Cytological grading of breast carcinomas correlating with ultrasonographic findings, histopathologic grade and Her2neu oncogene status in women of age group thirty to sixty five years

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Abstract

Background: Breast cancer is a malignant disease with a heterogeneous prognosis. It is the most common cancer in the urban Indian population. Women over 30 years of age are especially prone to develop this type of cancer. There are various prognostic factors that have been studied in invasive ductal carcinomas to predict the tumour behaviour. As fine needle aspiration cytology is an important investigation for diagnosis and grading of the tumour, preoperative evaluation of the tumour behaviour can be studied. Aim: The aim of the study was to diagnose and grade the tumours cytologically by fine needle aspiration cytology (FNAC), to study the tumour behaviour by ultrasonography, by histo pathological examination and also to study the her2neu oncogene overexpression of the malignant tumours. Materials & methods: The present study was carried out in Gauhati Medical College, Guwahati, Assam from May, 2010 to July, 2012. The patients presenting with breast lumps were selected for the study. FNAC smears were stained by Papaniculaou and May Grunwald Giemsa stain. Ultrasonography of the breast lumps were done. Histopathological examination was done using the H & E stain. Immunohistochemical study was done to know the her2neu oncogene overexpression by the tumours. Results & observations: The age of patients with malignant tumours were found in the pre and perimenopausal age group. 48% of the malignant tumours were infiltrating duct carcinoma and of high grade. Her2neu overexpression was seen in 35% of the malignant tumours. Conclusion: A good correlation exists between the cyto-histopathologic grade of the tumours (83.3%), BIRADS 5 category on ultra sonography with histopathologic grade (98.5%) and her2neu oncogene overexpression along with histopathologic grade (93.3%). Therefore, cytological grading indicates an important prognostic marker to know the behaviour of breast carcinomas.

Keywords: fine needle aspiration cytology, histopathology, duct carcinoma, immunohistochemistry

Introduction

Breast cancer is a malignant disease with a heterogeneous prognosis. It is the most common cancer in the urban Indian population based on National Cancer Registry Programme, ICMR1. Women over 30 years of age are especially prone to develop this type of cancer, but can happen to younger women also.

There are various prognostic factors that have been studied in invasive ductal carcinomas to predict the tumour behaviour and response to chemotherapy. Attempts are being made to assess these prognostic factors preoperatively on aspiration cytology material. As FNAC of breast lumps is one of the baseline investigation done on outpatient basis, attempts to grade the tumour on cytology material is being studied. In the era of neoadjuvant chemotherapy (NAT), the FNA material is often an important investigation for preoperative evaluation. NAT is potentially advantageous to down stage the tumour. This necessitates grading of the tumor on the aspiration material obtained before surgery.
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Materials and methods
The present study was carried out in Gauhati Medical College, Guwahati, Assam. This study was carried out from May, 2010 to July, 2012. The patients presenting with breast lumps were selected for the study.

Inclusion Criteria: Patients whose FNAC diagnosis was ductal carcinoma breast and cases with cytological features suspicious of malignancy and subjected for ultrasonography and histopathology were included in the study.

Exclusion criteria: Patients who did not have a histopathological confirmation of their cytology were excluded from the study.

Demographic and Clinical details
The following details of patient’s history and clinical details were obtained:
1. Age of the patients
2. Smoking
3. Alcohol
4. High fatty diet
5. Obesity
6. Early menarche
7. Breast feeding
8. Location of the tumours
9. Size of the tumours

Plan of the study
Routine investigations like Blood R/E were performed. Fine needle aspirations were performed on the presenting breast lumps and also on the lymphadenopathies after permission from the ethical committee and consent of the patient.

Study design
Fine needle aspiration cytology material diagnosed and malignant cases graded → ultrasonographic findings correlated → histopathological examination done and malignant cases graded → Immunohistochemical study done on paraffin blocks for Her2neu oncogene status of patients with breast carcinoma. For FNAC smears, Papanicolaou (PAP) and May Grunwald Giemsa (MGG) stains were used. Statistical analysis was done by using Spearman correlation coefficient and Chi square tests.

Results and observations
Out of 829 cases of breast lumps, 20% were Non neoplastic, 61% benign neoplastic and 19% were malignant neoplastic lesions. (Table No. 1).

Table No. 1: Table showing cytological diagnosis

<table>
<thead>
<tr>
<th>FNAC diagnosis</th>
<th>No. of Cases</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Non neoplastic</td>
<td>166</td>
<td>20%</td>
</tr>
<tr>
<td>Benign neoplastic</td>
<td>506</td>
<td>61%</td>
</tr>
<tr>
<td>Malignant neoplastic</td>
<td>157</td>
<td>19%</td>
</tr>
</tbody>
</table>

Table No. 2: Table showing age distribution of the patients.

<table>
<thead>
<tr>
<th>Age groups</th>
<th>Percentage of cases</th>
<th>Age in years</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pre &amp; Perimenopausal</td>
<td>45%</td>
<td>&lt;47</td>
</tr>
<tr>
<td>Menopausal</td>
<td>22%</td>
<td>47-52</td>
</tr>
<tr>
<td>Postmenopausal</td>
<td>33%</td>
<td>&gt;52</td>
</tr>
</tbody>
</table>

Table No. 3: Table showing correlation of age in years with histopathological grade of tumours.

<table>
<thead>
<tr>
<th>HPE grade</th>
<th>&lt;47</th>
<th>47-52</th>
<th>&gt;52</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>5</td>
<td>4</td>
<td>6</td>
<td>15</td>
</tr>
<tr>
<td>II</td>
<td>16</td>
<td>5</td>
<td>8</td>
<td>29</td>
</tr>
<tr>
<td>III</td>
<td>25</td>
<td>8</td>
<td>10</td>
<td>41</td>
</tr>
<tr>
<td>Total</td>
<td>46</td>
<td>15</td>
<td>24</td>
<td>85</td>
</tr>
</tbody>
</table>

Table No. 4: Table showing correlation of size of tumours with histopathological grade.

<table>
<thead>
<tr>
<th>Size of tumours</th>
<th>Gr. I</th>
<th>Gr. II</th>
<th>Gr. III</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;2 cm.</td>
<td>4</td>
<td>4</td>
<td>4</td>
<td>13</td>
</tr>
<tr>
<td>2-5 cm.</td>
<td>9</td>
<td>8</td>
<td>17</td>
<td>34</td>
</tr>
<tr>
<td>&gt;5 cm.</td>
<td>2</td>
<td>16</td>
<td>20</td>
<td>38</td>
</tr>
<tr>
<td>Total</td>
<td>15</td>
<td>29</td>
<td>41</td>
<td>85</td>
</tr>
</tbody>
</table>

Table No. 5: Table showing correlation of cytology – histopathologic grade.

<table>
<thead>
<tr>
<th>Cytologic grade</th>
<th>Histopathologic grade</th>
<th>Concordance rates (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Gr. I</td>
<td>Gr. II</td>
</tr>
<tr>
<td>Grade I</td>
<td>19</td>
<td>14</td>
</tr>
<tr>
<td>Grade II</td>
<td>27</td>
<td>1</td>
</tr>
<tr>
<td>Grade III</td>
<td>39</td>
<td>0</td>
</tr>
<tr>
<td>Total</td>
<td>85</td>
<td>15</td>
</tr>
</tbody>
</table>

Table No. 6: Table showing Her2neu positivity.

<table>
<thead>
<tr>
<th>Her2neu positivity</th>
<th>No. of cases</th>
</tr>
</thead>
<tbody>
<tr>
<td>Strongly positive</td>
<td>30</td>
</tr>
<tr>
<td>Weakly positive</td>
<td>4</td>
</tr>
<tr>
<td>Negative</td>
<td>51</td>
</tr>
</tbody>
</table>
Most of the patients with malignant tumours belonged to the pre and perimenopausal age group (<47 years). (Table No. 2). Malignant tumours were found to be mostly in the upper outer quadrant.

Out of 166 cases of non neoplastic lesions, 88 cases were reported as Benign proliferative lesions, 42 cases Mastitis, 26 cases Galactoceles/lactational change, eight cases Epidermal cysts and two cases were reported as Fat necrosis.

Benign neoplastic lesions were reported to be the maximum that is 506 no. of cases (61%). Out of them 491 (97%) were Fibradenomas, six cases were Benign phylloides tumour, four cases were Duct papilloma, three cases were Lipoma and two cases Capillary hemangioma.

Out of 157 cases of malignant neoplastic lesions, 139 (88.5%) cases were reported as Duct carcinoma. Rest of the cases were Infiltrating duct carcinoma, Lobular
carcinoma, Medullary carcinoma, Malignant phyllodes tumour, Metaplastic carcinoma and Mucinous carcinoma.

Grading of the tumours were done according to Robinson's grading system\(^1\). On grading the tumours on FNAC slides we reported 54% as high grade tumours, 30% Intermediate grade and 16% as low grade tumours.

**Histopathological diagnosis**

After excision of the tumours with or without mastectomy 340 cases were evaluated for histopathological examination using the routine hematoxylin and eosin (H & E) stain. The detailed gross and microscopic features were studied in each of the specimen. Report of the cases was as follows: 37 cases (11%) non neoplastic lesions, 218 cases (64%) benign neoplastic lesions and 85 cases (25%) as malignant neoplastic lesions. Out of the non neoplastic lesions majority of the cases was fibrocystic disease. Fibroadenomas were predominant among the benign neoplastic lesions and Infiltrating duct carcinoma NOS among the malignant neoplastic lesions.

There were 85 histologically proven cases of malignancy. 48% of the cases were with metastasis to the regional lymph nodes and 52% did not show any metastasis.

All these cases were further graded depending upon the histopathological features described by modified Bloom Richardson system\(^1\). It was seen that out of the 85 cases 41 cases (48%) were high grade lesions, 29 cases (34%) of intermediate grade and 15 cases (18%) of low grade features.

On correlating the age of the patients with grade of the tumours, it was found that high grade lesions were common among patients below 47 years. (Table No.3).

Considering the size of the tumours, it was observed that tumours of more than 5cm in size were predominantly high grade lesions. (Table No. 4).

There was a significant correlation in the cytohistopathologic grade of the malignant tumours. (Table No. 5).

Out of the total 422 number of cases subjected for ultrasonography, 92 cases (21.8%) were reported as BIRADS 5 category (BIRADS- Breast imaging reporting and data system).

Out of these 92 cases, the authors could correlate the findings with the histopathologic grade in 85 of them. It was observed that the concordance rate was 98.5%.

**Her2neu oncogene positivity**

Her2neu oncogene overexpression were seen in 35% of the malignant tumours. (Table No.6). Her2neu positive cases were mostly found in the age group of 41-50 years. The strongly positive cases were predominantly Infiltrating duct carcinoma and rest were Duct carcinoma in situ. There was a strong correlation of Her2neu positive cases with high grade tumours giving a concordance rate of 93.3%.

**Discussion**

In the present study, breast carcinomas were commonly found in the pre and perimenopausal age group. Considering the demographic and risk factors, breast cancer are seen to be related with smoking, early menarche, no breast feeding, fatty diet and location. No significant correlation has been found with alcohol intake and size of the tumours. On FNAC diagnosis, the predominant non neoplastic lesions were benign proliferative lesions, benign neoplastic lesions were mostly fibroadenomas and duct carcinomas among the malignant lesions.

Lymph node metastasis were found to be in lesser numbers of cases of the malignant tumours. The observation showed insignificant correlation between axillary lymph node metastasis and histopathologic grade (\(p \text{ value } 0.36\)). On HPE, the non neoplastic lesions were fibrocystic disease. Fibroadenomas were common among benign neoplastic lesions and malignant neoplastic lesions were mostly Infiltrating duct carcinoma NOS. High grade tumours are featured more commonly in the younger (Premenopausal) age group (<47 years). Overall the grade of the tumour did not show a significant correlation with the age of the patient (\(p \text{ value } >0.05\)).
Grading of the malignant tumours both on cytology and histopathology showed high grade lesions to be more common (Cytology -54% and HPE -48%). A strong correlation exists between the Cytohistopathologic grade of the tumours. The average concordance rate was found to be 85.3%. Correlating the data of ultrasonography (BIRADS-5) with the histopathologic grade gave a concordance rate of 98.5%.

Her2neu oncogene overexpression detected by immunohistochemical method was seen in 35% of the tumours with ductal carcinoma. Her2neu positive cases was found predominantly in the age group. Her2neu oncogene overexpression were seen in 93% of the cases of Infiltrating duct carcinoma NOS and rest were Duct carcinoma in situ. Her2neu over expression correlated well with the histopathologic grade of the malignant tumours.

Comparing the data with many of the authors gave a clear view of the demographic and risk factor which correlated well with the literatures having a significant p value of <0.001. High grade malignant tumours were predominant in our study which was similar to the literatures. The cyto-histopathologic correlation were comparable with many other literatures and showed a concordance rate of >80%.3,4,7,8

Breast ultrasonography correlating with histopathological grade show a high sensitivity.13,10,11

The results of her2neu oncogene status of the patients detected by immunohistochemical method was found to be variable compared with the data shown by different authors. (Her2neu overexpression:- our study -35%; Kamil et al12- 31.5%; Poonam et al13- 61.7%; Choi14, USA, US- 16% and Native Korea- 47.5%; Moriki et al15, -27%; ).

The present study had limitations because most of the patients failed to attend the OPD for follow-up. Therefore a more extensive study with follow-up of the patients is necessary.

Conclusion
A good correlation exists between the cytohistopathologic grade of the tumours (85.3%), BIRADS 5 category on ultrasonography and histopathologic grade (98.5%) and her2neu oncogene overexpression along with histopathologic grade (93.3%). Cytological grading indicates an important prognostic marker to know the behaviour of breast carcinomas.

Therefore, highlighting the biological behaviour of breast cancer would aid the clinicians to adopt proper clinical management strategies and pave the way for a quality survival of the patients.

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References
7. Meena SP, Hemrajani DK, Joshi N. Acomparative and evaluative study of cytological and histological grading system profile in malignant neoplasm of breast
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