ROLE OF CLASMATOSIS OF PHAGOCYTES IN IMMUNOGENESIS

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Cytological and cytochemical investigations showed that clasmatosis of phagocytes (micro- and macrophages) is characteristic primarily of active, phagocytosing cells and not of dying cells, as was previously considered. Experiments were carried out on guinea pigs in the course of vaccination against and infection with brucellosis. Clasmatosis of phagocytes, which is known to take place in intact animals also, was observed to increase considerably in the period of vaccination and infection. Fragments of cytoplasm which became detached from the phagocytes were shown to be saturated with acid phosphates and brucellosis antigen, whereas fragments of cytoplasm of microphages also contained RNA. There is reason to suppose that antigenic information is intensified and spread rapidly in the animal organism with the aid of clasmatosis of phagocytes.

KEY WORDS: *Clasmatosis; micro- and macrophages; phagocytosis.*

Numerous investigations have shown that macrophages containing phagocytosed and enzymatically processed antigenic material play an important role in antibody formation, by activating immunocompetent lymphocytes.

One group of workers [4] has shown that lysosomes of microphages (neutrophils) if present in a certain concentration, can induce blast transformation of lymphocytes. Fractionation of the lysosomal mass led to the isolation of a certain protein fraction which is responsible for this process. The authors cited consider that the lysosomal factors of lymphocyte transformation which they found are liberated during phagocytosis and activate metabolism of the lymphocytes, with the result that they undergo blast transformation.

It has also been shown [2] that, besides destruction of the antigen, a complex process of formation of specific immunogenic RNA, capable of inducing antibody synthesis in plasma cells, takes place in the macrophages.

It is well known that neutrophils (microphages) are the first cells to appear at the site of injection of an antigen, after which they are replaced by macrophages. It is also known that phagocytes possess the property of clasmatosis (detachment of fragments of cytoplasm of different sizes from the cell).

Until recently it was considered that clasmatosis is a sign of death of the cell and its commencing destruction. However, the authors' investigations lasting several years into both vaccination against and infection with brucellosis revealed that this is not the whole story.

The object of this investigation was to study the possible role of clasmatosis of phagocytes in the transmission of information of various sorts during the course of vaccination against and infection with brucellosis.

EXPERIMENTAL METHOD

Experiments were carried out on guinea pigs. The phenomenon of clasmatosis was studied both during vaccination (animals were immunized by subcutaneous injection of living brucello-

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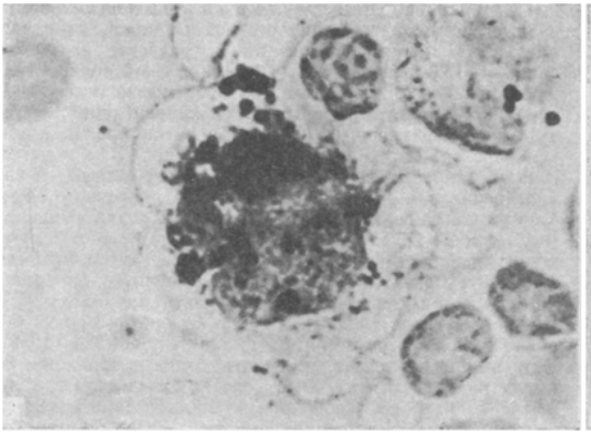

Fig. 1

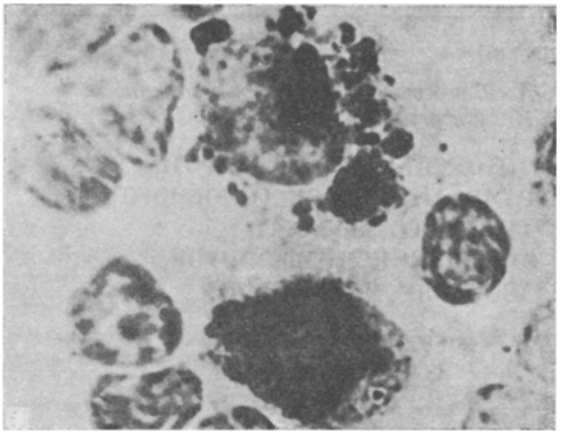

Fig. 2

Fig. 1. Intensive clasmatosis of macrophage from the spleen: different stages of detachment of fragments of cytoplasm, saturated with acid phosphatase, can be seen; many detached droplets of cytoplasm lie freely in the intercellular space. One day after immunization. Acid phosphatase revealed by Gomori's method, ocular 10, objective 100.

Fig. 2. Active clasmatosis of phagocytes in the spleen: six months after infection. Acid phosphatase detected by Gomori's method, ocular 10, objective 100.

sis vaccine composed of *Brucella abortus* 19-BA in a dose of $2 \cdot 10^9$ bacterial cells) and during infection (guinea pigs were infected by the same method with a virulent culture of *Brucella melitensis* 565 in a dose of 20 bacterial cells). Cytological impressions were obtained from the site of injection of the brucella cultures (3 and 6 h, 1, 3, and 5 days later), and also from a regional lymph node and the spleen (3 and 6 h, 1, 3, 5, and 15 days, 1, 3, and 6 months after inoculation with brucellas). Glycogen was detected in the cells by Shabadash's method, acid phosphatase by Gomori's method, RNA by Brachet's method, and brucellosis antigen by a luminescence method.

Survey preparations were stained with Azure II–eosin and hematoxylin–eosin.

EXPERIMENTAL RESULTS

Separation of fragments of cytoplasm from phagocytes is known to take place in the intact organism also. Such cells can be found as isolated examples most frequently in the lymph nodes and spleen. However, this process is greatly intensified in vaccinated and infected animals.

The most intensive clasmatosis was found at the site of injection of the brucellas: As early as after 6 h all phases of detachment of fragments of cytoplasm could be seen in many cells. The process was particularly intensive in neutrophils, less so in macrophages, and it reached its maximal intensity on the first to third day. At about the same time, or sometimes a little later (toward the first day) clasmatosis of phagocytes of the lymph nodes and spleen began to increase.

The process of detachment of a fragment of cytoplasm begins with the projection (as of pseudopodia) of one or several areas of cytoplasm at the same time, most frequently, as our observations showed, from a fully viable phagocyte. This cytoplasmic droplet is then gradually separated by a constriction band from the cell body and, finally, it becomes free-lying in the intercellular space (Figs. 1 and 2).

Because of its biological properties the spleen is one of the richest organs in phagocytic cells and clasmatosis was more marked in the spleen than in cells from the lymph nodes.

Comparison of clasmatosis of phagocytes in immunized and infected animals showed that it occurred more actively during vaccination than infection. This was evidently because of the larger number of brucellas of the vaccinal strain injected into the animal. However, it must be noted that increased clasmatosis of phagocytes in infected animals continued for a longer time: Whereas in the vaccinated animals it was already considerably subdued by 3

months, in the inflected animals its intensity exceeded that of the control (intact animals) even 6 months after inoculation with brucellas (period of observation). The reason for this is probably the longer preservation of the virulent strain of brucellas in the body of the receipient.

Previous investigations [1, 3] showed a high glycogen content in the detached fragments of cytoplasm. It was suggested that these droplets of cytoplasm saturated with glycogen were possibly ingested by subsequent generations of phagocytes and served to prime their energy metabolism.

The present observations showed that droplets of cytoplasm shed by both macrophages and microphages were highly saturated also with enzymes (for example, acid phosphatase), and the fragments of cytoplasm of macrophages also contained RNA. It must be emphasized that the content of glycogen and acid phosphatase in the detached fragments of cytoplasm was as a rule greater than in the remainder of the cytoplasm of the cell. By luminescence microscopy, considerable quantities of specific brucella antigen (more frequently soluble) were also found in them.

It is very probable that with the aid of clasmatosis of the phagocytes the amount of antigenic information in the recipient's body is increased and is spread rapidly (it may perhaps start in this way also) via the lymph and blood stream.

The facts described above suggest that clasmatosis is more frequently an indication of high functional activity of the phagocytes and that it plays an important role in the formation of immunity.

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