examined), the proportion possessing the Gm factor is 57.3%. We tested whether the fact that a patient has the Gm* factor may influence the serological reactions. First of all, we studied, together with F. Jacqueline, patients with chronic inflammatory rheumatism controlled for several years. In actual fact, rheumatoid arthritis is the main affection during which such a possibility may be foreseen. The observed divisions of Gm* were 56.8% in 271 patients suffering from P.C.E. This division amounted to 58.7% in patients with a positive Waaler-Rose reaction and to 53.3% with a negative reaction. There is, therefore, no correlation between the presence of Gm* factor and that of the agglutinating factor responsible for the Waaler-Rose reaction.

Study of the division of the Gm* factor was performed during another form of chronic inflammatory rheumatism, spondylitis, as well as during infectious mononucleosis, diabetes and diseases of the haemopoietic organs.

Antibodies Against Leucocytes in Sera of Pregnant Women

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The importance of antibodies against leucocytes as a cause of transfusion reactions has become a well established fact since the original work of Dausset and van Loghem, and it is now generally accepted that multiple transfusions are in most cases the stimulating factor in the formation of these antibodies1-4. This paper deals with observations which point to pregnancies as another causative factor with regard to the formation of leucocyte antibodies.

Mrs. H.-B. received a blood transfusion for a post partum bleeding. This transfusion was followed by a severe reaction. Red cell incompatibility could be excluded as a cause. Further investigations showed the existence of a strong leucocyte agglutinin which was also present in a pre-transfusion sample. The patient had never been transfused before5.

The possibility was considered that these leucocyte-agglutinins were true antibodies against leucocytes and that they had arisen through immunization during pregnancy.

For the demonstration of the leucocyte-agglutinins we made use of the technique as described by van Loghem8: two drops of the inactivated serum were mixed with two drops of leucocyte rich plasma obtained by dextran-sedimentation. After incubation during 1% hours at 37 C the supernatant serum was removed and replaced by one drop of 3% acetic acid. The results were read microscopically.
The following observations are in favor of the concept that these leucocyte-agglutinins are true antibodies against leucocytes.

1. The agglutinins can be absorbed specifically, i.e. they can only be absorbed with leucocytes which give a positive agglutination test.
2. The absorption is quantitative, i.e. the amount of leucocytes needed for the absorption correlates with the titre of the agglutinin and the amount of serum.
3. It is possible to elute the agglutinin from the sensitized leucocytes (Weiners method).
4. Agglutinins could be located in the gamma-globulin fraction.
5. The agglutinins resist heating at 56°C during 30 minutes.
6. The agglutinins are destroyed at 70°C in 15 minutes.
7. The agglutinin is not absorbed by BaSO4.

With regard to the formation of these antibodies the following points are worth mentioning.

1. On screening the sera of 122 pregnant women who had never received a blood transfusion, the presence of antibodies against leucocytes was revealed in 7 cases (table I).

Table I

2. The frequency of these leucocyte antibodies was significantly higher after the third pregnancy (table I).
3. Apart from the 7 patients mentioned above, we found leucocyte antibodies in the serum of 24 other patients, men and women suffering from various diseases. All these 24 patients had received blood transfusions. By comparing the patients which had received blood transfusions only with those which had been transfused and been pregnant, we found that the last group had leucocyte antibodies after a significantly lower number of transfusions (table II).
4. Another control group consisted of 213 donors of the blood transfusion service. We found antibodies against leucocytes in one case. This was a woman who had been pregnant three times, but who had never received a blood transfusion.

With regard to localization of the antigen responsible for the antibody formation the following points will be mentioned.

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Table II
1. In no instance the leucocytes of the patient were agglutinated by her own serum but in all cases studied, the serum of the wife agglutinated the leucocytes of the husband. It is highly improbable that this is due to chance alone, because the sera agglutinated from 88 to 11 % of random leucocyte samples. The same holds true for the sera not shown here of those women who had received less than 3 blood transfusions and had been pregnant at least 5 times (table III).

Table III

2. We were able to show that also the hemoglobin-free, repeatedly washed and homogenized placenta tissue can absorb leucocyte antibodies specifically. At the moment we would like to refrain from commenting on the exact relationship of these findings and the localization of the antigen responsible for the formation of the antibody (table IV).

From these observations it is concluded that pregnancy in itself can stimulate the formation of antibodies directed against antigens present in the leucocytes of the husband, and are therefore to be considered as iso-immune antibodies. This brings us to the next question : Is it possible to identify leucocyte groups with the aid of these sera?

We have studied 250 different leucocyte samples with 11 sera. From this study the following findings will be mentioned:

1. Only three of the sera gave less than 10 % weak reactions.

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Table IV

Results of the Leucocyte Agglutination Reaction

N.B. : The serum of Mother K. did not possess antibodies against leucocytes.

2. The titre of all sera was low (1/2-1/8) with one exception (1/128).
3. Absorption experiments showed that two of the sera contained at least two antibodies or perhaps more, while one serum might contain only one antibody. The antigen recognized with this antibody had a frequency of about 11%. We had several sera, which gave positive results in about 60%. It is possible that one of them identified the first leucocyte antigen described, the antigen Mac of Dausset. To minimise the chance of confusion with the erythrocyte antigens, we propose to call our antigen big II and its allele small 2.
4. Family studies revealed that the antigen-gen was able to express itself in a single dose (table V).

Table V
Regarding the clinical importance of these antibodies, the following can be reported.

1. As mentioned above, these antibodies can also give rise to severe transfusion reactions. The study of patients who had been pregnant and had received a blood transfusion revealed that in the presence of leucocyte antibodies the incidence of transfusion reactions was significantly higher (table VI).

Table VI

We could confirm the finding of Spielmann7, that leucocyte compatible blood does not, and leucocyte incompatible blood does give rise to a transfusion reaction.  

2. The question if these antibodies influence the development of the fetus unfavorably cannot be definitely answered at this moment. Preliminary data suggest that this might be the case. In this connection it is important to note that with the technique used incomplete antibodies escape detection.

Conclusions

1. Pregnancy may induce the formation of leucocyte agglutinins, which have to be considered as iso-immune antibodies directed against an antigen present in the leucocytes of the husband. 

It seems probable that the antigens can also be found in placenta tissue.

2. It is probably possible to use some of the sera of pregnant women containing leucocyte antibodies to establish the presence of group specific leucocyte antigens. An example is given proving the presence of an antigen called big II with a frequency of about 11 %.

3. These antibodies can cause transfusion reactions, and possibly have a deleterious effect on the fetus.

References