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Cytomorphology and diagnostic pitfalls of sebaceous and nonsebaceous salivary gland lymphadenoma: A multi-institutional study

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Abstract

Background: Salivary gland lymphadenoma (LAD) is a rare benign neoplasm comprising sebaceous (SLAD) and nonsebaceous (NSLAD) types. Despite established histologic criteria, limited data on cytomorphology, tumor heterogeneity, and overlap with other entities make the diagnosis of LAD by fine needle aspiration (FNA) challenging. We describe a multi-institutional cohort of 14 LADs with cytology, clinical, radiologic, and histopathologic data.

Methods: Our cohort included nine SLAD and five NSLAD with corresponding histopathology. Mean patient age and M:F ratio were 60.4 years (range 45-86 years) and 1:2 for SLADs and 57.4 years (range 42-80 years) and 1:1.5 for NSLADs, respectively. One NSLAD patient had a germline predisposition for Cowden syndrome. Glass slides and whole slide images of air-dried Diff-Quik (DQ), alcohol-stained Papanicolaou smears (Pap) and cellblocks were reviewed for key cytomorphologic findings.

Results: FNAs from SLAD and NSLADs demonstrated vacuolated and basaloid epithelial clusters within a lymphoid background. Vacuolated cells from SLAD showed sebaceous cells with microvesicular cytoplasm indenting a central nucleus. Vacuolated cells from NSLAD were columnar with eccentric nuclei, corresponding to abluminal glandular cells. SLADs were classified using the Milan System for Reporting Salivary Gland Cytopathology as nondiagnostic (11.1%), nonneoplastic (44.4%), atypia of uncertain significance (AUS) (22.2%), and salivary gland neoplasm of uncertain malignant potential (SUMP) (22.2%). NSLADs were classified as AUS (40%), SUMP (40%) and Benign Neoplasm (20%).

Conclusion: Although rare, knowing the cytologic features of salivary LAD is important to avoid diagnostic pitfalls. Vacuolated cells can be prominent in both SLAD and NSLAD aspirates. Diagnostic issues arise from insufficient sampling of all tumor components leading to marked variation in diagnostic classification of LAD.

KEYWORDS

fine needle aspiration, lymphadenoma, nonsebaceous, salivary, sebaceous

1 | INTRODUCTION

Among head and neck neoplasms, 6.5% arise in the context of a major or minor salivary gland[1]. In the initial preoperative workup of a salivary gland lesion, fine needle aspiration (FNA) remains the current standard of care as it is rapid, cost-effective, and maintains a high diagnostic accuracy[2-5]. In addition, preoperative cytology provides useful information that helps guide the clinical approach to a patient with a salivary gland neoplasm, including the decision for surgery or medical management[3,6]. However, disadvantages of the FNA technique include procurement of samples that lack available tissue architecture for evaluation, may inadequately sample tumors with heterogeneity, and exhibit cytomorphologic overlap among an ever-expanding portfolio of salivary gland lesions[7]. Given these challenges, and in an effort to standardize reporting of FNA cytology among institutions, a six-tiered reporting system known as the Milan System for Reporting Salivary Gland Cytopathology (MSRSGC) was published in 2018, and is gaining widespread acceptance[8].

Among the many primary epithelial salivary gland tumors recognized by the 2016 World Health Organization (WHO), sebaceous and nonsebaceous salivary gland lymphadenoma (LAD) are rare, painless, benign neoplasms that occur more often in older adults, although pediatric cases have been reported[9-11]. Moreover, both LAD subtypes occur predominantly in the parotid gland, with rare cases occurring in the minor salivary glands or in other uncommon locations, such as the submandibular gland and minor salivary glands[12,13]. In resections, both sebaceous and nonsebaceous LAD present as well-demarcated lesions. Histologically, nonsebaceous LAD demonstrates cords, nests, tubules or cystic spaces composed of basaloid, squamous or glandular cells[13,14]. With immunohistochemistry, the basaloid and squamous cell components of LAD demonstrate p63 and cytokeratin 5/6 staining, whereas when present, glandular cells can be highlighted with other cytokeratins. Myoepithelial cells are not typically present[13]. The epithelial clusters in LAD are typically embedded within a lymphoid-rich background and reactive lymphoid follicles may be present. The sebaceous type of LAD shows similar histologic characteristics with a few notable exceptions. First, sebaceous differentiation is exclusively associated with this subtype of salivary gland neoplasm. Second, a foreign body giant cell reaction or granulomatous inflammation towards extravasated sebum is more common with this neoplasm subtype. Malignant transformation has been rarely reported with both subtypes of LAD[13,15].

Despite their characteristic histopathologic features, FNA interpretation of sebaceous and nonsebaceous LAD is challenging for several reasons. Given the intrinsic biphasic nature of LAD[13,15], only certain tumor components may be sampled which can thereby lead to their misclassification using the MSRSGC. Second, the differential diagnosis for salivary gland LAD is broad and includes both benign and malignant entities that can create diagnostic confusion[16,17]. Uncommon presentations of this tumor, such as arising in younger age groups or abnormal location may further compound these diagnostic dilemmas[9,10,12,13]. Most importantly, however, given the rarity of this neoplasm LAD may be overlooked when considering the cytologic differential diagnosis of salivary gland lesions. In addition, the available cytomorphologic data in the literature on salivary LAD are limited and prior studies have primarily been single case reports[16-22]. Herein, we discuss a multi-institutional case series of sebaceous and nonsebaceous LAD describing their cytomorphologic features on FNA, differential diagnosis, and highlight potential pitfalls. To the best of our knowledge, this cohort on LAD represents the largest reported cytologic series.

2 | MATERIALS AND METHODS

A retrospective search of the pathology archives at the Massachusetts general hospital (Boston, MA), University of Michigan (Ann Arbor, Michigan), University of Pittsburgh medical center (Pittsburgh, Pennsylvania), and The Johns Hopkins hospital (Baltimore, Maryland) identified 14 matched FNAs and surgical resections of either sebaceous or nonsebaceous LAD cases. FNA specimens were processed as either air-dried Diff-Quik (DQ) stained, and/or alcohol-fixed Papanicolaou (Pap) stained slides, liquid-based preparations (Surepath or Thin-Prep), and/or hematoxylin and eosin (H&E)-stained cell block (CB) slides. Due to insufficient material, ancillary immunohistochemical stains could not be performed on the CB preparation. Whole slide images (scanned at 40× magnification on an Aperio AT2 scanner, Leica Biosystems) or glass slides were reviewed from each of the four medical institutions by their respective authors (William C. Faquin-MGH, Zahra Maleki-JHH, Richard Cantley-U of M, Liron Pantanowitz-UPMC), and key cytologic features were assessed specifically including (a) lymphoplasmacytic component, (b) sebaceous cells, (c) tingible-body macrophages, and (d) other epithelial components. Representative hematoxylin and eosin stained whole slide images (scanned at 40× magnification on an Aperio AT2 scanner, Leica Biosystems) or glass slides of sebaceous and nonsebaceous LAD resections were examined for established diagnostic criteria[13]. The authors were not blinded to the cytologic or histologic diagnoses. Immunohistochemistry for p63 and cytokeratin 5/6 were performed per manufacturer instructions. Clinical parameters and radiology imaging characteristics when available were obtained from the medical records and correlated with the cytomorphologic findings. FNA diagnoses were categorized using established MSRSGC criteria[8].

3 | RESULTS

3.1 | Clinical findings

Between the four medical institutions, our LAD cohort was comprised of 14 cases and included nine sebaceous LAD (SLAD) and five nonsebaceous LAD (NSLAD) cases, all definitively diagnosed on histologic resection. All cases, regardless of subtype, were unilateral and located in the parotid gland. The mean age of the patients for SLAD was 60.4 years (range 45-86 years) with a 1:2 male to female (M:F) ratio and an average tumor size of 2.4 cm (range 1-5.2 cm). Similarly, for

TABLE 1 Clinicopathologic parameters for the multi-institutional case series of sebaceous and nonsebaceous lymphadenoma

Institution	Case	Age	Gender	Laterality	Salivary gland location	Size (cm)	Imaging findings
Sebaceous lymphadenoma							
UPMC	1	51	F	Left	Parotid	1	NA
UPMC	2	45	F	Left	Parotid	3	NA
JHH	3	86	M	Right	Parotid	5.2	NA
JHH	4	62	M	Left	Parotid	2.5	NA
JHH	5	55	M	Right	Parotid	1	NA
MGH	6	79	F	Left	Parotid	2	MRI: 2 × 1.7 cm mass with differential including benign and malignant categories
MGH	7	50	F	Left	Parotid	2.3	MRI: slightly hypointense rim, round mass with multiple small internal areas of signal dropout
MGH	8	51	F	Right	Parotid	1.5	MRI: mildly T2 intense, mildly enhancing in tail of parotid
MGH	9	65	F	Right	Parotid	1.9	NA
Nonsebaceous lymphadenoma							
U of M	1	48	M	Right	Parotid	3	MRI: heterogenous, hyperenhancing, mixed cystic, and solid
UPMC	2	68	F	Right	Parotid	4.2	NA
UPMC	3	42	M	Left	Parotid	4	NA
JHH	4	80	F	Left	Parotid	3	NA
MGH	5	49	F	Right	Parotid	1	US: hypoechoic solid nodule with mild internal vascularity

Abbreviations: F, female; M, Male; MRI, magnetic resonance imaging; NA, not available; US-ultrasound.

NSLAD, the mean age was 57.4 years (range 42-80 years) with a 1:1.5 M:F ratio and an average tumor size of 3 cm (range 1-4.2 cm). Radiology imaging characteristics were available for 67% (n = 6) of sebaceous LAD and 60% (n = 3) of NSLAD. For SLAD with radiology imaging, 50% (n = 3) were T2 hyperintense whereas 16% (n = 1) were hypointense on magnetic resonance imaging (MRI). For NSLAD with radiology imaging, 67% (n = 2) demonstrated hypoechoic, homogeneous and solid features on ultrasound. On radiology imaging, all cases of SLAD and NSLAD were well-delineated. The clinicopathologic and imaging findings are summarized in Table 1.

The cytomorphologic findings along with the MSRSGC categorization and final surgical resection diagnoses for both SLADs and NSLADs are highlighted in Table 2.

3.2 | Cytologic findings of sebaceous LAD

FNA specimens of SLAD (n = 9) demonstrated low to moderate cellularity. Among the key features assessed, 89% (n = 8) showed a lymphoplasmacytic background, 67% (n = 6) had tingible-body macrophages, and 80% (n = 7) had a nonsebaceous epithelial component present that included either basaloid cells in one case, atypical epithelial clusters in two cases, and either bland epithelium or benign salivary gland elements (acini and ductal cells) in four cases. Only 44.4% (n = 4) demonstrated sebaceous cells on both DQ and Pap stained slides as evidenced by cells with a low nuclear to cytoplasmic ratio and microvesicular foamy cytoplasm indenting a centrally placed small nucleus (Figure 1A-D). Scant sebaceous cells were present in a CB slide from one case. When a basaloid component was present, the cells demonstrated a high nuclear to cytoplasmic ratio, scant cytoplasm, uniform round and regular nuclei with fine chromatin and without cytologic atypia. Using MSRSGC criteria, for SLAD (n = 9), 44.4% (n = 4) were diagnosed as nonneoplastic (NN), 22.2% (n = 2) were labeled as atypia of uncertain significance (AUS), 22.2% (n = 2) were categorized as salivary gland neoplasm of uncertain malignant potential (SUMP), and 11.1% (n = 1) were interpreted as being nondiagnostic.

3.3 | Cytologic findings of nonsebaceous LAD

For NSLADs (n = 5), on DQ and Pap smears the specimen cellularity ranged from paucicellullar to moderately cellular. Among the key cytologic features assessed, 80% (n = 4) showed a lymphoplasmacytic

TABLE 2 Preoperative fine needle aspiration cytology diagnoses, Milan system for reporting salivary gland cytology classification and follow-up surgical resection for sebaceous and nonsebaceous lymphadenomas

Site	Case	FNA Diagnosis	MSRSGC	Lymphoplasmacytic background	Tingible body macrophages	Vacuolated cells	Epithelial component features	Histologic follow-up
Sebaceous Lymphadenoma								
UPMC	1	Negative, few reactive lymphocytes & anucleate squamous cells, foamy cells (possible sebaceous cells) & debris	NN	Yes	No	Yes	NA	Unilocular cystic sebaceous lymphadenoma
UPMC	2	Positive for neoplasm, Neoplasm with basaloid features and lymphocytic background	SUMP	Yes	Yes	Yes	Basaloid epithelial clusters and basement membrane-like matrix	Sebaceous lymphadenoma
JHH	3	Rare clusters of atypical epithelium, lymphocytes and macrophages in a cystic background	AUS	Yes	Yes	No	Atypical epithelium	Sebaceous lymphadenoma
JHH	4	Benign, Epithelial inclusion cyst	NN	No	Yes	No	Bland appearing epithelial cells	Cystic sebaceous lymphadenoma
JHH	5	Polymorphous lymphocytes, ductal and acinar epithelium	NN	Yes	Yes	No	Benign ductal and acinar epithelial cells	Sebaceous lymphadenoma with atypia
MGH	6	Atypical neoplasm; question atypical pleomorphic adenoma	SUMP	Yes, scant	No	Yes	Atypical epithelium with prominent nucleoli	Sebaceous lymphadenoma
MGH	7	Negative; scant benign salivary gland, ductal cells, lymphocytes and macrophages; limited cellularity	AUS	Yes	Yes	Yes	scant benign salivary gland	Sebaceous lymphadenoma
MGH	8	Nondiagnostic; scant fibrous tissue and few lymphocytes and benign salivary gland acinar cells	ND	Yes, rare	No	No	scant benign salivary gland	Sebaceous lymphadenoma
MGH	9	Negative; mixed lymphocytes, histiocytes, blood, possible lymph node	NN	Yes	Yes	No	NA	Sebaceous lymphadenoma
Nonsebaceous lymphadenoma								
U of M	1	SUMP - clear cell features	SUMP	Yes	No	Yes	Abundant vacuolated cytoplasm, mild nuclear atypia, indistinct cell borders, architecture ranging from single cells to small clusters to branching groups	Nonsebaceous lymphadenoma with prominent fine needle aspiration site change
UPMC	2	Atypical cells, predominantly lymphocytes with benign appearing epithelioid cells	AUS	Yes	Yes	No	Bland basaloid epithelial clusters	Nonsebaceous lymphadenoma
UPMC	3	Atypical cells, cystic change with multinucleated giant cells and debris	AUS	Yes	Yes	No	NA	Cystic nonsebaceous lymphadenoma with reactive lymphoid hyperplasia

TABLE 2 (Continued)

Site	Case	FNA Diagnosis	MSRSGC	Lymphoplasmacytic background	Tingible body macrophages	Vacuolated cells	Epithelial component features	Histologic follow-up
JHH	4	SUMP, salivary gland neoplasm with basaloid features	SUMP	No	No	Yes	Bland appearing epithelial cells	Nonsebaceous lymphadenoma
MGH	5	Benign neoplasm; clusters of oval, bland cells differential includes myoepithelioma, pleomorphic adenoma, other	BN	Yes, minimal	No	No	Bland appearing epithelial cells	NA

Abbreviations: AUS, atypia of undetermined significance; BN-Neoplastic, benign; MSRSGC, Milan system for reporting salivary gland cytology; NA, not available; ND, nondiagnostic; NN, nonneoplastic; SUMP, salivary gland neoplasm of uncertain malignant potential.

background, 40% (n = 2) had tingible-body macrophages, 80% (n = 4) had an epithelial component, and 40% (n = 2) demonstrated vacuolated cells. Similar to the SLAD cases, the epithelial component in NSLAD was composed of basaloid tumor clusters with cells exhibiting high nuclear to cytoplasmic ratio, scant dense cytoplasm, uniform round and regular nuclei with fine chromatin and without significant cytologic atypia (Figure 2A-D). Using the MSRSGC, 40% (n = 2) of these cases were categorized as AUS, 40% (n = 2) as SUMP, and 20% (n = 1) as a benign neoplasm.

3.4 | Histopathologic and immunohistochemical findings

Among SLADs, gross examination of a representative case demonstrated a well-circumscribed, pale yellow-tan lesion containing scattered white foci (Figure 3A). Histologic examination of cases demonstrated basaloid epithelial clusters transitioning with sebaceous differentiation within a lymphoid-dense background. Neither the basaloid clusters nor the lymphoid population demonstrated any evidence of cytologic atypia, mitoses or necrosis (Figure 3B). Cystic change with proteinaceous debris was noted in 22.2% (n = 2) of cases.

On resection, for the NSLADs gross examination of a representative case revealed a well-circumscribed, cystic, yellow-tan mass (Figure 4A). On histologic examination of NSLADs there were cords, tubules and epithelial basaloid clusters of tumor cells situated within a lymphoid background. No sebaceous differentiation was present. There was no evidence of cytologic atypia, mitoses or necrosis in any case (Figure 4B). Cystic change was noted in 20% (n = 1) of NSLADs. Immunohistochemistry was available for 20% (n = 1) of NSLADs and showed that the basaloid epithelium was diffusely positive for p63 and cytokeratin 5/6 (Figure 4B).

4 | DISCUSSION

Although both sebaceous and nonsebaceous LADs are benign tumors without any known metastatic potential[13,14], there is an inevitable risk of a false positive cytological diagnosis as a malignant primary lesion such as sebaceous carcinoma or a secondary metastasis, leading to unnecessary surgery or inappropriate medical treatment. Diagnostic challenges arise when all of the cellular components are not represented in a cytologic preparation and/or when pertinent clinical data is unavailable. For example, should LAD occur in a pediatric patient or in an uncommon location such as the upper neck or in minor salivary glands of the oral cavity, it would likely add to the diagnostic dilemma[9,10,12,13]. In fact, based on the published literature, there have been a wide range of interpretations for both SLADs and NSLADs based on preoperative cytology (Tables 3 and 4).

The MSRSGC has structured the uniform reporting of salivary gland lesions into distinct categories, each with an associated risk of malignancy and recommended clinical or surgical management[8]. However, for salivary LAD, the MSRSGC categorization is challenging as it

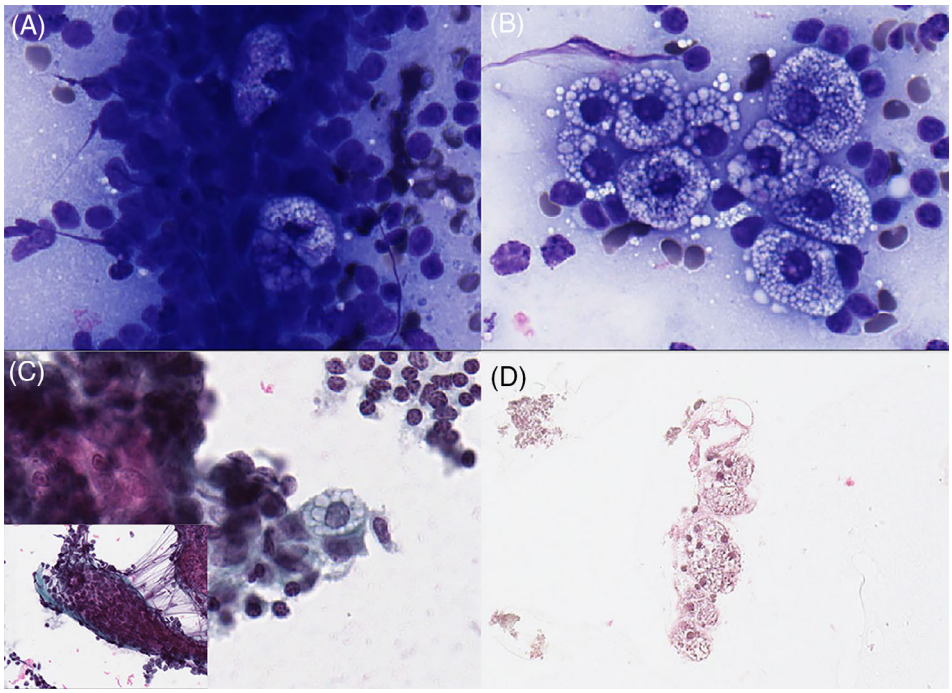

FIGURE 1 Representative Cytologic preparations of sebaceous lymphadenoma using conventional smears. (A and B) Diff-Quik stained smear preparations showing clusters of sebaceous cells with polygonal shapes, low nuclear to cytoplasmic ratios and microvesicular vacuolated cytoplasm that is impinging on a central small nucleus. These cells are noted either in association with basaloid cells or dispersed singly or in small clusters among lymphocytes or crushed lymphoid tangles (DQ stains, ×200, ×400 original magnification). C, Papanicoloau-stained smears show similar microvesicular, vacuolated sebaceous-type cells in close association with basaloid cells (Pap stain, ×400 original magnification). In addition, the inset demonstrates extracellular basement membrane-like matrix seen in association with this case (Pap stain, ×200 original magnification). The basaloid component shows no evidence of cytologic atypia, mitoses, or necrosis. D, Separate representative example of sebaceous cells in a cell block from a sebaceous LAD (case 3, UPMC, ×200 original magnification) [Color figure can be viewed at wileyonlinelibrary.com]

depends on the components represented in the FNA preparation. In our case series, it is not surprising given the rarity of this entity that neither salivary gland LAD subtype had a definitive preoperative diagnosis; rather, a descriptive comment was provided. In theory, SLADs and NSLADs would be classified either as a Neoplasm: Benign or in some cases as SUMP according to the MSRSGC. Based on our cohort, NSLADs were easier to accurately classify than the SLAD cases. A significant problem in the cytologic evaluation of LAD is obtaining an adequate specimen containing a representative sample of the epithelial component. For several of the cases in our cohort, the specimens were paucicellular with rare or no identifiable epithelial clusters in an otherwise predominantly cystic or lymphoid background. Such specimens raised a differential diagnosis of an inflammatory process or a reactive lymph node leading to their misclassification as Non-Neoplastic, or rarely as AUS when rare epithelial elements were present. Even among cases that had adequate cellularity with basaloid or vacuolated tumor cells present in a lymphoid background, the aspirates were classified as SUMP given the wide overlapping differential diagnostic considerations.

The variation that we found in the cytologic interpretation of LAD aspirates is consistent with prior studies on FNA from SLAD in which only 5/23 cases (21.7%) were specifically identified, with the remaining cases classified over the entire MSRSGC spectrum ranging from NonDiagnostic to Malignant (Table 3). Similarly, none of the seven reported NSLAD FNA cases in the literature was definitively diagnosed (Table 4). The challenge in classifying an FNA of LAD using the MSRSGC appears to relate to a combination of sampling issues and the broad differential diagnostic considerations. Given these limitations, a conservative approach incorporating clinical history and imaging findings would seem prudent, and an intraoperative frozen consultation could be considered for further diagnostic clarification for surgical resection cases.

The cytologic differential diagnosis for the two LAD subtypes includes a range of nonneoplastic to neoplastic benign and malignant conditions[16,17]. The differential diagnoses along with useful ancillary tests for distinguishing LAD from other tumor types are summarized in Table 5. The role of ancillary testing is limited as the diagnosis of salivary gland LAD is primarily made on cyto- and histomorphologic features especially since LAD does not have a specific immunocytologic signature [13,14,16,17]. That being said, judicious use of immunohistochemistry, flow cytometry, and in rare cases molecular testing can help to exclude certain entities from the differential diagnosis.

For cytologic preparations of NSLAD containing basaloid fragments in a background of dispersed lymphocytes or crushed lymphoid tangles (Figure 3A-C), and lacking cytologic atypia and extracellular matrix (Figure 3D), the differential diagnosis would include basal cell

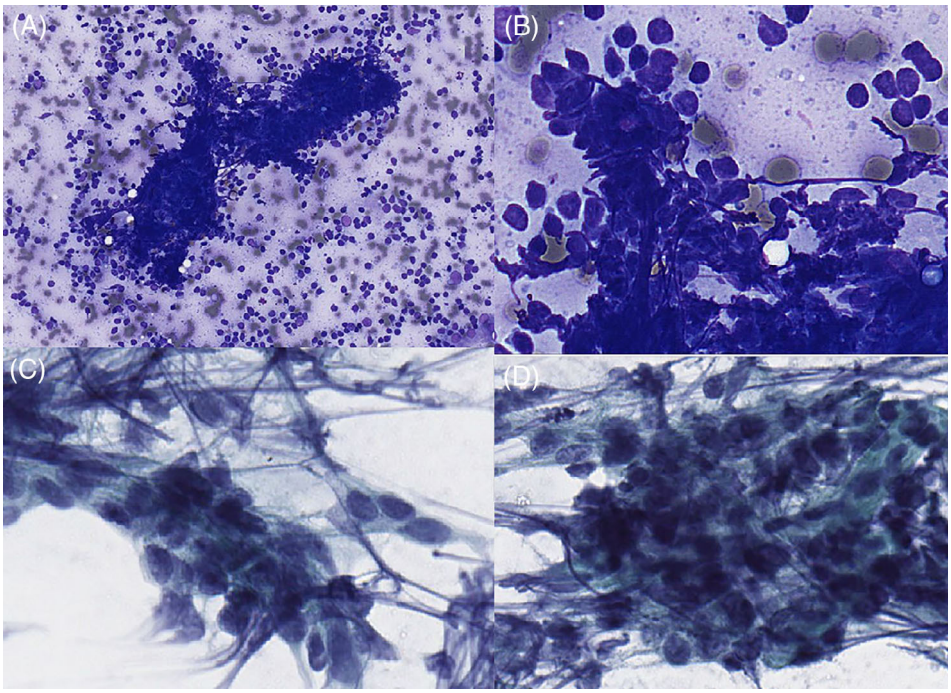

FIGURE 2 Cytologic preparations of nonsebaceous lymphadenoma in conventional smears. (A, and B,) Diff-Quik stained smears showing clusters of basaloid cells among dispersed lymphocytes or crushed lymphoid tangles (DQ stains, ×100, ×400 original magnification). (C, and D,) Papanicoloau-stained smears show similar basaloid cells organized in a cohesive three-dimensional cluster with uniform small nuclei without any cytologic atypia (Pap stain, ×200, ×400 original magnification) [Color figure can be viewed at wileyonlinelibrary.com]

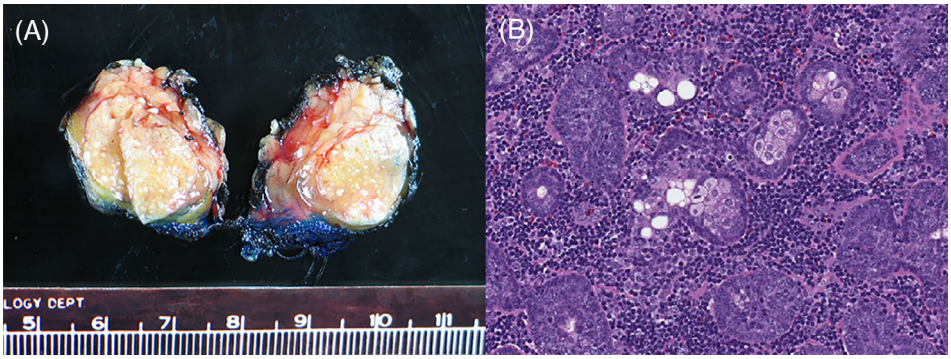

FIGURE 3 Representative gross pathology and light microscopy images of sebaceous lymphadenoma. A, Gross examination of sebaceous LAD (case 9) reveals a well-demarcated, pale yellow-tan lesion within the salivary gland with small white scattered foci. The pale areas within the lesion likely correspond to the lymphoid background whereas the scattered white foci likely reflect sebaceous differentiation and lipid accumulation. B, Light microscopy examination of a representative sebaceous lymphadenoma (case 2) highlighting basaloid clusters transitioning with sebaceous differentiation within a lymphoid background [Color figure can be viewed at wileyonlinelibrary.com]

neoplasia as well as entities such as lymphoepithelial sialadenitis[16]. Lymphoepithelial sialadenitis may similarly show clusters of ductal epithelium imparting a basaloid appearance in a background of lymphocytes. However, unlike LAD that presents as a well-delineated solitary mass, lymphoepithelial sialadenitis (as well as chronic sialadenitis) presents as a diffuse, indistinct, firm salivary gland enlargement, and lymphoepithelial sialadenitis is often found in association with autoimmune disorders. Lymphoepithelial cysts, both sporadic and those associated with human immunodeficiency virus (HIV), are usually distinguished not only by their radiologic appearance as a cystic or multi-cystic lesion, but also cytologically by the presence of abundant anucleate squamous cells in a lymphocyte-rich background. Basal cell adenoma and adenocarcinoma are distinguished from aspirates of LAD by their biphasic myoepithelial-epithelial composition. The latter can be highlighted immunohistochemically using markers for myoepithelial differentiation, whereas LAD lacks a myoepithelial component[13]. In addition, basal cell adenomas and adenocarcinomas are often positive for β-catenin or show cylindromatosis gene mutations[23]. Another distinguishing feature is that basal cell tumors are not typically associated with a lymphoid background as is found in aspirates from LAD. Basement membrane-type matrix may rarely be seen with LADs (Figure 1C inset)[16], and by itself may create diagnostic confusion when attempting to distinguish LADs from basal cell adenomas and adenocarcinoma.

Among malignant entities in the differential diagnosis with LAD, lymphoepithelial carcinoma shows overlapping features that include

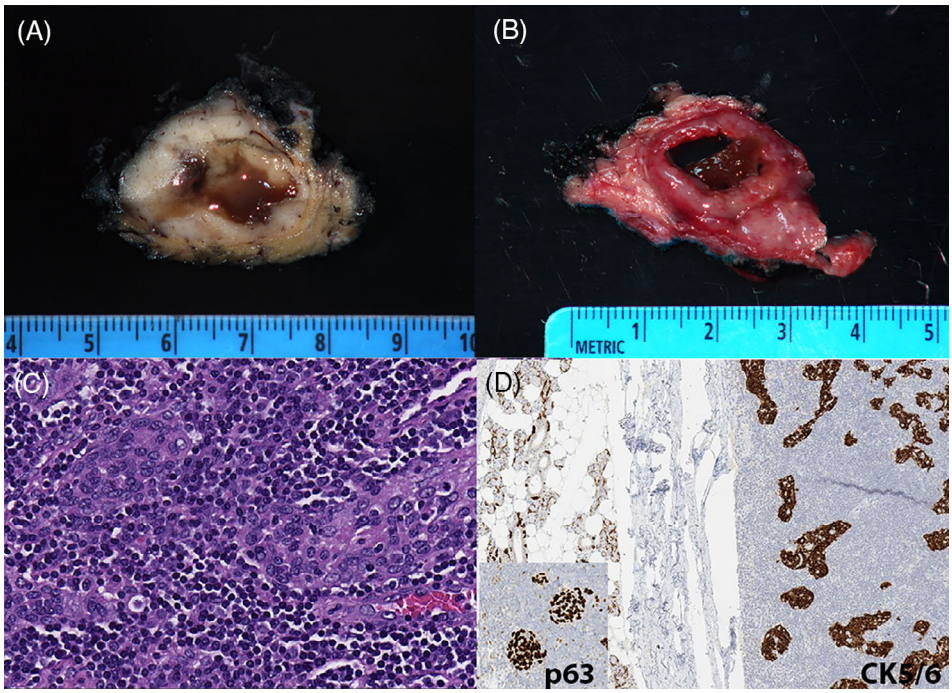

FIGURE 4 Representative gross pathology and light microscopy examples of nonsebaceous lymphadenomas. A, Gross examination of nonsebaceous LAD (case 1 and 2) reveals a well-demarcated, pale tan solid and cystic lesions within the salivary gland. B, Light microscopy examination of a representative nonsebaceous lymphadenoma (case 2) highlights basaloid clusters within a lymphoid background. The basaloid clusters are positive for CK5/6 and p63 (inset) on immunohistochemistry (×100 original magnification) [Color figure can be viewed at wileyonlinelibrary.com]

epithelial syncytia and background lymphocytes; however, the epithelial component of lymphoepithelial carcinoma exhibits marked "malignant-appearing" nuclear atypia. While ancillary marker studies of LAD are limited, Seethala et al demonstrated that none of their tested cases harbored Epstein-Barr Virus (EBV), human papilloma virus (HPV) or human herpes virus 8 (HHV8)[13]. By comparison, lymphoepithelial carcinomas characteristically demonstrate EBV expression using in situ hybridization[24]. Metastatic carcinoma, especially nonkeratinizing squamous cell carcinoma (SCC) has more cellular smears with basaloid epithelial groups of tumor cells in a lymphocytic background. The combination of nuclear atypia, background necrosis, positivity for high-risk HPV, and location of a tumor in Level II or III of the neck helps distinguish it from LAD[13]. For parotid LAD, care should be taken to distinguish this neoplasm from metastasis of a cutaneous neoplasm including keratinizing squamous cell carcinoma, melanoma, and sebaceous carcinoma. The latter may be the most difficult to figure out; however, sebaceous carcinomas show overt high-grade atypia which together with the clinical information of a prior eyelid cancer would help to exclude LAD.

For SLAD, the presence of vacuolated sebaceous cells further expands the differential to include neoplasms with a clear cell/vacuolated pattern[17]. Sebaceous cells are lipid-rich and thus stain positive with Oil Red O, and with immunohistochemistry they often stain for androgen receptor[25]. Mucoepidermoid carcinoma can show similar foamy cells and an associated lymphoid response on cytology. However, the glandular component in proximity to squamoid cells contains mucin, and the background will be mucinous. Moreover, the mucinous component will stain pink with mucicarmine and will be negative with Oil Red O. Molecular studies may also identify the MAML2 translocation characteristic of mucoepidermoid carcinoma[23]. Metastatic clear cell renal cell carcinoma in a salivary gland lymph node, albeit rare, will often show clear, vacuolated cells in association with background blood. However, these vacuolated renal cancer cells typically show significant cytologic atypia, lack of an accompanying basaloid component, and ancillary immunohistochemistry with PAX-8 and carbonic anhydrase (CA-IX) could be used to confirm the diagnosis. As mentioned previously, sebaceous carcinoma is exceedingly rare and can be challenging to diagnose cytologically, but the malignant cells show significant cytologic atypia, including pleomorphism and prominent nucleoli[7,13,15].

Finally, for both LAD subtypes if the associated lymphoid tissue is primarily sampled then an intra-parotid lymph node, chronic sialadenitis, lymphoepithelial lesion or low-grade lymphoproliferative disorder would need to be excluded. Even in the absence of atypical features, flow cytometric analysis and/or immunohistochemistry may be necessary to exclude lymphoma. As was observed in some of the cases in our cohort, a mixed population of lymphocytes together with negative flow cytometry will support a reactive lymphoid process. An integrated correlation with clinical and radiologic information and follow-up would be needed in this setting.

While reviewing our salivary gland LAD cases, we noted a potential diagnostic pitfall. In 50% (n = 2) of NSLAD FNAs there were vacuolated cells, which lead to a false-positive interpretation as a SLAD or another neoplasm. By DQ smear preparation, these vacuolated cells had a glandular appearance with a columnar morphology and basally placed nuclei (Figure 5A) and likely correspond to the sampled abluminal glandular component of a NSLAD (Figure 5B). Clear cell/vacuolated change has not been previously reported in NSLAD, and while the glandular component can be cytokeratin seven positive, the precise cytoplasmic contents imparting a delicate cytoplasmic appearance are unclear[13]. However, true sebaceous differentiation should show characteristic polygonal cells with a low nuclear to cytoplasmic ratio and microvesicular cytoplasm scalloping a central small nucleus.

TABLE 3 Summary of the fine needle aspiration cytology interpretations and MSRSGC equivalent of sebaceous salivary lymphadenomas in the literature

Reference	Gender and Age	Preoperative FNA diagnosis	MSRSGC equivalent	Histologic follow-up
Assor (1970)[26]	65 year-old woman	Mucoepidermoid carcinoma	Malignant	Sebaceous lymphadenoma
Mayorga et al (1999)[27]	78 year-old woman	Acinic cell carcinoma	Malignant	Sebaceous lymphadenoma
Firat et al (2000)[28]	56 year-old woman	Sebaceous lymphadenoma	Benign Neoplasm	Sebaceous lymphadenoma
Shukler and Panicker (2003)[22]	68 year-old woman	Pleomorphic adenoma	Benign Neoplasm	Sebaceous lymphadenoma
Boyle and Meschter (2004)[29]	75 year-old man	Sebaceous lymphadenoma	Benign Neoplasm	Sebaceous lymphadenoma
Hayashi et al (2007)	72 year-old man	Warthin tumor	Benign Neoplasm	Sebaceous lymphadenoma
Hayashi et al (2007)[30]	57 year-old man	Pleomorphic adenoma	Benign Neoplasm	Sebaceous lymphadenoma
Banich et al (2007)[19]	60 year-old man	Warthin tumor	Benign neoplasm	Sebaceous lymphadenoma
Chandrasekhar et al (2007)[31]	28 year-old man	Abscess contents	Nonneoplastic	Sebaceous lymphadenoma
Maffani et al (2007)[32]	67 year-old woman	Granulomatous inflammation	Nonneoplastic	Sebaceous lymphadenoma
Majeed et al (2008)[33]	60 year-old man	Nondiagnostic	Nondiagnostic	Sebaceous lymphadenoma
Sun et al (2009)[10]	16 year-old boy	Warthin	Benign neoplasm	Sebaceous lymphadenoma
While et al (2010)[34]	80 year-old woman	Lymphoid population with differential including non-Hodgkin lymphoma	Atypical	Sebaceous lymphadenoma
Jazaerly et al (2014)[20]	57 year-old man	Sebaceous lymphadenoma	Benign neoplasm	Sebaceous lymphadenoma
Liu et al (2014)[14]	73 year-old man	Pleomorphic adenoma	Benign neoplasm	Sebaceous lymphadenoma
Liu et al (2014)[14]	60 year-old man	Pleomorphic adenoma	Benign neoplasm	Sebaceous lymphadenoma
Liu et al (2014)[14]	72 year-old woman	Lymphadenoma	Benign neoplasm	Sebaceous lymphadenoma
Vande Haar et al (2014)[17]	81 year-old woman	Parotid epithelial neoplasm	Salivary gland neoplasm of uncertain malignant potential	Sebaceous lymphadenoma
Potenziani et al (2017)[35]	63 year-old man	Intraparotid lymph node with reactive changes	Nonneoplastic	Sebaceous lymphadenoma
Rastogi (2017)[36]	41 year-old woman	Suggestive of sebaceous lymphadenoma	Benign neoplasm	Sebaceous lymphadenoma
Sun et al (2018)[37]	82 year-old woman	Lymphoproliferative disorder	Malignant	Sebaceous lymphadenoma
Shekarkhar et al (2018)[21]	39 year-old man	Intrasalivary lymph node with reactive changes	Nonneoplastic	Sebaceous lymphadenoma
Al-Essa M (2020)[18]	55 year-old woman	Chronic inflammation	Nonneoplastic	Sebaceous lymphadenoma
This study	See Table 2			

Abbreviations: FNA, fine needle aspiration; MSRSGC, Milan system of reporting salivary gland cytology.

TABLE 4 Summary of the fine needle aspiration cytology interpretations and MSRSGC equivalent of nonsebaceous salivary lymphadenomas in the literature

References	Gender and Age	Preoperative FNA diagnosis	MSRSGC equivalent	Histologic follow-up
Gallego et al (2009)	58 year-old woman	Nondiagnostic	Nondiagnostic	Nonsebaceous lymphadenoma
Castelino-Prabhu et al (2009)[16]	80 year-old woman	Salivary epithelioid neoplasm with basaloid features	Salivary gland neoplasm of uncertain malignant potential	Nonsebaceous lymphadenoma
Yamanaka H et al (2019)	65 year-old woman	Polymorphous lymphoid population with mild nuclear atypia	Atypia of undetermined significance	Nonsebaceous lymphadenoma
Mabel Cedeno Diaz et al (2014)	57 year-old woman	Nondiagnostic	Nondiagnostic	Nonsebaceous lymphadenoma
Mabel Cedeno Diaz et al (2014)	64 year-old woman	Polymorphous lymphoid background	Nonneoplastic	Nonsebaceous lymphadenoma
Kato et al (2016)	54 year-old man	Abundant small lymphocytes	Nonneoplastic	Nonsebaceous lymphadenoma
Kato et al (2016)	61 year-old woman	Abundant small lymphocytes	Nonneoplastic	Nonsebaceous lymphadenoma
This study	See Table 2			

Abbreviations: FNA, fine needle aspiration; MSRSGC, Milan system of reporting salivary gland cytology.

TABLE 5 Diagnostic pitfalls for Salivary lymphadenoma (sebaceous and nonsebaceous) and their associated salient features and ancillary testing

Differential diagnoses	Salient cytomorphologic features	Ancillary diagnostic testing
Sebaceous lymphadenoma	• Sebacecous cells • Basaloid cells as clusters • Lymphoid background • No cytologic atypia, no mitoses, no necrosis	• Oil Red O positive • Androgen receptor positive • Negative for mucicarmine stain
Nonsebaceous lymphadenoma	• Basaloid cells as clusters • Lymphoid background • No cytologic atypia, no mitoses, no necrosis	• NA
Metastatic clear cell renal cell carcinoma	• Large cells with low nuclear to cytoplasmic ratio • Multivacuolated, wispy cytoplasm • Adherent metachromatic basement-membrane material • Nuclear membrane irregularities • No basaloid epithelial cells • Pertinent clinical history	• PAX-8 • CA-IX
Lymphoma	• Monomorphic lymphoid population • Nuclear enlargement, irregularities and more open chromatin	• Flow cytometry with appropriate markers
Sebaceous carcinoma	• Sebaceous cells with significant cytologic atypia • Nuclear enlargement, pleomorphism, prominent nucleoli	• NA
Keratinizing Squamous cell carcinoma	• Infiltrative border • Dirty necrotic background • Dyskeratotic atypical cells	• p40 positive • Usually HPV negative
Nonkeratinizing squamous cell carcinoma	• Infiltrative borders • Basaloid epithelial clusters with cytologic atypia, mitoses • Pertinent clinical history	• Usually HPV positive

TABLE 5 (Continued)

Differential diagnoses	Salient cytomorphologic features	Ancillary diagnostic testing
Adenoid cystic carcinoma	• Infiltrative borders • Three dimensional basaloid clusters associated with matrix globules	• Positive for myoepithelial markers • *MYB-NFIB* translocation
Mucoepidermoid carcinoma with TALP	• Mucinous background • Glandular mucinous cells and squamous cells • Muciphages	• Mucicarmine stain may highlight mucin-containing glandular cells • *MAML2* translocation
Lymphoepithelial carcinoma	• Syncytial groups of tumor cells with enlarged nuclei and prominent nucleoli • Admixed lymphocytes	• EBV positive
Lymphoepithelial sialadenitis	• Nonneoplastic acini and sheets of ductal epithelium • Lymphoid background • Pertinent clinical history of diffuse firm salivary gland	• NA
Acinic cell carcinoma with TALP	• Oncocytic cells with granular cytoplasm in three dimensional clusters • Stripped naked nuclei • Admixed lymphocytes	• DOG1, PAS-D to highlight zymogen granules • NR4A3 immunohistochemistry • Molecular testing
Warthin tumor	• Oncocytes • Proteinaceous debris • Lymphoid background	• NA
Lymphoepithelial cyst	• More common in HIV positive patients • Cyst contents • Anucleate squamous cells • No cytologic atypia	• NA
Benign intraparotid lymph node	• Lymphoid population • Sampling of germinal centers with tingible-body macrophages	• Flow cytometry will show a polymorphous nonneoplastic lymphoid population
Needle track contamination of sebaceous component	• Scant sebaceous cells by comparison to basaloid population • Lymphoid background	• NA

Abbreviations: EBV, Epstein Barr virus; HIV, human immunodeficiency virus; HPV, human papilloma virus; NA, not available.

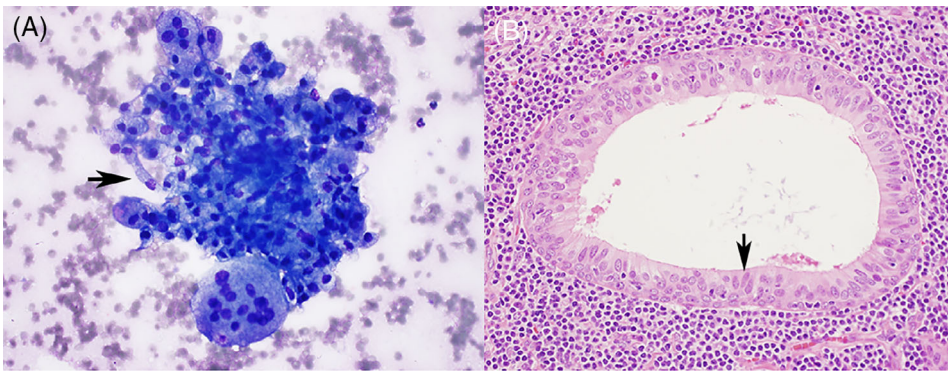

FIGURE 5 Abluminal glandular cells in a nonsebaceous lymphadenoma imparting a clear cell morphology can be a diagnostic pitfall. A, Unusual case of nonsebaceous LAD in the patient with Cowden syndrome with clear cells mimicking sebaceous cells. On further examination, few clear cells show a columnar appearance with eccentrically placed nuclei imparting a glandular appearance (DQ stain, ×400 original magnification, black arrow). B, Corresponding abluminal glandular cells in nonsebaceous LAD on histologic resection (H&E, ×400 original magnification, black arrow) [Color figure can be viewed at wileyonlinelibrary.com]

In conclusion, LAD is a benign primary salivary gland neoplasm that, although rare, is an important diagnostic consideration that can be overlooked in the differential diagnosis of a lymphoid-containing salivary gland lesion. We highlight the key diagnostic features for SLAD and NSLAD based on cytologic preparations. While lymphocytes, basaloid cell clusters, and proteinaceous debris can be seen with both LAD subtypes, true sebaceous cells are restricted to only the sebaceous variant. In addition, cytologists need to be aware of the broad and often overlapping differential diagnoses for both LAD subtypes, as well as the potential diagnostic pitfall that can occur in these cases due to sampling of only certain components such as clear cell/vacuolated change. The role of ancillary studies plays a limited role in diagnosing LAD. Hence, familiarity of the cytomorphologic features of SLAD and NSLAD is of paramount importance to ensure the correct categorization of these rare neoplasms when using the MSRSGC.

CONFLICT OF INTEREST

The authors declare that there are no conflicts of interest.

AUTHORS' CONTRIBUTIONS

All authors of this article declare that we qualify for authorship as defined by diagnostic cytopathology. Each author has participated sufficiently in the work and takes public responsibility for appropriate portions of the content of this article. Kartik Viswanathan carried out the background research and drafted the manuscript. Kartik Viswanathan and William C. Faquin conceived the idea for the manuscript and its design and coordination. Liron Pantanowitz, Zahra Maleki, Richard Cantley and William C. Faquin were involved in collation of cases. Kartik Viswanathan, Zahra Maleki, Richard Cantley, Liron Pantanowitz and William C. Faquin participated revising the manuscript critically for important intellectual content. All authors read and approved the final manuscript.

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REFERENCES

1. Seethala RR, LiVolsi VA, Baloch ZW. Relative accuracy of fine-needle aspiration and frozen section in the diagnosis of lesions of the parotid gland. *Head Neck*. 2005;27(3):217-223.
2. Rossi ED, Wong LQ, Bizzarro T, et al. The impact of FNAC in the management of salivary gland lesions: institutional experiences leading to a risk-based classification scheme. *Cancer Cytopathol*. 2016;124(6):388-396.
3. Barbarite E, Puram SV, Derakhshan A, Rossi ED, Faquin WC, Varvares MA. A call for universal acceptance of the Milan system for reporting salivary gland cytopathology. *Laryngoscope*. 2020;130(1):80-85.
4. Jayaram G, Verma AK, Sood N, Khurana N. Fine needle aspiration cytology of salivary gland lesions. *J Oral Pathol Med*. 1994;23(6):256-261.
5. Layfield LJ, Gopez E, Hirschowitz S. Cost efficiency analysis for fine-needle aspiration in the workup of parotid and submandibular gland nodules. *Diagn Cytopathol*. 2006;34(11):734-738.
6. Mairembam P, Jay A, Beale T, et al. Salivary gland FNA cytology: role as a triage tool and an approach to pitfalls in cytomorphology. *Cytopathology*. 2016;27(2):91-96.
7. Layfield LJ, Glasgow BJ. Diagnosis of salivary gland tumors by fine-needle aspiration cytology: a review of clinical utility and pitfalls. *Diagn Cytopathol*. 1991;7(3):267-272.
8. Rossi ED, Faquin WC. The Milan system for reporting salivary gland cytopathology (MSRSGC): an international effort toward improved patient care-when the roots might be inspired by Leonardo da Vinci. *Cancer Cytopathol*. 2018;126(9):756-766.
9. Rawlinson NJ, Almarzooqi S, Nicol K. Sebaceous lymphadenoma of the parotid gland in a 13-year-old girl: a case report. *Head Neck Pathol*. 2010;4(2):144-147.
10. Sun G, Hu Q, Huang X, Tang E. Sebaceous lymphadenoma of parotid gland in a child. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod*. 2009;107(2):253-255.
11. El-Naggar AK, Chan J, Grandis JR, Takata T, Slootweg PJ. *WHO Classification of Head and Neck Tumours*. 4th ed. Lyon, France: IARC; 2017.
12. Pattnaik N, Parmar D, Rao BR, Rao ES. Sebaceous lymphadenoma of submandibular gland. *J Indian Med Assoc*. 2013;111(1):62-63.
13. Seethala RR, Thompson LD, Gnepp DR, et al. Lymphadenoma of the salivary gland: clinicopathological and immunohistochemical analysis of 33 tumors. *Mod Pathol*. 2012;25(1):26-35.
14. Liu G, He J, Zhang C, Fu S, He Y. Lymphadenoma of the salivary gland: report of 10 cases. *Oncol Lett*. 2014;7(4):1097-1101.
15. Kara H, Sonmez S, Bagbudar S, Gulluoglu M, Basaran B. Malignant transformation of parotid gland non-sebaceous Lymphadenoma: case report and review of literature. *Head Neck Pathol*. 2020. https://doi.org/10.1007/s12105-020-01133-3. Online ahead of print.
16. Castelino-Prabhu S, Li QK, Ali SZ. Nonsebaceous lymphadenoma of the parotid gland: cytopathologic findings and differential diagnosis. *Diagn Cytopathol*. 2010;38(2):137-140.
17. Vande Haar MA, DeFrias D, Lin X. Fine-needle aspiration cytomorphology of sebaceous lymphadenoma of the salivary gland. *Diagn Cytopathol*. 2014;42(11):959-963.
18. Al-Essa M. Sebaceous lymphadenoma of parotid gland: a case report of a unique presentation in an immunocompromised patient. *J Family Med Prim Care*. 2020;9(2):1202-1205.
19. Banich J, Reyes CV, Bier-Laning C. Sebaceous lymphadenoma identified by fine needle aspiration biopsy: a case report. *Acta Cytol*. 2007;51(2):211-213.
20. Jazaerly T, Bandyopadhyay S, Almardini N, Husain M. Diagnosis of sebaceous lymphadenoma by fine needle aspiration in a patient with Cowden syndrome: case report and review of the literature. *Cytojournal*. 2014;11:25.
21. Shekarkhar G, Soleimanpour H, Jafari SH, Zamani F. Sebaceous Lymphadenoma of parotid: imaging, cytological, and histological findings in detail. *Case Rep Pathol*. 2018;2018:1–4.
22. Shukla M, Panicker S. Synchronous sebaceous lymphadenoma with squamous cell carcinoma - case report. *World J Surg Oncol*. 2003;1(1):30.

23. Jo VY, Krane JF. Ancillary testing in salivary gland cytology: a practical guide. *Cancer Cytopathol*. 2018;126(8):627-642.
24. Chan JK, Yip TT, Tsang WY, Poon YF, Wong CS, Ma VW. Specific association of Epstein-Barr virus with lymphoepithelial carcinoma among tumors and tumorlike lesions of the salivary gland. *Arch Pathol Lab Med*. 1994;118(10):994-997.
25. Bayer-Garner IB, Givens V, Smoller B. Immunohistochemical staining for androgen receptors: a sensitive marker of sebaceous differentiation. *Am J Dermatopathol*. 1999;21(5):426-431.
26. Assor D. Sebaceous lymphadenoma of the parotid gland. A case report. *Am J Clin Pathol*. 1970;53(1):100-103. https://doi.org/10.1093/ajcp/53.1.100
27. Mayorga M, Fernández N, Val-Bernal JF. Synchronous ipsilateral sebaceous lymphadenoma and acinic cell adenocarcinoma of the parotid gland. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod*. 1999;88(5):593-596.
28. Firat P, Hosal S, Tutar E, Ruacan S. Sebaceous lymphadenoma of the parotid gland: Journal canadien d'otolaryngologie. *J Otolaryngol*. 2000;29(2):114-116.
29. Boyle JL, Meschter SC. Fine needle aspiration cytology of a sebaceous lymphadenoma: *a case report*. *Acta Cytol*. 2004;48(4):551-554.
30. Hayashi D, Tysome JR, Boyei E, Gluckman P, Barbaccia C. Sebaceous lymphadenoma of the parotid gland: report of two cases and review of the literature. *Acta Otorhinolaryngol Ital*. 2007;27(3):144-146.
31. Chandrasekar T, Ramani P, Anuja N, et al. Unilocular cystic sebaceous lymphadenoma: a rare tumour. *Ann R Coll Surg Engl*. 2007;89(4):1-3.
32. Maffini F, Fasani R, Petrella D, et al. Sebaceous lymphadenoma of salivary gland: a case report and a review of the literature. *Acta Otorhinolaryngol Ital*. 2007;27(3):147-150.
33. Majeed M, Murray B, Hume W, Makura Z. Sebaceous lymphadenoma of the parotid gland. *Dentomaxillofac Radiol*. 2008;37(5):300-304.
34. While B, Whiteside OJ, Desai V, Gurr P. Sebaceous lymphadenoma: a case report and review of the literature. *Ear Nose Throat J*. 2010;89(8):E22-E23.
35. Potenziani S, Applebaum D, Krishnan B, Gutiérrez C, Diwan AH. Multiple clear cell acanthomas and a sebaceous lymphadenoma presenting in a patient with Cowden syndrome - a case report. *J Cutan Pathol*. 2017;44(1):79-82.
36. Ratsogi N. Sebaceous lymphadenoma of the parotid gland-a case report of infrequently encountered tumor. *Asian Pac. J. Health Sci.*, 2017;4(3):6-8.
37. Gallego L, Junquera L, Fresno MF. Non-sebaceous lymphadenoma of the parotid gland: immunohistochemical study and DNA ploidy analysis. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod*. 2009;107(4):555-558.
38. Mabel Cedeño Díaz O, Jesús Fernández Aceñero M, Mullor Nogales R, Pino Jiménez A, Lourdes Maniegas Lozano M, Martínez Pérez D. Non sebaceous lymphadenoma of the parotid gland: Two-case report. *Journal of Oral and Maxillofacial Surgery, Medicine, and Pathology*. 2014. 26(2):239-243.
39. Kato T, Nakamizo M, Yokoshima K et al. Non-sebaceous lymphadenoma of the parotid gland: a report of two cases of superficial parotidectomy. *Head and Neck Surgery*. 25(3):415-420, 2015.

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