Original Research Article

Indian Journal of Pathology: Research and Practice
Volume 9 Number 3, September – December 2020
DOI: http://dx.doi.org/10.21088/ijprp.2278.148X.9320.41

Correlation of HER2 Positivity with Clinicopathological Parameters in Colorectal Carcinoma Patients

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How to cite this article:
Shruthi Gowthami M R, Nirmala C. Correlation of HER2 Positivity with Clinicopathological Parameters in Colorectal Carcinoma Patients. Indian J Pathol Res Pract 2020;9(3):269–273.

Abstract

Context: Identification of her2 expression by Immunohistochemistry and correlating it with histopathological parameters in colorectal carcinoma may help know the prognosis and helps for the apt use of anti-Her2 targeted therapy.

Aims: To correlate Her2 positivity with clinicopathological parameters in colorectal carcinomas.

Settings and Design: Retrospective study.

Methods and Material: The study was carried out in the Department of Pathology in a Tertiary Care Centre. Total 30 patients studied. Tissue blocks from colonic biopsies and colectomy specimens of histologically proven colorectal carcinomas were retrieved for immunohistochemical analysis. Clinicopathological characteristics were obtained from patients' medical records and pathology reports. Hematoxylin and Eosin stained slides of biopsies and resected specimens of histologically proven colorectal carcinomas were retrieved and reviewed for histopathological parameters. HERACLES scoring was used for HER2 expression. Data was analysed with appropriate statistical tests.

Statistical analysis used: Appropriate statistical tests.

Results: Mean age of the patient was 48.32± 3.4 years. 8 (26.67%) patients were HER2 positive. No significant difference was observed in HER2 positivity and grade of tumour ($p>0.05$). Cytoplasmic expression was most commonly observed in low grade tumour 07(43.75%) but statistically not significant; ($p>0.05$). Similarly Membranous expression was also commonly observed in low grade tumour. Stage of tumour did not show any association with grade, HER2 expression or HERACLES score. ($p>0.05$).

Conclusions: Assessment of Her2/neu scoring irrespective of histopathological and clinicopathological parameters may help select the appropriate targeted therapy.

Keywords: Clinicopathological; Colorectal cancer; Her2; Immunohistochemistry.

Key Messages: Identification of her2 expression by Immunohistochemistry and correlating it with histopathological parameters in colorectal carcinoma may help know the prognosis and helps for the apt use of anti-Her2 targeted therapy. The study was carried out in the Department of Pathology in a Tertiary Care Centre to correlate Her2 positivity with clinicopathological parameters in colorectal carcinomas. Total 30 patients studied. We conclude that assessment of Her2/neu scoring irrespective of histopathological and clinicopathological parameters may help select the appropriate anti-Her2 targeted therapy. This brings effective personalized medicine into picture.

Introduction

The global cancer burden is significant and increasing. In this era of personalized medicine, various targeted therapies are available which target specific proteins responsible for the proliferation of cells in cancers.

HER2 is an oncogene which is over expressed in malignancies of breast, ovarian, gastric, Colorectal, pancreatic and endometrial cancers.[1,2]

The introduction of HER2-targeted therapy for breast and gastric patients with ERBB2 (HER2) amplification/overexpression has led to dramatic improvements in oncologic outcomes.[3] Limited literature about the expression of Her2 in colorectal cancer is available with prevalence ranging from 0 to 84%.[4]

Identification of her2 expression by Immunohistochemistry and correlating it with histopathological parameters in colorectal carcinoma may help know the prognosis and helps for the apt use of targeted therapies. Also location of Her2 expression in colorectal carcinoma whether membranous or cytoplasmic helps in the selection of suitable therapeutic agent; thus Her2/neu can also be utilized as a predictive marker.

So here is an effort to study the correlation of Her2 positivity with clinicopathological parameters in colorectal carcinoma patients.

Material and Methods

Retrospective study carried out in patients of colorectal carcinomas in a Tertiary Care Centre during January 2018 to December 2019.

Based on previous study by Shabbir A et al prevalence of colorectal carcinoma was 78.9%.[5] According to formula n = (Zα)pq/ d2 (Zα= Standard table value for 95% Confidence Interval, p= prevalence of her2 positivity in colorectal cancer in previous study, q= 100-p, d= relative precision of 20% of p, n= sample size) our sample size was 30.

Inclusion criteria: Patients diagnosed as Colorectal carcinomas in either biopsy or resected specimens received in the tertiary care centre during the study period.

Exclusion criteria : 1. Patients diagnosed as non-epithelial malignancies of colorectal region. 2. The patients diagnosed as colorectal carcinomas and whose blocks and slides are not available for the study. 3. Very small biopsies.

Tissue blocks from colonic biopsies and colectomy specimens of histologically proven colorectal carcinomas were retrieved and immunohistochemical analysis done.

Clinical features, gross and microscopic findings were obtained from patient's medical records and pathology reports. Hematoxylin and Eosin stained slides of biopsies and resected specimens of histologically proven colorectal carcinomas were retrieved and reviewed for histopathological parameters. Histopathological grading of tumors was performed according to the World Health Organization (WHO) criteria as grade I (well differentiated), grade II (moderately differentiated) and grade III (poorly differentiated). Pathological staging of colectomy cases were recorded as per the 7th edition of the American Joint Committee on Cancer Staging (stage I to stage IV).[6]

Histopathological scaling of tumors was performed according to nuclear pleomorphism, mitoses rate and tubule formation (grade system).

Tubule formation represents percent of cancer cells that are in tubule formation. Grade of 1 means more than 95% cells are in tubule formation (better). Grade of 2 means tubule formation is found in 50–95% cells and Grade of 3 indicates tubule formation in less than 50% cells (worst).

Nuclear pleomorphism indicates variation in size and shape of cancer cell nuclei. Scale 1 means nucleus closest to normal cells (better). Scale 3 is having more variations.

Mitotic rate describes how quickly cancer cells are multiplying. Scale 1 indicates slow multiplication (1–4 mitoses/100 tumor cells), scale of 2 is 5–7 mitoses/100 tumor cells and scale of 3 is 8–10 mitoses/100 tumor cells.

Her-2/neu stained slides were independently evaluated by the investigator using HERACLES HER2 scoring system.

Data was analysed using SPSS software version 22 and technique applied was Chi square or Fischer test for comparing categorical data. P value significance was seen at ≤0.05.

Results

In the present study, 30 patients with colorectal carcinoma were studied. Of them 60% population were males and 40% were females. HER2 positivity was seen in total 8(26.6%) cases and negative in 22(73.4%) cases (Fig. 1). Among males, HER2 was positive in 5(22.2%) cases and negative in 13(77.6%)

cases and in females 3(25%) cases were HER2 positive and 9(75%) cases were HER2 negative. (Table 1)

Table 1: Distribution of patients according to gender and HER2 staining.

Gender	HER2 positive	HER2 negative	Total
Male	05(62.5%)	13(59.09)	18 (60%)
Female	03(37.5%)	09(40.91%)	12(40%)
Total	08(100%)	22(100%)	30(100%)

P =0.86 not significant

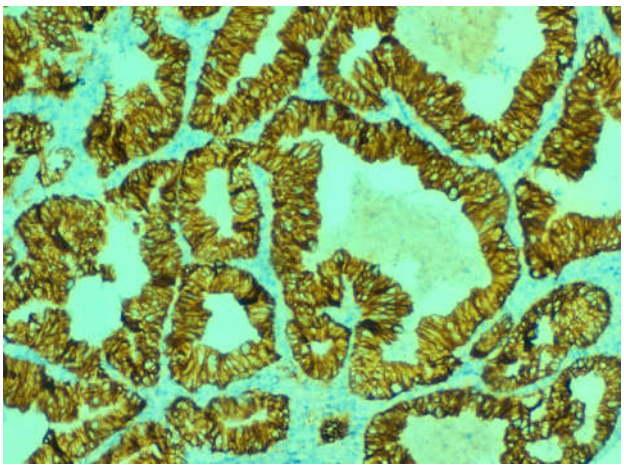

Fig. 1: Her2 positivity of Score 3+ in colorectal carcinomas; membranous immunostaining is incomplete and basolateral.

Mean age of the patient was 48.32± 3.4 years. Majority of the patients were from the age group of 51–60 years, 10 cases(33.34%) followed by 41–50 years 09 cases(30%). Among HER2 positive samples maximum were from age group of 31–40 years followed by 41–50 and 51–60 years (25%) each. HER2 negative samples were maximum seen in age group of 51–60 years. Difference between them is not significant (p>0.05) (Table 2)

Table 2: Distribution of patients according to age group and HER2 staining.

Age group	HER2 positive	HER2 negative	Total
31–40	03(37.5%)	01(4.55%)	04(13.33%)
41–50	02(25%)	07(31.83%)	09(30%)
51–60	02(25%)	08(36.36%)	10(33.34%)
61–70	00(0%)	03(13.64%)	03(10%)
>70	01(12.5%)	03(13.64%)	04(13.33%)
Total	08(100%)	22(100%)	30(100%)

P =0.2 not significant.

Of the total 30 colorectal cancer tissues, 28 (93.33%) showed Her-2/neu staining, among which 8 (26.67%) patients were HER2 positive according to HERACLES HER2 Scoring System.

Out of total 30 patients 27 (90%) presented with growth and 3 (10%) presented with intestinal obstruction. All HER2 positive samples presented with growth. (Table 3)

Table 3: Distribution of patients according to clinical history and HER2 staining.

Clinical history	HER2 positive	HER2 negative	Total
Growth	08 (100%)	19(86.36%)	27(90%)
Intestinal obstruction	00(0%)	03(13.64%)	03(10%)
Total	08(100%)	22(100%)	30(100%)

P =0.27 not significant.

Most common location of tumour was rectum and sigmoid colon in both HER 2 positive and HER 2 negative samples. Ascending colon was less observed site for tumour in both the groups. Right sided tumours were less than left sided one. (Table 4)

Table 4: Distribution of patients according to location of tumour and HER2 staining.

Location	HER2 positive	HER 2 negative	Total
Splenic flexure	01(12.5%)	00(0%)	01(3.33%)
Ascending colon	01(12.5%)	04(18.18%)	05(16.67%)
Transverse colon	00 (0%)	01(4.55%)	01(3.33%)
Sigmoid colon	02(25%)	06(27.27%)	08(23.67%)
Rectosigmoid junction	01(12.5%)	01(4.55%)	02(6.67%)
Rectum	03(37.5%)	10(45.45%)	13(43.33%)
Total	08(100%)	22(100%)	30(100%)

P =0.1 not significant.

Histologically 2(6.67%) specimen were mucinous and 28(93.34%) were non mucinous.

Majority of the tumours in our study were of Grade I 17(56.67%) followed by Grade II 07(23.33%). Grade III tumour were 06(20%). Among grade III, two cases were mucinous adenocarcinomas. HER2 positive tumours were mostly from grade I (75%) and 25% in grade II, none in grade III. Of the total 22 HER2 negative tumours 11(50%) were grade I, 22% grade II, 28% grade III.

HER2 Positive expression was commonly observed with low grade tumour but No significant difference was observed in HER2 positivity and grade of tumour (p>0.05) (Table 5)

Table 5: Distribution of patients according to grade of tumour and HER2 staining.

Grade of tumour	HER2 positive	HER 2 negative	Total
Grade I	06(75%)	11(50%)	17(56.67%)
Grade II	02(25%)	05(22.73%)	07(23.33%)
Grade III	00(0%)	06(27.27%)	06(20%)
Total	08(100%)	22(100%)	30(100%)

P >0.05 Statistically not significant.

Table 6: Distribution of patients according to Tubule formation and HER2 staining.

% Tubule formation	HER2 positive	HER 2 negative	Total
>95%(Grade 1)	06(75%)	11(50%)	17(56.67%)
50–95%(Grade2)	02(25%)	05(22.73%)	07(23.33%)
<50%(Grade 3)	00(0%)	06(27.27%)	06(20%)
Total	08(100%)	22(100%)	30(100%)

P >0.05 Statistically not significant.

Tubule formation of grade I was observed commonly in both positive and negative samples(Table 6). Majority of the cells have mitosis rate of scale 1 in both positive (62.5%) and negative (68.18%) HER 2 expression (Table 7). Majority of the cells have nuclear pleomorphism of scale 2 in HER 2 positive and negative tumours (Table 8). Thus Mitosis rate, tubule formation and Nuclear pleomorphism were not statistically associated with positive and negative expression of HER2 staining (p>0.05)

Table 7: Distribution of patients according to Mitoses rate and HER2 staining.

Mitoses rate	HER2 positive	HER 2 negative	Total
Scale 1(1–4 mitoses/100 tumor cells)	05 (62.5%)	15(68.18%)	20(66.67%)
Scale 2(5–7 mitoses/100 tumor cells)	02(25%)	04(18.18%)	06(20%)
Scale 3(8–10 mitoses/100 tumor cells)	01(12.5%)	03(13.64%)	04(13.33%)
Total	08(100%)	22(100%)	30(100%)

P >0.05 Statistically not significant.

Table 8: Distribution of patients according to Nuclear pleomorphism and HER2 staining.

Nuclear pleomorphism	HER2 positive	HER 2 negative	Total
Scale 1	00(00%)	03(13.63)	03(10%)
Scale 2	06(75%)	18(81.82%)	24(80%)
Scale 3	02(25%)	01 (4.54%)	03(10%)
Total	08(100%)	22(100%)	30(100%)

Table 9: Distribution of HER2 staining pattern and grade of tumour.

Grade of tumour	Cytoplasmic	Membranous	Cytoplasmic + Membranous	Negative
Grade I	07(43.75%)	08 (72.73%)	01(100%)	01(50%)
Grade II	04 (25%)	02 (18.18%)	00(0%)	01(50%)
Grade III	05(31.25%)	01 (9.09%)	00(0%)	00(0%)
Total	16(100%)	11(100%)	01(100%)	02(100%)

Table 9 shows Distribution of HER2 staining pattern and grade of tumour. Cytoplasmic expression was most commonly observed in low grade tumour 07(43.75%) but statistically not significant. (p>0.05). Similarly Membranous expression was also commonly observed in low grade tumour.

Out of 30 patients 6 were resected specimen. Table 10 shows characteristics of resected specimen. Out of 6 specimens 2 were of stage IIA, one of IIB and 3 were of stage IIIB. Size of tumour ranges from 4×1 to 7×4. Membranous expression was seen in 3 patients and cytoplasmic expression was seen in 3 patients. Stage of tumour did not show any association with grade, HER2 expression or HERACLES score. (p>0.05)

Table 10: Characterstics of resected specimens.

Sr no	Stage of tumour	Size of tumour (cm)	Grade	Membranous/ cytoplasmic expression	HERACLES Score
1	IIA (PT3N0Mx)	4×1	I	M	1+
2	IIA (PT3N0Mx)	5×3	II	C	1+
3	IIB (PT4N0Mx)	7×4	I	M	3+
4	IIIB (PT3N1Mx)	7×4	I	M	2+
5	IIIB (PT3N1Mx)	3×1	III	C	2+
6	IIIB (PT4N1Mx)	5x4	III	C	1+

Discussion

In this study of 30 cases of colorectal carcinomas, mean age of the patient was 48.32± 3.4 years. These results are relatively comparable with the findings of Terzi et al and Neklason et al.[7,8]

In our study Male to female ratio was 1.5:1. Similar findings were seen in previous studies.[9,10]

Out of 30 patients 25 (80%) of them were biopsy specimen and 6(20%) were colectomy specimen.

HER 2 Positive expression was commonly observed with low grade tumour but No significant difference was observed in HER2 positivity and grade of tumour (p>0.05)

In our study, Membranous Her-2/neu staining was observed maximum in low grade. Cytoplasmic staining was observed more in low grade colorectal cancer. Similar to our study Shabbir A et al found Pattern of Her-2/neu staining was significantly associated with the grade of colorectal cancer depicting cytoplasmic Her-2/neu expression higher in low grade (50 %) while membranous Her-2/neu expression more in high grade colorectal cancer (45 %)(p-value = 0.030).[5] A study by Half E et al, 63.5 % cases with cytoplasmic expression were observed and a significant association as found with low grade colorectal carcinoma.[11]

In our study, Mitosis rate, tubule formation and Nuclear pleomorphism were not statistically associated with Positive and negative expression of HER2 staining (p>0.05). Shabbir et al where they observed a significant association between percentage of cells stained and tumor type, with score 3+ maximum in non mucinous type of colorectal cancer (p-value = 0.006).[5]

In our study Stage of tumour did not show any association with grade, HER2 expression or HERACLES score. (p>0.05) It may be because of limited sample size.

Limitations of the study: 1) Among these 8 cases, 2 cases were confirmed Her2 positive with Score 3+ in cells >50%. Other 6 cases with Score 2+ need to be confirmed by FISH(Flourescent Insitu Hybridization) or any Hybridization techniques. 2) Limited sample size.

Conclusion

No significant correlation between Immunohistochemical Her2 positivity and clinicohistopathological prognostic parameters was found in our study.

Hence we conclude that assessment of Her2/ neu scoring irrespective of histopathological and clinicopathological parameters may help select the appropriate anti-Her2 targeted therapy. This brings effective personalized medicine into picture.

References:

1. English DP, Roque DM, Santin AD. HER2 expression beyond breast cancer: therapeutic implications for gynecologic malignancies. MolDiagnTher2013;17:85–99.
2. Wen W, Chen WS, Xiao N, Bender R, Ghazalpour A, Tan Zet al. Mutations in the Kinase Domain of the HER2/ERBB2 Gene Identified in a Wide Variety of Human Cancers. J MolDiagn 2015;17:487–95.
3. Meric-Bernstam F, Johnson AM, Dumbrava EEI,Raghav KP, Balaji K, Bhatt Met al. Advances in HER2-Targeted Therapy: Novel Agents and Opportunities Beyond Breast and Gastric Cancer. Clin Cancer Res2019;25:2033–41.
4. Blok EJ, Kuppen PJ, van Leeuwen JE, Sier CF. Cytoplasmic overexpression of HER2: a key factor in colorectal cancer.Clin Med Insights Oncol2013;7:41–51.
5. Shabbir A, Mirza T, Khalid AB, Qureshi MA, Asim SA. Frequency of Her2/neu expression in colorectal adenocarcinoma: a study from developing South Asian Country. BMC Cancer 2016;16:855.
6. Edge SB, Compton CC. The American Joint Committee on Cancer: the 7th edition of the AJCC cancer staging manual and the future of TNM. Ann SurgOncol2010;17:1471–4.
7. Terzi C, Canda AE, Sagol O, Atila K, Sonmez D, FuzunM, etal. Survivin, and Ki-67 as predictors of histopathologic response in locally advanced rectal cancer treated with preoperative chemoradiotherapy. Int J Colorectal Dis 2008;23:37–45.
8. Neklason DW, Kerber RA, Nilson DB, Anton-Culver H, Schwartz AG, Griffn CA, et al. Common familial colorectal cancer linked to chromosome7q31:agenome-wideanalysis.Cancer Res2008;68:8993–7.
9. Schuell B, Gruenberger T, Scheithauer W, Zielinski C, Wrba F. HER 2/neuprotein expression in colorectal cancer.BMCCancer 2006;6:123.
10. Messerini L, Cianchi F, Cortesini C, Comin CE. Incidence and prognostic significance of occult tumor cells in lymph nodes from patients with stageIIA colorectalcarcinoma. HumPathol2006;37:1259–67.
11. Half E, Broaddus R, Danenberg KD, Danenberg PV, Ayers GD, Sinicrope FA. HER-2 receptor expression, localization, and activation in colorectal cancer cell lines and human tumors. Int J Cancer 2004;108:540–8.