresponding author:**
Oytun O. ŞENEL
Phone: +90 312 317 0315
E-mail: oytunsenel@gmail.com

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Abstract

This study aims to evaluate animal-related and environmental risk factors influencing the development of prolapsed nictitating membrane gland (PNMG) in cats presented with complaints of conjunctivitis. A retrospective review of 65 cats diagnosed with PNMG, selected from a cohort of 318 cats presenting with conjunctivitis between 2019 and 2024. Data recorded included signalment, case history, ophthalmic examinations, intraoperative findings and outcomes. Direct ophthalmoscopy and slit-lamp biomicroscopy revealed no ocular abnormalities in cats with PNMG. Notably, 81.5% of cats with gland prolapse were identified as brachycephalic. Univariate logistic regression analysis indicated that breed, season, and history had a statistically significant effect on the occurrence of PNMG ($p<0.05$). Classification and regression tree analysis identified season as the most influential factor affecting PNMG, with the highest prevalence (40.5%) observed in brachycephalic breeds during spring season. The brachycephalic head structure was confirmed as a significant contributing factor to the development of PNMG in cats, consistent with findings in dogs. However, the seasonal variation and the role of ocular irritants in gland prolapse suggest that allergens may play a primary role in the etiology, as proposed by the authors.

Keywords: Brachycephalic, Cherry eye, Conjunctivitis, Hyperplasia, Ocular irritants, Third eyelid

Introduction

The third eyelid, also nictitating membrane, is a conjunctival fold located at the medial canthus of the eye, positioned between the lower eyelid and the globe in animals. Anatomically, in many animal species, including cats, it consists of a T-shaped cartilage and a prominent lacrimal gland situated at the base of this cartilage. This gland is anchored to the periorbital region by the fascia retinaculum. Known by various terms such as the glandula nictitans and tarsal gland, it is responsible for producing approximately 30-57% of the aqueous component of tears [1,2]. Its secretions play a crucial role in maintaining ocular health by supplying oxygen and nutrients to the eye. The T-shaped cartilage, with its horizontal segment running parallel to the eyelid margin and a vertical segment extending perpendicularly, forms the structural framework of the third eyelid. In cats, this cartilage exhibits elastic properties [2].

Prolapsed nictitating membrane gland (PNMG), predominantly observed in dogs [3], also occurs in cats [4], rabbits [5], wild animals [6], and bird species [7]. This condition involves the displacement of the lacrimal gland within the third eyelid from its anatomical position, accompanied by hyperplasia, which manifests as a visible red mass at the medial canthus. The condition, referred to as Harderian gland prolapse in rabbits and birds and as nictitating membrane gland prolapse in other species, is commonly known as cherry eye [1]. PNMG is significantly less common in cats than in dogs, and a review of the literature reveals that many studies on this condition are presented as case reports. In these studies, the most affected cat breeds are Burmese, British shorthair and Persian cats [8-11].

The prolapsed gland should be promptly surgically repositioned to its anatomical location to maintain essential lacrimal function and prevent desiccation and infection of the overlying conjunctival tissue. Neither the

removal of the third eyelid nor the gland is recommended, as this may lead to keratoconjunctivitis sicca later in life [2].

The aim of this study is to evaluate animal-related and environmental risk factors influencing the development of PNMG in cats presented with complaints of conjunctivitis.

Material and Methods

Ethical Statement

This study was approved by the Ankara University Animal Experiments Local Ethics Committee (Approval no: 2024-08-64). An "Informed Consent Form" was obtained from the animal owners before examination of animals.

Study Design

A retrospective review of a cohort of 318 cats of various breeds, ages, and genders presenting with conjunctivitis to the Ankara University Faculty of Veterinary Medicine Animal Hospital Ophthalmology Clinic between July 2019 and May 2024 identified 65 cases diagnosed with prolapsed nictitating membrane gland (PNMG). Statistical analyses primarily focused on these 65 cases, with additional comparisons involving the broader cohort to provide further context. Detailed records were maintained for all animals, encompassing signalment, history, ophthalmic examinations, the duration and localization of gland prolapse, the presence of third eyelid cartilage eversion, and postoperative outcomes. The ophthalmic examinations, including direct ophthalmoscopy, slit-lamp biomicroscopy, intraocular pressure measurements, Schirmer test type I and fluorescein staining were performed on all cats.

Specifically, antigen detection was conducted on tear samples from animals suspected of herpesvirus infection. Based on the anamnesis obtained from the owners, recommendations were made to eliminate environmental factors that could contribute to ocular irritation. Throughout the procedure, all animals were equipped with Elizabethan collars, and hyaluronic acid eye lubricants were prescribed for application to the prolapsed gland. Prophylactic treatment with topical tobramycin eye drops twice daily was started in all eyes a few days before surgery.

General anesthesia was induced with propofol and maintained with isoflurane. Periocular preparation was performed using diluted 10% povidone-iodine, and the conjunctival sac was irrigated. All cats underwent gland repositioning via a modified Morgan pocket technique [12]. Two parallel conjunctival incisions were made over the prolapsed gland to create a subconjunctival pocket. The intervening tissue was excised, and the gland was secured using a continuous Schmieden suture pattern with 5-0 absorbable monofilament suture. Knots were placed on the palpebral surface, and suturing was completed 3-4 mm from the incision margins. Light pressure was applied post-suturing. In cases of persistent cartilage eversion, a small horizontal conjunctival incision was made, the folded cartilage was dissected, and a strip was excised. All surgeries were performed by the same clinicians.

Postoperative treatment included topical tobramycin and hyaluronic acid eye drops twice daily, along with oral amoxicillin suspension (20 mg/kg) for one week. An Elizabethan collar was recommended for two weeks. Patients were monitored periodically for up to three years, during which no recurrences or complications were observed.

Statistical Analysis

The proportional distributions of conjunctivitis-affected cats were calculated based on breed, gender, neuter status, age, season, and medical history. The impact of these variables on the likelihood of gland prolapse occurrence was evaluated using univariate logistic regression analysis. To determine and classify the outcomes related to gland prolapse occurrence, including season, breed, and other historical factors, the classification and regression tree method was employed. A criterion of $P<0.05$ was used for all statistical comparisons. Data analyses were performed using the SPSS 30 software package.

Results

In the study, 318 cats complaining of conjunctivitis were categorized as brachycephalic (n=128) and non-brachycephalic (n=190) cats. Prolapsed nictitating membrane gland (PNMG) was diagnosed in 65 cats (20.4%). Of the 65 cats diagnosed with PNMG, 53 cats (81.5%) were identified as being of brachycephalic breeds. It was observed that cats under 2 years of age were most affected by PNMG (0-6 months: n=21, 6.1 months-2 years: n=29, 2.1-6 years: n=11, 6.1-10 years: n=2, 10.1-14 years: n=1, <14.1 years: n=1).

In the ophthalmic examinations of cats presenting with PNMG, fluorescein staining yielded negative results. Direct ophthalmoscopy and slit-lamp biomicroscopy revealed no ocular abnormalities or evidence of disease. Intraocular pressure was within normal limits for all animals (mean 20.60±2.02 mmHg). The mean Schirmer tear test value was 25.44±1.65 mm in the preoperative period, and measured 20.18±2.63 mm at the end of the first postoperative week.

Gland prolapse was bilateral in only one cat, while it was unilateral in all other animals (Left eye = 47.7%; Right eye = 50.8%) *(Fig. 1)*. The prevalence of cartilage eversion accompanying gland prolapse was determined to be 43.11% *(Fig. 2)*. In all animals affected by eversion,

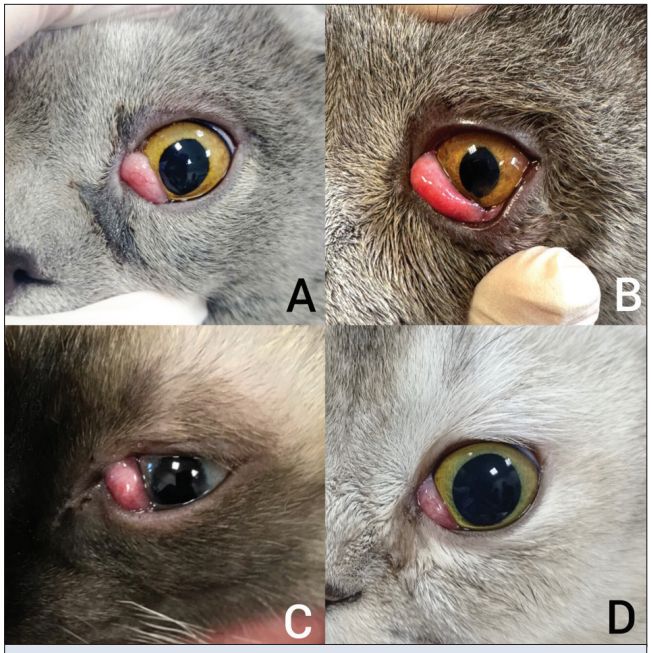

Fig 1. Preoperative appearance of prolapsed nictitating membrane gland with varying severity at the medial canthus

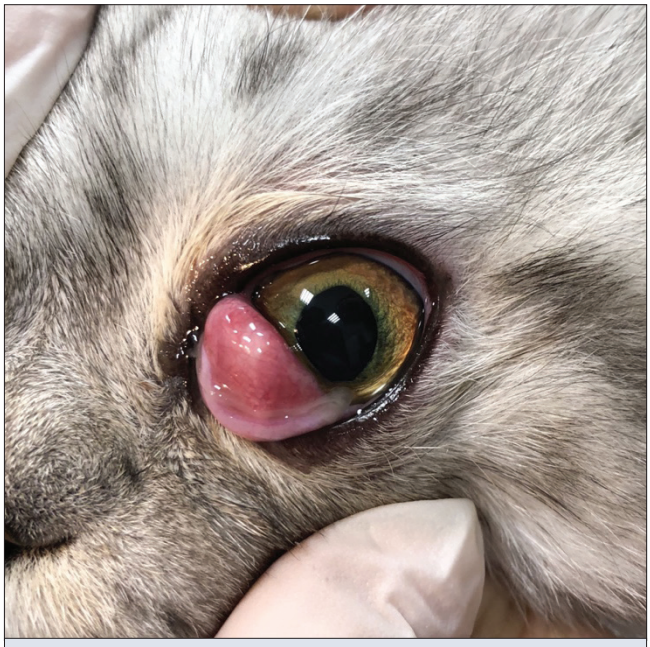

Fig 2. Cartilage eversion accompanied by prolapsed nictitating membrane gland in the left eye of a 3-year-old female Scottish Fold cat

the duration of prolapse development was observed to be two weeks or longer. In contrast, cases without cartilage eversion exhibited a variable duration, ranging from a few days to one week. Operative intervention was performed in 9 cats with persistent cartilage eversion following prolapse surgery. Throughout the entire follow-up period after the surgery, no complications or recurrence were observed in any of the cats *(Fig. 3; Fig. 4)*.

In the study, the effects of risk factors on the likelihood of gland prolapse in cats with conjunctivitis were evaluated

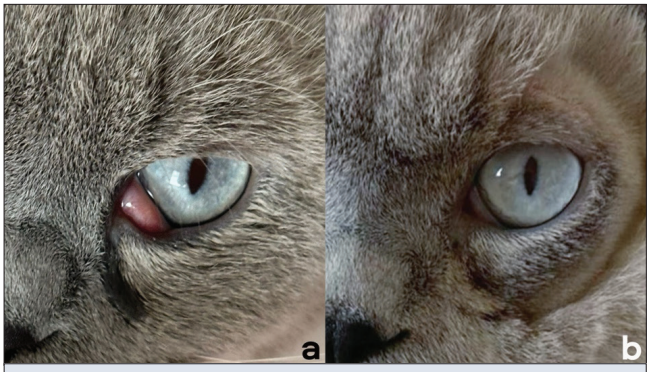

Fig 3. Prolapsed nictitating membrane gland in the left eye (a) and its appearance on postoperative day 5 (b) in an 8-month-old intact male Scottish Fold cat

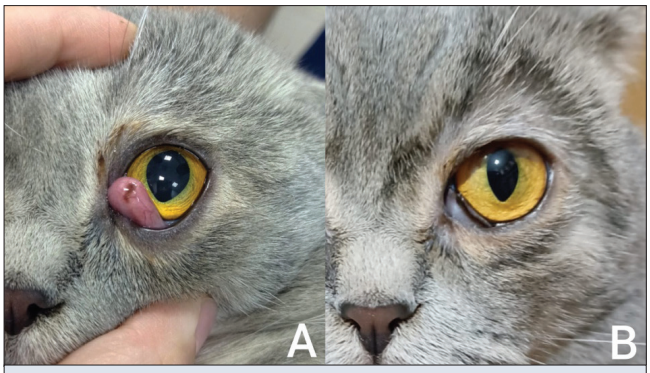

Fig 4. Preoperative (A) and postoperative 1st week (B) appearance of the prolapsed nictitating membrane gland in the left eye of a 1-year-old neutered male British Shorthair cat

using univariate logistic regression analysis. Based on the results of the univariate logistic regression model, breed, season, and history were found to have a statistically significant effect on the occurrence of PNMG in cats ($P<0.05$). Accordingly, the prevalence of PNMG was 6.556 times higher in brachycephalic breeds compared to non-brachycephalic breeds. As for the season of occurrence, the incidence during spring was noted to be 2.993 times higher compared to winter. Regarding animal history, the presence of ocular irritations and viral infections was observed. Notably, the incidence of the condition was found to be 3.113 times higher in cats exposed to ocular irritants compared to those with a history of infections *(Table 1)*. In conjunctivitis cases presented with complaints of herpesvirus infections, virus antigen (n=92) positivity was detected. Cats with PNMG were found to have the highest prevalence of herpesvirus antigen (12/17).

The study also utilized the classification and regression tree (CRT) method to classify cats with conjunctivitis based on factors influencing the occurrence of gland prolapse. According to CRT, the most influential factor affecting the occurrence of PNMG was found to be the season. The overall prevalence of PNMG was 17.0%, which increased to 27.4% in the spring season and was determined to be 40.5% in brachycephalic breeds. The

Table 1. *Univariate logistic regression analysis evaluating the effects of risk factors on the likelihood of gland prolapse in cats with conjunctivitis*

Variable	Variable Range	β	SE (β)	Wald	P	OR	95% Confidence Interval of OR	
Breed	Non-brachycephalic (Ref)	-	-	-	-	-	-	-
	Brachycephalic	1.88	0.34	30.672	<0.001	6.556	3.37	12.754
	Constant	-1.822	0.17	115.181	<0.001	0.162	-	-
Gender	Female (Ref)	-	-	-	-	-	-	-
	Male	0.514	0.283	3.308	0.069	1.672	0.961	2.91
	Constant	-	0.141	133.247	<0.001	0.196	-	-
Neutering status	Non-neutered (Ref)	-1.631	-	-	-	-	-	-
	Neutered	0.263	0.448	0.345	0.557	1.301	0.541	3.129
	Constant	-1.481	0.224	43.766	<0.001	0.227	-	-
Age	0-6 months	-	-	0.407	0.816	-	-	-
	6-24 months	0.2	0.314	0.407	0.524	1.222	0.66	2.26
	>24 months	0.118	0.37	0.102	0.75	1.125	0.545	2.322
	Constant	-1.594	0.141	127.912	<0.001	0.203	-	-
Season	Winter (Ref)	-	-	7.247	0.064	-	-	-
	Spring	1.096	0.409	7.168	0.007	2.993	1.341	6.679
	Summer	0.496	0.397	1.562	0.211	1.643	0.754	3.577
	Autumn	0.483	0.392	1.518	0.218	1.621	0.752	3.493
	Constant	-1.551	0.138	125.929	<0.001	0.212	-	-
History	FHV-I	-	-	13.987	0.003	-	-	-
	N/A	0.232	0.396	0.344	0.558	1.261	0.581	2.739
	Trauma	-19.192	7338.199	0.00	0.998	0.00	0.00	.
	Irritation	1.136	0.327	12.031	<0.001	3.113	1.639	5.913
	Constant	-6.467	1834.55	0.00	0.997	0.002	-	-

Categories indicated as 'Reference' are used as the baseline
Ref: *Reference;* ***β:*** *Estimated slope coefficient;* ***SE (β):*** *Standard error of the estimated slope coefficient;* ***Wald:*** *Wald statistic testing whether the slope coefficients are equal to zero for the model;* ***P:*** *P value associated with the Wald statistic;* ***OR:*** *Odds ratio*

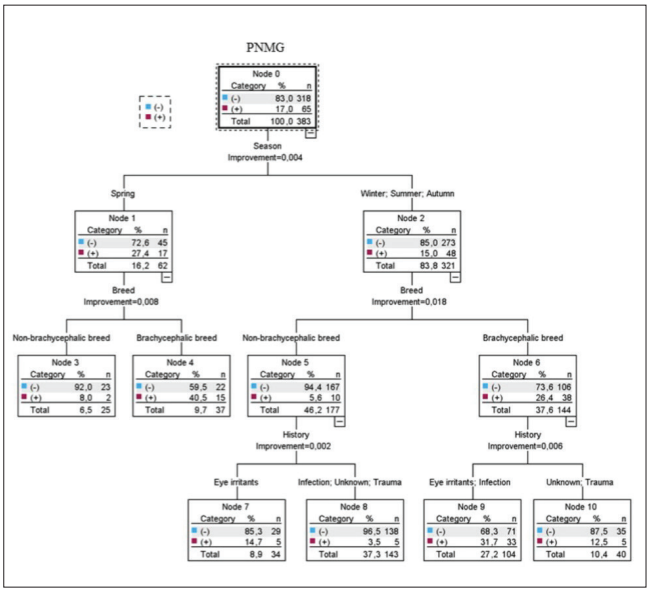

Fig 5. Classification and regression tree (CRT) used to classify cats with conjunctivitis based on factors influencing the occurrence of gland prolapse

prevalence of the condition was calculated as 15.0% during other seasons, increasing to 26.4% in brachycephalic breeds. Additionally, it was determined to be 31.7% in animals with a history of ocular irritation and infections. Upon classification results, the highest prevalence (40.5%) of PNMG was found to occur in brachycephalic breeds during the spring season *(Fig. 5)*.

Discussion

The conjunctival mucosa plays a crucial role in protecting the ocular surface through its anatomical, physiological, and immunological mechanisms. It supports ocular health while detecting and eliminating environmental irritants and pathogens. The nictitating membrane, a key component of the conjunctival surface, serves as a protective structure for the corneal surface due to its mobility across various species. Additionally, its glandular structure contributes significantly to normal tear production [2].

The exact etiology of prolapsed nictitating membrane gland (PNMG) remains unclear. Research has primarily focused on the deformation of the fascia retinaculum, which serves to anchor the gland in its anatomical position. It is hypothesized that the weakening of the gland's attachment to surrounding tissues also compromises its connection to the cartilage, thereby predisposing it to prolapse. Genetic and congenital factors are also suggested to play a role in the development of this condition. Notably, breeds such as the English Bulldog, Cocker Spaniel, Cane Corso, Neapolitan Mastiff, Lhasa Apso, Pekingese, and Shih Tzu have been reported to be predisposed to gland prolapse in dogs. A common characteristic among many of these breeds is the presence of disproportionately long eyelids relative to the size of the eyeball and orbit, resulting in a wide palpebral fissure. This condition is often associated with laxity of the lateral canthal ligament and supporting structures, with gland prolapse being particularly frequent in giant breeds [13-15]. Furthermore, skull morphology has been identified as a significant predisposing factor for PNMG. Numerous studies have established a strong correlation between brachycephalism and gland prolapse [13,16,17]. The narrow inferior orbital region in brachycephalic skulls has been proposed as a contributing factor to this predisposition [16,18]. Furthermore, the atypical positioning of the zygomatic salivary gland in the periorbital region, particularly in small and brachycephalic dog breeds, has been documented. The smaller size and steeper angle of the nictitating membrane gland compared to the zygomatic salivary gland suggest that this unusual placement, along with the resulting orbital congestion, may contribute to the development of prolapse [19].

However, the limited studies conducted in cats have not sufficiently established a breed-specific association with PNMG. An examination of the available literature, predominantly consisting of case reports, highlights brachycephalic breeds -characterized by their large, wide eyes, short noses, and disproportionate cheek widths- which have recently gained popularity [4,8,11]. The potential role of periorbital tissue laxity, a factor implicated in the etiopathogenesis of PNMG in brachycephalic dog breeds, should also be considered in brachycephalic cats. Notably, these cats are frequently reported to exhibit a predisposition to eyelid abnormalities such as entropion. While one previous study hypothesized that this may be related to increased laxity of the palpebral musculature, this remains speculative and lacks direct empirical confirmation [20]. In the present study, PNMG was most frequently observed in brachycephalic cats presenting with conjunctivitis. It is hypothesized that the development of this condition in brachycephalic cats may involve an abnormality characterized by the loosening of the nictitating membrane gland's attachment to surrounding tissues and cartilage, analogous to the palpebral laxity proposed in prior literature. The significant representation of brachycephalic cats within the study population further suggests a potential predisposition to PNMG. However, the condition is likely multifactorial in origin, underscoring the need for further investigative studies to clarify the underlying mechanisms.

The significant representation of brachycephalic cats within the study population further suggests a potential predisposition to PNMG. However, the condition is likely multifactorial in origin, underscoring the need for further investigative studies to clarify the underlying mechanisms. In this context, environmental contributors such as seasonal variation and ocular irritants, both of which showed statistically significant associations with PNMG development in the present study, warrant particular attention. The conjunctival mucosa, especially that of the third eyelid, contains conjunctiva-associated lymphoid tissue (CALT), a structured form of mucosa-associated lymphoid tissue (MALT) involved in local immune responses. In cats, CALT is most prominently located on the bulbar surface of the nictitating membrane and consists of lymphoid follicles lined by specialized epithelium containing cells with morphological characteristics of M cells [2,21]. These cells are known to transport antigens to underlying lymphoid structures, initiating mucosal immune cascades under inflammatory or allergenic conditions [22,23].

Given the observed seasonal and environmental correlations with PNMG in this study, it is plausible that conjunctival immune activation, potentially mediated by M cells, may contribute to inflammatory changes and glandular instability in predisposed individuals. Chronic antigenic stimulation due to allergens or irritants could exacerbate tissue remodeling processes in the third eyelid, particularly in cats with existing anatomic predispositions. While the present study was not designed to elucidate these immunopathogenic pathways, the findings underscore a plausible link between environmental immune triggers and gland prolapse. Future investigations incorporating immunohistochemical and molecular assessments of conjunctival immune activity under environmental stressors may provide deeper insights into the multi-factorial pathogenesis of PNMG.

In dogs, third eyelid gland prolapse is most commonly observed in young individuals [12,15]. While case reports in cats have referenced individuals under 2 years of age, no studies have specifically investigated the age-related incidence or prevalence of this condition. In the present study, age was found to be statistically insignificant in the development of gland prolapse in cats presenting with conjunctivitis. However, it was noted that cats with PNMG were numerically more likely to be under 2 years of age.

PNMG may occur independently or concurrently with the eversion of the T-shaped cartilage that forms the skeleton of the third eyelid [4,24]. In cats, the cartilage is elastic, with elastic fibers originating from connective tissue and extending into the perichondrium. In this region, the fibers form an extensive network that deepens, while the central cartilage is composed of similarly dense, finer elastic fibers. Notably, it has been observed that these elastic fibers form a regular network around chondrocytes exclusively in the cartilage of cats and horses. Additionally, the spade-like anatomical structure at the base of the cat's cartilage provides an advantage over species such as dogs and small ruminants, offering a more robust base for the cartilage [25,26]. In the present study, cartilage eversion was observed in nearly half of the cases alongside gland prolapse. Given the anatomical structure of this region in cats, the prevailing hypothesis is that gland prolapse contributes to the occurrence of eversion. In a small number of affected cases, surgical intervention was performed; however, in the majority of cases, gentle pressure applied for a few minutes was sufficient to correct the cartilage structure, either partially or completely. For mild cases of eversion, surgical intervention was generally avoided to prevent unnecessary manipulation of the cartilage, which forms the structural framework of the third eyelid and supports the gland. Notably, during the follow-up period, complete resolution of the eversion was observed in these cases.

Numerous surgical techniques have been developed for repositioning the third eyelid gland. These techniques primarily involve either repositioning the gland to its anatomical position using specialized sutures [27] or placing the prolapsed gland into a pocket created in the surrounding tissue [12]. Among these methods, the Morgan pocket technique remains the most commonly preferred approach due to its advantages, such as minimizing damage to the gland's ducts and preserving tear production [3,28,29]. In the present study, the modified Morgan pocket technique was utilized for the treatment of PNMG in cats. All cats exhibited excellent tolerance to the procedure, which resulted in a highly successful, complication-free outcome. Long-term follow-up findings confirmed complete success, supporting the conclusion that the modified Morgan pocket technique is a safe and effective method for managing PNMG in cats. Postoperative normalization of tear production was observed, likely due to the repositioning of the prolapsed gland and the resolution of inflammation achieved through medical therapy. This resolution appears to have alleviated conjunctival irritation, thereby restoring normal tear production levels.

Total or partial excision of the nictitating membrane gland was deliberately avoided, given the well-documented risk of severe complications -particularly keratoconjunctivitis sicca- associated with gland removal [30,31]. Additionally, the improper selection of suture materials or surgical techniques during gland repositioning may result in secondary complications, such as cyst formation, persistent inflammation, or recurrence of prolapse [13,16]. These findings underscore the critical importance of preserving the gland and adopting meticulous surgical practices to achieve favorable outcomes. Notably, the careful selection of suture materials and the precise application of the pocket technique in this study successfully prevented such complications, leading to complete and uneventful healing.

In conclusion, the inclusion of cats diagnosed with conjunctivitis in this study was based on clinical observations indicating that PNMG is frequently detected during evaluations of ocular surface irritation or inflammation. This case selection enabled a focused analysis of animal-related and environmental risk factors potentially contributing to PNMG development. Our findings confirmed a strong association between brachycephalic head conformation and PNMG, consistent with previous reports in dogs. Additionally, the pronounced impact of seasonal variation and ocular irritants on gland prolapse supports the hypothesis that environmental allergens may play a significant etiological role. While immunological mechanisms were not directly examined in this study, the dense lymphoid architecture of the third eyelid mucosa -together with the observed seasonal and environmental influences- raises the plausible involvement of mucosa-associated lymphoid tissue (MALT) and M cells in the pathogenesis of this condition. Future immunopathological investigations focused on antigenic stimulation and mucosal immune responses may further elucidate these mechanisms. Although the symptomatic nature of the study population may limit the generalizability of our results, large-scale studies incorporating asymptomatic individuals will be essential for identifying independent risk factors beyond clinical presentation.

Declarations

Availability of Data and Materials: The data that support the findings of this study are available on request from the corresponding author.

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Conflict of Interest: The authors declare that there is no conflict of interest.

Declaration of Generative Artificial Intelligence (AI): The authors declare that the article, tables and figures were not written/created by AI and AI-assisted Technologies.

Authors' Contributions: İE: Conceptualization, data curation,

investigation, methodology, writing-original draft, writing-review and editing. AA: Data curation, formal analysis, writing-review and editing. SS: Data curation, investigation, writing-review and editing. OOŞ: Conceptualization, writing-review and editing, supervision.

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