Clinicopathological Study of HER2/neu expression in Gastric Cancer

Mellonie P*[1], Anto J Richie[2]

[1]*Department of Pathology, G R Medical College, Mangalore, India*

[2]*Department of Radiology, Father Muller Medical College, Mangalore, India*

ABSTRACT

Introduction: Gastric cancer the fourth leading cause of cancer-related death. Overexpression of HER2 is seen in a significant number of gastric cancer cases leading to the development of targeted therapy with anti-HER2 antibodies like Transtuzumab. However, uncertainty persists regarding the association between the clinicopathological features of gastric cancer patients and HER2 overexpression.

Aim: This study aims to find the association between clinicopathological features of gastric adenocarcinoma and HER2 overexpression.

Materials and Methods: This descriptive observational study was conducted in the Department of Pathology of a tertiary care hospital laboratory in Mangalore from January 2018 to April 2021. In this study we analysed 75 consecutive specimens of gastric/gastroesophageal junction tumours. Histological types other than gastric adenocarcinoma were excluded from the study. Diagnosis was confirmed using hematoxylin and eosin (H&E) slides. IHC was done using mouse monoclonal antibody HER2 from Biogenex. Scoring for HER2 was done by Hoffmann scoring system. Tumours were classified according to Lauren's histological classification into Intestinal and diffuse type .Other relevant details like patient's age, gender, type of specimen, tumour location, histological type and tumour grade were collected and tabulated.

Results: Out of the 75 cases of adenocarcinomas analysed in this study 48 were biopsy specimens and 27 were gastrectomy specimens. In our study the rate of overall Her2 overexpression was 25.3%. Also, Her2 overexpression was associated with Intestinal type (26.9%), GEJ Tumours, moderately differentiated carcinomas and male gender .There was no correlation of HER 2 positivity with age. (30.6%, p = 0.006).

Conclusion: Since, status of HER2 expression by IHC plays pivotal role in the treatment of gastric cancer, testing for the same has increased in the recent times. Hence knowing the association of HER2 expression with clinicopathological features has a clear advantage.

Key words: Gastric cancer, HER2/neu, Prognosis, Transtuzumab.

Corresponding author: Mellonie P

e-mail✉: drmellonierichie@gmail.com

INTRODUCTION

Gastric cancer is one of the most common malignancies worldwide and is the fourth leading cause of cancer-related death [1]. Human epidermal growth factor receptor (HER2) protein overexpression plays an important role in tumour proliferation. The frequency of HER2 overexpression in gastric cancer varies with different geographical regions and ranges from 13% to 91% [2]. Currently anti-HER2 monoclonal antibody such as Transtuzumab is being used to treat gastric cancer patients with HER2 overexpression [3].Combination chemotherapy along with Transtuzumab, has increased the overall survival in Her2-positive, locally advanced gastric cancer [4, 5]. HER2 overexpression plays a key role in the treatment of gastric adenocarcinoma; hence it is important to study in detail about the HER2 overexpression by IHC and its association with clinicopathological features of gastric cancer [6].

AIM

To study the expression of HER2 in Gastric adenocarcinomas and to correlate it with clinico-pathological features like age, sex, location of tumour, histological type and tumour grade.

Method: This study was conducted in the Department of Pathology of a tertiary care hospital laboratory in Mangalore from January 2018 to April 2021. In this study we analysed 75 consecutive cases of gastric cancer cases excluding histological types other than gastric adenocarcinoma. The Haematoxylin and Eosin (H&E) stained slides were studied and tumours were classified according to Lauren's classification into Intestinal and Diffuse type [7]. Other relevant details like patient's age, gender, type of specimen, tumour location and tumour grade were collected and tabulated. Immunohistochemical Staining wasdone on poly-l lysine coated slides with 4 μm thick paraffin embedded sections. Tri sodium citrate buffer at pH-6 was used for antigen retrieval. Mouse monoclonal antibody HER2 from Biogenex, was used for HER2 antigen detection. HER2 scoringwas done using the Hofmann scoring system [8, 9]. All cases with score 3+ were considered positive.

Statistical Analysis

Study design-descriptive observational study

Sample size calculator-Confidence level-95%, Confidence interval 5, population- 100 patients with gastriccancer,Samplesizeneeded60 patients.

Formula used n = N/1+N*e2

Inclusion criteria- patients diagnosed with gastric adenocarcinoma in gastric biopsy or gastrectomy specimen from January 2018 to April 2021.

Exclusion criteria- Patients with gastric cancer who have undergone prior chemo/radiotherapy were excluded from the study.

Descriptive statistics was applied, chi-square test was used to analyse association between Her2 status and clinicopathological parameters.

P-value<0.05 was considered significant. The SPSS software, version 20.0 was used for data analysis.

RESULTS

Out of the 75 cases of adenocarcinomas analysed in this study 48 were biopsy specimens and 27 were gastrectomy specimens. Age of patients ranged from 33-87years. Majority of patients were in the age group of 61-70 years. Mean age of the patients was 65.65 years with males accounting to 59%. Rate of overall Her 2 overexpression was 25.3%. In our study HER2 over expression was seen more in gastric biopsies (37.5%) as compared to resected specimens (14.8 %). Also, Her 2-overexpression showed association with male gender (30.6%), Intestinal type (26.9%), Gastroesophageal Junction (GEJ) Tumours (45.5%) and moderately differentiated carcinomas (32.6%) [Table 1-4] [Figure 1, 2].

Table 1: Gender distribution *vs* Her2 neu expression.

Gender	Number of cases	Her2neu expression			
		0	1+	2+	3+
Male	44(58.6%)	29(65.9%)	1(2.3%)	3(6.8%)	11(30.6%)
Female	31(41.3%)	12(38.7%)	8(25.8%)	3(9.6%)	8(25.8%)
Total cases	75	41(54.6%)	9(12%)	6(8%)	19(25.3%)

Table 2: Specimen type distribution vs Her2neu expression.

Her2neu expression	Gastric biopsies	Gastrectomies	Total number of cases
0	25(52%)	21(77.7%)	41(54.6%)
1+	4(8.3%)	1(3.7%)	9(12%)
2+	3(6.3%)	1(3.7%)	6(8%)
3+	18(37.5%)	4(14.8%)	19(25.3%)
Total	48(100%)	27(100%)	75(100%)

Table 3: Correlation of Her2neu with Lauren's classification.

Lauren's classification	Total number of cases	Her2neu			
		0	1+	2+	3+
Diffuse	12(84%)	9(75%)	0	1(8.3%)	2(16.6%)
Intestinal	63(16%)	32(50.7%)	9(14.2%)	5(7.9%)	17(26.9%)
Total	75	41(54.6%)	9(12%)	6(8%)	19(25.3%)

Table 4- Correlation of Histological grade with Her 2 neu expression.

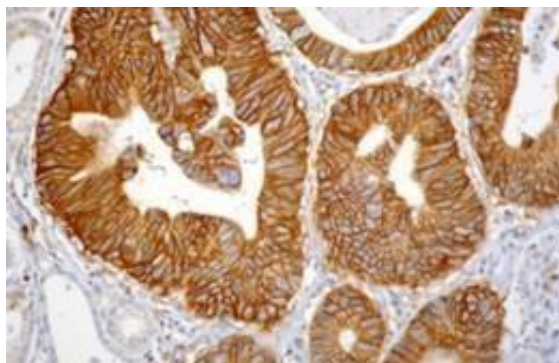| Grade	Total number of cases	Her2neu expression			
		0	**1+**	**2+**	**3+**
Well differentiated	11(14.6%)	9(81.8%)	1(9%)	0	1(9%)
Moderately differentiated	49(65.3%)	21(42.8%)	7(14.2%)	5(10.2%)	16(32.6%)
Poorly differentiated	15(20%)	11(73.3%)	1(6.6%)	1(6.6%)	2(13.3%)
Total cases	75	41(54.6%)	9(12%)	6(8%)	19(25.3%)

Figure 1: Moderately differentiated adenocarcinoma showing strong membranous positivity, Her2 score 3+.
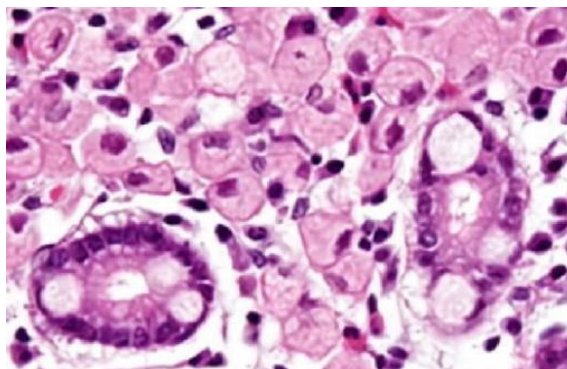

Figure 2: Signet ring cell carcinoma showing no Her2 staining, Her2 score 0.

DISCUSSION

Gastric cancer is one of the most common malignancies worldwide and is the fourth leading cause of cancer-related death. In India, gastric cancer ranks fifth among males and seventh among females. The etiopathogenesis of Gastric cancer is multifactorial and includes smoking, alcohol, dietary factors like high consumption of salty fish containing N-nitrosamines, family history and infections like Helicobacter pylori and Epstein Barr Virus. Improved food preservation techniques have led to a decrease in the incidence of gastric cancer over the years [10]. Recently, Human epidermal growth factor receptor 2 (HER2) proteins, has evolved as one of the main receptor for targeted therapy [11]. Results published from phase III ToGA trial provided first evidence towards the use of trastuzumab in combination with chemotherapy for advanced gastric cancer. There was clear evidence of increased overall survival of 13.8 months in trastuzumab arm as compared to 11.1 months in the chemotherapy only arm. As a result of this testing for HER2 in gastric cancer has sky rocketed to determine patient eligibility for targeted therapy [12]. Many studies have been done in recent times to study the association of clinicopathological features of gastric cancer with HER2 over expression.

In our study, HER2 overexpression was observed in 25.3% of cases, which is similar to study done which reported HER2 positivity in 26.7% of cases [13]. It is also similar to TOGA trial in which 22.1% cases of gastric cancer were HER2 positive. In our study Her2 over expression was seen more in gastric biopsy specimens (37.5%), as compared to resected specimens (14.8 %). This is similar to TOGA trial which also had a lower HER 2 positivity rate in resected samples

compared to biopsies [14]. This may be because of larger sample size of biopsies (n=48) when compared to gastrectomy (n=27). Another reason may be because of better fixation of biopsy specimens as suggested by Ruschoff J, et al [15]. In our study Her 2 overexpression was seen more in Intestinal type (26.9%, p = 0.051), when compared to diffuse type (16.6%). Similar observation was made by Koltz, et al in which the incidence of HER2 overexpression in gastric adenocarcinoma was reported to be 34% in intestinal type and 7% diffuse type [16]. The selective overexpression of HER2 in intestinal type of gastric cancer suggests that there is difference in tumorigenesis between the 2 histological types at a molecular level. Also, E-cadherin mutation was typically seen in diffuse type.

We did not find statistically significant correlation of HER2 positivity with age. This was similar to study done by Ling Shan, et al [17]. We found a statistically significant correlation of HER2 with male gender (30.6%, p = 0.006). This is similar to study done by Lei YY, et al [18]. In our study Her2 overexpression was seen more in gastroesophageal junction tumours (45.5%) as compared to primary gastric tumours (22.4%). This was in concordance with study done by Tafe LJ, et al [19]. We also observed higher rates of HER2 positivity (32.6%) in moderately differentiated carcinomas when compared to well and poorly differentiate cancers. This may be because our study group consisted of 66.7% of moderately differentiated carcinomas and hence the frequency of HER2 positivity was also more. However, demonstrated increasing HER2 expression in moderately differentiated Tumours [20]. To further evaluate moderate differentiation as an independent risk factor, a multicenter study including cases from each category (well, moderate and poorly differentiated) would be necessary. One of the drawbacks of our study is that we were not able to confirm IHC results by FISH due to budget constraints. However, many studies have shown very good concordance (95%) between IHC and FISH for HER2 score 0 and 3+. Some of the challenges we faced while evaluating IHC were the crush and edge artifacts especially in small biopsies [21].

CONCLUSION

Our study found an HER2 overexpression of 22.4% in gastric cancers which is similar to most of the studies in India. We found a statistically significant correlation of HER2 overexpression with male gender, intestinal-type and moderately differentiated gastric cancers. These clinicopathological features may be kept in mind while evaluating patients as they help in finding the right candidates to benefit from targeted therapy.

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