Original article:

Counjctival impression cytology of various ocular diseases

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Abstract:

Introduction: Impression cytology technique was used to study the cytological changes in various ocular diseases. It is very simple, noninvasive technique can be repeated if required and provides an good alternative to conjunctival diagnostic excision biopsy.

Material and Method : 100 patients were studied .Impression smear was taken with cellulose acetate filter paper and wet fixed and stained with H& E and periodic schiff stain.

Results: 100 patients were studied vary in age from 0 to 80 yrs . Out of 100 patients 47 were males and 53 were females.Impression cytology smear were graded based on epithelial cell morpholgy and goblet cell density according to Nelson and Adams grading. Findings observed in patients were compared with other studies .

Conclusion : Conjunctival impression Cytology is an easy, non invasive, quick, repeatable, techniques with minimal discomfort to patients and provides important diagnostic finding to support clinical diagnosis.

Key words : Impression cytology , Ocular disease

Introduction : Conjunctival impression cytology (CIC) is one of the techniques to study ocular surface disorders. CIC is a very simple, non invasive technique which can be repeated easily, can be done On OPD (out patient department) basis and does not require any specialized gadgets. Impression cytology, with cellulose acetate filters, was introduced in 1977 as a minimally invasive conjunctival biopsy. It provides an alternative to conjunctival diagnostic excision biopsy or conjunctival smears made from scrapes taken with a blunt spatula [1] .Applications for impression cytology include diagnosing a wide range of ocular surface disorders, documenting sequential changes in the conjunctival and corneal surface over time, staging conjunctival squamous metaplasia, and monitoring effects of treatment[2] .

Material and Method : This study comprised 100 patients of ocular surface disorder. Subjects were ranging from 0 to 80 years and all patients were submitted through a detailed ophthalmic examination. From each patient impression smear were taken from diseased eyes from both bulbar and palpebral regions. The impression was taken from temporal and bulbar conjunctiva. Upper lid was everted and impression were taken from palpebral conjunctiva, by applying rough surface of filter paper to conjunctiva and was gently pressed by a glass rod for 3-5 seconds and filter was removed with peeling motion. Strict sterilization was maintained during the

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procedure. This was then applied to a clean glass slide by which the impression smear was transferred to slide by applying gentle pressure over filter with rolling of glass rod and smears were fixed,

Smears were fixed in 50% ether and 50% alcohol.

The Smears were stained with –

(1) Haematoxylin and Eosin stain.

(2) Periodic Acid Schiff's stain.

Impression cytology specimens were graded based on epithelial cell morphology and goblet cell density

CYTOMORPHOLOGICAL FEATURES:

According to Nelson [3] and Adams [4] Grading :

Grade 0 – Small rounded epithelial cells with neucleocytoplasmic ratio of 1:2, lots of deeply stained goblet cells, intense PAS positive staining and good confluent sheet.

Grade 1 – Good cell sheet consisting of larger epithelial cells with decreased nucleocytoplasmic ratio 1:3. Goblet cells were slightly decreased but deeply stained with PAS stain.

Grade 2 – Large epithelial cells with decrease in nucleocytoplasmic ratio. Decreased number of goblet cells with reduced staining.

Grade 3 – Poor sheets of larger irregular epithelial cells with small nuclei, occasional goblet cells if seen stained palely.

Observation :

Patients between the ages from 0 to 80 years were studied. Out of 100 patients 47 were males and 53 were females.

Table 1 : No. of cases of various ocular disorders studied in present study

Condition	No. of cases
Nondiagnostic	09
Vitamin A deficiency	04
Conjunctivitis	18
Panophthalmitis	07
KCS	06
Post cataract surgery	12
Corneal ulcer	16
Corneal abscess	15
Contact lens wearer	08
Pterygium	02
Glaucoma : on long term Rx topical application of antiglaucoma drugs	02
Neoplastic	01
Total	**100**

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Table 2 Grading of smears studied in various ocular disorders

	Total	Grade 0	Grade 1	Grade 2	Grade 3
Nondiagnostic	09	00	00	--	--
Vitamin A deficiency	04	--	--	02	02
Conjunctivitis	18	05	09	04	--
Panophthalmitis	07	01	04	02	--
KCS	06	--	--	02	04
Post cataract surgery	12	02	07	03	--
Corneal ulcer	16	03	09	04	--
Corneal abscess	15	03	09	03	--
Contact lens wearer	08	07	01	--	--
Pterygium	02	02	--	--	--
Glaucoma : on long term Rx of topical antiglaucoma drugs	02	--	--	01	01
Neoplasia	01	NA	NA	NA	NA
Total	**100**	**30**	**41**	**21**	**07**

[NA- Not Applicable]

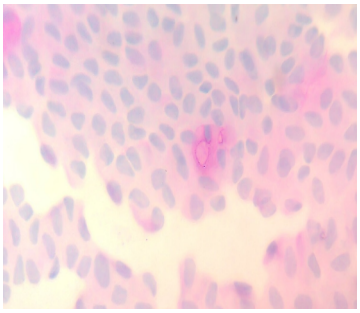

Figure 1 Vitamin-A deficiency Grade-II decreased goblet cells with reduced staining (PAS-H Stain X 400)

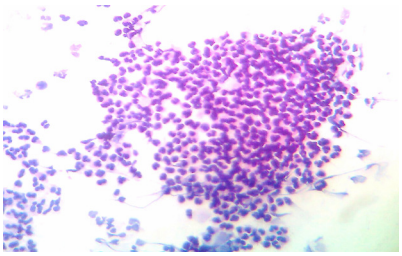

Figure 2 Many polymorphs observed in patient of acute conjunctivites (H & E 400X)

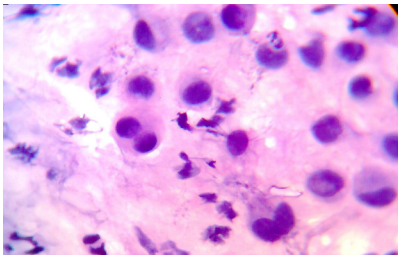

Figure 3 Binucleated cells obsereved in viral conjunctivitis (H & E 400X)

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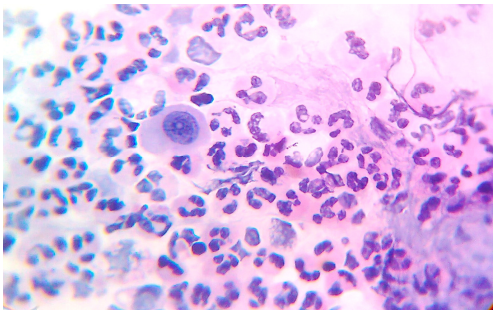

Figure 4 Many polymorphs observed with occasional mononuclear cells in patient of corneal abscess (H & E 400X)

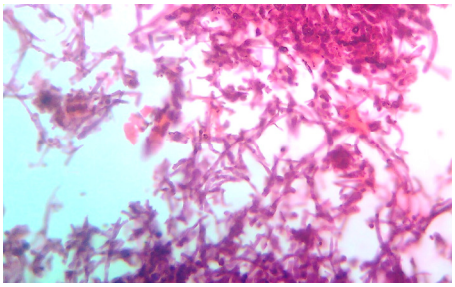

Figure 5 Fungal hyaphae with scattered polymorphs observed in patient of fungal corneal ulcer (H & E 400X)

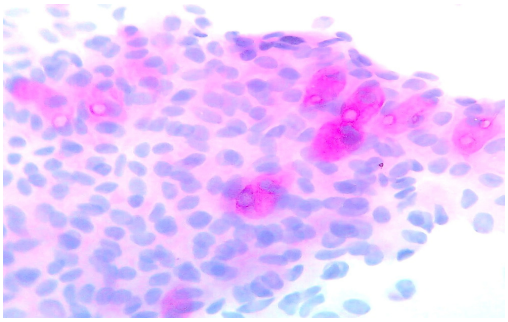

Figure 6 Increase in goblet cells and epithelial cells in contact lens wearer (PAS 400X)

Discussion : An impression cytology usually removes only 1–3 cell layers of the ocular surface and it is therefore ideal for studying the surface epithelium and method to determine the density of goblet cells in different areas of the conjunctiva. In Present study, Conjunctival Impression cytology from conjunctiva of 100 cases which were clinically diagnosed as ocular disorder were performed. Out of which 09 cases were observed with nondiagnostic materials. So sensitivity of these techniques was found to be 91%.

Sr. No.	Study	Year	Sensitivity
1	Natadisastra et al[36]	1988	93 %
2	Vedhal et al[2]	1998	83 %
3	Ersoz at al[49]	1998	90 %
4	Present study	2012	91 %

In pediatric age group (0 – 6 yrs) with vitamin A deficiency shows mainly Decrease goblet cell density (figure .1) correlates with study of Natadisastra et al[5] & Singh et al[6] In present study, 18 eyes of clinically diagnosed inflammatory lesions were studied. Out of them findings of acute inflammatory conjunctivitis suggested by inflammatory cells (figure 2), fibrinous mucoid material, with increased numbers of goblet cells. Binucleated epithelial cells were seen in viral conjunctivitis(figure 3) and degenerated epithelial cells in Panophthalmitis. Similar finding were observed by Dewan et al[7].In 2 eyes eosinophils were seen with few neutrophils suggestive of allergic conjunctivitis. Other workers[7] [8] obtained similar findings. 31 eyes studied showed plenty of polymorphs, few mononuclear cells and

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mucus material suggestive of corneal abscess (figure 4) and corneal ulcer (figure 5) showed necrotic debris with pus cells. Similar findings were observed by Adams et al[4]Smear collected from 12 eyes post cataract surgery showed regenerative cells as observed by G.N.Seal and S.K Seal[9]8 eyes of contact lens wearer (figure 6) studied showed an increase in number of goblet cell density. Similar finding were observed by other workers [10] [11]6 eyes of keratoconjunctivitis sicca showed squamous metaplasia decrease goblet cells and presence of polygonal cells. It was comparable to the study of Nelson [12] 2 eyes of clinically diagnosed pterygium showed squamous metaplasia, decrease in number of goblet cells. It was comparable to the study of Ranjana Bandyopdhyay et al[13](2010). Similarity in both studies is squamous metaplasia, which is constant finding in pterygium along with decrease in number of goblet cell.2 eyes of clinically diagnosed glaucoma patients on long term treatment of antiglaucoma drugs showed higher stage of metaplasia with decreased numbers of conjunctival goblet cells. It was comparable to the study of Rana K Sherwani et al[14](2008).

1 eye of clinically suspected malignancy studied, patient showed cytology of squamous cell carcinoma. Impression cytology has been used widely as a non-invasive method for conjunctival biopsy for suspected ocular surface squamous neoplasia [15]

As we can take multiple samples of the ocular surface at one sitting with minimal discomfort to the patient makes it an ideal method of investigating ocular disorders and providing useful supportive findings when the diagnosis is not clinically obvious or when the clinical diagnosis needs to be substantiated and documented. Ophthalmic clinics can intodue this technique into routine clinical practice with a team approach including the ophthalmologist and pathologist,

Conclusion : Conjunctival Impression Cytology is an easy, non invasive, quick, repeatable, techniques with minimal discomfort to patients and can be done on outpatient department basis. Conjunctival impression cytology is a very valuable tool in the understanding of ocular surface disorders and provides important diagnostic finding to support clinical diagnosis.

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