

Conjunctival impression cytology in patients with ectodermal dysplasia

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Summary

Ectodermal dysplasia is a group of symptoms affecting tissues and organs of ectodermal origin. In this study, conjunctival impression cytology technique which is a non-invasive method was performed in patients with ectodermal dysplasia to evaluate the effects on the conjunctiva. Six eyes of three patients with ectodermal dysplasia who were followed up by the Ophthalmology Clinic of Dicle University Faculty of Medicine and Prosthetic Dentistry Departments were included in the study. Bulbar conjunctiva impression cytology method was applied to the upper surface of the bulbar conjunctiva and the cell groups taken to the surface of cellulose acetate paper were fixed by a solution containing 70% ethyl alcohol, 37% formaldehyde and glacial acetic acid with a ratio of 20:1:1 and later stained by Periodic acid-Schiff and hematoxylin-eosin. Using impression cytology method a decrease in integrity of the cells, picnotic changes in the nucleus, epithelial cell-like changes and loss of goblet cells were observed in patients with ectodermal dysplasia. The signs of eye dryness in patients with ectodermal dysplasia were supported by the impression cytology technique. Impression cytology can be used as a non-invasive method in ophthalmologic follow-up of patients with ectodermal dysplasia. (*Turk Arch Ped* 2012; 47: 72-5)

Key words: Conjunctiva, ectodermal dysplasia, Impression cytology

Introduction

Ectodermal dysplasia is a group of diseases characterized by abnormalities of the epidermis and its appendages and lack of sweat glands or more frequently decrease in sweat glands which affects tissues and organs of ectodermal origin (1). The most common type is hypo- or anhydrotic ectodermal dysplasia described by Christ, Siemens and Touraine (2). The main characteristics of this rare autosomal recessive hereditary disease include abnormalities of the epidermis and its appendages (hair, nails, eccrine and sebaceous glands) together with lack of sweat glands or more frequently decrease in sweat glands which leads to increase in body temperature (1). Ocular findings including strabismus, telecanthus, blepharophymosis, entropion, dacriocystitis, blepharitis, conjunctivitis, meibomian gland dysfunction, corneal opacity and cataract may be observed in individuals with ectodermal dysplasia (3-9). Recurring corneal erosions, opacifications, vascularization and perforations affect various ocular surface disorders. However, the reason of ocular surface disorders in individuals with ectodermal dysplasia is not

known exactly (6,7). To prevent corneal damage these patients should be followed up with artificial tear.

Impression cytology is a rapid, convenient, economical and noninvasive technique which is used for the diagnosis of ophthalmologic disorders. Conjunctival impression cytology which is performed using cellulose acetate filter papers on the superficial epithelium of the eye has no side effects or contraindications (10-12). In this study, it was aimed to discuss the characteristics of ectodermal dysplasia syndrome which is observed rarely with three cases and to find conjunctival involvement in these cases using impression cytology which is a non-invasive method.

Cases

Six eyes of three patients with ectodermal dysplasia who were followed up in Dicle University Medical Faculty, Departments of Ophthalmology and Prosthetic Dentistry were included in the study. Impression cytology technique was performed after obtaining informed consent from the subjects.

The samples were taken onto a cellulose acetate filter paper with a pore diameter of 0,20 µm (Sartorius, 11107-50-N) following local anesthesia provided by dropping 0,4% "oxibupracaine hydrochlorid" into both eyes. Cellulose acetate filter papers cut as rectangles of 3 x 4 cm were impressed slightly for 3-4 seconds on the upper bulbar conjunctiva at 12 o'clock position with the lower edge 2 mm away from the limbus with the help of a smooth forceps and with the dull surface touching the conjunctiva. The filter paper was taken away slowly from the conjunctiva. The wetted filter papers which carried conjunctival epithelial samples at the cytological level were placed in vials containing a mixture of 70% ethyl alcohol, 37% formaldehyde and glacial acetic acid with a ratio of 20:1:1 for staining with the cell samples looking upwars with the aim of fixing and were kept at +4°C in the refrigerator to prevent evaporation of alcohol (10-12). After the samples were transferred from vials into cachets, they were stained with periodic acid schiff (PAS) and hematoxylin eosin (H&E). The samples were examined with Olympus BH-2 Light Microscope.

Results

Case 1

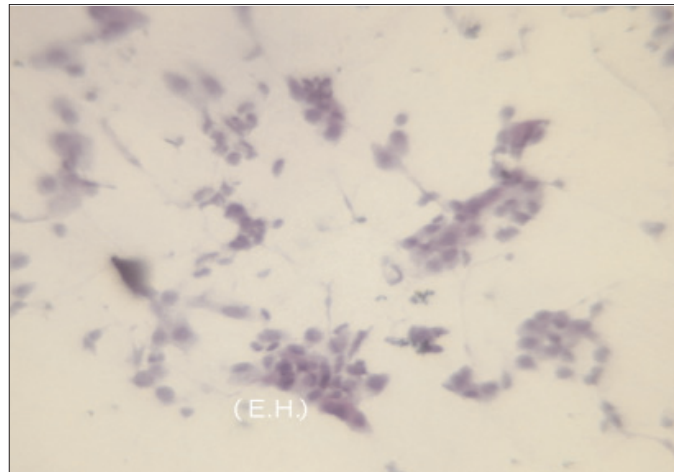
A 6-year-old boy presented to our hospital because of hair loss, skin dryness and dental problems. He had seven siblings from the consanguineous marriage (second degree) of his parents. His two brothers had ectodermal dysplasia (our cases number 2 and 3). His two sisters and two brothers were healthy. On physical examination of our case with ectodermal dysplasia the forehead was prominent, the hair were scarce, light-colored and dry, the eyelashes and eyebrows were scarce and a saddle nose and outward and thick lips were present. Oral examination revealed enamel hypoplasia in the teeth and hypodontia. Examination of the skin revealed severe hypohydrosis, dry skin and thermoregulation problem. Complete blood count and biochemical values were found to be normal.

Case 2

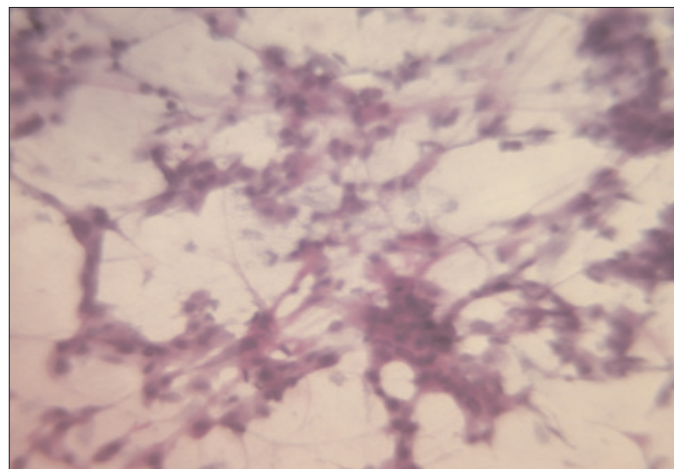
A 14 year-old boy had marked loss of hair, eyelashes and eyebrows, old appearance, a flattened nose root, outward and thick lips, dry mouth, severe hypodontia in the teeth on physical examination. Examination of the skin revealed hypohydrosis, dryness and hyperkeratosis in the palms and thin and bridged hand and foot nails. Complete blood count and biochemical values were found to be normal.

Case 3

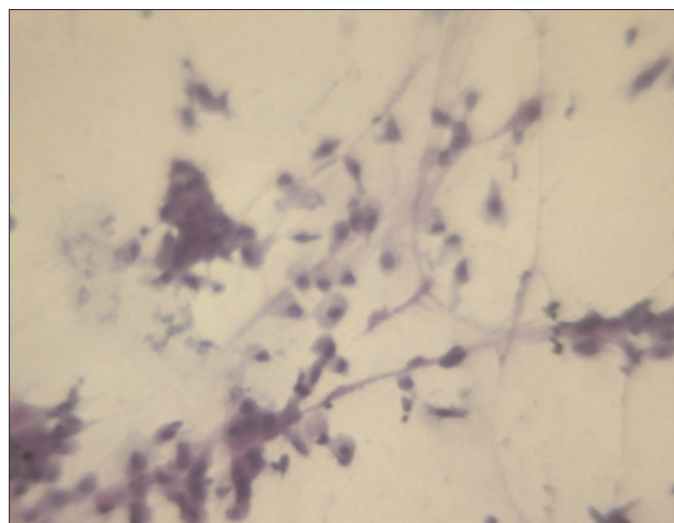
This 18 year-old boy was the first sibling with ectodermal dysplasia. On physical examination of this case, more prominent



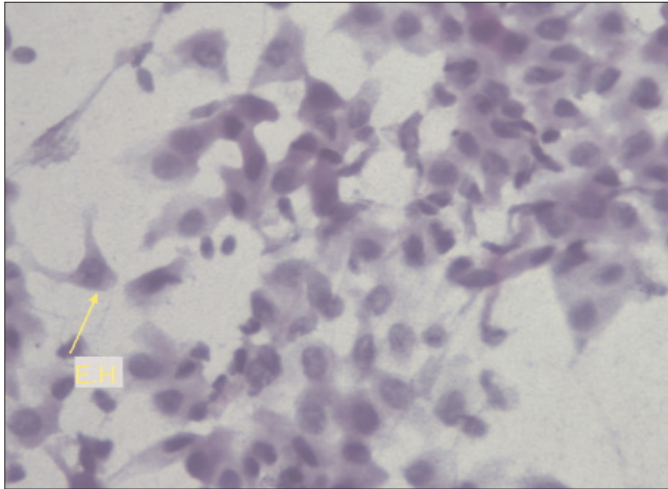
Picture 1. Conjunctival impression cytology (Case 1). E.H: Epithelial cell (PAS-Hematoxylin-original magnification X 82).



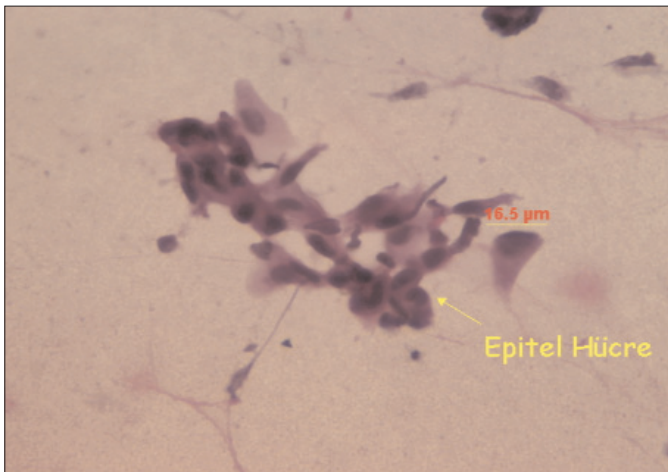
Picture 2. Conjunctival impression cytology (Case 1) (PAS-Hematoxylin-original magnification X 164)



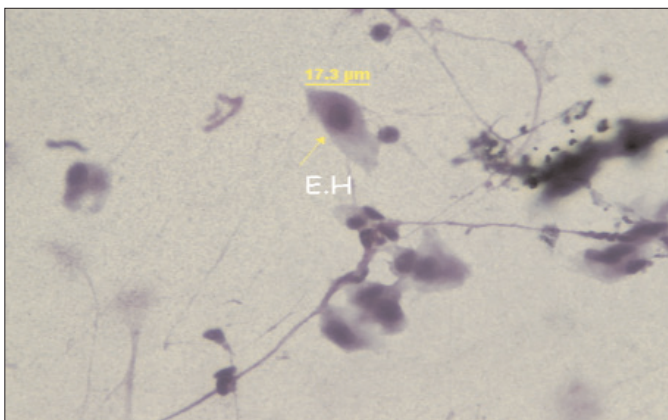
Picture 3. Conjunctival impression cytology (Case 2). (PAS-Hematoxylin-original magnification X 82)



Picture 4. Konjunktival impression cytology (Case 2). E.H: Epithelial cell (PAS-Hematoxylin-original magnificationX164).



Picture 5. Konjunktival impression cytology (Case 3) (PAS-Hematoxylin-original magnification X82).



Picture 6. Konjunktival impression cytology (Case 3). E.H: Epithelial cell (PAS-Hematoxylin–original magnificationX164).

loss of hair, eyelashes and eyebrows was found. Marked enamel hypoplasia and hypodontia were observed on oral examination and severe hypohydrosis, dry skin and marked thermoregulation problem were found on cutaneous examination. Complete blood count and biochemical values were found to be normal.

Conjunctival impression cytology samples of our cases are shown in Pictures 1-6. Generally, decrease in the intensity of epithelial cells, anisocytosis in the epithelial cells together with change of nucleus/cytoplasm ratio in favor of the nucleus were found. Observation of single nuclei in some cells and multiple nuclei with polygonal and fusiform shapes in some cells was significant in terms of cellular shape differences. Lack of goblet cells among epithelial cells was considered significant in terms of progression to metaplasia.

Discussion

In ectodermal dysplasia which is a group of disorders affecting tissues and organs of ectodermal origin during embryonal development, the hair, teeth, nails, sweat glands and eyes are affected (1). In cases with ectodermal dysplasia, ocular pathologies reported include loss of meibomian glands, poliosis, hypoplasia of the nasolacrimal canal, keratopathy, ankioblepharon and lid hypoplasia (3-8).

In cases with ectodermal dysplasia, the etiology of ocular superficial diseases is not clear (7). Infections including chronic conjunctivitis and dacryocystitis, decreased function or complete loss of function of the lacrimal glands and meibomian glands, possible primary developmental disorder of the corneal epithelium during embriogenesis are among the blamed factors (13-15).

Ota et al. (7) emphasized that lack of meibomian glands and imbalance of tear release would lead to epithelial problems on the ocular surface together with abnormalities in the lipid layer.

Cytological examination is significant in the diagnosis of conjunctival diseases. Appropriate samples should be taken from the ocular surface for a perfect cytological examination. Cytologic samples can be taken using scraping method, pipette method, spreading method with cotton-tipped applicator and impression cytology. Conjunctival scraping method is an invasive method and its use in the young age group is rather difficult and risky (16-18). Pipette method is rather difficult and requires experience. Mostly, shed dead cells are taken with this method and the results are usually inadequate, since the localization can not be determined. In the method of spreading with cotton-tipped applicator, the cells are lost in the cotton at the end of the applicator and adequate details can not be displayed (18,19). Impression cytology which was described as simple conjunctival biopsy in 1977 for the first time by Egbert (11) and Thatcher (20) has been reported to be a noninvasive and safe method which can be used in the diagnosis and follow-up of changes at the cellular level in the epithelium of the conjunctiva which develop due to

various pathologies. Cytologic impression of the conjunctiva is based on obtaining epithelial samples by sticking a cellulose acetate filter paper on the surface of the epithelium for the aim of examining the superficial layers of the conjunctiva at the cytologic level. Since this method gives information about the epithelial morphology of the conjunctiva, the cytoplasm/nucleus ratio, cellular morphology and especially intensity of goblet cells, it has been emphasized that it is significant in studying ophtalmologic diseases (11,18,20).

In the literature, no study related to conjunctival impression cytology in cases with ectodermal dysplasia was found. In our study, histologic findings due to eye dryness were found using impression cytology technique in cases with ectodermal dysplasia. Lack of goblet cells which help release of tear also supported this view. We believe our findings will strenghten in larger series with increased number of subjects. With this view we can conclude that impression cytology can be used as a noninvasive method in the ophtalmological follow-up of ectodermal dysplasia.

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