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Short Report

Report of VNTR with 13 Repeats Linked to Phenylalanine Hydroxylase Locus in Unaffected Members of Two PKU Families

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ABSTRACT

Phenylketonuria (PKU) is one of the most common metabolic inborn diseases caused by mutations in the phenylalanine hydroxylase (PAH) gene. This gene is linked to a variable number of tandem repeats (VNTR) region which is a polymorphic marker that facilitates the implementation of prenatal diagnosis and carrier screening. In this study, VNTR with 13 repeats that has not been reported previously was observed in 2 PKU families from Fars province, south of Iran. This allele showed 4% frequency in normal individuals. *Iran. Biomed. J. 7 (2): 89-90, 2003*

Keywords: Phenylketonuria (PKU), Phenylalanine hydroxylase (PAH), Variable number of tandem repeats (VNTR)

INTRODUCTION

Phenylketonuria (PKU) with autosomal recessive inheritance is one of the most common inborn metabolic diseases caused by a large number of mutations in a gene encoding a liver-specific enzymes phenylalanine hydroxylase (PAH). The PAH gene is linked to a variable number of tandem repeats (VNTR) region which is located on the 3′-end of the gene. Since the VNTR is a highly polymorphic genetic marker and is inherited in a Mendelain fashion, it is used to give a risk estimation of linked defective allele. These alleles contain 3, 6-9 and 12 copies of VNTR [1-9]. So far the VNTR with more than 12 repeats has not been reported. Here, we report alleles with 13 copies of VNTR in two PKU families and so normal individuals from Fars province, south of Iran.

MATERIALS AND METHODS

Whole blood samples were collected from two PKU families with at least one affected child. These families were Muslims from Fars province, south of Iran. The genomic DNA was extracted from whole blood. A simple and rapid one-step PCR-based procedure was used to find the number of copies of the repeated units of VNTR region linked to the PAH gene, according to the method described by Zschock *et al.* [3]. For evaluating the polymorphism status, the amplified products were analyzed using 3.5% agarose gel electrophoresis. DNA fragments with 325-595 bp length corresponded to 3 to 12 copies of VNTR units, respectively, as reported previously [3].

RESULTS AND DISCUSSION

As shown in Figure 1, DNA fragments of 625 bp corresponding to the 13 copies of the VNTR units were observed in the mothers and unaffected children of the both families. Therefore, the newly reported VNTR 13 was associated with normal alleles in both families.

A unit less than 3 or more than 12 repeats has not been observed in other studied populations of United State of America, Brazil, Italy, Russia, Poland, Sweden, England, Turkey, Ireland, Spain, Switzerland, and Denmark [1-9]. At present, the molecular mechanism for formation of VNTR 13 is unclear, however, it is suggested that the alleles of VNTR are produced by the gain or loss of a single VNTR unit, possibly by slippage during DNA replication [1, 9]. Finding of this new VNTR encouraged us to perform further study on a control

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group. Analysis of VNTR alleles in 50 normal individuals (100 alleles) from the general population was performed. The result showed that the frequencies of VNTR alleles, containing 3, 6, 7, 8, 9, and 13 repeats were 44, 4, 13, 26, 9, and 4 percent, respectively. Notably, the VNTR 13 was present only in four heterozygous individuals.

The observed heterozygosity of this VNTR system was 66 percent in the studied population, which is equal to that of European Caucasians and is higher than that of Chinese population [4]. This finding is of interest, because, based on population genetic studies, Iran has one of the most heterogeneous populations in the world [10, 11]. In this regard, frequencies of the VNTR alleles in other Iranian populations and DNA sequences of VNTR with 13 repeats are under investigation.

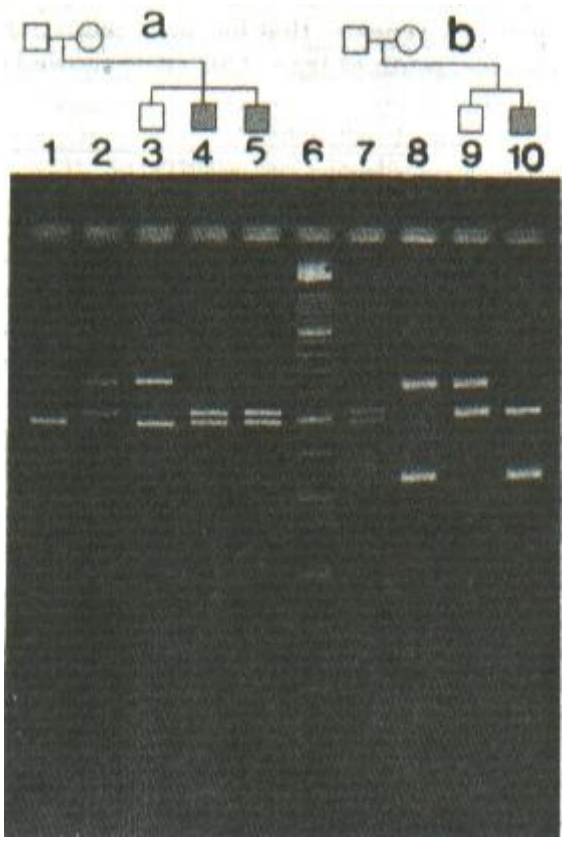

Fig. 1. VNTR analysis in the studied families. In both families, the mother and normal child show a VNTR allele with 13 repeats (lanes 2 and 3 in the family "a" and lanes 8 and 9 in the family "b"). The marker represents a 100-bp ladder.

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