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Immunohistochemical expression of beta-catenin in ampullary adenocarcinoma: a cross-sectional retrospective study

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Introduction: Ampullary carcinomas are uncommon malignant tumours of the digestive system, they usually are adenocarcinomas presenting histologically as three types: intestinal, pancreaticobiliary and mixed. β-catenin is a multifunctional protein involved in physiological homoeostasis and intracellular adhesion. Abnormal nuclear accumulation of β-catenin has been described in many malignancies such as colon, breast, liver and others. The relationships between the immunohistochemical expression of β-catenin and the subtype, the grade and the stage of ampullary carcinoma are studied.

Material and methods: A cross-sectional retrospective study was done on 25 formalin-fixed paraffin-embedded blocks of ampullary carcinoma: Cases were collected from the archives of the pathology department in the Gastroenterology and hepatology teaching hospital, medical city/ Baghdad from January 2019 to March 2022. The data of the patients and the characteristics of the tumour were derived from the pathological reports; additional sections from the block were stained with β-catenin immunohistochemically.

Results: 25 paraffin blocks from patients with ampullary carcinoma (12 males, 13 females) were included in the study. 64% of the cases are classified as pancreaticobiliary, 20% as mixed, 12% as intestinal and 4% as adenosquamous type. Eighty-four percent of the cases are moderately differentiated, and the remaining is poorly differentiated. Most cases show strong β-catenin membranous staining and 80% express 3+ staining of cytoplasmic β-catenin. Regarding nuclear β-catenin staining, 56% has negative staining.

Conclusion: No significant association was found between the cytoplasmic and the nuclear expression of β-catenin and the tumour type, size and lymph node status. The grade of the tumour showed a significant correlation with the cytoplasmic expression; while, no correlation was noted with the nuclear expression. This study results do not support the use of beta-catenin as a diagnostic marker or prognostic marker in ampullary cancers.

Keywords: Ampullary adenocarcinoma, beta-catenin, nuclear and cytoplasmic

Introduction

Ampullary carcinomas are uncommon malignant tumours of the digestive system and comprise only 7% of periampullary carcinomas[1]. They are usually detected earlier than other malignancies in the periampullary region as they cause obstructive jaundice[2]. As a result of their earlier presentation, the surgical resection of these tumours occurs at a higher rate than other tumours in the area, and their prognoses are good[2].

Ampullary cancers usually are adenocarcinoma presenting histologically as intestinal, pancreaticobiliary and mixed. Differentiating these subtypes by morphology and immunohistochemical markers is important as the pancreaticobiliary subtype has a worse prognosis than the intestinal subtype[3]. Regarding the molecular pathogenesis of ampullary carcinomas, WNT pathway alterations occur commonly in the intestinal

HIGHLIGHTS

- Ampullary carcinomas are uncommon malignant tumours of the digestive system and present histologically as three subtypes: intestinal, pancraeticobiliary and mixed.
- Abnormal nuclear accumulation of β-catenin has been described in many malignancies of various organs.
- The study evaluated the expression of cytoplasmic and nuclear immunohistochemical staining of β-catenin in ampullary carcinoma and its relation to the subtype, grade and stage of the tumour.
- The study was a cross-sectional retrospective study that evaluated 25 formalin-fixed paraffin-embedded blocks of ampullary carcinoma removed by whipple procedure.
- The results showed that nuclear β-catenin expression was significantly higher in the intestinal subtype compared to the pancreaticobiliary subtype.
- There was no significant difference in β-Catenin expression between different grades or stages of ampullary carcinoma.
- The study suggests that β-catenin may have a role in the pathogenesis and progression of ampullary carcinoma, particularly in the intestinal subtype, and may have potential as a therapeutic target.

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subtype of ampullary adenocarcinoma, while TP53-Rb and RTK-RAS changes are common in the pancreaticobiliary subtype. KRAS, TP53, CTNNB1, SMAD4 are described in both types[4].

MRI and magnetic resonance cholangiopancreatography represent valuable diagnostic tools in assessing ampullary tumours. However, to accurately detect the epicentre of the tumour, careful gross dissection of resection specimens is necessary[3]. The localized tumours are typically treated by surgery, most commonly via a Whipple procedure, followed by adjuvant chemotherapy. Conversely, combined chemotherapeutic drugs are the primary approach for advanced tumours[2]. Old age, high tumour grade, advanced stage, lymphovascular invasion and lymph node and distant metastasis represent unfavourable prognostic factors[5]

β-catenin is a multifunctional protein involved in physiological homoeostasis and intracellular adhesion. WNT pathway is the chief regulator of β-catenin level[6]. In the absence of WNT pathway activation, β-catenin is inhibited through ubiquitin-proteasome system. When the WNT pathway is stimulated by a ligand or a genetic mutation, β-catenin accumulates in the cytoplasm and the nucleus. In the nucleus, it activates the transcription of oncogenes, such as Jun, c-Myc and CyclinD-1 and promotes cellular proliferation[7]. Consequently, abnormal nuclear accumulation of β-catenin has been described in many malignancies such as colon, breast, liver and others[8–10].

In our study, we evaluate the expression of cytoplasmic and nuclear immunohistochemical staining of β-catenin in ampullary carcinoma and its relation to the subtype, the grade and the stage of ampullary carcinoma.

Material and methods

The study has been approved by the ethical committee of the scientific unit &medical ethics of Al-Kindy College of Medicine (approval no. 191 on the 23 March 2022). The study does not involve any harm or risk to any patient or disclose any information about them.

A cross-sectional retrospective study was done on 25 formalin-fixed paraffin-embedded blocks of ampullary carcinoma removed by Whipple procedure performed in a tertiary centre specialized in this type of operation; each diagnosed and staged by experienced gastrointestinal pathologists: 3 are intestinal subtype, 16 are pancreaticobiliary, 5 are mixed and 1 is adenosquamous variant of ampullary adenocarcinoma. The type of tumour was confirmed by doing CK-7, CK-20 and CDX-2. These blocks were collected from the archives of the pathology department in the Gastroenterology and hepatology teaching hospital, medical city / Baghdad/ Iraq, from January 2019 to March 2022. Endoscopic biopsies are excluded from our study due to incomplete data regarding the subtype, grade and stage of the tumour.

The demographic data of the patients and the characteristics of the tumour were derived from the pathological reports in the archives. Our study population was 12 males (48%) and 13 females (52%); the age range was 28–69 years (the mean was 52.72). All the biopsies were resections as part of Whipple procedures. The original hematoxylin and eosin stained slides were reviewed and additional sections from the block were stained with β-catenin immunohistochemically using an antibody to Beta-Catenin (Clone: EP35- Rabbit Monoclonal Antibody) which is affinity purified and diluted in antibody diluent with 1% bovine serum albumin and 0.05% of sodium azide (NaN3) (1:50–1:100). The detection System was HRP/ DAB detection system. The manufacturer is PathnSitu technologies (https://www.pathnsitu.com/productDetails.php?id=16).

The positive control was the membranous staining of the normal duodenal mucosa. The negative control is obtained by removing the step of adding a primary antibody for β-Catenin in the procedure.

Interpretation of the results of Immunohistochemical staining

Two pathologists examine the slides of immunohistochemistry for β-catenin and scores for the cytoplasmic and the nuclear staining of β-catenin in the hot spot were used as follows:

0: when there is no staining of the cells
1 + : when there are less than 10% of the cells are stained
2 + : when there are 10–50% of the cells are stained
3 + : when there are more than 50% of the cells are stained

Statistical analysis

The data was analyzed by using SPSS (Statistical Package for Social Sciences), version 24, IBM.

Numerical data were presented as mean ± SD, while those categorical data were presented as frequencies and percentages.

t-test and one-way ANOVA were used to analyze the relationship among different numerical variables, while the χ^2 test was used in the case of categorical ones, (a *P* value of 0.05 and less was considered to be significant).

Fisher, s exact test was used for cells with small counts; expected frequencies of less than 5.

Results

In our study, paraffin-embedded blocks from 25 patients with ampullary carcinoma (12 males, 13 females) were stained and

Table 1

Association between age of participants and other variables

Variables	Age (years) Mean ± (SD)	*P*
Type		
Adenosquamous	61 ± 0	0.439
Intestinal	55.33 ± 4.51	
Mixed	54.60 ± 8.62	
Pancreato-biliary	51.13 ± 10.31	
Grade		
Second	54.00 ± 7.96	0.572
Third	46.00 ± 14.14	
Tumour size		
T1	59.67 ± 8.08	0.214
T2	53.56 ± 8.35	
T3	50.54 ± 9.91	
Lymph nodes		
N0	54.63 ± 12.50	0.287
N1	52.75 ± 8.50	
N2	51.75 ± 7.42	
N x	45.00 ± 0	

The above table illustrates nonsignificant differences when comparing patients with different types, grades, tumour sizes and the number of involved lymph nodes, according to age variable, as *P* values were (0.439, 0.572, 0.214& 0.287), respectively.

Table 2

Association between sex of participants and other variables.

Variables	Sex		P
	Male, *N* (%)	Female, *N* (%)	
Type			
Adenosquamous	1 (8.33)	0	0.627
Intestinal	1 (8.33)	2 (15.38)	
Mixed	3 (25)	2 (15.38)	
Pancreato-biliary	7 (58.33)	9 (69.23)	
Total	12 (100)	13 (100)	
Grade			
Second	11 (91.67)	10 (76.92)	0.315
Third	1 (8.33)	3 (23.08)	
Total	12 (100)	13 (100)	
Tumour size			
T1	2 (16.67)	1 (7.69)	0.786
T2	4 (33.33)	5 (38.46)	
T3	6 (50)	7 (53.85)	
Total	12 (100)	13 (100)	
Lymph nodes			
N0	5 (41.67)	3 (23.08)	0.482
N1	3 (25)	5 (38.46)	
N2	3 (25)	5 (38.46)	
N x	1 (8.33)	0	
Total	12 (100)	13 (100)	

Nonsignificant differences were found when comparing males and females with regard to the type, grade, tumour size and the number of involved lymph nodes (*P* values were 0.627, 0.315, 0.786 and 0.482), respectively.

examined for the immunoexpression of β-catenin. The age range of the patients was from 28 to 69 years; with the male patients being older than the females. Regarding the type of ampullary carcinoma, 64% of the cases (16 out of 25) are classified as pancreaticobiliary type, 20% (5 out of 25) as mixed type, 12% (3 out of 25) as intestinal-type and 4% (1 out of 25) as an adenosquamous variant. Most cases were assigned as moderately differentiated (84% of the cases), and the remaining are poorly differentiated. Considering the tumour depth of invasion in the stage of ampullary carcinomas; 52% of the cases were T3, 36% were T2 and 12% were T1; while the lymph node status was as the following: N0, N1 and N2 were eight cases each (32%) with one case assigned as NX.

There was no significant association between the age of the patients and the type, grade and size of the tumours as well as the number of lymph nodes involved. The same comment applies to the sex of the patients (Tables 1 and 2, respectively).

Most ampullary carcinomas show strong β-catenin membranous staining. Most ampullary carcinomas in this study (80%) express 3+ staining of cytoplasmic β-catenin, 12% of them have 2+ staining, 4% (one case) have 1+ staining and 4% have negative cytoplasmic staining (score 0) (Fig. 1). Regarding nuclear β-catenin staining, 56% have negative staining (score 0), 28% have 1+ staining, 12% have a score of 2+ and one case (4%) shows 3+ nuclear staining (Fig. 2).

There was no significant association found between the cytoplasmic and the nuclear immunohistochemical expression of β-catenin and the tumour type, tumour size and the number of lymph node involved by the carcinoma (Tables 3, 4 and 5, respectively).

Patients with moderately differentiated ampullary carcinoma have significantly higher levels of cytoplasmic β-catenin (*P* value = 0.005); While patients with moderately differentiated ampullary carcinoma have nonsignificantly negative and lower levels of nuclear β-catenin (*P* value > 0.05) (Table 6).

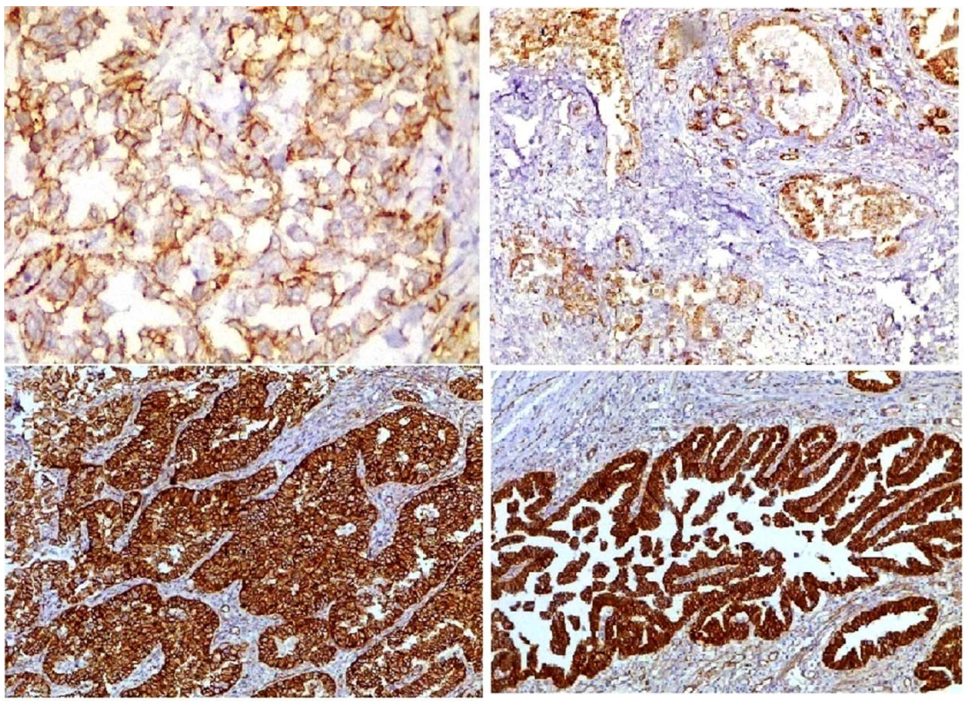

Figure 1. Cytoplasmic expression of beta-catenin; upper left (score 0), upper right (score 1+) , lower left (score 2+) and lower right (score 3+).

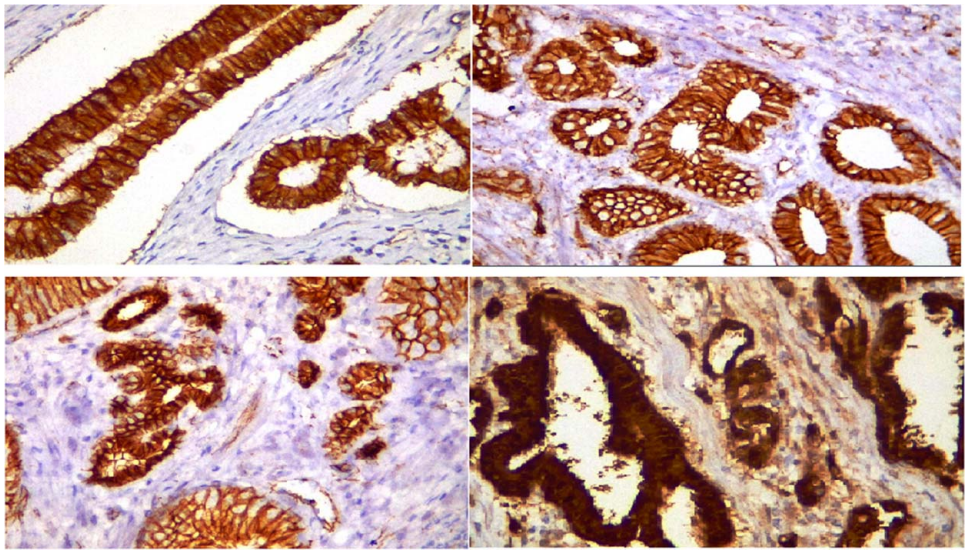

Figure 2. Nuclear expression of beta-catenin; upper left (score 0), upper right (score 1+) , lower left (score 2+) and lower right (score 3+).

Discussion

Ampullary carcinomas are infrequent cancer in comparison to other malignancies of the gastrointestinal tract. It has three types; each having variable biological characteristics as the ampullary region has multiple cell linings (intestinal, pancreaticobiliary). These tumours most commonly occur in sporadic settings; however, they may be a presentation of syndromes such as familial adenomatous polyposis and Gardner's syndrome[11]. Understanding the role of beta-catenin in the development of ampullary carcinoma may have diagnostic and therapeutic implications.

β-catenin is involved in the WNT signalling pathway that has a vital role in many types of cancer, especially colonic cancer[12]; however, β-catenin's role is less obvious in ampullary carcinomas as found in a study done by Kawakami *et al.*[13] who found β-catenin nuclear accumulation in 19% of ampullary carcinomas (21 cases included in their study). Another study done by Yamazaki reported 30% of ampullary carcinomas cases with nuclear staining of β-catenin (30 cases included in their study)[14]. In our study, 16% of ampullary carcinomas show 2+ to 3+ nuclear immunostaining which is comparable to that found in a study done by Kim *et al.*[11] in which they report 17.8% of 73 cases of ampullary carcinoma.

In our study, there was no significant association between the tumour type and nuclear staining of β-catenin; in contrast to the finding observed in Kim and colleagues' study with a high incidence of β-catenin nuclear accumulation in the intestinal-type, this is because of lower frequency of intestinal-type in our study (three cases only).

In a study accomplished by Sung and colleagues, membranous loss of E-cadherin and β-catenin was observed in the non-intestinal-type of ampullary tumours, while aberrant non-membranous β-catenin expression was observed in intestinal-type tumours. Dysregulation of E-cadherin, β-catenin and S100A4 expression may play a role in the carcinogenesis and tumour progression of ampullary adenocarcinomas as concluded in their study[14].

Table 3

Association between type of ampullary carcinoma and tumour marker (β-catenin)

	Type				
Tumour marker	**Adenosquamous, *N* (%)**	**Intestinal, *N* (%)**	**Mixed, *N* (%)**	**Pancreato-biliary, *N* (%)**	***P***
β-catenin (cytoplasmic)					
Negative	0	0	0	1 (6.25)	0.768
1+	0	0	0	1 (6.25)	
2+	0	0	2 (40)	1 (6.25)	
3+	1 (100)	3 (100)	3 (60)	13 (81.25)	
Total	1 (100)	3 (100)	5 (100)	16 (100)	
β-catenin (nuclear)					
Negative	0	2 (66.67)	4 (80)	8 (50)	0.576
1+	1 (100)	1 (33.33)	1 (20)	4 (25)	
2+	0	0	0	3 (18.75)	
3+	0	0	0	1 (6.25)	
Total	1 (100)	3 (100)	5 (100)	16 (100)	

Non-significant relationships were prevailing between the type of ampullary carcinoma and the tumour marker (β-catenin) both cytoplasmic and nuclear (*P* values were 0.768 and 0.576), respectively.

Table 4

Association between tumour size of ampullary carcinoma and tumour marker (β-catenin)

	Tumour size			
Tumour marker	**T1, *N* (%)**	**T2, *N* (%)**	**T3, *N* (%)**	***P***
β -catenin (cytoplasmic)				
Negative	0	0	1 (7.69)	0.762
1+	0	1 (11.11)	0	
2+	0	1 (11.11)	2 (15.38)	
3+	3 (100)	7 (77.78)	10 (76.92)	
Total	3 (100)	9 (100)	13 (100)	
β-catenin (nuclear)				
Negative	0	6 (66.67)	8 (61.54)	0.266
1+	2 (66.67)	3 (33.33)	2 (15.38)	
2+	1 (33.33)	0	2 (15.38)	
3+	0	0	1 (7.69)	
Total	3 (100)	9 (100)	13 (100)	

The above table shows a nonsignificant association between tumour size and the level of tumour marker β-catenin; both cytoplasmic and nuclear ones.

Hsu *et al.*[15] study showed that loss of membranous expression of E-Cadherin and β-catenin was associated with pancreatic invasion, recurrence and poor outcome. Dysfunction of β-catenin may contribute to the development of ampullary adenocarcinoma and relate to some poor prognostic clinicopathological features reported in other studies[13,15,16].

In another study done by Hsiao *et al.*[17], aberrant localization of beta-catenin was noticed in 10.7% of ampullary adenocarcinomas and 0% of periampullary/ duodenal adenocarcinoma, and 42.9% of jejunal/ ileal adenocarcinomas, these results indicate that WNT signalling pathway in a subset ampullary adenocarcinomas may be a possible marker for targeted therapy.

A Perysinakis *et al*. study was done to associate the immunohistochemical expression of beta-catenin, EGFR, CK7, CK20, MUC1, MUC2 and CDX2 in ampullary carcinoma with the grade of differentiation and prognosis, they concluded that the immunoreactivity against Ck20 and MUC1 in ampullary carcinoma is a helpful adjunct to histological examination to determine the type of ampullary cancer; however, none of the studied

Table 5

Association between number of involved lymph nodes in ampullary carcinoma and tumour marker (β-catenin)

	Lymph nodes				
Tumour marker	**N0, *N* (%)**	**N1, *N* (%)**	**N2, *N* (%)**	**Nx, *N* (%)**	***P***
β -catenin (cytoplasmic)					
Negative	1 (12.5)	0	0	0	0.677
1+	0	0	1 (12.5)	0	
2+	0	2 (25)	1 (12.5)	0	
3+	7 (87.5)	6 (75)	6 (75)	1 (100)	
Total	8 (100)	8 (100)	8 (100)	1 (100)	
β -catenin (nuclear)					
Negative	3 (37.5)	5 (62.5)	5 (62.5)	1 (100)	0.347
1+	2 (25)	3 (37.5)	2 (25)	0	
2+	3 (37.5)	0	0	0	
3+	0	0	1 (12.5)	0	
Total	8 (100)	8 (100)	8 (100)	1 (100)	

The above table demonstrates a nonsignificant association between the number of involved lymph nodes and the level of the tumour marker β-catenin both cytoplasmic and nuclear ones.

Table 6

Association between grade of ampullary carcinoma& tumour marker (β-catenin)

	Grade		
Tumour marker	**Second grade (moderately differentiated), *N* (%)**	**Third grade (poorly differentiated), *N* (%)**	***P***
β -catenin (cytoplasmic)			
Negative	0	1 (25)	0.005
1+	0	1 (25)	
2+	2 (9.52)	1 (25)	
3+	19 (90.48)	1 (25)	
Total	21 (100)	4 (100)	
β -catenin (nuclear)			
Negative	10 (47.62)	4 (100)	0.291
1+	7 (33.33)	0	
2+	3 (14.29)	0	
3+	1 (4.76)	0	
Total	21 (100)	4 (100)	

Patients with moderately differentiated ampullary carcinoma; had significantly higher levels of the cytoplasmic marker β-catenin, (3+). While patients with moderately differentiated ampullary carcinoma; had nonsignificantly negative and lower levels of the nuclear marker β-catenin, (*P* value > 0.05).

markers found to have prognostic significance[18]. The mutation of tumour suppressor genes (e.g. APC, GSK-3 and AXIN) in the WNT pathway was found to induce nuclear accumulation of beta-catenin in ampullary cancer[19].

Park S and colleagues studied 111 cases of ampullary carcinoma to investigate the immunohistochemical expression of E-cadherin and beta-catenin, 49 (44.1%) cases were associated with an adenomatous component; they found that 4 (8.2%) adenomas and 45 (40.5%) carcinomas showed abnormal staining pattern either as nuclear staining or loss of membranous staining. The cases with membranous loss of beta-catenin expression were correlated with less differentiated histology and poor prognosis[20].

The results of our study may conflict with the results of previous studies which may reflect the variability of the assessment method of beta-catenin immunohistochemistry between studies, the differences in patient populations and the need for a larger sample size as ampullary cancer is rare in Iraq. This study only focuses on the immunohistochemical expression of beta-catenin in ampullary carcinoma and does not investigate other potential biomarkers or molecular pathways involved in the disease. However, the study should be followed by future studies on other markers in this specific type of cancer.

Conclusion

Most ampullary carcinomas show strong β-catenin membranous expression, nuclear expression was lost in more than half of the cases. No significant association was found between the cytoplasmic and the nuclear expression of β-catenin with the tumour type, size and lymph node status. The grade of the tumour showed a significant correlation with the cytoplasmic expression; while no correlation was noted with nuclear expression. Our study results do not support the use of beta-catenin as a diagnostic marker or prognostic marker in ampullary cancers. As a result, a search for other helpful markers is indicated.

Ethical approval

The study has been approved by the ethical committee of the scientific unit &medical ethics of Al-Kindy College of Medicine (approval no. 191 on the 23 March 2022). It could be provided on request.

Patients consent

None.

Source of funding

None.

Author contribution

S.A.M.: supervising of writing the manuscript, interpretation of immunohistochemical findings. M.N.H.: writing the manuscript, interpretation of the immunohistochemical findings, using SPSS (Statistical Package for Social Sciences), version 24, IBM.. A.N. A.: sample and data collection, performance of immunohistochemical staining.

Conflicts of interest disclosure

None.

References

[1] Jemal A, Siegel R, Ward E, *et al*. Cancer statistics, 2008. CA Cancer J Clin 2008;58:71–96.
[2] Ahn DH, Bekaii-Saab T. Ampullary cancer: an overview. Am Soc Clin Oncol Educ Book 2014:112–5.
[3] Allard FD. Adenocarcinoma-ampulla. PathologyOutlines.com website. https://www.pathologyoutlines.com/topic/ampullaadenocarcinoma.html
[4] Tan J, Tan P, Teh BT. Defining the molecular alterations of ampullary carcinoma. Cancer Cell 2016;29:135–6.
[5] Li HB, Zhao FQ, Zhou J. Prognostic nomogram for disease-specific survival in patients with non-metastatic ampullary carcinoma after surgery. Ann Surg Oncol 2019;26:1079–85; Epub 2019 Jan 18.
[6] van Ooyen A, Nusse R. Structure and nucleotide sequence of the putative mammary oncogene int-1; proviral insertions leave the protein-encoding domain intact. Cell 1984;39:233–40.
[7] Shang S, Hua F, Hu ZW. The regulation of β-catenin activity and function in cancer: therapeutic opportunities. Oncotarget 2017;8: 33972–89.
[8] Khramtsov AI, Khramtsova GF, Tretiakova M, *et al*. Wnt/beta-catenin pathway activation is enriched in basal-like breast cancers and predicts poor outcome. Am J Pathol 2010;176:2911–20.
[9] Tao J, Calvisi DF, Ranganathan S, *et al*. Activation of beta-catenin and Yap1 in human hepatoblastoma and induction of hepatocarcinogenesis in mice. Gastroenterology 2014;147:690–701.
[10] Kobayashi M, Honma T, Matsuda Y, *et al*. Nuclear translocation of beta-catenin in colorectal cancer. Br J Cancer 2000;82:1689–93.
[11] Kim GIl, Kim JB, Park SB, *et al*. Genetic and epigenetic alterations of the wnt/beta-catenin signaling pathway in cancer of the ampulla of vater. Korean J Pathol 2007;41.4:224–31.
[12] Bienz M, Clevers H. Linking colorectal cancer to Wnt signaling. Cell 2000;103:311–20.
[13] Kawakami M, Kimura Y, Furuhata T, *et al*. beta-Catenin alteration in cancer of the ampulla of Vater. J Exp Clin Cancer Res 2002;21:23–7.
[14] Sung R, Kang L, Han J-H, *et al*. Differential expression of E-cadherin, β-catenin, and S100A4 in intestinal type and nonintestinal type ampulla of vater cancers. Gut Liver 2014;8:94–101.
[15] Hsu HP, Shan YS, Jin YT, *et al*. Loss of E-Catherin and beta catenin is correlated with Poor prognosis of ampullary neoplasms. J Surg Oncol 2010;101:356–62.
[16] Yamazaki K, Hanami K, Nagoa T, *et al*. Increased cyclin D1 expression in cancer of ampulla of Vater: relevance to nuclear beta catenin accumulation and K-ras gene mutation. Mol Pathol 2003;56:336–41.
[17] Hsiao SJ, Remotti HE. Beta-catenin and p53 expression in ampullary and small intestinal adenocarcinoma. J Clin Oncol 2013;31 doi:10.1200/jco.2013.31.15_suppl.e14659
[18] Perysinakis Iraklis, Minadou Emillia, Leontara Vasileia, *et al*. Differential expression of B- catenin, EGFR, CK7, CK20, MUC1, MUC2; and CDX2 in intestinal and pancreatobiliary- type Ampullary Carcinoma. Int J Surg Pathol 2017;25:31–40.
[19] Jayaramyya K, Balachandar V, Santhy KS. Ampullary carcinoma a genetic perspective. Mutat Res 2018;776:10–22.
[20] Park S, Kim SW, Lee BL, *et al*. Expression of E-cadherin and Beta catenin in adenocarcinoma- carcinoma sequence of ampullary of vater cancer. Hepatogastroenterology 2006;53:28–32.