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Research Article

ASSESSMENT OF CONJUNCTIVAL IMPRESSION CYTOLOGY AND ITS DIFFERING PATTERNS IN CONTROLLED AND UNCONTROLLED DIABETIC PATIENTS

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ABSTRACT

Background: Diabetes mellitus has a wide spectrum of effects on the ocular surface. Conjunctival impression cytology (CIC) is a relatively easy and non-invasive technique that allows for the evaluation of the morphology of the conjunctival epithelium and changes in the conjunctival goblet cells.

Objective: To assess conjunctival impression cytology patterns in controlled and uncontrolled diabetic patients and correlate cytological findings with HbA1c levels.

Materials and Methods: A comparative observational cross sectional study conducted in the Department of Ophthalmology, National Institute of Medical Sciences and Research, Jaipur, for 18 month. The total 216 patients with type 2 diabetes mellitus were divided into two groups: controlled (HbA1c level was ≤6.5%) and uncontrolled (HbA1c level was >6.5%). Ophthalmological detailed examination and impression cytology of the conjunctiva were carried out. The nasal bulbar conjunctiva was sampled under topical anaesthetic, stained for cytological examination and classified using Nelson's classification. Descriptive statistics and Student's t-test were used in SPSS and Microsoft software for data analysis.

Results: 216 subjects participated in the study, with a mean age of 59.3 ± 10.2 years. The goblet cells were more common in the controlled group (80.0%) than in the uncontrolled group (59.4%), while the metaplastic cells were more common in the uncontrolled group (24.5% vs 10.0%). Nelson Grade 0 was mainly seen in controlled patients and Grades 2 and 3 were more common in uncontrolled patients (p=0.001). There was a significant association among fundus findings and HbA1c status (p=0.001) with the uncontrolled group having more cases of proliferative DR and clinically significant ME. Nelson grade (p=0.008) was significantly associated with the duration of diabetes. Patients with HbA1c >6.5% were more likely to have reduced corneal sensation (10.18%) compared with patients with HbA1c <6.5% (p=0.001) who had no reduced corneal sensation.

Conclusion: There were positive associations between glycaemic control and duration of diabetic disease and increasing severity of abnormalities in the conjunctival epithelium. CIC may be a helpful non-invasive tool for early detection of changes in the ocular surface in diabetic patients.

KEYWORDS: Diabetes Mellitus, Conjunctival Impression Cytology, HbA1c, Goblet Cells, Squamous Metaplasia, Nelson Grading, Diabetic Retinopathy.

Introduction

Diabetes mellitus refers to a chronic metabolic disease in which blood sugar levels are consistently elevated, and which is linked with a wide variety of microvascular and macrovascular complications. Diabetic retinopathy is the most widely known complication of diabetes, but diabetes can also impact the ocular surface such as the conjunctival epithelium, goblet cells, corneal nerves and tear film[2,7]. Prolonged hyperglycaemia, microangiopathy, neuropathy, oxidative stress and inflammation adversely affect the function of the epithelium. The study documents these changes at the surface of the eye as being associated with, and perhaps even a natural consequence of, clinically observed retinal changes[6,11,12].

Goblet cells are found in the conjunctiva which produces mucin and stabilizes the tear film. Diabetic changes may include decreased goblet-cell numbers, enlargement of the epithelium, changes in the nuclear/cytoplasmic ratio, keratinization and squamous metaplasia[10,13-16]. The changes can be identified by using a minimally invasive technique, called conjunctival impression cytology, whereby very superficial conjunctival cells are collected onto a membrane and examined under the microscope[17]. The study highlights that CIC is able to identify subclinical changes of the ocular surface and that the severity of the cytological abnormalities may be linked to glycaemic control[3,4,8,9].

Conjunctival epithelial changes are graded according to Nelson's classification, which corresponds to the progressive squamous metaplasia and goblet-cell loss. The present study thus aimed to compare the CIC pattern in controlled diabetic patients with that of uncontrolled, and study the relationship between these patterns and HbA1C, duration of diabetes, DR and CS[1,5].

Materials and Methods

It was an observational cross-sectional study done in the department of ophthalmology, National Institute of Medical Sciences and Research, Rajasthan, Jaipur. Study was approved by ethical committee of our institute and adhered to the tenets of declaration of Helsinki. The patients with diabetes mellitus were stratified into two groups: Group 1 (HbA1c ≤6.5%) and Group 2 (HbA1c >6.5%). The stated study period was 1 July 2024 to 31 December 2025 (18 months).

Patients were included if they had known type 2 diabetes mellitus and either HbA1c ≤6.5% or HbA1c >6.5%, were able to give informed consent and were age >40, regardless of sex. The exclusion criteria were: use of contact lenses, smoking, vitamin A deficiency, previous eye trauma or surgery, lid/adnexal disorders or corneal pathology, topical/systemic eye medication, and anaemia.

Conjunctival Impression Cytology

The cellulose acetate filter paper disc (pore size 0.2 µm) was applied to the nasal bulbar conjunctiva, adjacent to the limbus, after topical anaesthesia with 0.5% proparacaine, and was gently pressed for about 10–15 seconds. Specimen placed on albumin coated slide, fixed prior cytopathological processing. Study describes staining with Papanicolaou stain in the processing protocol and also it reports that the samples were stained with PAS and H&E for cytological examination[18-20]. Microscopic examination was undertaken by a pathologist who had no knowledge of the clinical information. Cytological assessment was made on the basis of epithelial morphology and goblet-cell density, and classified as per Nelson's classification.

Nelson grading of goblet cells	
Grade	**Features**
0	>500 goblet cells / mm Small ,round epithelial cells with large nuclei
1-2	100-500 goblet cells/mm
3	<100 goblet cells/mm Large , polygonal epithelial cells with small nuclei

Statistical analysis was performed using IBM SPSS Statistics and Microsoft Excel.

Ethical approval for the study was obtained from the Institutional Ethics Committee of NIMS University/National Institute of Medical Sciences & Research, Jaipur. Written informed consent was obtained from all participants prior to their enrolment in the study.

Results

A total of 216 participants were included. The overall mean age was 59.3 ± 10.2 years. There were 150 males (69.4%) and 66 females (30.6%). The controlled group (group 1) had a mean age of 58.9 ± 9.8 years and the uncontrolled group (group 2) 59.7 ± 10.6 years.

Table 1: Demographic characteristics of study participants

Characteristic	HbA1c ≤6.5%	HbA1c >6.5%	Overall
Sample size	108	108	216
Male	77 (70.0%)	73 (68.9%)	150 (69.4%)
Female	33 (30.0%)	33 (31.1%)	66 (30.6%)
Mean age, years	58.9 ± 9.8	59.7 ± 10.6	59.3 ± 10.2

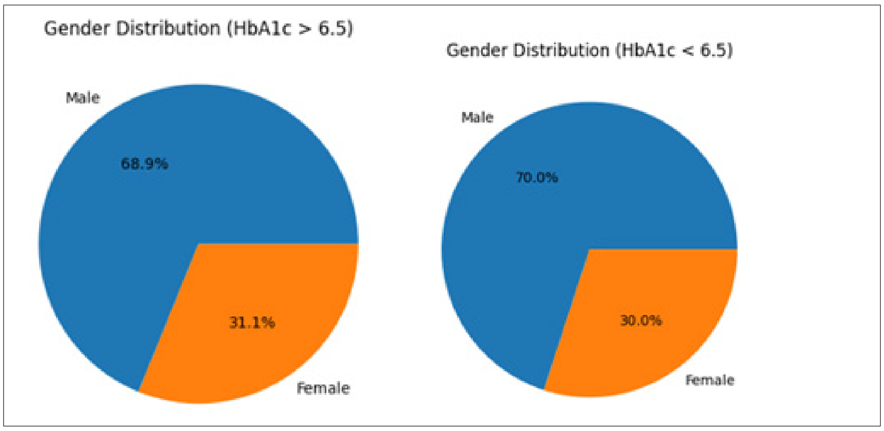

Figure 1. Gender distribution according to HbA1c level

Table 2: Distribution of conjunctival cell types according to HbA1c level

Cytological finding	HbA1c ≤6.5%	HbA1c >6.5%	Total
Goblet	88 (80.0%)	63 (59.4%)	151
Anucleate	33 (30.0%)	21 (19.8%)	54
Squamous	25 (22.7%)	24 (22.6%)	49
Dysplastic	25 (22.7%)	24 (22.6%)	49
Degenerative	14 (12.7%)	19 (17.9%)	33
Keratinized	16 (14.5%)	5 (4.7%)	21
Pigmented	11 (10.0%)	5 (4.7%)	16
Columnar	8 (7.3%)	12 (11.3%)	20
Metaplastic	11 (10.0%)	26 (24.5%)	37

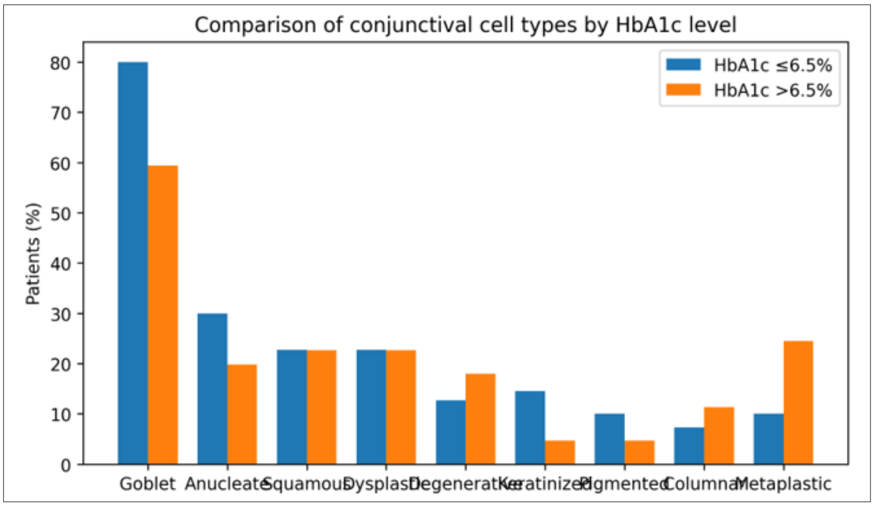

Figure 2.Comparison of conjunctival cell types by HbA1c level

Cytological examination demonstrated multiple epithelial cell types. Goblet cells were the most frequent finding, occurring in 88 (80.0%) patients in the group1 and 63 (59.4%) in group 2. Metaplastic cells were more frequent in group 2 (26; 24.5%) than in group 1(11; 10.0%). Degenerative cells were also somewhat more frequent in group 2.

Nelson grading showed a significant association with glycaemic status (p=0.001). Grade 0 was predominantly seen in the HbA1c ≤6.5% group (37; 88.1% of Grade 0 cases). In contrast, Grades 2 and 3 were more frequent in the HbA1c >6.5% group, accounting for 62.1% and 66.7% of cases within those grades, respectively.

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Table 3: Nelson grades according to HbA1c level

Nelson grade	HbA1c ≤6.5%	HbA1c >6.5%	Total	P value
Grade 0	37 (88.1%)	5 (11.9%)	42 (19.4%)	0.001
Grade 1	30 (50.0%)	30 (50.0%)	60 (27.8%)	
Grade 2	25 (37.9%)	41 (62.1%)	66 (30.0%)	
Grade 3	16 (33.3%)	32 (66.7%)	48 (22.2%)	

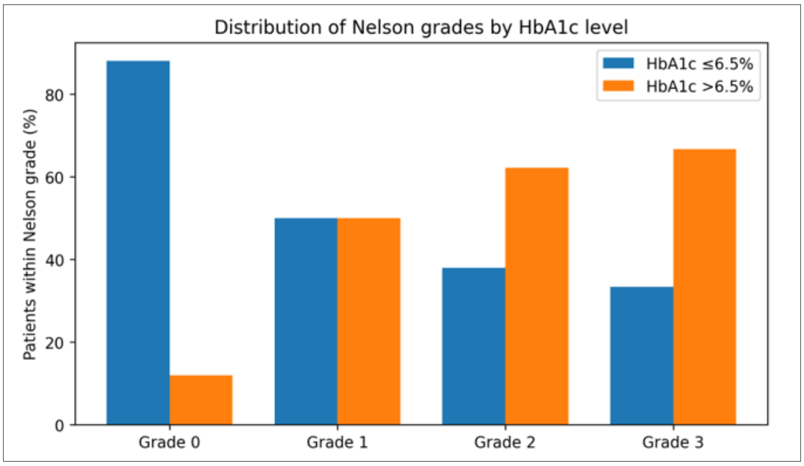

Figure 3.Distribution of Nelson grades by HbA1c level

Table 4: Nelson grades correlated with fundus findings

Fundus finding	Grade 0	Grade 1	Grade 2	Grade 3	Total	P
Normal	46 (38.3%)	30 (25.0%)	29 (24.2%)	15 (12.5%)	120	0.001
NPDR	6 (15.0%)	17 (42.5%)	17 (42.5%)	0 (0.0%)	40	
PDR	0 (0.0%)	0 (0.0%)	11 (39.3%)	17 (60.7%)	28	
CSME	0 (0.0%)	6 (21.4%)	11 (39.3%)	11 (39.3%)	28	

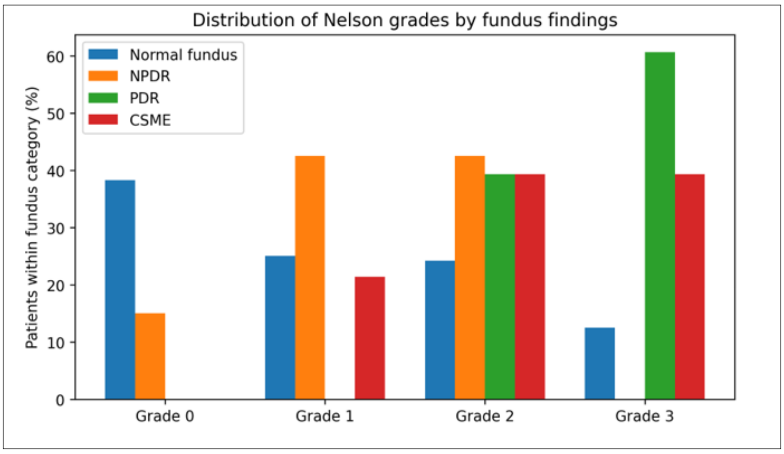

Figure 4.Distribution of Nelson grades according to fundus findings

Fundus findings were significantly associated with HbA1c status (p=0.001). Normal fundus findings were more frequent in group 1 (89; 82.4%) than in group 2 (28; 25.9%). Proliferative diabetic retinopathy was observed only in the HbA1c >6.5% group (25; 23.1%), while clinically significant macular edema was also substantially more frequent in group 2 (32; 29.62% versus 4; 3.7%). Nelson grades were also significantly associated with fundus findings (p=0.001). Patients with normal fundus findings showed a broader distribution with Grade 0 as the most frequent grade. In NPDR, Grades 1 and 2 were each present in 42.5% of patients. In PDR, no Grade 0 or Grade 1 cases were observed; Grade 2 occurred in 39.3% and Grade 3 in 60.7%. CSME was likewise associated with higher grades, with Grade 2 and Grade 3 each accounting for 39.3%.

Table 5: Association of duration of diabetes and corneal sensation with study findings

Parameter	Finding 1	Finding 2	P value
Duration of DM vs Nelson grade	<5 years: Grade 0 most frequent	5–10 and >10 years: higher Grade 2/3 pattern	0.008
Corneal sensation	HbA1c ≤6.5%: 108 (100%) normal	HbA1c >6.5%: 97 (89.8%) normal; 11 (10.18%) reduced	0.001

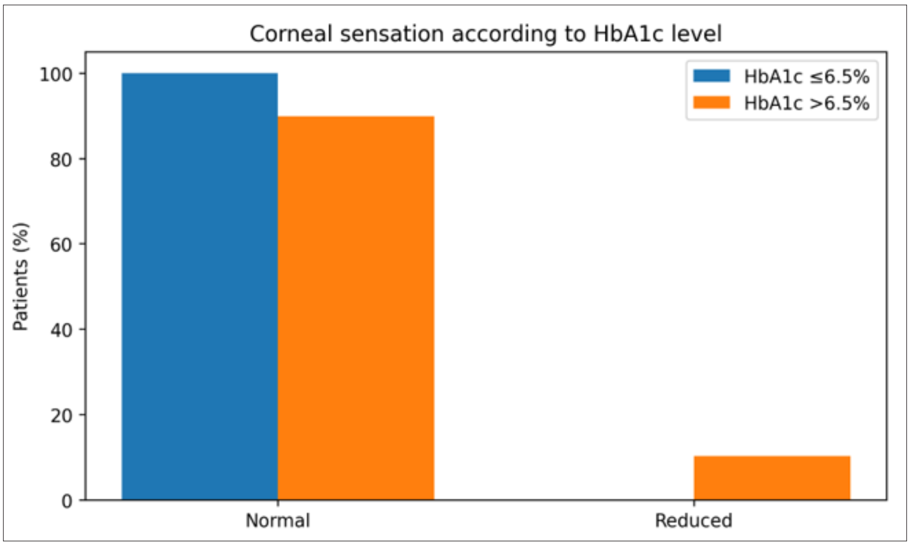

Figure 5.Corneal sensation according to HbA1c level

Duration of diabetes was significantly associated with Nelson grade (p=0.008). Among participants with diabetes for <5 years, Grade 0 was the most frequent category. In those with 5–10 years of diabetes, Grade 2 and Grade 3 predominated, while participants with diabetes for >10 years showed no Grade 0 cases and persistent Grade 1–3 abnormalities. Corneal sensation was assessed by the cotton-wisp test. All 108 patients in the HbA1c <6.5% group had normal corneal sensation, whereas 97 (89.8%) in the HbA1c >6.5% group had normal sensation and 11 (10.18%) had reduced sensation. The association with glycaemic status was statistically significant (p=0.001).

Discussion

The present study evaluated conjunctival epithelial changes by conjunctival impression cytology (CIC) and their association with glycaemic control, duration of diabetes mellitus (DM), severity of diabetic retinopathy (DR), and corneal sensation. Among 216 participants, the mean age was 59.3 ± 10.2 years, with a male predominance. The comparable age distribution between the controlled and uncontrolled groups suggests that the observed cytological differences were more likely related to glycaemic status than age-related changes. Similar demographic patterns have been reported by Dogru et al. and Sahu et al., who also observed male predominance in diabetic ocular surface studies. The higher representation of males in hospital-based studies may reflect differences in healthcare-seeking behaviour and access to medical services.

A significant association was observed between poor glycaemic control and conjunctival epithelial abnormalities. Patients with higher HbA1c levels demonstrated reduced goblet cell density, increased squamous metaplasia, keratinization, and higher Nelson grades. These findings are consistent with Saito et al. (2000) and Yoon et al., who demonstrated ocular surface changes in diabetes. Choudhary et al. (2020) and Mahadevan et al. similarly reported greater goblet cell depletion and more advanced squamous metaplasia in patients with poor glycaemic control. Furthermore, Gupta et al. (2022), as well as Zhang et al. (2021) and Li et al. (2022), demonstrated an association between increasing HbA1c levels and worsening conjunctival cytological changes, supporting a possible dose–response relationship between chronic hyperglycaemia and ocular surface damage.

The present study also demonstrated a significant relationship between duration of diabetes and CIC abnormalities, with increasing disease duration associated with higher Nelson grades, although the relationship was not strictly linear. This observation is comparable with the findings of Rossi et al. (2021) and Manchikanti et al., who reported progressive goblet cell loss and increasing ocular surface abnormalities with longer duration of DM. The absence of a consistent linear trend in the present study suggests that factors other than duration, particularly glycaemic control, may have a greater influence on conjunctival epithelial integrity.

A significant association was also observed between the

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severity of DR and cytological abnormalities. Figueroa-Ortiz et al. and Thaseen et al. (2019) reported increasing ocular surface damage and higher grades of squamous metaplasia with increasing severity of DR, while Sridhar et al. (2023) suggested that ocular surface changes may parallel the systemic microvascular burden of diabetes.

The mechanisms underlying these changes are likely multifactorial, involving diabetic microangiopathy, neuropathy, chronic inflammation, and tear-film instability. Di Zazzo et al. highlighted the role of inflammatory mechanisms in diabetic ocular surface disease, while Kim et al. (2020) demonstrated the contribution of diabetic neuropathy to reduced corneal sensation and impaired tear secretion. These mechanisms may collectively promote goblet cell loss, epithelial metaplasia, and ocular surface instability. Kheirkhah et al. (2020) similarly reported altered epithelial morphology and differentiation in diabetic patients. In the present study, symptoms such as dryness, burning, and foreign-body sensation were present in some patients, although several patients with cytological abnormalities remained asymptomatic. This is consistent with Dogru et al. and Hagan et al., who emphasized that diabetic ocular surface abnormalities may precede clinically evident symptoms.

Conclusion

A total of 216 patients with type 2 diabetes mellitus were included in this comparative cross sectional study; advanced cytological abnormalities of the conjunctiva were associated with poor glycaemic control. The goblet-cell representation was found to be lower, metaplastic changes and degenerative changes were higher and Nelson grades were higher in uncontrolled diabetic patients. HbA1c was significantly related to Nelson grading, fundus findings and corneal sensation whereas duration of diabetes showed significant association with cytological severity. CIC is an easy and non-invasive technique, which can be used as an additional tool in the work-up and early detection of changes in the ocular surface in diabetic patients.

Limitations

The study is cross-sectional and thus cannot prove a causal or temporal relationship for cytological changes. This was done at one tertiary-care center and might not be generalizable.

Ethics Approval and Consent

Permission was obtained from the Scientific and Ethics Committee of the National Institute of Medical Sciences & Research, Jaipur, and written informed consent was obtained from participants.

Conflict of Interest

No specific conflict-of-interest statement was provided in the study source; this should be confirmed by the authors before submission.

Data Availability

The underlying patient-level dataset was not provided as a public dataset in the study. Data-availability wording should be finalized according to the target journal's requirements.

References

1. Kinoshita S, Kiorpes TC, Friend J, Thoft RA. Basement membrane abnormalities in diabetic corneas. Prog Retin Eye Res. 2001;20(5):639–73.
2. Gipson IK. The epithelial basement membrane zone of the limbus. Invest Ophthalmol Vis Sci. 2007;48(10):4391–8.
3. Tseng SC. Staging of conjunctival squamous metaplasia by impression cytology. Ophthalmology. 1985;92(6):728–33.
4. Nelson JD. Impression cytology. Cornea. 1988;7(1):71–81.
5. Kaiserman I, Kaiserman N, Nakar S, Vinker S. Dry eye in diabetic patients. Am J Ophthalmol. 2005;139(3):498–503.
6. Dogru M, Katakami C, Nishida T. Tear function and ocular surface changes in non-insulin-dependent diabetes mellitus. Ophthalmology. 2001;108(3):586–92.
7. Yoon KC, Im SK, Kim HG, Park YG. Usefulness of impression cytology for evaluating the severity of dry eye disease. Korean J Ophthalmol. 2004;18(2):168–74.
8. Nelson JD, Wright JC. Conjunctival impression cytology. Arch Ophthalmol. 1984;102(7):1049–51.
9. Calonge M, Enriquez-de-Salamanca A, Diebold Y, et al. Impression cytology of the ocular surface: a review. Exp Eye Res. 2004;78(3):457–72.
10. Goebbels M. Tear secretion and tear film function in insulin dependent diabetics. Br J Ophthalmol. 2000;84(1):19–21.
11. Oshitari T, Yamamoto S, Roy S. Ocular surface changes in diabetic patients. Nippon Ganka Gakkai Zasshi. 2009;113(8):876–82.
12. Thomas J, Jacob GP, Srinivasan R, Sen S. Ocular surface changes in diabetes: A clinical study. Indian J Ophthalmol. 2019;67(1):64–70.
13. Mahadevan G, Hatti AS, Shashikala P, Arun Kumar S, Barki R. Conjunctival impression cytology and its varying patterns in controlled and uncontrolled diabetic patients. J Clin Ophthalmol Res. 2023;11(3):123–128.
14. Sahu J, Sahu S, Sahu S, Sahu S. Conjunctival impression cytology and tear film changes in diabetic retinopathy. J Clin Ophthalmol Res. 2024;12(1):45–50.
15. Citirik M, Demircan A, Ozdemir H, et al. Conjunctival impression cytology in non-proliferative and proliferative diabetic retinopathy. Cornea. 2014;33(6):595–600.
16. Manchikanti V, Manchikanti S, Manchikanti S, et al. Ocular surface disorder among adult patients with type II diabetes mellitus. J Clin Ophthalmol Res. 2020;8(2):101–106.
17. Figueroa-Ortiz LC, Figueroa-Ortiz J, Figueroa-Ortiz J, et al. Study of tear function and conjunctival surface in diabetic retinopathy. J Clin Ophthalmol Res.

2011;9(4):234–239.
18. Hagan S, Hagan S, Hagan S, et al. Biomarkers of ocular surface disease using impression cytology. Biomark Med. 2017;11(8):711–718.
19. Di Zazzo A, et al. Inflammatory and ocular surface mechanisms in diabetic disease. [As cited in the study bibliography].
20. Chotaliya R, Patel R, Patel S, et al. A review of ocular surface impression cytology along with its applications. Med J DY Patil Univ. 2025;18(1):1–8.

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