THE INFECTION OF MONKEYS WITH PARATYPHOID B

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(Received August 31, 1955. Presented by Active Member of the AMS of the USSR, I. M. Davydovsky)

Paratyphoid diseases, or salmonellosis, caused by diverse varieties of paratyphoid bacteria, are widespread among animals. Thus, paratyphoid is known in colts, calves, pigs, reindeer, birds, reptiles and many other animals, down to small laboratory animals [5,6,7].

All of these diseases are produced by different bacteria, only some of which are pathogenic to man. B. paratyphosus A and B are pathogenic to man, but not to other animals, although several carriers have been found among animals. These bacteria produce the so-called abdominal form of paratyphoid in man, although the same term is occasionally applied to the condition produced by other paratyphoid bacteria as well.

Foreign, as well as our own, literature contains reports of paratyphoid in monkeys. The microorganisms isolated in such cases are predominantly Bact. paratyphosus Breslau and paratyphosus B[2,5]. According to data available at the Sukhumi Medico-Biological Station and the Moscow Zoo, such infections develop in the form of dysentery, often fatal; autopsy shows diphtheroid ulcerous changes in the large intestines and catarrhal inflammation of the small intestine. Although etiologically the disease is paratyphoid, it differs anatomically from paratyphoid in man. At the same time, paratyphoid carriers are found fairly frequently among monkeys. E.V. Abramova found carriers of Bact. paratyphosus Breslau and paratyphosus B in monkeys at the Sukhumi nursery in 1937. The Laboratory of Infectious Pathology of the Medico-Biological Station found Bact. paratyphosus B carriers among monkeys imported from Ethiopia; their infectivity disappeared spontaneously. However, no cases of illness caused by these bacteria have been established.

I.I. Mechnikov and A.N. Bezredka succeeded in producing experimental typhoid and paratyphoid clinically similar to pediatric infection in anthropoid apes (chimpanzees) by the administration of typhoid and paratyphoid bacteria or of the excreta of patients. The same authors' attempts to produce paratyphoid in monkeys and other animals were unsuccessful.

More recently, A.A. Valdman [4] succeeded in producing typhoid and paratyphoid infections in small laboratory animals by intestinal infection.

In order to produce paratyphoid B in monkeys, we undertook the experimental infection of 7 monkeys – Macaca rhesus and Chinese macaca, averaging about one year old. The infecting agent used was a paratyphoid B culture from the Museum of the Laboratory of Infectious Pathology which had been isolated by A.S. Aksenova from the monkeys brought from Ethiopia in 1948 [1].

All 7 monkeys were kept under close surveillance for 17 days before infection in order to establish the normal blood indexes, to examine bone marrow punctures, to determine the normal body temperature, the normal flora of the intestine, and the phagocytic acitvity of the leucocytes. Examination of the blood for antibodies to the paratyphoid B culture gave negative results twice.

After the preliminary investigations, the 7 monkeys were given a dose of 30 billion bacteria from the paratyphoid B culture by mouth. One hour prior to ingestion of the bacteria, the monkeys each had been given 4 ml of ox bile, which, according to Bezredki [3], is conducive to the development of the disease.

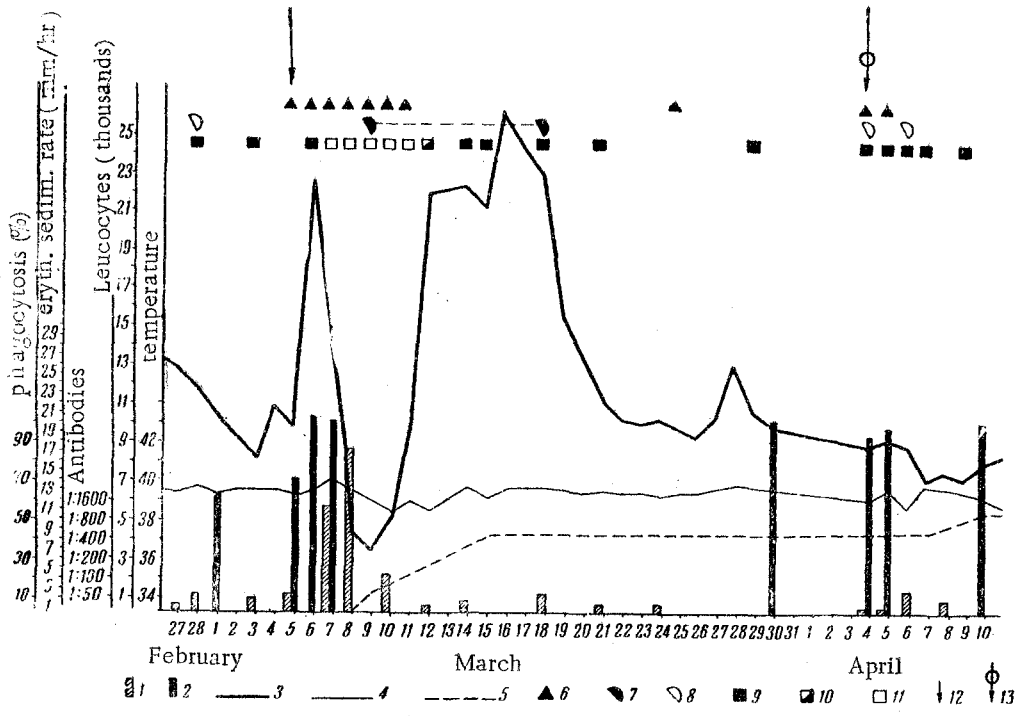

Figure 1. The course of the disease in the monkey Ruchei. No relapse was produced by reinfection. 1) Erythrocyte sedimentation rate; 2) phagocytosis; 3) leucocytes; 4) temperature; 5) antibodies in the blood; 6) excretion of the paratyphoid B rods in the stool; 7) enlarged spleen; 8) normal spleen; 9) formed stools; 10) soft stools; 11) liquid stools; 12) initial infection; 13) reinfection.

Figure 2. A portion of the small intestine with hypertrophied Peyer's patches.

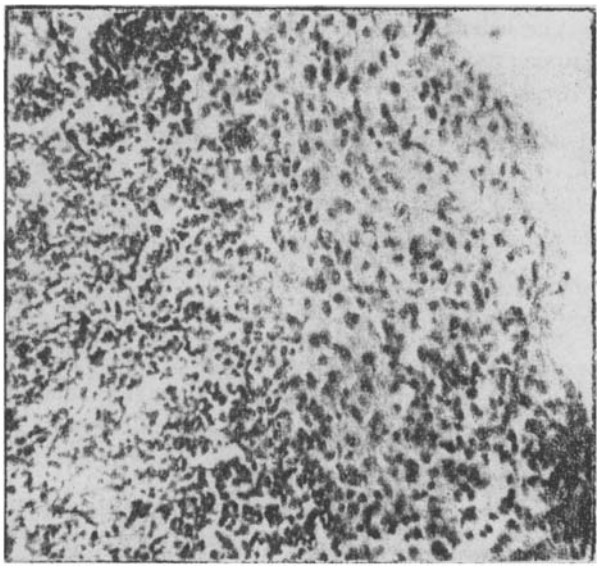

Figure 3. Growth of the reticular cells in the Peyer's patches of the ileum. Obj. 40, oc. 4.

The incubation period for the appearance of the first clinical symptoms was one day. By the second day the monkey became listless and anoretic. A wave of leucocytes (as many as 19,000-23,000 leucocytes) was observed, with marked lymphopenia and a shift to the left. This leucocytosis was replaced during the second day by a progressive decrease in leucocytes reaching leucopenia. The shift to the left remained.

The second day of the illness was characterized by a marked increase in the erythrocyte sedimentation rate (while the leucocytosis was decreasing) up to 12-30 mm per hour (base rate was 0.5-3 mm per hour); on the third day the sedimentation rate reached a maximum, then began to decrease until it returned to normal by the 7th day. On the second or third day, diarrhea developed; the stool was fluid at first but became pea-soup later, typical of the abdominal form of paratyphoid in man. The diarrhea continued approximately 5-6 days, after which the stools began to return to normal. Bact. paratyphosus B could be isolated from the intestine two hours after administration of the culture. The bacteria could be found frequently in the course of 5-6 days, then occasionally during the next 15-19 days. Only in the case of the one monkey, Iago, which died 3½ days after infection, could the Bact. paratyphosus B be isolated only two hours after the administration of the culture. However, this was the monkey from whose blood Bact. paratyphosus B was isolated two days after infection. Bacteria could not be isolated from the blood of the other animals.

On the third or fourth day of the illness, the spleen became enlarged and palpable, although it had been imperceptible earlier. At this time the monkeys ate practically nothing, lost weight sharply and, were very listless, sitting pressed against each other. Pallor was observed in the integument and the visible mucous membranes. The integument became dry; the eyes receded; By the eighth day, approximately, these symbols disappeared, with the exception that the spleen remained enlarged for 7-8 more days.

No clearly defined temperature change could be found in the sick monkeys, although the body temperature of two animals (Iago and Tozer) went down to 35° when they were critically ill.

The disappearance of clinical symptoms by the fifth day apparently did not indicate recovery, since a new wave of leucocytosis, greater than the rise at the beginning of the illness, was observed during this period. This rise lasted 3-4 days, following which the number of leucocytes gradually returned to normal. Thus, uncomplicated paratyphoid lasted approximately two weeks in the monkeys (Fig. 1).

However, the disease was without complications only in the case of two monkeys. Pneumonia and myocarditis complicated the other cases at various times.

Autopsy of one monkey, which had died 3½ days after administration of the culture, revealed large areas of pneumonia and the appearance of interstitial myocarditis, in addition to the changes characteristic of the abdominal form of paratyphoid. Arrhythmia and extrasystole were found in the monkey Tozer on the 11th day, the consequences, we believe, of myocarditis.

The paratyphoid B which we produced in the monkeys resembles human paratyphoid fairly closely, differing from it in the absence of fever and a shorter incubation period. However, it seems to us that illnesses identical to those in man cannot be produced in animals because of the considerable differences in the developmental level of the central nervous system, the metabolism, etc. Only illnesses or pathologic conditions more or less closely analogous to the human types can be discussed.

The fact that the monkey Nelson suffered two marked waves of the disease is not without interest. At the end of the first week, the leucocyte count returned to normal, the sedimentation rate began to fall, and the stool regained its normal consistency.

However, on the 8-9th day its condition deteriorated; there was a recurrence of diarrhea, leucocytosis, and increased sedimentation rate, and the second week was an exact repetition of the first.

Thus, during the second week one of the 7 monkeys suffered an early relapse which, in duration, clinical aspects, and symptomatology, did not differ from the typical course of a single attack.

Autopsy of the monkey Iago revealed systemic hypertrophy of the lymph nodes, most pronounced in the group of lymph nodes in the mucosa of the small intestine. Hyperplasia of the reticular elements in the lymph nodes was established microscopically, combined with a rounding of the nodes, which consisted of large light reticular cells; pinkish areas of necrosis were found in many. Peyer's patches were hypertrophied and protruded into the lumen of the intestine (Fig.2). Viewed microscopically, enlargement of the large light cells in the surface portion of Peyer's patches, directly below the epithelial layer, resulted in the displacement of the lymphadenoidal tissue (Fig. 3). In the deeper areas, groups of light reticular cells were apparent among the endothelial cells.

Large, solid, airless, grayish-white areas, ranging in size up to that of a pea and protruding under the pleura, were observed in the lungs. Microscopic examination revealed that aggregations of leucocytes had completely filled the alveolar spaces.

The spleen was enlarged and edematous in cross-section. Microscopically, the changes found in it were analogous to those found in the lymph nodes. An insignificant number of small granulomatous areas of overgrown reticular cells was evident along Glisson's capsules in various sections of the liver.

There were diffuse round-celled infiltrations in the interstices of the myocardium. The liver, kidneys, and myocardium presented a picture of parenchymatous and fatty degeneration, greatest in the liver where marked dissociation of the liver lobules was observed, together with small diffuse areas of necrotic liver tissue.

The changes observed through autopsy and microscopical analysis were consistent with the anatomicopathological changes in human abdominal paratyphoid. As regards the pneumonia, its etiology remains unknown. In spite of the fact that a pure culture of Bact. paratyphosus B was isolated from the pneumonic foci, we are not yet ready to draw conclusions regarding the etiological significance of this.

We can, we believe, discuss the normal course of paratyphoid B in the monkey on the basis of our clinical, bacteriological and anatomical examinations.

Since Bact. paratyphosus B was isolated from the blood of only one monkey, we ascertained the distribution of the bacteria in the monkey's system by means of tagged atoms. We observed Bact. paratyphosus B, marked with radioactive phosphorus P^{32}, in the urine only two hours after administration, and in the blood an hour later. The negative blood cultures may have been due to the fact that only 1 ml of venous blood was used per culture.

The experimentally-produced disease was accompanied by the development of a permanent immunity as indicated by the marked increase in the phagocytic activity of the leucocytes and the appearance and considerable rise of the specific antibody titer in the blood of the experimental animals. During the third day of the disease, specific antibodies against Bact. paratyphosus B began to appear, with a titer of 1:50; the 4th day the titer was 1:200 and the 9th, 1:400 and 1:800. The antibody titer did not change during the month after infection, once the maximum had been attained.

After 24 hours, the number of leucocytes, phagocytic microorganisms, increased from 60 to 100%, while the phagocytic index rose from 3 to 17.5. A month after the infection, the number of phagocytic leucocytes dropped somewhat (88-90%), but the phagocytic index rose higher, reaching 45-63 in individual cases.

Reinfection of 4 monkeys with paratyphoid B culture under the same conditions one month after the initial infection did not lead to development of the disease (Fig. 1). Moreover, the excretion of an insignificant number of Bact. paratyphosus B was observed only 4 hours after infection of the animal. Repeated bacteriological examination of rectal smears failed to show Bact. paratyphosus B.

Consequently, "human" cultures of Bact. paratyphosus B can be said to be pathogenic to monkeys and to cause a disease in them which is similar to the abdominal form of paratyphoid in man.

In view of the coincidence of the basic clinical and pathological changes, the experimentally-induced disease can be regarded as a good example of paratyphoid, useful for the study of problems of pathogenesis, prophylaxis and therapy of paratyphoid infections.

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