Original Research Article

A predictive and prognostic biomarker profile of carcinoma breast

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1. Introduction

Carcinoma of the breast is one of the leading sites of cancer all over the world.1 In India it is the top ranked cancer in females with age adjusted rate as high as 25.8 per 100000 women.2 Breast cancer is recognised as a heterogenous disease, with different biological properties across the different subtypes. Accordingly, the standard of care includes the immunohistochemical staining for every case, to classify it into different subtypes based upon the scoring for at least three biomarkers- Estrogen receptor, Progesterone receptor and Her 2 neu. These are called as predictive biomarkers because the neoadjuvant or adjuvant therapy of carcinoma breast is decided by the particular subtype. Additionally Her2 neu is also a prognostic biomarker which has bearing on the disease free survival as well as overall survival of the patient.

The hormone receptor profile of carcinoma Breast cases in India has been studied by many authors, but very few reports exist about the profile of patients from Central India.3–11 Genetic tests like Oncotype DX or Mammaprint are validated tests to predict the recurrence of disease. But the cost is beyond the reach of a majority of patients. Ki 67 is a surrogate predictive biomarker which is much more economical as compared to the genetic tests.12

**Context:** The immunohistochemical (IHC 4) biomarker profile is part of the standard histopathology report of all newly diagnosed and recurrent cases of carcinoma Breast. This profile is the basis for all neoadjuvant and adjuvant treatment planning in these cases.

**Aims:**
1. To study the IHC4 biomarker profile of Carcinoma Breast cases at our Institute.
2. To study the correlation of the five types of molecular subgroups with various clinical and histological parameters.

**Settings and Design:** 271 cases of carcinoma breast diagnosed and treated at our Institute, during the period 1st July 2017 till 30th June 2018. This is a prospective, observational study.

**Materials and Methods:** All the cases of biopsy proven carcinoma Breast were subjected to immunohistochemical staining for four markers- ER, PR, Her2, and Ki67. Formalin Fixed Paraffin Embedded tumor tissue was stained for 4 biomarkers and scored with appropriate method. (Interpretive Guide: ASCO - CAP Test Guidelines Recommendations 2013)

Manual method of staining was employed, using commercially available reagents. The cases were classified into five molecular subtypes.

**Results:**
Triple negative breast carcinoma was the most frequent subgroup, followed by the luminal B and A types and the Her2 enriched cases were lowest in number. A few cases showed triple positive staining pattern.

**Conclusions:** The IHC 4 biomarker findings in every case of carcinoma has a direct impact on the treatment decision making and also on risk stratification of the patients.

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With this background, the study was conducted to profile all the patients of carcinoma breast diagnosed at our institute.

2. Materials and Methods

271 newly diagnosed and recurrent cases of carcinoma breast registered from 1st July 2017 till 30th June 2018 form the subject material of the study.

All the cases were clinically examined and staged after radiological evaluation.

As per the NCCN guidelines, all Stage I, IIa, IIb, IIIa cases were subjected to FNAC and if necessary to a core needle biopsy to document the tissue diagnosis of Carcinoma Breast. All these cases underwent surgery as the first modality of treatment. The surgical specimens were processed, tissue sections studied and the appropriate sections were subjected to immunohistochemical staining for the biomarkers to help plan the adjuvant therapy.

In cases of locally advanced breast cancer (IIIa, IIIb, IIIc - LABC), the patients underwent a core needle biopsy to document the disease as well as to determine the molecular subtype by immunohistochemistry for biomarkers to plan the neoadjuvant therapy.

In every case, Formalin fixed paraffin embedded sections were stained by the manual method for the four biomarkers- ER, PR, Her2 neu and Ki67.

The standard clones were used for the primary antibodies.

All the cases were typed into five subtypes (St Gallen (Vienna) 2013 consensus classification)\textsuperscript{13}

1. Luminal A – ER positive, HER2 negative, Ki-67 < 15%, and PR high;
2. Luminal B (HER2 negative) – ER positive, HER2 negative, and either Ki-67 high or PR low;
3. Luminal B-like (HER2 positive) – ER positive, HER2 over expressed or amplified, any Ki-67, and any PR;
4. HER2 enriched – HER2 over-expressed or amplified, ER and PR absent;
5. Triple negative – ER and PR absent and HER2 negative.

3. Results

The majority of cases were seen in female breast (267), only four cases were found in male breasts (four). The pre and post menopausal age group distribution did not show significant difference. Infiltrating Duct Carcinoma-Not otherwise specified- was the most frequent histological type (263) and other variants like Infiltrating Lobular Carcinoma (three), Papillary Carcinoma (one), Medullary Carcinoma (one), Metaplastic Carcinoma (three) were very few. (Figure 1)

Triple Negative Breast Cancer was the most frequent molecular subtype, followed closely by Luminal A type.

Only Her2 positive was the least frequent. (Figure 2)

Of the triple negative group, significant number of cases were having T size 2 and 3 with N1 status and so more likely to show higher p stages. Table 1

The luminal A subgroup also showed T2 as the most frequent T size, but the node status was more likely to be N0 and thus showed a lower p stage. Table 2

Both the luminal B types and the Her 2 enriched subtype showed similar associations as the Luminal A subtype. Tables 3, 4 and 5

Most interesting was The Ki67 score which showed a significant concentration at different values between the subtypes, Luminal A showing the lowest score and TNBC the highest. Table 6

4. Discussion

Triple negative Breast cancer is the most frequent subtype (30%) found in this study. This observation is similar
Table 3: Correlation of Luminal B (Her2 Negative) with T size, N status and p Stage

| T size |     | Significant |
|--------|-----|-------------|
| T1     | 0.37|             |
| T2     | 14.02|            |
| T3     | 4.30|             |
| T4     | 0.37|             |
| N Status |    |             |
| N0     | 9.59| Significant |
| N1     | 7.38|             |
| N2     | 1.85|             |
| N3     | 1.11|             |
| Stage  |     |             |
| I      | 0.37|             |
| IIa,IIb| 13.65|           |
| IIIa   | 4.06|             |
| LABC   | 1.11|             |

Table 4: Correlation of Luminal B -like(Her2 Positive) with T size, N status and p Stage

| T size |     | Significant |
|--------|-----|-------------|
| T1     | 0   |             |
| T2     | 11.07|            |
| T3     | 1.85|             |
| T4     | 0   |             |
| N Status |    |             |
| N0     | 5.90| Significant |
| N1     | 6.27| Significant |
| N2     | 0.37|             |
| N3     | 0.37|             |
| Stage  |     |             |
| I      | 0   |             |
| IIa,IIb| 10.33|           |
| IIIa   | 1.85|             |
| LABC   | 0.74|             |

Table 5: Correlation of Her 2 enriched group with T size, N status and p Stage

| T size |     | Significant |
|--------|-----|-------------|
| T1     | 0.37|             |
| T2     | 9.59| Significant |
| T3     | 1.85|             |
| T4     | 0.37|             |
| N Status |    |             |
| N0     | 3.69| Significant |
| N1     | 6.64|             |
| N2     | 1.48|             |
| N3     | 0.37|             |
| Stage  |     |             |
| I      | 0.37|             |
| IIa,IIb| 8.86|           |
| IIIa   | 1.48|             |
| LABC   | 1.48|             |

to that reported previously. Since all three receptors are not expressed by this tumor subtype, these patients do not benefit from hormonal treatment, they have to be managed by chemotherapy. The reported prognosis of this group is poorer as compared to the hormone receptor positive subtype.14,15

The Ki 67 score in this study group was found to be the highest, prompting a close follow up to assess recurrence and progression.

Luminal A (26%) was the next most frequent group. Since these tumors express the hormone receptors, they respond to hormonal treatment directed against these targets. Consequently this group as a whole is reported to have a better prognosis.15 The Ki 67 scores in this group was the lowest, which also predicts a better outcome as compared to other subtypes.

The two Luminal B subtypes were the next in frequency (Her2 negative 19% and Her2 Positive13%). Amongst these subtypes, the luminal B/HER2– is reported to have had higher risk of mortality than the luminal B/HER2+.15

The Ki 67 scores in both these groups were low, without much statistical difference which predict a better outcome as compared to TNBC and Her 2 enriched subtypes.

The least number of cases were of the Her2 enriched subtype. This group of patients benefit by targeted therapy in the form of Herceptin. However, the Ki67 score of this group was found to be high, again prompting a close follow up to detect early recurrence.

The Ki 67 scores of different molecular types show interesting findings. The Luminal A cases show the lowest
Table 6: Correlation of Ki67 score - a significant concentration at different values between the subtypes, Luminal A showing the lowest score and TNBC the highest.

| S. No | Molecular type | Ki 67 score | 0-15 | 16-25 | 26-35 | 36-45 | 45 onwards | Total P value | Significance | Observations significantly concentrate at |
|-------|----------------|-------------|------|-------|-------|-------|-----------|--------------|-------------|---------------------------------|
| 1     | Luminal A      | 69          | 0    | 0     | 0     | 0     | 0         | 69           | 0           | Significant 0-15                   |
| 2     | Luminal B HN   | 8           | 21   | 8     | 9     |       | 6         | 52           | 0.0074      | Significant 16-25                 |
| 3     | Luminal B HP   | 8           | 15   | 2     | 4     |       | 6         | 35           | 0.0226      | Significant 16-25                 |
| 4     | Her 2 enriched | 3           | 3    | 17    | 5     |       | 5         | 33           | 0.0006      | Significant 26-35                 |
| 5     | TNBC           | 8           | 9    | 20    | 17    |       | 28        | 82           | 0.0023      | Significant >45                  |

Proliferation index whereas the TNBC type shows the highest Proliferation index. These findings will have to be correlated with the disease free survival rates in follow up.

5. Conclusion

Indian data on breast cancer is being published from different geographical regions of the country and gradually a clearer picture is emerging regarding the distribution of different molecular types of carcinoma breast. As more and more patients get tested and typed for the hormonal markers, and long term survival data emerges, we will have a better idea about the challenge of treating Breast cancer in our country.

6. Source of funding

None.

7. Conflict of interest

None.

References

1. Torre L, Islami F, Siegel R, Ward E, Jemal A. Global Cancer in women: Burden and trends. *Cancer Epide Biomark Prev*. 2017;26(4):1–15.
2. Malvia S, Bagadi S, Dubey U, Saxena S. Epidemiology of breast cancer in Indian women. *Asia-Pacific J Clin Oncol*;17(12):289–295.
3. Desai SB, Moonim MT, Gill AK, Punia RS, Naresh KN, et al. Hormone receptor status of breast cancer in India: A study of 798 tumours. *Breast*. 2000;9:267–270.
4. Shet T, Agrawal A, Nadkarni M, Palkar M, Havaldar R, Parmar V. Hormone receptors over the last 8 years in a cancer referral center in India: What was and what is? *Indian J Pathol Microbiol*. 2009;52:171–174.
5. Doval DC, Sharma A, Sinha R, Kumar K, Dewan AK, Chaturvedi H. Immunohistochemical profile of breast cancer patients at a tertiary care hospital in New Delhi. *India Asian Pac J Cancer Prev*. 2015;16:4959–4964.
6. Patnayak R, Jena A, Rukmangadha N, Chowkan AK, Sambasiviah K, et al. Hormone receptor status (estrogen receptor, progesterone receptor), human epidermal growth factor-2 and p53 in South Indian breast cancer patients: A tertiary care center experience. *Indian J Med Paediatr Oncol*. 2015;36:117–122.
7. Ghosh J, Gupta S, Desai S, Shet T, Radhakrishnan S, et al. Estrogen, progesterone and HER2 receptor expression in breast tumors of patients, and their usage of HER2-targeted therapy, in a tertiary care centre in India. *Indian J Cancer*. 2011;48:391–396.
8. Munjal K, Ambaye A, Evans MF, Mitchell J, Nandedkar S, et al. Immunohistochemical analysis of ER, PR, Her2 and CK5/6 in infiltrative breast carcinomas in Indian patients. *Asian Pac J Cancer Prev*. 2009;10:773–778.
9. Singh R, Gupta S, Pawar SB, Pawar RS, Gandham SV, et al. Evaluation of ER, PR and HER-2 receptor expression in breast cancer patients presenting to a semi urban cancer centre in Western India. *J Cancer Res*. 2014;10:26–28.
10. Ambroise M, Ghosh M, Mallikarjuna VS, Kurian A. Immunohistochemical profile of breast cancer patients at a tertiary care hospital in South India. *Asian Pac J Cancer Prev*. 2011;12:625–629.
11. Akhtar M, Dasgupta S, Rangwala M. Triple negative breast cancer: an Indian perspective. *Breast Cancer*. 2015;7:239–243.
12. Sahebjam S, Aloyz A, Plivdzic D, Bristson M, Bouganim N, et al. Ki 67 is a major but not the sole determinant of Oncotype Dx recurrence score. *Ar J of Cancer*. 2011;105(13):42–45.
13. Falck AK, Ferno M, Bendall PO, Ryden L. St Gallen molecular subtypes in primary breast cancer and matched lymph node metastases - aspects on distribution and prognosis for patients with luminal A tumours: results from a prospective randomised trial. *BMC Cancer*. 2013;13:558.
14. Dent R, Trudeau M, Pritchard KI, Hanna WM, Kahn HK, Sawka CA. Triple-negative breast cancer: Clinical features and patterns of recurrence Clini. *Cancer Res*. 2007;67(15):4429–4434.
15. Parise CA, Caggiano V. Breast Cancer Survival Defined by the ER/PR/HER2 Subtypes and a Surrogate Classification according to Tumor Grade and Immunohistochemical Biomarkers. *J Cancer Epidem*. 2014;469251:1–11.

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