

Immunohistochemical expression of CDX2, CK7, HER2 and HER4 in periampullary adenocarcinoma : implications for clinicopathology and patient outcomes

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Abstract

Background : Periampullary carcinomas originate from the pancreatic head, the ampulla, the distal bile duct, or the duodenum. The expression of CK7 and CDX2 has been used in the classification of periampullary carcinomas. There is prognostic value of human epidermal growth factor receptor (HER) 2 and HER 4, which have been linked to poor prognosis in several types of tumors, such as breast and gastric carcinomas. We aimed to evaluate the expression and prognostic value of CDX2, CK7, HER 2, and HER 4 in periampullary adenocarcinoma.

Patients and Methods : We retrospectively selected 98 patients who had undergone pancreatoduodenectomy for periampullary adenocarcinoma at our pathology department. The tumor location, pathological subtype, involvement of vessels and lymph nodes, perineural invasion, clinical follow-up, and tumorstage were noted. Immunohistochemistry was performed for CK7, CDX2, HER2, and HER4.

Results : CDX2 staining was predictive of perineural invasion. Additionally, there was a significant association between the overexpression of HER2 and HER4 and the presence of perineural invasion. HER4 was significantly positive in patients with the pancreatobiliary subtype compared with patients with the intestinal subtype. Patients with the pancreatobiliary subtype, lymph node involvement, and advanced pT and UICC stages had significantly lower median survival.

Conclusion : Our findings suggest that only pancreatobiliary subtype, lymph node involvement and advanced pT and UICC stages were independent predictors of short survival, but the ampulla tumor location predicted a significantly better survival time. The immunohistochemical expression of CDX2, CK7, HER4, and HER2, vessel involvement, and perineural invasion were not associated with the survival of patients with periampullary adenocarcinoma. (*Acta gastroenterol. belg.*, 2017, 80, 31-37).

Key words : CDX2, CK7, HER2, HER4, immunohistochemistry, periampullary adenocarcinoma.

Introduction

Periampullary carcinomas originate from the pancreatic head, ampulla, distal bile duct, or the duodenum, and the incidence of these carcinomas has gradually increased (1,2). Based on its tissue of origin and histopathological differentiation, periampullary carcinoma has traditionally been divided into intestinal and pancreatobiliary subtypes, of which the latter subtype shows less cellular differentiation, more local and distant invasion, and a worse prognosis (2,3).

Currently, surgery (pancreaticoduodenectomy) remains the only treatment modality for periampullary tumors. However, the recurrence rate is greater than 50% after surgery, indicating the need for effective adjuvant

therapy (1,4). Current evidence indicates that the optimal duration of adjuvant therapy and the extent of surgery should be based on the evaluation of prognostic factors including histopathological differentiation, gene expression profile, and immunohistochemical markers (1,5). Although the role of immunohistochemistry, i.e., expression of cytokeratin (CK) 7, CK20, caudal type homeobox 2 (CDX2), mucin (MUC) 1 and 2, etc., in histological subtyping and the differential diagnosis of periampullary carcinomas is well defined (6-12), studies of the association of immunohistochemical markers with the outcome of disease are limited (13,14). Furthermore, the prognostic value of human epidermal growth factor receptor 2 (HER2) and HER4, which have been linked to poor prognosis in several types of tumors, such as breast, lung, and gastric carcinomas, has not been extensively studied.

Standard and reproducible methods should be implemented to estimate the prognosis of patients with periampullary adenocarcinoma for planning the management of the disease. The expression of different proteins that play a role in the pathogenesis of periampullary adenocarcinoma should also be determined to identify new molecular therapeutic targets.

In this study, we aimed to evaluate the expression and predictive value of immunohistochemical markers and the expression of CDX2, CK7, HER2, and HER4 on the outcome of patients with periampullary adenocarcinoma.

Materials and Methods

Study design and population

Patients who had undergone pancreatoduodenectomy (Whipple procedure) for histologically confirmed periampullary adenocarcinoma at our pathology department between January 2005 and December 2010 were included in this study.

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Submission date : 18/02/2016
Acceptance date : 24/10/2016

Acta Gastro-Enterologica Belgica, Vol. LXXX, January-March 2017

This project was performed with the assistance of our Scientific Research Unit (No. KUAP (T)-2013/3) and was approved by the Institutional Ethics Committee (No. B.30.2.ULU.0.20.70.02-050.99/12). The study was conducted in accordance to the current version of the Helsinki Declaration, and all patients gave written informed consent before participation.

Histopathologic assessment and immunohistochemical histopathologic staining of specimens

The surgical specimens were evaluated in our department of pathology. Pathological subtypes were classified as described previously by Kimura et al. (3). Intestinal-type adenocarcinomas, similar to colorectal adenocarcinomas, were characterized by well-formed tubular or cribriform glands with tall, pseudostratified columnar cells that have oval nuclei located at the basal surface. Pancreatobiliary-type adenocarcinomas were characterized by simple or complex branching glands composed of low columnar epithelium with a single layer and rounded nuclei, set in abnormal desmoplastic stroma (Figure 1). The histologic parameters, including the involvement of vessels and lymph nodes, perineural invasion, pathological tumor (pT) stage, and the Union for International Cancer Control (UICC) stage were noted. All specimens were fixed in formalin and embedded in paraffin. The samples were cut into 4µm thick serial sections, one of which was stained with hematoxylin and eosin, and the others were used for immunohistochemistry.

Immunohistochemistry was carried out for a panel of markers, including CK7, CDX2, HER2, and HER4, using

a Leica Bond Max automated slide staining system (Leica, Bannockburn, IL). The primary antibodies used were NCL-L-CK7-560 (Novocastra, UK; 1:500 dilution) against CK7; NCL-CDX2 (Novocastra, UK; 1:100 dilution) against CDX2; P04626 (Abbotec, USA; 1:400 dilution) against HER2; and Q15303 (Abbotec, USA; 1:400 dilution) against HER4. The sections were counter-stained with hematoxylin.

For CK7, staining of the cytoplasm, the cell membrane, or both was considered positive. CDX2 expression with nuclear and strong staining was regarded as positive. Membrane staining was evaluated for HER2. Staining of HER2 was classified into four categories (0, no reactivity or membranous reactivity in less than 10% of tumor cells; 1+, faint or barely visible membranous reactivity in more than 10% of cells; 2+, weak to moderate complete or basolateral membranous reactivity in more than 10% of cells; and 3+, moderate to strong complete or basolateral membranous reactivity in more than 10% of cells) according to the immunohistochemistry (IHC) scoring system for gastric tumors created by Hofmann et al. (15). Cytoplasmic staining was also evaluated for HER4. Positive staining for HER4 was considered when at least 10% or more of tumor cells were stained, and cytoplasmic staining intensity was scored and graded from 0 to 3 (16). We considered only IHC 3+ staining as overexpression of HER2 and HER4 (Figure 2).

Statistical analysis

The study data were summarized using descriptive statistics (mean, standard deviation, frequency, or percentage). Groups of tumor specimens with expression

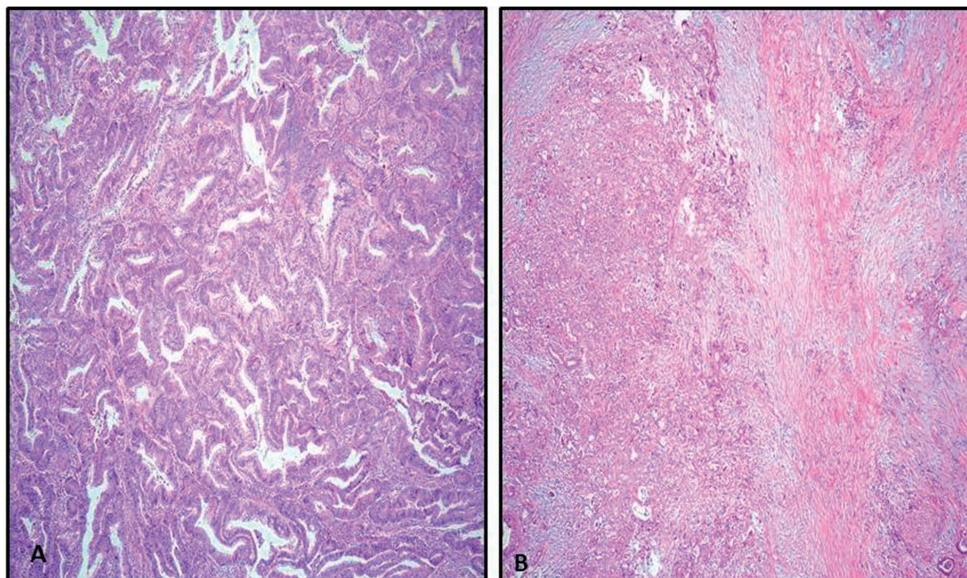


Fig. 1. — A) Adenocarcinoma of the intestinal type with well-formed tubular or cribriform glands with tall, pseudostratified columnar cells with oval nuclei located at the basal surface (H&Ex40) B) Adenocarcinoma of the pancreatobiliary type with simple or complex branching glands composed of low columnar epithelium with a single layer and rounded nuclei, set in abnormal desmoplastic stroma (H&Ex40).

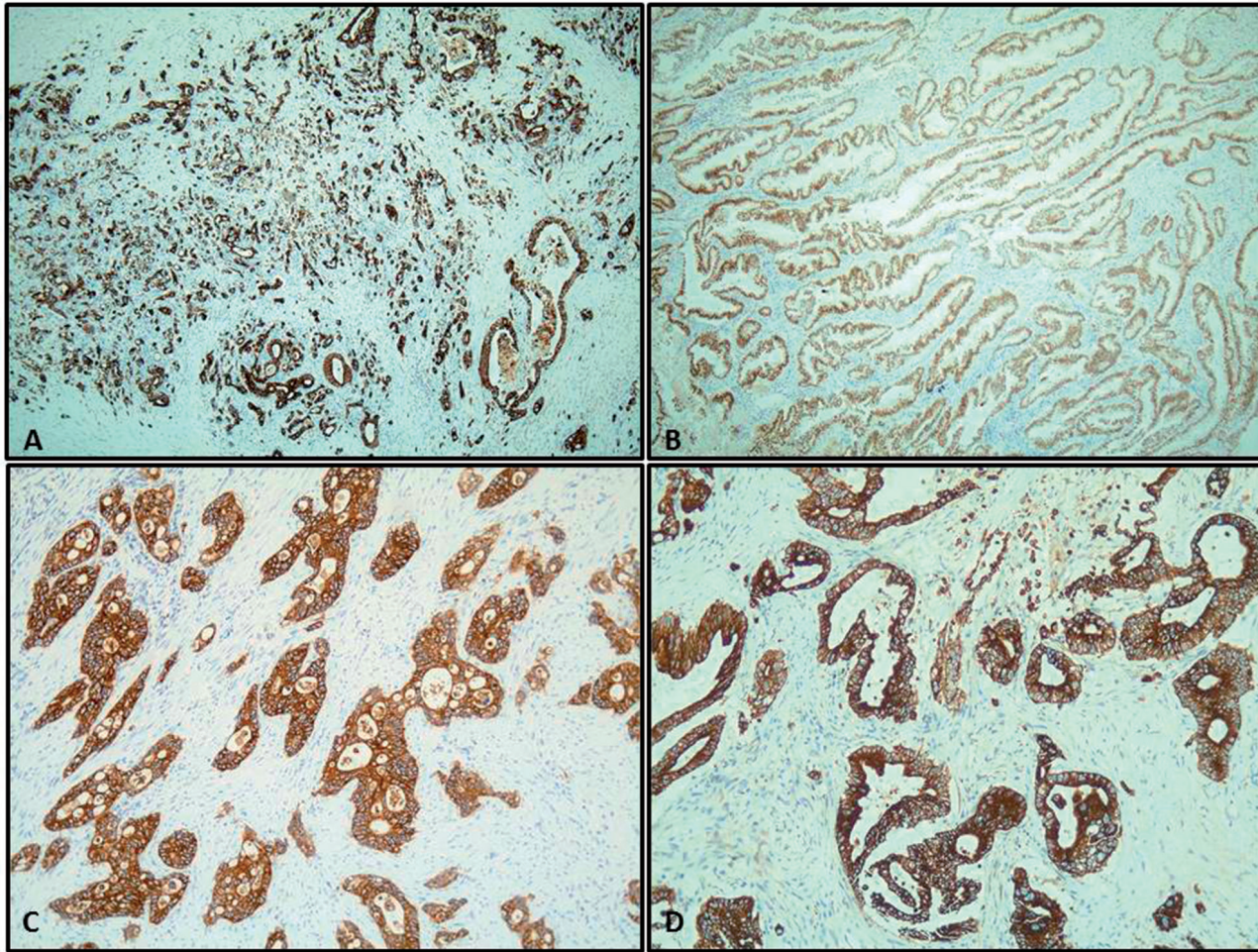


Fig. 2. — A) CK7 immunohistochemical staining is positive along the tumor cell membranes (x40); B) CDX2 nuclear staining positive cells (x40); C) HER2 immunostaining demonstrates strong membranous positive of tumor cells (x100) D) HER4 immunostaining shows strong cytoplasmic positive of the tumor cells (x100)

of CDX2, CK7, HER2, and HER4 were compared using the Kruskal-Wallis test for continuous variables, and Pearson's chi-square test and Fisher's exact test for categorical variables. Kaplan-Meier survival analysis was performed to evaluate the survival time and the effect of immunohistochemical and clinical variables on survival. The overall survival was expressed as the median survival in months with a 95% confidence interval (CI).

Statistical analysis was performed using the SPSS software (StatisticalPackageforSocialSciences, SPSS Inc., Chicago, IL, USA). The level of statistical significance was set to $p < 0.05$.

Results

Demographic and clinicopathological characteristics of the patients

A total of 98 patients (63 males, 35 females; mean age, 63.0 ± 11.6 years) were included in the study. Tumors were located in the duodenum in 12 cases (12.2%), in the distal bile duct in 7 cases (7.1%), in the head of pancreas

in 41 cases (41.8%) and in the ampulla in 38 cases (38.8%). There were 48 (49%) patients with intestinal and 50 (51%) patients with pancreatobiliary histologic types of differentiation. All cases had negative resection margins (circumferential parenchymal pancreas, retroperitoneum, vascular groove, and common bile duct). Vascular invasion was observed in 15 (15.3%) patients, and perineural invasion was present in 51 (52%) patients. Lymph node involvement was present in 37 cases (37.8%).

The rates of expression of CDX2 and CK7 were 80/98 (81.6%) and 34/98 (34.7%), respectively. Among the 98 periampullary adenocarcinomas, 38 (38.8%) cases showed negative immunoreactivity, 21 (21.4%) cases showed weak (IHC1+) positive expression, 21 (21.4%) cases showed intermediate (IHC2+) positive expression, and 18 (18.4%) cases showed overexpression (IHC 3+) of HER2. HER4 protein immunostaining was predominantly located in the cytoplasm. In 98 specimens, HER4 immunostaining results were: grade 0, 50 (51%); grade 1+, 11 (11.3%); grade 2+, 25 (25.5%); and grade 3+, 12 (12.2%).

CK7 positive patients were predominantly female compared to patients with CDX2 and both CDX2 and CK7 positivity (female rates, 55.6%, 35.9%, and 12.5%, respectively; $p=0.031$) (Table 1).

Patients who had positive staining for CDX2 alone and for both CDX2 and CK7 had a significantly higher rate of perineural invasion (57.8% and 62.5%, respectively) than those with pancreatobiliary marker CK7 expression alone ($p=0.019$, Table 1). Tumor location, pathological subtypes, involvement of vessels and lymph nodes, expression of HER2 or HER4, and pT and UICC stages did not significantly differ with CDX2 and CK7 positivity (Table 1).

We next examined the effects of HER2 and HER4 positivity on demographics, clinical parameters and the extent of the disease and found that there was a significant association between HER4 expression and the presence of perineural invasion ($p=0.039$). When the subgroup analysis was performed significant differences were observed in the comparison between HER4 overexpression and the negative subgroups ($p=0.007$). Additionally, HER4 was significantly positive in patients

with the pancreatobiliary subtype compared with patients with the intestinal subtype ($p=0.026$). Significant results were observed in the comparison between HER4 IHC2+ and the negative subgroups ($p=0.009$). Further, borderline significance was detected in the comparison between HER4 overexpression and the negative subgroups ($p=0.053$).

Furthermore, HER2 was significantly negative in the intestinal subtype, and the pattern of overexpression of HER2 was significantly associated with the pancreatobiliary type of differentiation ($p=0.004$). Moreover, overexpression of HER2 was highly associated with tumor location ($p=0.001$). In pancreatic head tumors, HER2 overexpression was observed 14 (77.8%) cases compared to in 2 (11.1%) cases of ampulla tumors ($p=0.009$). In addition, the HER2 negative subgroup was significantly associated with the ampulla location ($p=0.039$). Other parameters related to the extent of the disease (i.e., vessel and lymph node involvement, pT and UICC stages) did not show significant associations with HER2 or HER4 expression (Table 2).

Table 1. — Demographic and clinicopathological characteristics of patients with respect to CDX2 and/or CK7 expression

		CDX2 (n=64)	CK7 (n=18)	CDX2+/CK7+ (n=16)	Total (n=98)	P
Gender	Male	41(64.1%)	8 (44.4%)	14 (87.5%)	63(64.3%)	0.031
	Female	23(35.9%)	10 (55.6%)	2 (12.5%)	35(35.7%)	
Age (years)		64.1±11.8	60.4±12.9	61.6±9.2	63.0±11.6	0.25
Location	Duodenum	7(10.9%)	2(11.2%)	3(18.8%)	17(7.1%)	0.302
	Distal bile duct	4(6.3%)	2(11.1%)	1(6.3%)	2(12.2%)	
	Pancreatic head	32(50%)	4(22.2%)	5(31.3%)	41(41.8%)	
	Ampulla	21(32.8%)	10(55.6%)	7(43.8%)	38(38.8%)	
Pathological subtypes	Intestinal	29(45.3%)	11(61.1%)	8(50%)	48 (49%)	0.509
	Pancreatobiliary	35(54.7%)	7(38.7%)	8(50%)	50 (51%)	
Vessel involvement		10(15.6%)	1 (6.5%)	4 (25.0%)	15(15.3%)	0.312
Lymph node involvement		23(35.9%)	9 (50.0%)	5 (31.3%)	37(37.8%)	0.442
Perineural invasion		37(57.8%)	4 (22.2%)	10 (62.5%)	51(52.0%)	0.019
HER2 expression	0	23(35.9%)	8(44.4%)	7(43.8%)	38(38.7%)	0.958
	1+	16(25%)	3(16.7%)	2(12.5%)	21(21.5%)	
	2+	13(20.3%)	4(22.2%)	4 (25%)	21(21.5%)	
	3+	12(18.8%)	3 (16.7%)	3 (18.8%)	18(18.3%)	
HER4 expression	0	31(62%)	11(22%)	8 (16%)	50(51%)	0.826
	1+	6(54.5%)	2(18.2%)	3(27.3%)	11(11.3%)	
	2+	17(68%)	4(16%)	4(16%)	25(25.5%)	
	3+	10(83.3%)	1(8.3%)	1(8.3%)	12(12.2%)	
pT stage	pT2	19(29.7%)	6 (33.3%)	7 (43.8%)	32(32.7%)	0.882
	pT3	36(56.3%)	10 (55.6%)	7 (43.8%)	53(54.1%)	
	pT4	9 (14.1%)	2 (11.1%)	2 (12.5%)	13(13.3%)	
UICC stage	1B	13(20.3%)	4 (22.2%)	6 (37.5%)	23(23.5%)	0.799
	2A	23(35.9%)	5 (27.8%)	5 (31.3%)	33(33.7%)	
	2B	19(29.7%)	7 (38.9%)	3 (18.8%)	29(29.6%)	
	3	9 (14.1%)	2 (11.1%)	2 (12.5%)	13(13.3%)	

Table 2. — Demographic and clinicopathological characteristics of patients with respect to HER2 and HER4 expression

		HER2 expression				p	HER4 expression				
		0 (n=38)	1+ (n=21)	2+ (n=21)	3+ (n=18)		0 (n=50)	1+ (n=11)	2+(n=25)	3+ (n=12)	p
Gender	Male	26 (41.3)	12 (19)	16 (25.4)	9 (14.3)	0.300	31 (62)	7(63.6)	19(76)	6(50.0)	0.430
	Female	12 (34.3)	9 (25.7)	5 (14.3)	9 (25.7)		19 (38)	4 (36.4)	6(24)	6(50.0)	
Location	Duodenum	5(13.2)	2(9.1)	3(15)	2(11.1)	0.001	4(8)	2(18.2)	4(16)	2(16.7)	0.12
	Distal bile duct	1(2.6)	3(13.6)	3(15)	0(0)		4(8)	2(18.2)	1(4)	0(0)	
	Pancreatic head	9(23.7)	8(36.4)	10(50)	14(77.8)		16(32)	4(36.4)	13(52)	8(66.7)	
	Ampulla	23(60.5)	9(40.9)	4(20)	2(11.1)		26(52)	3(27.3)	7(28)	2(16.7)	
Pathological Subtypes	Intestinal	27 (56.2)	8 (16.7)	9 (18.8)	4 (8.3)	0.004	32 (64)	4 (36.4)	8(32)	4(33.3)	0.026
	Pancreato biliary	11 (22)	13 (26)	12 (24)	14 (28)		18 (36)	7 (63.6)	17(68)	8(66.7)	
Vessel involvement	Absent	31 (37.3)	17(20.5)	20(24.1)	15(18.1)	0.502	45 (90)	7(63.6)	21(84)	10(83.3)	0.160
	Present	7 (46.7)	4 (26.7)	1(6.7)	3 (20.0)		5 (10)	4 (36.4)	4(16)	2(16.7)	
Lymph node involvement	Absent	26(42.6)	12 (19.7)	11(18)	12(19.7)	0.600	33 (66)	5(45.5)	17(68)	6(50)	0.447
	Present	12 (32.4)	9 (24.3)	10(27)	6 (16.2)		17(34)	6(54.5)	8(32)	6(50)	
Perineural infiltration	Absent	24 (51.1)	7 (14.9)	8 (17)	8 (17)	0.101	30 (60)	4(36.4)	11(44)	2(16.7)	0.039
	Present	14 (27.5)	14 (27.5)	13 (25.5)	10 (19.6)		20 (40)	7(63.6)	14(56)	10(83.3)	
Pt stage	pT2	18 (56.2)	5 (18.8)	6 (18.8)	3 (9.4)	0.135	19 (38)	4(36.4)	7(28)	2(16.7)	0.866
	pT3	16 (30.2)	11 (20.8)	12 (22.6)	14 (26.4)		25 (50)	6(54.5)	14(56)	8(66.7)	
	pT4	4 (30.8)	5 (38.5)	3 (23.1)	1 (7.7)		6 (12)	1(9.1)	4(16)	2(16.7)	
UICC stage	IB	15 (65.2)	2 (8.7)	3 (13)	3 (13.0)	0.211	15 (30)	2 (18.2)	4(16)	2(16.7)	0.755
	IIA	9 (23.7)	7 (21.2)	8 (27.3)	9 (27.3)		17 (34)	2 (18.2)	10(40)	4(33.3)	
	IIB	10 (34.5)	7 (24.1)	7 (17.2)	5 (17.2)		12 (24)	6 (54.5)	7(28)	4(33.3)	
	III	4 (30.8)	5 (38.5)	3 (23.1)	1 (7.7)		6 (12)	7(9.1)	4(16)	2(16.7)	

Survival analysis

Using Kaplan-Meier analysis, the overall median survival for the entire study population was 25 months (95% CI, 15.1-34.9 months). There was no significant association between CDX2 and/or CK7, HER4, or HER2 expressions and survival time. Additionally, survival was not affected by gender, expression of CDX2 and/or CK7, HER4 expression, HER2 expression, vessel involvement, or perineural invasion in our study (Table 3). However, patients with the pancreatobiliary subtype, lymph node involvement, and advanced pT and UICC stages had a significantly lower survival time ($p=0.001$, $p=0.018$, $p=0.004$, and $p=0.007$, respectively; Table 3). Moreover, the median survival time of the patients with a duodenum tumor location was 94 months ($p=0.030$), which was much longer than in the patients with tumors located at the distal bile duct (median survival time 37 months) or the pancreatic head (median survival time 50 months).

Discussion

In this study, we demonstrated that in periampullary adenocarcinoma, the rates of overexpression of CDX2, CK7, HER2 and HER4 were 81.6%, 34.7%, 18.4%, and 12.2%, respectively. CDX2 and HER4 staining was predictive of perineural invasion. Additionally, we detected that there was a significant association

between HER2 overexpression, HER4 IHC2+ subgroup and pancreatobiliary subtype, but none of the markers we studied had a significant relationship with patient survival.

Together with morphology, the expression of immunohistochemical markers, such as CK7, CK20, transcription factor CDX2, MUC 1 and 2, HepPar-1, and the epithelial growth factor receptor family, have been used in the classification of periampullary and ampullary carcinomas (6-12). Intestinal type periampullary tumors generally show expression of CK20 and MUC2, whereas the pancreatobiliary type expresses CK7 (17). Kumari et al. (6) reported that intestinal and pancreatobiliary types of periampullary carcinomas could be differentiated in 84% of cases by morphology alone and in 88% cases with immunohistochemistry. Among the immunohistochemical markers, CDX2 showed the highest sensitivity and specificity for the intestinal type of periampullary carcinomas (6). In contrast, in our study the immunohistochemical markers CK7 and CDX2 were not useful for determining the histologic subtype. On the basis of previous reports, we studied the immunohistochemical expression of CDX2 and CK7 in periampullary adenocarcinoma and evaluated their predictive value on the outcome of the disease.

Studies on the association of immunohistochemical markers and outcome of disease reveal conflicting results. de Paiva Haddad et al.(14) found no association

Table 3. — Median survival time in months by Kaplan-Meier analysis with respect to demographic and clinicopathological characteristics

		Survival time (months) (95% confidence interval)	P
Gender	Male (n=63)	25 (11.5-38.4)	0.338
	Female(n=35)	22 (9.3-34.7)	
Location	Duodenum	94 (45.4-143.5)	0.030
	Distal bile duct	37 (25.3-49)	
	Pancreatic head	50 (40.9-60.9)	
	Ampulla	57 (46.8-68)	
CDX2 and/or CK7 expression	CDX2(n=64)	24 (14.7-33.3)	0.76
	CK7 (n=18)	30 (0-68.5)	
	CDX2+/CK7+ (n=16)	22 (0-49.7)	
HER4 expression	0 (n=50)	25 (14.9-35.1)	0.642
	1+ (n=11)	26 (21-40.5)	
	2+ (n=25)	19 (8.9 -28.8)	
	3+ (n=12)	23 (0-64.5)	
HER2 expression	0 (n=38)	26 (15.0-36.9)	0.177
	+1 (n=21)	23 (8.3-37.7)	
	+2 (n=21)	17 (7.0-26.9)	
	+3 (n=18)	22 (0-45.6)	
Vessel involvement	Absent (n=83)	25 (15.9-34.1)	0.152
	Present (n=15)	15 (11.3-18.7)	
Lymph node involvement	Absent (n=61)	30 (22.9-37.0)	0.018
	Present (n=37)	14 (10.6-17.4)	
Perineural invasion	Absent (n=47)	28 (21.5-34.5)	0.286
	Present (n=51)	16 (12.2-19.8)	
pT stage	pT2 (n=32)	43 (27.5-58.5)	0.004
	pT3 (n=53)	16 (6.9-25.2)	
	pT4 (n=13)	14 (8.9-19.2)	
UICC stage	IB (n=23)	45 (21.9-68.1)	0.007
	IIA (n=33)	23 (8.3-37.7)	
	IIB (n=29)	15 (10.8-19.2)	
	III (n=13)	14 (8.8-19.2)	
Pathological subtypes	Intestinal (n=48)	30 (22.9-37.0)	0.001
	Pancreatobiliary (n=50)	17 (7.0-26.9)	
Total (n=98)		25 (15.1-34.9)	

between CDX2 expression and outcome, whereas Kumari et al.(6) reported CDX2 was an independent prognostic marker for longer survival of patients with periampullary carcinoma. Zhao et al. (13) showed that the immunohistochemical classification of ampullary carcinomas according to their CK7 and CK20 status along with osteopontin status could be used to estimate prognosis. Furthermore, reduction in the expression of Raf-1 kinase inhibitory protein (18) and an increase in the expression of hepatocyte nuclear factor 4-alpha (5) and cyclin D (19) have been shown to be favorable prognostic factors in periampullary carcinoma originating from the ampulla. Yamamoto et al. (20) evaluated the association of histologic type and the expression of CK20 and/or CDX2 to the location of periampullary carcinomas and concluded that the intestinal type and CK20 positivity indicated non-extension of the tumor into the common duct. On the other hand, de Paiva Haddad et al.(14) reported that although the intestinal type showed better prognosis in univariate analysis, only lymph node metastasis, lymphatic invasion, and stage were independent risk factors for survival in multivariate analysis, but not the histological classification or immunohistochemistry results. Our results were similar

to those of de Paiva Haddad et al.(14), in that patients who had positive staining for CDX2 alone and for both CDX2 and CK7 had a significantly higher rate of perineural invasion, and neither CDX2 nor CK7 were associated with overall survival.

Lymph node and vessel involvement, perineural invasion, and advanced pT and UICC stages of tumors are adverse predictors of survival in periampullary carcinomas, particularly in those with pancreatobiliary differentiation (2). Accordingly, we also found that patients with pancreatobiliary differentiation, lymph node involvement, and advanced pT and UICC stages had a significantly lower median survival. However, survival was not affected by vessel involvement or perineural invasion in our study.

HER2 and HER4 are members of the epidermal growth factor receptor family of transmembrane receptors, which play a role in tumor cell proliferation, adhesion, migration, and survival (21,22). Overexpression of HER2 and HER4 has been linked to poor prognosis in several types of tumors such as breast, lung, and gastric carcinomas. However, the expression of HER2 or HER4 and their correlation with survival in pancreatic tumors has been poorly studied and with inconsistent results.

Komoto et al. (23) reported that over 60% of pancreatic cancers demonstrated HER2 overexpression, which is an independent prognostic factor indicating shorter survival (hazard ratio, 1.806; $p=0.0258$). Other studies report a wide range of HER2 overexpression in pancreatic cancers (16% to 69%). Lei et al., (24) Yamanaka et al., (25) and Saxby et al. (26) also report poor survival outcome in patients with HER2 overexpression in pancreatic cancer. On the other hand, Koka et al. (27) found that the overall survival was longer in pancreatic adenocarcinoma cases with HER2 overexpression than in those without HER2 overexpression. In our series, we found that 18.4% of periampullary carcinoma showed HER2 overexpression, which was highly associated with the pancreatobiliary subtype and pancreatic head tumor location, but did not affect the overall survival.

There are even fewer studies of HER4 expression in the literature, probably due to the physiological role of HER4 in normal pancreatic tissue (15). However, te Velde et al. (15) reported loss of HER4 expression in 18% of pancreatic cancers, which was not associated with survival, stage or tumor grade. In the present study, our cases had a rate of HER4 overexpression of 12.2%, which was associated with perineural invasion and pancreatobiliary subtype. However, HER4 positivity was not associated with patient survival, as reported previously.

In conclusion, our findings suggested that only pancreatobiliary subtype, lymph node involvement and advanced pT and UICC stages were independent predictors for short survival, but the ampulla tumor location had a significantly better survival time. Immunohistochemical expression of CDX2, CK7, HER4, and HER2, vessel involvement, and perineural invasion were not associated with survival of patients with periampullary adenocarcinoma. Our findings warrant confirmation with new studies using a larger cohort of case analyses.

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