INTRODUCTION

The Kell blood group system:
- Consists of 35 antigens and more than 70 alleles (see table at www.isbtweb.org)
- The K+k− phenotype primarily results from homozygosity for a c.578C>T change encoding p.Thr193Met
- At least 28 silenced KEL*02 alleles have been reported and most encode premature stops, frame shifts or alternative spliced products (see table)
- Only three KEL*02 alleles (KEL*02N.05, *02N.28 and *02N.29) encode a k− phenotype resulting from a single amino acid change

CASE STUDY

- Group O Rh-negative Caucasian RBC donor who phenotyped K+k− on two different donations
- Donor sent for HEA PreciseType™ testing to predict an extended phenotype and to confirm the k− phenotype
- The HEA result predicted K+k+ and additional DNA-based was performed

MATERIALS AND METHODS

- Standard hemagglutination methods were used for antigen typing and adsorption/elution studies
- Acid eluates were prepared using Gamma ELU-KIT II
- Genomic DNA was isolated from WBCs
- HEA PreciseType™ testing was performed
- KEL exons 1 to 19 were amplified and sequenced

INITIAL RESULTS

KEL HEA PreciseType™ Results

<table>
<thead>
<tr>
<th>Polymorphism</th>
<th>Result</th>
<th>K</th>
<th>k</th>
<th>Js a</th>
<th>Js b</th>
<th>Kp a</th>
<th>Kp b</th>
</tr>
</thead>
<tbody>
<tr>
<td>c.578T&gt;C</td>
<td>AB</td>
<td>+</td>
<td>+</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>c.841T&gt;C</td>
<td>BB</td>
<td>0</td>
<td>+</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>c.1790C&gt;T</td>
<td>BB</td>
<td>0</td>
<td>+</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

- Discrepancy between the HEA PreciseType K+k+ prediction and serology K+k− results

RESULTS

Serology Results

- The donor’s RBCs typed K+ (3+) and k− with commercial (Immucor) and single donor source polyclonal anti-k
- Adsorption/elution studies of the donor RBCs with two sources of polyclonal anti-k (Immucor and single donor source) did not detect k antigen on the donor RBCs; control eluates reacted as expected

KEL Sequencing Results

<table>
<thead>
<tr>
<th>Exon</th>
<th>Nucleotides present</th>
<th>Amino acids predicted</th>
<th>Predicted phenotype</th>
</tr>
</thead>
<tbody>
<tr>
<td>6</td>
<td>c.578C/T</td>
<td>p.193Met/Thr</td>
<td>K+k+</td>
</tr>
<tr>
<td>8</td>
<td>c.841C/C, c.842G/G</td>
<td>p.281Arg</td>
<td>Kp(a−b+)</td>
</tr>
<tr>
<td>10</td>
<td>Heterozygous for novel c.1130C&gt;T</td>
<td>p.377Leu/Pro</td>
<td>k−</td>
</tr>
<tr>
<td>17</td>
<td>c.1790T/T</td>
<td>p.597Leu</td>
<td>Js(a−b+)</td>
</tr>
</tbody>
</table>

- No other changes found in remainder of KEL
- The c.1130C>T change (p.Leu377Pro) has not been previously reported
- Based on the adsorption/elution results, it is predicted to silenced k antigen expression
- The change was not found in the rs database

CONCLUSIONS

- We identified a new KEL*02 allele, c.1130C>T in exon 10 encoding p.Leu377Pro in a Caucasian blood donor whose RBCs typed K+k−
- HEA testing predicted a K+k+ phenotype
- To determine if this change is associated with greatly reduced or silenced k antigen expression, encoded by a KELnull or KELmod allele, respectively, adsorption/elution studies of the RBCs were performed
- No detectable k antigen was detected when adsorption/elution studies were performed with the donor’s RBCs, indicating that this single nucleotide change results in a silenced, KEL*02N (null) allele and a serological k− phenotype
- It is now becoming clear from recent reports in the literature that K+k− donors rather than genotyping as KEL*01/01 are not infrequently KEL*01/02, and have silenced KEL*02
- Confirming the molecular basis of apparent historical null phenotypes in rare donors can help to distinguish true null phenotypes from potentially weak phenotypes and provides insight into the prevalence of altered alleles in the population

REFERENCES