THE DATA SHOWING ANTIGENIC RELATIONSHIP BETWEEN *Salmonella typhi* AND HUMAN LEUCOCYTES

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Antigenic structure of organisms is considered one of the most important indicators of their species and individual specificities. However, along with this it should be pointed out that in certain cases a particular antigen, characteristic of one animal species, is found – in the same or in an altered state – in other species. Thus, for example, antigen A (of the ABO system), characteristic of free and fixed cells, as well as of human secretions, is also found in certain man-resembling monkeys and other mammals (horses and pigs) and even in some microorganisms,

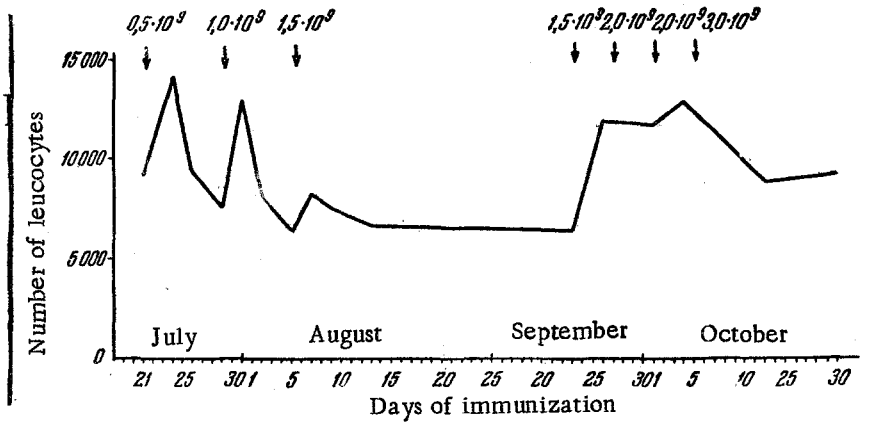

Fig. 1. Leucocytic response in treatment with alcohol Vi-typhoid vaccine. A combined curve.

for example, in type XIV pneumococci (Goebel, Beeson and Hoagland [9]; Beeson and Goebel [17]). As a proof of certain cellular and microbial antigens, one must consider the studies of N. N. Zhukov-Verezhnikov and G. Guseva [4], which established (by means of agglutination reaction) the existence of a common antigen in the plaque bacillus and human erythrocytes.

We consider the studies presented in this report showing antigenic relationship of human leucocytes and *S. typhi* in the light of these currently available facts.

These investigations are directly related to our previously published results pertaining to the antigenic properties of leucocytes and specificity of anti-leucocytes antibodies (P. Popivanov and V. Kh. Vilchanov [5,6,10,11]) and to the role of Vi-vaccine in experimental production of immune leucopenia (V. Kh. Vilchanov and P. Popivanov [1,2]), which studies indicated that anti-Ty (Vi)-sera have an effect on leucocytes in vitro analogous to the effect produced by antileucocyte sera, obtained by immunization of rabbits with leucocytes or pus [6,11].

EXPERIMENTAL METHOD

Experiments were carried out in the following manner: 20 male rabbits were immunized three times (7 days apart) by intravenous injection of $0.5 \cdot 10^9$, $1 \cdot 10^9$, and $1.5 \cdot 10^9$ microbes in 0.5 ml of alcohol typhoid Vi-vaccine

(strain Ty_2 Glatt). Samples of serum were obtained on the 10th and 40th day after the 3rd injection. A new cycle of immunization began on the 50th day (4 intravenous injections of the same vaccine into the same animals, 4 days apart, in stronger doses – $1.5 \cdot 10^9$, $2.0 \cdot 10^9$, $2.0 \cdot 10^9$, and $3.0 \cdot 10^9$ bacteria). Serum samples were obtained 10 days after the last (7th) injection. Sera from 26 normal rabbits served as controls.

Total and differential leucocyte counts were determined at appropriate intervals during the course of immunization. The results of these investigation have been published elsewhere [2]. At this time we present diagrammatically only the scheme of immunization and reaction of leucocytes at different times of immunization with typhoid Vi-vaccine (Fig 1).

All sera were studied by means of complement fixation reaction. Alcoholic and saline extracts of human leucocytes, in particular, of the sterile empyemal pus of tuberculous patients having O or A blood groups, were used as

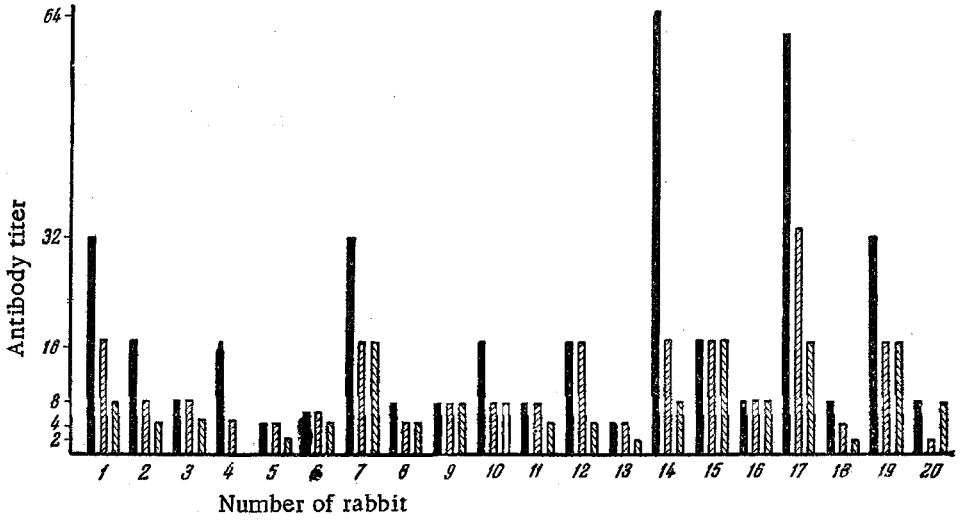

Fig. 2. Complement fixing titers of antibodies in sera of rabbits #1-20.

antigens. Five sera were studied to extablish the relationship of antisera to extracts of human, rabbit, and guinea pig erythrocytes, as well as to the extract of rabbit leucocytes.

EXPERIMENTAL RESULTS

The experimental results demonstrated that sera of control rabbits (not treated with typhoid Vi-vaccine) do not have complement fixing leuco-antibodies.

On the other hand it has been established that sera obtained from 20 immunized rabbits (on the 10th and 40th days after first series of injections as well as on the 10th day after second cycle of injections of typhoid Vi-vaccine), without exception contain complement fixing antibodies in relation to leucocytes. Therefore, in this case Vi-vaccine produced an immunogenic effect similar to the effect of leucocytic antigens, a facts indicating that S. typhi and leucocytes contain similar components producing the same reaction. However, it should be pointed out that leucoantibodies (Ty (Vi)-antibodies) produced as a result of immunization with Vi-vaccine, react with leucocyte antigens in a much lower titer than do the leucoantibodies obtained as a result of immunization with leucocyte mass or pus [6, 11]. It is also established that the above mentioned complement fixing leucoantibodies (Ty (Vi)-antibodies) are not group specific; i.e. they react with leucocytes of group O as well as with leucocytes of group A.

The dynamics of variation in complement fixing leucoantibodies is shown on figure 2, from which it is seen that on the 10th day after the 3rd injection the titer of complement fixing leucoantibodies in anti-Ty (Vi)-sera varies between 4 and 64. On the 40th day after the 3rd injection leucoantibody titer was lowered on the average by 50 % in 11 animals, particularly in those which showed higher titers on the 10th day; in 9 animals having initially lower titers, the titer remained at the same level.

On the 10th day after the second series of injections of higher doses of the vaccine, at shorter intervals, the titer became lower in 12 animals, and leucoantibodies were absent in one animal; 4 animals maintained a constant

titer from the 40th day after the first series of injections; the titer did not change in 2 animals, and one animal had an increase in leucoantibody titer.

In single experiments (carried out for orientation) dealing with the relation of anti-Ty (Vi)-sera to erythrocytes, it has been established that anti-Ty (Vi)-sera react (but in much lower titers of 4 to 16) also with alcoholic and saline extracts of human, rabbit, and guinea pig erythrocytes and with rabbit leucocytes.

Analysing these results one may conclude that in reacting with human and rabbit leucocytes, anti-Ty (Vi)-rabbit sera show the presence of hetero- and isoleuco-antibodies; i.e., they have the properties of antileucocyte antibodies obtained by immunization with leucocytes [6, 11]. However, reaction of anti-Ty (Vi) sera with extracts of erythrocytes shows that these antisera demonstrate not only antileucocytic, but also, express to a lesser degree, antierythrocytic activity. This consideration leads to even greater similarity of anti-Ty (Vi) antibodies to the properties of the investigated antileucocytic sera obtained by immunization with pus and leucocytes.

The antigenic similarity established by us between S. typhi, on the one hand, and leucocytes and erythrocytes on the other, considered in the light of the common antigens between the plaque microbes and erythrocytes, could be brought forward as a new proof of definitely existing antigenic ties between certain microbial and cellular antigens.

SUMMARY

As a result of the immunization of 20 rabbits with alcohol typhoid Vi-vaccine (strain Ty_2 Glatt) the authors obtained in all cases antisera containing complement-fixing leucoantibodies (against the leucocytes of man). The sera of 26 nontreated rabbits did not show the presence of such antibodies. Additional investigations proved that the anti-Ty (Vi)-sera bind (in the complement fixation test) not only extracts of human and rabbit leucocytes – thus showing the presence of hetero- and iso-leucoantibodies – but also bind (at lower titers) extracts of human, rabbit and guinea pig erythrocytes, showing the presence of hetero- and iso-erythroantibodies. These results are considered as a further evidence of the possible antigenic relationships between some cell and microbe antigens.

LITERATURE CITED

1. V. S. Gostev, Chemistry of Specific Immunity [in Russian] (Moscow, 1959).
2. N. Zhukov-Verezhnikov and G. Gusev, Zhurn Mikrobiol., Epidemiol. i Immunobiol., No. 3 (1944) p. 14.
3. V. Kh. Vilchanov and P. Papivanov, Dokl. Bolg. AN, 13, 2 (1960) p. 207.
4. P. Popivanov and V. Kh. Vilchanov, Izvestia In-ta Mikrobiologii Bolgorsk., AN, 11 (1960) p. 169.
5. P. Popivanov and V. Kh. Vilchanov, Trudove na Visshiya Meditsinski In-ta Sofia, 7, No. 1 (1960).
6. P. Popivanov and V. Kh. Vilchanov, Z. Immun.-Forsh., No. 119 (1960) p. 383.
7. P. Popivanov and V. Kh. Vilchanov, C. R. Acad. Bulg. Sci., 12 (1959) p. 463.
8. P. Beeson and W. Goebel, quoted by V. S. Gostev.
9. J. Dausset, Biological and Clinical Immuno-Hematology [in French] (Paris, 1956).
10. W. F. Goebel, P. B. Beeson, and C. I. Hoagland, J. Biol. Chem., 129 (1939) p. 445.
