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IMMUNOCHEMISTRY. -- IMMUNOCHEMICAL STUDY OF SERUM FROM RATS RAISED STERILELY

[Following is the translation of an article by Michel Gleye, Edmond Sacquet and Georges Sandor, in the form of a note presented at the 5 March 1962 session of the Academy of Sciences by Jacques Trefouel, in the Frenchlanguage publication <u>Comptes rendus de l'Academie des</u> <u>Sciences</u>, (Reports of the Academy of Sciences) 1962, 751. 254, No. 11, pp. 2100-2102, 12 Mar 63

Gamma globulins that contain serum from adult rats raised sterilely have a high electrophoretic mobility.

The immunochemical study of the serum of animals raised sterilely has not yet been made, to our knowledge. It has merely been confirmed several times since the work of Thorbecke and his colleagues [See Ncte] that the serum of these animals has a relatively low content of electrophoretic gamma globulins. The present immunochemical study of serum from rats raised sterilely provides some specific data on this subject.

([Note:] G. J. Thorbecke, H. A. Gordon, B. Wostmann and J. A. Reyniers, <u>J. Infect. Diseases</u>, Vol CI, 1957, p. 237.)

We used adult rats produced by raising them sterilely in the Centre de Selection des Animaux de Laboratoire [Center for Selecting Laboratory Animals] of the C.N.R.S. Their serum was studied both by immunoelectrophoresis and by Ouchterlony's methods, in comparison with rabbit serum antiserum of a rat raised naturally.

In Figure 1 we reproduced the results of the saturation test performed with the serum of a "sterile" rat in comparison with "natural" antirat serum. We see that although all the serous antigens are indeed present in the serum of the "sterile" rat, one of them is present only in a very small amount. In fact, a very weak trace appears slowly between the cupping-glass containing the "natural" antirat serum satured with "sterile" rat serum on the one hand, and the cupping-glasses containing either serum from a "sterile" rat or serum from a "natural" rat on the other hand. Consequently, a complex, an antigen-antibody, solubis in the presence of a great excess of antibodies, has diffused commencing with the cupping-glass containing the saturated antiserum (see the article by two of us [See Note]).

([Note:] G. Sandor and M. Gleye, <u>C. R. Soc. Biol.</u>, Vol CLIV, 1960, p. 725.)

From a superficial examination of the immunoelectrophoretic diagram (Fig. 2) one might conclude that gamma globulins are completely absent from the serum of a "sterile" rat. But a more detailed study proves that only the relatively slow electrophoretic mobility component of the gamma globulins is missing. One can count, in fact, toward the cathode,

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four components in the serum from a "sterile" rat as well as in the serum from a "natural" rat. Three of these traces amount to β and $\alpha_{eglobu-}$ lins and are easy to identify equally in the two types of serum. The fourth trace is the only one that is different: its electrophoretic mobility characteristic of gamma globulins in the case of the serum from a "natural" rat is, on the contrary, characteristic of beta globulins in the case of the serum from a "sterile" rat (Fig. 3). There is reason to believe that the component present in a small amount, revealed by Ouchterlony's method, is precisely this gamma globulin that has been cut off from its "slow" components.

Some interesting problems are raised in connection with the fact brought out in this study. One may wonder, in particular, if the gamma globulins with a high cataphoretic velocity that are the only ones persisting in animals raised sterilely do not result from a base synthesis that is independent of antigenic stimuli.



Figure 1. -- Immunochemical comparison between serum from a "sterile" rat and serums from a "natural" rat. (Ouchterlony's method).

> Upper cupping-glass: "natural" antirat serum, saturated with "natural" rat serum;

> Center cupping-glass, left: serum from a "natural" rat;

Center cupping-glass, right: serum from a "sterile" rat;

Bottom cupping-glass: "natural" anti-rat serum, saturated with "sterile" rat serum.

On detailed examination, a very weak continuous trace of precipitation is discovered between the two center supping-glasses and the bottom one.

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Figure 2. -- Immunoelectrophoregram of "natural" rat serum and of "sterile" rat serum, compared with "natural" antirat serum.

Top: "Natural" rat serum; bottom: "sterile" rat serum.



Figure 3. -- Graphic representation of the immunoelectrophoregram in the area of low mobilities. 1, gamma globulins; 2, beta globulins; 3 and 4, beta or alpha 2 globulins.

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