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Research Paper

Evaluation of Human Epidermal Growth Factor Receptor 2 (HER2) expression in gastric and gastroesophageal junction adenocarcinoma

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Abstract

Introduction: Gastric and gastro-oesophageal junction (GOJ) adenocarcinoma is the fifth most common cancer and fourth leading cause of cancer death globally. HER2 overexpression occurs in 13-22% of gastric and 30% of GOJ adenocarcinoma.

Objective: This was a retrospective study of HER2 (4B5) expression in gastric, oesophageal and GOJ adenocarcinoma reported at a single centre in the United Kingdom. HER2 (4B5) immunohistochemistry (IHC), in situ-hybridization(ISH) status and other pathological data of gastric and GOJ adenocarcinomas reported in both biopsies and resection specimens were collected from January 2014 to December 2022.

Results: A total of 481 patients were included in the study. The median age of patients at the time of surgery was 67.7 years; 68.2% were male. The proportion of patients with of HER2 (4B5) positivity was 13.30% (64/481) . Most HER2 positive tumours were grade 2 (G2) and of intestinal subtype (50%). Of the HER2 positive tumours, most were located at GOJ/Cardia (31; 48.43%), were pT1b (9/64, 39.13%) stage and showed nodal positivity (pN1) (12/64, 52.2%). HER2 assessment had been performed on both biopsies and subsequent resection specimens in 100 patients of which 95 (95%) cases showed concordance of HER2 results. The discordance was mainly due to tumour heterogeneity.

Conclusion: The overall prevalence of HER 2 positivity was 13.30%. HER2 positivity was associated with intestinal subtype rather than diffuse subtype of adenocarcinoma. Tumour heterogeneity was identified as a cause for cases of discordance of HER expression between biopsy and resection specimens.

Keywords: gastric adenocarcinoma, gastroesophageal junction adenocarcinoma, oesophageal adenocarcinoma, HER2

Introduction

Gastric cancer (including cancer of the gastro-oesophageal junction) is the fifth most common cancer worldwide and fourth leading cause of cancer death globally (1).

Oesophageal adenocarcinoma is reported to be associated with Barrett's oesophagus. Excess body weight, gastroesophageal reflux disease and *Helicobacter pylori* infection play a role in cardiac gastric cancer pathogenesis. Non-cardia gastric cancer is considered to be caused by chronic *H. pylori* infection (1).

Human epidermal growth factor receptor 2 (HER2) is a proto-oncogene, located on chromosome 17, encoding for a membrane-bound tyrosine kinase receptor (2). HER2 is a key driver of tumorigenesis, where it is implicated in tumor cell proliferation, apoptosis, adhesion, migration, and differentiation (3)

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The pathogenesis and progression of several solid organ epithelial malignancies including gastric, oesophageal, colorectal, breast, ovarian, prostate, lung, pancreatic, bladder, uterine carcinomas and osteosarcoma is driven by amplification of the HER2 gene leading to overexpression of the membrane bound HER2 receptor (4).

HER 2 overexpression occurs in 12-17% of the oesophageal and 13-22% of the gastric adenocarcinoma and ranges up to 30% in gastro-oesophageal junction (GOJ) adenocarcinoma (2). HER2 overexpression is associated with poor prognosis in gastric cancer (5). Trastuzumab for Gastric Cancer (ToGA) trial demonstrated that trastuzumab, a humanized monoclonal antibody against HER2, in combination with chemotherapy, resulted in improved median overall survival and progression-free survival, compared with chemotherapy alone in patients with HER2 positive advanced gastric cancer (6).

Our objective was to determine the prevalence of HER2 positivity in upper gastrointestinal (GI) adenocarcinoma and to describe the pathological characteristics of HER2 positive upper GI adenocarcinoma in surgical resection specimens.

Methods

This was a retrospective case series. All the biopsies and resection specimens (oesophagogastrectomy, partial/total gastrectomy - including cases with and without neoadjuvant chemotherapy) with oesophageal, gastric or GOJ adenocarcinoma reported at Royal Surrey Hospital form January 2014 to December 2022 in which HER2 (4B5) status was assessed were included. Resection specimens following neoadjuvant chemotherapy that showed positive lymph nodes with a complete pathological response in the primary tumour site (ypT0 yN1) in which where the HER2 status has been tested on the positive lymph node were excluded. The study received the approval from the Chief of Service, Audit Department, Royal Surrey Hospital (No: DCSS-CA-23-24-055).

Pathological features		Number of cases (%)
Tumour site	GOJ/ Cardia	31(48.43)
	Lower oesophagus	27(42.18)
	Stomach non-cardia	6 (9.37)
Tumour subtype/grade	G1 Intestinal	5(7.81)
	G2 Intestinal	32(50)
	G3 Intestinal	21(32.30)
	G3 Diffuse	3(4.68)
	G3 Mixed	3(4.68)
T stage	T1a:	0 (0%)
	T1b	9 (39.13%)
	T2	7 (30.43%)
	T3	6 (26.08%)
	T4a	1 (4.34 %)
	T4b	0 (0%)
N stage	N0	7 (30.4%)
	N1	12 (52.2%)
	N2	2 (8.7%)
	N3	2 (8.7%)
Tumour regression grade	TRG3	3 (16.6%)
	TRG4	11 (61.1%)
	TRG5	4 (22.2%)

Table 1: Pathological features of HER2 (4B5) positive upper GI adenocarcinoma

Three hundred and seventy-five biopsies and 206 resection specimens of oesophageal, GOJ and gastric adenocarcinomas fulfilling the inclusion and exclusion criteria were identified. HER 2 (4B5) had been performed on both biopsy samples and subsequent resection specimens in 100 patients. Four hundred and eighty-one patients [(375-100) + 206 = 481] were included in the study.

Demographic and pathological data were obtained from Winpath data base in Department of Histopathology.

Tumour site was determined by assessing the macroscopic description of tumor location in the histology report of resection specimens and comparing it with the Royal College of Pathologists, Standards and datasets for histopathological reporting of oesophageal and gastric carcinoma October 2019 (7) and TNM classification of malignant tumours 8th edition (8). Histological tumour type was determined based on the Laurén classification and categorized as diffuse, intestinal and mixed/unclassifiable types (9).

HER2 status assessment was performed by using IHC staining, and silver-in situ hybridization (SISH) techniques in HER2 equivocal cases. HER2 had been scored using the modified scoring system for gastro-oesophageal adenocarcinoma by Hofmann et al/ Ruschoff et al (10), as used in the ToGA trial (3). HER 2 IHC scores of 3+ was taken as positive. Score values of 0 and 1+ were taken as negative. HER2 IHC score 2+ cases were considered as equivocal and were further tested for gene amplification by SISH method. SISH had been performed on all of the HER2 2+ cases for assessment of amplification.

Results

A total of 481 patients were tested for HER2 status in this cohort. The median age of patients was 67.7 years, with 68.2% of patients being male. The overall proportion of HER2 (4B5) positivity in the cohort was 13.30% (64/481).

Among HER2 positive cases, most were grade 2 (G2) and of intestinal subtype (32/64, 50%) (Table 2). Most tumours were located in the gastroesophageal junction/cardia (31/64; 48.43%) (Table 1).

Of the 18 post-neoadjuvant HER2 positive resection specimens the majority were of tumour regression grade 4 (TRG4) comprising 11 specimens (61.1 %). TRG Grade 5 and 3 were seen in 4 (22.2%) and 3 (16.6) specimens respectively.

With respect to TNM staging, of the HER2 positive tumours, the majority were of the T1b category (9/64,; 39.13%), signifying an early stage of tumor progression and had a nodal stage of N1 stage (12/64; 52.2%) (Table 1).

HER2 (4B5) had been performed on both biopsy samples and subsequent resection specimens in 100 patients. Ninety-five (95/100, 95%) showed concordance for HER2 IHC and SISH interpretation. The details of the five discordant cases are described in Table 2.

Discussion

The overall prevalence of HER2 (4B5) positivity in this cohort was 13.3% (64/481), which is closely related to rates of HER2 positivity reported in other studies. The distribution of tumour subtypes, grade and T and N stage among HER2 positive cases in this study are compared with other studies in table 3.

HER 2 (4B5) had been performed on both biopsy samples and subsequent resection specimens in 100 patients in the present study and 95% cases showed concordance in the results. Intratumoral HER2 heterogeneity was the major cause for discordant HER2 status between biopsy and resection specimens. It is suggested that a minimum of five biopsy fragments should be assessed to accurately predict HER2 status (7), which may reduce the impact of tumour heterogeneity.

Year	HER2(4B5) status on biopsy	Chemotherapy (CT) given / not given	HER2 (4B5) status on resection specimen	Comment of the project lead after reviewing slides	Explanation for discordance
2017	HER2 3+ in G2 areas and negative (score 0) in G3 areas	Not given	Negative (score 0)	Block taken for assessment in resection specimen contains predominantly G3 areas	Tumour heterogeneity
2018	Negative (HER2 1+)	Not given	Positive Focal HER2 2+ and HER2 3+ areas. HER 2 SISH amplification.	Biopsy showed both HER2 1+ areas and HER2 2+ areas. SISH should have been done on the biopsy.	Tumour heterogeneity
2019	Negative	CT given	Positive Patchy HER 2 2+ and focal HER2 3+ staining in high grade dysplasia and intramucosal adenocarcinoma components (<10%). HER2 SISH amplification.	SISH shows heterogeneity. HER2 3+ positivity is seen very focally in <10% of the tumour.	Tumour heterogeneity
2020	Negative (HER2 1+)	CT given	Positive HER2 3+	Post CT clone that remained behind is HER2 3+	Residual HER 2 positive clone
2021	Positive (HER2 3+)	CT given	Negative (score 0)	Positive clone is not seen in the block sent for HER2 (4B5) staining. Repeat HER2 (4B5) on another block revealed HER 2+ and HER2 3+ staining in G2 areas and negative staining in G3 areas.	Tumour heterogeneity or down regulation of HER2 (4B5) post CT

Table 2: Discordance for HER2 (4B5) staining in biopsy samples and resection specimens

Study	**Van-Cutsem et al (3) (HER2 screening data from ToGA trial)**	**Koopam et al (11)**	**Fazlollahi et al (12)**	**Smolinska et al (13)**	**Ngaiza et al (6)**	**Present study**
Published year	2014	2017	2018	2019	2022	2023
Country	Asia Pacific, Europe, Central/South America	Netherlands	USA	Poland	Tanzania	United Kingdom
Sample size	3803 screened. HER2 IHC and FISH testing successful in 3665	319	100	104	150	481
Specimen type	Biopsies: 2492 Resections: 1173	Biopsies 232 Resection: 87	Biopsies only: 12 Resection only: 69 Both: 19	Gastrectomy specimens:104	Biopsies: 41 Resections: 109	Biopsies: 275 Resection:206
Number of HER2 positive cases	810	48	13	20	9	64
HER2 positive rate across analysed sample	810 / 3665 (22.1%)	48/319 (15%)	13% (13/100)	20/104 (19. 23%)	9/150(6.0%)	64/481 (13.30%)
Tumour location	GOJ: 208 (5%) Stomach: 2195 (58%) Metastatic sites: 322 (8%) Primary tumor (exact location of specimen not recorded) 1046 (28%) Not assessed 32 (<1%). *Tumour location among HER2 positive cases* *Stomach: 451 (56%)* *GOJ: 65 (8%)* *Metastatic sites: 51 (6%)* *Primary tumor (exact location of specimen not recorded 232 (29%)* *Not assessed 9 (1%).*	Distal oesophagus 54, (16.9%) GOJ 97 (30.4%) Cardia: 28 (8.8%) Non cardia: 133(41.7%) Unknown: 7 (2.2%)	Oesophagus: 10 (10%) GOJ: 44 (44%) Stomach: 46 (46%)	Cardia: 33 (31.8%) Fundus: 38 (36.5%) Antrum: 12 (11.5%) Pylorus: 21 (20.2%) *Tumour location among HER2 positive cases.* *Cardia: 7 (21.21%)* *Fundus: 6 (15.79%)* *Antrum: 1 (8.33%)* *Pylorus 6 (28.57%)*	Gastric: 123 GOJ: 27 *Tumour location among HER2 positive cases.* *Gastric: 8 (6.5%)* *GOJ: 1 (3.7%)*	*Tumour location among HER2 positive cases* *Lower oesophagus: 27 (42.18%)* *GOJ/Cardia: 31(48.43%)* *Stomach: 6 (9.37%)*
Tumour type	Intestinal: 1916 (50%) Diffuse: 1117 (29%) Mixed: 652 (17%) Not assessed: 118 (3%) *Tumour type among HER2 positive cases.*	Intestinal: 188 (58.9%) Diffuse: 85 (26.6%) Mixed: 38 (11.9%) Indeterminate: 8 (2.2%)	Intestinal: 60 (60%) Diffuse: 33 (33%) Mixed: 4 (4%) Other 3 (3%)	Intestinal:54 (51.92%) Diffuse: 41 (39.42%) Mixed: 9 (8.65%) *Tumour type among HER2 positive cases.*	Intestinal: 77 (51.3 %) Diffuse: 41 (27.3 %) Mixed: 32 (21.4 %) *Tumour type among HER2 positive cases.*	*Tumour type among HER2 positive cases.* *G1 Intestinal: 5 (7.81%) G2 Intestinal: 32 (50%)* *G3 Intestinal: 21 (32.30%)* *G3 Diffuse: 3 (4.68%)*

	Intestinal: 606 (75%) *Diffuse: 68 (8%)* *Mixed: 131 (16%)* *Not assessed 5 (<1%)*			*Intestinal: 12 (60%)* *Diffuse: 6 (30%)* *Mixed: 2 (10%)*	*Intestinal: 9 (11.7%)* *Diffuse: 0 (0%)* *Mixed: 0 (0%)*	*G3 mixed: 3 (4.68%)*
Pathological grade	-	-	G1: 2 (2%) G2: 41 (41%) G3: 57 (57%)	G1: 2 (10%) G2 45 (43.27%) G3 57 (54.81%) *Pathological grade among HER2 positive cases* *G1: 2 (10%)* *G2: 8 (17.78%)* *G3: 10 (17.54%)*	G1: 46 (30.7%) G2: 54 (36.0%) G3: 50 (33.33%) *Pathological grade among HER2 positive cases.* *G1: 8 (17.4%)* *G2: 1 (1.9%)* *G3: 0 (0%)*	*Pathological grade among HER2 positive cases.* *G1: 5 (7.81%)* *G2: 32 (50%)* *G3: 27 (42.19%)*
T Stage	-	-	T1: 29 (29%) T2: 16 (16%) T3: 38 (38%) T4: 17 (17%)	T0: 8 (7.69%) T1 – T2: 31 (39.81%) T3-T4: 65 (62.50%) *T stage among HER2 positive cases* *T0: 0 (0%)* *T1-T2: 7 (22.58%)* *T3-T4: 13 (20.0%)*	-	*T stage among HER2 positive cases* *T1a: 0%* *T1b: 9 (39.14 %)* *T2 7 (30.43 %)* *T3 6 (26.08 %)* *T4a 1 (4.34%)* *T4b 0%*
N stage	-	-	Nx: 5 (5%) N0: 41 (41%) N1: 24 (24%) N2: 9 (9%) N3: 21 (21%)	N0: 40 (38.46%) N1-N3: 64 (61.54%) *N stage among HER2 positive cases.* *N0: 5 (12.5%)* *N1-N3: 15 (23.44%)*	-	*N stage among HER2 positive cases.* *N0 7 (30.4%)* *N1 12 (52.2%)* *N2 2 (8.7%)* *N3 2 (8.7%)*
Results	HER2 expression varied by tumour location and type. Concordance between HER2 and FISH was 87.2%	-	Concordance of HER 2 positivity between biopsy and paired resection was 78.9%.			Concordance of HER 2 positivity between biopsy and paired resection was 95%

Table 3: Comparison of findings of present study with studies on clinicopathological features of HER2 positive upper GI adenocarcinoma

Conclusion

Accurate assessment of HER2 status in gastric, gastroesophageal junction, and oesophageal adenocarcinomas is of great importance because anti-HER2 monoclonal antibody targeted therapy, in combination with with chemotherapy, has shown to improve median overall survival of patients with HER2 positive adenocarcinoma.

HER2 testing should be performed on resection specimens when the initial biopsy has a negative HER2 IHC (scores 0, 1+) or equivocal 2+ and amplification-negative results if patients are being considered for anti-HER2 monoclonal antibody–targeted therapies because biopsy testing alone may miss potential cases with HER2 positivity because of tumor heterogeneity.

References

1. Bray F, Ferlay J, Soerjomataram I, Siegel RL, Torre LA, Jemal A. Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. *CA Cancer J Clin*. 2018;68(6):394-424. doi:10.3322/caac.21492.

2. Thijs L, Peeters P, Maes B, Tejpar S. HER2 overexpression in gastroesophageal adenocarcinoma,
prognostic and predictive value
and diagnostic approach. Belgian Journal of Medical Oncology 2019;13(4):135-141).

3. Van Cutsem E, Bang YJ, Feng-Yi F, et al. HER2 screening data from ToGA: targeting HER2 in gastric and gastroesophageal junction cancer. *Gastric Cancer*. 2015;18(3):476-484. doi:10.1007/s10120-014-0402-y

4. Nagaraja V, Eslick GD. HER2 expression in gastric and oesophageal cancer: a meta-analytic review. *J Gastrointest Oncol*. 2015;6(2):143-154. doi:10.3978/j.issn.2078-6891.2014.107.

5. Yang T, Xu R, You J, Li F, Yan B, Cheng JN. Prognostic and clinical significance of HER-2 low expression in early-stage gastric cancer. *BMC Cancer*. 2022;22(1):1168. doi:10.1186/s12885-022-10262-7

6. Ngaiza A, Vuhahula E, Yahaya J, et al. Evaluation of Human Epidermal Growth Factor Receptor 2 Expression in Gastric and Gastroesophageal Cancers in Tanzania. *Arch Pathol Lab Med*. 2022;146(12):1523-1529. doi:10.5858/arpa.2021-0394-OA

7. Grabsch HI, Mapstone NP, Novelli M. Standards and datasets for reporting cancers. Dataset for histopathological reporting of oesophageal and gastric carcinoma October 2019 [Internet]. 2019 Oct.

8. Daiko H, Kato K. Updates in the 8th edition of the TNM staging system for esophagus and esophagogastric junction cancer. *Jpn J Clin Oncol*. 2020;50(8):847-851. doi:10.1093/jjco/hyaa082

9. Chen YC, Fang WL, Wang RF, et al. Clinicopathological Variation of Lauren Classification in Gastric Cancer. *Pathol Oncol Res*. 2016;22(1):197-202. doi:10.1007/s12253-015-9996-6

10. Rüschoff J, Hanna W, Bilous M, et al. HER2 testing in gastric cancer: a practical approach. *Mod Pathol*. 2012;25(5):637-650. doi:10.1038/modpathol.2011.198

11. Koopman T, de Bock GH, Buikema HJ, et al. Digital image analysis of HER2 immunohistochemistry in gastric- and oesophageal adenocarcinoma: a validation study on biopsies and surgical specimens. *Histopathology*. 2018;72(2):191-200. doi:10.1111/his.13322

12. Fazlollahi L, Remotti HE, Iuga A, Yang HM, Lagana SM, Sepulveda AR. HER2 Heterogeneity in Gastroesophageal Cancer Detected by Testing Biopsy and Resection Specimens. *Arch Pathol Lab Med*. 2018;142(4):516-522. doi:10.5858/arpa.2017-0039-OA

13. Smolińska M, Grzanka D, Antosik P, et al. HER2, NF-κB, and SATB1 Expression Patterns in Gastric Cancer and Their Correlation with Clinical and Pathological Parameters. *Dis Markers*. 2019;2019:6315936. Published 2019 Oct 14. doi:10.1155/2019/6315936