OncoTargets and Therapy

Dovepress
open access to scientific and medical research

Open Access Full Text Article

CASE REPORT

c.1439delA frameshift deletion mutation in familial adenomatous polyposis

This article was published in the following Dove Press journal:
OncoTargets and Therapy

De-Jian Ma[1]
Bi-Shi Wang[2]
Jin-Bo Yue[3]
Zeng-Jun Li[2]
Yan-Lai Sun[2]

[1]Department of Surgical Oncology, School of Medicine and Life Sciences, University of Jinan-Shandong Academy of Medical Sciences, Jinan 250062, China; [2]Department of Gastrointestinal Cancer Surgery, Shandong Cancer Hospital Affiliated to Shandong University, Shandong Academy of Medical Sciences, Jinan 250117, China; [3]Department of Radiation Oncology, Shandong Cancer Hospital Affiliated to Shandong University, Shandong Academy of Medical Sciences, Jinan 250117, China

Abstract: Familial adenomatous polyposis (FAP) is a rare autosomal dominant genetic disease related to germline mutations of the *APC* gene. The clinical features of this disease most commonly include hundreds of adenomas or polyps. If not treated in a timely fashion, FAP can eventually result in colorectal carcinoma. In this report, clinical manifestations, family history, relevant auxiliary examinations and gene detection from patient blood led us to discover a novel frameshift mutation in exon 12 of the *APC* gene. The deletion of adenine in c.1439 resulted in the formation of codon 480. The occurrence of this frameshift deletion may lead to inexpressibility of the main functional regions in *APC* and may affect gene function. In addition, colonoscopy and histopathology showed malignant changes in the colon and rectum. There have been no reports of this frameshift mutation, but it can be considered in case of *APC* mutations and FAP in patients with clinical manifestations; auxiliary examination may be related, and it may be used as a reference for preventive clinical treatment in the future.

Keywords: familial adenomatous polyposis, mutation, *APC* gene, exon 12, colorectal cancer

Core tip

Familial adenomatous polyposis (FAP) is an autosomal dominant condition affecting nearly 1 in 5,000 people and accounts for only about 1% of all colorectal cancers. There are many mutations in the *APC* gene, but the *APC* locus c.1439 adenine deletion in this study also led to the formation of codon 480, leading to disease. This locus has not been recorded to date.

Introduction

Familial adenomatous polyposis (FAP) is an autosomal dominant genetic disease related to germline mutations of the *APC* gene. The clinical manifestations of FAP are found both in and outside the colon; colonic manifestations include hundreds of adenomas or polyps, and external manifestations include polyps and adenomas in the upper gastrointestinal tract, desmoid tumors, congenital hypertrophy retinal pigment epithelium, osteomas, sebaceous cysts (epidermoid cysts), hepatoblastoma, fundic gland polyposis, dental abnormalities and malignancies of the central nervous system.[1] The *APC* gene is a large "housekeeping" gene. This gene contains an 8,535 bp open readable frame with 15 exons and 6 mutable expressions and is located on the 5q21 chromosome. Exon 15 only contains 6,571 bp, constituting 77% of the coding region. It is the largest known exon, encoding 2,843 amino acids.[2] Mutation types of the *APC* gene include micro-deletions and frameshift mutations. The former include nonsense mutations, dislocation mutations and splicing errors, and the latter include deletions and insertions.[3] There are many mutations in the *APC* gene, and more than 60% of the

Correspondence: Yan-Lai Sun
Department of Gastrointestinal Cancer Surgery, Shandong Cancer Hospital Affiliated to Shandong University, Shandong Academy of Medical Sciences, 440 Jiyan Road, Jinan 250117, China
Tel/fax +86 531 6762 6830
Email sunyanlai@126.com

submit your manuscript | www.dovepress.com
Dovepress
http://dx.doi.org/10.2147/OTT.S183153

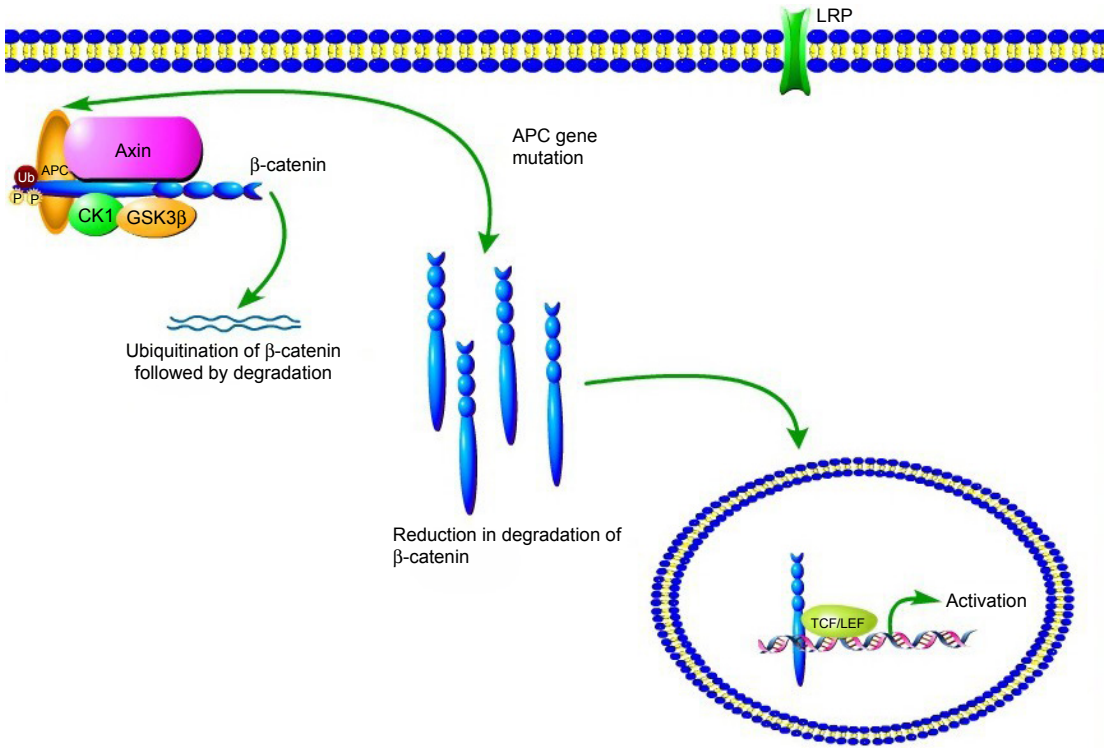

Figure 1 Wnt signal transduction mediated by β-catenin.
Notes: β-catenin is separated by a polyprotein "destruction complex" and is phosphorylated and ubiquitinated, leading to degradation. However, after mutation of the *APC* gene, the degradation of β-catenin decreased, leading to the accumulation of β-catenin in the cytoplasm, eventually transferring to the nucleus and interacting directly with the TCF/LEF transcription factor and other transcriptional coactivators. Thus, the expression of target genes is initiated.
Abbreviation: TCF/LEF, T-cell factor/lymphoid enhancer factor.

mutations are located at the 5′ end of exon 15. Approximately 65% of somatic cell mutations are concentrated between codons 1281 and 1556, ~10% of the coding region; this is called a mutation cluster region. A variety of mutations are caused by the deletion or insertion of 1–8 bp. Approximately 95% of the mutations lead to the formation of an early termination codon downstream, resulting in the truncation of *APC* protein changes, thus weakening the inherent inhibition of *APC* on cell proliferation and leading to the dysfunction of *APC* protein.[4] The *APC* protein combines with β-catenin to form complexes, leading to β-catenin degradation. Therefore, as a result of the *APC* gene mutations, *APC* protein function is damaged and β-catenin not destroyed by ubiquitin accumulates in the nucleus. Thus, reducing the degradation of β-catenin activates the WNT signaling pathway activating transcription factors T-cell factor/lymphoid enhancer factor, and increasing the expression of cyclinD1, MMP9 and other cancer genes. Ultimately, this process leads to uncontrolled proliferation and the development of colon cancer (Figure 1).[5]

Case report

A 33-year-old male patient was admitted to the hospital with abdominal distension, abdominal pain and altered bowl habits, and stool frequency of every 2–3 days. Digital rectal examination revealed a mass 7 cm from the anus, presumed to be the lower edge of a protuberant lesion, the upper edge of which could not be palpated; the rectal mucosa revealed multiple polyps on palpation of ~0.8 cm. There was a family history of FAP and colon cancer in first-degree relatives (but total proctocolectomy had been performed). Colonoscopy revealed several rectal and colorectal polyps, as well as ~60 cm of colon protuberant lesions (Figure 2). The pathological manifestations of colon and rectum tumor were adenocarcinoma. The colonic tumor was located in the hepatic flexure of the colon, of a hard protruding type, and a rectal tumor was located in the middle part of the rectum, of a hard protruding type. The remainder of the colon and rectum were covered with polyps (0.2–1.5 cm in diameter) (Figure 3). Pathology revealed a large number of tubular villous adenomas between the differentiated adenocarcinoma of the colon and the middle-differentiated adenocarcinoma of the rectum. Lymph node metastases around the mesentery were seen in four cases (4/27) (Figure 4). TNM classification of the hepatic flexure of the colon tumor showed pT4aN2M0, IIIC and the rectum tumor showed pT4bN0M0, IIB.

We performed KRAS/NRAS/BRAF joint genetic detection on two malignant lesions; the KRAS exon 2 of hepatic

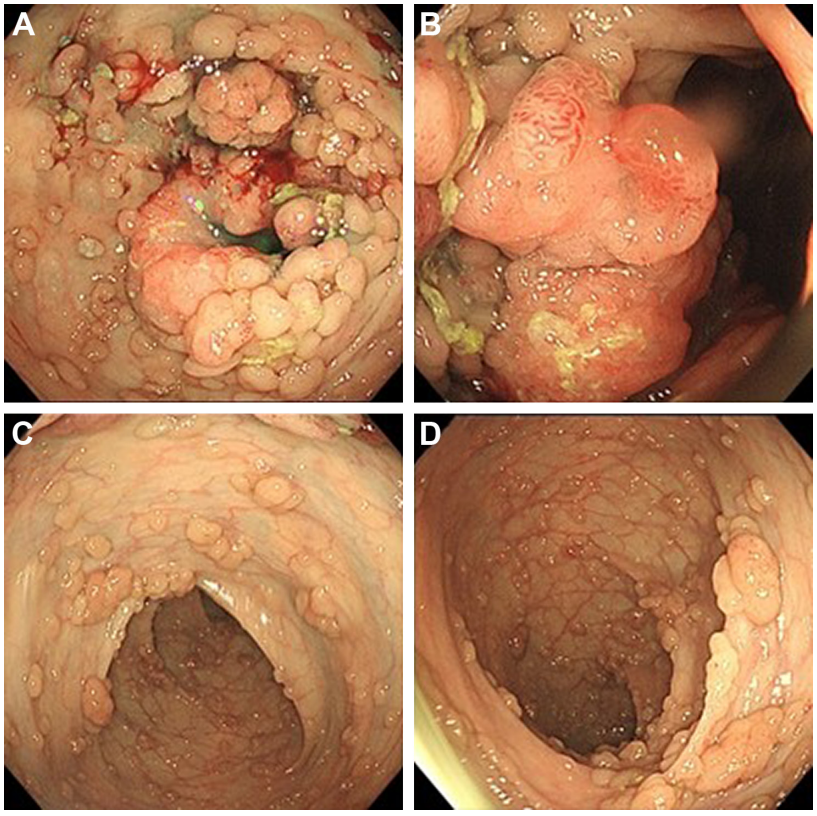

Figure 2 Colonoscopy.
Notes: (**A**) Hepatic flexure of colon malignant lesions; (**B**) rectal malignant lesions; (**C**, **D**) a large number of colorectal polyps.

flexure of colon malignant lesions was a mutant and the remainder were wild type (Table 1); the malignant rectal lesions were wild type (Table 2). Peripheral blood gene detection showed *APC*, AXIN2, EPCAM, MLH1, MLH3, MSH2, MSH2, MSH6, MUTYH, PMS1, PTEN, SMAD4, BMPR1A and TP53 genes. Only exon 12 in the *APC* gene had a deletion mutation (Table 3). At present, this mutation had not been recorded in the COSMIC database, and functional studies of amino acid number 480 are unknown (Table 4). However, a frameshift deletion may lead to the non-expression of the major functional regions and may influence the gene function of *APC*. The frequency of de novo mutations in the region near c.1439delA is very high. In addition, we found that a deletion site NM_000038.5(*APC*): c.(1409−?)_(1743+?)del Simple (variant ID217926) near the region was pathogenic in the ClinVar database. Therefore, in this analysis, the mutation detected was likely a pathogenic mutation.

The patient was cleared on preoperative examination without contraindications. Total colorectal resection was performed with laparoscopic assistance. After surgery, the patient's clinical symptoms were relieved and the surgery was well tolerated. But there was no accident. He eventually

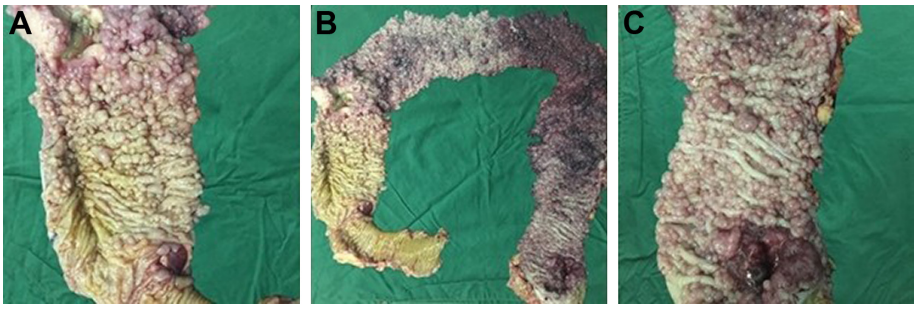

Figure 3 Surgical specimens.
Notes: (**A**) Malignant tumor found in the hepatic flexure of the colon; (**B**) a large number of polyps were found in the rest of the colon and rectum; (**C**) malignant tumor found in the rectum.

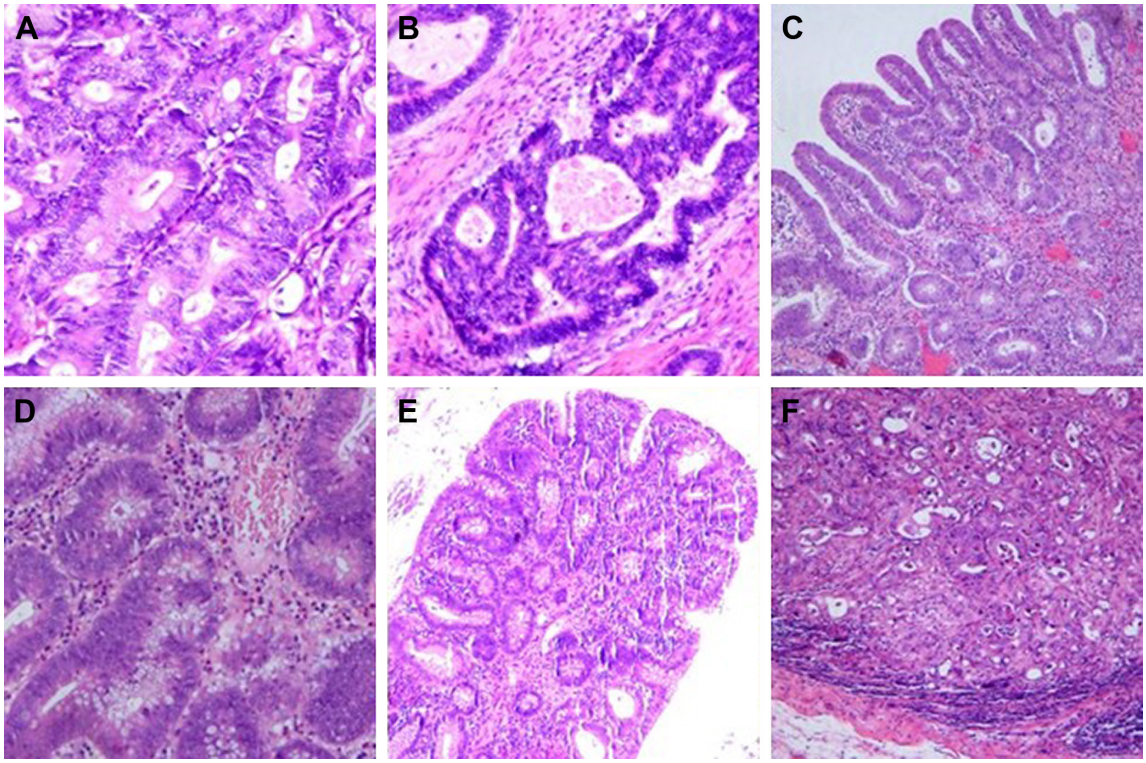

Figure 4 Histopathology.
Notes: (**A**, **B**) Carcinoma infiltrating the serosa, with ulcerative moderately differentiated adenocarcinoma; (**C**–**E**) a tubular-villous adenoma colonic polyp; (**F**) lymph node metastasis. Scale bar 100 μm; magnification ×400.

recovered and was transferred from the hospital. So far, he has lived for 8 months.

Discussion

FAP is an autosomal dominant hereditary disease, more common with familial inheritance. This disease also arises sporadically in patients with no family history.[6] The main pathological change in FAP is the presence of dozens to hundreds of polyps of various sizes in the intestine, with thousands of polyps present in severe cases.[7] There are no substantial symptoms at the onset of the disease. With the increase and expansion of polyps, the patients may develop abdominal discomfort, abdominal pain and mucopurulent stool, as well as an increase in defecation frequency.[8] Over time, colorectal cancer can result, in which case genetic detection is useful because some mutations in the *APC* gene may be closely linked to the genetic heterogeneity and severity of the disease.[9] The mutations of the *APC* gene are numerous and variable, but the most common changes are code mutations and single base-pair changes, premature termination codons, which lead to the production of a truncated protein.[10] The main role of the *APC* protein

Table 1 Hepatic flexure of colon malignant lesions KRAS/NRAS/BRAF joint mutation detection

Test items	Exons/codons	Type of mutation	Test results
KRAS/NRAS/BRAF joint mutation detection	KRAS Exon-2	G12S, G12D	Wild type
	KRAS Exon-2	G12C, G12R, G12V, G12A, G13C	Mutant
	KRAS Exon-2	G13D	Wild type
	KRAS Exon-3	Q61L, Q61R, Q61H	Wild type
	KRAS Exon-4	K117N, A146T, A146V, A146P	Wild type
	NRAS Exon-2	G12D, G12S	Wild type
	NRAS Exon-2	G13D	Wild type
	NRAS Exon-2	G13R, G12C, G12V, G12A, G13V	Wild type
	NRAS Exon-3	Q61R, Q61K, Q61L, Q61H	Wild type
	NRAS Exon-4	A146T	Wild type
	BRAF Exon-15	V600E	Wild type

Table 2 Rectal malignant lesions KRAS/NRAS/BRAF joint mutation detection

Test items	Exons/codons	Type of mutation	Test results
KRAS/NRAS/BRAF joint mutation detection	KRAS Exon-2	G12S, G12D	Wild type
	KRAS Exon-2	G12C, G12R, G12V, G12A, G13C	Wild type
	KRAS Exon-2	G13D	Wild type
	KRAS Exon-3	Q61L, Q61R, Q61H	Wild type
	KRAS Exon-4	K117N, A146T, A146V, A146P	Wild type
	NRAS Exon-2	G12D, G12S	Wild type
	NRAS Exon-2	G13D	Wild type
	NRAS Exon-2	G13R, G12C, G12V, G12A, G13V	Wild type
	NRAS Exon-3	Q61R, Q61K, Q61L, Q61H	Wild type
	NRAS Exon-4	A146T	Wild type
	BRAF Exon-15	V600E	Wild type

is to interact with β-catenin and E-cadherin to affect cell adhesion and intercellular signal transduction; *APC* is a negative regulator of β-catenin. High levels of β-catenin can cause *APC* protein to be phosphorylated through GSK3β, thereby increasing the efficiency of β-catenin degradation and keeping the β-catenin level in a state of equilibrium in the cytoplasm. One of the crucial consequences of the mutated *APC* gene in tumorigenesis is a lack of regulation of β-catenin. Because the β-catenin-binding site of *APC* protein is highly conserved, it has been proven that the ability of the mutant *APC* protein to form complexes with β-catenin is very important in tumorigenesis.[11] In addition, the *APC* protein binds to microtubules and plays an important role in cell division and migration. *APC* regulates the cell cycle by regulating the activity of cyclin-dependent cell cyclin kinase complex; it also mediates colon adenoma formation by inducing apoptosis. Therefore, *APC* is known as the molecular "gatekeeper" of the integrity of the colonic epithelium. Mutations of the *APC* gene can change the balance between the *APC* protein, and β-catenin and E-cadherin, resulting in changes in the adhesion and contact inhibition signal transduction between cells and the cell matrix. The imbalance between cell division and cell death leads to uncontrolled growth, becoming a rate-limiting molecular factor in colorectal cancer.[12] The genetic and phenotypic analyses of FAP show that more than 5,000 polyps have severe polyposis with mutations usually occurring between codons 1250 and 1464. The mutation of codon 1309 is likely to cause particularly severe phenotypes and early onset. In contrast, attenuated FAP(AFAP) of less than 100 adenomatous polyps (*APC*) is caused by mutations in the 5′ or 3′ terminals of the *APC* gene or by mutations in the splice region of exon 9.[13] Congenital retinal pigment epithelial cell hypertrophy occurs only in patients with mutations between codon 457 and codon 1444.[14] Mutations with extra-polyp phenotypes, such as dermatofibroma, osteoma, epidermoid cysts and upper gastrointestinal polyps, were found to be the most common mutations between codons 1445 and 1578 or codons 1395 and 1493.[15] In our patient, we found masses of various sizes distributed over the colon and rectum, and malignant lesions were noted in the colon and the rectum. We used laparoscopic-assisted proctocolectomy and ileal pouch–anal anastomosis because it completely removed the colonic mucosa as a potential source of cancer, reducing the risk of cancer and maintaining the intestinal function.[16]

Table 3 Hereditary colorectal cancer gene detection

Test items	Detection gene	Test results
Hereditary colorectal cancer gene detection	*APC*	+
	AXIN2	–
	EPCAM	–
	MLH1	–
	MLH3	–
	MSH2	–
	MSH6	–
	MUTYH	–
	PMS1	–
	PMS2	–
	SKT11	–
	PTEN	–
	SMAD4	–
	BMPR1A	–
	TP53	–

In our genetic analysis, we found that there was a new frameshift mutation in exon 12 of the *APC* gene. The deletion

Table 4 The frameshift mutation in exon 12 c.1439 is related to the deletion of adenine

Gene	Exon	Nucleotide change	Amino acid change	Mutation type	Zygosity
APC	12	c.1439delA	p.Q480fs	Frameshift deletion	Heterozygous mutation

of adenine in c.1439 resulted in the formation of amino acid 480. According to the HGMD dataset (http://www.hgmd.cf.ac.uk/ac/index.php), the primary causes of mutations in the *APC* gene are micro-deletions, leading to changes in the open reading frame and the formation of truncated gene products. In the ZJU-CGGM database (http://www.genomed.org/lovd2/variants_statistics.php), we found that the *APC* gene in c.(1313–?)_(1408+?)del caused a change in the 470 codon; in the ClinVar database (https://www.ncbi.nlm.nih.gov/clinvar/), we found the NM_000038.5(*APC*): c.(1409–?)_(1743+?)del Simple (variant ID217926). Therefore, it can be seen that the frequency of new mutations is very high in the vicinity of c.1439delA, and some new mutations in the vicinity may be pathogenic. The *APC* gene c.1438C>T (p.Gln480*) was found in the LOVD database (http://www.lovd.nl/3.0/home), resulting in the early termination of codon 480, followed by the formation of truncated gene products; Tsukanov et al reported a mutation of the *APC* gene c.1438C>T (p.Gln480X); c.1438 thymidine replaced cytosine in the formation of termination codon 480 and caused the formation of truncated proteins.[17] Similarly, the *APC* locus c.1439 adenine deletion in this study also led to the formation of codon 480, leading to disease. This locus has not been recorded to date.

Finally, we can conclude that a pathogenic mutation was identified, according to the relevant databases and literature reports and the clinical manifestations of the patient and the postoperative histopathological and pathological examination reports. For the first-degree family members, they can understand their genetic background through genetic testing. They would be able to get personalized health consultation management services. Genetic testing is necessary for early detection, early prevention and early treatment of diseases. This method can improve the quality of life and prolong life. We believe that the *APC* mutation spectrum increased with the increase in FAP history. These type of studies will facilitate early identification of FAP patients from diverse populations and will lead to preventative treatment of patients with associated *APC* gene mutations to improve survival rates.

Ethical approval and consent

Written informed consent was obtained from relatives of the patient and patient for publication of this case report and accompanying images. Ethical approval for the publication of this case was obtained from the ethical committee of the Cancer Center of Shandong Cancer Hospital Affiliated to Shandong University.

Acknowledgments

We express our thanks to grants from science and technology development plan of Jinan, No. 201401253; medical and health technology development program of Shandong province, No. 2015WS0149 and No. 2015WS0197; Postdoctoral Science Foundation of China, No. 2017M612317; national key research and development plan, No. 2018YFC0114707; Shandong province key research and development, No. 2018GSF118047.

Disclosure

The authors report no conflicts of interest in this work.

References

1. Russo A, Catania VE, Cavallaro A, et al. Molecular analysis of the *APC* gene in Sicilian patients with familial adenomatous polyposis (F.A.P.). *Int J Surg*. 2014;12(Suppl 2):S125–S129.
2. Schirosi L, Pellegrino M, Tarantino P, Mauro S, Tinelli A, Greco M. A new germline stop codon mutation in exon 15 of the *APC* gene predisposing to familial adenomatous polyposis. *Int J Biol Markers*. 2013;28(4):405–408.
3. Eser B, Yıldar M. Two different nucleotide substitutions of *APC* gene in a family with familial adenomatous polyposis. *J Surg Case Rep*. 2015;2015(9):rjv118.
4. Moreira-Nunes CA, Alcântara D, Lima-Júnior SF, et al. Presence of c.3956delC mutation in familial adenomatous polyposis patients from Brazil. *World J Gastroenterol*. 2015;21(31):9413–9419.
5. Park MG, Kim JS, Park SY, et al. MicroRNA-27 promotes the differentiation of odontoblastic cell by targeting APC and activating Wnt/β-catenin signaling. *Gene*. 2014;538(2):266–272.
6. Li FF, Liu Z, Yan P, et al. Identification of a novel mutation associated with familial adenomatous polyposis and colorectal cancer. *Int J Mol Med*. 2015;36(4):1049–1056.
7. Leoz ML, Carballal S, Moreira L, Ocaña T, Balaguer F. The genetic basis of familial adenomatous polyposis and its implications for clinical practice and risk management. *Appl Clin Genet*. 2015;8:95–107.
8. Jahng J, Yoon SJ, Park H. A novel germline mutation in Exon 15 of the *APC* gene in attenuated familial adenomatous polyposis: a report of two cases. *Gut Liver*. 2013;7(1):120–125.
9. Li BQ, Liu PP, Zhang CH. Correlation between the methylation of APC gene promoter and colon cancer. *Oncol Lett*. 2017;14(2):2315–2319.
10. Rashid M, Fischer A, Wilson CH, et al. Adenoma development in familial adenomatous polyposis and *MUTYH*-associated polyposis: somatic landscape and driver genes. *J Pathol*. 2016;238(1):98–108.
11. Grainger S, Willert K. Mechanisms of Wnt signaling and control. *Wiley Interdiscip Rev Syst Biol Med*. 2018;10(5):e1422.
12. Hankey W, Frankel WL, Groden J. Functions of the APC tumor suppressor protein dependent and independent of canonical WNT signaling: implications for therapeutic targeting. *Cancer Metastasis Rev*. 2018;37(1):159–172.
13. Chen QW, Zhang XM, Zhou JN, et al. Analysis of small fragment deletions of the *APC* gene in Chinese patients with familial adenomatous polyposis, a precancerous condition. *Asian Pac J Cancer Prev*. 2015;16(12):4915–4920.
14. Liu Q, Li X, Li S, et al. Three novel mutations of *APC* gene in Chinese patients with familial adenomatous polyposis. *Tumour Biol*. 2016;37(8):11421–11427.
15. Li H, Zhang L, Jiang Q, Shi Z, Tong H. Identification a nonsense mutation of *APC* gene in Chinese patients with familial adenomatous polyposis. *Exp Ther Med*. 2017;13(4):1495–1499.

16. Dalavi SB, Vedpalsingh TH, Bankar SS, Ahmed MH, Bhosale DN. Familial adenomatous polyposis (FAP) – A case study and review of literature. *J Clin Diagn Res*. 2015;9(3):PD05–PD06.

17. Tsukanov AS, Pospekhova NI, Shubin VP, et al. Mutations in the *APC* gene in Russian patients with classic form of familial adenomatous polyposis. *Russ J Genet*. 2017;53(3):369–375.