

Fine-needle aspiration cytology and scrape cytology of conjunctival squamous cell carcinoma: Report of a rare case

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ABSTRACT

The utility of fine-needle aspiration cytology (FNAC) in the diagnosis of conjunctival tumor is not well-described in the literature. Here, we report a case of conjunctival squamous cell carcinoma (SCC) of the left eye in a 47 year old man. The case was initially diagnosed by FNAC using 24-gauge needle fitted in a 10 ml syringe. Scrape cytology was also done after FNAC. Smears from fine needle aspirates show malignant squamous epithelial cells arranged in aggregates. Intracellular keratin was discernible. Scrape cytology showed similar type of cytomorphological features. A cytodiagnosis of ocular surface squamous neoplasia favoring SCC was made and biopsy was suggested for confirmatory diagnosis. Surgical excision of the tumor was done and histopathological examination confirmed the diagnosis of SCC.

Key words: Conjunctiva, fine-needle aspiration cytology, scrape cytology, squamous cell carcinoma

INTRODUCTION

Conjunctival squamous cell carcinoma (SCC) is uncommon worldwide, and the incidence varies geographically from 0.2 to 3.5/100,000.^[1] The term ocular surface squamous neoplasia (OSSN) was introduced to encompass the spectrum of conjunctival intraepithelial neoplasia and SCC.^[2] Clinically, it is difficult to distinguish dysplasia, carcinoma *in situ* and SCC. Macroscopic patterns of SCC, include gelatinous, papilliform or velvety, nodular, leukoplakic or diffuse.^[2]

Fine-needle aspiration cytology (FNAC) is useful in diagnosing SCC and other tumors of the conjunctiva. It may be used as a guide for performing a biopsy.^[3] But observers experience is needed for cytodiagnosis.^[3] For impression cytology cellulose acetate was used initially which was

modified later on. Recently, Thiel *et al.* have described the use of a biopore membrane device for obtaining conjunctival cells.

In a retrospective analysis of 26 cases of conjunctival SCC, McKelvie, *et al.* found that it occurs in the sun damaged ocular surface, usually at the limbus in the elderly men.^[2]

CASE REPORT

A 47-year-old man presented to the ophthalmology outpatient department with complaints of chronic redness, irritation and photophobia of left eye for 7-8 months. He also noticed a rapidly growing red swelling in the left eye for 3-4 months. On examination, a red-colored nodular mass (2 cm × 1.8 cm) was found in the bulbar conjunctiva (temporal side) near the limbus of left eye. His past medical history was insignificant. Patient was sent to the pathology department for FNAC.

Fine needle aspiration cytology was done using a 24-gauge needle fitted with a 10-ml syringe. Six passes were made and aspirates spreaded over glass slides. Scrape smears were also made after FNAC using a scalpel blade with great caution. Multiple slides were prepared for alcohol fixed Papanicolaou (Pap) stain and air-dried may grunwald

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giemsa stain. Smears from the fine-needle aspirates show malignant squamous epithelial cells, mostly arranged in aggregates though few discretely arranged cells were also found. These cells have anisonucleosis and hyperchromatic nuclei. Intracellular keratin was discernible. Smears prepared from scrapes showed similar cytomorphological features [Figure 1a-c]. Based on cytomorphology a provisional diagnosis of OSSN, favoring SCC was made. Histopathological examination was suggested for correlation with cytologic diagnosis and confirmatory diagnosis of this particular case [Figure 1d]. Immunohistochemistry (IHC) showed pan-cytokeratin, epithelial membrane antigen positivity among the tumor cells. Ki-67 index was 24%. Thus, IHC analysis also supported the cytological and histological diagnosis.

The conjunctival mass was excised surgically and the specimen was sent for histopathologic examination. Microscopic examination revealed malignant squamous epithelial cells arranged in sheets and infiltrated the subepithelial layer after rupturing basement membrane. Focal necrosis was also noted. Hence, a final diagnosis of SCC was established.

DISCUSSION

Ocular surface impression cytology was first introduced into ophthalmology by Egbert *et al.* in 1977. These authors used cellulose acetate filter paper for collection of conjunctival superficial layer of cells. Several authors modified this technique and used it for investigation of dry eyes, diagnosis of vitamin A deficiency, staging of conjunctival squamous metaplasia etc., Nolan *et al.* first reported the application of impression cytology for diagnosis of conjunctival neoplasm.^[4] Later on, Thiel *et al.* introduced use of a new biospore membrane device for obtaining conjunctival cells to diagnose superficial viral infections.^[5] This biopore membrane remains transparent in wet state that allows for a detailed cytological examination.^[6] Subsequently biospore membrane was used to diagnose different conjunctival lesions, including OSSN and SCC.^[7]

Till date, there are only few published articles in the literature regarding FNAC as a diagnostic tool of conjunctival neoplasms. Grossniklaus *et al.* opined aspiration cytology of the conjunctival surface was useful in obtaining diagnostic material. It might be used as a guide to where a biopsy should be performed.^[3] In our case, we also obtained good diagnostic material through fine needle aspirates. Moreover, patient did not have any complication after the procedure. This might be due to fact that we used 24-gauge needle instead of conventional 21-22 gauge needle. Scrape material also yielded good diagnostic material in the present case. Ersöz *et al.* suggested that both impression and brush

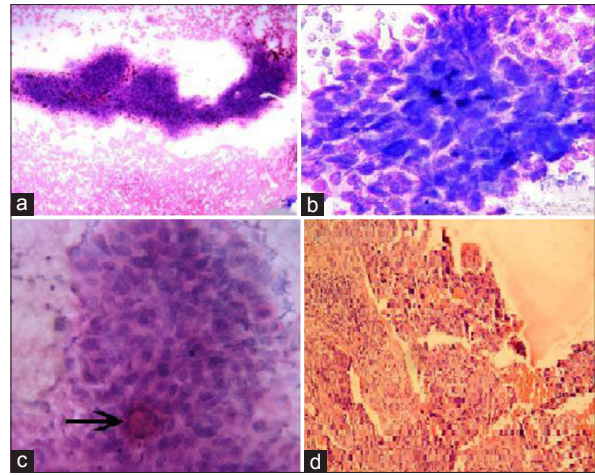


Figure 1: (a) Scrape cytology showing clusters of malignant squamous epithelial cells. (Papanicolaou [Pap], ×100). (b) Fine needle aspiration cytology (FNAC) showing malignant squamous epithelial cells having nuclear overlapping, anisonucleosis and prominent nucleoli. (May Grunwald Giemsa, ×400). (c) FNAC showing malignant squamous epithelial cells having hyperchromatic nuclei with conspicuous nucleoli. One keratin pearl formation is also noted (marked by arrow) (Pap, ×400). (d) Histology showing malignant squamous epithelial cells invading the basement membrane and are arranged in sheets (Hematoxylin and Eosin, ×100)

cytology are fast, cost-effective, reliable and noninvasive diagnostic tools for OSSN. However, they observed the brush technique had several advantages over impression cytology.^[8]

A prospective study on 31 conjunctival lesions was performed by Lima *et al.* They used Kimura spatula for sample collection.^[9] The Pap stain was used for cytologic examination and histologic examination was done to compare cytodiagnosis. They reported high specificity (100%) and sensitivity (92.9%). The positive predictive value was 100% and the negative was 94.4%.^[9] In their study, Tananuvat *et al.* found the positive and negative predictive accuracy of impression cytology was 97.4% and 52.9%, respectively when cytologic findings were compared with histologic findings.^[7] They observed SCC had the highest correlation with histology for squamous neoplasm.^[7] In another study, 91% concordance on cytologic smears and 98% concordance in subsequent biopsies were reported as no dysplasia versus any degree of dysplasia.^[10]

The transformation of the stratified epithelium of conjunctiva into SCC as a result of mechanical irritation like poorly fitting prosthesis has been described. However in our case, there was no such medical history.

Differentiating *in situ* carcinoma from infiltrating carcinoma is difficult in impression cytology.^[11] Although the presence of prominent nucleoli has been suggested as a feature of invasive carcinoma, but it is not always reliable.^[10] Some authors described the presence of a tumor diathesis (blood and necrotic debris) is suggestive of an invasive lesion

as claimed by Midena and colleagues. But with impression cytology, which only samples the surface/superficial layer of conjunctival epithelium, such tumor diathesis is usually absent.^[2,11] Perhaps scrape cytology and FNAC are superior to impression cytology in this regard as if sample the deeper layer of conjunctiva.

CONCLUSION

To conclude, FNAC can be employed for quick and early diagnosis of conjunctival SCC. But caution should be taken to avoid complications, and it should be done only by experienced hands. Final diagnosis of SCC should be rendered only after clinical correlation and histologic examination.

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