[14] – creates favorable conditions for their death and either their subsequent replacement by adipose tissue or the development of immunologic conflict, which lies at the basis of progression of Sjögren's disease. The present investigation shows that a virus disease of the salivary glands can play an important role in the triggering of autoimmune diseases of the salivary glands.

LITERATURE CITED

1. V. S. Kolesov, Vrach. Delo. No. 3, 96 (1983).
2. G. N. Maskalenko, The Principal Stomatologic Diseases [in Russian], Moscow (1981), pp. 153-156.
3. P. S. Polyanskii, Mumps [in Russian], Kiev (1979), p. 112.
4. I. F. Romacheva, L. A. Yudin, V. V. Afanas'ev, and A. N. Morozov, Moscow (1976).*
5. O. V. Rybalov, Abstracts of Proceedings of a Scientific Conference of Stomatologists [in Russian], Poltava (1981), p. 154.
6. O. V. Rybalov, Stomatologia, No. 6, 35 (1982).
7. M. G. Rybakova, Arkh. Patol., No. 11, 85 (1979).
8. G. V. Savel'ev, Z. G. Aprosina, T. I. Lopatkina, and Z. N. Vavilova, Arkh. Patol., No. 1, 44 (1978).
9. L. Sazama, Diseases of the Salivary Glands [in Russian], Prague (1971).
10. V. F. Semenkov, Arkh. Patol., No. 10, 77 (1973).
11. A. C. Thackray, Histological Typing of Salvary Gland Tumours, WHO, Geneva (1972).
12. C. Bimes, D. Barthe, and T. David, Bull. Assoc. Anat. (Nancy), 59, 333 (1975).
13. K. Donath and G. Seifert, Allergologie, 4, 3 (1981).
14. G. Seifert and K. Donath, Beitr. Pathol., 159, 1 (1976).

*As in Russian original; a search of "Ezhegodyne Knigi SSSR" (which lists all books published annually in the USSR) revealed nothing by these authors in 1972-82.

CHANGES IN RESPIRATORY ORGANS IN EXPERIMENTAL ADENOVIRUS INFECTION

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KEY WORDS: alveolar and bronchial epithelium; electron microscopy; adenovirus infection

There have been few studies of the pathomorphology of adenovirus infection [1, 2]. The reason is evidently the difficulty of experimental reproduction of this infection [3-12].

The aim of this investigation was to reproduce adenovirus infection experimentally and to describe the structural changes in the respiratory organs.

EXPERIMENTAL METHOD

Experiments were carried out on six African green monkeys weighing 1.5-2 kg. Type AVL-4 adenoassociated virus, in combination with simian adenovirus S-15 (strain L-1) was instilled into the nasal passages and rubbed into the mucous membrane of the tonsils and conjuctiva of the eyes of these animals. Reinfection with the adenovirus was carried out on the 23rd and 40th days, and was preceded by administration of the immunodepressant cyclophosphamide. In another experiment 286 newborn cotton-tail rats were used and were infected intranasally, under superficial ether anesthesia, with adenoviruses of types 1 and 2. The respiratory organs were studied 6 h and 1-7, 10, 14, and 21 days after infection by histological,

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immunofluorescence and electron-microscopic methods. For histological study the lungs were fixed in 10% neutral formalin solution and embedded in paraffin wax. Sections were stained with hematoxylin and eosin, by the Gram-Weigert, by Van Gieson's method with counterstaining with fuchselin for elastic tissue, by Foot's impregnation method, and by the Feulgen and Branchet reactions. Squash preparations taken from the tracheal mucosa and the lungs were studied in the ML-2 luminescence microscope. The electron-microscopic investigation was carried out by the usual method with the Tesla ABS-500 instrument (Czechoslovakia).

EXPERIMENTAL RESULTS

The temperature of the green monkeys was raised on the 5th day after infection and catarrhal manifestations were observed. On reinfection combined with cyclophosphamide the clinical picture was more marked. The infection followed a latent course in the cottontail rats. All animals excreted adenovirus, and as shown by the immunofluorescence test, adenoviral antigen also. The latter was found in the early period of development of the infection, only in nuclei of the bronchial and alveolar epithelium, and after the 3rd day diffuse fluorescence of the antigen was observed in both nuclei and cytoplasm of the epithelium. Microorganisms could not be found in the lungs of any of the animals after staining by the Gram-Weigert method. No differences in principle were found by analysis of morphological changes in the respiratory organs of the green monkeys and cottontail rats. The characteristic morphological picture of this infection was observed mainly on the 5th-7th days of development of the disease.

No significant changes were found at the light-optical level in the respiratory organs 6 h after injection. Histological study of the trachea and main-trunk bronchi 1-2 days after infection revealed dystrophy of the epithelium, focal desquamation of the epithelium into the lumen and, in some areas, basal cells. Marked circulatory disorders were present in the lung tissue: congestion of the vessels, edema of their walls and of the interalveolar septa. The alveoli contained solitary large cells with an enlarged, basophilic nucleus, staining positively by Feulgen's method.

Electron-microscopic study of animals killed 6 h and 1 day after infection revealed endotheliocytes with redistribution of their nuclear chromatin, in the form of islets and bands, with a narrow perinuclear border of cytoplasm. Processes of the endotheliocytes contained single vacuoles, and their cytoplasm formed cytoplasmic outgrowths, and bridges, protruding into the lumen of the capillaries and partitioning it. The basement membranes were widened to 0.33 μ, with thickening and homogenization of the diffuse layer. Processes of type 1 pneumocytes also formed multiple cytoplasmic outgrowths, projecting into the lumen of the alveoli. In the type 2 pneumocytes most of the osmiophilic bodies lost their typical structure and contained thin laminae; electron-dense areas could be detected in the center of some of these bodies (possibly arising as a result of dystrophic processes and conglomeration of the laminae: Fig. 1a). In other type 2 pneumocytes, in place of the osmiophilic bodies there were vacuoles, and the contents of the osmiophilic bodies could be seen to be ejected into the alveolar space (Fig. 1b). The type 1 pneumocyte had a paddle-shaped nucleus, islets of chromatin, and a narrow band of cytoplasm, distributed perinuclearly, and containing numerous polysomes and ribosomes.

On the 5-7th day of development of the infection large cells with a large basophilic nucleus, almost completely filling the cytoplasm, and staining positively by Feulgen's method, appeared in the lung tissue, and Feulgen-positive inclusions were sometimes visible in the nuclei. Starting with the 2nd day of infection, the interalveolar septa were thickened due to infiltration by round cells and proliferation of septal cells. Focal atelectases were discovered in the lungs on the 7th-14th days. After the 14th-21st day, the cellular structures were restored in two-thirds of the animals, proliferation of collagen fibers were observed in the peribronchial region and in the interalveolar septa.

Electron-microscopic investigation at these same times of development of the infection showed that, compared with earlier times, the endotheliocytes had numerous microprojections with vacuolated cytoplasm. Lymphocytes, monocytes, and eosinophils were frequently discovered in the lumen of the capillaries and in the interstices of the alveoli. Most alveolar spaces appeared as narrow clefts, sometimes the alveolar walls were firmly apposed, the basement membranes were convoluted, and in some areas their outline was indistinct, and they incorporated newly formed collagen fibers (Fig. 2).

Few osmiophilic bodies were observed in the type 2 pneumocytes. The nuclear membranes

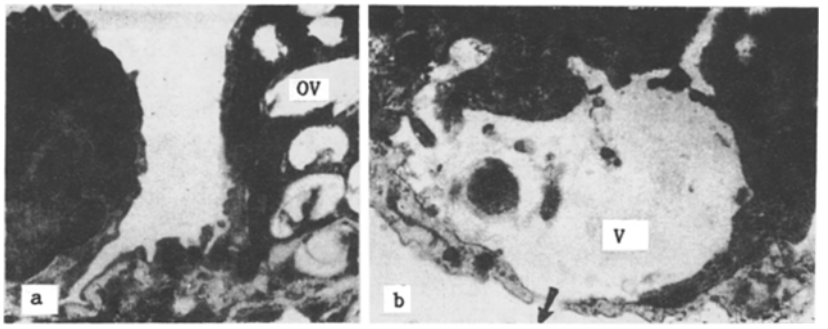

Fig. 1. Type 2 pneumocytes 24 h after infection of animals. a) Nucleus (N) displaced toward apical part of cell, atypical structure of osmiophilic bodies (OB) with electron-dense center. 19,000 ×; b) Vacuoles (V) at site of osmiophilic bodies, contents expelled into alveolar space (arrow). 10,000 ×.

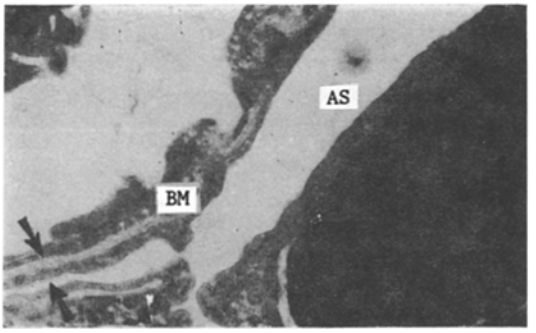

Fig. 2

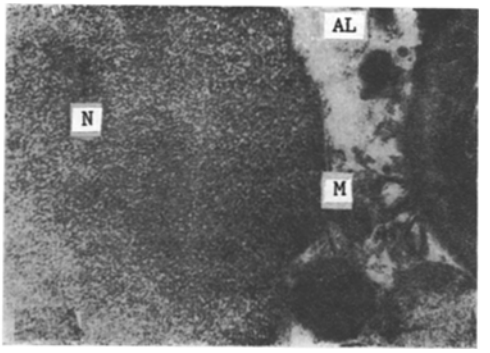

Fig. 3

Fig. 2. Development of adenovirus infection on the 5th day. Alveolar space (AS) appears as narrow cleft (sites of firm apposition indicated by arrows), basement membrane (BM) convoluted, reduced in thickness, and with indistinct outlines. 10,000 ×.

Fig. 3. Development of adenovirus infection on 14th day. "Bare" nuclei of destroyed cells, individual organelles and mitochondria (M) in alveolar lumen (AL). Electron micrograph. 10,000 ×.

were separated into layers and underwent lysis, in some parts the outer nuclear membrane could not be distinguished, the nucleus was enlarged, and almost completely filled the cytoplasm, in which there were a few mitochondria; the diameter of the cells was up to about 12 μ. Desquamated type 2 pneumocytes or individual "bare" nuclei, remnants of osmiophilic bodies, mitochondria, and sometimes lattice-like structures were found in the majority of alveoli (Fig. 3). The thickness of the type 1 pneumocytes was 3 μ, chromatin in the nucleus was arranged in the form of islets and agglomerations around the periphery of the nucleus, and in the perinuclear zone and cytoplasmic processes there were many small mitochondria, polysomes, and ribosomes. Fibroblasts, which contained collagen fibers, were frequently seen beneath the basement membrane. In some observations monocytes and numerous collagen fibers were present in the interstitial tissue, in the interalveolar space.

In experimental adenovirus infection ultrastructural changes thus began to appear 6 h after infection and they continued to be observed until the 10th day of its development. A characteristic feature of this process was the distinctive change in structure of the type 2 pneumocytes: enlargement of the nucleus to 2-2.5 times its normal size, an increase in diameter of the cells to 12 μ and more. These changes were most marked on the 5th-7th day after infection, and after the 14th day of its development the nuclei of these cells began to appear in the lumen of the alveoli. At all times of development of the infection, there was a marked decrease in the number of osmiophilic bodies, with a small number of atypically structured lamellae. Even in the early stages the number of mitochondria in the cytoplasm of

pneumocytes of types both 1 and 2 was reduced, whereas large numbers of polysomes and ribosomes and an endoplasmic reticulum were present. Monocytes, eosinophils, and lymphocytes constantly appeared in the capillaries, especially from the 5th-7th day after infection, evidence of definite allergization of the animals. From the 5th day after infection the formation of collagen fibers was intensified and sclerosis was most marked on the 14th-21st days. It is interesting to note that protein metabolism and protein synthesis were intensified in all cell components of the air-blood barrier, evidently as a result of the action of the virus, as shown by an increase in the number of polysomes and cisterns of the endoplasmic reticulum, and separation of the intermembranous nuclear space, involved in the structure of the endoplasmic reticulum, into separate layers. At the same time, the type 2 pneumocytes contained few osmiophilic bodies, and this state of affairs may have led to a decrease in the production of surfactant and the onset of atelectasis. After the 21st day of experimental infection, no changes were observed in the respiratory organs.

LITERATURE CITED

1. L. O. Vishnevetskaya, Vopr. Okhr. Mat., No. 11, 50 (1964).
2. Z. I. Gavrilov, N. N. Dodonova, and E. S. Voronin, Respiratory Virus Infections [in Russian], Moscow (1963), pp. 176-177.
3. R. S. Dreizin and V. M. Zhdanov, Adenovirus Infections [in Russian], Moscow (1962), p. 287.
4. M. A. Kostyukov, "Adenovirus infection in monkeys," Dissertation for the Degree of Candidate of Medical Sciences, Moscow (1965).
5. O. T. Oganesyan, "Study of the role of maternal adenovirus and influenzal infection in fetal and neonatal pathology," Dissertation for the Degree of Candidate of Medical Sciences, Moscow (1967).
6. A. V. Tsinzerling, E. V. Polonskaya, A. P. Tarasova, et al., Arkh. Patol., No. 10, 21 (1965).
7. R. W. Atchison, B. C. Lasto, and W. M. Hammon, Virology, 29, 353 (1966).
8. A. O. Betts, A. R. Jennings, P. Lamont, et al., Nature, 193, 45 (1962).
9. G. S. Boyer, F. W. Denny, and H. S. Ginsberg, J. Exp. Med., 109, 85 (1959).
10. H. Handa, H. Shiroki, and H. Shomigo, Replication of Mammalian Parvoviruses, New York (1978), pp. 77-86.
11. R. N. Hull, J. R. Minner, and C. C. Mascoli, Am. J. Hyg., 68, 31 (1958).
12. H. G. Pereira and B. Kelly, Nature, 180, 615 (1957).

STEREOLOGIC ANALYSIS OF THE MYOCARDIUM IN ALCOHOLIC CARDIOMYOPATHY

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Acute alcoholic poisoning and long-term alcohol consumption lead to a disturbance of structural protein synthesis [15], inhibition of biosynthesis and the respiratory function of cardiomyocyte mitochondria [14], and reduced contractility of the myocardium as a whole [12], but do not themselves induce the formation of alcoholic cardiomyopathy [11]. An essential, but as yet incompletely studied role in the pathogenesis of human alcoholic cardiomyopathy is played by vitamin deficiency (mainly thiamine) and changes in protein metabolism. Hence it follows that in order to create an adequate model of experimental alcoholic cardiomyopathy,

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