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Dr. Deepti Arora
Assistant Professor, Department of Pathology, Teerthankar Mahaveer Medical College and Research Center, Moradabad, Uttar Pradesh, India

Dr. Rashmi Chauhan
Assistant Professor, Department of Pathology, Teerthankar Mahaveer Medical College and Research Center, Moradabad, Uttar Pradesh, India

Dr. Ankita Mittal
Associate Professor, Department of Pathology, Teerthankar Mahaveer Medical College and Research Center, Moradabad, Uttar Pradesh, India

Dr. Faiyaz Ahmad
Professor, Department of Pathology, Teerthankar Mahaveer Medical College and Research Center, Moradabad, Uttar Pradesh, India

Dr. Seema Awasthi
Professor, Department of Pathology, Teerthankar Mahaveer Medical College and Research Center, Moradabad Uttar Pradesh, India

Dr. Shyamoli Dutta
Professor, Department of Pathology, Teerthankar Mahaveer Medical College and Research Center, Moradabad, Uttar Pradesh, India

Corresponding Author:
DR. Rashmi Chauhan
Assistant Professor, Department of Pathology, Teerthankar Mahaveer Medical College and Research Center, Moradabad, Uttar Pradesh, India

Practical implication of risk based categorization of salivary gland lesions on FNAC by Milan system: A two year retrospective study

Dr. Deepti Arora, Dr. Rashmi Chauhan, Dr. Ankita Mittal, Dr. Faiyaz Ahmad, Dr. Seema Awasthi and Dr. Shyamoli Dutta

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Abstract
Milan system was introduced in 2018 with an objective to standardize and bring uniformity in the reporting of salivary gland lesion on fine needle aspiration. The present study is conducted retrospectively over a period of two years to reclassify salivary gland lesions according to Milan system and to calculate risk of malignancy in each category. Out of total 106 salivary gland aspiration cases, histologic follow up was available for 57 cases. On re-categorizing these lesions, maximum cases belong to neoplastic benign category. Risk of malignancy in each category were-non diagnostic (0%), non-neoplastic (25%), atypia of undetermined significance AUS (33.3%), neoplastic benign (5.3%), suspicious for malignancy (75%) and malignant (100%). Three false negative and one false positive case was identified in the study. In conclusion, Milan system is very helpful in providing a uniform reporting system for salivary gland lesions, enhancing diagnostic accuracy and providing valuable impact on their management.

Keywords: Milan, salivary gland, risk of malignancy, cytology, surgical follow up

Introduction
Salivary gland neoplasms are relatively uncommon and constitute about 2%-6.5% of all the head and neck lesions [1, 2]. Fine Needle Aspiration Cytology (FNAC) is considered first line of modality for investigating salivary gland lesions due to its easy accessibility and superficial location of the lesions [3]. Its role is well established in diagnosing and management of salivary gland lesions. It has high sensitivity (86%-100%) and high specificity (90%-100%) as reported in many studies [4-9]. However, despite being a useful technique, minimal invasion and cost-effectiveness cytopathologists continuously face challenges in reporting due to diversity and heterogeneity of salivary gland lesions [10-13]. Another problem with the reporting of these lesions across multiple institutions worldwide is that they lack a standard terminology along with the few descriptive reports which create a diagnostic confusion and pose difficulty for the clinician to decide the management of the lesion [10, 13-15].

After the successful implementation of The Bethesda System for reporting Cervical Cytopathology (TBSRCC) AND The Bethesda System for Reporting Thyroid Cytopathology (TBSRTC), American Society of Cytopathology (ASC) and the International Academy of Cytology (IAC) proposed a new standardized reporting system for salivary gland FNA specimens in 2015 [10, 13, 16-18]. This finally led to the development of six tiered international classification system: Milan System for Reporting Salivary Gland Cytopathology" (MSRSGC). This evidence based classification system is based on the same rationale as TBSRCC and TBSRTC of improved patient care and is user friendly, practical, universally accepted. It aimed at cyto-histo correlation, promote enhanced sharing of data between institutions and better communication between clinicians and pathologists as gave implied risk of malignancy (ROM) and recommended clinical management [10, 19-22].

The present study was carried out with the aim of reclassifying the cytological diagnosis of salivary gland lesions according to the MSRSGC and to calculate the risk stratification of malignancy in each category in our institute.

Material and methods

The present study is a retrospective study conducted in the department of pathology for two years (October 2017-September 2019). Ethical approval was taken from institutional ethical committee prior to the commencement of study. All the patients with salivary gland swellings who were referred for FNAC in the pathology department of our institute were enrolled in the study. Demographic details like age, gender, relevant clinical data and anatomical location of the lesion were obtained from the hospital medical records. Cytological diagnosis along with the slides of FNAC specimens and histopathological reports wherever available were retrieved from the pathology department.

FNAC from both major and minor salivary gland was done by trained cytopathologists under aseptic precautions with the help of 10 ml disposable syringe and 23/ 24 gauge needle through direct percutaneous or intraoral route with or without ultrasound guidance. Aspiration was done 2-4 times randomly in different directions. The character of the aspirated material was noted. Smears were prepared. Air dried smears were stained with May-Grunwald Giemsa stain while those fixed in 95% alcohol were stained by Papanicolaou method.

Cytological features were evaluated and cases were reclassified according to the criteria of Milan system:

1. Non-diagnostic
2. Non-neoplastic
3. Atypia of undetermined significance (AUS)
4. (a & b) Neoplasm: benign and uncertain malignant potential
5. Suspicious for malignancy (SFM)
6. Malignant. (Table 1)

Histopathologic examination was considered as gold standard to calculate ROM (Risk of Malignancy). For histological examination, biopsy specimens were fixed in 10% neutral buffer formalin, processed and stained with H&E stain (hematoxylin and eosin stain). Both cytologic and histopathology reports were compared and ROM was calculated for each category.

Table 1: Milan system for reporting salivary gland cytopathology (MSRSGC) with risk of malignancy and recommended clinical management

	Diagnostic category	Risk of malignancy (ROM)%	Management
I	Non-diagnostic	25	Clinical & Radiological correlation/Repeat FNA
II	Non-neoplastic	10	Clinical follow up & Radiological correlation
III	Atypia of undetermined significance (AUS)	20	Repeat FNA/Surgery
IV	Neoplasm		
IV a	Neoplasm: Benign	<5	Surgery/Clinical Follow up
IV b	Neoplasm: Salivary gland neoplasm of uncertain malignant potential (SUMP)	35	Surgery
V	Suspicious for Malignancy (SFM)	60	Surgery
VI	Malignant	90	Surgery

Results

A total of 106 FNAC cases of salivary gland lesions were included in the study in a period of two years. The age of these studied cases range from 6 to 80 yrs. Maximum number of cases were seen in the age group of 41-50 years followed by 21-30 years age group. In the study population, males were commonly affected with male to female ratio of 1.7:1. Table 2 shows distribution of cases according to age and sex. Parotid gland was most commonly involved gland in 48.1% followed by submandibular gland in 34.9% and minor salivary gland in 17% (Table 3).

FNAC results were re classified according to Milan system in six categories as depicted in Table 4. Non diagnostic (ND) category included 5 cases (4.7%). 37 cases (34.9%) were reported in Non-neoplastic (NN) category. Chronic sialadenitis was the most common non neoplastic lesion. AUS category was seen in 3 cases (2.8%). Neoplastic benign (NB) category constituted maximum number of 49 cases (46.2%). Of these, pleomorphic adenoma was the most common neoplasm constituting about (87.7%) of all benign neoplasms. Salivary gland neoplasm of uncertain malignant potential (SUMP) category had only 2 cases (3.8%). 4 cases were identified in Suspicious for malignancy (SFM) category while 6 cases (5.7%) were noted in Malignant (M) category. The most common malignant neoplasm seen in our study was mucoepidermoid carcinoma.

Histopathological correlation was available in 57 cases. Risk of malignancy (ROM) was calculated for individual category as shown in Table 5. Maximum ROM was 100% for M category followed by 75% for SFM category. NN, AUS, NB had ROM of 2.5%, 33.3% and 5.3% respectively. Discordance was noted in 4 cases. One case of NN category (mucocele) was found to be malignant (mucoepidermoid carcinoma) on surgical follow up. Two cases of NB category were reported as pleomorphic adenoma on FNAC and were diagnosed histologically as low grade mucoepidermoid carcinoma and adenoid cystic carcinoma. This amounted to 3 false negative cases. One false positive case was identified in SFM category which was reported as suspicion of mucoepidermoid carcinoma cytologically while on histopathological examination, it turned out to be pleomorphic adenoma.

Table 2: Distribution of cases according to Age and Gender

Age (Years)	**0-10**	**6**
	11-20	11
	21-30	26
	31-40	16
	41-50	28
	51-60	9

	61-70	9
	71-80	1
Sex	Male	67
	Female	39

Table 3: Distribution of cases according to gland involved

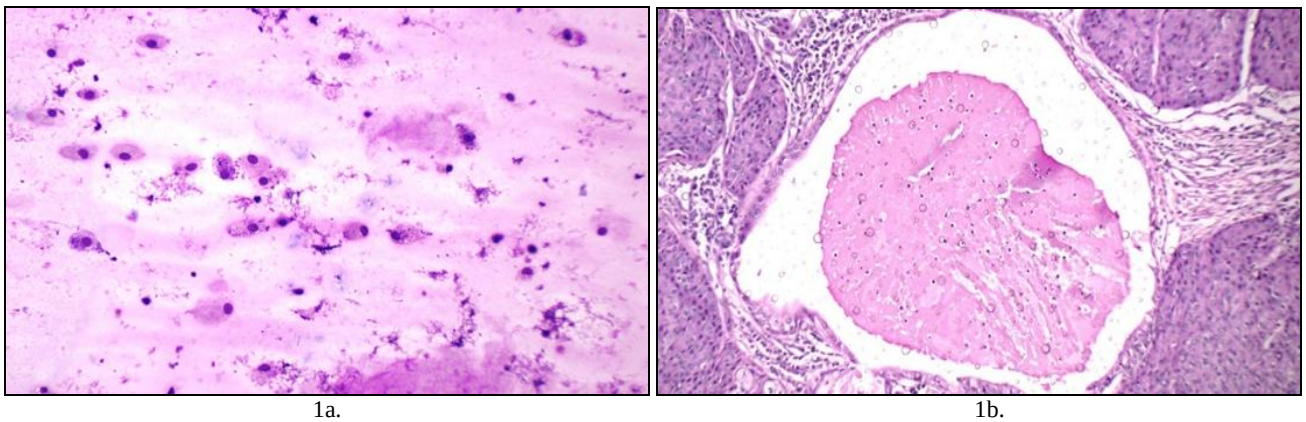
Gland Involved	N = Number of cases (%)
Parotid	51 (48.1)
Submandibular	37 (34.9)
Minor salivary gland	18 (17)

Table 4: Spectrum of reclassified cytological diagnosis by MILAN system (N=106)

Cytological Diagnosis		N = Number of cases (%)
Non Diagnostic		5(4.7)
Non Neoplastic		37(34.9)
	Acute sialadenitis	17(45.9)
	Chronic sialadenitis	7(18.9)
	Necrotizing sialadenitis	2(5.4)
	Sialadenosis	4(10.8)
	Abscess/Suppurative lesion	3(8.1)
	Benign Cystic lesions	3(8.1)
	Others	1(2.7)
AUS		3(2.8)
Neoplastic		51(48.1)
Benign		49(46.2)
	Pleomorphic adenoma	43(87.7)
	Warthin's tumor	4(8.1)
	Myoepithelioma	1(2)
	Basal cell adenoma	1(2)
UMP		2(1.9)
Suspicious for Malignancy(SFM)		4(3.8)
Malignant		6(5.7)
	Mucoepidermoid carcinoma	2(33.3)
	Adenoid cystic carcinoma	1(16.7)
	Poorly differentiated carcinoma	1(16.7)
	Non-Hodgkin's lymphoma	1(16.7)
	Metastatic squamous cell carcinoma	1(16.7)

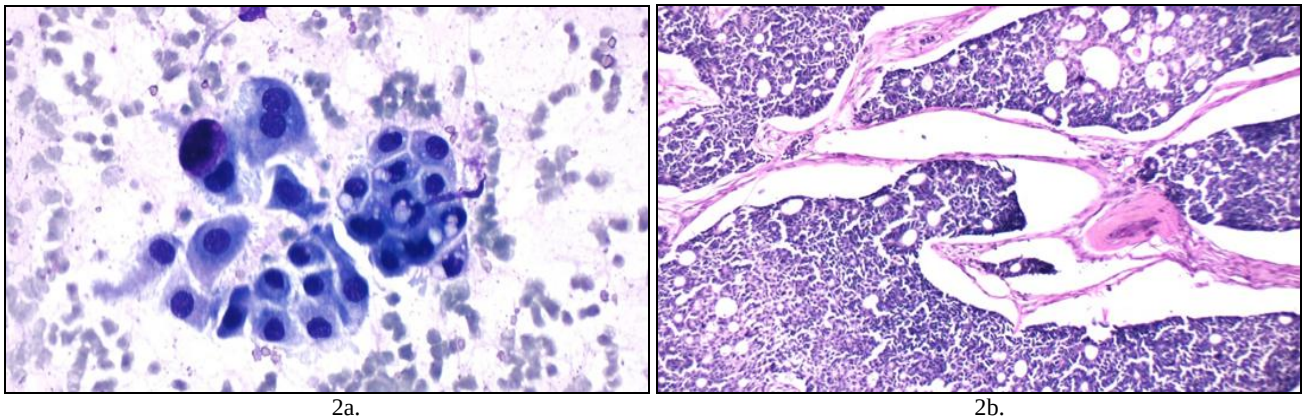
Table 5: Histological follow up of categories of MSRSGC (N=106)

Category	Cat 1	Cat 2	Cat 3	Cat 4a	Cat 4b	Cat 5	Cat 6	Total
Number of cases	5	37	3	49	2	4	6	106
Number of cases with surgical follow up	3	4	3	38	-	4	5	57
Non neoplastic	3	3	0	0	-	0	0	6
Benign: Neoplastic	0	0	2	36		1	0	39
Malignant	0	1	1	2	-	3	5	12
ROM (%)	0	25	33.3	5.3	-	75	100	



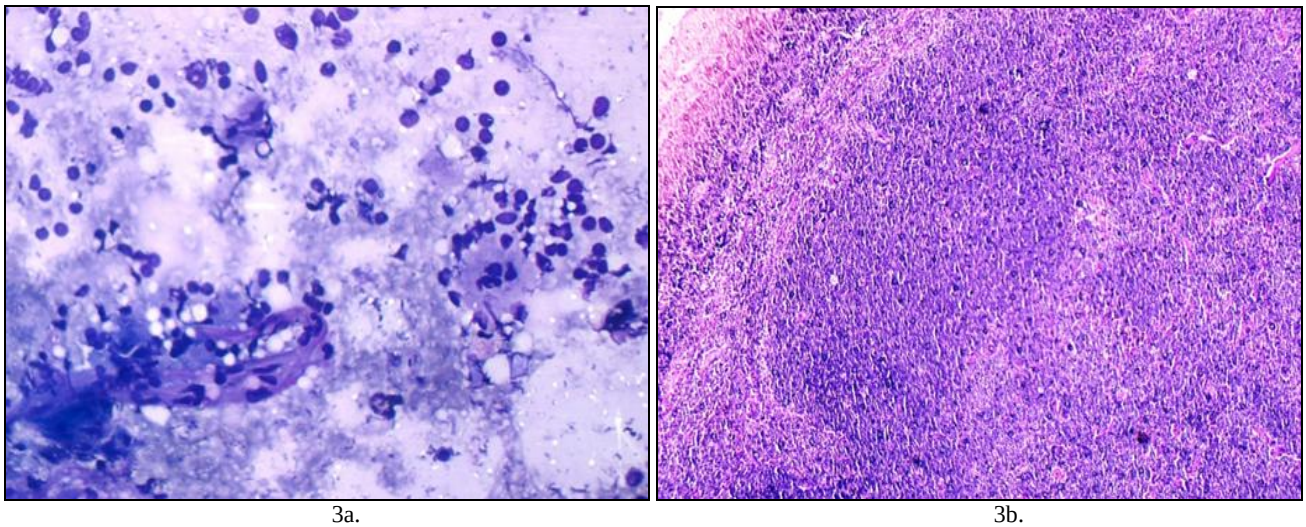
1a. 1b.

Fig 1 a & b: Shows bland epithelial cells with macrophages on background of abundant mucin interpreted as mucocele (400X, MGG)shows features of low grade mucoepidermoid carcinoma (100X, H &E)



2a. 2b.

Fig 2 a & b: Shows epithelial cells around hyaline stromal globule (400X, MGG) diagnosed as adenoid cystic carcinoma with cribriform pattern on histopathology (100X, H&E).



3a. 3b.

Fig 3 a & b: case of atypia of undetermined significance (AUS) with presence of few atypical lymphoid cells (400X, MGG) shows features of Non Hodgkin Lymphoma on histology (100X, H&E)

Discussion

After introduction of FNAC, it has gained widespread popularity and acceptance due to its easy technique, ability to perform in out-patient setting and rapid results. Despite of its importance in deciding the origin and nature of lesion, its role has few limitations in diagnosing and differentiating salivary gland lesions due to complexity and significant overlapping of cytological features in various lesions [21-23]. The development of MSRSGC is an effort to standardize and bring uniformity in the reporting of salivary gland lesions and to avoid descriptive reports in difficult cases leading to diagnostic confusion to the treating clinician [9, 14-16].

In the current study, the age of the patients ranged from 6 yrs to 80 yrs with maximum number of patients in the age group of 41-50 yrs. The study group showed male preponderance with male to female ratio of 1.7:1 which is similar to other studies [21, 24, 25]. Majority of the salivary gland lesions involve parotid gland (48.1%) followed by submandibular gland (34.9%) and minor salivary gland (17%). It is similar to the findings of Karuna V [21] and Jain R *et al.* [26].

In the present study, non-diagnostic group (Cat I) comprised of 5 cases (4.7%) of total study group. These were attributed to insufficient material aspirated. The histologic follow up was done for 3 cases. Two of them were diagnosed as chronic sialadenitis while one was diagnosed as obstructive pathology due to fibrosis which is the possible explanation of low cellularity despite of repetitive aspirations in this case. No malignant lesion was found in this group similar to the study of Savant D *et al.* [27].

In the study group, majority cases (48.1%) were in benign neoplastic group (Cat IV) followed by 34.9% in non-neoplastic group (Cat II) and 5.7% in malignant group (Cat VI). These results were in concordance with the results of studies by Karuna V, Subrata P and Sheetal GG *et al.* [21, 28, 29]. Most common non-neoplastic, benign and malignant neoplasm in the present study was chronic sialadenitis (45.9%), pleomorphic adenoma (87.7%) and mucoepidermoid carcinoma (33.3%). The predominance of these lesions is in concordance with few other previous studies [11, 30].

Non-neoplastic (Cat-II) category included 37 cases (34.9%) out of total 106 FNAC cases. In previous studies, this value ranges from 5.1% to 53.4% while low incidence of non-neoplastic cases was reported by Savant D *et al.* [27]. The calculated ROM for this category was 25% which was higher than the proposed ROM by MSRSGC. It was consistent with the study of Rohilla *et al.* [31] while other studies demonstrated lower ROM for this category [18, 23, 32]. Out of 4 cases which underwent surgical pathology follow up, one false negative case was identified and was

diagnosed as mucoepidermoid carcinoma. It was interpreted on cytology as mucocele as smears were paucicellular with abundant mucoid material with presence of benign looking epithelial cells and scattered macrophages (Image 1a & b). Mucoepidermoid carcinoma is notorious for cystic degenerative changes and on aspiration there may be paucicellular due to cystic fluid and thus misleading diagnosis can be given. Therefore, it is essential to reaspirate the residual mass in cases of cystic lesion to reduce sampling errors.

Neoplastic benign category (Cat-IVa) comprised of maximum cases. Histological follow up was available for 38 out of 49 cases. ROM calculated in this category was 5.3% which was near to MSRSGC criteria and slightly higher than Kala C *et al.* [10]. Few other studies have reported higher ROM that is, 7.1% and 7.14% [18, 23]. Two cases were found to be malignant histopathologically and were misdiagnosed as pleomorphic adenoma. These false negative cases were diagnosed histopathologically, one as low grade mucoepidermoid carcinoma and other as adenoid cystic carcinoma (AdCC). Cytologic smears of the case which was diagnosed as AdCC showed low cellular smears with occasional hyaline globules and foci of myxoid areas showing few cells melting in it (Image 2a & b). Although Mucoepidermoid carcinoma is the most common malignant salivary gland tumor, it poses a diagnostic challenge for cytopathologists due to presence of two types of cells, subtle cytomorphological features of low grade mucoepidermoid carcinoma and degenerative changes. On re-examination of slides, smears showed dominance of intermediate squamous cells which was mimicking myoepithelial cells and cystic change was also seen. Cystic degenerative changes can be found in both the neoplasms and in this particular case this was the reason behind misdiagnosis. According to Ramya Katta and DP Chaganti [19] intermediate squamous cells of low grade MEC may resemble myoepithelial cells so we should take at least 2-3 aspirates from different sites of lesion for proper yield of representative cells. The lower ROM in this category is attributed to the fact that morphological features of benign tumors are well described in literature.

Even in the presence of adequate smears, few cases pose diagnostic dilemma to the most experienced cytopathologists and definitive diagnosis is still not possible. These cases with uncertain diagnosis are placed in indeterminate category which encompasses "AUS", "SUMP" and "suspicious for malignancy" categories of MSRSGC [23, 33].

AUS category (Cat III) includes cases which lack the adequate features for either category i.e., non-neoplastic and neoplastic with presence of atypia were placed into AUS category [33]. In the present study, all the three cases were examined histopathologically and one of them showed discordance amounting to 33.3% ROM in this category. This case was diagnosed as Non Hodgkins Lymphoma on histopathological follow up while a descriptive report of chronic sialadenitis with few atypical lymphoid cells was given cytologically (Image 3a & b). Chronic sialadenitis is a known diagnostic pitfall due to the presence of reactive atypia which emphasizes the importance of using ancillary techniques like flow cytometry and immunohistochemistry in cases of suspicion of atypical lymphoid cells to determine the clonality of these cells [14, 16, 23]. In the current work, ROM of AUS category (33.3%) was higher than the proposed ROM by MSRSGC (10%) but lies within the range of 10%-35% provided in the MSRSGC atlas published in 2018. Our results were comparable to those of Hollyfield *et al.* [16] with ROM of 33%. Viswanathan *et al.* [23] reported higher ROM of 38% and Thirayi *et al.* [32] reported 100% ROM. The reason for variable ROM in this category could be attributed to interobserver variability.

The category SUMP includes cases which cannot be clearly differentiated into benign or malignant neoplasm based on its cytological features [10, 14]. Only 2 cases of total study population (106) in the present study were identified in this category. However no histological follow up was available.

The category of suspicious for malignancy (SFM) also lies in the indeterminate category. This represents FNAC specimens where cytological features are suggestive for malignancy but a specific diagnosis cannot be made [22]. 4 cases (3.8%) were identified in this category. On surgical follow up of all these cases, only one case was found to be benign histopathologically. This case was given detailed descriptive cytological report with suspicion of mucoepidermoid carcinoma but on histopathology it was diagnosed as pleomorphic adenoma with scant stroma, squamoid metaplasia along with mucus cell metaplasia. It was identified as a single false positive case in our study.

Six cases were included in the Malignant category (Category VI) out of which histopathological follow up was available for 5 cases. All these five cases were diagnosed as malignant. These included two cases of mucoepidermoid carcinoma, one case each of adenoid cystic carcinoma, poorly differentiated carcinoma and Non Hodgkins lymphoma.

Calculated Risk of malignancy in SFM is 75% and malignant category is 100%. Different studies showed >90% ROM in both of these categories [12, 17, 18, 20, 21, 23, 27, 32].

Limitation

Although categorization of salivary gland lesion by Milan system is of practical importance but there must be some adequacy criteria like in Bethesda system in cervical and thyroid lesion to effectively rule out false negative and false positive diagnosis.

Conclusion

FNAC is very effective and rapid mode of diagnosis with some limitations depending on the type of lesion and sampling. Due to unavailability of architectural details, low grade malignancies can be reported as benign or vice versa. So, the categorization makes it very effective and universal mode of reporting by which clinicians and patients get non-subjective information regarding the disease, further investigations and treatment required. Thus, MSGSRC enhances diagnostic accuracy of salivary gland lesions with valuable impact on clinical management.

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