

Hereditary Persistence of Alpha-Fetoprotein Is Associated with the $-119G>A$ Polymorphism in *AFP* Gene

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ABSTRACT

Alpha-fetoprotein (AFP) is a glycoprotein that is produced by the liver and yolk sac during fetal development. Its levels are usually raised in malignant conditions. Hereditary persistence of AFP (HPAFP) is a rare benign condition with elevated levels of AFP. It is inherited in a dominant mode with complete penetrance and is usually not associated with any clinical disability. We report two individuals with elevated levels of AFP harboring the $-119G>A$ polymorphism in the *AFP* gene. A genetic screening to rule out variants in the *AFP* gene is advised in cases with unexplained persistent AFP levels to avoid inappropriate treatment and surgical options.

INTRODUCTION

Circulating levels of alpha-fetoprotein (AFP), a glycoprotein produced by the liver and the yolk sac during fetal development, decrease upon birth and are negligible in normal adults (>7 ng/mL).¹ However, it is found to be elevated under certain conditions such as pregnancy, neural tube defects, liver cirrhosis, hepatocellular carcinoma, and other malignancies.² Hereditary persistence of AFP (HPAFP) is a rare disorder that does not manifest clinically. The first case of HPAFP was identified in 1983 during an antenatal screening program for spina bifida in a pregnant woman.³ Further testing in other members of the family revealed that the condition is inherited as an autosomal dominant trait with complete penetrance, without being associated with any disease. A later study revealed that a G to A substitution in the 5' untranslated region of the *AFP* gene ($-119G>A$) is associated with HPAFP.⁴ Another study reported 2 other polymorphisms in the 5'UTR of the *AFP* gene ($-55C>A$ and $-65C>T$).⁵ All 3 polymorphisms are found in a region of the 5'UTR that is a potential binding site for the hepatocyte nuclear factor 1 (HNF-1).^{2,6} Since these findings, HPAFP has been reported in more than 20 families worldwide, and persistently elevated serum AFP levels are often misconstrued as serious malignant disorders.^{2,7} A recent study that sequenced 5 regions that are critical for AFP expression in 4 healthy adults with persistently elevated AFP levels of South Korean ethnicity suggested that persistent elevation of AFP could be a heterogeneous condition with or without a hereditary component and may be caused by changes in regions other than transcription regulatory regions.⁸

CASE REPORT

A 64-year-old male from Bangladesh, identified to have elevated levels of serum AFP in 2010 by electrochemiluminescence immunoassay (ECLIA), presented with consistently elevated AFP levels ($>1,100$ ng/mL), with the most recent evaluation showing 1,830 ng/mL. Physical examination revealed no abnormality or swelling anywhere in the body. There was no loss of appetite, sudden weight loss, history suggestive of liver disease, or family history of

ACG Case Rep J 2017;4:e33. doi:10.14309/crj.2017.33. Published online: March 1, 2017.

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malignancy. He underwent cholecystectomy in 2003 for gallstone disease and complained of asthma and sinusitis. He had also recently been diagnosed for type 2 diabetes mellitus. His liver function tests were normal except for a slight elevation of alanine aminotransferase (41 IU/L), and he tested negative for all other viral infections suggestive of hepatitis. Computed tomography of the abdomen and examination of the upper gastrointestinal series and colon were all normal. Ultrasound of the abdomen revealed hepatomegaly with a grade III fatty liver.

Given the consistent elevation of AFP without any associated abnormality, the proband was subjected to genetic analysis to ascertain HPAFP. His immediate family (wife and 2 sons) were also evaluated for elevated serum AFP and other routine medical examinations. The older son had hepatomegaly with grade II fatty infiltration of liver, elevated alanine aminotransferase, and normal serum AFP. The younger son had an elevated serum AFP level (174.2 ng/mL) with normal abdominal scan, while the wife's results were normal. Because two of the family members had fatty infiltration, the family was also evaluated for variants that are associated with non-alcoholic fatty liver disease (*PNPLA3*; rs22811357, rs7384098 and *TM6SF2*; rs585429267).

Whole blood (3 mL) was collected from the family members, and DNA was isolated. The promoter region of the *AFP* gene was amplified.⁴ A touch-down polymerase chain reaction was initially incubated at 95°C for 2 min, followed by 14 cycles, where the annealing temperature was decreased by 0.5°C per cycle (denaturation at 95°C for 30 seconds; annealing for 61.2°C, with a decrease of 0.5°C per cycle for 30 seconds; extension 72°C for 40 seconds), followed by another round of amplification (denaturation at 95°C for 30 seconds; annealing at 54.2°C for 30 seconds; extension at 72°C for 40 seconds for 19 cycles). An amplicon of 294 bp was generated and sequenced on Beckman GeXP system. Variants associated with non-alcoholic fatty liver disease were genotyped using Taqman probes (rs58542926, rs2281135) and sequencing (rs738409). All protocols used in the study were approved by the institutional ethics committee, and procedures followed have been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki and its later amendments.

DISCUSSION

We report a family with HPAFP. The proband and his younger son showed elevated levels of serum AFP and were found to be heterozygous for the $-119G>A$ polymorphism (rs587776861) in the promoter region of the *AFP* gene (Figure 1), while the proband's wife and older son were of wild type (Figure 1). Two other polymorphisms $-55C>A$ and $-65C>T$, reported in the same region, were of wild type in the family. Father and elder son were heterozygous for

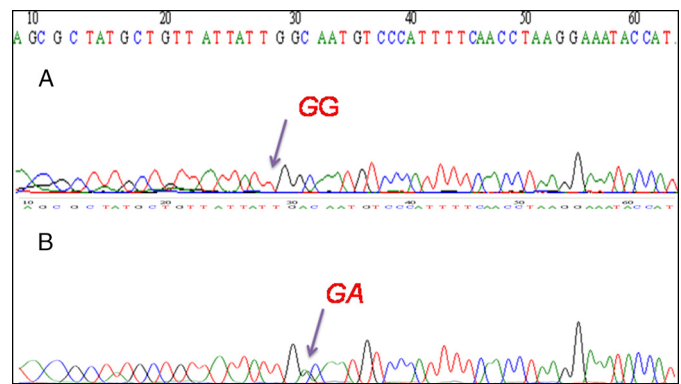


Figure 1. Representative electropherogram of (A) normal genotype (GG) and (B) heterozygous genotype (GA) for $-119G>A$ polymorphism in *AFP* gene.

variants in the *PNPLA3* (rs22811357 and rs7384098) gene and both had hepatomegaly. It has been well documented that these polymorphisms are associated with fatty infiltration of liver.^{9,10}

It was reported in 2010 that there were 19 known cases of HPAFP worldwide, of which 6 families had HPAFP associated with polymorphisms in the promoter region of the *AFP* gene.² All 6 families carried the $-119G>A$ polymorphism, and one family had the $-55C>A$ and $-65C>T$ polymorphisms. Two more cases of HPAFP have been reported in Japan and Italy.^{11,12} Recently, a new case of HPAFP associated with $-119G>A$ and $-55C>A$ polymorphisms has been reported in South Korea.¹³ These polymorphisms lead to increased binding of transcription factor HNF-1 to the promoter region of the *AFP* gene, leading to an increased rate of transcription, which causes the serum AFP levels to rise.⁶ HPAFP is a rare benign disorder that is inherited dominantly and, more often than not, elevated serum AFP levels are suspected to indicate malignancies. There are documented cases where unnecessary treatments or surgical procedures were performed on people with HPAFP; in 5 out of 19 cases of HPAFP, patients underwent unnecessary chemotherapy or surgery.² When patients still have elevated serum AFP after surgery or therapy, they are subjected to further procedures, based on the assumption that the earlier treatment was insufficient. This practice is common in both adults and children.¹⁴

Before resorting to extraneous procedures, the existence of hereditary persistence of AFP should be considered for patients, both children and adults, with elevated serum AFP levels. This report presents the first case of HPAFP from Bangladesh.

DISCLOSURES

Author contributions: All authors contributed to the manuscript creation. VV Ravikanth is the article guarantor.

Financial disclosure: None to report.

Informed consent was obtained for this case report.

Received September 20, 2016; Accepted November 14, 2016

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