Original Article

The Role of Fine Needle Aspiration Cytology in Triple Assessment of Patients with Malignant Breast Lumps

A. Ugwu-Olisa Ogbuanya[1,2], S. Nnamdi Anyanwu[3,4], E. Festus Iyare[5,6], C. Gregory Nwigwe[1,2]

[1]Department of Surgery, Alex Ekwueme Federal University Teaching Hospital, Abalaliki, Ebonyi State, Nigeria, [2]Department of Surgery, Ebonyi State University, Abakaliki, Ebonyi State, Nigeria, [3]Department of Surgery, Nnamdi Azikiwe University Teaching Hospital, Nnewi, Anambra State, Nigeria, [4]Department of Surgery, Faculty of Medicine, Nnamdi Azikiwe University, Awka, Anambra State, Nigeria, [5]Department of Pathology, Alex Ekwueme Federal University Teaching Hospital, Abalaliki, Ebonyi State, Nigeria, [6]Department of Pathology, Ebonyi State University, Abakaliki, Ebonyi State, Nigeria

Abstract

Background: Breast cancer is the leading female malignancy in the world and now the most common cancer in Nigeria. The evaluation of patients with breast cancer requires stepwise diagnostic approach to achieve a combined diagnostic information required to make an enhanced decision on how best to approach management. The aim of this study is to evaluate the role of fine needle aspiration cytology (FNAC) in the triple assessment of patients with malignant breast lumps in our center. **Patients and Methods:** This is a descriptive prospective study of patients with palpable breast lumps over an eighteen-month period. The patients were sequentially subjected to FNAC and open biopsy. Those whose lumps were malignant were further evaluated. **Results:** There were 88 (44.9%) and 108 (55.1%) biopsy confirmed breast cancers and benign lumps, respectively. From cytology reports, there were 12 (6.1%) unsatisfactory (C1), 96 (49%) benign (C2), 8 (4.1%) atypical (C3), 10 (5.1%) suspicious of malignancy and 70 (35.7%) unequivocally malignant (C5) smears. FNAC performed better than clinical examination in the validity tests for breast malignancy. The diagnostic results for breast malignancies were 97.2% (sensitivity), 98.9% (specificity), 1.4% (false positive rate []), 2.1% (false negative rate), 98.6% (positive predictive value), 97.9% (negative predictive value), and overall diagnostic accuracy of 98.2%. **Conclusion:** Considering the high performance of diagnostic cytology noted above, FNAC has proved itself to be useful and significantly accurate in making diagnosis of breast cancers in our center.

Keywords: *Accuracy,breast,cancer, cytology, fine needle aspiration*

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Introduction

Cancer is expected to rank as the leading cause of death and the single most important barrier to increasing life expectancy in every country of the world in the 21st century.[1] In 2018, an estimated 18.1 million new cancer cases and 9.6 million cancer deaths were projected.[1] Globally, about 25% and 15% of all new cancer cases and cancer deaths, respectively, among females were due to breast cancer.[2] Breast cancer is the leading female malignancy in the world and is now the most common cancer in Nigeria.[2-5]

Considering the pathetic state and magnitude of the ravage caused by breast cancer in the Developing nations, the importance of studies aimed at the evaluation of the quality of diagnostic services for breast cancer in our population cannot be overemphasized.[4,6,7] Managing breast cancers entails major surgical and oncologic implications and therefore requires firm pathological

Address for correspondence: Dr. A. Ugwu-Olisa Ogbuanya, Department of Surgery, Alex Ekwueme Federal University Teaching Hospital, PMB 102, Abakaliki, Ebonyi State, Nigeria. E-mail: ogbuanyaa92@gmail.com

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evaluation. Failure to do this may lead to inappropriate decision such as performing mastectomy for tuberculous or chronic pyogenic mastitis.[7,8]

The benefit of a multidisciplinary approach to the management of patients with breast cancers has been emphasized.[9,10] Under the triple assessment approach, Bishop and colleagues reported that triple test is positive if any of the three components is positive and negative if all the components are negative.[9] Its sensitivity and specificity have been shown to be as high as 99.6% and 93% respectively.[9] In resource poor communities like ours, triple assessment approach on the first clinic visit is commonly unachievable due to inherent unavailability of dedicated imaging suit and the difficult task of conducting core needle biopsy (CNB) on the often poor, ill-guided, anxious patients. These limitations can be attenuated through the use of clinical and fine needle aspiration cytology (FNAC) assessments with imaging component playing a secondary role.[6,8,11] However, studies have shown that combined diagnostic information from FNAC and clinical diagnosis is equally effective.[6,9]

The utilization of FNAC for pre-operative diagnosis of breast cancer has many advantages. Under this scenario, the test is done as an office (outpatient) procedure, without need for anesthesia.[7,9] It is much more minimally invasive, maximally diagnostic, easier to execute, rapid and cheaper compared to formalin fixed, paraffin-embedded histology diagnosis from core needle or open biopsy specimens.[6,7,9,12]

Experience from Calabar, Nigeria showed that a combined diagnostic accuracy (FNAC and clinical examination) exceeds the accuracy of each component when viewed alone.[6] In a prospective study involving 102 patients with breast disease, sensitivities of 90% and 92.4% and specificities of 98.4% and 80.7% for FNAC and clinical examination, respectively, were reported.[6] When both modalities were considered, and found to be in agreement, the combined sensitivity and specificity rose to 97.1% and 100%, respectively, with overall diagnostic accuracy (ODA) of 98.8%.[6] Indeed, since the start of United Kingdom National Health Services Breast Screening Program in the mid and late 80s, FNAC has superseded the use of frozen section examination in the diagnosis and management of patients with breast cancers.[7,13] The aim of this study is to document the role of FNAC in the combined diagnostic assessment of patients with breast cancer.

Patients and Methods

Design and setting

This study is an 18-month prospective study of all consecutive patients with malignant breast lumps seen at the general surgery clinics of our hospital, from October 2011 to March 2013.

Patients

All consecutive 196 patients with palpable breast lumps who gave consent were subjected to clinical examination, breast imaging, FNAC, and open biopsy. However, only the 88 patients with histologically malignant breast lesion and who gave written informed consent were further evaluated. Patients who were below 16 years of age and those without discretely palpable lumps were excluded from the study. Each of the 196 patients was seen, interviewed, and examined by a specialist general surgeon. The biodata and other relevant sociodemographic details were obtained and entered into a pro forma. Each patient was subsequently subjected to a standardized clinical examination, with special emphasis on the breasts and axillae.

The findings on breast examination and an initial clinical impression were entered into the pro forma. Breast ultrasonography was routinely done, but only seventeen breast cancer patients did mammographic studies. This is due to the absence of a dedicated mammographic suit in the breast clinic. Patients with fungating lesions and those with extensive skin and chest wall involvement were not considered for breast imaging on the ipsilateral breasts, but their contralateral breasts were evaluated. Basic and special investigations (when indicated) were equally requested, and their results were recorded.

Before the FNAC procedure, an information sheet [Appendix I], describing the procedure, its benefits and possible complications, was routinely distributed to the patients. Majority of the FNAC tests were carried out by the corresponding author, the rest were done by the pathologist. During the procedure, a 23G disposable hypodermic needle and 10 ml plastic syringe were routinely used. With the needle *in situ* at the designated location of the breast lump and the syringe attached to it, "needling" continued until adequate yield was observed at the needle hub. The material in the needle lumen was expelled onto the center of a pretabled glass slide.

The smears were prepared by gently spreading the aspirate using another slide inclined at 45°. Subsequently, wet-fixed slides were obtained by placing the slides into a coplin jar containing 95% ethanol. These slides were transported to pathology laboratory for staining using Hematoxylin and Eosin (H and E) method and immediate reporting by the cytopathologist. Whenever possible, the lesions were subclassified using specific cytological features. Smears reported as "unsatisfactory" were repeated immediately. The results of the FNAC were discussed with the patients and entered into a pro forma.

The open biopsy used in this study was either excisional, incisional, or wedge biopsy, and majority were done on day case basis. All the open biopsy procedures were done by a specialist general surgeon under local infiltrative anesthesia (majority) or general anesthesia or through combined procedure.

The resection specimens were preserved with 10% formal saline, labeled, and transported to histology laboratory. A single pathologist microscopically examined and reported all the H and E stained slides. Follow-up visits were ordered to review the clinical, radiological, and pathological results. The combined diagnostic information and the strength of each of the triple test components were evaluated and informed decisions were made. Follow-up of available patients were done for 5 years.

Statistical data analysis

This was done using Statistical Package for Social Sciences, software version 22.0 (IBM Chicago, IL,USA, 2015). Descriptive statistics was employed to calculate categorical variables like percentages. The results were presented in tables. Mean, median, and standard deviation (SD) were used to summarize continuous variables. Where appropriate, Chi-square test was used to test for the level of significance of the variables. Confidence interval was calculated at 95% level and significance at 5% probability level ($P < 0.05$). The FNAC validities for breast cancer were calculated using the standard statistical formulae.[14]

Ethical approval

The proposal for the study was approved by the research and ethical committee of the hospital before commencement of the study. We also attest that all human studies from which this manuscript was derived are in compliance with regulations of this institution and with universally accepted guidelines governing such work.

Results

During the study period, a total of 196 (75.4%) of 260 patients with breast complaints presented with palpable lumps and were seen and sequentially subjected to clinical assessment, breast imaging, FNAC, and open biopsy. However, 88 (44.9%) of the 196 patients earlier recruited had malignant diagnosis following histopathology examinations of the breast tissue. Among the breast cancer patients, male breast cancer represents 2.3% (2 patients), but 1% of all patients with breast disease during the period of study. The ages of the 88 breast cancer patients ranged from 18–85 years with a mean of 45.8 ± SD 15.7 years [Table 1].

The histopathological diagnoses of the 88 breast cancers examined in this study are shown in Table 2 below. Over three-fourth (76.1%) of the cancers were due to invasive ductal carcinomas. The distribution of FNAC diagnosis for the 196 patients using open biopsy as reference standard is shown in Table 3 below.

Of the 88 biopsy confirmed breast cancers, clinical examination showed that 7 (7.9%) had benign and 81 (92.1%) had malignant disease. Similarly, FNAC falsely reported that 2 (2.3%) of these cancer patients harbored benign lumps rather than cancers; these misdiagnoses include one fibroadenoma and one mastitis. These were later confirmed by histology to be each malignant phyllodes and papillary carcinoma, respectively. Of the remaining 86 smears, 69 (78.4%) were correctly designated malignant while 17 (19.3%) could not be typed benign or malignant. These 17 undefined FNAC smears (C1, C3, and C4) were later confirmed histologically to consist of six low-grade invasive ductal cancer, three low-grade invasive lobular carcinoma, three

Table 1: Age distribution of breast cancer patients

Age range (years)	Frequency (%)
10-19	2 (2.3)
20-29	10 (11.4)
30-39	18 (20.4)
40-49	24 (27.3)
50-59	20 (22.7)
60-69	8 (9.1)
70-79	4 (4.5)
80-89	2 (2.3)
Total	88 (100.00)

Table 2: Frequency distribution of histological diagnosis

Histology diagnosis	Frequency (%)
Invasive ductal carcinoma	67 (76.1)
Invasive lobular carcinoma	8 (9.1)
Mucinous carcinoma	2 (2.3)
Papillary carcinoma	2 (2.3)
Medullary carcinoma	5 (5.7)
Burkitt's lymphoma	2 (2.3)
Malignant phyllodes	1 (1.1)
Paget's disease of breast	1 (1.1)
Total	88 (100.0)

Table 3: Fine needle aspiration cytology and histopathologic reports of breast lumps

FNAC test		Histology	
Cytology report	Frequency	Benign	Malignant
Inadequate (C1)	12	8	4
Benign (C2)	96	94	2
Atypical (C3)	8	4	4
Suspicious (C4)	10	1	9
Malignant (C5)	70	1	69
Total	196	108	88

FNAC: Fine needle aspiration cytology

medullary carcinoma, two Burkitt's lymphoma, and one each of Paget's disease of breast, papillary carcinoma, and mucinous carcinoma. The diagnostic validities of FNAC and clinical assessment for breast cancers were calculated and their findings recorded [Table 4].

When FNAC was utilized to subclassify the breast cancers into definitive pathologic entities, an overall concordance rate of 87.7% was documented for the 57 slides reported while immense difficulties were encountered in the remaining 31 (35.2%) slides leading to "no diagnosis" in these categories. The three incorrect typing recorded against invasive ductal carcinoma were confirmed by histology to comprise of two invasive lobular cancers and one medullary cancer [Table 5]. The validity results for malignant tumors ≤5 cm include sensitivity (83.3%), specificity (100.0%), false positive rate (FPR) (0.0%), false negative rate (FNR) (2.3%), and ODA of 97.9%. The validities for lumps >5 cm are sensitivity (98.0%), specificity (94.7%), FPR (2%), FNR (5.3%), and ODA of 97.1%.

Of the 17 cancer patients that had mammography done, 11 were highly suspicious of malignancy, two benign while the remaining four received equivocal diagnosis. This gives a concordance of 64.7% and an equivocal rate of 23.5%. Similarly, of the 51 cancer patients with breast ultrasound reports, 28 were highly suspicious of malignancy, seven benign, and the rest (16) equivocal. Overall, ultrasound achieved concordance of 54.9% and equivocal rate of 31.4%.

Table 4: Comparison of diagnostic validities of fine needle aspiration cytology and clinical diagnosis for breast malignancy

Validity test (%)	Sensitivity	Specificity	FPR	FNR	PPV	NPV	ODA
FNAC	97.2	98.9	1.4	2.1	98.6	97.9	98.2
Clinical diagnosis	90.9	86.1	15.8	7.9	84.2	92.1	88.3
NHSBSP[13]	>800	>60.0	4.0	<6.0	>95.0	-	-

FPR: False positive rate, FNR: False negative rate, PPV: Positive predictive value, NPV: Negative predictive value, ODA: Overall diagnostic accuracy, NHSBSP: National Health services Breast Screening programme, FNAC: Fine needle aspiration cytology

Table 5: Sub-classification of breast cancers by fine needle aspiration cytology

FNAC sub-classification		Histopathology	
Cytology report	Frequency	True	False
Subclassified			
Invasive ductal carcinoma	50	47	3
Invasive lobular carcinoma	2	1	1
Medullary carcinoma	2	1	1
Mucinous carcinoma	1	1	0
Fibroadenoma	1	0	1
Mastitis	1	0	1
Total	57	50	7
Not subclassified	31	0	31
Grand total	88	50	38

FNAC: Fine needle aspiration cytology

Discussion

Majority (75.4%) of the patients with breast disease presented with palpable lumps and accepted both FNAC and surgical excision biopsy for the assessment of their breast problems. Although there was preponderance of benign lumps, patients with malignant diagnoses were more heterogeneous in age and method of presentation. Reports from China, Hong Kong, and Ile-Ife, Nigeria, conform with the above findings.[15-17] Traditionally, histopathology is utilized to make diagnosis of breast lesions; but with combined diagnostic approach, FNAC has now become an established definitive diagnostic procedure for breast diseases worldwide.[9,10,16,18-21] Considering the significant apprehension from patients and their families, FNAC and clinical assessment with or without breast imaging can be utilized to fast track diagnosis and clinical decision on the first clinic day or shortly afterward.[7,9,22,23] In concert, FNAC and histopathology showed that male breast cancer represented 2.3% among breast cancer patients and 1% of all breast lumps, respectively. Previous studies support these reports.[3,4]

Overall, FNAC misdiagnosed 2 (2.3%) of the 88 biopsy confirmed breast cancers and labeled them benign and was equivocal (C3 and C4) in 13 cases (14.8%). In addition, there were no specific diagnoses in four cases (4.5%) due to inadequate cytological sample (C1). However, of the 69 (78.4%) cases that were unequivocally malignant (C5) and confirmed microscopically, there were 61 invasive ductal carcinomas, only two of which were low-grade tumors. The import of this finding lies with the fact that the preponderance of high-grade tumors among the invasive ductal carcinomas probably contributed to the high diagnostic yield by FNAC test seen in this study. Studies have shown that the equivocal (C3 and C4) and inadequate (C1) smears constitute the gray areas of FNAC that may be due to carcinoma in-situ, low grade or sclerotic tumors or atypical ductal hyperplasia.[7,9,10,20] Perhaps, this explains why the seventeen unclassified smears in this study variously comprised of low-grade invasive ductal and lobular carcinomas, Burkitt's lymphoma, and the less common histologic variants of breast cancers (mucinous, medullary, papillary, Paget's disease of breast) which are frequently sclerotic, hypocellular or limited by the lack of full cytological features to define either benign or malignant diagnosis. These findings on FNAC mandate

CNB rather than repeating the slides or the use of CNB as an initial diagnostic procedure in these special circumstances if they have been suspected through clinical or mammographic approaches.[7,9] Excision biopsy is recommended if CNB subsequently fails.[7,9]

Other striking observations were two cases of pregnancy coexisting with invasive breast cancers; these were reported as atypical (C3) diagnosis by FNAC. Elsewhere, it has been shown that pregnancy, use of contraceptive pills or hormone replacement therapy are associated with elevated C3 diagnosis.[7,9,20] The clinical implications of these reports warrant selection of CBN or open biopsy rather than FNAC for diagnosis of breast lesions in the setting of pregnancy or hormone manipulation.[9]

FNAC proved itself relevant in subclassifying breast cancers in this present study. An overall concordance rate of 87.7% for 57 histologically confirmed cancer cases suggests that the test is a useful tool for breast cancer diagnosis when done by experienced pathologist. Moreover, there was no attempt to subclassify the seventeen smears that previously fell into the gray areas of FNAC. The reasons adduced for the disharmony and failed subclassification include the fact that breast cancers share a common spectrum of cytological features ranging from lack of cell cohesiveness, hyperchromasia to frank cellular pleomorphism and atypia.[7,9,16,19,24]

A case of malignant phyllodes tumor was subclassified by FNAC as fibroadenoma, validating earlier reports that fibroepithelial breast lesions (fibroadenoma, phyllodes tumor, fibrocystic change) share common cytological and histopathological features and may require pathologic examination of the entire resection specimen to make correct diagnosis.[7,9,16,19] In conclusion, sub-classification of breast cancers using FNAC was good for invasive ductal carcinomas (94%), but not suitable for uncommon tumors such as mucinous, medullary, and lobular carcinomas. It has been observed that in selected cases, FNAC can diagnose, type, prognosticate, and grade breast cancers with a degree of accuracy that will allow the surgeon to make an informed decision on the best line of therapy.[7,9,16,20,25]

The validity results recorded in the current study showed that FNAC is generally superior to clinical evaluation for diagnosis of breast cancers. This is comparable to reports from other studies, but contrasts with findings culled from Lagos, Nigeria, where clinical diagnosis showed a diagnostic accuracy of 95.7% and superseded FNAC (89.9%).[6,8,10,11] The reason may be due to a higher inadequate (C1) rate (17%) reported in Lagos series compared to an impressive 6.1% recorded in this study.

Interestingly, a FPR of 1.4% recorded with FNAC in this study is clinically significant. A false positive diagnosis is a source of concern for any diagnostic test because it can lead to inordinate anxiety, unnecessary overinvestigation and overtreatment for a truly benign pathology. False positive diagnosis in FNAC is frequently attributable to difficulty in interpretation of some peculiar breast lesions.[7,9]

Similarly, the FNR of 2.1% documented by FNAC in this study indicates that about 2.1% of breast cancers could be missed if diagnosis is based on FNAC alone. The consequences of this are false confidence by both patient and clinician and subsequent presentation with advanced malignancy in a patient previously diagnosed with a benign breast disease. The causes of false negative diagnosis in this study include a malignant phyllodes tumor masquerading as fibroadenoma and an invasive papillary carcinoma misdiagnosed as mastitis on FNAC. False negative diagnosis in FNAC is commonly due to sampling error, but at times due to interpretation errors.[9] It ranges from 3% to 24% in various studies.[8,16,20] Remarkably, even open biopsy, the gold standard against which clinicians judge all other diagnostic techniques has been reported to have a FNR of 1.4%, as it is also subject to sampling and interpretation errors.[9,20,22]

Only 17 out of the 88 patients with biopsy confirmed breast cancer did mammography for evaluation of the contralateral breasts and delineation of structural details of ill-defined palpable breast masses. The concordance of 64.7% supports previous reports that mammography is a useful component of the triple assessment approach in patients with breast cancers.[7,9] However, from our findings, it is less reliable than FNAC (concordance of 78.4%); perhaps this supports the reason while FNAC has become established as a diagnostic rather than a screening tool.[9] The occurrence of breast cancer a decade earlier in African women compared to their Caucasian counterparts is clinically relevant considering the fact that mammography has low diagnostic yield in women below 35–40 years.[3,4,18] In our resource limited setting, dedicated ultrasound and mammography machines are scarcely available, thereby limiting the pool of breast cancer patients with imaging reports that can aid early diagnosis.

In the current survey, FNAC was more sensitive for malignant lump >5 cm compared to smaller tumors (98% and 83.3%, respectively). It has been shown that small malignant lumps are more likely to be low grade, sclerotic and hypocellular which subsequently limit the cytological features necessary to diagnose cancer when cancer cells are present.[9] False positive and FNRs were also higher for malignant tumors >5 cm expressing the need for greater caution when FNAC is utilized in suspicious breast masses with extensive sizes. The

reasons for lower performance of FNAC in large tumors include hemorrhagic necrosis, increased vascularity and fibrosis inherent in these tumors.[8,9,12,16]

Conclusion

FNAC in this study proved to be significantly accurate in making a diagnosis of malignancy compared to clinical examination and imaging studies in the assessment of patients with breast cancers. However, the unacceptably high false positive and negative diagnoses recorded with clinical examination reduced reliance on it. A histological confirmation remains the gold standard when clinical and FNAC are not in harmony. A combined diagnostic approach with FNAC playing a dominant role would provide a suitable option.

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Conflicts of interest

There are no conflicts of interest.

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Appendix

Appendix I

Information Sheet for Patients Undergoing Fine Needle Aspiration Cytology Produre

1. Fine needle aspiration cytology (FNAC) is used to investigate a breast lump. The triple test approach is the recommended way of investigating a patient with breast lump
2. The triple test includes three main steps;
   a. Clinical breast examination after medical history
   b. Imaging tests; mammography and/or ultrasound and
   c. FNAC.
3. FNAC involves placing a thin needle into the breast to obtain a small sample of cells from the lump. An experienced surgeon, breast physician, pathologist or radiologist takes the sample
4. The decision for treatment of the breast disease will be based on histopathology result not FNAC because its reliability has not been established in our center. The needle is typically inserted several times. The test itself takes only one to two minutes
5. The sample obtained is then sent to a pathologist, who will study the cells and provide a detailed report on the type of cells present. In some situations, the sample can be immediately checked to see if enough cells have been obtained. If the cells are deemed inadequate, it can be repeated at the same sitting
6. The final result will be available within a few hours to few days
7. The test may be uncomfortable, but it is rarely painful. It will sometimes cause bruising, but infection is rare. Firm pressure be used to ease any discomfort and reduce swelling
8. Definition of terms:
   Cyto = Cell
   Cytology = Study of cells
   Mammography = X-ray of the breast
   Ultrasound = Special imaging study.