Study of HER2 expression and its relation to tumor characteristics among gastric adenocarcinoma patients of Firoozgar hospital, Tehran, Iran; in 2010 and 2011

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Abstract

Background: One million people are diagnosed with gastric cancer (GC) annually and the prognosis for patients with advanced GC or gastroesophageal junction (GEJ) cancer is usually poor with a median survival time of 8 to 10 months. Recent reports have identified that anti HER2 target therapy improved the median survival rate in patients with HER2 positive GC; but a wide range of HER2 overexpression incidence has been reported in different studies. This study was conducted to determine the incidence of HER2 overexpression and its relationship to tumor characteristics.

Methods: All gastric or GEJ adenocarcinoma patients who underwent a curative surgical resection in Firoozgar Hospital, Tehran, Iran between 2010 and 2011 were entered into this cross-sectional study. The formalin-fixed and paraffin wax-embedded tissue blocks used in the present study were surgical resection specimens obtained from 115 patients. All tumor samples were examined for HER2 expression by an immunohistochemistry (IHC) assay and HER2 IHC score 3+ was considered positive.

Results: The study sample consisted of 115 subjects, 81 males and 34 females with a mean age of 61.01 years (30-85 years, SD=11.605). HER2 overexpression was detected in 13 (11.3%) of the patients. HER2 overexpression was significantly higher in tumors ≥5cm compared to those <5cm (P=0.01). But, there was no significant relationship between tumor histological subtype, GC stage or tumor location and HER2 overexpression (P=0.607, 0.745, 0.491).

Conclusion: The findings of the present study indicated that the incidence of HER2 overexpression in gastric adenocarcinoma was 11.3%. The higher incidence of HER2 overexpression in patients with tumors ≥5cm maybe indicate that anti HER2 target therapy administration for these patients could be more beneficial.

Keywords: HER2, Gastric adenocarcinoma, Gastroesophageal junction adenocarcinoma

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Introduction

About one million people are diagnosed with gastric cancer (GC) annually all around the world, and both gastric and gastroesophageal junction (GEJ) adenocarcinomas are major causes of cancer morbidity and mortality (1,2). Additionally, subsequent locoregional and distant recurrence of GC after complete surgical tumor resection is frequent; moreover, the prognosis of patients with advanced GC or GEJ cancer is poor with a median survival time of 8 to 10 months (2,3). Actually, prognosis and survival vary among patients at the same stages of GC or GEJ cancer; therefore, it is important to detect subgroups that are more responsive and sensitive to new therapeutic strategies (4).

Trastuzumab is the main treatment for human epidermal growth factor receptor 2 (HER2) positive breast cancer patients (5-7). Furthermore recent studies have reported an

improved median survival rate and reduced the risk of death by adding trastuzumab to the standard chemotherapy treatment of HER2 positive advanced GEJ or gastric adenocarcinomas patients (8-10).

The HER2 gene is located on the human chromosome 17q21 and it encodes a transmembrane glycoprotein receptor that has partial homology with the epidermal growth factor receptor. In addition, HER2 receptor dimmers autophosphorylation to activate the intracellular signaling pathways, which could cause cell proliferation, differentiation, migration and oncogenesis(11,12).

Reports from various studies have shown a wide range in the incidence of HER2 overexpression (7% to 53.4%) and some studies reported higher frequencies in the intestinal subtype of gastric adenocarcinoma (13-16). This heterogeneity could be explained by numerous factors such as differences in the study population, use of various assays for detection of genetic amplification, as well as different scoring criteria and degrees of resection.

The purpose of this study was to evaluate the frequency of HER2 overexpression in gastric adenocarcinoma, using immunohistochemistry (IHC) assay and determining its relationship with different tumor characteristics

Materials & Methods

All primary gastric or GEJ cancer patients who underwent a curative surgical procedure in Firoozgar Hospital, Tehran, Iran, between 2010 and 2011, were included in this cross-sectional study. The formalin-fixed and paraffin wax-embedded tissue blocks used in the present study were surgical resection specimens obtained from 115 patients. Data on age, sex, pathologic stage, histological subtype, lymph node involvement, tumor size and location were extracted from patients' records held at the Research Center for Gastrointestinal Diseases, Firoozgar Hospital. Tumor slides were reviewed to determine histological subtypes of tumors by an expert pathologist.

Three tumor cores were carefully selected by H&E coloring in each formalin-fixed and paraffin wax-embedded tissue blocks. Then, sampling from these cores was done using a tissue microarray (TMA) devise at the Cancer and Pathology Research Center of Tehran University of Medical Science (Hemmat campus).

Seven TMA recipient blocks were made from 115 donor blocks (each with three tumor cores). The slices were then made from these TMA blocks, using H&E and IHC coloring to determine HER2. Colored slices were seen through field light microscopy with a typical magnification of x 40 and interpretation was made according to the recommendations of the consensus panel on HER2 scoring of gastric carcinoma. Briefly, scores were as follows: samples with strong complete or basolateral cell membrane reactivity in ≥10% of tumor cells scored IHC 3+ (positive staining), weak to moderate complete or basolateral membrane reactivity in ≥10% of tumor cells scored IHC 2+ represented equivocal staining and should be surveyed further with FISH or CISH, but in this study this group registered negative staining. Samples with faint/barely perceptible partial membrane reactivity in ≥10% of tumor cells scored IHC 1+ (negative staining) and finally, samples with levels of membrane reactivity less than 10% of tumor cells scored IHC 0 (negative staining).

Data were analyzed with SPSS software version 16 using chi-square and independent sample T test. P values less than 0.05 were considered statistically significant.

Results

One-hundred and fifteen gastric adenocarcinoma cancer patients were studied. The study group consisted of 81 males and 34 females, with a mean age of 61.01 (30-85 years). Most of the gastric tumors were found in the stomach (n=99, 86.1%) and others were located at the gastroesophageal junction (GEJ) (n=16, 13.9%). According to the Lauren classification, more than half of the patients had intestinal adenocarcinoma (n=76, 66.1%). The frequencies of other types are shown in Table1.

According to these results, among 101 patients whose tumor pathologic state, lymph node (LN) involvement and tumor size were available, the frequency of pathologic stage IIIA tumors were most common (n=34, 33.7%). LN involvement was detected in 68patients (67.3%) and mean tumor size was 5.351 cm. More information about these factors is shown in Table 1.

table 1: Baseline patient characteristics and their relation with HER2 overexpression

Baseline characteristics	NO(percent)	P-value
All patients	115	
Mean age, y (range)	61.01 (30-85)	0.466
Male: Female (%)	81:34 (70.4:29.6)	0.920
Tumor location, n (%) Stomach GEJ	 99(86.1) 16(13.9)	0.491
Tumor type, n (%) Intestinal Diffuse Mixed	 76(66.1) 31(27) 8(7)	0.607
Pathologic stage, n(%)[total 101] IA IB II IIIA IIIB	 6(5.9) 11(10.9) 29(28.7) 34(33.7) 21(20.8)	0.745
LN involvement, n(%)[total 101] Positive Negative	 68(67.3) 33(32.7)	0.429
Mean Tumor size, cm (range)[total 101]	5.351(0.5-12.0)	0.135
HER2, n (%) Positive Negative	 13(11.3) 102(88.7)	
HER2 score, n(%) 0 1+ 2+ 3+	 67(58.3) 20(17.4) 15(13.0) 13(11.3)	

As the results of this study show, HER2 overexpression (HER2 IHC score 3+) was detected in 13 patients (11.3%). There was not a significant relationship between gastric adenocarcinoma subtypes and HER2 overexpression (p= 0.607). In Table 2, HER2 states in different subtypes of GC tumor were found.

There was also no significant relationship between stages of gastric adenocarcinoma and HER2 overexpression (P= 0.745). Although there was not a significant relationship between tumor size and HER2 overexpression, there was a significant difference between HER2 overexpression in groups with tumor size ≥5cm and tumor size <5cm (P=0.01).

According to these results, 67 patients (58.3%) had an HER2 IHC score of 0 and patients with HER2 IHC scores of 1+ were the second most frequent group (n=20, 17.4%). More information is presented in Table 1. Furthermore, there was no significant difference between gastric adenocarcinoma subtype of groups with 1+, 2+ and 3+ HER2 IHC score(P= 0.292).

Results of this study showed that there was no significant correlation between age and HER2 overexpression and age was not an independent predictor of HER2 as either positive or negative.

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Table2: the relation between HER2 and type of adenocarcinoma (p-value = 0.607)

HER2 overexpression	Tumor type			Total
	Adenocarcinoma (intestinal)	Adenocarcinoma (diffuse)	Adenocarcinoma (mix)	
Positive	10	2	1	13
Negative	66	29	7	102
Total	76	31	8	115

Table3: the relation between HER2 overexpression and Tumor size

HER2 overexpression	Tumor size		Total
	≥5	<5	
Positive	12	1	13
Negative	48	40	88
Total	60	41	101

Discussion

In this cross-sectional study, 115 patients with curative resection of primary GC or GEJ cancers were evaluated to determine the incidence of HER2 overexpression by IHC method and its relationship with clinicopathological features of cancer. According to these results, the incidence of HER2 overexpression considered as IHC 3+ was 11.3%. There is a great deal of variation among different studies on the incidence of HER2 overexpression, ranging from 7% to 53.4%. This may be attributed to different populations in various studies, methodology or scoring criteria (13-16). In a recent review of 24 studies (n=6542) the mean value of HER2

positivity was reported 19% (13). Moreover, the results of some studies imply that HER2 overexpression is reportedly less common in the early stages of gastric cancer than in more advanced stages (17, 18). The high ratio of patients with a diagnosis of an early stage of the disease in this study may have contributed to the low rate of HER2 overexpression exhibited here. In a study by Yan, Et al. on 124 gastric cancer patients which had the same definition of HER2 overexpression as the patients in this study HER2 overexpression was reportedly 9.4% which was in consistent with the results of the present study (19). Moreover, in a similar study by Park DI, et al. HER2 overexpression defined as IHC 2+ and 3+ was found 15.9% (20). Actually, if the present study included HER2 2+ as overexpression the incidence of HER2 positive in patients would be increased.

Results of the current study showed that HER2 overexpression varies according to histological subtypes and it was higher in the intestinal subtype than the diffuse one (13.1 vs 6.4), but no significant difference of HER2 overexpression was found in different histological subtypes of tumor. The results of the current study are consistent with the findings in prior studies conducted by Gravalos C et al. (15). Conversely, other studies have reported a significant relationship between HER2 positivity and histological subtype of tumor (16, 21-24).

There was no significant relationship between HER2 overexpression and tumor location in this study, but a significant relationship was reported in a study by Tnner M, et al. (16). Maybe the lower proportion of GEJ cancers in this study explains this difference.

In the current study, there was no significant relationship between pathologic stage and HER2 overexpression and this result is consistent with other reports (18, 19, 25, 26). Conversely, some other studies reported a significant relationship between these two factors (17, 18). Moreover, in the present study HER2 overexpression was higher in pathologic stages III and II (14.5%, 13.7%) than stage I (5.8%).This maybe because the present study was deprived of pathologic stage IV so, no distinct conclusion about the relationship between these two factors could be made.

There was not a significant relationship between HER2 overexpression and tumor size (cm) in this study (P: 0.135). In contrast, a significant difference in HER2 overexpression was found between GCs with tumor sizes ≥5cm and <5cm (P: 0.01). Maybe this explains the poor survival rate in patients with a tumor size of more than 5 cm as reported in some other studies (20).

In conclusion, according to the results, the incidence of HER2 overexpression in gastric adenocarcinoma was 11.3%. The higher incidence of HER2 overexpression in patients with tumors ≥5cm maybe indicate that anti-HER2 targeted therapy administration for these patients could be more beneficial.

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