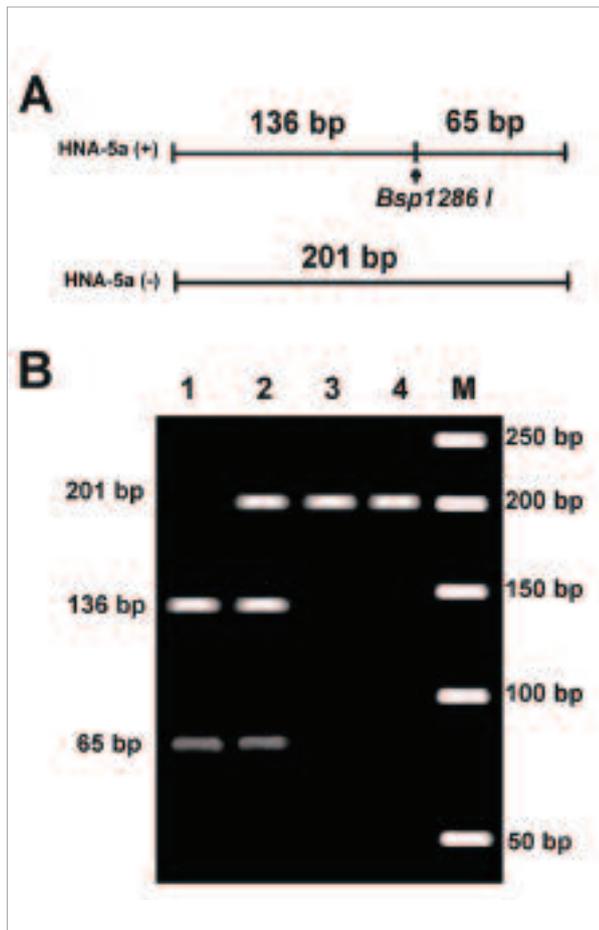


### CLINICAL AND MOLECULAR ANTIGENS AND ANTIBODIES RELATED TO BLOOD CELLS

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PCR-RFLP method and typical results of HNA-5a genotyping.

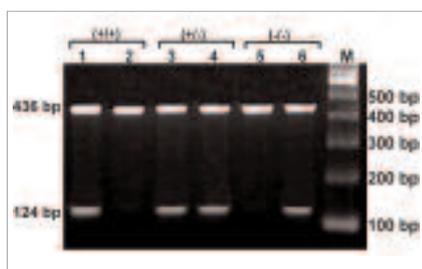
A) A region (201 bp) in the genomic DNA, in which the HNA-5 polymorphism is located, was amplified by PCR. The sizes of the fragments produced by digestion with Bsp1286 I are shown.

B) Typical RFLP patterns Bsp1286 I-treated PCR product. Lane 1: Homozygous HNA-5a (+/+). Lane 2: Heterozygous HNA-5a (+/-). Lane 3: Homozygous HNA-5a (-/-). Lane 4: not digested. Lane M shows the DNA molecular weight marker

Red cell alloantibodies may cause hemolytic disease of the newborn and hemolytic transfusion reactions, while red cell autoantibodies participate in the immune destruction of red cells seen in autoimmune hemolytic anemia. Neutrophil alloantibodies may produce neonatal alloimmune neutropenia and transfusion related acute lung injury (TRALI), on the other hand granulocyte autoantibodies provoke neutrophil immune destruction observed in patients with autoimmune neutropenia. Platelet alloantibodies may induce neonatal alloimmune thrombocytopenia, post-transfusion purpura, and platelet transfusion refractoriness, conversely platelet autoantibodies induce primary or secondary immune thrombocytopenia purpura. The aim of the present immunohematological project is to examine not only the molecular basis of the blood cells alloantigen systems, but also calculate the risk associated to the exposition to allogenic blood cells by blood component transfusion, transplantation, or pregnancy.

## SUMMARY OF RESULTS TO DATE AND PERSPECTIVES

The major project consists of 5 subprojects designed to study blood group systems: 1 subproject connected to platelet alloantigen systems; and 6 subprojects associated with granulocyte alloantigen systems. We propose to study immune mechanisms which take place in autoimmune hemolytic anemia, in hemolysis post-kidney transplant, and in vaso-occlusive crisis seen in patients with sickle cell disease. We will also investigate molecular aspects of the Rh blood group system in Brazilian individuals, the gene frequency of some platelet



Typical result HNA-4a genotyping by PCR-SSP. Lanes 1, 3 and 5 contain HNA-4a-positive-specific reaction, and lanes 2, 4 and 6 contain HNA-4a-negative-specific reaction. Lanes 1 and 2 contain HNA-4a homozygous-positive, lanes 3 and 4 heterozygous and lanes 5 and 6 homozygous-negative. Presence or absence of product at 124 bp determines genotype. All reactions also contained control HGH primers that resulted in a 432 bp product. Lane M shows the DNA molecular weight marker (from 100 bp)

alloantigen systems, and the platelet alloimmunization risk in Brazilian ethnic groups. Finally, we will determine the phenotypic and genotypic distribution of the granulocyte alloantigen systems in different Brazilian ethnic groups, the relevance of some polymorphisms in the expression of such antigens, and the role of granulocyte specific alloantibodies in some transfusion reactions.

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