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Welcome to the Int J Cancer and Biomedical Research (IJCBR)**!**

It is with great pleasure that I write this editorial to welcome you to the IJCBR. This journal provides a platform for publication of original and reviews research articles, short communications, letter to editor, thesis abstract, conference report, and case studies. These types of publication are directed at the interface of the fields of cancer and biomedical research.

The IJCBR relies on a distinguished expert of the Advisory and Editorial Board Members from the top international league covering in depth the related topics. They timely review all manuscripts and maintain highest standards of quality and scientific methodology and ethical concepts. Meanwhile, we take all possible means to keep the time of the publication process as short as possible.

I take this chance to welcome your contributions to the IJCBR and have every expectation that it will soon become one of the most respected journals in both the fields of cancer and biomedical research.

Mohamed L. Salem,

Editor in Chief

INTERNATIONAL JOURNAL OF CANCER AND BIOMEDICAL RESEARCH (IJCBR)

RESEARCH ARTICLE

The role of Her 2 neu and N-cadherin expression in hepatocellular carcinoma

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ABSTRACT

Background: Despite advances in diagnosis and surgery, patients with hepatocellular carcinoma (HCC)show bad prognosis and high recurrence rate. Therefore, it is important to search for new targeted therapy and prognostic markers. **Aim**: To evaluate the expression levels of Her2 neu and N-cadherin in HCC and whether they could be used as prognostic markers. **Materials and Methods:** This retrospective study included 40 HCC specimens retrieved from the archival cases of Pathology Department, National liver institute, Menoufia University, Egypt, spanning the period between January 2010 to December 2017. Overall survival time was available for 28 patients only. All specimens were stained by Her2 neu and N-cadherin antibodies using the streptavidinbiotin- peroxidase technique. **Results:** There was significant association between high Her2 neu expression and good prognostic parameters as absent lymphovascular invasion, low grade (P value = 0.028 and 0.049 respectively) and also was associated with low pathological stage but the results didn't reach the level of significance. However there was significant association between low N-cadherin score of expression and good prognostic parameters as low grade (p value = 0.022). Also, there is inverse association between Her 2 neu and N- Cadherin expression in HCC (P value = 0.000). **Conclusion:** The anti-Her 2 target therapy might not be valuable for HCC patients and also larger studies are recommended to explore potential alternative targeted Her2 enhancer or promoter as well as to understand the exact biological role of Her 2 neu in HCC.

Keywords: Hepatocellular carcinoma; Her 2 neu; immunohistochemistry; N-Cadherin; prognosis

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INTRODUCTION

Hepatocellular carcinoma (HCC) is the commonest adult liver primary malignant tumor and represents 85% - 90% of liver tumors (Gharib et al., 2014). It represents the sixth most frequent malignancy and the fifth leading cause of cancer deaths worldwide (Siegel et al., 2019), liver cancer incidence constitutes 14.1% in males and 5.2% in females (Sung et al., 2021) and it is the most common cancer in males and the second in females (Bray et al., 2018). Human Epidermal Growth Factor Receptor 2 (HER2) is a member of the epidermal growth factor receptor which is involved in the transmission of proliferation and differentiation signals. Its overexpression has been documented in some cancer types as breast cancer and is associated with worse biologic behavior and poor prognosis (Kim et al., 2020). There is conflicting data in the literature regarding HER2 expression, and the clinical significance of HER2 in HCC is still unclear. As such, studying the expression and the further mechanism of HER2 in HCC has been discussed to help find a new target as a treatment option for HCC as well as tumor recurrence after surgery (Shi et al., 2019).

There are more than 80 members of cadherins which are calcium-dependent molecules responsible for cell-cell junctions. Classic cadherins is a subfamily of cadherins that

mediate adherence junction between epithelial cells maintaining the tissue integrity and cellular polarity in addition to their important role during embryogenesis. N-cadherins is one of classic cadherins family and is found in neural tissue, skeletal, cardiac muscles and fibroblasts together with endothelial cells (Abdallah et al., 2019 and Liu et al., 2014)

Regarding E-cadherin expression, in spite of being decreased in many kinds of epithelial tumors, few reports showed direct association between tumor progression and E-cadherin loss, especially in vivo (Shimada et al., 2012). Regarding HCC, E-cadherin has vital roles in homeostasis maintenance and carcinogenesis suppression in the liver. As E-cadherin loss leads to induction of EMT, stem cell markers upregulation, which leads to enhanced carcinogenesis and an invasive phenotype (Maeda and Nakagawa, 2015).

Epithelial-mesenchymal transition (EMT) is a key event in the tumor invasion process, whereby epithelial cell layers lose polarity and cell-cell contacts and undergo a dramatic remodeling of the cytoskeleton (Zhang et al., 2019). The hallmark of EMT is loss of E-cadherin while N-cadherin is expressed (the so-called cadherin switch) in epithelial cells, which is accompanied by loss of tight cell-cell adhesion and acquisition of a fibroblastic morphology (Zhang et al., 2019).

This study aimed to examine the expression of Her2 neu and mesenchymal marker N-cadherin in HCC patients and their correlation with the available clinicopathological data.

MATERIAL AND METHODS

This retrospective study included 40 HCCs obtained from 40 Egyptian patients, retrieved from the archival material of Pathology Department, National Liver Institute, Menoufia University, during the period between January 2010 and December 2017.

Tissue Microarray (TMA) Construction

Multiple tissue cores (not 1 core only) with a diameter of 1.5 mm from the predefined regions of each specimen in donor paraffin block were punched manually using a tissue arrayer's needle set provided by the TMA instrument manufacturing company (Breecher Instrument), as a large area of the studied cases could be represented (Eskaros et al., 2017). We used the TMA needles with a simple handheld holder with great success without the need to use the expensive tissue arrayer instrument (Abdel-Rahman et al., 2014).

Immunohistochemistry

Paraffin-embedded tissue sections (5 mm) were deparaffinized in xylene and rehydrated. The sections were treated with 10 mM citrate buffer, pH 6.0, at 961C for 10 to 20 minutes, followed by 10mL of Tris-EDTA for 10 to 20 minutes. Endogenous peroxidase was blocked with peroxidase-blocking reagent (cat. #TP-015-HD) (Lab Vision Cooperation, Fremont, CA) using HER2 rabbit monoclonal antibody (c-erbB-2), clone GR011, ready to use, Genemed Biotechnologies, Inc. and N-cadherin mouse antihuman cadherin antibody (Neural cadherin, 13B154, US Biological). It is received as 0.1 concentrated and diluted by phosphate buffer saline (PBS) in a dilution 1:150

A positive reaction was revealed using the streptavidinbiotin- peroxidase technique (cat. #TP-015-HD) (Lab Vision Cooperation) with chromogen DAB. The sections were then counterstained with Mayer's hematoxylin (cat. No. 94583; Bio Genex) for 30 to 60 seconds. Sections were washed in tap water for 5 minutes. Positive controls for the reaction were performed with specific paraffin-embedded sections of breast carcinoma and melanoma for HER 2 neu and N-cadherin primary antibodies respectively, and negative controls were made by substituting the primary antibody with non-immune serum.

Interpretation of Immunostaining Results

Positive cells were identified by the presence of brownish cytoplasmic coloration for both Her 2 neu and N-cadherin detected by DAB reaction.

Interpretation of Her2 neu Immunohistochemical results

Tumor cells were considered positive when they showed brownish coloration of the cytoplasm **(**Shi et al., 2019 and Döring et al., 2021)**..** We determine the intensity of immunostaining as 0–3 (0, negative; 1, weak; 2, moderate; 3,

strong) and the percentage of immunoreactive cells as 0 (less than 5%), 1 (6%–25%), 2 (26%–50%), 3 (51%–75%), and 4 (76%–100%). Multiplication of the intensity and the percentage resulted in an immunoreactivity score ranging from 0 to 12 For categorical analyses, the immunoreactivity was graded as low level (total score ≤4) or high level (total score > 4) **(**Shi et al., 2019).

Interpretation of N-cadherin Immunohistochemical results

Tumor cells were considered positive when they showed brownish coloration of the cytoplasm. Semi-quantitative assessment of the immunohistochemical results was performed in terms of intensity of staining (range, 0–3; 0= absent, 1= weak, 2= moderate, 3= strong) and percentage of stained cells (range, 0–3; <10%=score 1, 10%–50%= score 2, >50%=score 3). Multiplication of the intensity and the percentage resulted in an immunoreactivity score ranging from 0 to 9. A final total score ≤3 was considered low expression, and score >3 was recorded as high expression (Jiang et al., 2018). All the immunostained slides were analyzed by two experienced pathologists working independently without knowing the patients' characteristics.

Statistical Analysis

The statistical analysis was conducted using SPSS "Statistical Package for the Social Sciences" program for windows, version 20 (SPSS Inc., Chicago, IL). Mann- Whitney (U) and Kruskal-Wallis tests were used to compare nonparametric data, and the X^2 and Fisher exact tests for assessing the association between the clinicopathologic parameters and both antibodies expression. P ≤ 0.05 was considered to indicate statistical significance in all tests (Dawson and Trapp, 2001). Ethical approval was not required as we used archival tissue blocks.

RESULTS

Clinicopathological data of the studied HCC cases (Table 1). Immunohistochemical results of N-cadherin and Her 2 neu in the studied HCC cases are shown in Table 2.

Relationship between Her 2 neu expression and clinicopathological data of the studied HCC cases: The current study showed statistical significant association between high Her2 neu score of expression and strong intensity of expression and good prognostic parameters as absent lymphovascular invasion, low tumor grade (P value = 0.028, 0.036, 0.049 and 0,02 for score and intensity of expression respectively). Also high Her2 neu score of expression was statistically associated with focal Hep par 1 expression (P value= 0.033) and strong intensity of Her 2 neu expression was statistically associated with trabecular tumor pattern (P value= 0.02) (Tables 3 and 4) and (Plate 1).

On the other hand, strong intensity of Her 2 neu expression was statistically associated with involved parenchymal margin and diffuse Hep par 1 distribution of expression (P = 0.045 and 0.03 respectively) Furthermore high Her 2 neu score of expression was associated with low pathological stage but the results didn't reach the level of significance.

Relationship between N-cadherin score of expression and clinicopathological data of the studied HCC cases: The current study showed statistical significant association between low N-cadherin score of expression and good prognostic parameters as low tumor grade and normal adjacent non neoplastic liver (p value = 0.022 and 0.02 respectively) (Table 5) and (Plate 1).

Association between Her2 neu score of expression and N cadherin expression in the studied HCC cases: There was a highly statistical significant association between high score and strong intensity of expression of Her 2 neu and low score and mild intensity of expression of N-cadherin (P value = 0.000 for both) (Table 6).

Overall survival

When revising the patients' files for HCC overall survival time was available for 28 out of 40 (70%) patients. The range of survival time was 1 to 36 months with 23.393± 11.318 as mean ± SD of months and a median of 27.5 months.

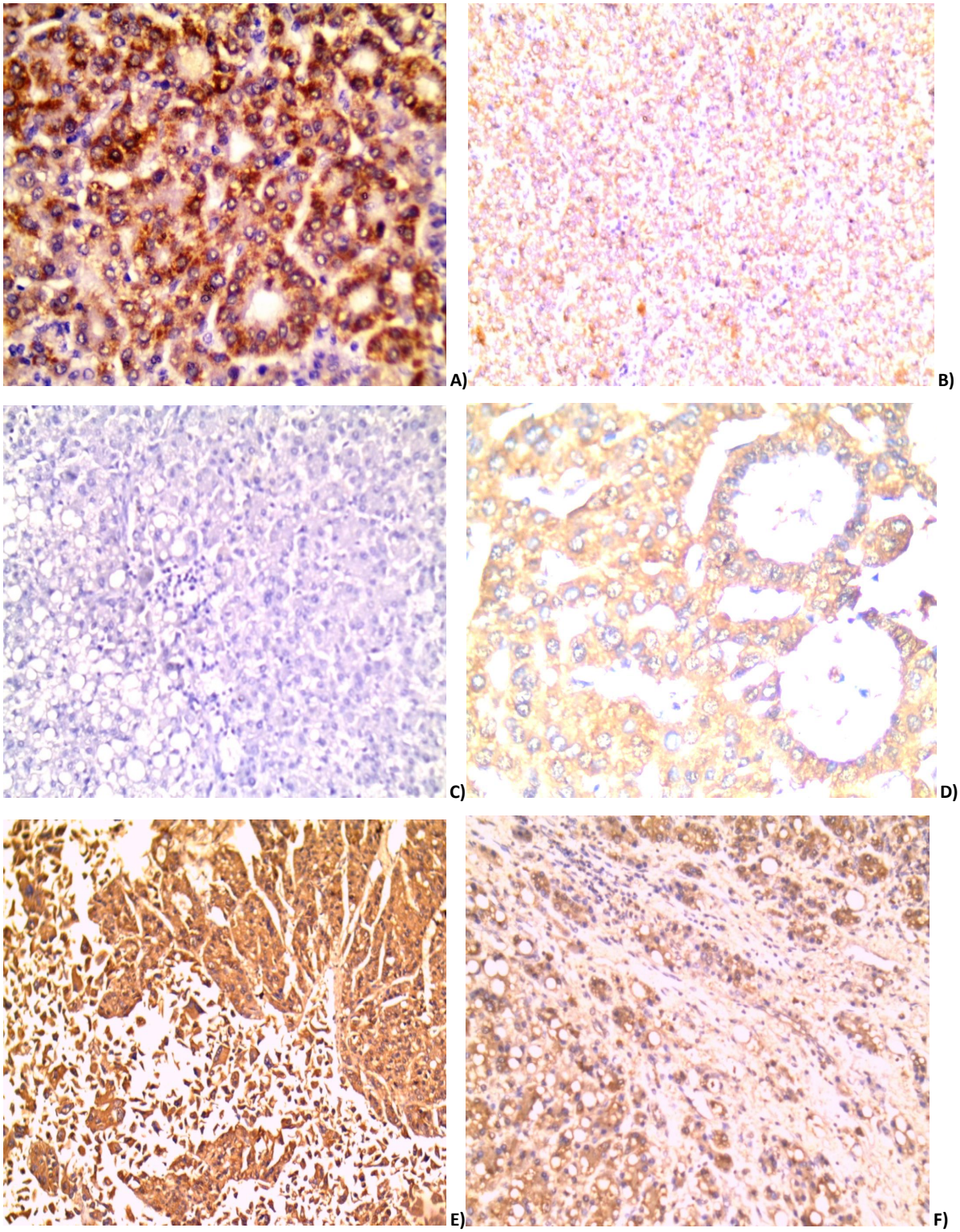

Plate1. Immunohistochemical (IHC) expression of Her 2 neu and n-Cadherin in hepatocellular carcinoma (HCC) cases.(A) Strong Her 2 neu expression in a case of moderately differentiated HCC (IHCx200). (B) Mild Her 2 neu expression in a case of poorly differentiated HCC (IHCx200). (C) Negative Her 2 neu expression in a case of poorly differentiated HCC (IHCx200). (D) Mild n-Cadherin expression in a case of moderately differentiated HCC (IHCx200). (E) Strong n-Cadherin expression in a case of poorly differentiated HCC (IHCx200). (F) Strong n-Cadherin expression in a case of poorly differentiated HCC.

Table 1. Clinicopathological data of the studied HCC cases.

Variable	Hepatocellular carcinoma	
	No	%
Age		
Mean±SD	57.3±5.75	
Median	58	
Range	45-70	
Gender		
Male	31	77.5
Female	9	22.5
M/F	3.4:1	
AFP(ng/ml)		
Mean±SD	428.595 ±216.115	
Median	28.8	
Range	3-2000	
HCV (39)		
Positive	38	95
Negative	1	5
Tumor focality		
Single	33	82.5
Multiple	7	17.5
Tumor site		
Right lobe	21	52.5
Left lobe	19	47.5
Tumor size		
Mean±SD	5.775±3.561	
Median	5	
Range	2-17	
Adjacent non-neoplastic liver (36)		
Cirrhotic	30	75
Chronic hepatitis	5	12.5
Normal liver	1	2.5
pattern of HCC		
Trabecular	10	25
Acinar	3	7.5
Solid	11	27.5
Acinar and trabecular	16	40
Parenchymal margin (Edge of excision)		
Uninvolved by tumor	30	75
Involved by tumor	10	25
LV invasion		
Present	20	50
Absent	20	50
Tumor grade		
I	6	15
II	23	57.5
III	10	25
IV	1	2.5
Tumor grade		
Low grade	29	72.5
High grade	11	27.5
Stage of HCC		
T1	15	37.5
T2	23	57.5
T3	1	2.5
T4	1	2.5
Regional LN		
Involved	1	2.5
Uninvolved	39	97.5
Hep Par1 distribution		
Diffuse	33	82.5
Focal	7	17.5

No= Number, SD=Standard deviation, M: F= Male to female ratio, AFP= Alpha fetoprotein, HCC= Hepatocellular carcinoma

Table 2. Immunohistochemical results of N-cadherin and Her 2 neu in the studied HCC cases.

Variables	No	%
N- cadherin intensity		
Mild	7	17.5
Moderate	23	57.5
Strong	10	25
N-cadherin expression		
Low	19	47.5
High	21	52.5
N-cadherin H score		
Mean±SD	148±58.14	
Median	160	
Range	40-240	
Her 2 neu expression		
Negative	12	30
+ (mild)	7	17.5
++ (moderate)	10	25
+++ (strong)	11	27.5
Her 2 neu percentage		
< 5%	12	30
26-50%	5	12.5
51-75%	11	27.5
76-100%	12	30
Her 2 neu score of expression		
Low expression		
High expression	19	47.5
Her 2 neu H score	21	52.5
Mean±SD	107.25±91.20	
Median	130	
Range	0-240	

Univariate survival analysis for HCC cases

There was no statistically significant association between overall survival and any of the clinicopathological parameters but the data is not tabulated.

DISCUSSION

Cytoplasmic HER2 staining was observed in various types of neoplasms such as thyroid neoplasm (Wu et al., 2013), pancreatic carcinoma (Shibata et al., 2015), adrenal tumors (Saeger et al., 2002) and prostatic cancer. Circumstantially, it seems that tumors arising from endocrine organs are likely to have cytoplasmic HER2 expression (Horiguchi et al., 2010). Immunostaining expression of Her 2 neu protein in HCC tissues in most of the articles showed negative expression (Xian et al., 2005 and Döring et al., 2021).

The present study showed that 70% (28/40) of HCC resected tissues showed cytoplasmic overexpression of Her 2 neu in contrast to Her 2 neu expression in breast tissue, however, Horiguchi et al., (2010) found that cytoplasmic HER2 reactivity characterized by granular cytoplasmic staining and mosaic pattern distribution was closely correlated with neuroendocrine differentiation of breast carcinoma Horiguchi et al., (2010). These results were near that of Shi et al., (2019) who detected membranous and cytoplasmic Her 2 neu overexpression in 82% of HCC cases but these results come in contrary with Xian et al., (2005) who found positivity in 21 (2.42%) of primary HCCs by immunohistochemistry and one case only showed gene amplification by FISH and this might be explained by difference in sample size and in the used technique.

Regarding N-cadherin expression, the present study showed that all HCC cases were positive for N-cadherin and this result was near that of Tajima et al., (2010) who reported that 14 out of 15 (93.3%) of HCC cases were positive for N-cadherin.

The current study showed significant association between high Her2 neu score and strong intensity of expression and good prognostic parameters such as absence of lymphovascular invasion, low tumor grade and trabecular HCC pattern. These results are in line with Shi et al., (2019) who found inverse association between Her2 neu expression and HCC grade. This results may explain the increases in the growth and invasion of the tumor through increasing heterodimerization with other members of EGF receptor and β-catenin/SMAD3 pathways.

On the other hand, the results in the present study disagree with Xian et al., (2005) who could not find any significant associations between Her-2 neu expression and the clinicopathological parameters and find also that there is no role of anti-Her-2 neu trastuzumab in the treatment of HCC patients and this may be explained by difference in sample size as this study contained 868 primary HCCs while the present study contained 40 cases only. Also disagree with Zhang et al., (2010) who found that the patients positive for Her2 neu in the HCC tissues have generally poor prognosis.

Table 3. The relationship between Her2neu score of expression and clinicopathological data in the studied HCC cases

Variables	Her 2 score				Test	P-value
	Low ≤4 (No=19)		High>4 (No=21)			
	No=	%	No=	%		
Age						
Mean±SD	59.88±7.511		57.07±6.603		U= 234.00	0.7
Median	59.5		58			
Range	47-58		36-70			
Gender						
Male	15	48.4	16	51.6	X^2= 0.043	0.56
Female	4	44.4	5	66.6		
AFP(ng/ml)						
Mean±SD	200.62±367.48		324±493.3		U= 86.500	0.66
Median	12		57.65			
Range	7-2000		23-1565			
HCV (39)						
Positive	17	44.7	21	55.3	FE=2.3	0.31
Negative	1	100	**0**	**0**		
Tumor focality						
Single	17	51.5	16	48.5	FE = 1.22	0.24
Multiple	2	28.6	5	71.4		
Tumor site						
Right lobe	8	38.1	13	61.9	FE= 1.57	0.17
Left lobe	11	57.9	8	42.1		
Tumor size						
Mean±SD	8.69±5.62		4.81 ±2.15		U=17.09	0.3
Median	7.25		4.5			
Range	3-17		1.3-13			
Adjacent non-neoplastic liver						
Cirrhotic	14	46.7	16	53.3	x^2 = 3.84	0.28
Chronic hepatitis	4	80	1	20		
Normal liver	0	0	1	100		
pattern of HCC						
Trabecular	3	30	7	70		
Acinar	2	66.7	1	33.3	x^2 = 5.12	0.163
Solid	8	72.7	3	27.3		
Acinar and trabecular	6	37.5	10	62.5		
Parenchymal margin (Edge of excision)						
Uninvolved	17	56.7	13	43.3	FE=4.043	0.048*
Involved	2	20	8	80		
LV invasion						
Present	13	65	7	35	FE=4.91	0.028*
Absent	6	30	14	70		
Tumor grade						
I	0	0	6	100		
II	11	47.8	12	52.2	x^2 = 8.56	0.036*
III	7	70	3	30		
IV	1	100	0	0		
Tumor grade						
Low grade (I,II)	11	37.9	18	62.1	FE=3.87	0.049*
High grade (III, IV)	8	72.7	3	27.3		
Stage of HCC						
T1	5	33.3	10	66.7		
T2	13	56.5	10	43.5	x^2 = 3.96	0.26
T3	0	0	1	100		
T4	1	100	0	0		
Regional LN						
Involved	1	100	0	0	FE=1.13	0.47
Uninvolved	18	46.2	21	53.8		
Hep Par1 distribution						
Diffuse	13	39.4	20	60.6	FE=7.8	0.033*
Focal	6	85.7	1	14.3		
N cadherin score						
Low	2	10.5	17	89.5	FE=19.83	0.000*
High	17	81	4	19		

No=Number, SD=Standard deviation, AFP=Alpha fetoprotein, HCC= Hepatocellular carcinoma, LV= lympho-vascular invasion, HepPar1= Hepatocyte Paraffin1, FE=Fisher's Exact, U=Mann-Whitney, x^2=Chi-Square, * =Significant.

Table 4. The relationship between Her2neu intensity of expression and clinicopathological data in the studied HCC cases

Variables	Her 2 intensity of expression								Test	P-value
	Negative (No=12)		Mild (No=7)		Moderate (No=10)		Strong (No=11)			
	No=	%	No=	%	No=	%	No=	%		
Age										
Mean±SD	39.82±6.23		58.52±9.24		60.22±12.27		46.82±19.22		U= 40.00	0.76
Median	55		61		56		58			
Range	49-68		45-67		50-65		53-67			
Gender										
Male	10	32.3	5	16.1	8	25.8	8	25.8	X^2= 0.562	0.90
Female	2	22.2	2	22.2	2	22.2	3	33.3		
AFP(ng/ml)										
Mean±SD	316.12±425.53		256.15±537.57		386.19±739.47				U= 86.500	0.65
Median	28.8		228.7		629.5					
Range	398-1456		456-1578		586-1348					
HCV (39)										
Positive	10	26.3	7	18.4	10	26.3	11	28.9	X^2=2.3	0.31
Negative	1	100	0	0	0	0	0	0		
Tumor focality										
Single	11	33.3	6	18.2	6	18.2	10	30.3	X^2 = 4.79	0.18
Multiple	1	14.3	1	14.3	4	57.1	1	14.3		
Tumor site										
Right lobe	6	28.6	2	9.5	7	33.3	6	28.6	X^2= 2.88	0.41
Left lobe	6	31.6	5	26.3	3	15.8	5	26.3		
Tumor size										
Mean±SD	5.72±4.66		6.73±4.57		7.85±4.66		6.34±3.52		U=65.54	0.45
Median	7		9		8		10			
Range	4-13		5-12		7-11		3-15			
Adjacent non-neoplastic liver										
Cirrhotic	9	30	5	16.7	8	26.7	8	26.7	x^2 = 7.19	0.6
Chronic hepatitis	2	40	2	40	1	20	0	0		
Normal liver	0	0	0	0	0	0	1	100		
pattern of HCC										
Trabecular	1	10	2	20	1	10	6	60		
Acinar	1	33.3	1	33.3	0	0	1	33.3	x^2 = 19.9	0.02*
Solid	7	63.6	1	9.1	1	9.1	2	18.2		
Acinar and trabecular	2	18.8	3	18.8	8	50	2	12.5		
Parenchymal margin (Edge of excision)										
Uninvolved	10	33.3	7	23.3	8	26.7	5	16.7	x^2 =8.03	0.045*
Involved	2	20	0	0	2	20	6	60		
LV invasion										
Present	9	45	4	2	5	25	2	10	x^2=7.59	0.049*
Absent	3	15	3	15	5	25	9	45		
Tumor grade										
I	0	0	0	0	1	16.7	5	83.3		
II	5	21.7	6	2.1	8	34.8	4	17.4	x^2 = 19.8	0.02*
III	6	60	1	10	1	10	2	20		
IV	1	100	0	0	0	0	0	0		
Tumor grade										
Low grade	7	63.6	1	9.1	1	9.1	2	18.2	x^2 =8.35	0.03*
High grade	5	17.2	6	20.7	9	31	9	31		
Stage of HCC										
T1	3	20	2	13.3	3	20	7	46.7		
T2	9	39.1	4	17.4	7	30.4	3	13	x^2 = 12.8	0.17
T3	0	0	0	0	0	0	1	100		
T4	0	0	1	100	0	0	0	0		
Regional LN										
Involved	1	100	0	0	0	0	0	0	x^2 =2.39	0.49
Uninvolved	11	28.2	7	17.9	10	25.6	11	28.2		
Hep Par1 distribution										
Diffuse	7	20	7	20	10	28.6	11	31.4	x^2 =13.3	0.03*
Focal	3	100	0	0	0	0	0	0		

No=Number, SD=Standard deviation, AFP=Alpha fetoprotein, HCC= Hepatocellular carcinoma, LV= lympho-vascular invasion, HepPar1= Hepatocyte Paraffin1, FE=Fisher's Exact, U=Mann-Whitney, x^2=Chi-Squares, * =Significant.

Table 5. The relationship between N-cadherin score of expression and clinicopathological data in the studied HCC cases

Variables	N cadherin score				Test	P-value
	Low ≤ 3 (No=19)		High > 3 (No=21)			
	No=	%	No=	%		
Age						
Mean±SD	54.3±8.75		67.17±8.61		U= 234.00	0.7
Median	53		59			
Range	55-69		50-67			
Gender						
Male	14	45.2	17	54.8	FE= 0.301	0.43
Female	5	55.6	4	44.4		
AFP(ng/ml)						
Mean±SD	416.18±828.54		368.24±535.3		U= 86.500	0.66
Median	528.8		227.65			
Range	367-1345		123-1497			
HCV (39)						
Positive	18	47.4	20	52.6	FE=2.3	0.31
Negative	0	0	**1**	100		
Tumor focality						
Single	15	45.5	18	54.5	FE = 0.316	0.44
Multiple	4	57.1	3	42.9		
Tumor site						
Right lobe	10	47.6	11	52.4	FE= 000	0.6
Left lobe	9	47.4	10	52.6		
Tumor size						
Mean±SD	6.15±4.54		7.81 ±2.15		U=17.09	0.3
Median	6		7.5			
Range	5-16		4-13			
Adjacent non-neoplastic liver						
Cirrhotic	15	50	15	50	x^2 = 6.92	**0.02***
Chronic hepatitis	0	0	5	100		
Normal liver	1	100	0	0		
pattern of HCC						
Trabecular	6	60	4	40		
Acinar	2	66.7	1	33.3	x^2 = 1.70	0.63
Solid	4	36.4	7	63.6		
Acinar and trabecular	7	43.8	9	56.2		
Parenchymal margin (Edge of excision)						
Uninvolved by tumor	13	43.3	7	56.7		
Involved by tumor	6	60	4	40	FE=0.835	0.29
LV invasion						
Present	7	35	13	65	FE=2.50	0.10
Absent	12	60	8	40		
Tumor grade						
I	6	100	0	0		
II	9	39.1	14	60.9	x^2 = 9.611	**0.022***
III	3	30	7	70		
IV	1	100	0	0		
Tumor grade						
high grade	4	36.4	7	63.6	FE=0.755	0.30
low grade	15	51.7	14	48.3		
Stage of HCC						
T1	9	60	6	40		
T2	9	39.1	14	60.9	x^2 = 0.356	0.30
T3	1	100	0	0		
T4	0	0	1	100		
Regional LN						
Involved	0	0	1	100	FE=0.918	0.52
Uninvolved	19	48.7	21	51.3		
Hep Par1 distribution						
Diffuse	17	51.5	16	48.5	FE=1.219	0.24
Focal	2	28.6	5	71.4		

No=Number, SD=Standard deviation, AFP=Alpha fetoprotein, HCC= Hepatocellular carcinoma, LV= lympho-vascular invasion, HepPar1= Hepatocyte Paraffin1, FE=Fisher's Exact, U=Mann-Whitney, x^2=Chi-Square,* =Significant.

Table 6. Association between Her 2 neu score of expression and n- Cadherin expression in the studied HCC cases.

	Her 2 neu score				Kappa Test	P-value
	Low (No=19)		High (No=21)			
	No=	%=	No=	%=		
N cadherin score						
Low	2	10.5	17	89.5	19.83	0.000**
High	17	81	4	19		
N cadherin intensity						
mild	1	14.3	6	85.7	15.64	0.000**
moderate	8	34.8	15	65.2		
sever	10	100	0	0		

No= Number, x^2=Chi-Square , ** = Highly Significant.

Moreover, our results disagree with Jørgensen and Hersom, (2012) who reported that Her2 neu is a negative prognostics factor in gastric cancer and associated with clinicopathological factor of poor prognosis. Ourresults contrast thoses of Konecny et al., (2004) who found association between poor prognosis (advanced stage, poor histological differentiation, and resistance to drug treatment) and Her2 neu overexpression in breast cancer and Foker et al., (2016) who also reported poor prognostic effect Her 2 neu positivity on breast carcinoma. This contraversry might be explained by difference in tissues used in both papers. Our results, however, did not show significant correlation between high Her2 neu score of expression and low pathological stage. Our results are in line with those of Shi et al., (2019) who found that tumor stage was negatively correlated with Her2 neu expression.

The current study showed statistical significant association between low N-cadherin score of expression and good prognostic parameters such as low tumor grade and normal adjacent non-neoplastic liver. These results are in line with Hazan et al., (2004) who reported that increased N-cadherin is associated with poor prognosis. As switch in cadherin from E- to N-cadherin activates and promotes the invasive and survival abilities of tumor cells. It also enhances the interaction between tumor cells and the surrounding microenvironment which is critical in metastasis process metastasis (Hazan et al., 2004). Ourresults are in contrast with Liu et al., (2015) who reported that absence of expression of both N- and E-cadherin associates with poor prognosis in HCC patients. The results in the present study agree with those of Zhou et al., (2017) who reported that overexpression of N-cadherin significantly associates with poor prognostic parameters of HCC.

The results of the current study showed a highly statistical significant association between high score and strong intensity of expression of Her 2 neu and low score and mild intensity of expression of N-cadherin. These results agree with those of Nami et al., (2020) who detected negative correlation between EMT markers and Her 2 neu expression in breast carcinoma. Our results are also in line with those of Nilsson et al., (2014) who reported that Her2 neu signaling in human breast epithelial cells leads to decreased EMT markers expression as vimentin and N-cadherin. In contrast, our results disagree with those of Nami et al., (2017) who showed that overexpression of Her 2 neu induces EMT and maintains the mesenchymal phenotype in breast cancer cells. They this correlation as a result of the ability of Her2 neu receptor to stimulate the stemness signaling pathways towards EMT in mammary epithelial cells. The contrast of this results to the current results could be explained by the difference in the used techniques as they used breast cancer cell line and we used immunohistochemistry tecnique (Nami and Wang, 2017).

The current study showed that the overall survival is not associated with any of the clinicopathological parameters or any of the used markers including Her 2 neu and N-Cadherin. These results agree with those of Xian et al., (2005) who reported that no significant associations between Her-2 neu overexpression and any of clinicopathological parameters including survival. However, these

results disagree with those of Zhou et al., (2017) and Liu et al., (2015) who reported that Her-2 neu overexpression associates with poor survival of the patients

The survival results of the present study are in contrast to those of Hazan et al., (2004) who reported increases in N-cadherin which was associated with poor prognosis and metastatic progression. It lso contrats the results of et al., (2015) who reported loss of both N- and E-cadherin expressions which was associated with poor surgical outcomes of HCC patients.

Hepatocellular carcinoma still has poor prognosis inspite of new modalities in surgical techniques and early diagnosis and the poor patients' survival even after resection because of the high incidence of recurrence so new treatments are needed to improve this poor prognosis. Given that trastuzumab is used for treatment of breast cancer patients who show overexpression of Her-2 neu (Nahta et al., 2004), our results indicate that this treatment which is anti Her-2 neu might not be useful for HCC patients since we found correlation between Her-2 neu overexpression and good prognostic factors . As such, new treatments are needed to increase the expression of Her-2 as well as its function (Her-2 neu agonist). However, the feasibility of our suggestion requires further wide scaled studies on Her2 neu expression and function in HCC.

CONCLUSIONS

The present study indicates significant association between high Her2 neu expression and good prognostic parameters such as absence of lymphovascular invasion, low grade and low stage. Therefore, the anti-Her-2 target therapy might not be valuable for HCC patients. also larger studies are recommended to discover potential targets for Her 2 enhancer or promoter and to understand the exact biological role of Her-2 neu in HCC.

CONFLICT OF INTEREST

All authors declared no conflicts of interest.

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