Journal of Advanced Medical and Dental Sciences Research

@Society of Scientific Research and Studies

Journal home page: www.jamdsr.com doi: 10.21276/jamdsr ICV 2018= 82.06 UGC approved journal no. 63854

(e) ISSN Online: 2321-9599; (p) ISSN Print: 2348-6805

Original Research

Ki-67 as a prognostic marker in patients with Primary Breast Cancer

Siddharth Gangwar[1], (Col). Vijai Singh[2]

[1]Junior Resident, [2]Associate Professor, Department of Pathology, Hind Institute of Medical Sciences, Safedabad, Barabanki, U.P., India

ABSTRACT

Introduction: Breast cancer is amongst most common cancer in females and is 2nd most common reason of death in patients with cancer in the world. Cell proliferation has an important function in the clinical behavior of invasive breast cancer. We aimed to assess the status of Ki-67 in patients with primary breast cancer and evaluated the association of this tumor marker with other clinico-pathologic and prognostic factors. **Materials and Method:** The current study consisted of 50 patients with primary breast cancer admitted to the surgical ward of Hind Institute of Medical Sciences Safedabad, Barabanki. Evaluation of Ki-67 IHC slides were done and reported. Among 50 patients, 16developed grade 2 tumors, and21 were below 50 years age. 29 cases were Ki-67 positive with more than 1% tumor nuclei stained, and 15 cases had tumors with more than 15% of Ki-67 expression. **Results**: There was no significant correlation between Ki-67 and patient's age, tumor size and grade however and there was a marginally significant relationship between lymph node status and Ki-67 expression. **Conclusion**: A reliable estimation of different prognostic factors in breast cancer patients is required for the selection of an optimal therapeutic strategy. The attention has been focused on the markers of tumor biology.

Key words: Breast Cancer, Grade, Lymph Node, Prognosis, Tumor Markers.

Received: 10 February, 2019 Revised: 27 March, 2019 Accepted: 29 March, 2019

Corresponding author: Dr. (Col.) Vijai Singh, Associate Professor, Department of Pathology, Hind Institute of Medical Sciences, Safedabad, Barabanki, U.P., India

This article may be cited as: Gangwar S, Singh V. Ki-67 as a prognostic marker in patients with Primary Breast Cancer. J Adv Med Dent Scie Res 2019;7(4): 125-128.

INTRODUCTION:

Breast cancer is amongst most common cancer and the second most common cause of death in women. Worldwide, one out of fourteen women develops breast cancer amid 0-79 years of age. Prognostic factors are very important breast cancer diagnosis as they help in identification of high-risk patients. In search for the potential prognostic indicators of breast cancer, focus is being shifted on tumor markers. Cell proliferation has a significant role in the clinical behavior of invasive breast carcinoma. Increased cell proliferation is associated with poor prognosis.[1]Ki-67 is a nuclear antigen, which exists in proliferative cells. Numerous studies have shown that the immune response of Ki-67 is closely associated with the cell cycle. Also Ki-67 may predict the pathological remission rate in breast cancer patients. Other key biological markers in primary breast cancer are tumor size, axillary lymph node involvement, nuclear grade, progesterone receptor, and HER2 status. Uncontrolled proliferation (such as Ki- 67) is an important characteristic of malignant tumors so, tumor proliferation is one of the major factors associated with prognosis.[2,3] The aim of this study was to evaluate the use and value of Ki-67 as a prognostic marker in breast cancer and associations between Ki-67, clinical, and histopathological parameters were estimated.

MATERIALS AND METHOD:

This study included 50 patients with primary breast cancer. The data of this analytical–descriptive study were obtained from patients documents in the college. The levels of ER, PR, Ki-67, LN status, and the tumor grades and sizes were determined after diagnosis. Pathologist re-evaluated all smearing stain of Ki-67 by IHC, and the exact levels of Ki-67 were determined. The grade of tumors was confirmed by an expert pathologist, and the lymph node status was confirmed clinically using imaging techniques after surgery.

IHC ANALYSIS IHC staining for ER, PR, and Ki-67 was performed in all cases. The sample sections were de-paraffinized in alcohol and xylene and then heated in EDTA buffer solution (PH=9) to 100◦C. After cooling for about 15minutes, the samples were rinsed in tris buffer solution (PH = 7.6) for 5 minutes. Endogenous peroxidase was quenched with 3% hydrogen peroxidase in methanol toblock nonspecific binding for 10 minutes. The slides were then incubated for 30 minutes with primary antibodies. The primary antibodies used for estrogen receptor was ER, Dako, clone ID 5, for progesterone receptor was PR, Dako, clone PgR636, and for Ki-67 was Dako, clone MIB-1. The tumor grade was reported based on the H&E (hematoxylin and eosin) smears, which were considered in 3 parameters: the number of mitoses, the nuclear polymorphism rates, and the gland formation structures. If the total score of them were 3 - 5, 6 -7, and 8 - 9, we considered the grades as 1, 2, and 3 respectively. The ER, PR, and Ki-67 status were defined based on the intensity and the percentage of nuclear stain. Negative Ki-67 was defined as less than 1% stain, and positive Ki-67 was greater than 1%. Patients with positive Ki- 67 were divided into 3 groups: 1-5%, 6-14% and 15% of Ki-67staining.

STATISTICAL ANALYSIS We used the Mann-Whitney U test to determine the correlation between Ki-67 and ER and PR expression because the goal was to evaluate the relationship between an ordinal variant Ki-67 and two unpaired sample variants, ER and PR. To determine the relationship between patient's age, tumor size and grade, and LN invasion in patients with primary BC, Spearman's rho and the Kendall rank correlation coefficient tests were used. SPSS 21 Statistical Analysis Software (SPSS Inc., Chicago, IL.) was used to perform all statistical analysis. Written consent forms were provided to all patients enrolled in the study, and all patients' data and information were confidential.

RESULTS:

Graph 1: Levels of Ki67 biomarker as IHC staining in 50 breast cancer patients

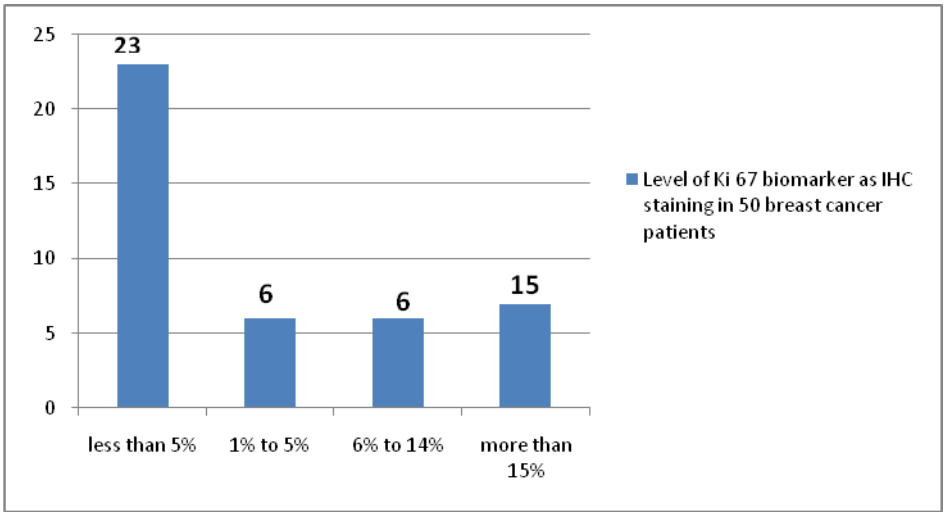


Table 1: The results of Ki-67 association with clinicopathological aspects in 50 breast cancer patients.

Variables		Number	Spearman rho	Tau kendall
Age	<50 >50	21 29	0.121	0.311
Tumor size (cm)	≤2 2 to 5 >5	14 30 6	0.498	0.534
Grade	I II III	15 16 19	0.564	0.458
Lymph node invasion	No 1-3 >3	15 6 29	0.081	0.086

Figure 1: Ki-67 expression in Grade I invasive ductal carcinoma.

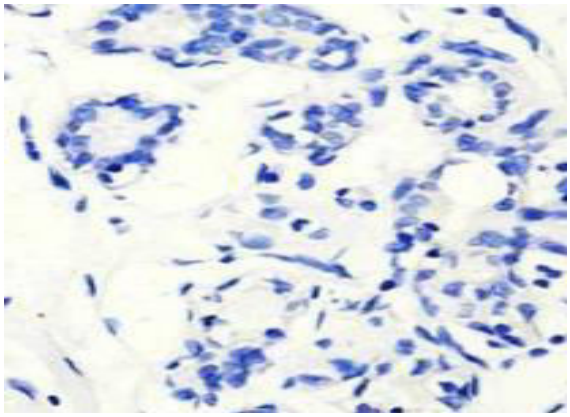


Figure 2: Ki-67 expression in Grade II invasive ductal carcinoma.

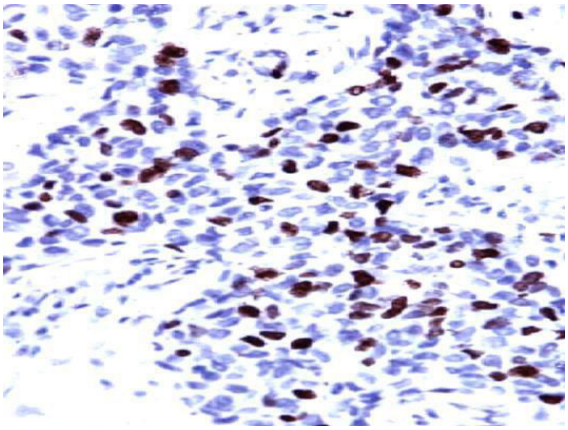
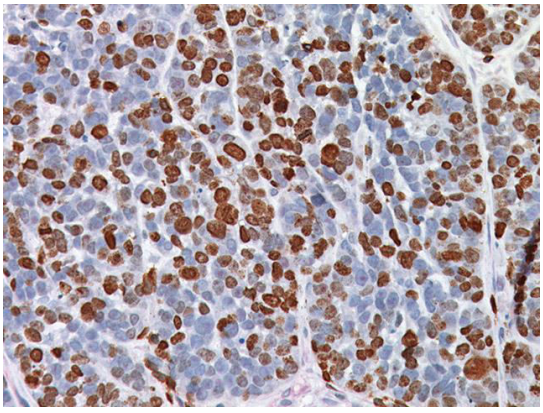


Figure 3: Ki-67 expression in Grade III invasive ductal carcinoma.



DISCUSSION:

In present study we observed that 29 cases were Ki-67 positive with more than 1% tumor nuclei stained, and 15cases had tumors with more than 15% of Ki-67 expression. There was an insignificant relationship between Ki-67 and the age of patient as well as the size and grade of tumors. Though, a significant relationship was seen in between the lymph node status and Ki-67expression.There was a significant relationship between estrogen receptor (ER) and progesterone receptor (PR) with Ki-67 status, a dependable estimation of prognostic factors in breast cancer patients is required to select optimal therapeutic strategy. Now a days, data has shown that the Ki-67 labelling index is an independent prognostic factor for the survival and recurrence of tumors. These investigations examined more than 4600 cases and proved that Ki- 67 labelling index is a significant prognostic factor.[4] De Azambuja et al in 2007 retrieved the disease free survival (DFS) data from 46 studies and confirmed that high Ki-67 levels conferred a worse prognosis in the studied cohorts.5 A number of adjuvant trials did not support the predictive role for the benefit of applying chemotherapy over endocrine treatment alone in patients with high tumor Ki-67 expression.[6] However, it is well-documented that higher levels of the proliferation marker Ki-67 are significantly associated with poor survival, high relapse and mortality rate.[7,8] A few studies revealed a significant association between the pre-therapy Ki-67 and histological grade of tumors and an inverse association between Ki-67 with ER status.[9]

A significant relationship was reported for Ki-67 and other tumor markers (ER, PR), which showed that increased Ki-67 levels were correlated with increased tumor grades.[9,10] However, in a review by Yerushalmi (2010), it was suggested that further studies are required before any recommendations can be made about using the relationship of tumor grade and Ki-67.6 In our study, however, no significant correlation was observed between tumor grade and Ki-67 levels (The Spearman rho = 0.564, tau Kendall = 0.458). In a survey by Altintas (2009), the correlation between Ki-67and other biologic markers used and it was found that highly proliferative lesions were more likely to be ER negative and PR negative.[11] In another study, there was a significant negative relationship between Ki-67 levels and the expression of estrogen and progesterone receptors. Additionally, Bouzubar (1989) suggested that, conversely, although the Ki-67 status of breast tumors and their percentage are not correlated with the ER status of breast cancer, ER positive tumors contain a slightly higher proportion of Ki-67 negative cells then ER negative tumors we observed a significant correlation between Ki-67 and ER (p=0.05) and a marginally significant correlation with PR (P = 0.07) by Mann-Whitney U test. Our findings showed that there was a negative correlation between Ki-67and ER and PR. In another study, a significant correlation was observed between the median Ki-67 staining and patient age and tumor nuclear grade. Tumors from patients younger than 50 years showed a higher level of Ki- 67 than those of older patients.[13] Other than that, none of similar studies has shown significant correlation between Ki-67 and patient's age, which is consistent with our result.[14,15] The status of the LN remains to be the most important determinant of the overall survival, and node negative BC patients have a favorable prognosis.[16-18] Molino et al. found that there was a positive relationship with nodal status, as node negative tumors are more likely to have a low proliferation index.[14] A correlation between the histological grade of malignancy of breast tumors and

their Ki-67 status was previously reported and no significant association was observed between tumor size, LN status, patients age, ER, and Ki-67 status. Although large tumors often contained an increased number of Ki-67 positive cells (up to 20%) [12], some events showed a positive association between Ki-67 staining and tumor size, in which, the smaller tumors had lower Ki-67 values, and the larger tumors (> 2 cm) were associated with poorer prognosis.[14,16]

REFERENCES:

1. Kermani T A, Kermani I A, Faham Z, Dolatkhah R. Ki-67 status in patients with primary breast cancer and its relationship with other prognostic factors. Biomed. Res. Ther.; 6(2):2986-2991.
2. Colozza M, Azambuja E, Cardoso F, Sotiriou C, LarsimontD, Piccart MJ. Proliferative markers as prognostic and predictive tools in early breast cancer: Where are we now? Ann Oncol 2005;16:1723-39.
3. Stuart-Harris R, Caldas C, Pinder SE, Pharoah P. Proliferation markers and survival in early breast cancer: A systematic review and meta-analysis of 85 studies in 32,825 patients. Breast 2008;17:323-34.
4. Scholzen T, Gerdes J. The Ki-67 protein: from the known and the unknown.J Cell Physiol.2000;182(3):311-22.
5. de Azambuja E, Cardoso F, Jr GdC, Colozza M, Mano MS, Durbecq V, et al. Ki-67 as prognostic marker in early breast cancer: a meta-analysis of published studies involving 12,155 patients. Br J Cancer. 2007;96(10):1504–13.
6. Yerushalmi R, Woods R, Ravdin PM, Hayes MM, Gelmon KA. Ki67 in breast cancer: prognostic and predictive potential. Lancet Oncol. 2010;11(2):174–83.
7. Liu Y, Zhang X, Yu F, Liu J, Zhang M, Zhang S, et al. [Prognostic significance of Ki-67 expression before and after neoadjuvant neoadjuvant chemotherapy in different biological breast cancer phenotypes]. Zhonghua Zhong Liu Za Zhi. 2014;36(9):671–6.
8. Matsubara N, Mukai H, Fujii S, Wada N. Different prognostic significance of Ki-67 change between pre- and postneoadjuvant chemotherapy in various subtypes of breast cancer. Breast Cancer Res Treat. 2013;137(1):203–12.
9. Petrelli F, Viale G, Cabiddu M, Barni S. Prognostic value of different cut-off levels of Ki-67 in breast cancer: a systematic review and meta-analysis of 64,196 patients. Breast Cancer Res Treat. 2015;153(3):477–91.
10. Chen Q, Wu K. Research progress on the clinical value of Ki-67 in breast cancer and its cut-off definition. Zhonghua Wai Ke Za Zhi. 2015;53(8):634–7.
11. Altintas S, Lambein K, Huizing MT, Braems G, Asjoe FT, Hellemans H, et al. Prognostic significance of oncogenic markers in ductal carcinoma in situ of the breast: a clinicopathologic study. Breast J. 2009;15(2):120–32.
12. Bouzubar N, Walker KJ, Griffiths K, Ellis IO, Elston CW, Robertson JF, et al. Ki67 immunostaining in primary breast cancer: pathological and clinical associations. Br J Cancer.1989;59(6):943–7.
13. Sahin AA, Ro J, Ro JY, Blick MB, el Naggar AK, OrdonezNG, et al. Ki-67 immunostaining in no denegative stage I/II breast carcinoma. Significant correlation with prognosis. Cancer. 1991;68(3):549–57.
14. Molino A, Micciolo R, Turazza M, Bonetti F, Piubello Q,Bonetti A, et al. Ki-67 immunostaining in 322 primary breast cancers: associations with clinical and pathological variables and prognosis. Int J Cancer. 1997;74(4):433–7.
15. Kim EK, Noh WC, Han W, Noh DY. Prognostic significance of young age (<35 years) by subtype based on ER, PR, and HER2 status in breast cancer: a nationwideregistry-based study. World J Surg. 2011;35(6):1244–53.
16. Kuru B, Camlibel M, Gulcelik MA, Alagol H. Prognostic factors affecting survival and disease-free survival in lymph nodenegative breast carcinomas. J Surg Oncol. 2003;83(3):167–72.
17. Bonneau C, Bendifallah S, Reyal F, Rossi L, Rouzier R. Association of the number of sentinel lymph nodes harvested with survival in breast cancer; 2015.
18. Yang J, Long Q, Li H, Lv Q, Tan Q, Yang X. The value of positive lymph nodes ratio combined with negative lymph node count in prediction of breast cancer survival. J Thorac Dis.2017;9(6):1531–7.