PRIMATOLOGY

Yersinia Infection in Monkeys

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Pathomorphological picture and agents of spontaneous Yersinia infection in monkeys are described. The infection was characterized by polymorphic and generalized changes (usually in *Y. pseudotuberculosis* infection) or mainly local changes involving the intestine and asymptomatic carriership (in *Y. enterocolitica* infection). Of 8 monkey species, red monkeys were the most sensitive to Yersinia. Similarity of human and simian yersiniases allows to use monkeys as the experimental model for studies of this pathology.

Key Words: *spontaneous Yersinia infection; monkeys*

Yersinia infection is a term denoting infections with representatives of *Yersinia* genus, family *Enterobacteriaceae* (except *Yersinia pestis* causing plague). Two representatives of this genus, *Y. pseudotuberculosis* and *Y. enterocolitica,* are most important for medicine. Yersinia infection belongs to saprozoonoses. Yersinia infections and epizooties have been described for numerous species of domestic and wild mammals and birds [5]. First reports about human disease induced by these agents appeared recently [1]; such diseases are highly prevalent in many countries at all continents and at present became a serious problem of public health. The most important characteristics of Yersinia from epidemiological viewpoint ensuring their high prevalence are their resistance to environmental factors and capacity to grow at low temperature in water, soil, foodstuffs, including those kept in home refrigerators. This seems to be one of the most important factors responsible for increased incidence of these diseases in humans.

The main natural reservoirs of the agent are small rodents which can infect foodstuffs, soil, and water by their excretions. In humans Yersinia infection usually involves the gastrointestinal tract. In generalized forms the patterns of the disease vary involving different organs and tissues [1].

Pseudotuberculosis was described in various monkey species much earlier than in humans; there are many reports about susceptibility of many monkey species to this agent [2,6,7,10,12,14]. Reports about infection of various monkey species with *Y. enterocolitica* appeared in many countries at the end of the 60s [1,13]. In some breeding centers this agent is more prevalent than *Yersinia pseudotuberculosis.* Infection with *Y. enterocolitica* was not associated with morphological changes in many cases.

For three decades we recorded cases of Yersinia infection in various species of monkeys in Sukhumi and Adler Breeding Centers. Our results are summed up in this report.

MATERIALS AND METHODS

Monkeys are kept under similar conditions in the Sukhumi and Adler Breeding Centers [4]. All dead animals are subjected to pathomorphological and bacteriological examinations. Generalized Yersinia infection, which could be diagnosed macroscopically, was observed in 52 monkeys, and 14 cases were diagnosed by pathomorphological and bacteriological studies. Specimens for histological studies were fixed in 10%

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neutral formalin, embedded in paraffin, and sections were stained with hematoxylin and eosin. Postmortem bacteriological analyses were carried out routinely. Blood, urine, bile, organs, tissues (liver, spleen, kidney, lung, lymph nodes, and bone marrow), and the contents of the small and large intestine were analyzed. The material was inoculated in common, sugar, serum broths, agar, Ploskirev's bactoagar, 5% blood agar, meat-peptone agar, and cultured at 28 and 37°C. Enzyme activity was studied by 28 tests recommended by International Committee for *Enterobacteriaceae*. Biotyping was performed according to Nilehn protocol. Agglutination test was performed by the volumic method with plague agglutinating serum (Mikrob Institute, Saratov). Serological typing was performed in the indirect hemagglutination (IH) test with commercial O-antigen-typing sera.

Pathogenic properties of isolated strains were studied in guinea pigs, random-bred albino mice, rabbits, and albino rats, which were intraperitoneally infected with 10% suspension of organs of dead monkeys (0.5-1.0 ml).

RESULTS

Pseudotuberculosis started at the end of December 1969 as a group disease, when 12 red monkeys died in 2 months. After that only sporadic cases (no more than 3 in a year) were recorded always in December-March. A total of 49 monkeys died from pseudotuberculosis: 23 red monkeys, 11 *Papio hamadryas,* 9 green monkeys, 2 red macaques, 2 pig-tailed macaques, 1 crab-eating macaque, and 1 *Papio anubis.*

Y. enterocolitica was first isolated in Sukhumi Breeding Center in 1980 from the intestine of an adult *Papio hamadryas* died from hemoblastosis without intestinal symptoms. Later this infection was observed as sporadic and group enteric disease of newly brought monkeys. Seventeen monkeys developed the disease (5 *Papio hamadryas,* 5 crab-eating macaques, 3 rhesus macaques, 2 green monkeys, 1 pig-tailed macaque, and 1 red monkey).

Hence, Yersinia infection was detected in 66 monkeys (24 *Erythrocebus patas,* 16 *Papio hamadryas,* 11 *Cercopithecus aethiops,* 6 *Macaca fascicularis,* 3 *Macaca mulatta,* 3 *Macaca nemestrina,* 2 *Macaca speciosa,* and 1 *Papio anubis*).

A total of 130 strains were isolated from 45 of 66 monkeys. By enzymatic activity 21 cultures were identified as *Y. enterocolitica* and 109 as *Y. pseudotuberculosis.*

Y. enterocolitica was found mainly in the intestinal contents and only in 3 of 17 monkeys in the viscera (in generalized infection). All *Y. enterocolitica* strains were identical. The absence of indole formation, fermentation of tregalose and xylose together with inability to metabolize esculin and salicin classified all these strains as biovar 3 (Nilehn scheme), serotype 0:3 (serotyping was carried out by G. V. Yushchenko at the Central Institute of Epidemiology, Ministry of Health of Russian Federation).

All 109 *Y. pseudotuberculosis* strains isolated from 28 monkeys virtually from all studied organs, blood, bile, urine, bone marrow, and lymph nodes were characterized by typical cultural and biochemical properties. All strains agglutinated in anti-plague serum diluted 1:1280-1:2560 at serum titer of 1:10,240. All strains except 6 were identified as serotype 1.

Biological material and isolated cultures were pathogenic for guinea pigs, albino mice, and rabbits and apathogenic for albino rats. Animals died 5-11 days after infection. Pathological process was characterized by hyperplasia of the spleen and mesenteric lymph nodes with the appearance of numerous yellowish-gray necrotic nodes in these organs and in the liver, lungs, and visceral peritoneum.

Clinical symptoms of Yersinia infection in monkeys were atypical. Several days before death motor activity in some animals decreased, they lost appetite, and sometimes developed petechyal hemorrhages on the skin. Monkeys very rapidly died from pseudotuberculosis with symptoms of severe toxicosis. Enteric Yersinia infection was associated with diarrhea in half of animals.

All cases with pseudotuberculosis and 3 cases with *Y. enterocolitica* infection were generalized and pathomorphologically similar. In cases with generalized disease autopsy showed enlarged liver and spleen with numerous necrotic nodules 0.1-2.0 cm in diameter in these organs (Fig. 1, *a, b*). Hemorrhages were seen in various organs, suppurative processes was observed in some animals. In septic variant of the generalized form (n=12) macroscopic changes were confined to the above-described alterations. In the most frequent enteric variant (n=33) intestinal involvement presented as catarrhal ulcerative colitis, terminal ileitis, and ulcerative colitis. Ulcers were located mainly near Peyer's plaques and solitary follicles. Mesenteric and especially ileocecal lymph nodes were enlarged (Fig. 1, *c*). In pulmonary variant (n=4) necrotic foci were primarily seen in the lungs.

Histological examination showed multiple necrotic foci in the liver, in addition to degeneration and circulatory disorders. Cell detritus and hematoxylin-stained Yersinia colonies were seen in the center of such foci, while the periphery was moderately infiltrated with histiocytes and lymphocytes, sometimes with an admixture of epithelioid and giant polynuclear cells, but the demarcation reaction was poorly expressed.

Large necrotic foci in the spleen involved the white and red pulp and contained Yersinia colonies (Fig. 1,

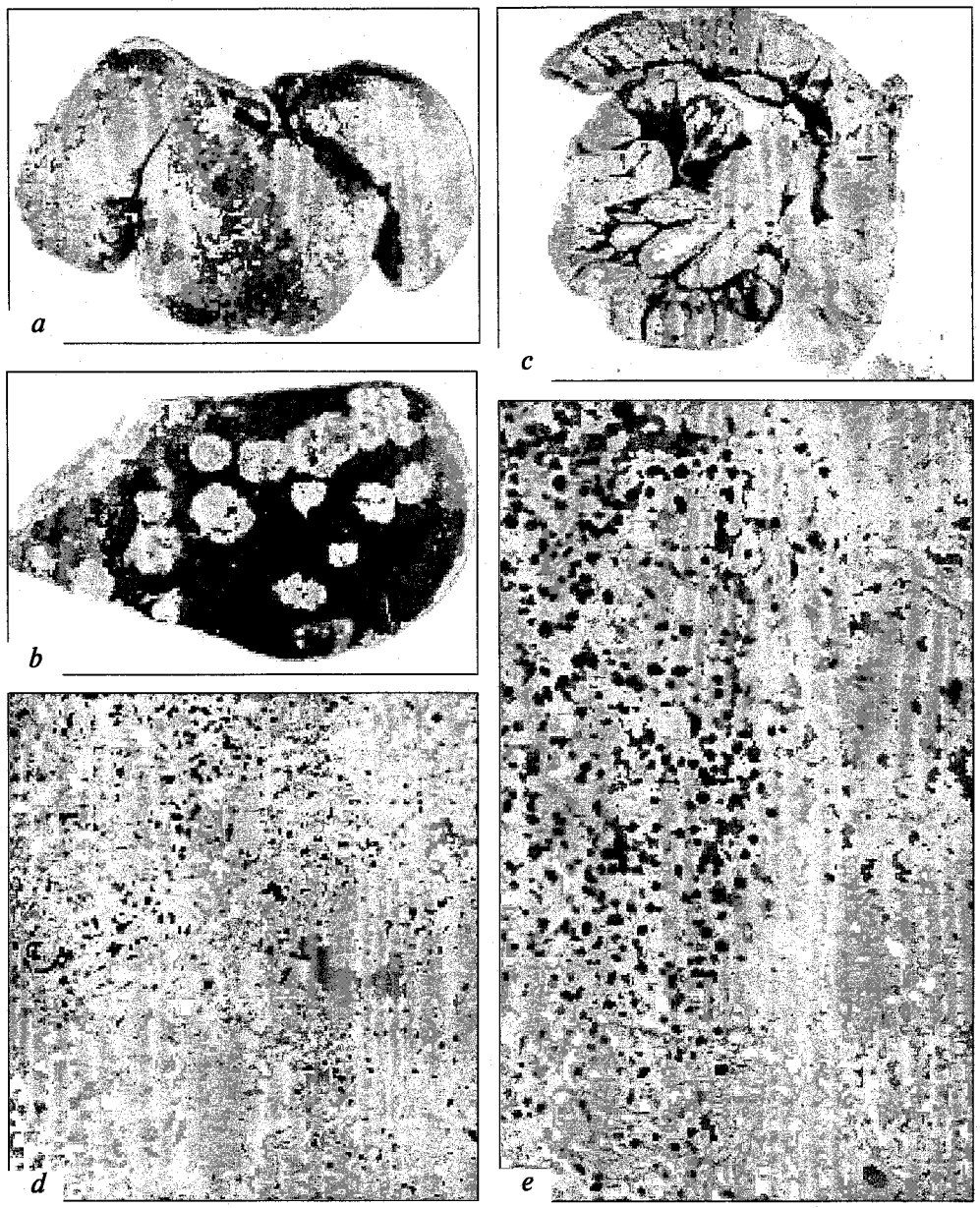

Fig. 1. Pseudotuberculosis in red monkeys. Hematoxylin and eosin staining (*d, e*). *a*) numerous necrotic foci in the liver; *b*) enlarged spleen with large necrotic foci; *c*) terminal ileitis, lymph node hyperplasia in the ileocecal angle; *d*) devastation of splenic follicles, necroses, bacterial colonies, ×80; *e*) necrosis of ileac mucosa (right), rudiments of devastated Peyer's plaque (left), ×180.

d), lymphoid tissue reduction, circulatory disorders, and hemorrhages were seen.

Circulatory disorders predominated in the lymph nodes. Lymphoid tissue, particularly cortical layer, was reduced; solitary necrotic foci were seen. In the enteric variant, necrotic foci and abscesses were seen in the mesenteric lymph nodes, as well as lymphoid cell hyperplasia with the appearance of numerous macrophage histiocytes and sometimes granulomas formed by these cells.

Changes in the gastrointestinal tract in the enteric variant of generalized pseudotuberculosis were characterized by catarrhal desquamative and catarrhal ulcerative forms of gastroenteritis, enteritis, and enterocolitis. Changes in the ileus presented as hemorrhagic inflammation of its terminal portion with hemorrhagic necrosis of the ileocecal valve. The necrotic foci were usually located near intestinal lymphoid tissue (Fig. 1, *e*). The bottom of the ulcer after removal of necrotic masses were presented by follicle residues and bacterial colonies. Reparative processes were poorly expressed. Follicular hyperplasia of enteric lymphoid tissue was more pronounced in minor surface necroses.

Large focal hemorrhages and necrotic foci with bacteria were seen in cases (n=4) with the pulmonary variant of septic form. Bacteria were seen in vessels

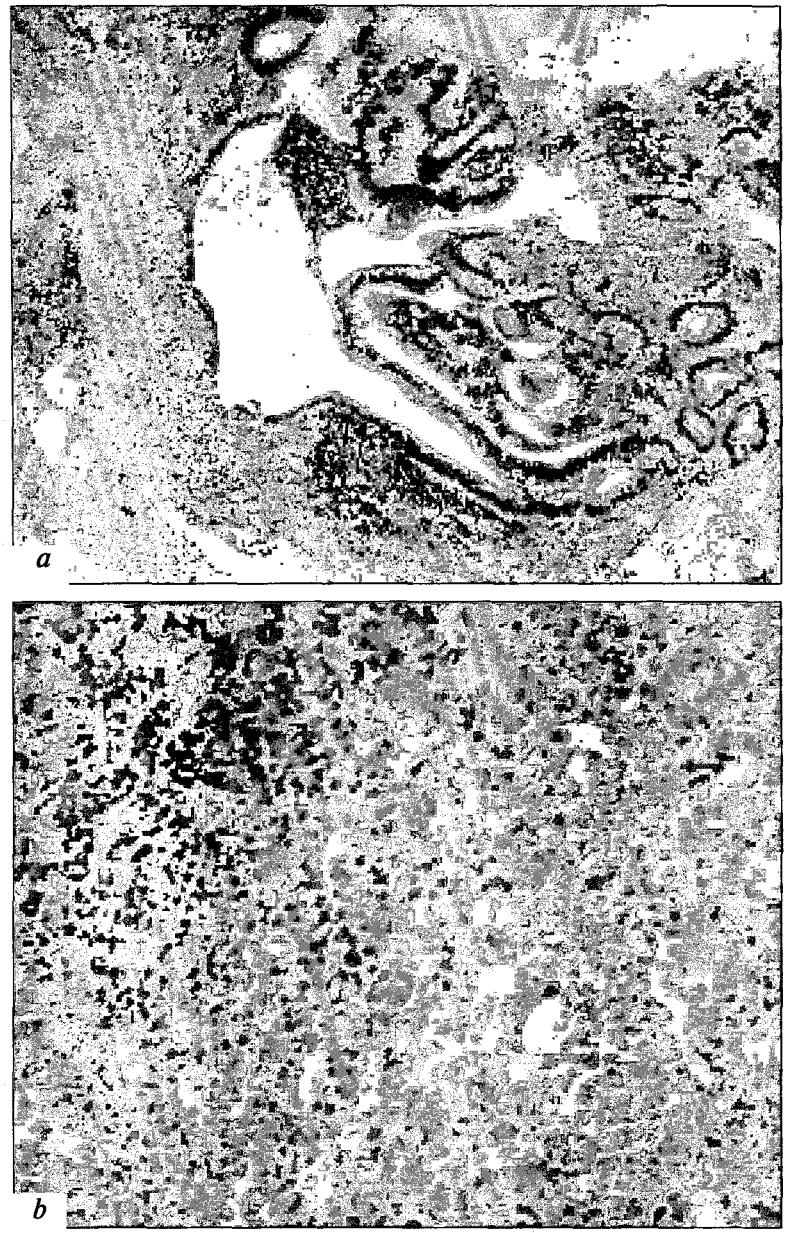

Fig. 2. Enteric Yersinia infection in monkeys. Hematoxylin and eosin staining. *a*) follicular colitis in a *Papio hamadryas*, ×40; *b*) lymphoid tissue hyperplasia in the mesenteric lymph node of a crab-eating macaque (histiocytic reaction), ×400.

as bacterial emboli. Interalveolar septae were slightly infiltrated with lymphocytes and segmented leukocytes.

Infection with *Y. enterocolitica* was local in the majority (10 of 13 manifest forms) of cases. Generalized enteric Yersinia infection was observed in 3 animals and did not differ morphologically from pseudotuberculosis. Macroscopically local Yersinia infection was characterized by the development of catarrhal changes: mostly colitis (n=8) and sometimes gastroenterocolitis and enterocolitis (1 case each) and hyperplasia of mesenteric lymph nodes. Histologically enteric Yersinia infection in monkeys was not characterized by specific features, except that changes were most pronounced in the intestinal lymphoid tissue (Fig. 2, *a*). The reaction of mesenteric lymph nodes with the appearance of numerous histiocytes was more expressed than in common enteric inflammatory processes (Fig. 2, *b*); it was observed in sinuses and pulp cords; permeability of the vascular wall was increased, the walls were impregnated with plasma and fibrin, perivascular hemorrhages were seen. Only occasional necrotic foci in the lymph nodes were found.

No pathomorphological changes except manifestations of lethal pathological processes (trauma, pathological labor, hemoblastosis) were found in 4 mon-

keys. Postmortem isolation of *Y. enterocolitica* in these cases (from the lungs of a green monkey and from intestinal contents in 1 rhesus macaque and 2 *Papio hamadryas*) was regarded as bacterial carriership [10].

Hence, enteric Yersinia infection ran a more benign course than pseudotuberculosis: generalization of the process occurred in more than 20% cases, in the susceptible red monkeys, in old animals, or during gestation. Lethal forms of enteric Yersinia infection were observed mainly in immunocompromized animals: in baby monkeys or during acclimatization stress. In some animals (4 of 17) dead from other diseases the infection presented as asymptomatic carriership.

Species resistance to Yersinia was different. Red monkeys proved to be the most susceptible. They developed generalized infection with pronounced pathomorphological changes after inoculation with both Yersinia variants. Usually it was the only pathological process. Green monkeys were close to them in susceptibility; they also often developed generalized forms, but it occurred mainly in animals with impaired immune defense, in old animals (2 monkeys were aged over 20 years), or severe diseases (hemoblastosis in 2 monkeys). Green monkeys can carry *Y. enterocolitica. Papio hamadryas* are more resistant to infection. Adult animals develop pseudotuberculosis only in the presence of manifest hemoblastosis, and adolescent monkeys only after overcooling. Enteric Yersinia infection was local. *Y. enterocolitica* carriership was observed in adult monkeys. It seems that macaques are also resistant to Yersinia. We observed no pseudotuberculosis in rhesus macaques, 5 cases we observed occurred in other species (during cold time of the year in compromised animals). Outbreaks of spontaneous pseudotuberculosis in rhesus macaques were usually observed during acclimatization [2,14]. Enteric Yersinia infection in these animals courses as a common enteric infection and leads to death only in weak animals.

Symptoms of Yersinia infection in monkeys are similar to those in humans, but monkeys seem to be more susceptible to Yersinia. Pronounced involvement of organs and similarity of the main manifestations of Yersinia infection (particularly of pseudotuberculosis) in monkeys and humans and high susceptibility of monkeys to the agents suggest [3] that monkeys represent a good model for studies of epidemiology, pathogenesis, pathogenicity of various species and serotypes of Yersinia, and postinfectious complications.

REFERENCES

1. A. P. Avtsyn and A. A. Zhavoronkov, *Arkh. Patol.*, No. 5, 4-13 (1980).
2. D. Sh. Beniashvili, *Ibid.*, No. 10, 71-75 (1976).
3. I. V. Marakulin, I. V. Darmov, I. P. Pogorel'skii, *et al., Byull. Eksp. Biol. Med.*, **118,** No. 7, 59-62 (1994).
4. Z. K. Stasilevich, E. K. Dzhikidze, R. I. Krylova, *et al., Lab. Zhivotnye,* No. 6, 217-225 (1996).
5. G. V. Yushchenko, *Natural Focal Anthroponotic Infections* [in Russian], Omsk (1976), pp. 114-115.
6. R. Bronson, B. D. May, and B. H. Ruebner, *Am. J. Pathol.*, **30,** No. 1, 135-142 (1972).
7. E. K. Dsikidse, S. M. Pekerman, I. I. Gorislawets, *et al., Z. Versuchstierkd.*, **14,** 147-153 (1972).
8. B. M. Flynn, P. A. Brown, M. A. Holt, *et al., Lab. Anim. Sci.*, **36,** No. 5, 552-554 (1986).
9. N. S. Mair, G. D. White, F. K. Shubert, *et al., Vet. Rec.*, No. 86, 69-71 (1970).
10. H. M. McClure, *Adv. Vet. Sci. Comp. Med.*, **28,** 267-304 (1984).
11. H. H. Mollaret, W. Giorgi, A. Matera, *et al., Res. Med. Vet.*, **146,** No. 9, 919-924 (1970).
12. J. Mortelmans and P. Kageruka, *Lab. Anim. Handlb.*, No. 4, 95-98 (1969).
13. P. A. Skavlen, H. F. Stills, and C. C. Middleton, *Lab. Anim. Sci.*, **33,** No. 5, 481-483 (1983).
14. H. L. Strickland, B. Rader, and N. D. M. Lehner, *Ibid.*, **32,** No. 4, 426-428 (1982).