

Original Research Article

Role of HER-2/Neu Expression in Premalignant and Malignant Lesions of Uterine Cervix

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Abstract

Background: HER-2/neu is a gene localized on chromosome 17q21 which encodes a growth factor receptor like molecule with tyrosine kinase activity and is known to encode a transmembrane glycoprotein. *Aim:* To study the expression of HER2/neu in benign and malignant lesions of cervix. *Material and Methods:* The present study was conducted on 100 cases of resected cervical tissue specimen suspected to have neoplastic lesions and hysterectomy specimen already diagnosed as carcinoma on cytology for a period of 12 months (September 2018 to August 2019). *Results:* Majority of benign cases were negative for HER-2/neu except few cases and positivity was higher in malignant than premalignant. Positivity was also higher in adenocarcinoma as compared to squamous cell carcinoma. *Conclusion:* HER-2/neu serves as a biological marker to identify high risk subgroups and can be useful as target for new treatment modalities.

Keywords: Her-2 /neu; Epithelial Cells; Cervical Cancer.

Introduction

Cervical cancer, a preventable disease, is the second most common malignancy in women worldwide and major cause of morbidity and mortality particularly in developing countries where it accounts for 15% of all new female cancers [1]. It is the most common cancer of female genital tract in India [2]. Cervical cancer is third largest cause of cancer mortality in India after cancer of oral cavity and esophagus, accounting for nearly 10% of all cancer related deaths in the country [3]. Although the introduction of papanicolaou test in developed countries has been effective in reducing cervical cancer mortality and morbidity rates, the efficacy of the Pap test is hampered by high

interobserver variability and high false negative and false positive rates that range between 20-30% [4] and 5-70% [5] respectively. Technical improvements of the pap test such as the Liquid Based Cytology (LBC) have not been shown to improve sensitivity or specificity for detection of high grade cervical intraepithelial neoplasm (CIN) compared to conventional cytology [6]. Histopathology determines treatment of malignant and premalignant through classifying into a diagnosis the patterns of microscopic organization of cells in tissue sections from biopsy or surgical specimens. Immunohistochemical studies allow the identification of the molecular changes as tumor marker and contributed to improve our capacity in diagnosis and evaluation of progression.

HER-2/neu is known to encode a transmembrane glycoprotein with tyrosine kinase activity and shows a extensive homology to the human epidermal growth factor receptor i.e. ERBB2 protein [7]. In most cases of benign lesions, HER-2/neu is negative. Also over expression rate was significantly higher in malignant cases as compared to premalignant cases. The over expression of HER-2 has been found to be related to aggressiveness of disease, increased mortality and higher relapse ratio [8].

Material and Methods

The present study was conducted on 100 cases of cervical lesions. The study was carried out to find out the histological type of lesion along with the expression of HER-2/neu for a period of one year. Inclusion criteria for histopathological examination are: 1. clinically suspected precancerous and cancerous lesions of cervix of all ages. 2. patients who agreed to sign on consent form. The tissues were processed for routine histopathological examination and immunohistochemical stained with HER-2/neu antibody. Standard streptavidin-biotin peroxidase method of immunohistochemical staining was followed. Positive staining was normally expressed in the membrane and cytoplasm of epithelial cells as golden brown reaction and was considered negative when it was completely unstained or staining positively in <5% of cells. The intensity of expression was graded according to the 2014 ASCO/CAP guidelines for HER-2/neu reporting.

The 2014 ASCO/CAP HER-2/neu reporting guidelines

Intensity of HER-2 Expression	Characteristic Features
0	No staining is observed or shows membrane staining that is incomplete and is faint perceptible and within ≤10% of tumor cells
1+	Membrane staining that is incomplete and is faint perceptible and within >10% of tumor cells

2+	Circumferential staining that is incomplete and/or weak/moderate within >10% of tumor cells.
3+	Complete intense circumferential membrane staining within >10% of tumor cells

Result

Out of 100 cases of cervical lesion, 58% were of benign lesions, 12% of cervical intraepithelial neoplasia where as 30% were invasive carcinoma. The age group of total 100 cases ranged between 21-80 yrs. Maximum number of cases presented above the age of 30 yrs. Maximum number of CIN cases were seen in age group of 41-50 yrs with CIN-II as the predominant grade and was mainly of squamous cell origin.

Among invasive carcinoma cases, majority of invasive carcinoma cervix were observed in sixth decade (40.00%) followed by 26.66% in more than 60 yrs. The study of parity wise distribution of these patients showed that highest incidence of cervical intraepithelial lesions and invasive carcinomas were noted in women who had 3 or more children. No case of invasive carcinoma was seen in single parity. Most of the cases 92 (92%) were Hindus and rest 08 (08%) were Muslims. Cases from rural areas predominated over those from urban areas with a rural urban ratio of 1.43:1. Among benign group, the most common presenting symptoms were white discharge per vagina (74.13%). The commonest presenting symptoms in low grade CIN was white discharge per vagina (50%).

Punch biopsy (74%) was the most common surgical specimen received in pathology department followed by simple hysterectomy (12%) and core biopsy (11%). Among benign cases, chronic non specific cervicitis (90.93%) was the most common lesion followed by acute cervicitis. Out of 30 cases of invasive carcinoma, squamous cell carcinoma was the commonest histological type (73.33%) and most of them were moderately differentiated (63.63%).

On analyzing the expression of HER-2/neu, overall HER-2/neu positivity was observed in 31% of cases. Majority of benign cases (93.11%) were negative for HER-2/neu expression except

Table 1: Expression of HER-2/Neu in Benign, Preinvasive and Invasive Carcinoma of Cervix

Cases	Total no. of cases	Her-2/neu positivity	Percentage	Chi- square = 37.8
Benign	58	4	6.89	p-value < 0.001
CIN(Preinvasive)	12	7	58.33	
Malignant(invasive)	30	20	66.66	

four cases (6.89%) which shows faint positivity only. Stroma was negative in all cases. HER-2/neu expression was observed in 58.33% of total CIN cases. The difference of expression of HER-2/neu between benign and CIN was found to statistically highly significant. ($p < 0.001$). Among invasive carcinoma group, 66.66% of cases showed HER-2/neu oncoprotein expression (Table 1).

These malignancies exhibited variable degree of expression of HER-2/neu oncoprotein with higher

expression in high grade carcinoma. In the present study, over expression rate was significantly higher ($p < 0.001$) in malignant cases (66.66%) as compared to premalignant cases (58.33%). Higher HER-2/neu positivity was observed in adenocarcinoma (85.71%) as compared to squamous cell carcinoma (59.09%). However association was not significant statistically. ($p\text{-value} = 0.301$). Overall stronger positivity was observed in high grade SIL

Table 2: Intensity of Her-2/Neu Staining in Premalignant Cases (CIN)

Intensity	CIN-I No. (%)	CIN-II No. (%)	CIN-III No. (%)	Chi-square=1.23 p-value=0.539
Negative	01(25)	02(40)	02(66.67)	
1+	03(75)	02(40)	0	
2+	0	01(20)	01(33.33)	
3+	0	0	0	

Table 3: Her-2/Neu Expression in Various Grades of Squamous Cell Carcinoma of Cervix.

Intensity	1+	2+	3+
Well differentiated(1)	1(7.69%)	0	0
Moderately differentiated(9)	2(15.38%)	5(38.48%)	2(15.38%)
Poorly differentiated(3)	0	2(15.38%)	1(7.69%)

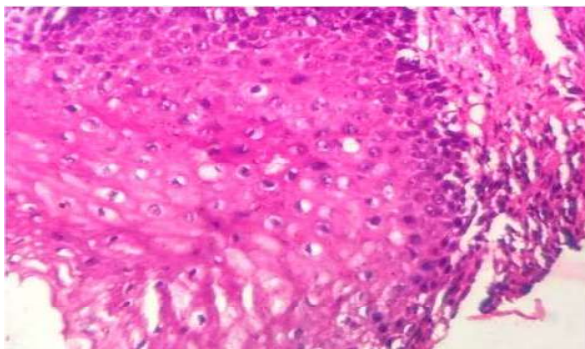


Fig. 1: Microphotograph of CIN II showing atypia and hyperchromatic nuclei in 2/3rd layer of cells of cervical squamous epithelium (H&E X 100)

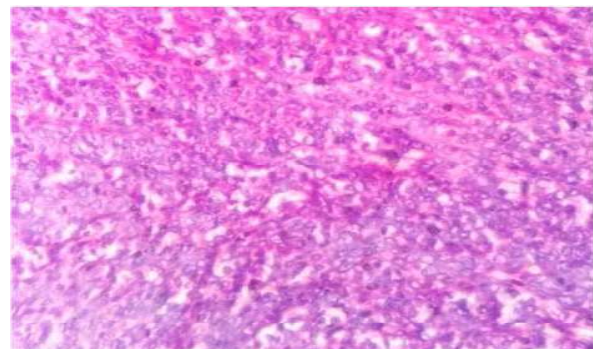


Fig. 3: Microphotograph of poorly differentiated squamous cell carcinoma of cervix (H&E stain X 100)

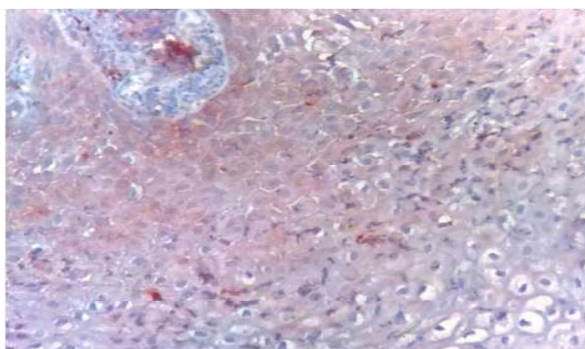


Fig. 2: Microphotograph of CIN II showing strong diffuse positivity of cytoplasm & cytoplasmic membrane of HER-2/ neu (Her2/ neu immunostain X100)

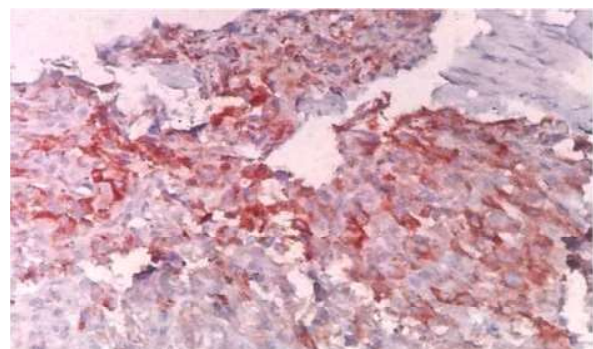


Fig. 4: Microphotograph of Poorly differentiated squamous cell carcinoma showing strong HER-2/ neu immuno positivity (Her-2/ neu immunostain X 100)

(CIN-II and CIN-III), but HER-2/neu scores did not show significant association with CIN grade. (p-value=0.539) (Table 2).

In the present study, HER-2/neu positivity rate showed an incremental trend from well differentiated (20%) to poorly differentiated (100%) squamous cell carcinoma (Table 3).

However the association was not considerable statistically. Among adenocarcinoma cases, reaction intensity was higher in moderately differentiated adenocarcinoma as compared to well differentiated adenocarcinoma. There was no case of poorly differentiated adenocarcinoma in the present study. In the present study, HER-2/neu positivity rate was 62.5% in FIGO stage 2 tumor, while stage 3 and stage 3 and stage 4 tumor shows 100% positivity. A statistically significant association was established between positive HER-2/neu expression and higher clinical stage of presentation. A significant correlation was also observed between lymph node positive cases and expression of HER-2/neu oncoprotein. (p value= 0.037) However association between intensity of staining and presence of parametrial extension was not found to be statistically significant. (p value= 0.389). The over expression is associated with poor prognosis, metastatic potential and aggressive biological behavior. The sensitivity and specificity of HER-2/neu expression detecting preinvasive lesion were 58.33% and 93.10% respectively with positive predictive value and negative predictive value 63.63% and 91.52% respectively.

The sensitivity and specificity of HER-2/neu staining in detecting invasive carcinoma were 66.66% and 93.10% respectively with positive predictive value and negative predictive value 83.33% and 84.37% respectively.

Discussion

In the present study out of total 100 cases, benign lesions (58%) were the commonest lesion of the cervix, 12 cases (12%) were of cervical intraepithelial neoplasia and 30 cases (30%) were malignant. Our results correlate well with the study of Kumari K et al. (2017) [9]. Maximum number of cervical intraepithelial neoplasia cases (50%) were observed in the age group of 41-50 years with CIN-II (41.6%) as the predominant grade. This was in accordance with the study of Poste P et al. (2015) [10]. Among invasive carcinoma, squamous cell carcinoma was the most common histological type accounting for 73.33% of total

invasive carcinoma followed by adenocarcinoma (23.33%) and adenosquamous carcinoma (3.34%). Our results are comparable with the study done by Gupta N et al. (2009) [11]. Expression of HER-2/neu in various lesions of cervix was seen in 31% of cases and rest 69% did not show any positivity for HER-2/neu. This was accordance to reported by Bajpai S et al. (2017) [12]. Out of total 12 CIN cases, HER-2/neu positivity was 58.33%. Overall stronger positivity was observed in CIN-II and CIN-III cases. Our findings are in accordance with study of Gupta N et al. (2009) [11]. HER-2/neu expression was significantly higher in malignant lesions as compared to premalignant lesions (p< 0.001). Our study is in agreement with the findings of Sarvade P et al. (2016) [13]. HER-2/neu positivity rate showed an incremental trend from well differentiated (20%) to poorly differentiated (100%) squamous cell carcinoma lesion. This is accordance with Mandai et al (1995) [14]; Joseph T et al. (2015) [15] and Ray et al. (2002) [16].

Conclusion

HER-2/neu expression level in the premalignant lesion appears to be a sensitive factor in predicting the neoplastic potential of dysplastic tissue. This suggest that HER-2/neu may serve as a biological marker to identify high risk subgroups and can be useful as target for new treatment modalities. The biology of HER-2/neu in cervical cancer must be studied further to understand how to predict response to targeted treatments and how to match individual patients characteristic to appropriate HER-2/neu inhibitor.

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