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Cell Block and Its Impact in the Diagnosis of Jaw Lesions over Fine Needle Aspiration Cytology

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Keywords
Cell block · Diagnosis · Fine needle aspiration cytology · Jaw lesions

Abstract
Objectives: To determine the role and efficacy of fine needle aspiration cytology (FNAC) and cell block in diagnosis of jaw lesions and compare the agreement between FNAC and cell block to predict the diagnosis. ***Method:*** The sample comprised 51 cases, including 12 odontogenic keratocysts (OKCs), 8 ameloblastomas, 22 radicular cysts, 7 dentigerous cysts, and 1 each of intraosseous mucoepidermoid carcinoma (MEC) and adenomatoid odontogenic tumor (AOT). FNAC samples remaining after hematoxylin and eosin (H&E)-stained cytosmear diagnosis were centrifuged at 3,000 rpm for 10 min. The supernatant was discarded and sediment mixed with 2–3 mL alcohol and filtered. To this, 10% formalin was added, filtered, taken for routine processing, and stained with H&E. The result of FNAC smear and cell block was compared with histopathological diagnosis. ***Results:*** On cytological examination of the smears, 7 OKCs and 22 radicular cysts were diagnosed, whereas ameloblastomas, AOT, intraosseous MEC, and dentigerous cysts were not. This gave an agreement of 56.8% with the biopsy reports. Cell block sections stained with H&E of 12 OKCs, 22 radicular cysts, 1 MEC, and 3 cases of ameloblastoma offered a diagnosis in accordance with the biopsies giving an agreement of 74.5%, while dentigerous cyst and AOT failed to do so. In comparison with FNAC, additionally 5 cases of OKC and 1 of MEC could be detected, and in ameloblastoma, out of 8 cases, only 3 yielded a concordant diagnosis through the cell block technique. ***Conclusion:*** In comparison with FNAC, the architectural pattern and the morphology of the cells were better preserved by the cell block technique. This substantiates that cell block could be used as an ancillary technique to aid in definitive diagnosis of head and neck swellings.

Introduction

The lesions of the jaw could be odontogenic cyst and tumors, neoplasms of salivary gland origin, metastatic lesions, and nonodontogenic lesions. Their early diagnosis is important to provide appropriate treatment [1]. In this regard, a diagnostic tool often used is the fine needle aspiration cytology (FNAC) but has a low accuracy of diagno-

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Table 1. Clinical details of all cases

Lesions	Cases, *n*	Age, years	Gender	
			female	male
Odontogenic cysts				
OKC	12	11–68	4	8
Dentigerous cyst	7	5–22	2	5
Radicular cyst	22	16–70	10	12
Odontogenic tumors				
Ameloblastoma	8	12–35	3	5
AOT	1	17	0	1
Salivary gland tumor				
MEC	1	16	0	1
Total	51		19	32

OKC, odontogenic keratocyst; MEC, mucoepidermoid carcinoma; AOT, adenomatoid odontogenic tumor.

sis due to lack of architectural context of FNAC material, nonrepresentative sampling, and nonspecific morphological features on cytosmear [2, 3]. These drawbacks of FNAC could be overcome by use of cell block preparation [4], for which, the FNAC material is fixed, centrifuged, and the cell pellet is transferred for paraffin embedding [5]. The cell block while maintaining the cellular architecture also provides multiple additional sections for special stains, immunostaining, ultrastructural analysis, and molecular testing [6]. Thus, cell blocks act as useful adjuncts to smears, especially in categorization of tumors, and a more definitive cytopathologic diagnosis can be drawn [1, 4]. There are many studies done to compare the usefulness of cell blocks with that of smears from fine needle aspiration material, but only a few in case of jaw lesions [1, 4, 7, 8].

In this context, the present study was undertaken to assess the utility of the cell block preparation method in increasing the sensitivity of cytodiagnosis of jaw lesions. We tried to evaluate the cellularity, architectural patterns, and morphological details both in the cytological smear and the section from the cell block method. Further, we also compared the cell block findings with biopsy diagnosis, and the accuracy of cell block in the diagnosis of various jaw lesions over cytopathology was assessed.

Materials and Methods

After approval from the Institutional Review Board of SDM College of Dental Sciences and Hospital, Dharwad (IRB No. 2017/S/OP/52), a total of 51 cases of jaw lesions (Table 1) were studied by cytopathology, cell block sections prepared from the residual FNAC material, and the subsequent incisional or excisional biopsy. The cases of jaw lesions diagnosed provisionally based on the clinical and radiographic features with FNAC and biopsy were included in the study. Both FNAC and biopsy were performed by oral surgeons. All the lesions were intraosseous, and a 5-mL syringe was used for FNAC. The cases having sufficient samples to prepare cell block and cases with and without recurrence were included. Nearly 10–12% of the samples with <1 mL of the aspirate were considered inadequate because the fluid left after smear preparation was insufficient to be centrifuged for cell block preparation. Also, the samples with only blood, pus, etc., were excluded from the study.

The clinical details including age, sex, site, clinical presentation, radiographic findings, and description of the FNAC sample were noted. From the FNAC sample, 2 cytosmears were prepared and stained with hematoxylin and eosin (H&E), following the diagnosis. The remaining sample was used for cell block preparation.

For FNAC smear, the smears were alcohol-fixed and stained with hematoxylin and eosin. For cell block preparation, the alcohol and formalin-fixed method was used. The FNAC fluid was centrifuged at 3,000 rpm for 10 min, the supernatant was discarded, and the sediment was mixed with alcohol (2–3 mL), filtered, and mixed in 4–5 mL of 10% formalin. This was again filtered using filter paper, and the sediment was taken for routine processing.

The biopsy specimens received were routinely processed and stained with H&E. The results of the FNAC smear, cell block, and histopathological data of the biopsy as well as the clinical and radiographic data of the lesions were analyzed.

Results

In the present study, 51 cases included 12 odontogenic keratocysts (OKCs), 7 dentigerous cysts, 22 radicular cysts, 8 ameloblastomas, and 1 case each of adenomatoid odontogenic tumor (AOT) and intraosseous mucoepidermoid carcinoma (MEC). The age of the patients ranged between 5 and 70 years with a mean age of 32. Of the 51 cases, 19 were females and 32 males (Table 1).

The FNAC diagnosis and the cell block results were compared with the histopathologic diagnosis (Table 2). Among the odontogenic cysts, all 22 radicular cysts showed a predominantly chronic inflammatory cell infiltrate in a necrotic background, and 7/12 OKCs showed anucleated epithelial cells and keratin debris with few chronic inflammatory cells as shown in Figures 1a and 2a, respectively, giving a concordant FNAC diagnosis, whereas 5 OKCs and all dentigerous cysts with sparse chronic inflammatory cells as shown in Figure 3a failed to do so. In these cases, a diagnosis of an inflammatory lesion was considered on FNAC. But, when the cell block sections were analyzed, all 22 radicular cysts revealing chronic inflammatory cells arranged in a compact manner with areas of cholesterol cleft formation and 12 OKCs with abundant keratin flakes and few nuclei as shown in Fig-

Table 2. Comparison of FNAC and cell block diagnosis with the histopathological diagnosis

Lesions	Histopathological diagnosis, *n*	FNAC diagnosis		Cell block diagnosis	
		concordant, *n* (%)	nonconcordant, *n*	concordant, *n* (%)	nonconcordant, *n*
Odontogenic cysts					
OKC	12	7 (58)	5	12 (100)	0
Dentigerous cyst	7	0 (0)	7	0 (0)	7
Radicular cyst	22	22 (100)	0	22 (100)	0
Odontogenic tumors					
Ameloblastoma	8	0 (0)	8	3 (37.5)	5
AOT	1	0 (0)	1	0 (0)	1
Salivary gland tumor					
MEC	1	0 (0)	1	1 (100)	0
Total	51	29 (56.8)	22 (43.2)	38 (74.5)	13 (25.5)

OKC, odontogenic keratocyst; MEC, mucoepidermoid carcinoma; AOT, adenomatoid odontogenic tumor; FNAC, fine needle aspiration cytology.

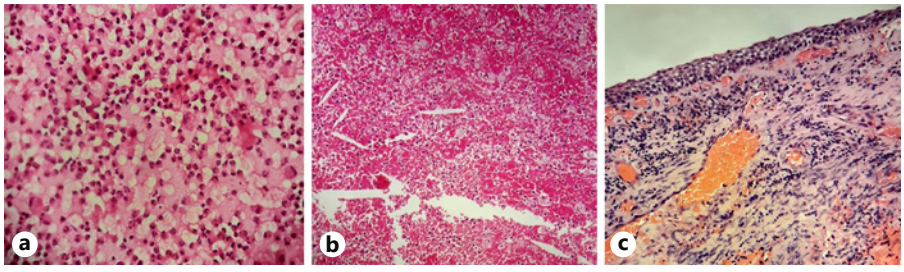

Fig. 1. Photomicrograph of cytosmear showing predominantly chronic inflammatory cell infiltrate in a necrotic background (H&E. ×40) (**a**), cell block section showing chronic inflammatory cells arranged in a compact manner with areas of cholesterol cleft formation (H&E. ×20) (**b**), and tissue section showing cystic lumen lined by nonkeratinized stratified epithelium with subepithelial chronic inflammation in the connective tissue (H&E. ×20) (**c**).

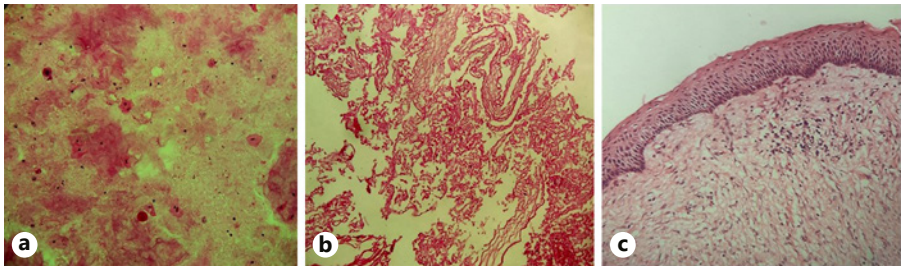

Fig. 2. Photomicrograph of cytosmear showing anucleated epithelial cells and keratin debris with few chronic inflammatory cells (H&E. ×40) (**a**), cell block section with abundant keratin flakes and few nuclei (H&E. ×20) (**b**), and tissue section showing cystic cavity lined by stratified squamous parakeratinized epithelium with surface corrugations. The basal cells have palisaded appearance and hyperchromatic nuclei. The interface between the epithelium and connective tissue capsule is nearly flat (H&E. ×20) (**c**).

ures 1b and 2b, respectively, could be diagnosed. The cytosmears of odontogenic tumor cases showed only inflammatory infiltrate as shown in Figures 4a and 5a, giving a nonconcordant FNAC diagnosis. However, 3 ameloblastomas detected by the cell block method as shown in Figure 4b revealed peripheral clumps of cuboidal to columnar cells with few stellate reticulum-like cells in a fibrous stroma having sparse chronic inflammatory cells, giving a diagnosis in accordance with that of the histopathological diagnosis. Cytosmear and cell block sections of AOT cases consisted of chronic inflammatory cell infiltrate, chiefly lymphocytes as shown in Figure 5a and b, respectively, giving a nonconcordant diagnosis. On comparing the intraosseous MEC, we noted that it could

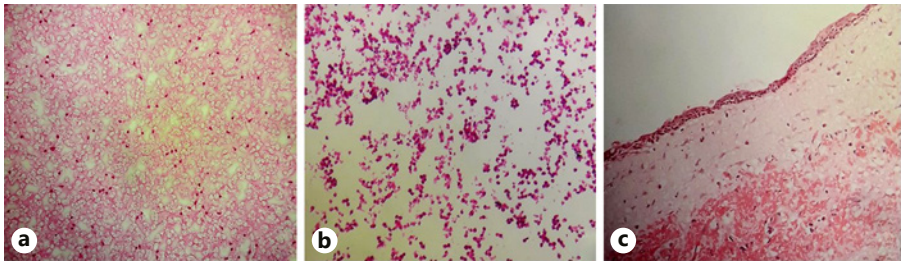

Fig. 3. Photomicrograph of cytosmear (H&E. ×20) (**a**) and cell block section (H&E. ×40) (**b**), showing sparse chronic inflammatory cells, and tissue section showing cystic cavity lined by a thin nonkeratinized stratified squamous odontogenic epithelium. Underlying cystic capsule is fibrous with few inflammatory cells and hemorrhagic areas (H&E. ×20) (**c**).

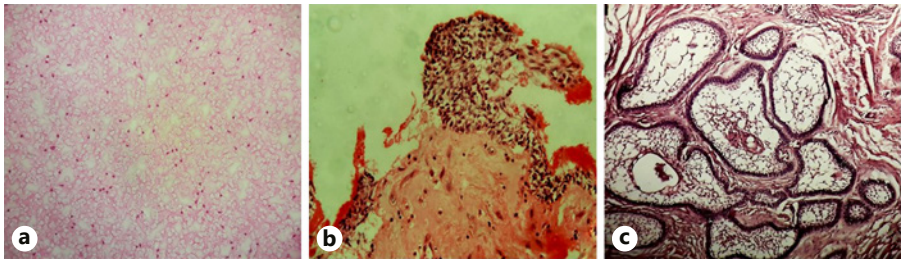

Fig. 4. Photomicrograph of cytosmear showing minimal inflammatory cell infiltrate (H&E. ×40) (**a**), cell block sections showing peripheral clumps of cuboidal to columnar cells with few stellate reticulum-like cells in a fibrous stroma having sparse chronic inflammatory cells (H&E. ×40) (**b**), and tissue section showing the odontogenic epithelium arranged in follicles of varying sizes with peripheral columnar cells with hyperchromatic nuclei exhibiting reversal of polarity and central stellate reticulum cells with squamous metaplasia, keratin formation, and some cystic degeneration. The stroma is fibrous to fibrocellular (H&E. ×40) (**c**).

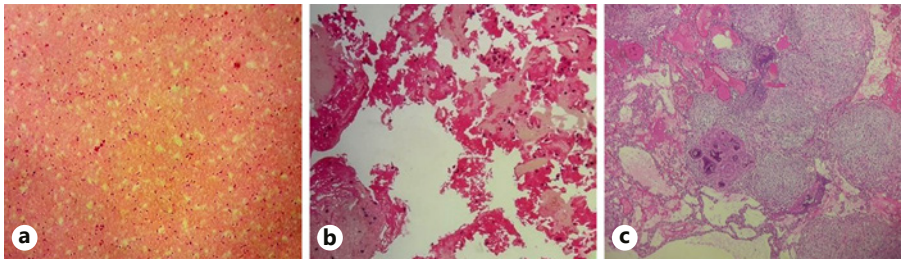

Fig. 5. Photomicrograph of cytosmear (H&E. ×20) (**a**) and cell block sections (H&E. ×20) (**b**) showing chronic inflammatory cell infiltrate, chiefly lymphocytes. **c** Tissue section showing solid areas supported by fibrous connective tissue. The solid component shows the odontogenic epithelium in the form of ducts, whorls, and interlacing pattern with eosinophilic secretion with areas of calcification (H&E. ×20).

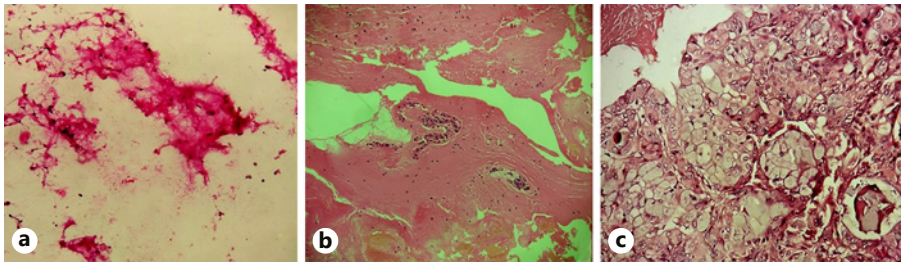

Fig. 6. Photomicrograph of cytosmear showing diffuse inflammatory cells in a mucinous background (H&E. ×20) (**a**) and cell block section showing benign round to ovoid cells in clusters with basophilic nucleus in a mucinous background. Few mucous cells and inflammatory cells are also noted (H&E. ×20) (**b**). **c** Tissue section shows epidermoid-like cells with vesicular and few hyperchromatic nuclei and many mucous like cells with eccentrically placed vesicular nuclei. The connective tissue stroma is densely fibrous (H&E. ×40).

be diagnosed by cell block as shown in Figure 6b due to the presence of benign round to ovoid cells in clusters with basophilic nucleus in a mucinous background and few mucous cells and inflammatory cells. Since the FNAC had only diffuse inflammatory cells in a mucinous background as shown in Figure 6a, the cases were diagnosed as an inflammatory lesion.

These findings aided in arriving at an agreement of 74.5% between the histopathological diagnosis and the cell block method. However, the agreement between the histopathological diagnosis and FNAC was 56.8%.

Discussion

Jaw lesions are among the frequent complaints for which patients consult dentists [9]. While biopsy is the mainstay for precise diagnosis, FNAC is a widely used more conservative alternative [10]. To overcome the disadvantages of FNAC, the cell block technique, introduced by Bahrenburg, has proved to be of great utility, which along with the advantages of FNAC has other advantages like maintaining of cellular architecture because of lesser dispersion of cells and providing multiple sections thus aiding in better diagnosis by means of special stains and immunohistochemistry [5, 8, 10]. The slides obtained from paraffinized cell blocks can be preserved, and the sections are akin to the tissue sections from paraffin blocks, with better cellular details for a definitive diagnosis [4]. There are limited studies comparing the cell block technique with the FNAC in jaw lesions [1, 4, 7, 8]. With an interest to unveil this, we conducted the present study to assess the role of both these techniques in the diagnosis of jaw lesions and compare their positive predictive value in histopathological diagnosis and also correlate the diagnosis with the clinical, radiographic, and histopathological features.

Among the odontogenic cyst, we had cases of radicular cyst, OKC, and dentigerous cyst. Radicular cyst is an odontogenic cyst of inflammatory origin [11]. The cystic fluid is brown in color, odorless, and nonsticky. The age range of our cases was 16–70 years. The cytosmears and cell block sections had predominantly chronic inflammatory cell infiltrate in a necrotic background, and the cells were arranged in a dispersed manner. However, cell block sections revealed cells in a compact manner, and areas of cholesterol cleft formation were also noted. These findings and a clinical and radiographic correlation aided to arrive at a diagnosis of a radicular cyst in all 22 cases and was confirmed with the excisional specimen. In an FNAC study by Goyal et al. [2], it was noted that of the cases considered to be odontogenic cysts by clinical and radiographic findings, 20% cases, and in a study on cell block sections by Rivero et al. [4], it was seen that 53% of the cases had features similar to our findings.

OKC is a developmental odontogenic cyst having a high recurrence [11]. The cystic fluid is creamish with thick cheesy keratin. The cytosmear and cell block sections in the present study showed varied degrees of anucleate epithelial cells and keratin debris with few chronic inflammatory cells. Cell block sections had abundant keratin flakes and some even contained nuclei. Histopathological examination revealed features suggestive of OKC. Of the 12 cases between the age range of 11 and 68 years, 7 FNACs and all cell blocks gave a diagnosis of keratinizing lesion which was in concordance with the histopathologic diagnosis. The other 5 FNAC lacked keratin and could be diagnosed only as an inflammatory lesion. We could diagnose all cases by the cell block technique because for preparing a cell block, the entire sample is used; however, for cytosmear preparation, only few drops of fluid is used. The findings of our study were in accordance with the study by Oenning et al. [7] where 33% cell block sections had similar findings. Pallavi et al. [8] compared FNAC and cell block findings with the histopathological findings. The authors noted that of the 7 OKCs, cell block sections of 5 cases showed keratin and epithelial cells with or without inflammatory cells giving a positive cell block diagnosis. The cytopathological features of these 5 cases consisted of mixed inflammatory cells and hemorrhagic areas.

Dentigerous cyst is an odontogenic cyst enclosing crown of an unerupted tooth at the cementoenamel junction [11]. The cystic fluid is thin and straw colored. The present study revealed sparse chronic inflammatory cells in cytosmear and cell block sections. Histopathology showed all features of the dentigerous cyst. Our study consisted of 7 cases within the age range of 5–22 years but all failed to be diagnosed as a dentigerous cyst both by FNAC and cell block. Nevertheless, the absence of exfoliated epithelial cells and secretory material helped us, as a diagnosis of exclusion. On the basis of this along with a correlation of clinical and radiographic features and the histopathology, a dentigerous cyst was considered. In a study by Pallavi et al. [8], 2 out of 3 dentigerous cysts could be diagnosed by the cell block technique due to the presence of epithelial cells and inflammatory cells. However, FNAC had only mixed inflammatory cells. In our cases, though the dentigerous cyst could not be diagnosed, it could be used as a diagnosis of exclusion especially in cases of follicular variant of OKC.

We had 9 cases of odontogenic tumors which included ameloblastoma and AOT. Ameloblastoma is an epithelial tumor of odontogenic origin [12]. The fluid consists of a yellow straw colored fluid with or without pungent odor. The cytosmears showed minimal inflammatory cell infiltrate. Cell block sections showed peripheral clumps of cuboidal to columnar cells with few stellate reticulum cells in a fibrous stroma. Histopathological examination confirmed the diagnosis of ameloblastoma. Among the 8 cases between the age of 12 and 35 years, on correlating with the histopathological diagnosis, none showed features of ameloblastoma with FNAC, while 3 cell block sections gave a concordant diagnosis. The other cases only had features of inflammation both in FNAC and cell block. The 3 cases diagnosed using cell block sections show that the amount of material examined in cell block could be more representative, aiding in better diagnosis. In contrast to our findings, a study by Kaliamoorthy et al. [12] showed that 50% of FNAC smears were reported as ameloblastoma with features such as cohesive cluster of basaloid epithelial cells with peripheral palisading and polygonal squamous cells with dense cytoplasm. A study by Belatto et al. [1] on 9 cell blocks of ameloblastoma had features similar to our findings, and the presence of epithelial cells was further confirmed by immunohistochemistry. AOT is a benign slow-growing odontogenic tumor. Of the 3 variants, the follicular AOT is frequently mistaken for a dentigerous cyst [13]. The 1 case of AOT in the present study failed to be diagnosed both by FNAC and cell block. The aspirate was blood tinged, odorless, and nonsticky. Cytology and cell block examination revealed only chronic inflammatory cells. The histopathologic features were suggestive of AOT. Epari and Hanumanthu [9] reported 3 cases of AOT falsely diagnosed as ameloblastoma (1 case) and odontogenic cyst (2 cases) on FNAC. The histopathology showed cuboidal to columnar cells in sheets with some degree of peripheral palisading in some clusters indicative of AOT. Pati et al. [14] reported of a case of extrafollicular AOT of the anterior maxilla. FNAC revealed clusters and sheets of basaloid cells having scanty to moderate cytoplasm with round-oval benign nuclei, fine chromatin, and indistinct nucleoli in a mucoid matrix and peripheral palisading suggestive of a benign neoplasm probably ameloblastoma or AOT or basal cell adenoma. The excisional biopsy confirmed the diagnosis of extrafollicular type of AOT.

The only salivary gland tumor included in the study was an intraosseous MEC. MEC accounts for 5–10% of all salivary gland neoplasms and majorly involves the parotid gland. The low-grade MEC which consists more of cystic lesions poses a difficulty in arriving at a cytological diagnosis because of failure in obtaining diagnostic material [15]. In the present study, the cytosmear of intraosseous MEC revealed mucinous material with inflammatory cells failing to arrive at a diagnosis of MEC. But, the cell block section could be diagnosed as intraosseous MEC, with benign round to ovoid cells with basophilic nucleus. The cells were in clusters in a mucinous background. Few inflammatory cells were also seen. Epari and Hanumanthu [9] got a consistent MEC diagnosis with FNAC in all 3 cases. In contrast, Joseph et al. [15] on studying the diagnostic challenges in cytology of MEC in 6 cases found a definite cytological diagnosis only in 2 cases. Among the rest, 2 were broadly diagnosed as neoplasm with cystic degeneration and 2 were underdiagnosed as pleomorphic adenoma. Supreetha et al. [16] discussed 5 salivary gland lesions where cytological diagnosis was missed. In 1 case, the patient who previously had been treated for MEC presented with nodular growth in the parotid region. Aspirate consisted of a thick abundant mucoid material with numerous cyst macrophages. Cytosmear had no malignant cells and hence was not diagnosed as MEC on FNAC. A cell block from residual material too did not reveal any tumor cells. But, on histopathology, there were small foci of low-grade MEC which was missed on cytology. In another case, a 60-year-old male patient complained of a swelling on the left cheek. Monomorphic epithelial cells arranged in acinar patterns with scant inflammatory infiltrate suggested a diagnosis of monomorphic adenoma. While on histopathology, presence of sheets of epidermoid cells, intermediate cells, and mucous cells suggested MEC. The authors inferred that since diagnosis of dysplasia was missed on aspirated cells, it resulted in a wrong diagnosis of monomorphic adenoma.

The agreement between the histopathological diagnosis and cell block (74.5%) was more as compared to that of FNAC diagnosis (56.8%). This indicates the cell block sections definitely provide with more details and aid in maintaining cell architecture. The cases which could not be diagnosed by cell block and FNAC were because of paucicellularity, secondary infection, blood mixed aspirate, and nonrepresentative sampling. Also, in cases where the aspirated material was minimal, there was insufficient material for cell block preparation. We observed that when more amount of fluid is aspirated, the cell yield is better resulting in diagnostic accuracy. A back and forth motion in different directions during needle aspiration may further increase the yield. As seen in our findings, in case of odontogenic cysts, the accuracy with cell block is much higher as compared to FNAC diagnosis. Considering the correctness of the cell block method

in diagnosis, treatment can be performed without any need of an incisional biopsy. But, owing to the need for laboratory procedures, the cell block technique is time consuming, and material may be lost during processing. Thus, the findings of the present study suggest that the cell block method can be used as a valuable preoperative diagnostic method along with the clinical and radiographic details and as an ancillary technique to FNAC.

Conclusion

We found the positive predictive value of cell block to be better in terms of maintaining the architectural pattern and the morphology of the cells. The technique was especially useful in diagnosing cases of OKC, which needs to be distinguished from other cysts at the earliest because of its high recurrence rate and in at least a few cases of ameloblastoma as compared to the FNAC. Thus, the cell block technique can be efficiently used as an adjunctive technique to aid in establishing a diagnosis for therapeutic planning.

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Statement of Ethics

The study has been approved by the Institutional Review Board of SDM College of Dental Sciences and Hospital, Dharwad (IRB No. 2017/S/OP/52).

Conflict of Interest Statement

The authors have no conflicts of interest to declare.

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Author Contributions

K.H. and B.B. proposed the study concept, and K.H. designed the study. All authors defined the intellectual content and examined the cases clinically; B.B., A.S., and R.M. collected the data and carried out literature search. All authors performed the study and analyzed the results. Statistical analysis was done by K.H. and B.B. Manuscript was prepared and reviewed by all the authors. K.H. edited the manuscript and will act as the guarantor.

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