Original Article

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Pathology Section

Expression of Beta-Catenin in Colorectal Carcinoma and its Association with Various Clinicopathological Parameters

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ABSTRACT

Introduction: Colorectal Carcinoma (CRC) is the third most commonly diagnosed cancer and the second leading cause of cancer related mortality globally. It is a source of concern for researchers worldwide and hence, a lot of emphasis is being given towards early detection and targeted drug therapy to improve the survival rate.

Aim: To study the expression of beta-catenin in colonic polyps, adenomas and CRC and to associate beta-catenin expression with various clinicopathological features.

Materials and Methods: This was a prospective cross-sectional study conducted in the Department of Pathology, Kalinga Institute of Medical Sciences, Bhubaneswar, Odisha, India from September 2018 to August 2020. Colonoscopic biopsies, mucinous carcinoma and poorly preserved tissue were excluded. Histopathological study and Immunohistochemistry evaluation of beta-catenin was done. Statistical analysis was done by using appropriate tests. A p-value of less than 0.05 was taken as statistically significant.

Results: Out of 80 cases, 40 cases were benign lesions Non neoplastic polyp and adenoma) and 40 cases were adenocarcinoma. It was observed that benign lesions had maximum cases with preserved membranous expression (36/40) and very few cases (4/40) showed cytoplasmic expression of beta-catenin. But in carcinoma, high cytoplasmic expression was seen in 20/40 (50%) whereas 8/40 (20%) cases had nuclear positivity. Membranous beta-catenin expression was significantly higher in benign lesions than in the malignant lesions (IS:8.75±3.09 versus 4.30±2.70) respectively; (p<0.0001). But cytoplasmic beta-catenin expression was low in benign lesion as compared to malignant lesion (IS: 2.07±3.46 versus 5.35±3.14), respectively; (p<0.0001). However, nuclear beta-catenin expression was extremely low in benign lesions than in malignant lesions (0.08±0.47 versus 1.90±3.49), respectively; (p=0.0016), this difference was statistically significant.

Conclusion: The present study demonstrates the change in beta-catenin expression with gradual transition from predominantly membranous pattern to cytoplasmic or nuclear as we progress from normal colorectal tissues to polyps, benign premalignant lesions and malignant neoplasms. This property of beta-catenin helps in determining malignant potential of various premalignant neoplasms of large intestine which in turn helps in initiating early prophylactic treatment.

Keywords: Adenomatous polyp, Colorectal neoplasms, Membranous expression

INTRODUCTION

The Colorectal Carcinoma (CRC) is the third most commonly diagnosed cancer and the second leading cause of cancer related mortality globally [1,2]. As per the World Health Organisation (WHO) GLOBOCAN database, there were 1,849,518 estimated new CRC cases and 880,792 deaths occurred due to CRC [3]. The CRC is the fourth most common cause of cancer in males and third most common cause of cancer in females in India [4]. The factors influencing outcome in CRC patients are tumour invasion, lymphovascular invasion, status of lymph nodes and serum Carcinoembryonic Antigen (CEA) level [5,6]. Continuous efforts are being put for early diagnosis and targeted therapy for the tumour. Adjuvant therapy is administered basing on individual patient risk and hence, prognostic factors are essential for risk stratification. Adenomatous Polyposis Coli (APC) mutations are acquired early in the pathogenesis of around 80% colon cancers. This leads to cytosolic accumulation of β-catenin which in combination with T cell Transcription Factor (TCF)/Lef1 shuttles to the nucleus where it functions as a transcription factor and promotes cellular proliferation [7,8]. Beta-catenin also has prognostic implications in carcinoma of esophagus, stomach, gall bladder, ovary, endometrium as well as in leukaemia and melanoma [7,8]. This study directs us towards risk stratification, early detection of premalignant lesions and proper categorisation by differential expression of beta-catenin. This was a novel approach towards early and rational targeted therapy of colorectal neoplasms.

Objectives

- To study the expression of beta-catenin in colonic polyps, adenomas and CRC and to compare its expression with that of normal colon.
- Association of beta-catenin expression with various clinicopathological features.

MATERIALS AND METHODS

This was a prospective cross-sectional study conducted in the Department of Pathology, Kalinga Institute of Medical Sciences, Bhubaneswar, Odisha from September 2018 to August 2020 after approval from Institutional Ethics Committee (IEC No. 90/2018).

Inclusion criteria: Radical excision specimens of histologically confirmed cases of CRC and all specimens of adenoma and colorectal polyps were included.

Exclusion criteria: Colonoscopic biopsies, mucinous carcinoma and poorly preserved tissue were excluded.

Study Procedure

This study included 40 cases of CRC, 20 cases of adenoma and 20 cases of non neoplastic polyps. Along with this, 20 cases of normal colon were also studied. Out of the total cases, 40 were endoscopy guided biopsies and 40 were colectomy specimens. Sample size was calculated after consulting with statistician. Specimens were routinely processed and fixed overnight in 10% buffered formalin.

Grossing of the specimens was done as per the recent American Joint Committee on Cancer (AJCC) guidelines [4]. Four to five micrometer thick formalin fixed, paraffin embedded tumour sections were stained with Haematoxylin and Eosin (H&E) stain. Histologic examination of tumour type and grade were performed routinely according to criteria outlined in WHO classification of tumours 5th edition, 2019 [3]. For colectomy specimen, tumour depth, Lymphovascular Invasion (LVI), Perineural Invasion (PNI) and lymph node status was assessed according to AJCC 8th edition [4].

Evaluation of beta-catenin was done on formalin fixed paraffin embedded tissue sections (4-5 µ thick) on poly L-lysine coated slides by using polymers in two steps (indirect method). Desmoid tumour block served as external positive control whereas normal colon was taken as internal positive control from the same tumour block under study. The IHC staining was done using monoclonal ready-to-use beta-catenin antibody manufactured by Dako (code IS 702). The degree of IHC staining in the tissue sections was scored independently by two pathologists who were blinded to the clinical and pathological data. Staining intensity was graded using a scale of 0-3 as follows:

0: No staining;

1: Weak staining;

2: Moderate staining and

3: Strong staining [9,10]

The extent of staining was graded on a scale as the following:

0: ≤5%;

1: 6-25%

2: 26-50%

3: 51-75%

4: 76-100% [9,10]

According to the percentage of the section exhibiting positive staining relative to the entire carcinoma involved area, more than 50% of tumour area showing moderate and strong staining was considered as positive. The intensity and extent scores were multiplied to generate the immunoreactivity score (IS; range, 0-12) for each case.

<6 was considered as low (Negative)

>6 was considered as high (Positive) [11]

STATISTICAL ANALYSIS

Data was entered into Microsoft excel spreadsheet 2010 and analysed using EpiInfo software version 7.1.3. Results were expressed as mean, median, standard deviation, percentages, frequencies and proportions. Statistical analysis was done by Chi-square test, Fisher's exact test and t-test which were used as tests of association. A p-value of less than 0.05 was taken as statistically significant.

RESULTS

In this study, the total number of subjects were 80 which included 40 cases (50%) of adenocarcinoma, 20 (25%) cases of adenoma and 20 (25%) cases of non neoplastic polyps. The age range of patients was from 1-96 years with mean age being 54.73±21.15 years. Age group of 51-60 years had highest number of cases i.e., 20 (25%) followed by the age range of 61-70 years i.e., 16 (20%). Out of 40 cases of adenocarcinoma, maximum belonged to the age group of 51-60 years and 71-80 years of age with mean age of 61.75±11.62 years, whereas maximum number of adenoma cases belonged to age group of 61-70 years of age.

For statistical analysis, the total cases were divided into two age groups- less than 50 years and more than 50 years. A 42.86% of patients below 50 years had preserved membranous expression and 54.54% of patients above 50 years had preserved membranous beta-catenin expression. Nuclear expression was seen in 28.57% patients with age less than 50 years and in 51.15% patients with age more than 50 years. However, there was no significant association of these results with age [Table/Fig-1]. Among the total 40 adenocarcinoma cases, 70% (28 cases) were males and 30% (12 cases) were females with male:female ratio being 2.3:1 [Table/Fig-1].

A 46.42% of males showed preserved membranous beta-catenin expression while it was seen in 58.43% of females. A 53.57% males had increased cytoplasmic beta-catenin expression whereas, 41.66% females had strong cytoplasmic expression. Increased nuclear expression of beta-catenin was seen in 21.42% males and in 25% females. These results were not statistically significant [Table/Fig-1].

Out of 80 cases, there were total number of 60 male patients (75%) and 20 (25%) female patients with male to female ratio being 3:1. Out of 80 cases, 40 cases (50%) were adenocarcinoma. Adenoma with low-grade dysplasia constituted 17 cases (20%) and with high-grade dysplasia constitute 3 cases (5%). Out of 20 cases of non neoplastic polyps, maximum were juvenile polyps, i.e., 10 cases (12.5%) and only one case was hamartomatous polyp (1.25%). Maximum number of adenocarcinoma were located in the right colon 17 cases (42.5%) followed by left colon 14 cases (35%) and minimum in the rectum nine cases (22.5%). A total of 29 cases (72.5%) of adenocarcinoma cases belong to grade I followed by seven cases (17.5%) in grade II and only four cases (10%) of carcinoma belong to grade III. A 45% (18 cases) of carcinoma belong to stage II, 30% (12 cases) belong to stage IIIB and only 5% (02 cases) belong to stage IIIC. LVI was present in 23 cases (57.5%) adenocarcinoma cases and PNI was present in10 cases (25%). A 45% (18) of adenocarcinoma cases had positive metastatic nodes [Table/Fig-2].

There were 45.2% cases in right colon, 35% cases in left colon and 22.5% cases in rectum. High beta-catenin positivity was seen in left colon (21.42%) and rectum (33.33%). These results showed statistically significant association with p-value=0.03 [Table/Fig-2].

According to histological grading, the CRC cases were divided in two groups grade I, II and grade III. A 47.22% of grade I, II tumours had preserved membranous expression while 50% of grade III tumours had increased nuclear expression. High cytoplasmic beta-catenin expression was seen in 52.78% of grade I, II tumours and in 50% of grade III tumours. Increased nuclear expression was seen in 16.66% of grade I, grade II tumours and 25% of grade III tumours. Membranous, cytoplasmic and nuclear expression of beta-catenin did not show any statistically significant association with histological grade of CRC [Table/Fig-2].

Variable value	Total	Membranous expression			Cytoplasmic expression			Nuclear expression		
		Preserved	Reduced	p-value	Low	High	p-value	Low	High	p-value
Age (in year)										
< 50	7	3	4	0.67*	3	4	0.69*	5	2	0.58*
>50	33	18	15		18	15		28	5	
Gender										
Male	28	13	15	0.73**	13	15	0.73**	22	6	1.00*
Female	12	7	5		7	5		9	3	

[Table/Fig-1]: Beta-catenin expression levels Adenocarcinoma cases in association with age and gender of patients.
*Applied fisher's-exact t-test; **Applied chi-square test

Variables	Total	Membranous expression			Cytoplasmic expression#			Nuclear expression##		
		Preserved	Reduced	p-value	Low	High	p-value	Low	High	p-value
Tumour site										
Right colon	17	11	6	0.31*	11	6	0.40*	17	0	0.03*
Left colon	14	6	8		6	8		11	3	
Rectum	9	3	6		3	6		6	3	
Adenoma										
LGD	17	14	3	0.02	14	3	0.02	0	0	1.00
HGD	3	0	3		0	3		2	1	
Grade										
Well-moderated	36	17	19	1.0	17	19	1.0	30	6	0.55
Poor	4	2	2		2	2		3	1	
TNM stage										
I-II	22	8	14	0.05**	8	14	0.05**	16	6	0.02*
III	18	13	5		13	5		16	2	
Lymphnode metastasis										
Present	18	14	4	0.72	14	4	0.72	16	2	0.23
Absent	22	15	7		15	7		16	6	
LVI										
Present	23	11	12	0.71	11	12	0.71	20	3	1.00
Absent	17	10	7		10	7		14	3	
PNI										
Present	10	5	5	0.72	5	5	0.72	9	1	0.66
Absent	30	15	15		15	15		24	6	

[Table/Fig-2]: Beta-catenin expression levels of the tumour in association with clinicopathological parameters.
Statistically significant (p<0.05); *applied Fisher's-exact t-test; **applied Chi-square test; LVI: Lymphovascular invasion; PNI: Perineural invasion; no nuclear expression was seen in LGD; #Total cytoplasmic expression was seen in 20 adenoma cases; ##Total nuclear expression was seen in 3 adenoma cases

Based on staging, tumours were categorised into two groups- Stage I, II and Stage III. A 36.36% tumours of Stage I, II had membranous beta-catenin expression while the same was observed in 72.22% cases of stage III. Increased cytoplasmic expression was found in 63.63% of stage I, II tumours and in 21.77% of stage III tumours. A 27.27% cases of stage I, II have high nuclear beta-catenin expression while no case among stage III tumours had nuclear expression of beta-catenin. These differences were statistically significant with p-value=0.02 [Table/Fig-2].

Preserved membranous beta-catenin expression was seen in 77.77% cases with nodal metastasis. High cytoplasmic expression was seen in 22.22% cases with nodal metastasis. Nuclear expression was seen in 11.11% cases with nodal metastasis. No statistically significant association was observed between beta-catenin expression and lymph node metastasis [Table/Fig-2].

A 23 cases of carcinoma showed vascular invasion and 10 cases showed perineural invasion. A 47.82% of tumours showing LVI and 50% of tumours showing PNI had preserved membranous beta-catenin expression. A 52.17% tumours with LVI and 50% of tumours with PNI had cytoplasmic expression. A 13.04% of tumours with LVI and 10% of tumours with PNI had nuclear beta-catenin expression. There was no statistically significant association found between LVI with beta-catenin expression and PNI with beta-catenin expression [Table/Fig-2].

Out of 80 total cases, 40 cases were benign lesions (Non neoplastic polyps and adenoma) and 40 cases were adenocarcinoma. We observed that maximum benign lesions had preserved membranous expression (36/40); very few cases (4/40) showed cytoplasmic expression of beta-catenin and only one case of adenoma with high grade dysplasia showed nuclear positivity. But in carcinoma, preserved membranous expression was in 20/40 (50%), high cytoplasmic expression was in 20/40 (50%), whereas 8/40 (20%) cases had nuclear positivity. Membranous beta-catenin expression was significantly higher in benign lesions than in malignant lesions. (IS:8.75±3.09 versus 4.30±2.70), respectively; (p<0.0001). But cytoplasmic beta-catenin expression was low in benign lesion as compared to malignant lesion (IS: 2.07±3.46 versus 5.35±3.14), respectively (p<0.0001).

However, nuclear beta-catenin expression was extremely low in benign lesions than in malignant lesions (0.08±0.47 versus 1.90±3.49), respectively; (p<0.0016) This difference was statistically significant [Table/Fig-3].

Beta-catenin score (Mean±SD)	Colorectal lesions		p-value
	Polyp and adenoma (n=40)	Colorectal cancer (n=40)	
Membranous score	8.75±3.09	4.30±2.70	<0.0001#
Cytoplasmic score	2.07±3.46	5.35±3.14	<0.0001#
Nuclear score	0.08±0.47	1.90±3.49	0.0016#

[Table/Fig-3]: Comparison of membranous, cytoplasmic and nuclear beta-catenin Immunoreactivity Score (IS) with benign and malignant lesions (N=80).
#applied independent t-test

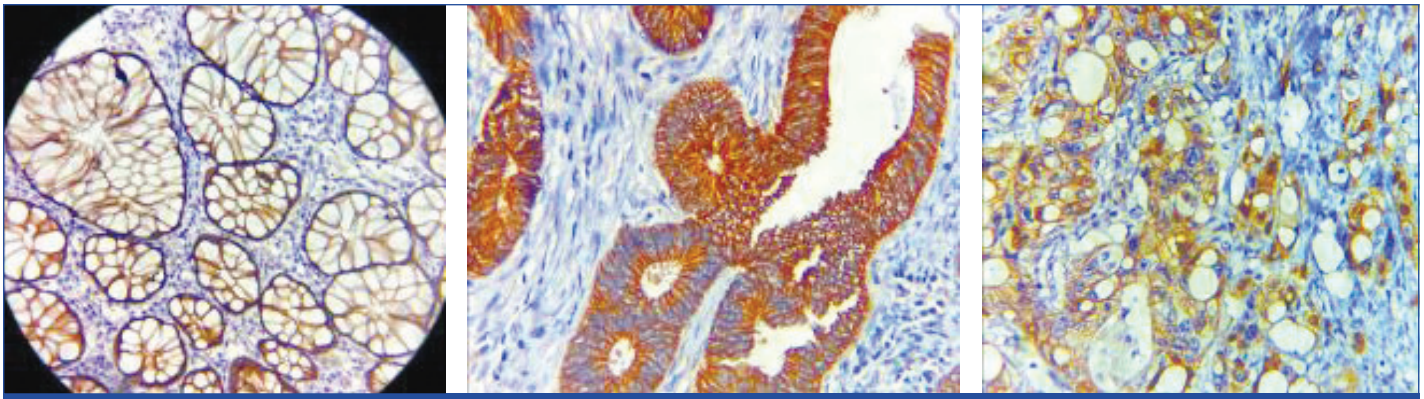
DISCUSSION

The various patterns of IHC expression of beta-catenin and its association with age, sex, site, histological grade, pathological stage, LVI, PNI and LN metastasis were studied.

Beta-catenin has a major role in development of CRC. Its function is controlled by Wingless-related integration site (Wnt) signaling pathway [12]. Normally beta-catenin is degraded by APC-beta catenin complex, thus avoiding its intracellular accumulation [13,14].

Mutations of APC and beta-catenin genes cause over accumulation of intracellular beta-catenin level which translocates to nucleus causing activation of transcription factor and uncontrolled growth of tumour cells.

Out of 40 cases of adenocarcinoma, maximum belonged to the age group of 51-60 years (27.5%) and 71-80 years (27.5%) of age with mean age of 61.75±11.62 years, whereas maximum number of adenoma cases belonged to age group of 61-70 years of age (25%). In this study, we found that patients with >50 years of age showed



[Table/Fig-4]: IHC for beta-catenin (400x): Normal benign intestinal glands showing membranous expression; **[Table/Fig-5]:** IHC for beta-catenin(400x): Well-differentiated (Grade-I) adenocarcinoma of colon displaying membranous expression score 3^{+}; **[Table/Fig-6]:** IHC for beta-catenin(400x): Poorly differentiated carcinoma (Grade-III) with both nuclear and cytoplasmic expression. (Images from left to right)

high cytoplasmic and nuclear expression of beta-catenin. But these results were statistically insignificant. Abdul rahaman ZA et al., similarly found that beta-catenin was frequently expressed in >50 years of age [14]. Gao ZH et al., also found that >60 years of patients had more cytoplasmic and nuclear beta-catenin expression [10].

There was a male preponderance among the patients with adenocarcinoma in this study with male to female ratio being 2.3:1. Males had increased cytoplasmic beta-catenin expression while females had a higher nuclear expression of beta-catenin. No significant association was found between beta-catenin expression and gender of patients. Gao ZH et al., and Yoshida N et al., had findings similar to this study [10,15].

A 42.5% cases of adenocarcinoma were located in right colon and 35% in left colon. Right colon carcinomas had preserved membranous expression whereas increased nuclear and cytoplasmic expression was seen in rectal carcinomas. This result was statistically significant with p-value=0.03. Present study findings were agreeable with Gao ZH et al., [10]; but showed discordant results with studies by Abdulrahaman ZA et al., and Wantisawan W et al., [14,16].

In the present study, out of 40 adenocarcinoma cases, 29 had histological grade I, seven had grade II and only four had grade III. No significant association was found between beta-catenin expression and histological grading of tumour. Wantisawan W et al., had maximum grade I tumours having highest nuclear beta-catenin expression [16]. Gao ZH et al., found increased cytoplasmic expression in grade-I and II tumours [10].

A 45% of adenocarcinoma cases in the study had pathological stage II, 30% had stage IIIB, 10% had stage IIIA and 5% had stage IIIC. There was statistically significant association of beta-catenin expression with pathological stage with p-value=0.02. Wantisawan W et al., found that maximum tumours had stage II having highest nuclear beta-catenin positivity [16]. Abdulrahaman ZA et al., found low cytoplasmic expression of beta-catenin in stage I and II tumours than in those of stage III and IV [14]. Gao ZH et al., found more number of cases with preserved membranous expression along with high nuclear positivity in tumours of stage I and II [8,10]. These studies were discordant with our study.

Nodal metastasis was seen in 18 cases of adenocarcinoma. However, there was higher cytoplasmic and nuclear beta-catenin expression in cases without nodal metastasis. But no significant association was observed. Gao ZH et al., and Wanistsawan W et al., had coherent results with this study [10,16]. On the contrary, Abdulrahaman ZA et al., found that node positive cases had higher beta-catenin expression than node negative patients [14].

Vascular invasion was present in 23 cases of carcinoma. A 77.77% of patients with vascular invasion had preserved membranous expression and high cytoplasmic expression. There was similar expression of membranous and cytoplasmic beta-catenin in cases both with and without perineural invasion. Gao ZH et al., observed that only 3 out of 182 cases showed vascular invasion [10]. All these cases showed membranous expression and only one case had high cytoplasmic expression. However, this was not consistent with the study.

It was observed that most of the benign lesions (36/40) had preserved membranous expression [Table/Fig-4] and very few cases (4/40) had cytoplasmic expression of beta-catenin. Only one case of adenoma with high-grade dysplasia showed cytoplasmic beta-catenin positivity. Among carcinoma cases, preserved membranous expression was seen in 50% cases [Table/Fig-5], high cytoplasmic expression in 50% whereas nuclear positivity was observed in 20% cases [Table/Fig-6].

Membranous beta-catenin expression was significantly higher in benign lesions than in the malignant lesions. (IS:8.75±3.09 versus 4.30±2.70) respectively; ($p<0.0001$). But cytoplasmic beta-catenin expression was low in benign lesion as compared to malignant lesion (IS): 2.07±3.46 versus 5.35±3.14), respectively; ($p<0.0001$). However, nuclear beta-catenin expression was extremely low in benign lesions than in malignant lesions (0.08±0.47 versus 1.90±3.49) respectively; ($p<0.0016$). This difference was statistically significant [Table/Fig-3].

Bhattacharya I et al., observed statistically significant correlation between IHC score and localisation of beta-catenin showing gradual shift from membrane localisation to nuclear positivity ($p<0.004$) [17]. This study was concordant with present study. Gao ZH et al., observed that membranous expression was exclusively seen in tumour center whereas in invasive fronts, nuclear beta-catenin level was significantly increased [10].

Limitation(s)

Lack of significant association between beta-catenin expression and grading of tumour may be due to presence of lesser number of grade III tumours in our study.

CONCLUSION(S)

In the present study, beta-catenin nuclear expression was found to be high in left colon and rectum. Beta-catenin scoring was found to be have statistically significant difference with stage of tumour. High cytoplasmic and nuclear positivity was observed more in early stages of tumour. Membranous expression of beta-catenin was exclusively preserved in benign lesions whereas most of the malignant lesions showed cytoplasmic and/or nuclear expression. This was statistically significant. To conclude, the study demonstrated the gradual transition in expression of beta-catenin from a predominantly membranous expression to subsequent positivity in cytoplasmic or nuclear location as it progressed from normal colorectal tissue to polyps and benign, premalignant lesions to malignant neoplasms.

REFERENCES

[1] Bray F, Ferlay J, Soerjomataram I, Siegel RL, Torre LA, Jemal A. Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. Cancer J Clin. 2018;68:394-424.

[2] Siegel RL, Miller KD, Jemal A. Cancer statistics, 2019. CA Cancer J Clin. 2019;69:07-34.
[3] International Agency for Research on Cancer WHO. Cancer today. IARC Web site. https://gco.iarc.fr/today/. Accessed January 21, 20197.
[4] Compton C, Fenoglio-Preiser CM, Pettigrew N, Fielding LP. American Joint Committee on Cancer Prognostic Factors Consensus Conference: Colorectal Working Group. Cancer. 2000;88:1739-57.
[5] Boonpipattanapong T, Chewatanakornkul S. Preoperative carcinoembryonic antigen and albumin inpredicting survival in patients with colon and rectal carcinomas. J Clin Gastroenterol. 2006;40:592-95.
[6] Morin PJ. Beta-catenin signaling and cancer. Bioessays. 1999;21:1021-30.
[7] Behrens J, von Kries JP, Kuhl M, Bruhn L, Wedlich D, Grosschedl R, et al. Functional interaction of beta-catenin with the transcription factor LEF-1. Nature. 1996;382:638-42.
[8] Weisburger JH. Causes, relevant mechanisms, and prevention of large bowel cancer. Seminar in Oncol. 1991;18:316-36.
[9] Sinicrope FA, Ruan SB, Cleary KR, Stephens LC, Lee JJ, Levin B. bcl-2 and p53 oncoprotein expression during colorectal tumourigenesis. Cancer Res. 1995;55:237-41.
[10] Gao ZH, Lu C, Wang MX, Han Y, Guo LJ. Differential β-catenin expression levels are associated with morphological features and prognosis of colorectal cancer. Oncology Letters. 2014;8(5):2069-76.
[11] Zhu JL, Song YX, Wang ZN, Gao P, Wang MX, Dong YL, et al. The clinical significance of mesenchyme forkhead 1 (FoxC2) in gastric carcinoma. Histopathology. 2013;62:1038-48.
[12] Morin PJ, Sparks AB, Korinek V, Barker N, Clevers H, Vogelstein B, et al. Activation of β catenin-Tcf signaling in colon cancer by mutations in β-catenin or APC. Science. 1997;275:1787-90.
[13] Wong SC, Lo ES, Lee KC, Chan JK, Hsiao WL. Prognostic and diagnostic significance of β catenin nuclear immunostaining in colorectal cancer. Clin Cancer Res. 2004;10(4):1401-08.
[14] Abdulrahman ZA, Ismael AT, Jalal JA, Alnuaimy WM. Significance of B-catenin Immunohistochemical Expression in Colorectal Carcinoma. Journal of Kurdistan Board of Medical Specialties. 2018;1(4):51-56.
[15] Yoshida N, Kinugusa T, Oshima K, Yuge K, Ohchi T, Fujino S, et al. Analysis of Wnt and beta-catenin expression in advanced colorectal cancer. Anticancer Research. 2015;35(8):4403-10.
[16] Wanitsawan W, Kanngurn S, Boonpipattanapong T, Sangthong R, Sangkhathat S. Overall expression of beta-catenin outperforms its nuclear accumulation in predicting outcomes of colorectal cancers. World J. Gastroenterol: WJG. 2008;14(39):6052.
[17] Bhattacharya I, Barman N, Maiti M, Sarkar R. Assessment of beta-catenin expression by immunohistochemistry in colorectal neoplasms and its role as an additional prognostic marker in colorectal adenocarcinoma. Medicine and Pharmacy Reports. 2019;92(3):246.

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