ASIAN JOURNAL OF PHARMACEUTICAL AND CLINICAL RESEARCH

INNOVARE ACADEMIC SCIENCES
Knowledge to Innovation

Vol 17, Issue 4, 2024

Online - 2455-3891
Print - 0974-2441

Research Article

RAPID ON-SITE EVALUATION AND MICRONUCLEUS SCORING IN BREAST LESIONS FINE NEEDLE ASPIRATION CYTOPATHOLOGY BY NEW IAC YOKOHAMA SYSTEM OF REPORTING

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Received: 10 March 2023, Revised and Accepted: 10 December 2023

ABSTRACT

Objective: The objective of the study is to report all fine needle aspiration cytopathology (FNAC) cases of lump breast as per New Yokohama classification 2016, to highlight the importance of rapid onsite evaluation (ROSE) in breast cytology at the time of FNAC, and to assess the relationship between micronucleus (MN) scoring and various breast lesions in cytology.

Methods: The study was conducted on 70 patients with breast lumps in the Department of Pathology in Government Medical College, Patiala, and comprised evaluation of ROSE and MN scoring in breast FNAC by the use of New Yokohama system of reporting.

Results: On ROSE, 11 out of 70 patients were categorized under category I according to the Yokohama system of reporting and on the same sitting with repeat FNAC after performing the ROSE; nine cases were upgraded for cytological reporting and two remained in category I. According to as per the International Academy of Cytology (IAC) Yokohama system of reporting (2016) cases were categorized as category I – 2 (2.9%), category II – 46 (65.7%), category III – 7 (10.1%), category IV – 5 (7.1%), and category V – 10 (14.2%). In MN scoring, MN with the highest score of 14–17/1000 epithelial cells was in category V (malignant), and the lowest score was 0–1/1000 epithelial cells in category II (benign).

Conclusion: ROSE is an easy, safe, and cost-effective method. IAC Yokohama system for reporting (2016) provides a comprehensive way of categorizing various breast lesions on FNAC with clinical correlation. MN score is a good biomarker in differentiating benign, atypical, and malignant breast lesions.

Keywords: Rapid onsite evaluation, Micronucleus, Fine needle aspiration cytology, Yokohama.

 DOI: http://dx.doi.org/10.22159/ajpcr.2024v17i4.47786. Journal homepage: https://innovareacademics.in/journals/index.php/ajpcr

INTRODUCTION

Breast lesions are heterogeneous diseases that are mainly seen in females. In general, benign breast lesions are more common than malignant breast lesions. 81.6% comprised benign lesions whereas 18.3% constituted malignant lesions. The most common benign lesion is fibroadenoma and the most common malignant lesion is invasive ductal carcinoma (IDC) [1]. Breast fine needle aspiration cytology (FNAC) is the most common, rapid, and minimally invasive procedure used in developing countries for diagnosing breast lesions. The International Academy of Cytology (IAC) developed the Yokohama system for reporting breast FNAC in 2016. It defines five categories based on the risk of malignancy: (1) Insufficient/inadequate. (2) Benign. (3) Atypical. (4) Suspicious of malignancy. (5) Malignant [2,3]. During the past few decades, micronucleus (MN) assay has become an important method to assess genotoxicity [4]. The International Human MN project was launched in 1997 to predict genomic damage by MN in lymphocytes and exfoliated buccal cells in humans [5-7]. MN is counted in 1000 epithelial cells on breast cytology smears under oil immersion [8]. Rapid onsite evaluation (ROSE) obtained through fine needle aspiration (FNA) can have a positive impact on the time and accuracy of diagnostic procedures and reduce the number of patient visits [9,10].

METHODS

All patients who attended the surgery outpatient department with complaints of breast lumps and were referred to the Pathology Department at Rajindra Hospital, Patiala, for fine needle aspirates were included in our study.

Inclusion criteria

All patients coming in the Department of Pathology with breast lumps.

Exclusion criteria

Already diagnosed cases of CA breast and on therapy.

Procedure

In this study, FNAC smears of breast lesions were collected from 70 female patients who reported to the Department of Pathology. Patients written consent was taken and FNA was done using a 22-gauze needle, 20 ml disposable syringe. Cellular material was aspirated into the syringe and smeared onto slides. One slide of each patient was prepared for ROSE method with the help of a Diff-Quick stain. The smears prepared were observed and the final diagnosis was made and classified as per IAC New Yokohama system for reporting MN was counted on the prepared slide (MN in 1000 epithelial cells under oil immersion) for further evaluation.

RESULTS AND DISCUSSION

Table 1 shows after performing ROSE on FNAC 11 cases out of 70 came out to be unsatisfactory (Fig. 1).

Table 2 shows that nine out of 11 category 1 cases after repeat FNAC were upgraded and two remained unsatisfactory.

Table 3 shows that the majority of the patients in the study population fall in category II (65.7%) followed by category V (14.2%).

Table 4 shows the cytological diagnosis of IAC Yokohama categories. Maximum MN score observed in category V (malignant) (Figs. 2 and 3) ranging from 14 to 17 followed by category IV (suspicious for malignancy).

Table 5 shows the majority of the patients in the study population falls in 0–1 MN score (68.6%) (Fig. 4).

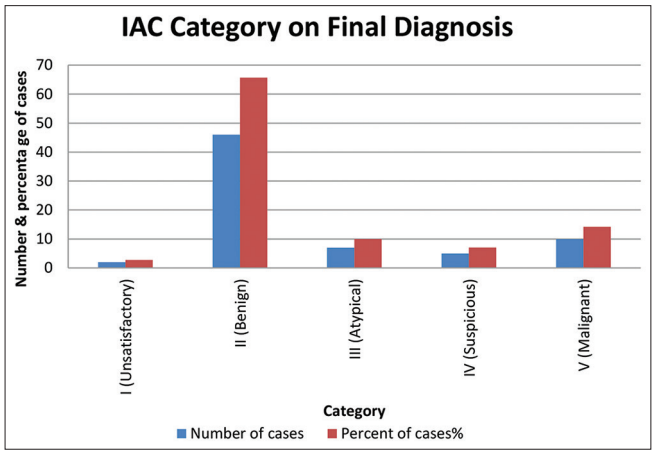

Fig. 1: Distribution of cases according to the International Academy of Cytology Yokohama classification on final diagnosis

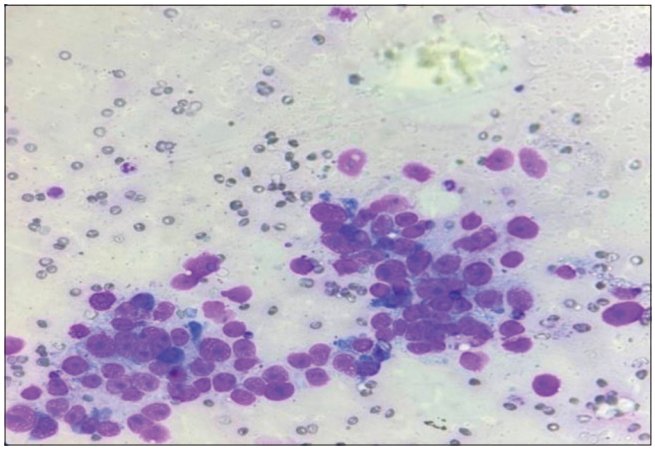

Fig. 2: Photomicrograph showing malignant cells. (Diff-Quick stain; ×40)

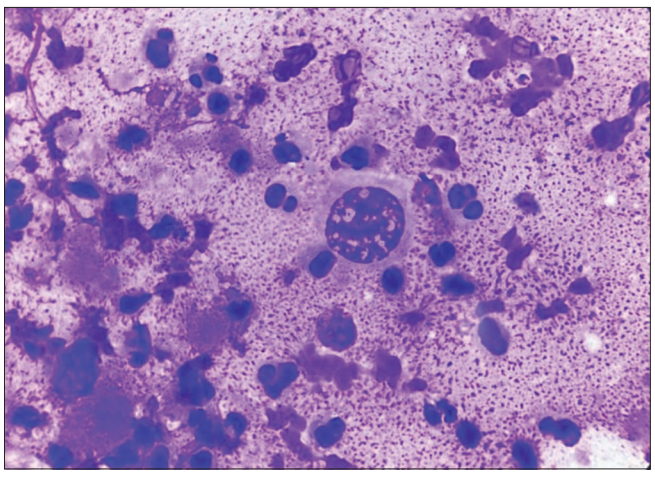

Fig. 3: Photomicrograph showing micronucleus in category V (malignant). (CS; MGG ×1000 Oil immersion)

Table 6 shows the comparison between ROSE and final diagnosis by IAC Yokohama showing that category I (unsatisfactory) cases decreased after performing ROSE and category V cases increased after performing ROSE. Overall p-value by Fisher exact test came out to be 0.016 (<0.05) showing statistically significant results.

Table 7 shows a comparison between IAC Yokohama categories and MN scoring in different categories. In category I (unsatisfactory), p-value

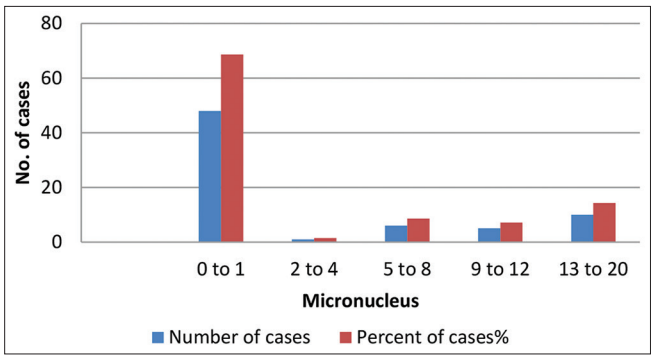

Fig. 4: Distribution of the cases according to micronucleus scoring

Table 1: Distribution of the study population after performing rapid on-site evaluation on fine needle aspiration cytopathology

ROSE on 70 cases	Unsatisfactory (category I)	Satisfactory	Total
Number of cases	11	59	70

ROSE: Rapid on-site evaluation, FNAC: Fine needle aspiration cytopathology

Table 2: Distribution of the study population after repeat fine needle aspiration cytopathology on cases which were under category I

On repeat FNAC on 11 unsatisfactory cases	Unsatisfactory	Satisfactory
Number of cases	2	9

FNAC: Fine needle aspiration cytopathology

Table 3: Distribution of the study population according to the International Academy of Cytology Yokohama classification on 70 cases of fine needle aspiration cytopathology for final diagnosis

IAC category	Number of cases	Percent of cases %
I (unsatisfactory)	2	2.9
II (benign)	46	65.7
III (atypical)	7	10.1
IV (suspicious)	5	7.1
V (malignant)	10	14.2
Total	70	100

IAC: International Academy of Cytology

came out to be 0.154 (statistically not significant) and in category II, III, IV, and V cases, p-value came out to be <0.05 showing statistically significant results.

Table 8 shows that micronucleus scoring can be used effectively to differentiate benign, atypical, suspicious malignancy, and malignant breast lesions (p≤0.001).

A MN is a small additional nucleus readily identifiable by light microscopy because it is morphologically identical to but smaller than the main nucleus [11]. This study showed that the maximum number of cases was 70 on which ROSE was performed and showed that 11 cases were unsatisfactory (category I) by IAC Yokohama classification and 59 were satisfactory for evaluation which was in comparison to the study done by: Liew *et al.* [12] and Wong *et al.* [9] According to IAC Yokohama reporting system, the cases were

Table 4: Distribution of the study population according to International Academy of Cytology Yokohama classification with cytological diagnosis and micronucleus score

Category	Number of cases (%)	Cytological diagnosis	Number of cases (%)	MN score/1000 epithelial cells
Category I	2 (2.8)	Unsatisfactory	2 (2.8)	0
Category II	46 (65.7)	Fibroadenoma	16 (22.8)	0
		Cellular fibroadenoma	3 (4.2)	0–1
		Fibrocystic disease	9 (12.8)	0
		Benign breast disease	10 (14.2)	0–1
		Mastitis	8 (11.4)	0–1
Category III	7 (10)	Atypical ductal hyperplasia	7 (10)	5–8
Category IV	5 (7.1)	Suspicious for malignancy	5 (7.1)	9–10
Category V	10 (14.2)	Malignant	10 (14.2)	14–17

MN: Micronucleus

Table 5: Distribution of the study population according to micronucleus scoring

MN scoring	Number of cases	Percent of cases
0–1	48	68.6
2–4	1	1.4
5–8	6	8.6
9–12	5	7.1
13–20	10	14.3
Total	70	100

MN: Micronucleus

Table 6: Comparison between rapid on-site evaluation and final diagnosis by International Academy of Cytology Yokohama category

IAC categories	ROSE	Final diagnosis
I	11 (15.7)	2 (2.9)
II	40 (57.1)	46 (65.7)
III	3 (4.3)	7 (10.0)
IV	11 (15.7)	5 (7.2)
V	5 (7.2)	10 (14.2)
Total	70 (100.0)	70 (100.0)
Fisher's exact test	12.054	
p-value	0.016* (<0.05)	
Significance	S	

*Statistically significant difference using threshold P=0.05. ROSE: Rapid on-site evaluation, IAC: International Academy of Cytology

Table 7: Comparison between the International Academy of Cytology Yokohama categories and micronucleus scoring

Categories	IAC Yokohama results	MN results	p	Significance
I	2	0	0.154	NS
II				
Fibroadenoma	16	0	<0.001*	HS
Cellular fibroadenoma	3	2	0.005*	Significant
Fibrocystic	9	0	0.003*	HS
Benign breast cancer	10	1	0.009*	HS
Mastitis	8	3	0.005*	Significant
III	7	7	0.001*	HS
IV	5	5	0.001*	HS
V	10	10	0.001*	HS
Total	70 (100.0)	70 (100 0)		

The statistically significant difference using threshold *P*=0.05. IAC: International Academy of Cytology, MN: Micronucleus, HS: Highly significant

arranged in the descending order as Benign – category II – 65.7%, malignant – category V–14.2%, atypical – category III – 10%, suspicious of malignancy – category IV – 7.1%, and insufficient – Category I – 2.8%. The distribution of the study population according to the Yokohama Classification was similar to the studies done by Ibikunle *et al*. [13], Montezuma *et al*. [14], Chauhan *et al*. [15], Agrawal *et al*. [16], and Dixit *et al*. [17]. Distribution of the study population according to IAC Yokohama classification with cytological diagnosis and MN score which was comparable to the study done by Sylvia *et al*. [8]. Comparison between ROSE and final diagnosis by IAC Yokohama classification in the study population was comparable to the studies done by Wong *et al*. in 2019 [9] and Agrawal *et al*. 2021 [16]. Comparison between IAC Yokohama results and MN results were comparable to the study done by Sylvia *et al*. [8], Hemalatha *et al*. [18], Samanta *et al*. [11] showing results to be statistically significant (p<0.05). On comparative analysis, the present study found that MN scoring can be used effectively in differentiating various benign and malignant breast lesions, benign and atypical, benign and suspicious for malignancy (p<0.001) and was comparable to the study done by Katta *et al* [19].

Table 8: Comparative analysis of micronucleus scores between various categories of breast lesions

Cytological categories	Number of cases	MN score cases	p*-value
Benign	46	6	<0.001
Malignant	10	10	
Benign	46	6	<0.001
Atypical favoring benign	7	7	
Benign	46	6	<0.001
Suspicious for malignancy	5	5	

*Statistically significant difference using threshold *P*=0.05. MN: Micronucleus

SUMMARY

1. A total of 70 FNAC were done on patients with palpable breast lump in the age range of 10–80 years in the Department of Pathology, GMC Patiala for a period of 2 years (2020–2022)
2. The most common age group of breast lesions was from 21 to 30 years (34.2%). The youngest female was 13 years and the oldest was 78 years
3. Both ROSE and conventional cytology smears were prepared for all 70 fine needle aspiration samples and categorized for reporting with the IAC Yokohama System (2016) and MN scoring was done
4. In 70 ROSE cases, 11 cases were in category I (unsatisfactory) and on same sitting with repeat FNAC after performing the ROSE; 9 cases were upgraded for cytological reporting and 2 remained in category I of IAC Yokohama system of reporting (2016).
5. According to the IAC Yokohama system of reporting (2016) these cases were categorized as category I – 2 (2.9%), category II- 46 (65.7%), category III – 7 (10.1%), category IV – 5 (7.1%), and category V – 10 (14.2%).
6. In MN scoring, MN with the highest score of 14–17/1000 epithelial cells was in category V (Malignant) followed by 9–10/1000 epithelial cells in category IV (Suspicious for malignancy),5–8/1000 epithelial cells in category III (atypical), 0–1/1000 epithelial cells in category II (benign), 0/1000 epithelial cells in category I (Unsatisfactory). Hence

MN score proved as an important indicator of chromosomal damage and served as additional criteria for differentiating benign, atypical, and malignant breast lesions
7. Statistical comparison of ROSE and final diagnosis on IAC Yokohama (2016), was found to be significant ($p \leq 0.05$)
8. Significant Statistical comparison was also seen in the final diagnosis on IAC Yokohama and MN score ($p \leq 0.05$).

CONCLUSION

ROSE is an easy, safe, and cost-effective method as it reduces the visits of the patients to the laboratory and benefits in improving the adequacy rate and diagnosis. IAC Yokohama System for reporting (2016) provides a comprehensive way of categorizing various breast lesions on FNAC with clinical correlation. MN score is a good biomarker in differentiating benign, atypical, and malignant breast lesions.

CONFLICTS OF INTERESTS

None.

AUTHORS FUNDING

Nil.

REFERENCES

1. Yogalakshmi S, Kavitha M. A study of histopathological spectrum of breast lesions. Int J Sci Stud. 2019;7(1):1-5.
2. Field AS, Raymond WA, Rickard M, Arnold L, Brachtel EF, Chaiwun B, *et al*. The international academy of cytology Yokohama system for reporting breast fine-needle aspiration biopsy cytopathology. Acta Cytol. 2019;63(4):257-73. doi: 10.1159/000499509, PMID 31112942
3. Field AS, Schmitt F, Vielh P. IAC Standardized reporting of breast fine-needle aspiration biopsy cytology. Acta Cytol. 2017;61(1):3-6. doi: 10.1159/000450880, PMID 27806362
4. Sommer S, Buraczewska I, Kruszewski M. Micronucleus assay: The state of art, and future directions. Int J Mol Sci. 2020 Feb 24;21(4):1534. doi: 10.3390/ijms21041534, PMID 32102335
5. Fenech M, Holland N, Zeiger E, Chang WP, Burgaz S, Thomas P, *et al*. The HUMN and HUMNxL international collaboration projects on human micronucleus assays in lymphocytes and buccal cells--past, present and future. Mutagenesis. 2011;26(1):239-45. doi: 10.1093/mutage/geq051, PMID 21164208
6. Holland N, Bolognesi C, Kirsch-Volders M, Bonassi S, Zeiger E, Knasmueller S, *et al*. The micronucleus assay in human buccal cells as a tool for biomonitoring DNA damage: The HUMN project perspective on current status and knowledge gaps. Mutat Res. 2008 Jul-Aug;659(1-2):93-108. doi: 10.1016/j.mrrev.2008.03.007, PMID 18514568
7. Fenech M, Holland N, Chang WP, Zeiger E, Bonassi S. The HUman MicroNucleus Project--An international collaborative study on the use of the micronucleus technique for measuring DNA damage in humans. Mutat Res. 1999;428(1-2):271-83. doi: 10.1016/s1383-5742(99)00053-8, PMID 10517999
8. Sylvia MT, Baskaran L, Bhat RV. Micronucleus study on breast cytology aspirate smears and its diagnostic utility. J Cytol. 2018;35(1):22-6. doi: 10.4103/JOC.JOC_160_16, PMID 29403165
9. Wong S, Rickard M, Earls P, Arnold L, Bako B, Field AS. The International Academy of Cytology Yokohama System for reporting breast fine needle aspiration biopsy cytopathology: A single institutional retrospective study of the application of the system categories and the impact of rapid onsite evaluation. Acta Cytol. 2019;63(4):280-91. doi: 10.1159/000500191, PMID 31108486
10. Schmidt RL, Witt BL, Lopez-Calderon LE, Layfield LJ. The influence of rapid onsite evaluation on the adequacy rate of fine-needle aspiration cytology: A systematic review and meta-analysis. Am J Clin Pathol. 2013;139:300-8.
11. Samanta S, Dey P, Nijhawan R. The role of micronucleus scoring in fine needle aspirates of ductal carcinoma of the breast. Cytopathology. 2011 Apr;22(2):111-4. doi: 10.1111/j.1365-2303.2010.00773.x, PMID 20553316
12. Liew PL, Liu TJ, Hsieh MC, Lin HP, Lu CF, Yao MS, *et al*. Rapid staining and immediate interpretation of fine-needle aspiration cytology for palpable breast lesions: Diagnostic accuracy, mammographic, ultrasonographic and histopathologic correlations. Acta Cytol. 2011;55(1):30-7. doi: 10.1159/000320869, PMID 21135519
13. Ibikunle DE, Omotayo JA, Ariyibi OO. Fine needle aspiration cytology of breast lumps with histopathologic correlation in Owo, Ondo State, Nigeria: A five-year review. Ghana Med J. 2017;51(1):1-5. doi: 10.4314/gmj.v51i1.1, PMID 28959065
14. Montezuma D, Malheiros D, Schmitt FC. Breast fine needle aspiration biopsy cytology using the newly proposed IAC Yokohama system for reporting breast cytopathology: The experience of a single institution. Acta Cytol. 2019 Feb 15;1:1-6. doi: 10.1159/000492638, PMID 30783035
15. Chauhan V, Pujani M, Agarwal C, Chandoke RK, Raychaudhuri S, Singh K, *et al*. IAC standardized reporting of breast fine-needle aspiration cytology, Yokohama 2016: A critical appraisal over a 2 year period. Breast Dis. 2019;38(3-4):109-15. doi: 10.3233/BD-190393, PMID 31524134
16. Agrawal N, Kothari K, Tummidi S, Sood P, Agnihotri M, Shah V. Fine-needle aspiration biopsy cytopathology of breast lesions using the international academy of cytology Yokohama system and rapid on-site evaluation: A single-institute experience. Acta Cytol. 2021;65(6):463-77. doi: 10.1159/000518375, PMID 34515039
17. Dixit N, Trivedi S, Bansal VK. A retrospective analysis of 512 cases of breast fine needle aspiration cytology utilizing the recently proposed IAC Yokohama system for reporting breast cytopathology. Diagn Cytopathol. 2021 Sep;49(9):1022-31. doi: 10.1002/dc.24808, PMID 34133084
18. Hemalatha A, Suresh TN, HarendraKumar ML. Micronuclei in breast aspirates. Is scoring them helpful? J Cancer Res Ther. 2014 Apr-Jun;10(2):309-11. doi: 10.4103/0973-1482.136588, PMID 25022383
19. Katta R, Rao KN, Murthy BK, Kalam SR. Micronucleus scoring in fine needle aspiration cytology of breast lesions-a retrospective analytical study. J Clin Diagn Res. 2022 Aug;16(8):EC52-5.