A second example of anti-Es\textsuperscript{a}, an antibody to a high-incidence Cromer antigen

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A blood sample contained an antibody to a high-incidence antigen that reacted with all red blood cells (RBCs) tested by the indirect antiglobulin test (IAT). The antibody reacted with papain, ficin, and trypsin-treated RBCs, but not with \(\alpha\)-chymotrypsin-treated RBCs. This pattern of reactivity suggested the possibility that the antibody was recognizing an antigen in the Cromer blood group system. Tests against RBCs deficient in decay-accelerating factor (which carries the Cromer antigens) were weakly positive. Tests with antibodies to high-incidence Cromer antigens and with RBCs lacking high-incidence Cromer antigens led to identification of the second example of anti-Es\textsuperscript{a} in an Es(a\textsuperscript{a}+) person. The antibody was IgG1 and reacted by the IAT to a titer of 64. The monococyte monolayer assay indicated potential clinical significance of this antibody in relation to transfusion. Immunohematology 1996;12:112-114.

Antigens of the Cromer blood group system are carried on the complement regulatory glycoprotein decay-accelerating factor (DAF; CD55).\textsuperscript{1-3} The molecular basis of some of the Cromer blood group antigens has been determined to be due to a nucleotide substitution in the DAF gene, leading to an amino acid substitution in DAF.\textsuperscript{4,5} The Cromer blood group system consists of seven high-incidence and three low-incidence antigens that are all destroyed by \(\alpha\)-chymotrypsin and pronase treatment of red blood cells (RBCs), but not by trypsin, papain, ficin, or sialidase treatment of RBCs.\textsuperscript{6-8} RBCs from individuals with the null phenotype (INAB) lack Cromer antigens and DAF\textsuperscript{6}

DAF is attached to the RBC membrane by a glycosphosphatidylinositol (GPI) linkage. A variable proportion of RBCs from patients with paroxysmal nocturnal hemoglobinuria (PNH) have reduced or absent DAF because of a deficiency in PIG-A, an enzyme necessary for the synthesis of glycosphosphatidylinositol anchors. Affected RBCs have a reduction in or a deficiency of all GPI-anchored proteins, including DAF.\textsuperscript{9} For this reason, RBCs from some patients with PNH have weak or absent Cromer antigens.\textsuperscript{10}

Although RBCs lacking any of the high-incidence antigens in the Cromer blood group system are rare, two such antigens have been found to be missing in a single person (UMC),\textsuperscript{11} and in a single family (Es(a\textsuperscript{a})). Anti-Es\textsuperscript{a} was found in the serum of an Es(a\textsuperscript{a}+) woman of Mexican descent who had two Es(a\textsuperscript{a}-) siblings and one Es(a\textsuperscript{a}+) sibling.\textsuperscript{12} The parents were first cousins. Es\textsuperscript{a} was placed in the Cromer blood group system by the nonreactivity of anti-Es\textsuperscript{a} with RBCs of the null (INAB) phenotype.\textsuperscript{13} This article discusses the second example of an anti-Es\textsuperscript{a}.

Case Study

An 86-year-old African American male was being treated in a convalescent home for Alzheimer’s disease, diabetes, and anemia. Two units of blood were ordered for transfusion. The patient had no history of having been transfused, but his serum reacted 1+ to 2+ in the indirect antiglobulin test (IAT) with all reagent and donor RBCs tested. No family members were available for testing.

Materials and Methods

Standard tube hemagglutination tests were used throughout. Enzyme treatment and dithiothreitol (DTT) treatment of RBCs were performed as described.\textsuperscript{14} Antisera and RBCs with unusual phenotypes were from our collections and had been received from patients or sent by colleagues through the Serum, Cell, and Rare Fluid (SCARF) Exchange Program.

IgG subclass determination

IgG subclassing was performed by a centrifugation antiglobulin test as reported.\textsuperscript{15} Anti-IgG1, IgG2, IgG3 and IgG4 were obtained from the Central Laboratory of The Netherlands Red Cross, and standardized as described.\textsuperscript{15}

Monocyte monolayer assay

The monocye monolayer assay (MMA) was performed by the method of Nance et al.\textsuperscript{16} Two examples
of antigen-positive RBCs were sensitized with the patient’s antibody and then incubated with a monolayer of monocytes. Reactivity of monocytes with sensitized RBCs was expressed as the percentage of adherence and phagocytosis when 200–600 monocytes were counted.

Results

The patient’s RBCs were group A, D+C+E−c+c+ (most probable Rh genotype R1r), and the direct antiglobulin test (DAT) was negative. Other typings were: M+N+S+s+, P1+, Le(a−b−), K−k+, Fy(a−b−), and Jk(a+b+). The patient’s RBCs expressed numerous high-incidence antigens and its serum reacted with RBCs known to lack different high-incidence antigens. The antibody reacted with RBCs by LISS-IAT, albumin-IAT, PEG-IAT, ficin-IAT, and papain-IAT. Reactivity was not abolished when tests were performed with RBCs treated with DTT or by prewarmed IAT procedures. However, since the antibody did not react with α-chymotrypsin-treated RBCs, we investigated the possibility of an antibody to an antigen in the Cromer blood group system.

The patient’s serum failed to agglutinate RBCs lacking GPI-linked proteins from a patient with PNH. It caused weak agglutination of RBCs from a person previously determined to express approximately 10 percent of normal levels of DAF and RBCs of the Dr(a−) phenotype, which are also known to have reduced amounts of DAF. Since these results were consistent with an antibody to a Cromer antigen, we typed the patient’s RBCs with antibodies to high-incidence Cromer antigens. The RBCs typed Cr(a+), Tc(a+), IFC+, Dr(a+), WES(b+), and UMC+, but Es(a−). The patient’s serum was then tested and found to be compatible with Es(a−) RBCs from the proposita and her brother.

The patient’s anti-Es made a titer of 64 by the albumin IAT and was determined to be IgG1. Results of the MMA, shown in Table 1, suggest the antibody would cause accelerated destruction of antigen-positive RBCs in vivo. No other alloantibodies were detected in the patient’s serum. The patient recovered without need of transfusion.

Discussion

This is the second reported case of anti-Es in the serum of an Es(a−) individual. Attempts to relocate the proposita or her compatible siblings were unsuccessful. Unlike the Dr(a−) phenotype, which has only been found in Russian Jews, it is possible (because of the Mexican and African ancestry of the two probands) that the Es(a−) phenotype may not be restricted to a particular ethnicity. However, the possibility of a common racial background should be further investigated as both Mexicans and African Americans may have mixed ancestry.¹⁸

<table>
<thead>
<tr>
<th>Patient’s serum + RBC sample</th>
<th>MMA (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patient’s serum + RBC sample #1</td>
<td>15.0</td>
</tr>
<tr>
<td>Patient’s serum + RBC sample #1 + FNS¹⁵</td>
<td>29.5</td>
</tr>
<tr>
<td>Patient’s serum + RBC sample #2</td>
<td>48.5</td>
</tr>
<tr>
<td>Patient’s serum + RBC sample #2 + FNS</td>
<td>53.0</td>
</tr>
<tr>
<td>Patient’s serum + patient’s RBCs</td>
<td>0.7</td>
</tr>
</tbody>
</table>

¹ Indirect antiglobulin test using anti-IgG.

Using the monoclonal–antibody-specific immobilization of erythrocyte antigens (MAIEA) assay, Petty et al.³ showed that the Es antigen is likely to be located in the first short consensus repeat (SCR) domain of DAF. Studies are under way to define the molecular basis of the Es(a−) phenotype.

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References


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