# Leucocyte grouping : a method and its application

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## Chapter six

## SUMMARY

This study describes the development of a method for the recognition and definition of leucocyte group Four. The method can also be applied to the detection of other leucocyte groups. Essential factors in the method were:

a) the use of leucocyte agglutinins formed during pregnancy;
b) an insight into the shortcomings of the agglutination test;
c) the use of statistical methods to overcome these shortcomings; and
d) the use of a panel consisting of the leucocytes of the women who had formed the agglutinins, and of those of their husbands.

In chapter one the pertinent literature is reviewed, especially that concerning the role of pregnancy in the formation of leucocyte agglutinins and the arguments in favour of the existence of leucocyte groups.

In chapter two a detailed description of the materials and methods is given.

Chapter three is subdivided into three sections. The first section, after reviewing the literature, details the influence of time, temperature and number of leucocytes on the outcome of the reaction. Furthermore the two agglutination tests, i.e. with leucocytes from defibrinated and from EDTA blood, are compared with regard to the percentage of dubious results, the percentage of positive results and the reproducibility. The second section describes the experiments which showed that these agglutinins are in all probability iso-immune antibodies directed against an antigen absent from the leucocytes of the women who had formed the agglutinins but present on the leucocytes of their husbands. Some of the physico-chemical and immunological properties of the agglutinins are described. The third section contains the data concerning the correlation between the number of cases in which leucocyte agglutinins were demonstrable and the number of previous pregnancies, the influence of previous blood transfusions, and the techniques used. No correlation was found between the presence of leucocyte agglutinins and a history of abortion.

Chapter four is divided into two sections. The first section describes the methods used to select, from a group of 66 sera, those which might recognise a leucocyte group. These methods included the use of Fisher's 2 x 2 test and of electronic computers. Twelve sera selected in this way were studied and it was shown that they recognised a leucocyte group, designated as leucocyte group Four because the existence of three other leucocyte groups had been postulated before. However, it is not impossible that leucocyte group Four is identical with the leucocyte group Mac described by Dausset (23). Leucocyte group Four is determined by the antigens $4^a$ and $4^b$ which behave as if they are determined by a single pair of alleles. Both can be recognised expressed in the heterozygote but there is evidence of an antigen dosage effect. In the second section the leucocyte group substance

is demonstrated to be present not only on leucocytes but also on platelets, and in placenta and kidney tissue.

In chapter five the extent is discussed to which the procedure followed was rational. A review is also given of the possible clinical significance of the formation of leucocyte agglutinins during pregnancy in general, and the possible usefulness of the recognition of leucocyte group Four in particular.