

Two novel *KEL* alleles encoding K_0 phenotypes in Brazilians

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K_0 (null) is a rare phenotype in which no Kell antigens are expressed on the red blood cell (RBC) membrane.¹ Anti-Ku seen in K_0 individuals has previously been shown to cause hemolytic disease of newborn and severe hemolytic transfusion reactions.^{2,3}

Several molecular mechanisms associated with the K_0 phenotype have already been described, including those that lead to alternative RNA splicing, premature stop codons, and amino acid changes that affect protein trafficking.^{4,5} We herein report two novel *KEL* alleles encoding K_0 phenotypes in Brazilians.

We analyzed genomic DNA of two unrelated Caucasian females referred to our reference laboratory. Both patients were phenotyped as K-, k-, Kp(a-), Kp(b-), Js(a-), Js(b-) and presented an antibody reactive 2+ in gel indirect antiglobulin test with all RBCs except their own.

BRIEF METHODS

Genomic DNA was isolated from peripheral blood with a commercially available purification kit (QIAamp, blood mini kit, Qiagen, Inc., Valencia, CA). *KEL* genotyping was performed by polymerase chain reaction–restriction fragment length polymorphism and the HEA BeadChip according to manufacturer’s instructions. Sequence analysis was performed on all 19 exons of *KEL* and on three exons of *XK* genes. Coding and intron-exon splice regions were sequenced as previously described.⁶

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TRANSFUSION **,**,**,*

RESULTS

Proband 1

KEL genotyping showed *KEL**02/02, *KEL**04/04, and *KEL**07/07 genotypes. Sequence analysis revealed a new homozygous change in *KEL* Exon 2, a guanine to an adenine substitution at Position 71 (c.71G>A, NCBI_ss#: 831878329) leading to a premature stop codon (Trp24Stop; Fig. S1A, available as supporting information in the online version of this paper). No other changes were identified and no mutations were found on *XK* gene.

Proband 2

Molecular testing showed *KEL**02/02, *KEL**04/04, and *KEL**07/07 genotypes. Analysis of coding and intron-exon splice regions of *KEL* of this K_0 individual showed a change of a guanine to an adenine at the first nucleotide of Intron 16 (IVS16+1g>a, NCBI_ss#: 831878330). The mutation changes the conserved *gt* sequence at the donor splice site to *at*, altering the splice donor site, and preventing the removal of the intron. This event creates a premature stop codon, two codons downstream from the SNP. Unfortunately, the patient sample was not available to perform further RNA analysis. The changes are summarized in Table 1 and the electropherograms are included in Fig. S1.

CONFLICT OF INTEREST

The authors report no conflicts of interest or funding sources.

TABLE 1. Phenotype, nucleotide and amino acid changes, GenBank database submitting numbers, and ISBT allele numbers

Proband	Phenotype	Nucleotide change	Amino acid change	NCBI_ss#	ISBT allele
1	K-, k-, Kp(a-), Kp(b-), Js(a-), Js(b-)	71G>A	Trp24Stop	831878329	<i>KEL</i> *02N.30
2	K-, k-, Kp(a-), Kp(b-), Js(a-), Js(b-)	IVS16+1g>a	Alternative Splicing	831878330	<i>KEL</i> *02N.31