Cytopathologic Review of Breast Lesions and Correlation with Histopathologic Results at Tikur Anbessa Specialized Hospital; A Six Years and Eight Months Retrospective Study from 2015 - 2021.

Thesis submitted to the Department of Pathology, Tikur Anbessa Specialized Hospital, School of Medicine, College of Health Sciences, Addis Ababa University for partial fulfillment of the requirements for post graduate diploma in pathology

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Abstract

Background: A palpable breast lump is always a cause of concern to the patient. Hence, accurate pathological diagnosis is crucial for further management and to evaluate the outcome. Clinical examination, imaging and FNAB are implemented in the assessment of breast lesions in the Triple Test. In this regard the role of FNAB is vital in the diagnoses of breast lesions particularly in identifying malignant lesions.

Objectives: The aim of this study was to determine cytopathologic patterns of breast lesions in TASH and to correlate Fine Needle Aspiration Biopsy results with the corresponding histopathologic results as to determine the validity of FNAB.

Methods: A retrospective descriptive cross-sectional study of 1327 patients who underwent FNAB of whom 145 had histopathology results over six years and eight months between January 2015 and August 2021 in TASH. Descriptive statistics of the cytology reports as well as diagnostic performance analysis of malignant and benign FNAB categories made using SPSS version 23.

Results: A total number of 1327 patients were included out of which 1214 (91.5%) were females and 113 (8.5%) were males. According to the IAC Yokohama system the malignant and benign categories of FNAB account 44% each of the total cases and the rest contributed by the suspicious, atypical and insufficient categories. Most of the Malignancies present in the 4^{th} and 5^{th} decades of life whereas benign cases happened a decade earlier. 145 of the cases were followed by histopathology. FNAB of benign and malignant lesions showed 96.0% accuracy, 96.3% sensitivity and 97.7% specificity.

Conclusion: The burden of malignancy in this setup (TASH) is high yet, FNAB remains to be a reliable and acceptable method for the diagnosis of breast lesions.

Acronyms and Abbreviations

FNAB - Fine Needle Aspiration Biopsy

FNA- Fine Needle Aspiration

GLOBOCAN - Global Cancer Observatory

CNB - Core Needle Biopsy

IHC - Immunohistochemistry

CBHI - Community Based Health Insurance

SHI - Social Health Insurance

IAC - International Academy of Cytology

SPSS - Statistical Package for the Social Sciences

UK - United Kingdom

NHSBSP - National Health Service Breast Screening Program

TASH -Tikur Anbessa Specialized Hospital

PPV - Positive Predictive Value

NPV- Negative Predictive Value

AAUMF – Addis Ababa University Medical Faculty

IRB – Institutional Review Board

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1. Introduction

1.1. Background

The breast or mammary gland is covered by skin and subcutaneous tissue and rests on the pectoralis muscle, from which it is separated by a fascial layer. The morphofunctional unit of the breast is a complex branching structure made up of the terminal duct–lobular unit (TDLU) and the large duct system. The TDLU is made up of acini and the terminal ductile and represents the secretory portion of the gland. It connects with hierarchical segmental ducts to reach the collecting (lactiferous) duct, which empties into the nipple [1]. Mammary stroma is composed of intralobular and interlobular stroma constituted by fibrous connective and adipose tissue. The most common breast problems for which women seek medical advice are breast pain, nipple discharge and a palpable mass. Most women with these complaints have benign breast disease [2]. The majority of breast masses are benign conditions like fibroadenoma as evidenced by multiple studies [3] [4] [5]. Nevertheless, it's imperative to thoroughly assess breast masses for cancer. According to GLOBOCAN 2020, breast cancer is the most common cancer in women, with nearly 2.2 million and 16133 new cases worldwide and in Ethiopia, respectively [6] .

Clinical examination combined with different diagnostic modalities including mammography, FNAB, CNB and IHC are employed in the assessment of breast lumps. In resource poor settings like Ethiopia, FNAB comes readily useful for its obvious advantages. It is a cheap, fast, and reliable diagnostic method [7].

Early diagnoses of breast lesions help to guide individualized management plan and to have better long term survival of patients and alleviate the disease burden to the health care system.

This retrospective study will be undertaken to determine Cytopathologic patterns of breast lesions and to correlate with the corresponding histopathologic results (Gold standard) at Tikur Anbessa Specialized Hospital, Department of Pathology, Faculty of Medicine, Addis Ababa University between January 01, 2015 and August 30, 2021.

1.2. Statement of the problem

With more than 112 million people (2019), Ethiopia is the second most populous nation in Africa after Nigeria. It is one of the poorest, with low health care service accessibility[8]. In recent years, Ethiopia has been successful in reducing deaths related to communicable, maternal, neonatal, and nutritional deficiency diseases and injuries by 65%. However, the country's performance regarding non-communicable diseases and cancer is minimal, causing these diseases to join the commonest causes of premature mortality and death rates [9]. Among which breast cancer is the most common cancer in women worldwide, with nearly 2.2 million new cases according to GLOBOCAN 2020. This represents about 24.5% of all new cancer cases making it the leading causes of cancer among women. The Ethiopian estimation shows 16133 new breast cancer cases accounting for 31.9% of the total cases which makes it the leading cancer among Ethiopian women. According to the Addis Ababa cancer registry data from 2012-2013, breast cancer constitute 33% of cancers in women and 23% of all cancers in Ethiopia. As a result, every woman presenting with a breast mass should be evaluated to exclude or establish a diagnosis of cancer [10].

A triple test consisting of clinical examination, imaging and FNAB is considered the standard in the assessment of breast lumps. Kun of Strasbourg reported the earliest usage of fine needle aspiration in 1847 and it was in the 1930s that FNA of breast lesions became widely accepted as a first-line diagnostic procedure [11]. In Ethiopia, the practice of cytopathology and fine-needle aspiration began sometime after the establishment of the department of pathology in Addis Ababa University, Faculty of Medicine, Tikur Anbessa Hospital, in 1965. Additional centers are now staffed with pathologists who practice cytopathology [12]. In medically under- resourced developing countries, which represent more than 80% of the world's population, breast is one of the most common FNA sites and FNA is the most appropriate first line test for all palpable breast lesions when core needle biopsy (CNB) and histopathology are not readily available and expensive options [13]. FNAB is a cheap, fast, and reliable diagnostic method with minimal complications, such as bleeding or infection. It also reduces the frequency of open biopsies [12].

Of course in developed (resourceful) countries, CNB is preferable over FNAB since it offers greater specificity in the diagnosis of lesions associated with microcalcifications and the diagnosis of certain proliferative lesions and low-grade DCIS and for confirming invasive carcinoma [13] [14]. Yet, CNB has limitations similar to FNAB like sampling error, in distinguishing papillomas from intraductal papillary cancer, cellular fibroadenomas from low-grade phyllodes tumors, and atypical ductal proliferations from low-grade DCIS. CNB is a more invasive procedure with a higher rate of complications, a less rapid turnaround time to diagnosis, greater expense for equipments and the requirement for a surgical pathology laboratory to process and interpret the samples. As a result, CNB is particularly inappropriate in a low resource setting for the diagnosis of the most common lesions [13].

The reporting and diagnoses of breast FNAB shows differences among institutions and individuals. This lack of uniform reporting may confuse clinicians and can result in inconsistent patient management [11]. In an effort to establish standard terminology and a uniform approach for the reporting of breast cytology, the IAC Yokohama System for Reporting Breast Fine Needle Aspiration Biopsy was devised that uses five clearly defined categories described by specific terms, and each has its own risk of malignancy[13]. The five categories are insufficient/inadequate, benign, atypical, suspicious of malignancy, and malignant. Each category and its risk of malignancy are linked to management recommendations, which include several options [13]. Like other tests, FNAB has its own limitations and diagnostic problems. The limitations include false negative, false positive and suspicious results. Sample adequacy, aspiration techniques and skill of performing physician, the experience of the pathologist interpreting the smear and the overlapping cytological features between some benign and malignant neoplasms are some of the factors affecting the outcome of FNAB. The accuracy of FNAB is optimized when the FNAB is performed by cytopathologists or experienced radiologists or clinicians in multidisciplinary clinics, utilizing ultrasound guidance as required with rapid on-site evaluation (ROSE) of Giemsa- stained slides to triage cases [13] [16]. The FNAB provisional results can be correlated immediately with the clinical and imaging findings in the ‘triple test’. In these settings, the sensitivity and specificity rates for FNAB and CNB are comparable [13] [16].

1.3. Significance of the study

Breast masses are common clinical presentation of patients with significant portion contributed by malignancy at Tikur Anbessa Specialized Hospital. Fine needle aspiration biopsy done commonly, but only few studies done years back are out there showing cytopathologic patterns of breast lesions and compare with histopathology reports in TASH and Ethiopia. The purpose of this study is to review the cytopathologic patterns of breast lesions and correlate cytology reports with the corresponding histopathologic reports as to determine the utility of FNAB in the diagnosis of breast lesions in our setup. As a result, this study hopefully will add something to the existing knowledge (practice) in this area and can be used as a reference for other studies in the future.

2. Literature review

The technique and diagnostic interpretation of fine needle aspiration biopsy (FNAB) of the breast has developed over the past 60 years into an extremely useful, accurate, highly specific and sensitive, and cost-effective test for the diagnosis of benign and malignant breast lesions. On literature search of various studies on spectrum of breast lesions; most of the cytologic diagnoses were classified into 5 categories according to the National Cancer Institute Consensus Conference on Breast FNA (1997): unsatisfactory, benign, atypical probably benign, suspicious probably malignant, and malignant [17]. But since 2016 based on a review of the literature and the expertise opinions of the IAC breast group a standardized reporting system was introduced as discussed above. There is a very low false-positive rate usually related to FNAB of fibroadenomas, papillomas and papillary lesions, and a low false-negative rate usually related to low-grade ductal and lobular carcinomas[15]. Published data regarding breast cancer detection for breast FNA indicate in the ranges: sensitivity, 66.7 to 99.4%; specificity, 60 to 100%, NPV, 66.7 to 100%; diagnostic accuracy, 81 to 97.2%. As a part of the 'triple test', breast FNAB misses less than 1% of breast cancers. With concordance of pathologic, radiologic, and clinical impressions, patients can proceed to definitive surgery. If any two of the three 'triple test' parameters are discordant, patients should undergo excisional biopsy for further histopathologic evaluation. Although CNB is reported to have sensitivities ranging from 84.7 to 100% and specificities ranging from 85 to 100% ; breast ultrasound paired with FNAB, as an alternative to breast mammography with CNB, can provide patients with substantial cost savings without compromising diagnostic accuracy in resource-limited healthcare settings[18][19][20][21][22].

According to a study done between January 1991 to December 1994 to review the cytologic features of breast lesions and to elucidate the clinical use of fine needle aspiration in Tikur Anbessa Hospital Department of Pathology; 1,211 fine needle aspirations of breast lesions revealed 255 (21%) malignant, 901 (74%) benign, 15 (1.2%) suspicious and 40 (3.3%) unsatisfactory cases. The mean age of patients with malignant lesions was 43 years (range: 24 to 80 years), while it was 27 years (range: 15 to 50 years) for those with benign lesion. There were 1132 (93.5%) females and 79 (6.5%) males [23].

In another prospective study of 138 cases done during June 2013 - June 2014 at Tikur Anbessa Hospital pathology laboratory; FNA diagnoses and biopsy of breast masses of patients who underwent subsequent histopathologic evaluation were evaluated. The sensitivity and specificity of FNAB was found to be 86.7% and 90.3% respectively. The positive and negative predictive values are 86.7% and 90.3% respectively. Age of patient varied from 11-65yrs. Most of the benign cases were in patients below 30yrs of age and malignant cases were in 40-50 yrs age group. 138 cases were categorized as inadequate 11(84.6%), benign 56(90.3%), suspicious 5(27.7%) and malignant 6(13.3%). Fibroadenoma 31(29%) was the commonest benign lesion followed by phyllodes tumor 14(13.3%) and papillary neoplasm 7(6.5%). Amongst malignancies, commonest lesion was ductal carcinoma, 43(40.2%) followed by lobular carcinoma, 2(1.87%). Majority of the cases reported as suspicious(72.3%) turned out to be malignant on histological examination [24].

In a study done in southwest Ethiopia of Jima University Specialized Hospital; 735 patients were included to determine the cytomorphologic patterns of palpable breast lesions and the overall distribution of the lesion was benign neoplastic (51%) , non-neoplastic (28%), malignancy (17%) and suspicious (4%) . The most common lesions in male were non-neoplastic and in female were benign neoplastic groups. The cytological diagnoses included malignancy 123 (16.7%), suspicious 28 (3.8%), benign neoplastic 378(51.4%), and non-neoplastic 206 (28%) cases. Among benign neoplastic lesions, the two maximum cases were fibroadenoma 152 (40.2%) and fibrocystic changes 117 (31.0%) [25].

A comparative study of the fine needle aspiration biopsy and excisional biopsy of breast lesions done at Ethiopian Health Nutrition Research Institute, Addis Ababa showed that fine-needle aspiration biopsy as a routine method in the management of breast diseases was compared to excisional biopsy of the same patients. Sixteen patients (15.7%) had carcinoma of the breast and 86 (84.3%) cases were found to have fibroadenomas of the breast. The sensitivity was 94.3% and the specificity was 78.6%. The positive and negative predictive values were 68.8% and 96.5%, respectively. There were three false positive and false negative cases[26].

In rural Kenya of African Inland Church (AIC) Kijabe Hospital, electronic pathology files were retrospectively reviewed from 1/1999-9/2017. A total of 695 breast FNAB cases were identified

in the 18 year 9 month interval. Five-hundred forty one of these (78%) were performed during the more recent 7 year 9 month period from January 2011 through September 2017. Two-hundred nineteen (219/695, 31.5%) cases from 196 patients had subsequent surgical pathology. The average patient age was 39 years (range, 13 to 88 years; median, 37 years). Ninety-five percent of the patients (186/196) were female. Retrospective FNAB interpretive categorization according to the Yokohama system for the 219 breast FNAB cases with subsequent surgical pathology was as follows: 20 (9%) insufficient, 103 (47%) benign, 16 (7%) atypical favor benign, 24 (11%) suspicious favor malignant and 56 (26%) malignant. On histology, there were 141 (64%) benign cases and 78 (36%) malignancies. The probability of malignancy for each of the five categories was calculated using histopathologic interpretations as the diagnostic gold standard. Probabilities of malignancy are as follows: insufficient, 0%; benign, 12%; atypical favor benign, 25%; suspicious favor malignant, 46%; malignant, 91%. With interpretations of atypical and above on cytology (i.e., atypical favor benign, suspicious favor malignant, and malignant) considered a non-benign interpretation due to the triggering of subsequent clinical work-up, the sensitivity of FNAB for detecting malignancy was 85% and the specificity was 75%. Positive and negative predictive values were 69% and 88%, respectively. Overall diagnostic concordance between FNAB and surgical pathology was 79%. The false positive rate was 2.3% (5/219 cases) and the false negative rate was 5.4% (12/219 cases) [18].

In a Correlation study between Fine-Needle Aspiration Biopsy and Histology for Palpable Breast Masses in a Nigerian Tertiary Health Institution from January 2007 to December 2011; FNAB was requested for a total of 1790 breast lumps. Of these, 1301 were benign, 250 were malignant, 181 were equivocal, and 58 were inadequate. Four hundred and thirty-six (436) of the 1790 cases subsequently had biopsies, a biopsy rate of 24.4%. Absolute sensitivity 95.4%, complete sensitivity 99.2%, full specificity 88.9%, positive predictive value 99.6%, false-negative rate 0.8% (<4%), false-positive rate 0.4%, inadequate rate 3.2%, and suspicious rate 10.2%. Over 90% of cases from FNAB (96.3%) were confirmed to be malignant by histology, and 99.5% were confirmed to be benign. 82.8% suspicious but possibly benign on FNAB were found to be benign on histology, and 60%of cases suspected to be malignant were found to be malignant [7].

Four hundred and twenty five (425) patients were evaluated at the Department of Pathology King Edward Medical University, Lahore in four years for FNAB of their palpable breast masses from

June 2006 to June 2010. FNAB diagnosis was compared with histological diagnosis to see the accuracy of fine needle aspiration biopsy for neoplastic lesions. There were 271/425 benign, 120/425 malignant, and 32/425 suspicious smears. Inadequate samples were repeated twice or thrice, and the degree of success was improved with consecutive repeating approaches. FCC, fibroadenoma and inflammatory breast lesions in decreasing order dominate benign conditions with maximum incidence in the 3rd, 4th and 5th decades of life. Among malignancies invasive ductal carcinoma was the most commonly reported lesion with maximum incidence in the 4th, 5th, and 6th decades followed by invasive lobular carcinoma and other malignant lesions. The sensitivity, specificity, accuracy, negative predictive value, and the positive predictive value of FNAB was 98%, 100%, 98%, 100%, and 97%, respectively [27].

At the Maharaj Nakorn Chiang Mai Hospital, Chiang Mai, Thailand, 2,375 cases of breast lesions were sampled by fine-needle aspiration (FNA) from 1994 –1999. Cytologic diagnoses were: benign (48%), suspicious for malignancy (5%), malignant (15%), and unsatisfactory (32%). Comparison with histology was possible in 721 cases. The diagnoses obtained by FNA showed a sensitivity of 84.4%, specificity of 99.5%, positive predictive value of 99.8%, negative predictive value of 84.3%, and overall diagnostic accuracy of 91.3%. In another study in northern Thialand during January 2003 -December 2006 a series of 182 breast masses revealed: benign lesions in 98 (53.9%); suspicious for malignancy in 31(17.0%); and malignant in 53 (29.1%). From the subsequent histopathologic diagnoses, 6/98 cases of benign cytology turned out to be malignant lesions; 22/31 cases of suspicious cytology were truly malignant while the other nine were benign; and only 1/53 with malignant cytology was benign (false positive), the lesion being a fibroadenoma. The overall accuracy, sensitivity, specificity, positive predictive value, and negative predictive value were 91.2% (95% confidence interval [CI], 87.6%-94.8%), 92.5% (95% CI, 88.7%-96.3%), 90.2% (95% CI, 85.9%-94.5%), 88.1% (95% CI, 83.4%-92.8%) and 93.9% (95% CI, 90.4%-97.4%), respectively [22].

A study conducted in India at Smt. N.H.L. Municipal Medical College, Ahmedabad, on 222 patients with breast lesions; among which cyto-histopathological correlations were obtained in 91 cases. Among 222 patients, 217 were females and 5 were males. Benign breast lesions were found in 144 cases (64.87%); among which fibroadenoma (30.18%) was the commonest lesion. Malignancy was observed in 69 cases (31.08%); among them, ductal carcinoma was the

predominant lesion (29.28%). All 45 malignant aspirates were confirmed by histopathology. Benign reports were confirmed in 45 out of 46 cases by doing histological examinations; except one case of malignant phyllodes. Sensitivity and specificity of FNAB in breast lesions were reported to be 97.82% and 100% respectively, with 100% positive predictive value, 97.85% negative predictive value and 98.90% diagnostic accuracy. This study documented the fact that benign breast lesions were the most common lesions in young females and the malignant lesions were common in fourth and fifth decades of life [4].

In Peoples Republic of China, Jinan Military General Hospital a study was conducted from January 1995 to December 2005 to verify the role of FNAB in preoperative diagnosis of breast carcinoma; 1238 aspirates from patients with a breast lump were involved. In total, 1071 breast carcinomas were diagnosed with postoperative histological diagnosis. One thousand and forty-six of these 1071 breast carcinomas were definitely diagnosed by FNAB. Only one breast carcinoma identified by FNAB was not finally verified by histological diagnosis postoperatively. The diagnostic sensitivity, diagnostic specificity, overall accuracy, and the pseudo-negative and pseudo-positive results of FNAB for diagnosing breast carcinoma are 97.72%, 99.4%,97.94%, 2.28%, and 0.6%, respectively [28].

3. Objective of the study

3.1. General objective

- To determine cytopathologic patterns of breast lesions and to correlate with their corresponding histopathologic results.

3.2. Specific objective

- To determine cytopathologic patterns of breast lesions.
- To see the relationship of different breast lesions with age and sex of patients
- To correlate FNAB reports with the corresponding histopathologic results.
- To compare cytologic nuclear grade of with the histopathologic grade.
- To compare and discuss the result with different studies.

4. Methods and materials

4.1. Study area

The study was conducted at Tikur Anbessa Specialized Hospital, pathology department which is found in Addis Ababa, Ethiopia. It is the one of the largest tertiary teaching hospital in the country with the vast majority of cases investigated at this department are patients referred from all over the country. The department gives cytopathology, surgical pathology, hematology and neonatal autopsy services. All of the pathologist staff and residents rotate to work through the FNAB service.

4.2. Study Design

The study applied a retrospective cross-sectional descriptive method to review all the Fine needle aspiration biopsy of breast lesions and their corresponding mastectomy/lumpectomy histopathology reports performed at Tikur Anbessa Specialized Hospital. Data was retrieved from the archive of department of pathology that was seen from January 01, 2015 to August 30, 2021.

4.3. Study population

All patients who had Fine needle aspiration biopsy performed for breast lesions at Tikur Anbessa specialized teaching Hospital during the study period.

4.4. Operational definitions

- FNAB results were classified in to five diagnostic categories for descriptive statistics.
- Histopathologic results classified into benign and malignant categories.
- Calculation of FNAB diagnostic performance tests was applied on benign and malignant cytological categories only whereas indeterminate suspicious, atypical and insufficient categories were not included.
- The association of FNAB with histopathology reports was also assessed using chi-square test with level of significance set to be less than 0.05 ($p<0.05$).
- Cytology-Histology grade agreement determined using kappa Chone's statistic.

Diagnostic approach according to the IAC Yokohama system was defined as follows

Insufficient or inadequate: descriptive and inconclusive reports. **Benign:** Fibroadenoma, Fibrocystic disease, Proliferative breast lesion without atypia, Intraductal papilloma, Mastitis/Abscess, Granulomatous mastitis, Tuberculous Mastitis, Fat necrosis, Duct ectasia, Lactational changes, Gynecomastia, Benign cyst, lipoma. **Atypical:** ADH, Low/intermediate grade phyllodes, Proliferative breast lesion with atypia

Suspicious for malignancy: low grade DCIS

Malignant: Ductal carcinoma, Lobular carcinoma, Invasive breast carcinoma, NST, High grade DCIS, Invasive lobular carcinoma, Mucinous carcinoma, Tubular carcinoma, Papillary carcinoma, Invasive micropapillary carcinoma, Metaplastic carcinoma, Carcinoma with apocrine differentiation, Salivary type tumors, Phyllodes tumor (malignant), Stromal sarcoma, lymphoma, Secondary of malignancy

Cytologic nuclear grade: Annexed

Histologic grade: Annexed

4.5. Study variables

In the study, the variables included for analysis and interpretation are depicted as the following.

- Independent variables
    - Age of the patient
    - Sex of the patient
- Dependent variables
    - FNAB diagnostic category
    - FNAB specific diagnosis
    - Histopathologic diagnosis
        - Cytologic nuclear grade
        - Histologic grade

4.6. Inclusion and exclusion criteria

4.6.1. Inclusion criteria

- All patients with breast mass who underwent Fine needle aspiration biopsy (including ultrasound guided).

4.6.2. Exclusion criteria

- Those patients with recurrence of tumor
- Unclear cytopathologic and/or histopathologic diagnosis

4.7. Data management and analysis

4.7.1. Sample size estimation

All patients fulfilling the criteria during the study period were included.

4.7.2. Sampling procedure

All the hard copy as well the electronic digital data recording of breast Fine needle aspiration biopsy and their corresponding histopathology reports from January 01, 2015 to August 30, 2021 were reviewed from the archive of pathology department.

4.7.3. Data collection tools and procedures

Demographic data, fine needle aspiration biopsy categorical and specific diagnosis including nuclear grade, corresponding histopathology diagnoses including histologic grade was extracted from the hard copy or electronic data recording using data extraction sheet.

4.7.4. Data analysis

The data coding, entry and analysis was done using the Statistical Package for the Social Sciences (SPSS-23).

Ethical consideration

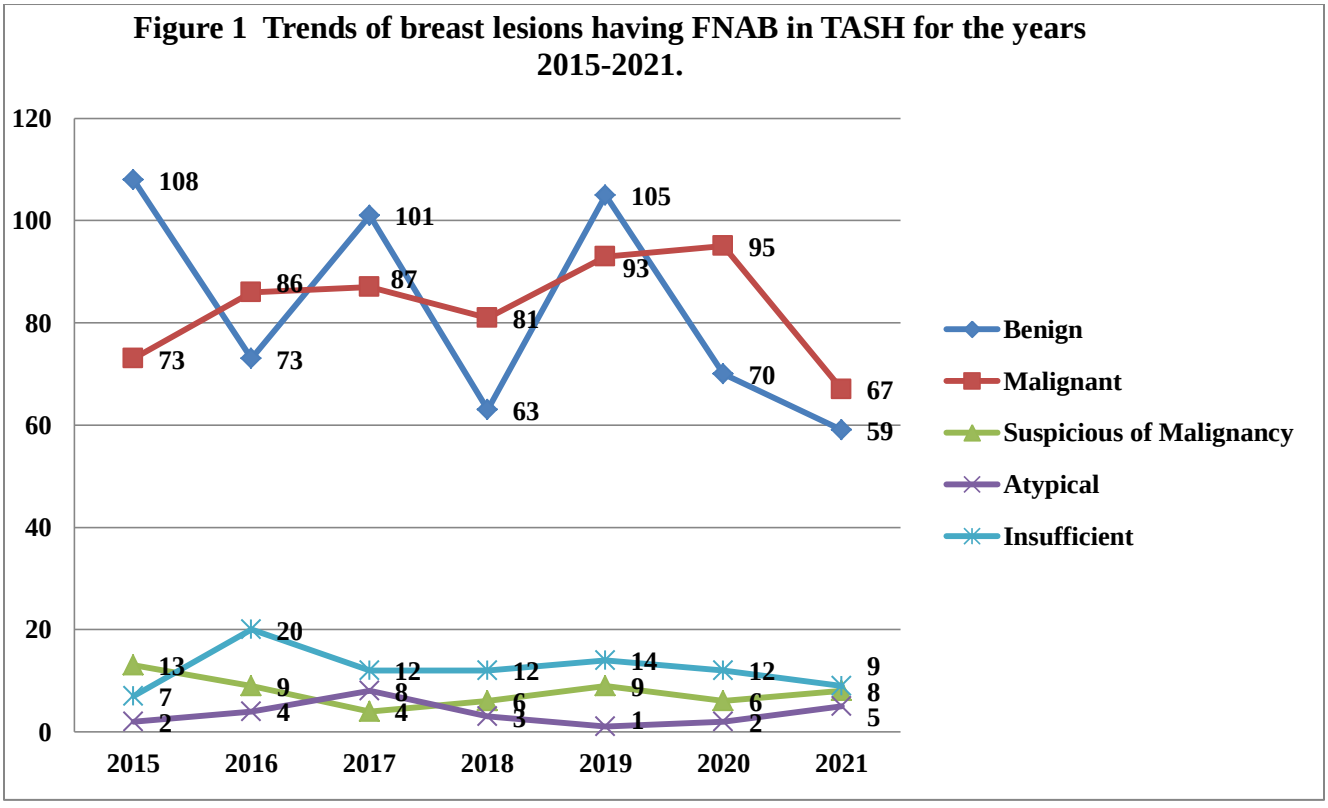
Ethical approval for this study was obtained from the Department of Pathology, College of Health Sciences, Addis Ababa University, IRB committee at Tikur Anbessa Specialized Hospital.

5. Results

5.1. General

A total of 1327 FNAB breast cases fulfilling the study criteria were identified between Jan. 2015 – Aug 2021; 203, 192, 212,165, 222, 185 and 148 cases seen in the years 2015, 2016, 2017, 2018, 2019, 2020 and 2021, respectively. As shown in figure 1 the benign and malignant cases are alternately the most frequent reports with minimum of 59 in the year 2021 and maximum of 108 in 2015 whereas the atypical, suspicious and insufficient reports accounted lower than 20 cases each year throughout the study period.

Figure 1 Trends of breast lesions having FNAB in TASH for the years 2015-2021.



Of the total 1327 cases, 1214 (91.5%) were females and 113 (8.5%) were males with age range of 6 months to 90 years. The mean age was 37.0±13.2 years for females and 48.2±18.9 years for males. 41 (3.1%) of the cases have no age documentation.

Table 1 Sex distribution with mean and std. deviation of breast lesions

Sex distribution		
sex	**Frequency**	**Percent**
F	1214	91.5
M	113	8.5
Total	**1327**	**100.0**

Mean and standard deviation					
	Mean age	**Std. Deviation**	**Minimum age**	**Maximum age**	**Frequency (percent)**
F	37.0	13.2	1.5	90.0	1181(89%)
M	48.2	18.9	0.6	88.0	105(7.9%)
Total	37.5	14.1	0.6	90.0	1286(96.9%)
Missing	-	-	-	-	41(3.1%)
Total	-	-	-	-	**1327(100%)**

5.2 Patterns of FNAB diagnosis

The frequency of cases according to the IAC Yokohama diagnostic categories were as follows; 582 (43.9%) were malignant, 55 (4.1%) were suspicious of malignancy, 25(1.9%) were atypical, 579 (43.6%) were benign and 86 (6.5%) were insufficient/inconclusive (Table 2). In each category specific diagnosis or differential diagnosis were made in 97.2% of malignant, 27.8% of suspicious of malignancy, 24% of atypical and 88.1% of benign reports. The peak age incidence was in the 3rd decade with a mean of 32.4±13.3 years for the benign and 4th decade with a mean age of 43.6±13.2 years for the malignant category.

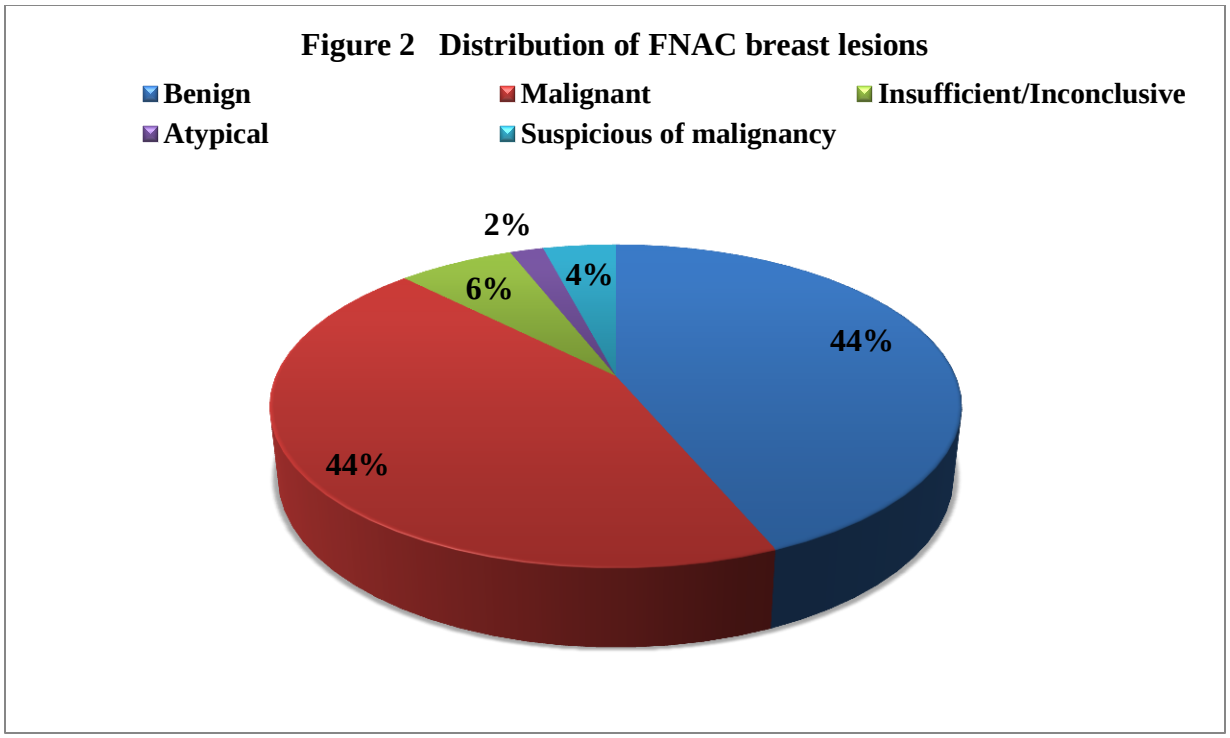


Table 2 Distribution of breast lesions by category and their mean age			
Category	**Frequency**	**Percent**	**Mean age ± SD**
Malignant	582	43.9	43.6±13.2
Suspicious of malignancy	55	4.1	37.4±9.9
Atypical	25	1.9	37.6±12.3
Benign	579	43.6	32.4±13.3
Insufficient	86	6.5	36.9±12.7
Total	**1327**	**100**	**37.9±14.1**

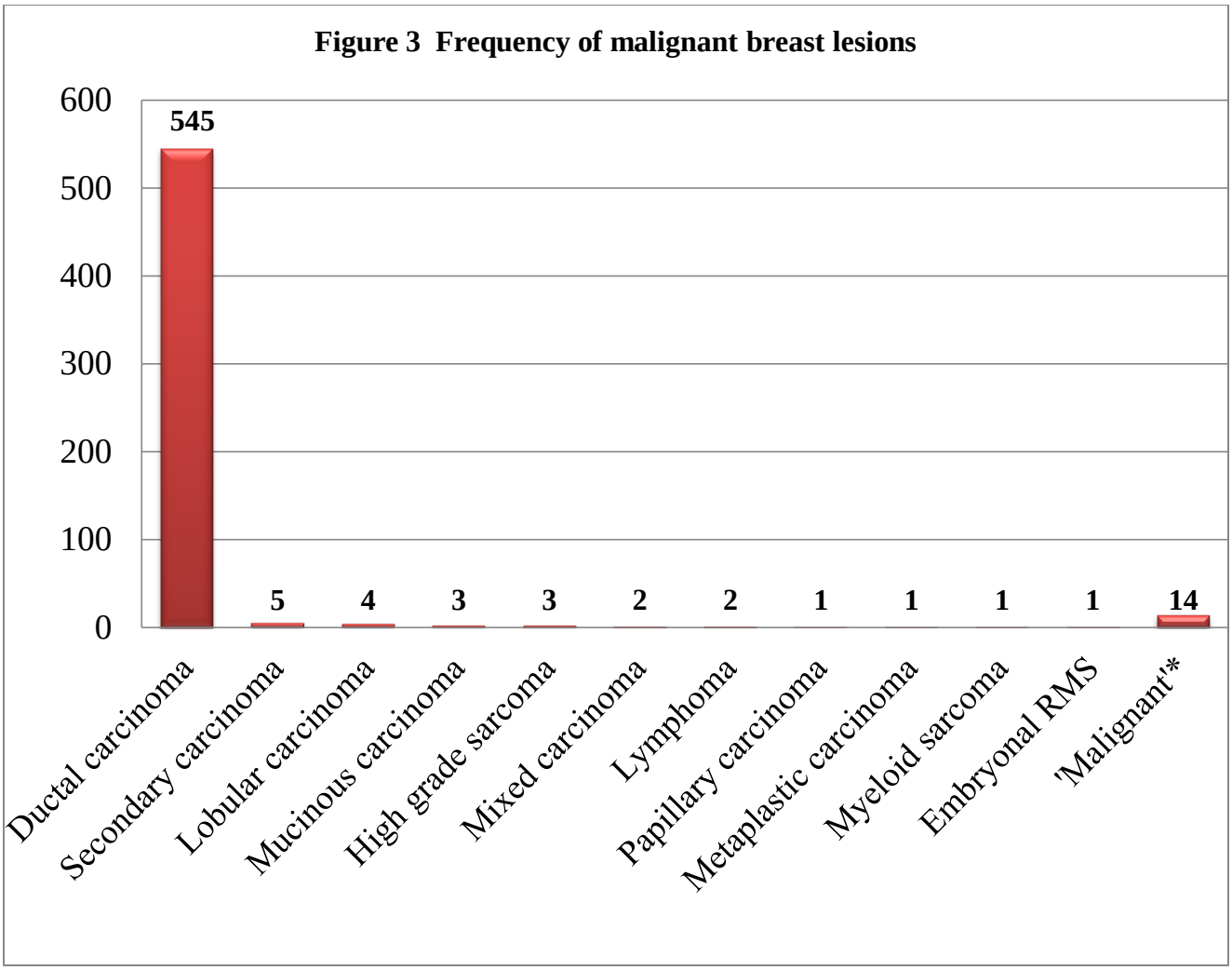
Among the malignant cases, ductal carcinoma (93.6%) accounted the most followed by secondary carcinoma (0.9%) and lobular carcinoma (0.7%) as sown in table 3. Of the benign cases to which specific diagnosis was mentioned, 255(44%) were non-neoplastic and 243 (42%) were neoplastic lesions. Overall fibroadenoma (41.1%) was the most common benign breast lesion followed by FCC (11.4%) and gynecomastia (10.4%). Pyogenic Abscess/Mastitis (7.1%) is the most common among inflammatory lesions (Table 4). Gynecomastia 60 (51.3%) followed by ductal carcinoma 34 (29%) were the most frequent findings among male patients.

Table 3 Distribution of malignant breast lesions

Malignant Category	Frequency			Percent	Valid Percent
	Female	Male	Total		
Ductal carcinoma	511	34	545	41.1	93.6
Lobular carcinoma	4	0	4	.3	.7
Secondary carcinoma	4	1	5	.4	.9
Mucinous carcinoma	3	0	3	.2	.5
High grade sarcoma	3	0	3	.2	.5
Mixed carcinoma	1	1	2	.2	.3
Lymphoma	2	0	2	.2	.3
Papillary carcinoma	1	0	1	.1	.2
Metaplastic carcinoma	1	0	1	.1	.2
Myeloid sarcoma	1	0	1	.1	.2
Embryonal RMS	1	0	1	.1	.2
'Malignant'*	12	2	14	1.1	2.4
Total	**545**	**38**	**582**	**43.9**	**100.0**

** Results reported as malignant without indicating a specific diagnosis.*

Figure 3 Frequency of malignant breast lesions

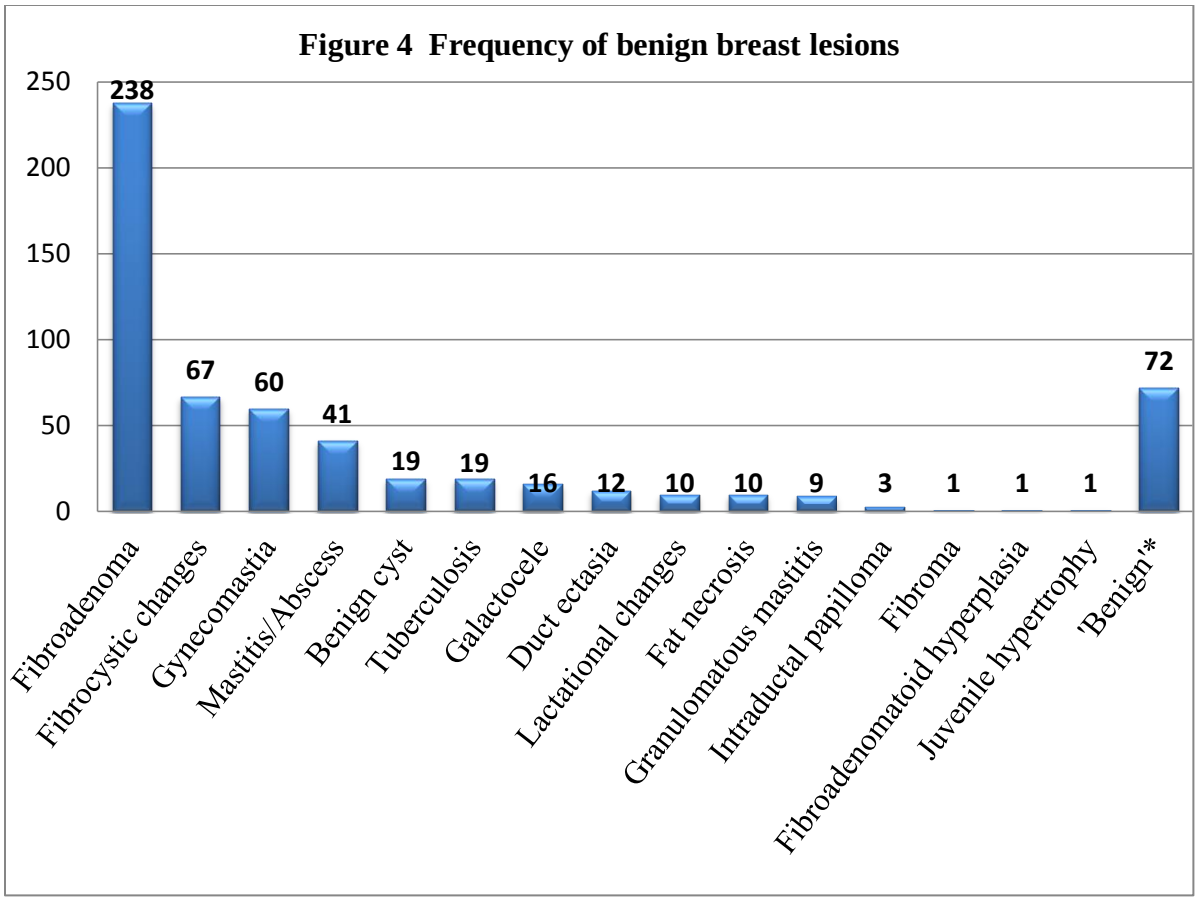


** Results reported as malignant without indicating a specific diagnosis.*

Among 55 cases in the suspicious category one case was reported as DCIS (1.8%), 15(27.3%) were reported as suspicious of ductal carcinoma and the rest simply suspicious of carcinoma (30.9%) and suspicious of malignancy (40%).

Table 4 Distribution of benign breast lesions						
Benign Category		Frequency			Percent	Valid percent
		Female	Male	Total		
Neoplastic	Fibroadenoma	238	0	238	17.9	41.1
	Intraductal papilloma	3	0	3	.2	.5
	Fibroadenomatoid hyperplasia	1	0	1	.1	.2
	Fibroma	1	0	1	.1	.2
Non-neoplastic	Fibrocystic changes	66	1	67	5.0	11.6
	Gynecomastia	0	60	60	4.5	10.4
	Mastitis/Abscess	38	3	41	3.1	7.1
	Tuberculosis	18	1	19	1.4	3.3
	Benign cyst	18	1	19	1.4	3.3
	Galactocele	16	0	16	1.2	2.8
	Duct ectasia	12	0	12	.9	2.1
	Fat necrosis	9	1	10	.8	1.7
	Lactational changes	10	0	10	.8	1.7
	Granulomatous mastitis	9	0	9	.7	1.5
	Juvenile hypertrophy	1	0	1	.1	.2
Miscellaneous	'Benign'*	69	3	72	5.4	12.5
	Total	**509**	**70**	**579**	**43.6**	**100.0**

** Results reported as benign without indicating a specific diagnosis.*



**Results reported as benign without indicating a specific diagnosis.*

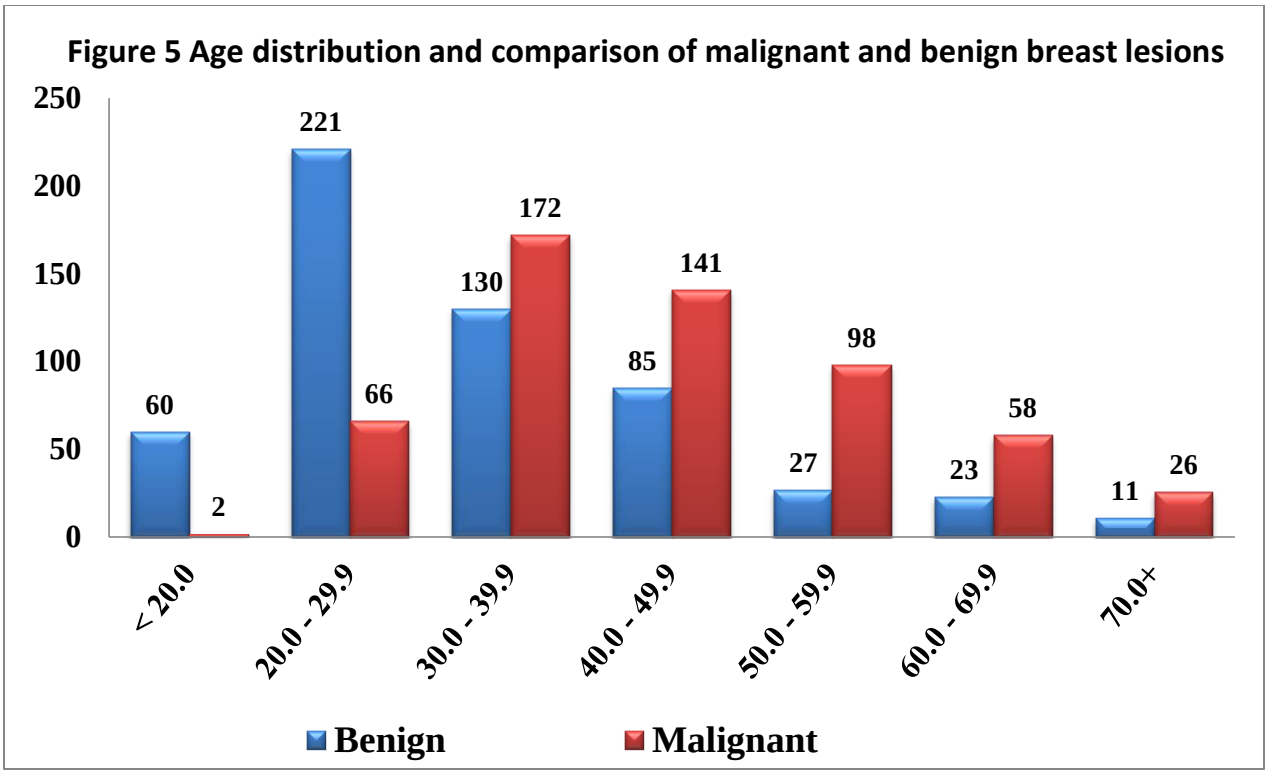
Of 25 cases in the atypical category 20 (80%) were reported as benign proliferative breast lesions with atypia followed by 4(16%) cases of low/intermediate grade phyllodes tumor and a case of intraductal papillary neoplasm (4%).

Table 5 Distribution of breast lesions in the atypical category

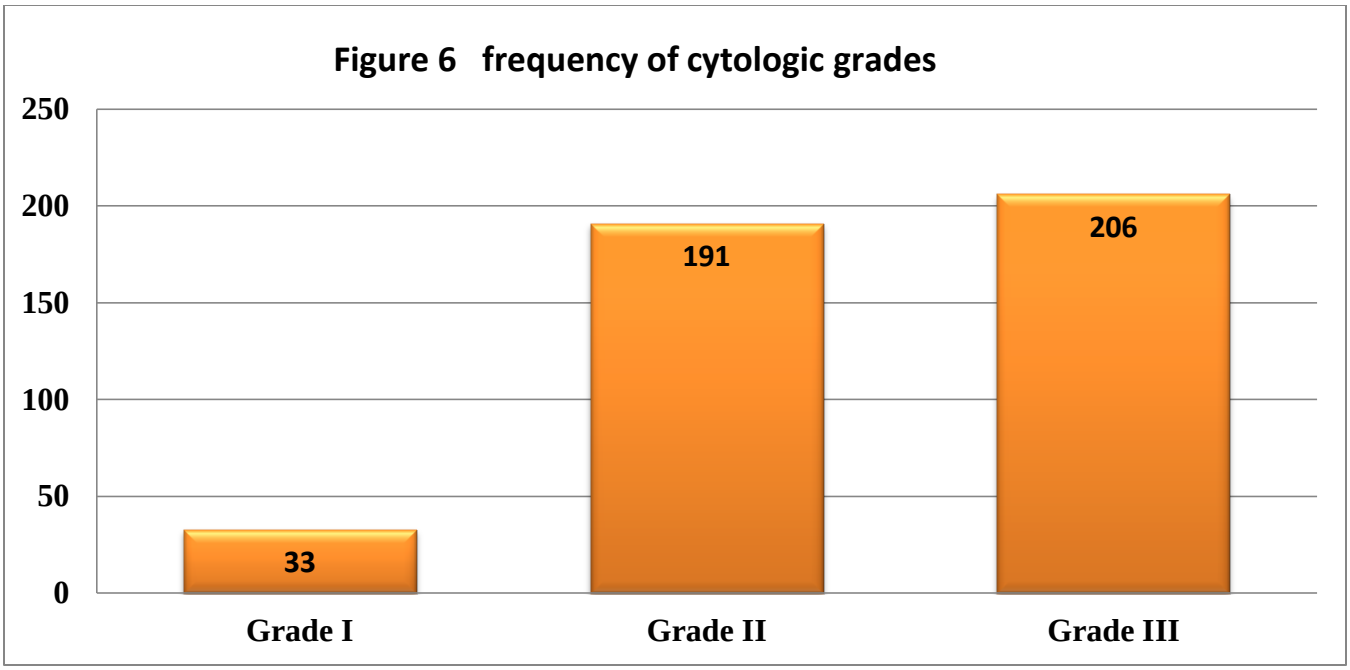
Atypical category	Frequency	Percent	Valid Percent
'Atypical' (Proliferative breast lesion with atypia)	20	1.5	80.0
Low/ grade phyllodes tumor	4	0.3	16.0
Intraductal papillary neoplasm	1	0.1	4.0
Total	**25**	**1.9**	**100.0**

Table 6 Distribution of FNAB breast lesions by age and sex

Diagnostic category	Frequency (valid %) and sex		Age category							
			< 20.0	**20.0 - 29.9**	**30.0 - 39.9**	**40.0 - 49.9**	**50.0 - 59.9**	**60.0 – 69.9**	**70.0+**	**Total**
Malignant	Frequency	Female	2	65	169	132	91	51	18	528
		Male	0	1	3	9	7	7	8	35
		Total(%)	2(0.3%)	66(11.7%)	172(30.5%)	141(25.0%)	98(17.4%)	58(10.3%)	26(4.6%)	**563(100%)**
Suspicious of malignancy	Frequency	Female	2	7	27	10	4	3	0	53
		Male	0	0	1	0	1	0	0	2
		Total(%)	2(3.6%)	7 (12.7%)	28 (50.9%)	10 (18.1%)	5 (9.0%)	3(5.4%)	0(0.0%)	**55(100%)**
Atypical	Frequency	Female	1	5	9	5	1	2	0	23
		Male	0	0	0	1	1	0	0	2
		Total(%)	1(4.0%)	5(20.0%)	9(36.0%)	6(24.0%)	2(8.0%)	2(8.0%)	0(0.0%)	**25(100%)**
Benign	Frequency	Female	53	211	124	74	15	11	5	493
		Male	7	10	7	11	12	12	6	65
		Total(%)	60(10.8%)	221(39.6%)	131(23.5%)	85(15.2%)	27(4.8%)	23(4.1%)	11(2.0%)	**558(100%)**
Insufficient	Frequency	Female	1	25	26	16	11	4	1	84
		Male	1	0	0	0	0	0	0	1
		Total(%)	2(2.4%)	25(29.4%)	26(30. 6%)	16(18.8%)	11(12.9%)	4(4.7%)	1(1.2%)	**85(100%)**
Total	**Frequency**	**Female**	**59**	**313**	**355**	**237**	**122**	**71**	**24**	**1181**
		Male	**8**	**11**	**11**	**21**	**21**	**19**	**14**	**105**
		Total(%)	**67(5.2%)**	**324(25.2%)**	**366(28.5%)**	**258(20.1%)**	**143(11.1%)**	**90(7.0%)**	**38(2.9%)**	**1286(100%)**



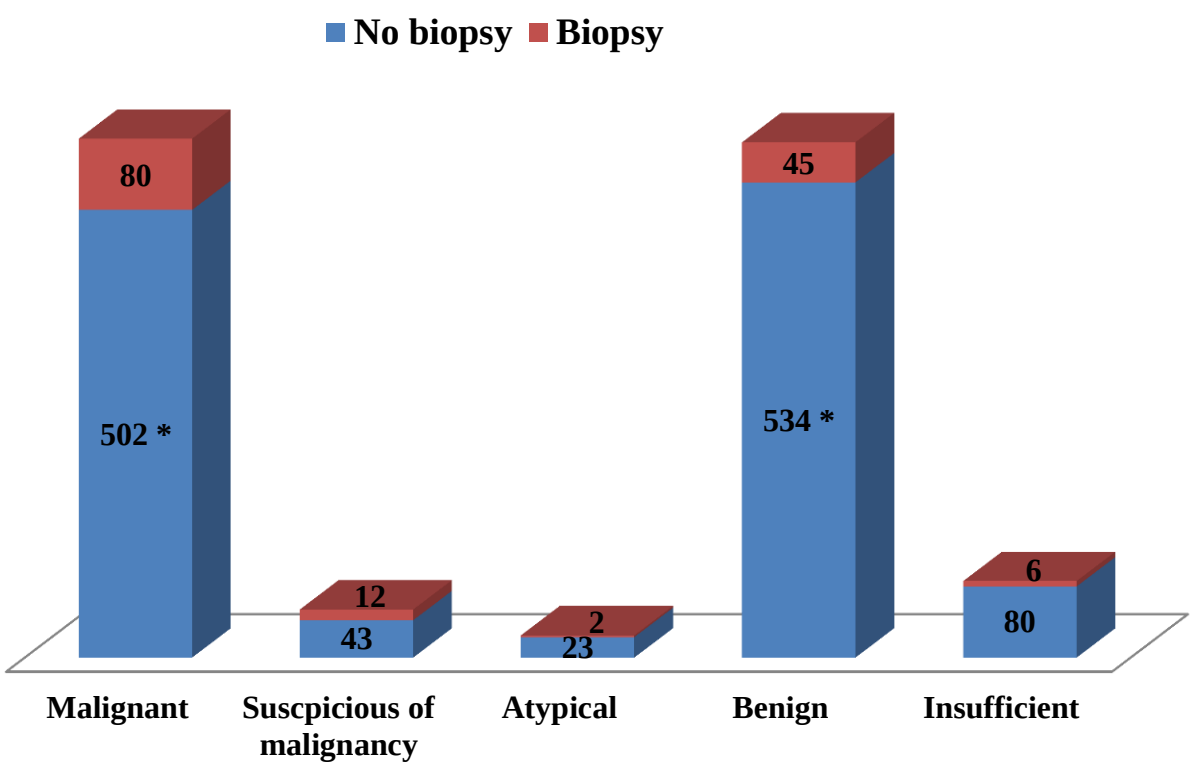
Cytologic nuclear grading was assessed in 430(74.8%) of primary breast carcinomas resulted in 33(7.7%) grade I, 191(44.4%) grade II and 206 (47.9%) of them grade III.



5.3. Comparison of FNAB and histopathology

145 of the 1327 FNAB cases subsequently had biopsies, a biopsy rate of 9.2%. On subsequent histopathology 92 (63.4%) were malignant and 53 (36.5%) were benign cases (Table 7).

Figure 7 A bar chart showing FNAC cases followed by biopsy



**The false positive and false negative rates are related to the number of cases with subsequent histopathology and hence an effect on diagnostic performance outcomes.*

Out of 80 cases 79 (98.75%) of them confirmed to be malignant; the other single case turned out to be fibroadenoma. Among the 45 benign cases 42 (93.3%) of them confirmed to be benign on histopathology. One case of fat necrosis and benign reports became ductal carcinoma and one case of fibroadenoma became DCIS. Out of 12 suspicious of malignancy cases, seven (58.3%) of them became malignant and the rest benign lesions. Only two atypical cases had subsequent histopathology where both of them became malignant (Table 7).

Table 7 Crosstabulation of FNAB and histopathology results			
FNAB	Histopathology		Total
	Malignant	Benign	
Malignant	79 (TP)	1 (FP)	**80**
Suspicious	7	5	**12**
Atypical	2	0	**2**
Benign	3 (FN)	42 (TN)	**45**
Insufficient	1	5	**6**
Total	**92**	**53**	**145**

Out of 86 insufficient/inconclusive cases 6 had subsequent histopathological examination resulted in five benign and one malignant reports; FCC and intraductal papilloma two cases each, a case of fibroadenoma and a case of ductal carcinoma.

A total of 41 cases had both cytologic and histologic grade reports of which 22 of them showed agreement, a concordance rate of 53.7%.

Table 8 Crosstabulation of Cytologic and Histlogic grade agreement					
		Histlogic grade			Total
		Grade I	Grade II	Grade III	
Cytologic grade	Grade I	2	1	0	**3**
	Grade II	2	11	5	**18**
	Grade III	1	10	9	**20**
	Total	**5**	**22**	**14**	**41**

An inter-rater reliability analysis using Kappa statistic (Table13) was performed to determine the agreement between the pathologists assessing the cytologic and histologic grade which was found to be Kappa=0.21 (21% (fair agreement)) without statistical significance (p=0.08).

5.4. Diagnostic performance of FNAB

In an effort to determine the diagnostic performance of FNAB; Sensitivity, Specificity, Positive Predictive Value, Negative Predictive Value and Accuracy were calculated as shown in Table 9 and Table 10.

Table 9 Crosstabulation of histopathology and FNAB for malignancy

		Gold standard (Histopathology)	
		Positive	**Negative**
Test (FNAB)	**Positive**	**TP** (79)	**FP** (1)
	Negative	**FN** (3)	**TN** (42)

Table10 FNAB diagnostic performance of breast lesions

Diagnostic Parameter	Result
Sensitivity (TP/TP+FN)	96 .3%
Specificity (TN/FP+TN)	97.7%
Positive Predictive Value (TP/TP+FP)	98.7%
Negative Predictive Value (TN/TN+FN)	93.3%
Accuracy (TP+TN/P+N)	96 .0%

A chi-square test was also performed to verify association between FNAB and their subsequent histopathology towards to detecting malignancy in which a strong association was found (p value < 0.001).

6. Discussion

Breast cancer is the most common cancer among women in the world and Ethiopia and its incidence is rising especially in low and middle income countries [29] [6]. Breast lesions are one of the diseases commonly seen on day to day basis in our department as T. Gemechu et al. estimated it to be 12% of the total cytology cases [23]. FNAB has been established as an important tool in the evaluation of breast lesions. It has been a preferred technique of pre-op diagnosis for breast pathologies in areas where resources are limited [5].

In this retrospective study female patients were by far the dominant as compared to males accounting for 91.5% of the total cases similar to studies by T. Gemechu et al. (93.5%) [23] , W. Abera et al. at Jimma University medical centers (90.3%) [3] and Ahmed et al.(94.9%) [30]. Considering the distribution of breast lesions with respect to age, more than half of the cases (55.7%) in the malignant category occur in the 4^{th} and 5^{th} decades whereas 63% of benign cases occur a decade earlier in the 3^{rd} and 4^{th} decades of life. These findings are in agreement with studies done by Ahmed et al., 2014 and M. H. Bukhari et al., 2011 [19] [27]. A study done in TASH, oncology unit from 2014 to 2015 showed also a similar result with 61% of breast cancer patients in their 4^{th} and 5^{th} decades of life [31]. However close to 70% of breast cancer cases in the US occur at more advanced age of 50 years and above generally a reflection of breast cancer trend in developed countries [31] [32]. Studies done by Brinton et al. and Allicock et al. on Africans particularly west and middle Africans and African-Americans uncovered important biological differences for the racial disparity as more basal like breast cancer cases are more common in patients of African origin. Patients of this type usually present at younger age and aggressive disease behavior with poor outcome [33] [34]. In spite of this, studies done on molecular subtyping and genetic analysis of breast cancer among Ethiopian women found out more similarity to whites than to Africans (west and middle) and African-Americans [35] [34] . Few authors including Hadgu et al. attributes the young age presentation of breast cancer in Ethiopia to the younger population pyramid [35], yet the age and tumor type paradox could be area of interest for further studies.

According to the IAC Yokohama classification system, our study reveals the dominance of malignant and benign categories as compared to the atypical, suspicious and insufficient categories with a comparable number of cases between the malignant and benign categories amounting to 583 (43.9%) and 579 (43.6%) cases, respectively. As shown in table 1 the overall occurrence is similar for the two categories in the study period even though alternating fluctuations are seen. In contrary to this, many studies show predominance of patients having benign breast lesions coming for cytological evaluation. A study done at AAUMF Department of Pathology between 1994 – 1998 showed superiority of benign lesions which was (74%) [36] [23]. A study done in Jimma University Medical Center showed 79% benign and 16.7% malignant cases from a total of 735 cases [25]. Other studies done in Africa also show the predominance of benign lesions; Embiye et al. (76.7%) [5], Almobarak et al. (55.7%) [19] and Daramola et al. (72.7%) [7]. However, in a study done over sixteen years between1997 – 2012 in TASH, oncology unit found out below 50 cases of breast cancer in 1997 to reach to around 400 by the year 2012 which showed increase in the trend of breast cancer in the hospital [31]. This increased trend of malignancy at the oncology unit is in accordance to this study where the proportion of malignant cases is also increasing in comparison to the earlier study done by T. Gemechu et al as discussed above [36] [23]. This increased trend of malignancy in the department could be due to referral of more patients with malignancy to TASH where it is the only oncology center in the country.

Looking for specific diagnoses made, ductal carcinoma is the most frequent malignant disease in both sexes (93.5%) whereas fibroadenoma (41.1%) and gynecomastia (10.4%) are frequent benign diseases in female and male populations, respectively. Many literatures done in various countries are compatible to our findings with the implication Ethiopian population is probably not different from the rest of the world in this regard even if it is a hospital based study [27] [4] [5].

In this retrospective study, we have found that breast FNAB in TASH is an efficient diagnostic modality with diagnostic accuracy of 96.0%. In addition, the sensitivity, specificity, PPV and NPV are 96.3%, 97.7%, 98.7% and 93.3%, respectively. These findings are comparable and by some parameters superior to a prospective study done at Ethiopian Health Nutrition Research Institute on 102 patients which showed sensitivity, specificity, PPV and NPV of 94.3%, 78.6%,

68.8% and 96.5%, respectively [26]. Similar studies done in different parts of the world particularly in Africa and Asia Showed comparable findings to our study with sensitivity (84.4% - 99.4%), specificity (99.5% -100%), PPV(99.8% - 100%) and NPV (66.7% - 98.5%) [22] [37] [14]. As a part of the 'triple test', breast FNAB misses less than 1% of breast cancers. With concordance of pathologic, radiologic, and clinical impressions, patients can proceed to definitive surgery [18]. Although CNB is reported to be better than FNAB with sensitivities ranging from 85 to 100% and specificities ranging from 86 to 100%; breast ultrasound paired with FNAB can provide patients with substantial cost savings without compromising diagnostic accuracy in resource-limited healthcare settings[18] [38].

The cytologic grade provides relevant prognostic information regarding the aggressiveness of the tumor and lymph node metastasis hence helps to guide the management options [39]. In light of this, a quarter of primary breast carcinomas in this study lack cytologic grade reports. Cyto-histologic agreement shows an overall concordance rate of 53.7% which is low as compared to Indian studies by J. Phukan et al. (72.2%) [40] and Das et al. (71.2%) [41]. Besides, the Cohen's kappa inter-rater reliability statistic of 0.2 represent only fair agreement with kappa below 0.6 indicating inadequate agreement between grade rating pathologists [42]. The national cancer institute recommends to incorporate cytologic grading as integral part of FNAB reports preferably using the Robinson system which is comparable to the Nottingham histologic grading [43] [44].

The occurrence of atypical and suspicious of malignancy cases in this study are 1.9% and 4.1%, respectively which is lower than a study done in Kenya (7.0%, 11.0%) [18], and Montenegro (4.0%, 9.3%) [45]. An Indian study found also 4.8% of atypical and 8.4% of suspicious cases with subsequent histopathology of most atypical to became FCC and most suspicious to became well differentiated malignant tumor [46]. UK NHSBSP in its recommendation puts suspicious rate to be preferably less than 15% [7]. Seven out of twelve suspicious cases confirmed to be malignant in this study. However, in these two categories, it is difficult to do conclusive analysis due to fewer number of corresponding histopathology results.

This study revealed a 6.1% of insufficient/inadequate cases which is a higher figure than the cutoff point less than 5% by the IAC Yokohama system [13]. It is also a higher than 3.3% as

shown by W. Ergete at this department earlier [36].The rate of inadequate aspiration ranges from 0.7% - 47% in various literatures and this is influenced by the nature of the lesion, the available technology and the experience of the operator [13] [47]. It was reported that the nature of the lesion was the most common cause of inadequacy of FNAB accounting for 68% of the inadequate aspirates followed by the experience of the aspirator that accounted for 32% of the inadequacy rate [47]. Both benign and malignant smaller, scirrhous and difficult to stabilize lesions have higher inadequacy rates [13]. A Swiss study (n=1472) found out a higher rate of inadequacy in low stage breast carcinomas as 9.5% in pT1, 5% in pT2 and 0% in pT3 tumors with an overall inadequate rate of 16.2% [48]. Non-palpable lesions are also associated with high rate of inadequacy as high as 34-58% in some reports [47].

The reason for increased inadequate rate in our setup remains to be area of further study. In spite of this, the IAC Yokohama system suggestion to reduce inadequacy rate includes

- adequate training, supervision and mentoring of FNAB operators.
- use of ultrasound/CT guidance for small/non-palpable lesions.
- optimization of smearing and staining qualities.
- performing ROSE or repeating FNAB up to three times.
- triage patient for CNB when needed and correlation with the imaging and clinical findings in the triple test in all cases [13].

7. Conclusion

On a whole, Cases in the malignant and benign categories have comparable frequencies of occurrence and dominant over the atypical, suspicious and insufficient categories. Most of the Malignancies present in the 4^{th} and 5^{th} decades of life whereas benign cases happen a decade earlier. With high sensitivity and specificity, most benign and malignant breast lesions can be reliably diagnosed by FNAB. In conjunction, the insufficient, suspicious of malignancy and atypical reports needs to be supported by image guided aspiration and/or invasive procedures like excisional biopsy and CNB not to miss malignancy. In general, Fine needle aspiration continues to be an acceptable and reliable method for the preoperative diagnosis of breast lesions particularly in resource limited countries.

8. Limitations of the study

- Relatively limited (small) number of data having both cytologic and histopathololologic reports especially in the atypical, suspicious and insufficient cases since most of them done in different centers.
- The low rate of cyto-histopathology correlation which results in low rate of false positive and false negative results may have an effect in showing the actual diagnostic performance of FNAB
- There was difficulty in collecting data due to disorganized manner of data keeping.
- Incomplete and faulty documentation of the print result and request paper.
- Some of the cytologic reports lack the standard IAC Yokohama category, specific diagnosis and/or differential diagnosis.

9. Recommendations

- Future study involving larger sample size having both cytology and histology reports to affirm the importance of FNAB is advised.
- Following the IAC Yokohama system of reporting as to produce a universal way of communication is recommended.
- Patients and clinicians should be encouraged to have imaging study as part of the triple test.
- More studies need to be done to investigate pattern of breast lesions in the other parts of the country.
- Focused studies in collaboration with other disciplines to better understand the disease condition in the Ethiopian context and to optimize the patient care is recommended.
- Multidisciplinary approach of patient evaluation to implement ROSE and improve the efficiency of FNAB should be in the plan.
- Proper and organized data keeping scheme should be established by the department.
- As a media of communication reports/results and requests should be clear, neat and complete.
- Developing and introducing quality assurance mechanisms in aspiration, smearing and staining technique and implementation should be done to improve the quality of FNAB.

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11. ANNEXES

The International Academy of Cytology Yokohama System for Reporting Breast Fine Needle Aspiration (Diagnostic category)

1. Insufficient/Inadequate: This category is used when the smears are too sparsely cellular or distorted to allow a microscopic diagnosis or the aspirate is inconsistent with the clinical and image findings. Since this is a subjective diagnosis an explanation of why the sample is insufficient/inadequate should be given.
2. Benign: This diagnostic category is used when the sample is adequate and shows no evidence of malignancy.
3. Atypical : This category is used with the presence of cytological features seen predominantly in benign processes or lesions, but with the addition of some features that are uncommon in benign lesions and which may be seen in malignant lesions
4. Suspicious of malignancy: This diagnostic category is used when the smears show features suggestive of but not diagnostic of malignancy. The malignant cells may be too scanty, obscured by artifact or show atypical features more marked than in the atypical or indeterminate category but not diagnostic of malignancy.
5. Malignant: This diagnostic category is used when the aspirate is clearly malignant. This diagnostic category includes invasive breast carcinoma, ductal carcinoma in-situ and other malignancies.

Table 11 Diagnostic features according to cytologic grade of breast cancer[49].

Feature	Score
Tubule and gland formation	
Majority of tumor (75%)	1
Moderate degree (10-75%)	2
Little or none (<10%)	3
Nuclear pleomorphism	
Small, regular, uniform cells	1
Moderate increase in size and variability	2
Marked variation	3
Mitotic count	
Dependent on microscopic field area	1-3
Total score **(Add gland formation, nuclear pleomorphism, and mitotic count)**	**Final grading**
3-5	**Grade I**
6 or 7	**Grade II**
8 or 9	**Grade III**

Table 12 Semiqualitative method for assessing histological grade in breast cancer [50].

	Grade I	Grade II	Grade III
Nucleus			
Size	Small	Medium	Large
RBC diameter	<2x	2-3x	>3x
Pleomorphism	Slight	Moderate	Marked/Bizzare
Membrane	Smooth	Slightly irregular	Irregular
Nucleoli	Inconspicous	Conspicous	Prominent, multiple
Chromatin	Fine	Granular	Coarse, irregular
Mitosis/10HPF	0-1	2-4	>5
Cell			
Size	Small	Medium	Large
Variation	Monomorphic	Moderate variation	Pleomorphic
Groups			
Gland formation	>75%	10-75%	<10%
Clusters	Predominate	Increased single cells	Many single cells

Table 13 Interpretation of Chone's kappa to assess inter-rater reliability agreement

Value of kappa	Level of agreement
<0.00	None
0.00-0.20	Slight
0.21-0.40	fair
0.41-0.60	Moderate
0.61-0.80	Substantial
0.81-1.00	Near perfect