ABSTRACT
Background: In 1996, National Cancer Institute (NCI) proposed five categories for the diagnosis of breast cytology in order to bring a degree of uniformity to the diagnostic reporting. Of these, categories 3 and 4 were sparsely studied.
Aims: The present study was undertaken for the evaluation of the fine needle aspiration cytology (FNAC) categories of C3 and C4 in the breast lump and its histopathological correlation.
Materials and Methods: In the retrospective study, a total number of 728 FNACs were categorized according to the NCI; of these, 28 cases of category C3 and 65 cases of category C4 were compared with histopathological diagnoses.
Results: On histopathological examination of category C3, 18 (64.3%) cases showed benign lesions and 10 (35.7%) cases showed malignancy and among the C4 category, benign lesions found in nine (13.8%) and malignancy found in 56 (86.2%) cases. There was a significant statistical difference between the number of benign and malignant diagnoses for cytological categories of C3 (64.3%) and C4 (86.2%) ($P < 0.001$). The sensitivity, specificity, positive predictive value, and negative predictive value of C4 category in the diagnoses of malignancy were 84.8%, 66.7%, 86.2%, and 64.3%, respectively.
Conclusion: FNAC of the breasts is a simple, cost-effective, accurate, quick, and relatively less painful procedure, and it continues to play an integral part in the management of breast lesions. Our study concluded that there was a statistically significant difference between the number of benign and malignant diagnoses for categories of C3 and C4.

Key words: Atypical, suspicious; breast lump; breast masses; fine needle aspiration cytology (FNAC); histopathological correlation

Introduction

The application of fine needle aspiration cytology (FNAC) in the diagnosis of palpable breast masses was first introduced by Martin and Ellis in 1930 and since then has been established as an important tool in the evaluation of breast lesions.[1] Breast cancer is the second most common cancer among Indian females after cancer cervix.[2] FNAC is a simple, cost-effective, less traumatic, sensitive, and rapid diagnostic method.[3]

According to the National Cancer Institute (NCI) guidelines in 1996, FNAC of the breasts were categorized into inadequate (C1), benign (C2), atypical, probably benign (C3), suspicious, favor malignancy (C4), and malignant (C5).[4] NCI recommended these categories in order to bring a degree of uniformity to the diagnostic reporting. Categories such as C1, C2, and C5 are usually straightforward and do not generally pose difficulties to pathologists. However, interpretation of C3 and C4 categories are difficult and
confusing, because they do not have strict criteria for diagnosis. Studies done by Kanhoush et al.[5] and Howell[6] suggested the use of the single term called equivocal to describe the category C3 and C4. The present study was undertaken for the evaluation of the FNAC categories of C3 and C4 in the breast lump and its histopathological correlation.

Materials and Methods

The present retrospective study was carried out at the Department of Pathology in our institution, a tertiary care hospital, conducted from March 2012 to April 2014, after obtaining approval from the Institutional Ethics Committee. May Grünwald Giemsa and hematoxylin and eosin stained smears of 728 patients, who had undergone FNAC for breast lump were included.

NCI guidelines (1996) was used for cytological evaluation[4] and patients diagnosed as atypical, probably benign (C3), suspicious, favor malignancy (C4) were selected for further study. Biopsies from subsequent operations by excisional biopsy or mastectomy specimens on these cases were reviewed for correlation with the FNAC diagnoses. When the smears have characteristics of a benign aspirate with any or a combination of nuclear pleomorphism, some loss of cellular cohesiveness or nuclear and cytoplasmic changes was considered category C3.[7] When the aspirates showing some cells with features of malignancy, where the material is not diagnostic of malignancy due to scanty, poorly preserved specimen or aspirates showing some malignant features of greater than those observed in category C3 without the presence of overtly malignant cells or aspirates showing an overall benign pattern with large numbers of naked nuclei and/or cohesive sheets of cells, but with occasional cells showing distinct malignant features was considered category C4.[7] On correlation of FNAC diagnoses of categories C3 and C4 with histopathological diagnoses, the sensitivity, specificity, accuracy, false positive rate, false negative rate, positive predictive value, and negative predictive value of category C4 of FNAC were calculated. Chi-square test was used to assess the diagnostic value of cytological diagnosis by comparing the percentage of benign and malignant histological diagnosis in categories C3 and C4. The P value of 0.05 or less was considered for statistical significance. All the statistical analysis was performed using International Business Machines Corporation (IBM) Statistical Package for the Social Sciences (SPSS) Statistics for Windows (version 20.0. Armonk, New York: IBM Corporation).

Results

In total, 728 cases of FNAC slides were reviewed and categorized according to the NCI guidelines (1996). Of these, categories C3 and C4 consisted of 29 (4%) and 68 (9.3%) cases, respectively. Histopathological follow-up was available in 28 (96.6%) cases of category C3 and 65 cases (95.6%) of category C4, and were selected for the present study.

The mean age of patients was 32 years with a range from 26-74 years. On histopathological examination of category C3, 18 (64.3%) cases showed benign lesions and 10 (35.7%) cases showed malignancy [Figure 1a and b]. Among the C4 category, nine (13.8%) cases showed benign lesions [Figure 2a and b] and 56 (86.2%) cases showed malignancy [Table 1]. In the benign lesions of both categories C3 and C4 (18, 64.3% and 9, 13.8%), fibroadenoma (10, 55.6% and 6, 66.7%) was most common followed by fibrocystic disease

| Table 1: Correlation of FNAC diagnoses with histopathological diagnoses |
|-----------------------------------------------|
| Histopathological diagnoses        | FNAC Category | Total |
|-----------------------------------------------|
| Benign                              | C3            | C4     | Total |
| Benign                              | 18 (64.3%)    | 9 (13.8%) | 27    |
| Malignant                           | 10 (35.7%)    | 56 (86.2%) | 66    |
| Total                               | 28            | 65     | 93    |

Figure 1: False negative case — Atypical, probably benign (C3) turned out to be malignancy. (a) Cytosmear showing ductal epithelial cluster with some loss of cohesion and mild nuclear atypia diagnosed as atypical, probably benign (C3) (H and E, x100). (Inset, upper right H and E, x400). (b) Follow-up histopathological section showing invasive ductal carcinoma — Not otherwise specified (H and E, x400)

Figure 2: False positive case — Suspicious, probably malignant (C4) turned out to be benign. (a) Cellular cytosmear exhibiting slight loss cohesiveness of ductal epithelial cell clusters with moderate nuclear atypia suggestive of suspicious, probably malignant (C4) (H and E, x400) (b) Subsequent histopathological examination revealing features of typical fibroadenoma (H and E, x400)
with atypia (8, 44.4% and 3, 33.3%) and in cases of malignancy of categories C3 and C4 (10, 35.7% and 56, 86.2%), invasive ductal carcinoma (IDC) (8, 80% and 54, 96.4%) was most common followed by invasive lobular carcinoma (ILC) (2, 20% and 2, 3.6%).

There was a significant statistical difference between the number of benign and malignant diagnoses for categories C3 (64.3%) and C4 (86.2%) (P < 0.001). The sensitivity, specificity, positive predictive value, and negative predictive value of C4 category in the diagnosis of malignancy were 84.8%, 66.7%, 86.2%, and 64.3%, respectively.

Discussion

FNAC of breast masses is widely accepted as a reliable diagnostic tool with high sensitivity and specificity. Correlation of FNAC results with the clinical and imaging studies is referred as the triple test[8-10] and using this approach, some studies have demonstrated 100% diagnostic accuracy.

The age range of the patients in this study was 26-74 years, almost comparable to the study by Chaiwan et al.,[11] in which the age range was 21-9 years. Categories C3 and C4 comprised of 97 (13.3%) among 728 cases in the present study. This proportion were in the range (4-17.7%) as reported by others,[5,11-17] indicating that these categories were not underused or overused in our institution.

Histopathological examination of category C3 showed malignancy in 10 (35.7%) cases which was considered false negative in the present study. This result was comparable with most of the published studies that range from 8.6% to 52% and in most of these studies, the proportion of malignancy in category C3 were over 30%.[5,13,17,18] Small tumor size, hypocellularity, sampling error during FNAC, few histological tumor types viz low nuclear grade, lobular carcinoma, scirrhous carcinoma, and well differentiated intracystic carcinoma are contributing factors for false negative results. Sampling error particularly in small tumor is the most common cause of false negative results.[19] We reviewed our false negative cases (eight IDC and two ILC) meticulously after histopathological examination and all of these cases showed predominantly benign-appearing cohesive monolayered epithelial cells and scattered bare nuclei with few discohesive clusters revealed nuclear atypia and nuclear crowding. The false negative rate can be reduced or eliminated by triple test that correlates the FNAC results with clinical and radiological studies.[8,9] Hence, the present study additionally supports the recommendation of other workers,[11,17,20] that use of triple test is important for the management of patients in category C3. The remaining 18 (64.3%) cases followed the original cytological diagnoses similar to Chaiwan et al. (14, 64%) and Goyal et al. (15, 62.5%) studies [11,17]

In the category C4, 56 (86.2%) cases were turned out to be malignant on histopathological examination. This result were comparable to previously published studies that range from 81% to 97%.[5,13,15,18] The most common malignancy found in both categories C3 and C4 were invasive ductal carcinoma followed by invasive lobular carcinoma, similar to recently published studies.[11,17] Because the rate of malignancy in the category C4 was high, the lesion should always be subjected to histopathological examination for the confirmation of malignancy, as suggested by some authors.[5,17] Nine (13.8%) cases of category C4 revealed benign lesions on histopathological examination, which was considered as false positive in the current study. Of these, six cases were diagnosed as fibroadenoma and three cases were fibrocystic disease with atypia. FNAC of these cases showed high cellularity and slight loss of cohesion with moderate nuclear atypia made us diagnose malignancy.

Some experts found that maintenance of category C3 and C4 is beneficial, because it identifies groups of patients who are more likely to have either benign (C3) or malignant outcomes. Large studies with histopathological correlation of category C3 revealed 48-64% of benign lesions, whereas category C4 showed malignancy in 81-87.5%. A significant proportion of category C3 showed malignancy (36-52%) and smaller percentage (12.5-19%) of category C4 revealed benign lesions as well.[5,11,17] Furthermore, patients with category C3 lesions may not necessarily be subjected to surgical biopsies if their clinical examination and mammographic findings are suggestive of benign outcomes as well. A repeat FNAC or core needle biopsy may be done after at least 1 month allowing for reactive changes to subside. If the second biopsy is benign, then no surgical management or close follow-up are an option.[11,18] However, patients with category C4 lesions should always be subjected for histopathological examination because of a higher malignancy rate. In our study, we found 64.3% of benign lesions in category C3 and 86.2% of malignancy in category C4. This difference was statistically significant (P = 0.001).

The sensitivity, specificity, positive predictive value, and negative predictive value of C4 category in the diagnosis of malignancy were 84.8%, 66.7%, 86.2%, and 64.3%, respectively. Comparison of sensitivity, specificity, positive predictive value, and negative predictive value of C4 category in the diagnosis of malignancy with published studies is shown in Table 2.
Conclusion

FNAC of the breasts is a simple, cost-effective, accurate, quick, and relatively less painful procedure, and it continues to play an integral part in the management of breast lesions. Our study concluded that there was a statistically significant difference between the number of benign and malignant diagnoses for categories C3 and C4.

Financial support and sponsorship
Nil.

Conflicts of interest
There are no conflicts of interest.

References

1. Ariga R, Bloom K, Reddy VB, Kluskens L, Francescatti D, Dowlat K, et al. Fine-needle aspiration of clinically suspicious palpable breast masses with histopathologic correlation. Am J Surg 2002;184:410-3.

2. Chopra R. The Indian scene. J Clin Oncol 2001;19(Suppl):106-11S.

3. Nasuti JF, Gupta PK, Baloch ZW. Diagnostic value and cost-effectiveness of on-site evaluation of fine-needle aspiration specimens: Review of 5,688 cases. Diagn Cytopathol 2002;27:1-4.

4. The uniform approach to breast fine-needle aspiration biopsy. National Cancer Institute Fine-Needle Aspiration of Breast Workshop Subcommittees. Diag Cytopathol 1997;16:295-311

5. Kanhough R, Jorda M, Gomez-Fernandez C, Wang H, Mirzabeigi M, Ghorab Z, et al. Atypical and suspicious diagnoses in breast aspiration cytology-is there a need for two categories? Cancer 2004;102:164-7.

6. Howell LP. Equivocal diagnoses in breast aspiration biopsy cytology: Sources of uncertainty and the role of “atypical/indeterminate” terminology. Diag Cytopathol 1999;21:217-22.

7. Shabb NS, Boulos FI, Abdul-Karim FW. Indeterminate and erroneous fine-needle aspirates of breast with focus on the ‘true gray zone’: A review. Acta Cytol 2013;57:316-31.

8. Kachewar SS, Dongre SD. Role of triple test score in the evaluation of palpable breast lump. Indian J Med Paediatr Oncol 2015;36:123-7.

9. Ghimire B, Khan MI, Bihhusal T, Singh Y, Sayami P. Accuracy of triple test score in the diagnosis of palpable breast lump. JNMA J Nepal Med Assoc 2008;47:189-92.

10. Ahmed I, Nazir R, Chaudhary MY, Kundi S. Triple assessment of breast lump. J Coll Physicians Surg Pak 2007;17:535-8.

11. Chaiwun B, Sukhamwang N, Lekawanyijit S, Sukapan K, Rangdaeng S, Muttarak M, et al. Atypical and suspicious categories in fine needle aspiration cytology of the breast: Histological and mammographical correlation and clinical significance. Singapore Med J 2005;46:706-9.

12. Nguansangiam S, Jesdapatarakul S, Tangjitgamol S. Accuracy of fine needle aspiration cytology from breast masses in Thailand. Asian Pac J Cancer Prev 2009;10:623-6.

13. Deb RA, Matthews P, Elston CW, Ellis IO, Pinder SE. An audit of “equivocal” (C3) and “suspicious” (C4) categories in fine needle aspiration cytology of the breast. Cytopathology 2001;12:219-26.

14. Lim JC, Al-Masri H, Salhadar A, Xie HB, Gabram S, Wojcik EM. The significance of the diagnosis of atypia in breast fine-needle aspiration. Diagn Cytopathol 2004;31:285-8.

15. Chaiwun B, Settakorn J, Ya-In C, Wisedmongkol W, Rangdaeng S, Thorner P. Effectiveness of fine-needle aspiration cytology of breast: Analysis of 2,375 cases from northern Thailand. Diagn Cytopathol 2002;26:201-5.

16. Kim A, Lee J, Choi JS, Won NH, Koo BH. Fine needle aspiration cytology of the breast. Experience at an outpatient breast clinic. Acta Cytol 2000;44:361-7.

17. Goyal P, Sehgal S, Ghosh S, Aggarwal D, Shukla P, Kumar A, et al. Histopathological Correlation of Atypical (C3) and Suspicious (C4) Categories in fine needle aspiration cytology of the breast. Int J Breast Cancer 2013;2013:965498.

18. Yeoh GP, Chan KW. Fine needle aspiration of breast masses: An analysis of 1533 cases in private practice. Hong Kong Med J 1998;4:283-8.

19. Silverman JF, Saad RS. Breast. In: Bibbo M, Wilbur D, editors. Comprehensive Cytopathology. 3rd ed. Philadelphia, PA: Saunders Elsevier; 2008. p. 713-72.

20. Brenner RJ, Bassett LW, Fajardo LL, Dershaw DD, Evans WP 3rd, Hunt R, et al. Stereotactic core-needle breast biopsy: A multi-institutional prospective trial. Radiology 2001;218:866-72.

21. Yusuf I, Aranda AT, Imam MI. Cyto-morphologic correlation of equivocal C3 and C4 breast lesions. Arch Int Surg 2014;4:131-5.

Table 2: Comparison of results of present study with previous study

| Study          | Year of study | Number of patients | Sensitivity (%) | Specificity (%) | PPV (%) | NPV (%) |
|----------------|--------------|--------------------|-----------------|-----------------|---------|---------|
| Goyal et al.[17]| 2012         | 40                 | 60.8            | 88.2            | 87.5    | 62.5    |
| Yusuf et al.[21]| 2012         | 47                 | 76.7            | 76.5            | 85.2    | 65      |
| Present study  | 2014         | 93                 | 84.8            | 66.7            | 86.2    | 64.3    |

PPV: Positive predictive value, NPV: Negative predictive value