Identification of p.V388M mutation in the PAH Gene in Phenylketonuria Patients from South Brazil.

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Phenylketonuria (PKU) is an autosomal recessive metabolic disorder caused by deficiency of phenylalanine hydroxylase. This deficiency is associated to a large number of mutations at the phenylalanine hydroxylase (PAH) gene. The objective of this work was to perform molecular identification of p.V388M mutation in 30 unrelated PKU patients. DNA samples were isolated from peripheral blood using standard techniques. Region of interest (exon 11 and adjacent regions) was amplified by PCR using specific primers. The p.V388M mutation was then identified by RFLP analysis, considering that the p.V388M mutation destroys a restriction site for *Bsa*AI in the PAH gene. Thus, PCR fragment of a mutant allele does not show smaller fragments following restriction digest while normal allele is represented by two smaller fragments following RFLP. Based on this study we found 9 alleles carrying p.V388M in a total of 60 alleles tested. Genotypic distribution was 3 homozygotes and 3 heterozygotes PKU patients for p.V388M mutation. Among the heterozygous cases, one of them had the mutation in the other mutant allele defined previously. We are currently extending the molecular analysis of these patients in order to define a complete genotype in all of them. In addition, this preliminary work will be the basis for further work on effect of these alterations on PAH protein structure, caused by this mutation (p.V388M) as well as other molecular changes in the PAH gene.

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Key words: Phenylketonuria, phenylalanine hydroxylase, RFLP and PCR.