Sporadic desmoid tumor in an Ashkenazi patient homozygous for the *APC*I1307K* gene mutation

PETER ZAUBER[1], MARLENE SABBATH-SOLITARE[2], STEPHEN P. MAROTTA[3], RONALD CHAMBERLAIN[4], GEORGE CHONG[5], WILLIAM D. FOULKES[6] & TIMOTHY BISHOP[7]

[1]*Department of Medicine, Saint Barnabas Medical Center, Livingston, New Jersey, USA, Department of Pathology, Saint Barnabas Medical Center, Livingston, New Jersey, USA,* [2]*Department of Pathology, Saint Barnabas Medical Center, Livingston, New Jersey, USA,* [3]*Department of Surgery, Saint Barnabas Medical Center, Livingston, New Jerey, USA,* [4]*Molecular Diagnostics Laboratory, Jewish General Hospital, Montreal, Canada,* [5]*Division of Medical Genetics, McGill University, Montreal, Canada and* [6]*Genetic Epidemiology Division, St. James's University Hospital, Leeds, England, and* [7]*Cancer Research, UK*

To the Editor

Desmoid tumors are benign growths of myofibroblasts that occur in musculoaponeurotic tissue. They occur in the general population with an incidence of approximately only 2 cases per million. However, 12 to 15% of patients with Familial Adenomatous Polyposis (FAP) develop a desmoid tumor. FAP is a dominantly inherited disorder due to an inactivating germ line mutation occurring within the Adenomatous Polyposis Coli (*APC*) gene. The majority of desmoid tumors, both sporadic and in FAP patients occur within the abdomen. We report a sporadic desmoid tumor in a patient determined to be homozygous for the *APC*I1307K* gene mutation.

Observation

The patient is a 28-year-old Ashkenazi female who developed the sudden onset of fever and right lower abdominal pain. Her past medical history was remarkable only for gastrointestinal reflux disease. One grandfather had colon carcinoma and one grandmother had breast carcinoma. She presented to an emergency room where a diagnosis of acute appendicitis led to a laparoscopic appendectomy. The pain persisted after surgery and a CT scan showed an 8 cm by 9 cm mesenteric mass. This was excised during an open procedure. Pathology revealed a mesenteric desmoid tumor (Figure 1).

Subsequent colonoscopy revealed no adenomas or non-adenomatous polyps.

Methods

APC Gene Studies

DNA was available from peripheral blood white cells as well as normal colonic tissue and the desmoid tumor. The *APC* gene was sequenced between codons 1270 to 1492 using the overlapping primer sets 5′-CCA AGA AAC AAT ACA GAC TTA TTG TG-3′ (sense 1) plus 5′-ATG AGT GGG GTC TCC TGA AC-3′ (antisense 1) and 5′-TTC TTC AGG AGC GAA ATC TC-3′ (sense 2) plus 5′-TCC ATC TGG AGT ACT TTC TGT G-3′ (antisense 2). Multiplex ligation dependent probe amplification (MLPA) was performed on both peripheral blood lymphocyte DNA as well as normal tissue DNA. The probe mix included probes for each of the 15 coding exons of the APC gene, alternative exon, and three probes for the promotor region of the *APC* gene. Thirteen probes for other loci located on different chromosomes were used as control. Quantification was performed by SoftGenetics using Gene Mapper software. The protein truncation test (PTT) was also applied to the two normal DNA samples. The exon 3 region of the beta-catenin gene was amplified using PCR with the primers 5′-TTT CCA ATC TAC TAA TGC TAA TAC TG-3′ and 5′-CTG CAT TCT GAC TTT CAG TAA GG-3′.

Correspondence: Peter Zauber, 22 Old Short Hills Road, Livingston, New Jersey 07039, USA. Tel: 973 533 9299. Fax: 973 992 7648. E-mail: pzauber@gmail.com

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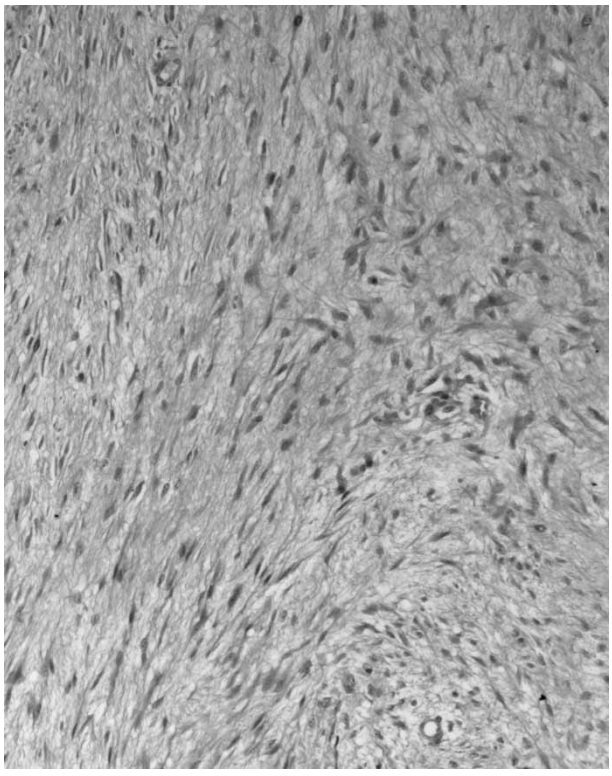

Figure 1. Hematoxylin and eosin stained tissue from abdominal desmoid showing a proliferation of bland spindle cells in a connective tissue background (×40).

Results

Normal tissue

Our patient is homozygous for a germ line mutation at codon 1307, a T to A transversion at nucleotide 3920 (Figure 2). No other mutations were detected in this region. DNA from peripheral blood cells was homozygous for several APC markers but was heterozygous for the marker D5S492. Sequencing confirmed the presence of one or two copies of the I1307K mutation but no wild type sequence. No duplication or deletion was detected by MLPA, thereby indicating that the mutation at codon 1307 was indeed homozygous and not hemizygous. Normal tissue DNA analyzed by the protein truncation test revealed no small deletions, insertions or frameshifts. Additional testing indicated the patient was not a carrier of the *MSH2** or *MLH1*D132H* germ line mutations.

Desmoid tissue

The desmoid tumor DNA showed only the *APC*I1307K* mutation and no other mutation or loss in the region of exons 15G and 15H of the *APC* gene. The patient's tumor and normal colonic tissue DNA were sequenced for the two known 'hot spots', codons 41 and 45, of the *beta-catenin* gene. The

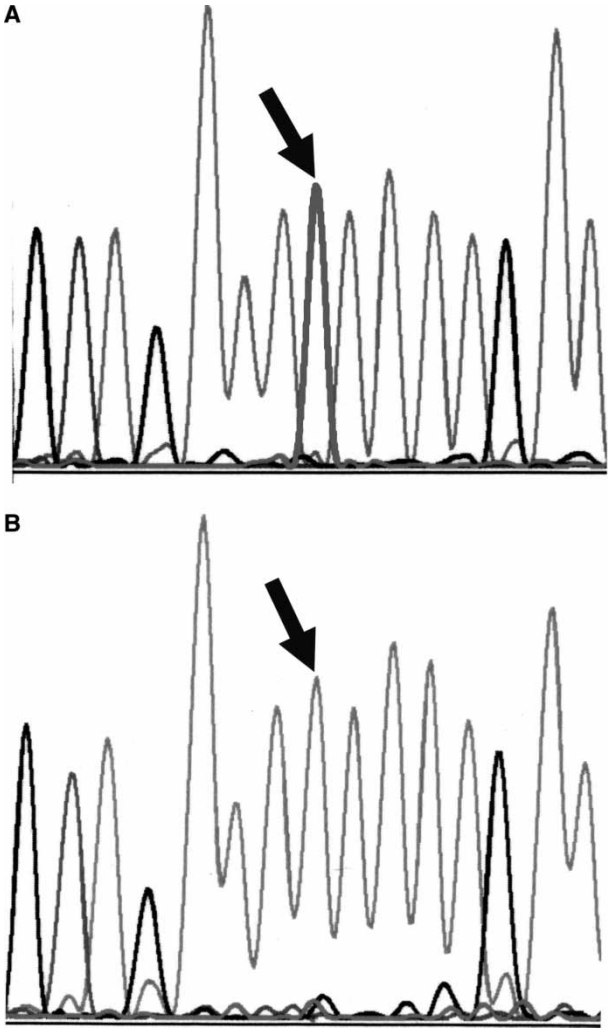

Figure 2. DNA sequence analysis. A. Normal DNA. The arrow indicates the wild-type pattern with a single thymine peak at the second nucleotide position of codon 1307. B. Patient's desmoid DNA with germ line homozygous mutation. The arrow indicates only the presence of adenine at the second nucleotide position of codon 1307.

desmoid tumor DNA revealed a transition in codon 41 of adenine to guanine in the first position, ACC to GCC (data not shown) that was not present in the normal colonic tissue.

Discussion

The *APC*I1307K* mutation is a T to A transversion at nucleotide 3920 that converts the sequence AAATAAAA to (A)8, resulting in a substitution of lysine for isoleucine at codon 1307. This mutation by itself is not associated with a stop codon and does not appear to affect the protein function. However, the (A)8 tract is unstable and may increase the rate of somatic mutations within this region, leading to frame shifts and APC gene inactivation [1]. Studies have suggested the *APC*I1307K* mutation confers

an increased risk for the development of colorectal neoplasms [2]. Additionally, codon 1307 is within the region coding for the segment of the APC protein that binds to and exerts control over *beta-catenin* gene expression [3]. Loss of APC protein control may result in accumulation of beta-catenin protein and increased expression of several proliferation genes controlling cell adhesion and cell signaling. Desmoid tumors are frequent in FAP patients with germ line *APC* gene mutations occurring beyond codon 1309, particularly in the region of codons 1445–1578 [4,5]. Latchford et al. demonstrated both germ line and somatic *APC* mutations in 17 of 32 FAP patients with demoid tumors but did not report *beta-catenin* gene analyses on these tumors [6]. They describe the loss of those areas of the APC protein that function as beta-catenin degradation sites resulting from a truncating APC mutation. These sites are coded by codons in the 1265–2000 region of the APC gene. Our data do not suggest a truncating mutation in this region.

Desmoids have been reported in non-FAP families with germ line *APC* gene mutations at codons 1924 and 1962 [7,8]. Somatic mutations in the *beta-catenin* gene also have been identified as important in the development of desmoid tumors [9,10]. Tejpar et al. performed mutational analysis of both the *APC* and *beta-catenin* genes in 42 sporadic desmoids, the largest reported study [11]. Nine tumors had mutations in the *APC* gene, all between codons 1324 and 1567 and resulting in an early stop codon. In addition, ten tumors revealed a mutation in the *beta-catenin* gene, a codon 41 transition of adenine to guanine. This substitution has been shown to produce a stabilized beta-catenin protein product [12]. Twelve additional tumors contained a point mutation in codon 45 of the *beta-catenin* gene.

It is possible that the *APC* gene mutation in our patient is uninvolved in the oncogenic process. However, the development of many tumors involves the accumulation of several genetic influences. Since the effects of *beta-catenin* and *APC* gene mutations can be additive, it is reasonable to postulate that there is an interactive oncogenic effect between the *beta-catenin* gene point mutation and the *APC*I1307K* mutation in our patient.

Our observation would be strengthened had we performed full mutational analysis of the *APC* and *beta-catenin* genes. However, the effect of the *APC*I1307K* mutation is limited to the area around codon 1307, which we did sequence. We also sequenced the major hot-spot for *beta-catenin* gene mutations. Immunohistochemical staining for beta-catenin has been shown to be sensitive but not a specific test for dermoids [13] and therefore we did not use immunohistochemistry.

The mechanism by which homozygosity for the *APC*I1307K* mutation may have contributed to the development of a desmoid tumor is not yet clear. It is possible that homozygosity, similar to heterozygosity, fosters additional *APC* point mutation(s) that we did not detect, but this is unlikely given our detailed analyses of both the somatic and germ line *APC* genes. We suggest the possibility that homozygosity of the *APC*I1307K* mutation may have a qualitatively different effect than heterozygosity and may lead directly to abnormal *APC* function. Homozygosity for the *APC*I1307K* mutation is infrequent, just two of 5081 Ashkenazi Jews in one large study [2] and two of 429 in another [14]. A full description of the associated phenotype and genotype of *APC*I1307K* homozygosity has yet to be defined. In conclusion, we suggest the possibly that homozygosity of the germ line *APC*I1307K* gene mutation in our patient contributed, along with the observed somatic *beta-catenin* gene mutation, to the development of the desmoid tumor.

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Capecitabine-induced oromandibular dystonia: A case report and literature review

JOANNE Y. Y. NGEOW[1], KUMAR M. PRAKASH[2], BALRAM CHOWBAY[3], SWEE TIAN QUEK[4] & SU-PIN CHOO[1]

[1]*Department of Medical Oncology, National Cancer Centre, Singapore,* [2]*Department of Neurology, Singapore General Hospital, Singapore,* [3]*Laboratory of Clinical Pharmacology, Division of Medical Sciences, National Cancer Center, Singapore and and* [4]*Department of Diagnostic Imaging, National Cancer Centre, Singapore*

We report a rare case of capecitabine-induced oromandibular dystonia and a summary of the cases reported in the literature.

A 57-year-old Chinese male was diagnosed with a pT3N2 stenosing adenocarcinoma of the rectum in January 2007, for which he underwent a low anterior resection. During staging evaluation, he was found to have a subcentimetre segment 2 liver lesion which was confirmed to be a solitary liver metastasis on positron emission tomography (PET). The plan was to give him two cycles of "neoadjuvant" capecitabine and oxaliplatin given every 3 weeks before resection of the liver metastasis, followed by further chemotherapy and radiotherapy. He was started on cycle 1 of capecitabine 2500 mg/m^2 (days 1–14) and oxaliplatin 130 mg/m^2 (day 1) on February 9, 2007.

Nine days after consuming capecitabine he developed sudden onset of disability to talk and swallow. Clinical examination revealed he had dystonia of tongue and pharyngeal muscles as well as involuntary jaw clenching. He did not have any other focal neurological signs nor associated metabolic abnormalities. He did complain of fatigue and anorexia but did not have features of hand-foot syndrome, haematological nor gastrointestinal toxicities.

Computer tomography of his brain done on the same day showed no obvious abnormalities. However, a magnetic resonance imaging (MRI) scan was done which showed non-enhancing abnormalities with restricted diffusion involving bilateral corona radiate, the centrum semiovale and splenium of corpus collosum with scattered brainstem lesions noted (Figures 1 and 2) consistent with multifocal leukoencephalopathy.

Capecitabine was discontinued on admission. A feeding tube had to be inserted as he had difficulty swallowing and he required a writing board to communicate. After 3 days, his symptoms completely resolved. Blood taken from the patient was analysed for TS 5′ gene polymorphisms and showed class 3 (3RG/3RC) genotype.

He was switched to raltitrexed and oxaliplatin as further therapy and no further neurological events occurred. A repeat MRI of his brain was done on March 20, 2007 (Figure 3), which showed significant improvement in the periventricular leukoencephalopathy. He underwent an uneventful liver resection on April 17, 2007 and remains neurologically asymptomatic.

Capecitabine is a proven oral chemotherapy agent in breast and colorectal cancer, which is increasingly

Correspondence: Su-Pin Choo, Medical Oncology, National Cancer Centre, Singapore 11, Hospital Drive, Singapore 169610. Tel: +65 64368000. Fax: +65 62272759. E-mail: choosupin@gmail.com

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