SOME IMMUNE REACTIONS IN EXPERIMENTAL BURNS

(UDC 616-001.17-07:616.15-097]-092.9)

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Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 58, No. 8, pp. 105-107, August, 1964
Original article submitted June 1, 1962

At the present time a great deal of attention is directed to a new and effective method of the treatment of burns which consists in transfusion of serum and plasma obtained from patients themselves recovering from burns. The immunological changes occurring under the influence of the burn are of special interest. In particular it is important to study changes in the indices of natural immunity produced under the influence of the burn, and to determine the immune mechanisms underlying the action of the sera obtained from convalescent patients.

The object of the present work has been to study some immune blood changes related to experimental burns.

EXPERIMENTAL METHOD

The experiments were carried out on 9 dogs. As an index of natural immunity we measured the phagocytic activity of the blood. To determine the presence of specific "burn" antibodies or antigens we made use of the reaction of precipitation in agar.

In the I experiment a burn was inflicted, in the II experiment a burn was inflicted and the animal was then treated by transfusion of plasma from convalescent animals, and in the III experiment the burn was followed by a transfusion of plasma from healthy animals.

The burn was inflicted by application of the flame from a soldering lamp for $1\frac{1}{2}$ min to the side of the back and belly, the area of the burn comprising 20-25% of the total cutaneous surface. In terms of the accepted classification the burn was of the degree IIIa.

The phagocytic activity of the blood and the presence of specific "burn" antibodies in the blood serum was studied at various times after infliction of the burn and infusion of plasma.

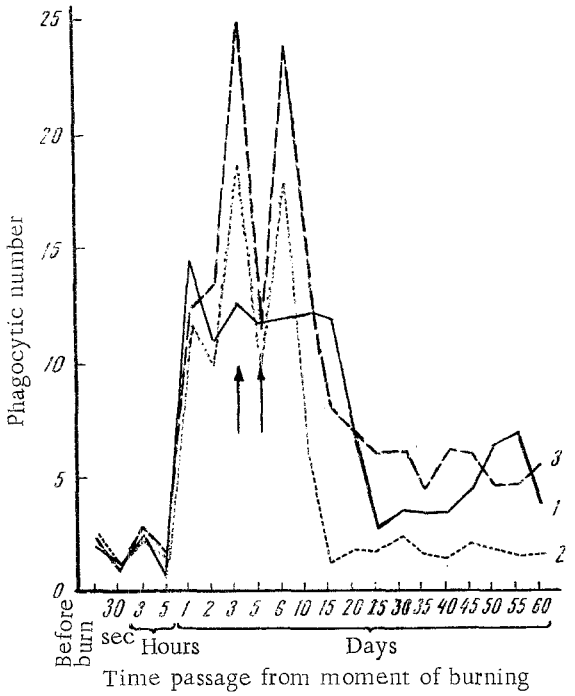
The phagocytic activity of the leucocytes was determined as follows. Blood taken from a vein was mixed in a test tube with an equal volume of 2% sodium citrate. To the mixture was added half the volume of a 1 billion 24 h culture of Staphylococcus aureus (strain No. 209). The mixture was incubated at 37°.

After 30 min a smear was made, and after fixation and staining it was stained by Romanowsky-Giemsa. In the stained preparation we counted 100 neutrophils; the percentage of phagocytic leucocytes represented the phagocytic activity, while the mean number of phagocytized microbes per leucocyte was the phagocytic number.

To determine the "burn" antibodies and the antigen we used a modification of the method of precipitation in agar of Oakley and Fulthorpe, because it allowed small quantities of reagents to be used. The reaction was carried out in narrow tubes at the bottom of which was placed a layer of agar containing serum with the presumed antibodies, and the upper part contained a layer of agar containing the presumed antigen: between them was a layer of pure agar in which the precipitation line was to be formed.

The external reagent or antigen was serum taken from dogs within a few hours after burning, while serum obtained 20 or more days later was used in the reaction to test for the corresponding antibodies. Sera obtained from blood taken before burning were used as controls.

EXPERIMENTAL RESULTS



Mean indices of phagocytic activity of leucocytes after infliction of a burn. 1) Burn; 2) burn and infusion of normal plasma; 3) burn and infusion of plasma from a convalescent animal. The arrows indicate the time of the plasma infusion.

During the first 5 h after the burn had been inflicted (the period of nervous-reflex shock) the phagocytosis was either not marked, or even somewhat suppressed (see figure). This effect may be accounted for by the influence of the shock due to the burn, or alternatively by the absence at this time of resorption from the damaged area. With the appearance of a damaged area and resorption from it non-specific phagocytosis was sharply increased, so that phagocytic activity quite frequently preceded the visible changes in the burnt area. Both clinical observations and studies of phagocytic activity leave us to conclude that in these experiments the duration of the period of toxemia and infection was 15-25 days.

At the end of this period the necrotic masses sloughed off, and a granulating surface was exposed; at the same time resorption was sharply reduced, with the result that there was a rapid reduction of non-specific phagocytosis, which however, reached a level higher than had obtained during the period of shock which followed the burn.

After 40-60 days the burn had almost completely healed; at this period there was some increase in the intensity of phagocytosis.

Infusion of fresh plasma caused a nearly two-fold increase in phagocytic activity, but the effect lasted only 2-3 days. A still greater elevation of phagocytic activity ($2^1/_2$-3-fold) was observed after infusion of plasma from convalescent dogs, and in this case the high phagocytic indices were recorded for as long as 8-10 days (see figure).

No "burn" antigen or any corresponding antibodies could be revealed by precipitation in agar. We made a simultaneous attempt to obtain heterogenous serum specific to the "burn" antigen. For this purpose rabbits were twice immunized, with dog sera obtained before the burn, and 6 h afterwards. The immunization was carried out by successive intravenous injection of 0.5 ml, 1 ml, 1 ml, and 1 ml of serum at 3 day intervals. Seven days after the end of the first and second cycle of immunization tests were made for the presence of "burn" antibodies in the rabbit sera by the method of precipitation in agar. In this experiment we obtained two clear precipitation lines, though the line appeared whether the dog serum was taken before or after the experiment. Therefore, in this experiment again we failed to reveal the presence of any specific "burn" antigen.

The results of these experiments made to identify a "burn" antigen by precipitation in agar by no means constitute proof of the absence of any such antigen, but merely demonstrate that the method is not sufficiently sensitive to reveal it.

SUMMARY

In the experimental burns there is a change in phagocytic activity which is related to a specific stage in the healing process. If the burnt animal is transfused with the plasma from a convalescent animal there is a marked rise in the phagocytic activity. When the method of precipitation in agar was used to determine whether or not specific "burn" antibodies were produced, no positive results were obtained.

LITERATURE CITED

1. C. Oakley and A. Fulthorpe. Path. Bact. (1953), v. 65, p. 49.