

EFFECT OF VACCINATION ON NONSPECIFIC PHAGOCYTTIC
ACTIVITY OF PERIPHERAL BLOOD LEUKOCYTES

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UDC 612.112.3.014.46:615.37

The phagocytic activity of the peripheral blood leukocytes against *Escherichia coli* was investigated in experiments on rabbits immunized with DPT, smallpox, BCG and antirabies vaccines. The phagocytic index of the leukocytes and the index of complete phagocytosis were determined at various times after immunization. In every case a statistically significant decrease in both indices was found, on the average for 14 days (from the 7th to the 21st day after vaccination). Toward the end of the period of observation (28th day) normal phagocytic activity was restored.

Previous investigations, the results of which are summarized by Pletsityi [8], showed that the humoral mechanisms of natural immunity (the bactericidal properties of the serum, the serum properdin, lysozyme, and complement levels, the lytic activity of the saliva) are temporarily depressed by various types of vaccination. Investigation of the effect of vaccination on the cellular mechanism of nonspecific resistance, reflected in the phagocytic activity of the peripheral blood leukocytes, is of great interest. It was decided to study nonspecific phagocytosis directed against microorganisms which differ antigenically from the vaccines used for immunization.

Although the literature on this problem is not extensive [2-7, 10-12] and despite substantial differences in the techniques used to obtain the observations, an unambiguous conclusion can be drawn: the changes in nonspecific phagocytic activity are phasic in character, with clear predominance of the stage of inhibition of phagocytosis. A disadvantage of the investigations cited above is that in most cases no consideration was paid to the completeness of phagocytosis as an index of its effectiveness.

The object of this investigation was to study, in experiments on animals, the effect of immunization on nonspecific phagocytic activity of the leukocytes, using vaccines widely administered in practice: combined diphtheria-pertussin-tetanus (DPT), smallpox, BCG, and antirabies vaccines.

EXPERIMENTAL METHOD

Observations were made on 60 gray rabbits weighing 2.5-3 kg. The action of each vaccine was tested on 8-10 rabbits, and each determination of phagocytosis was accompanied by a control test on unimmunized animals. DPT vaccine was given to the rabbits as a single intramuscular injection in a dose of 0.7 ml. The animals were immunized with smallpox vaccine by a single intradermal injection in a volume of 0.1 ml. BCG vaccine was given as a single intradermal injection into the rabbits in a dose of 0.1 ml. Antirabies vaccine (Fermi type) was injected subcutaneously into the rabbits in a dose of 1 ml daily for 12 days. Ordinary production batches of all the vaccines were used to immunize the rabbits.

The phagocytic activity of the leukocytes was determined by a modified method of Berman and Slavskaya [1]. The test organism was *Escherichia coli*. The phagocytic index (percentage of leukocytes with

Laboratory of Physiology of Immunity, Institute of Normal and Pathological Physiology, Academy of Medical Sciences of the USSR, Moscow. (Presented by Academician of the Academy of Medical Sciences of the USSR A. M. Chernukh.) Translated from *Byulleten' Éksperimental'noi Biologii i Meditsiny*, Vol. 75, No. 3, pp. 76-79, March, 1973. Original article submitted May 22, 1972.

TABLE 1. Changes in Index of Nonspecific Phagocytic Activity of Leukocytes in Rabbits Immunized with DPT, Smallpox, BCG, and Antirabies Vaccines

Vaccine	Phagocytic index				
	before vaccination	after vaccination (time in days)			
		7	14	21	28
DPT	55±0,9	52±2,2	47,7±1,4	47,9±0,7	53±1,0
P		<0,5	<0,01	<0,001	<0,1
Smallpox	56±0,7	49±1,5	48±1,8	51±0,8	53,7±0,8
P		<0,01	<0,01	<0,001	<0,1
BCG	56,6±1,0	50±1,3	45,5±1,3	46,8±2,0	56,8±0,56
P		<0,01	<0,001	<0,02	
Antirabies	55,8±0,45	48±0,7	45±1,2	52±0,7	54±0,9
P		<0,001	<0,001	<0,01	

ingested bacteria) and the index of complete phagocytosis (percentage of leukocytes having completely digested microorganisms) were determined. The criterion of death of the microorganisms ingested by the leukocytes was the ability (or otherwise) of the phagocytosed organisms to grow. The tests were carried out as follows.

To a test tube containing 0.1 ml sodium citrate 0.2 ml of the blood for testing was added, the contents were stirred, and 0.1 ml of a 2×10^9 emulsion of *Escherichia coli* was then added; the tube and its contents were incubated at 37°C for 30 min. One drop of the mixture was then spread uniformly over the surface of a plain agar plate. When the film was dry, the first impression was made on a slide. After incubation of the Petri dish for 2 h a second impression was taken. The impressions were fixed for 3-5 min in methyl alcohol and stained by the Romanowsky-Giemsa method. The slides with the impressions were dried and counts of 100 neutrophils were made.

The phagocytic activity of the peripheral blood leukocytes was investigated for each animal separately before immunization and on the 7th, 14th, 21st, and 28th days after vaccination (in the case of rabies vaccine, after the first injection). Statistical analysis of the results was carried out with the aid of Student's tables.

EXPERIMENTAL RESULTS

Spontaneous changes in the mechanism of natural resistance were found to be negligible. Immunization with all the vaccines used induced similar changes in the nonspecific phagocytic activity of the leukocytes of the experimental animals: their ability both to ingest and to digest was distinctly weakened (Tables 1 and 2).

A substantial decrease in the phagocytic index (number) was observed mostly for 14 days, the greatest fall being found 2 weeks after vaccination. Toward the end of the period of observation the phagocytic activity of the leukocytes was restored virtually to its original level, but not above it. Only for rabbits immunized with DPT vaccine was the decrease in the phagocytic index statistically significant 7 days after vaccination. In that case, however, this function was inhibited most strongly on the 21st day, i.e., at a time when after the other types of vaccination the phagocytic index had started to return to normal.

The dynamics of the index of complete phagocytosis was similar, and the similarity extended not only to the times of the decrease in this index and its return to normal and the corresponding times for the phagocytic index, but also to their absolute values. Irrespective of the properties of the vaccines used for immunization, the pattern of changes was in general standard. In all groups of animals the index of complete phagocytosis was low on the 7th, 14th, and 21st days after vaccination, returning completely (or partly, in the case of rabbits receiving DPT vaccine) to normal by the 28th day.

Two features attract attention when these results are analyzed. First, the very short times taken for the initial and final phases of the observed changes to develop. Since in most cases there was already a significant decrease in the phagocytic activity of the leukocytes 7 days after vaccination, it is reasonable to assume that this reaction began very early, perhaps immediately after immunization. The opposite reaction—restoration of the normal phagocytic activity of the leukocytes—took place equally rapidly, for in most cases it was complete in not more than 7 days.

TABLE 2. Changes in Index of Complete Phagocytosis of Leukocytes in Rabbits Immunized with DPT, Smallpox, BCG, and Antirabies Vaccines

Vaccine	Index of complete phagocytosis				
	before vaccination	after vaccination (time in days)			
		7	14	21	28
DPT	59,4±1,3	52,8±1,2	48,7±1,4	48±0,4	53,5±0,8
P		<0,01	<0,001	<0,001	<0,02
Smallpox	55±0,6	49±1,4	52±1,0	52±1,0	55,8±0,9
P		<0,01	<0,02	<0,02	
BCG	58,6±0,8	48±0,8	47±1,2	50±1,0	58,5±0,7
P		<0,001	<0,001	<0,01	
Antirabies	56±0,35	46±0,7	44,5±0,7	53±0,5	54,5±0,7
P		<0,001	<0,001	<0,01	<0,1

The second feature is connected with the more marked effect of vaccination with antirabies and BCG vaccines on the phagocytic activity of the leukocytes. Whereas in the first case this can be explained by the prolonged administration of massive doses of the vaccine, in the second case either some especially strong primary effect of the BCG vaccine or a rapid proliferation of the attenuated microorganisms in the immunized animal must be assumed.

The mechanism of the phenomenon observed in those experiments is evidently composed of several factors. The first and most important is the action of the vaccine preparations as extraordinary stimuli of the nervous system, causing a disturbance of the normal activity of the higher visceral centers, which must inevitably lead ultimately to a change in the functional state of the autonomic nervous system, with a firmly established role in the regulation of phagocytosis [9].

The second probable component is connected with the possible depression during vaccination of the synthesis of opsonins, one type of what are described as normal antibodies, with the property of increasing the phagocytic activity of leukocytes. However, special investigations will be required to solve these problems.

These investigations thus showed that immunization of animals with the vaccines in large-scale practical use leads to a temporary, yet definite, inhibition of the nonspecific phagocytic activity of the peripheral blood leukocytes actually during the stage of energetic development of specific immunological transformation. This "negative" phase coincides with the time of inhibition of the humoral mechanisms of natural immunity (nonspecific resistance) by vaccination.

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