AN EPIDEMIC IN A MOUSE COLONY DUE TO THE VIRUS OF ACUTE LYMPHOCYTIC CHORIOMENINGITIS

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PLATES 41 AND 42

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In a preliminary publication (1) the detection of a disease due to a filtrable virus in our colony of white mice was reported. The purpose of the present paper is to describe this disease in greater detail.

The Disease in Mice

Based on the number of immune mice, it was estimated that about 50 per cent of the mice of the colony were infected. The rates of morbidity and mortality cannot be given accurately; the former, however, was less than 20 per cent, and the latter less than 2 per cent of the number of infected mice. The majority of the infected stock mice showed no definite symptoms, and the infection was determined by inoculating their blood or emulsions of their brains into guinea pigs, or by testing the mice for immunity by intracerebral inoculation with virus, using equal numbers of mice from a virus-free stock as controls. Stock mice which proved resistant to intracerebral inoculation with virus very probably had acquired a true immunity following natural infection and were not merely naturally resistant to the virus, since mice of the same strain bred from virus-free parents are all susceptible to intracerebral inoculation with virus. That a subclinical, latent infection is the rule in mice became apparent from several experiments to be described in a following paper in which disease-free mice were infected by placing them in contact with infected ones.

In the infected colony it was noted that a number of 2 to 6 week old mice were emaciated and drowsy. Their fur was ruffled and they were often seen sitting in corners of the cage by themselves. Their movements were rather slow and stiff, and their legs appeared long in

proportion to their thin bodies. Nineteen mice presenting such symptoms (Group C, Table I) were examined for virus by inoculating suspensions of their brains or blood or both intracerebrally into guinea pigs, and the virus was recovered from fourteen of them. Four mice presenting such symptoms and carrying virus in their blood were put in a cage by themselves and observed for 3 weeks, during which time they made a complete recovery.

The symptoms most frequently presented by naturally infected young mice cannot be regarded as pathognostic. The tremors and

TABLE I

Symptoms Presented by Naturally Infected Mice

Group	Condition of mice	No. of mice examined	Test for virus by inoculating blood or brain or both into guinea pigs	
			No. positive	No. negative
A	No symptoms; normal rate of growth	At least 13*	9	4
B	Conjunctivitis and photophobia; no other symptoms	5	2	3
C	Emaciation; ruffled fur; somnolence; slow, stiff movements; slow rate of growth	19	14	5
D	Found dead in colony	3	2	1

* The exact figure cannot be given, because in ten cases the pooled blood of from five to thirty healthy mice was injected. Each of these cases is counted as one mouse.

spastic convulsions which characterize the experimental disease in intracerebrally injected mice have not been observed in naturally occurring cases.

In Table I a list is given of mice which were examined for virus by inoculating heart blood or suspensions of brains or both intracerebrally into guinea pigs, and of the symptoms noted in the mice.

As mentioned before (1), a striking clinical picture can be produced in some mice which have a latent infection by an intracerebral injection with sterile bouillon. In 3 to 13 days after the inoculation such mice present symptoms indistinguishable from those shown by mice inoculated intracerebrally with virus: marked tremors, passing into

spastic convulsions of the hind legs when the animals are lifted by the tail.

The Experimental Disease in Mice

Intracerebral Inoculation.—In mice from the infected stock the rate of morbidity following intracerebral inoculation with virus was about 60 per cent, and the rate of mortality about 40 per cent, while in mice of the same strain which were bred from noninfected parents the rates of morbidity and mortality were practically 100 per cent. The incubation period is on the average 6 to 7 days and may vary from 5 to 12 days according to the amount of virus injected. It has never been shorter than 5 days, no matter how much virus was inoculated. On the 6th or 7th day the mice appear ill and show signs of general malaise. They are no longer lively and often sit quietly alone. Their fur is usually ruffled. When the animals are lifted by the tail, rapid motions followed by very distinct tremors of the front and hind legs result, the latter being somewhat retracted. As the disease progresses such tremors often pass into a striking spastic convulsion of the hind legs. The front legs are rarely involved in this convulsion. When the animals are dropped during the convulsion, they may lie on the side, the hind legs being stiffly stretched out, the tail rigid, and the back appearing humped. The front legs carry out a series of very rapid motions. Animals that gain the erect posture may drag themselves about the table by means of the front legs, the hind legs remaining stretched out and rigid (Fig. 1). The convulsions usually last from one to several minutes. Many mice die during the first convulsion brought about by lifting them by the tail. Others recover from the convulsion and are then able to walk about normally. Several minutes usually elapse before a second convulsion can be induced. Practically all affected mice ultimately die in convulsions, as evidenced by the position of their hind legs during rigor mortis. Death occurs in 1 to 2 days after the onset of symptoms.

Intranasal Instillation.—The intranasal instillations were carried out as follows: About 5 cc. of a 1 per cent suspension (in saline) of infective mouse brain were placed in a sterile Petri dish, and the nostrils of a slightly etherized mouse were dipped once or twice into the suspension. By this procedure a small drop of suspension became attached to the nostrils and was inhaled by the mouse immediately. In this manner the dosage could not be accurately measured, but the method is probably more similar to the natural mode of infection (if that takes place by way of the nasal passages) than the instillation of a large amount of suspension.

In the first experiment eight 6 week old mice bred from disease-free mothers were used. They were bled from the heart and their pooled defibrinated blood was inoculated into a guinea pig (0.2 cc. intracerebrally and 0.5 cc. subcutaneously into each planta), with negative result. The eight mice were then inoculated intranasally with virus. From each of two mice, 0.2 cc. blood was drawn on the 7th day after the exposure and inoculated immediately (not defibrinated) into the brain of a guinea pig. Both guinea pigs developed the disease and died on

the 14th and the 17th day, respectively, after the inoculation. Two mice were bled in the same manner on the 11th day after the exposure, and their blood was also virulent for guinea pigs. The four remaining mice were bled similarly on the 11th day and again on the 16th day. All blood samples obtained from them were avirulent for guinea pigs. None of the eight mice showed the slightest symptoms. 3 weeks after the exposure they were tested for immunity by intracerebral inoculation with virus, and the four mice which carried virus in their blood were immune; while the four others, as well as the eight normal control mice, died.

In another experiment, five half grown mice bred from disease-free parents were inoculated intranasally with virus. The animals showed no symptoms. On the 10th day after the inoculation each mouse was bled by heart puncture, and 0.2 cc. defibrinated blood was inoculated immediately into the brain of a guinea pig. None of the guinea pigs showed fever or symptoms; but one became immunized by the blood inoculation, while the four others died following the test inoculation. The mice were tested for immunity by intracerebral inoculation with virus on the 23rd day following the inoculation. One mouse, as well as six normal control mice, died; while four mice, including the one whose blood immunized a guinea pig, showed no symptoms. This result indicated that four of the five mice had become infected by the virus given intranasally.

Intraperitoneal Inoculation.—Of mice bred from virus-free, susceptible mothers about 60 per cent showed symptoms 5 to 7 days after intraperitoneal inoculation with virus. The symptoms differed from those presented by mice inoculated intracerebrally in that there was markedly labored breathing and no tremors or convulsions. The symptoms lasted for about a week, and all such mice finally recovered.

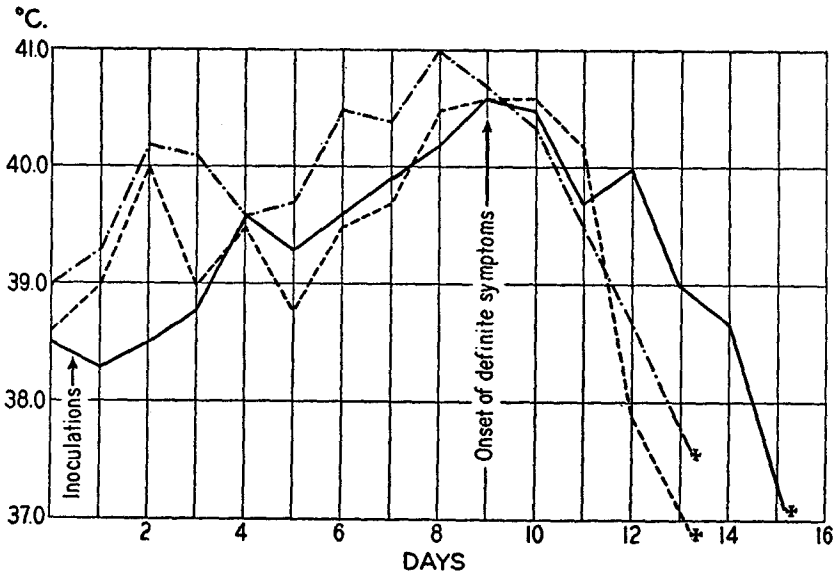
Intravenous Inoculation.—About 70 per cent of the disease-free mice injected intravenously with the supernatant fluid of a virulent guinea pig brain emulsion showed symptoms after 6 to 10 days. The symptoms were similar to those exhibited by intraperitoneally inoculated mice, except that the labored respiration was more striking. Spastic convulsions and tremors were not noted. Three out of 42 mice died; the remaining sick ones recovered slowly or were sacrificed for histological examination when they showed definite symptoms.

The Experimental Disease in Guinea Pigs

The virus has been transmitted to guinea pigs by intracerebral and subcutaneous inoculation[1] and by intranasal instillation. In a comparative titration experiment the minimal infective dose was equal for intracerebral and subcutaneous inoculation. The rates of morbidity and mortality have varied considerably with different strains

[1] All subcutaneous inoculations were made into one or both footpads in the plantar region.

of virus, but every strain produced at least a marked febrile reaction followed by a very solid immunity to a highly virulent virus. We have two strains of virus, maintained by passages through guinea pigs, which kill over 80 per cent of the guinea pigs following intracerebral and subcutaneous inoculation, and produce a very severe disease in the remaining animals. These two strains were used in neutralization tests.



TEXT-FIG. 1

—— Guinea pig inoculated intranasally with bacteria-free Berkefeld N filtrate.

-·- Guinea pig inoculated intracerebrally with mouse brain suspension.

--- Guinea pig inoculated subcutaneously with mouse brain suspension.

The course of the disease in guinea pigs is rather slow, and the symptoms, when marked, are pathognostic. Unlike the disease in mice, there is practically no difference in the incubation period nor in the clinical picture following intracerebral, subcutaneous, and intranasal inoculations with virus. The most constant clinical feature is fever (a body temperature of 40°C. and above is considered febrile, except on hot summer days when normal guinea pigs may show temperatures of 40.5°C.). In Text-fig. 1 three typical temperature curves of experimentally infected guinea pigs are given. Other evidences of disease seldom

appear before the 9th day. There is a considerable loss of weight, and a markedly labored breathing, which may last for a few days, and gradually pass into dyspnea if the animal dies, or slowly subside if the animal recovers. In severe cases the emaciation is very pronounced, and recovering guinea pigs require a long period of time to regain the lost weight. Somnolence, salivation, and a marked seropurulent conjunctivitis are also present in severe cases. Death rarely occurs before the 12th day, and it may be delayed as long as 30 days.

The pathogenicity of a sample of the virus may vary for different guinea pigs. One animal may merely show fever and no other symptoms, while another of the same stock and the same size, inoculated with the same amount of virus at the same time, may develop a most severe, fatal disease. In all cases in which fever was the only sign of the disease, the guinea pigs were tested for immunity, usually 3 weeks after the first inoculation, by an intracerebral or subcutaneous injection with a highly virulent virus, using an adequate number of normal guinea pigs as controls.

Inoculation of Other Animals

Rats.—Five of seven white rats inoculated intracerebrally with virus died in 6 to 9 days following the inoculation. One rat became very ill on the 7th day and recovered, and one rat showed no symptoms. The clinical picture in rats was characterized by clonic-tonic spasms of the muscles of the legs and neck. The symptoms on the whole were somewhat similar to those presented by intracerebrally injected mice.

Rabbits.—Two rabbits inoculated with infective mouse brain suspension intracerebrally, intranasally, intracutaneously, intratesticularly, and by rubbing virus into the scarified corneae showed no fever and no symptoms during 1 month's observation. 3 weeks after the inoculation the rabbits were bled, and in their pooled serum neutralizing antibodies were detected. Two more rabbits were inoculated intracerebrally with infectious mouse brain suspensions. One rabbit showed no fever and no symptoms. The other had fever (41.0°C.) on the 3rd day following the inoculation. On the 4th day its temperature was again normal, and the animal never showed symptoms.

Chicks and Pigeons.—Four chicks, 6 weeks old, and two pigeons inoculated intracerebrally with virus showed no symptoms.

The Pathological Changes Produced by the Virus

Technique.—The organs examined were fixed in Zenker's fluid or, less frequently, in formalin or a combination of both. The fixed tissues were embedded in paraffin. The following staining methods were used: Iron alum hematoxylin and Van Gieson (particularly for sections of brain and spinal cord examined for changes of nerve cells), hematoxylin and eosin, Giemsa stain, eosin and methylene blue (for sections of tissues examined for acidophilic inclusion bodies), and phloxine and methylene blue. All animals (except one mouse inoculated intranasally which showed no symptoms) were killed for histological examination when they presented definite symptoms, or the organs were removed immediately after death.

Naturally Infected Mice.—The pathological changes in naturally infected mice presenting definite symptoms were scanty. Macroscopically no lesions were noted. Histologically the majority of such mice showed changes in the liver: small collections of round cells in the vicinity of blood vessels and scattered lymphocytes, single or in small groups, in the interstitial tissue. Patchy reticuloendothelial hyperplasia was frequent in the liver. In the lungs of two mice slight peribronchial and perivascular infiltrations with round cells and a slight thickening of the alveolar walls were noted. Only one of the twelve mice examined presented a slight meningitis, the exudate cells being predominantly lymphocytes. The chorioid plexuses were not involved.

Mice Inoculated Intracerebrally with Virus, and Naturally Infected Mice Inoculated Intracerebrally with Sterile Bouillon.—No definite macroscopical lesions occurred regularly in mice killed in the stage of spastic convulsions or autopsied immediately after death. In some cases the liver had a more or less pronounced nutmeg color, and the spleen was slightly enlarged. The lungs appeared normal in every case.

Microscopically a more or less marked infiltration of the meninges with round cells was noted in all of seventeen mice examined histologically. The predominating cells in the meningeal exudate were lymphocytes. Mononuclear cells were next in frequency, and a small percentage of the cells were polymorphonuclear leucocytes.[2] The meningitis was usually most intense at the base of the brain (Fig. 2). In many mice round cell infiltration was also noted in the pia-arachnoid of the spinal cord. In thirteen of the seventeen mice the chorioid plexuses were infiltrated with round cells, and in four cases the plexuses were not involved. In ten cases the infiltration was very intense. The plexuses of the third and fourth ventricles showed infiltration more frequently and more intensely than the side ventricles. In some cases many round cells had accumulated in the ventricles outside the plexuses, and in one case the third ventricle was almost completely filled with such cells. The ependyma was frequently infiltrated, and there was a moderate degree of subependymal gliosis. Very rarely the ependyma was pro-

[2] Cultures of all brains saved for histology showed no growth.

liferating. Round cell infiltrations of the perivascular lymph spaces of submeningeal and subependymal vessels were noted in ten of fifteen brains. In cross-sections of such vessels and of the vessels of the pia mater unusually large numbers of lymphocytes were often present. Some round cells in the exudate of the meninges, chorioid plexuses, and in the perivascular lymph spaces were in mitosis.

In three brains more or less large areas of the stem were infiltrated with round cells. Small collections of oligodendroglia cells were seen in a few mice in the cerebral cortex at the base of the brain. In the cerebellum of many mice small numbers of pycnotic and shrunken Purkinje cells were scattered in the rows among perfectly normal cells of this type. In the spinal cord one or two degenerated ventral horn cells surrounded by oligodendroglia and microglia cells were occasionally seen. On the whole, nerve cell degeneration and neuronophagy were not frequent, and the changes in the nervous tissue proper were few.

No inclusions were found in any types of cells.

In other organs no definite changes were noted. In several liver sections there were patchy reticuloendothelial hyperplasia and a few small collections of lymphocytes in the vicinity of blood vessels. A slight thickening of the alveolar walls in areas of the lungs and a slight bronchitis and peribronchitis (round cell collections) were noted in a few mice, but the pulmonary lesions were not nearly so marked as in guinea pigs. No changes were found in the salivary glands of mice, and it is believed that the virus is not related to the salivary gland viruses. Blood counts on a number of intracerebrally injected mice revealed no definite abnormalities.

Intranasally Injected Mice.—The organs of a mouse which had contracted a latent infection by intranasal instillation of virus were examined histologically (mouse killed on the 7th day after exposure) and no definite changes were noted. There was no evidence of meningitis or pneumonia.

Intraperitoneally Injected Mice.—In mice killed when the symptoms were very marked the spleen was often enlarged (1½ to 2 times normal size), and the liver had a nutmeg color and was slightly swollen. The lungs contained no consolidated areas. About 20 per cent of the mice autopsied presented a serous pleuritis and peritonitis, the pleural and peritoneal cavities containing an almost clear, serous exudate, films of which stained with Giemsa stain revealed a considerable number of lymphocytes and macrophages.

The organs of six mice were examined microscopically. In only one case was a slight meningitis noted; the chorioid plexuses were not involved. In all other cases the meninges appeared normal. In liver sections of all mice patchy proliferation of reticuloendothelial elements was noted, giving the section a spotted appearance when examined with lower magnification. Single or small groups of round cells were scattered throughout the interstitial tissue. More or less extensive collections of lymphocytes and mononuclears around blood vessels were frequent findings. In mice presenting serous pleuritis and peritonitis the pleural or peritoneal endothelial cells were swollen, and the pleura and peritoneum were slightly infiltrated with round cells. In the spleen the Malpighian bodies appeared

enlarged, and there was some reticuloendothelial hyperplasia. In two cases a definite interstitial pneumonia with small round cell collections around blood vessels was noted, while in four cases no definite pneumonia was present. Other organs showed no marked changes.

Blood counts on a limited number of mice injected intraperitoneally revealed no marked changes of the blood picture.

Intravenously Injected Mice.—The changes in mice showing symptoms following intravenous inoculation with virus were similar to those in intraperitoneally injected mice but more marked. In all cases the spleen was considerably enlarged, its volume being from 3 to 6 times greater than normal. The enlargement of the spleen could be readily detected in living animals by palpation. The liver appeared somewhat pale and slightly swollen. In one case one kidney was much enlarged. The lymph nodes were not definitely enlarged. A few mice presented serous pleuritis. The lungs appeared normal macroscopically. In the spleen the Malpighian bodies were enlarged, and the red pulp was more or less infiltrated with lymphocytes and mononuclear cells. The reticuloendothelial elements showed hyperplasia. In some lymph nodes reticuloendothelial hyperplasia was also noted. In the liver large round cell collections were present around the larger blood vessels, and smaller collections around the central veins and in association with proliferated Kupffer cells. Necrosis of some liver cells was noted in the immediate vicinity of the round cell aggregates. Round cell collections were also present in the kidneys, and in the case in which a marked enlargement of one kidney was noted macroscopically, huge round cell collections were present in the atrophic renal cortex and in the renal pelvis. Lung sections revealed an interstitial pneumonia in every case, and more or less extensive round cell collections around blood vessels and bronchi. In cases of pleuritis the pleural cells were swollen, and the pleura was moderately infiltrated with round cells. The endocardium and epicardium contained more or less large round cell collections. In some cases the meninges and chorioid plexuses were moderately infiltrated with round cells.

Blood counts revealed a marked increase of white blood cells (up to 55,000 per c.mm.) in three of seven cases. A more or less pronounced monocytosis (4 to 23.5 per cent) was noted in six of seven cases, and a lymphocytosis (62.5 to 80 per cent) in five cases.

Guinea Pigs.—The outstanding lesions produced by the virus in guinea pigs following intracerebral or subcutaneous inoculation or intranasal instillation are in the lungs and, less regularly, in the heart. At autopsy in an advanced stage of the disease (dyspnea) more or less extensive consolidated areas are usually present in the lungs. Such areas may be found in several lobes with no special site of predilection. In many cases lung tissue from pneumonic areas was sterile, while from others bacteria of different types were cultivated. The heart may be slightly enlarged and the ventricles slightly dilated. A few cc. of clear serous fluid were present in the thoracic cavities of some guinea pigs, and such animals usually presented a marked subcutaneous edema at the lower parts of the abdomen,

the subcutaneous tissue consisting of a thick, gelatinous mass. Some guinea pigs showed small necrotic areas of light color and rather hard consistency in the liver. The central nervous system presented nothing abnormal except a definite excess of cerebrospinal fluid in some cases.

Material from 31 guinea pigs infected by different routes was studied histologically. The dominant lesion was a typical virus pneumonia which was present in every animal sacrificed in a late stage of the disease. Lungs which appeared normal macroscopically presented pneumonic areas microscopically. A marked pulmonary edema was noted in many guinea pigs.

Heart changes, present in the majority of the guinea pigs, consisted mainly of subendothelial infiltrations with round cells. Very small, scattered round cell collections were also seen in the myocardium and in the subepicardial layer. The cardiac muscle tissue was not affected.

The infiltration of the meninges with lymphocytes and mononuclear cells (polymorphonuclear leucocytes were entirely absent) was much less intense than in mice. A meningitis was noted in four of seven guinea pigs inoculated intracerebrally with virus, and in nine of twelve guinea pigs inoculated subcutaneously. The infiltration of the chorioid plexuses was never very marked, the inflammatory cells, if present, consisting of a few lymphocytes and mononuclear cells.

In four of eight guinea pigs inoculated subcutaneously, and in four of six guinea pigs inoculated intracerebrally, intranuclear eosinophilic bodies were seen in cells of the pia mater, in mononuclear cells scattered along the meninges, and in some adventitial cells of meningeal vessels, as well as in glia cells at the periphery of the cerebral cortex. The majority of these intranuclear bodies were small, and not always so distinct as those shown in the photograph (Fig. 4). Many of them appeared as deep red, small granules, single or in small groups in a nucleus. When the bodies were as large as those in the picture, there was a definite hyperchromatosis of the nuclear membrane. In sections from five normal guinea pigs and from a number of guinea pigs inoculated with equine encephalomyelitis virus such intranuclear bodies were not found. In one of six guinea pigs inoculated intracerebrally with mouse virus, inclusion bodies were also present in a few epithelial cells of the chorioid plexus of a side ventricle. These bodies were round and stained faintly red with eosin. Because of the small number of normal brains examined, the relation of these intranuclear bodies to the mouse virus is not definitely established. Their etiological significance is doubtful, since they are absent in mice and in the lungs of guinea pigs.

Perivascular cuffs were rarely seen in sections of the brains of guinea pigs. If present, they consisted of one layer of mononuclear cells and lymphocytes. Glia proliferation was never noted, and no damage to nerve cells was detected except in a few scattered Purkinje cells which appeared pycnotic and shrunken when stained with iron hematoxylin in combination with van Gieson's method.

As a rule many liver cells showed vacuoles in the cytoplasm, and small round cell collections around blood vessels were seen in a number of liver sections. In about 20 per cent of the guinea pigs examined necrosis of groups of adjacent liver

lobules was noted. It is questionable, however, whether such necrosis was attributable to the action of the virus. In all guinea pigs presenting conjunctivitis the conjunctival epithelium was slightly infiltrated with polymorphonuclear exudate in the conjunctival sac. No definite changes were detected in the spleen, kidneys, pancreas, salivary glands, adrenals, or lymph nodes.

Rats.—Histologically a marked infiltration of the chorioid plexuses of the side ventricles with round cells was noted (Fig. 3). The meningitis was less intense than in mice but more marked than in guinea pigs.

The Distribution of the Virus in the Body of Mice and Guinea Pigs

In mice showing symptoms following intracerebral, intraperitoneal, or intravenous inoculation the virus was invariably present in the brain and blood stream, and often in the urine and nasal secretions.

In guinea pigs showing definite symptoms following subcutaneous or intracerebral inoculation virus was present in the blood, brain, lung, and urine. The virus content of the brain of a subcutaneously infected guinea pig which died about a minute before blood was drawn, was higher than that of the blood.

Serological Relationship of the Virus to Other Viruses

Because of the similarity from a clinical and pathological viewpoint of the mouse virus and the virus of acute lymphocytic choriomeningitis described by Armstrong and Lillie (2), samples of immune serum prepared against the respective viruses were exchanged with Dr. Armstrong. Cross-neutralization tests revealed the serological identity of the two viruses. These tests are recorded in a paper by Armstrong and Dickens (3). Shortly after the isolation of our virus from mice, Rivers and Scott (4) obtained from two human patients suffering from acute meningitis a filtrable virus which those authors have concluded is closely related to ours and to that of Armstrong and Lillie or immunologically identical with them.

There is no similarity between this disease and other known virus diseases of mice. Infectious ectromelia (5) and spontaneous encephalitis of mice (6) are quite different both clinically and pathologically.

Tests were made to determine the relationship of this virus to those human viruses that are readily transmitted to mice. Sera of horses immune to the human or the swine influenza virus[3] failed to neutralize

[3] These sera were obtained by Dr. R. E. Shope from Dr. C. H. Andrewes of Hampstead, England.

the virus under consideration. The relationship to lymphogranuloma inguinale (climatic bubo) was tested as follows:

A guinea pig recovered from typical experimental bubos in the inguinal lymph nodes, and two mice recovered from lymphogranuloma infection following intracerebral inoculation with virus were obtained through the courtesy of Dr. A. W. Grace of the New York Hospital. They were inoculated with our virus and all succumbed, as did the normal control animals. Later seven mice which had recovered in our laboratory from lymphogranuloma infection induced by intracerebral inoculation with virus obtained from Dr. Grace, were injected intracerebrally with mouse virus. Five of them died in 7 to 8 days, and two became typically ill and recovered. All ten control mice died in 6 to 8 days. Ten mice solidly immune to intracerebral inoculation with mouse virus, and ten normal control mice were inoculated intracerebrally with lymphogranuloma virus. Of the immune mice, six became sick 8 to 14 days after the inoculation and recovered, and four showed no symptoms. Of the control mice, seven became sick at the same time and recovered, and three showed no symptoms. These experiments suggest that the mouse virus and lymphogranuloma virus do not cross-immunize. Cross-neutralization tests gave no clear cut results, because our mice are apparently not susceptible enough to lymphogranuloma, and the serum (drawn before the test inoculation) of the guinea pig immune to lymphogranuloma did not have sufficient neutralizing power on lymphogranuloma virus to permit a conclusion as to its effect on the mouse virus.

DISCUSSION

The disease caused by the virus under natural conditions in mice is mild, with a rather high rate of infection, but with a low rate of morbidity and a very low rate of mortality. Symptoms were presented by only a small percentage of young, 2 to 6 week old mice, and were not striking. The disease is not readily recognized unless transfers of material are made from infected mice to guinea pigs, or to normal mice by intracerebral inoculation.

On the other hand, mice inoculated intracerebrally with infectious material show very striking symptoms which are probably pathognostic. Diseased guinea pigs also present symptoms more or less specific for the virus. It is suggested that for diagnostic purposes intracerebral inoculations in mice and guinea pigs be carried out, and that the body temperatures of the latter be recorded daily for 2 weeks.

Different species of rodents are differently affected by intracerebral injection of this virus. Rabbits are apparently resistant. Guinea pigs develop pneumonia and a mild meningitis. White rats respond

with a marked meningitis and inflammation of the chorioid plexuses. White mice as a rule show a marked meningitis and a more or less pronounced round cell infiltration of the plexuses and ventricles, and the pulmonary lesions, if present, are usually not very marked. The symptomatology in guinea pigs, rats, and mice corresponds to the pathological picture. In the first, respiratory symptoms dominate; while in rats and mice spastic convulsions are the most striking clinical feature, for which the anatomical basis seems to be the inflammation of the meninges and chorioid plexuses and a possible overproduction of cerebrospinal fluid and consequently increased intracranial pressure.

In general, the pathological changes produced by this virus are inflammation and hyperplasia. Necrosis is very rare.

Besides serological identity, the virus of acute lymphocytic choriomeningitis and our virus have other common features, such as the striking symptoms produced by them in mice inoculated intracerebrally. The description of these symptoms given by Armstrong and Lillie (2) can be applied to mice injected intracerebrally with our virus almost word for word, and the pathological picture presented by such mice is similar to that described by Lillie (2). Both viruses are present in the brain and blood of the mice, and the pathogenicity of the viruses for laboratory animals as far as tested differs for only one species, the white rat. While Armstrong and Lillie could not infect rats, our virus caused symptoms in six of seven rats inoculated intracerebrally. It is possible that a difference in the strains of rats used was responsible for this discrepancy rather than a difference in the viruses. With both viruses intranasal instillations into mice produced no symptoms.

On the whole the evidence presented is believed to justify the conclusion that our virus is identical with the virus of acute lymphocytic choriomeningitis.

The name lymphocytic choriomeningitis does not describe the disease naturally occurring in mice, since choriomeningitis is rare in such cases. The designation is, however, adequate for the experimental disease produced in mice by intracerebral inoculation.

SUMMARY

A filtrable virus, identical with that which causes acute lymphocytic choriomeningitis, has been found to cause a disease in white mice.

Naturally infected mice usually show no symptoms, but such animals inoculated intracerebrally with sterile bouillon or other materials develop characteristic symptoms. The same symptoms are produced by intracerebral injection of the virus into mice from a disease-free stock. Guinea pigs are very susceptible and are therefore useful for detecting the virus and for neutralization tests. The disease in both naturally infected and inoculated animals is discussed and the pathological findings given.

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EXPLANATION OF PLATES

PLATE 41

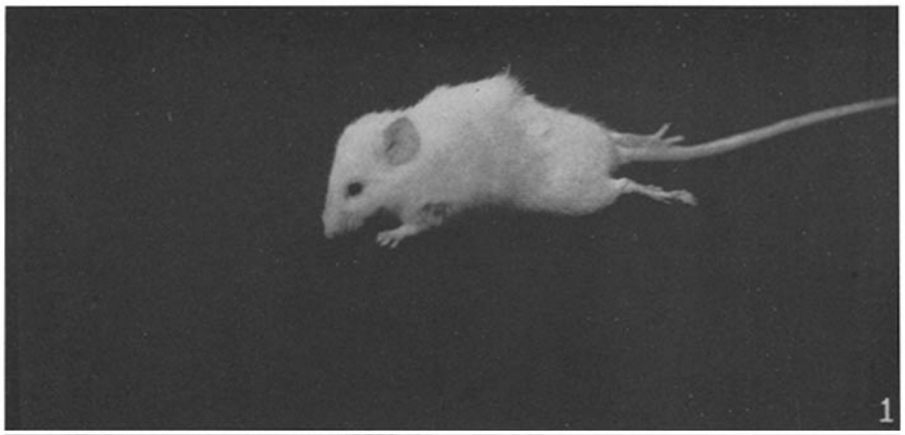
FIG. 1. Mouse showing typical symptoms on the 7th day following intracerebral inoculation with virus. Spastic convulsion of hind legs after being lifted by the tail. The mouse is dragging itself about the table with the front legs.

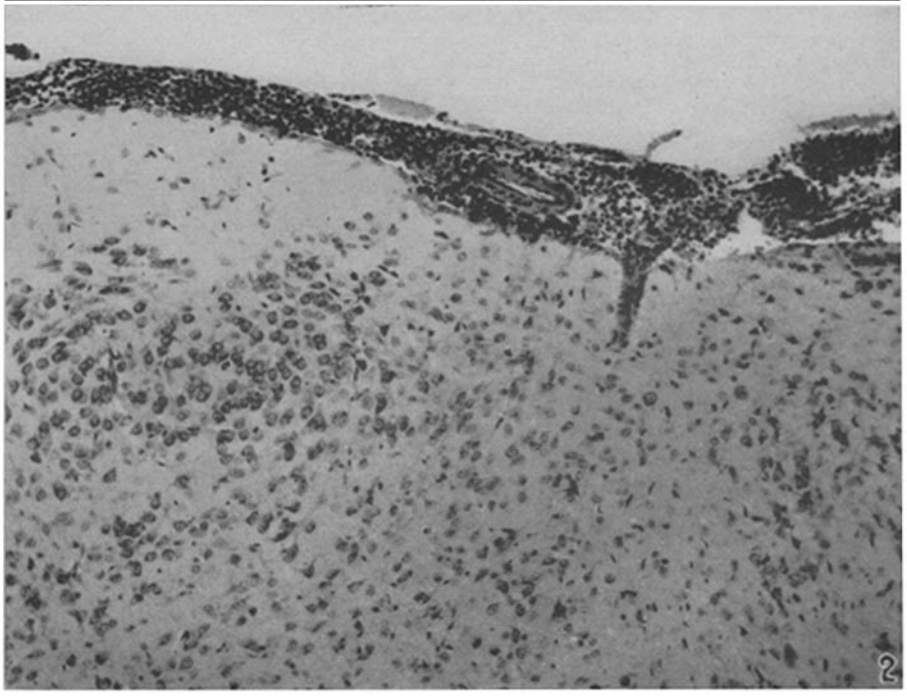
FIG. 2. Mouse, intracerebral inoculation with virus. Marked meningitis at base of brain. Eosin and methylene blue. ×130.

PLATE 42

FIG. 3. White rat, intracerebral inoculation. Inflammation of the chorioid plexus of a side ventricle. Eosin and methylene blue. ×100.

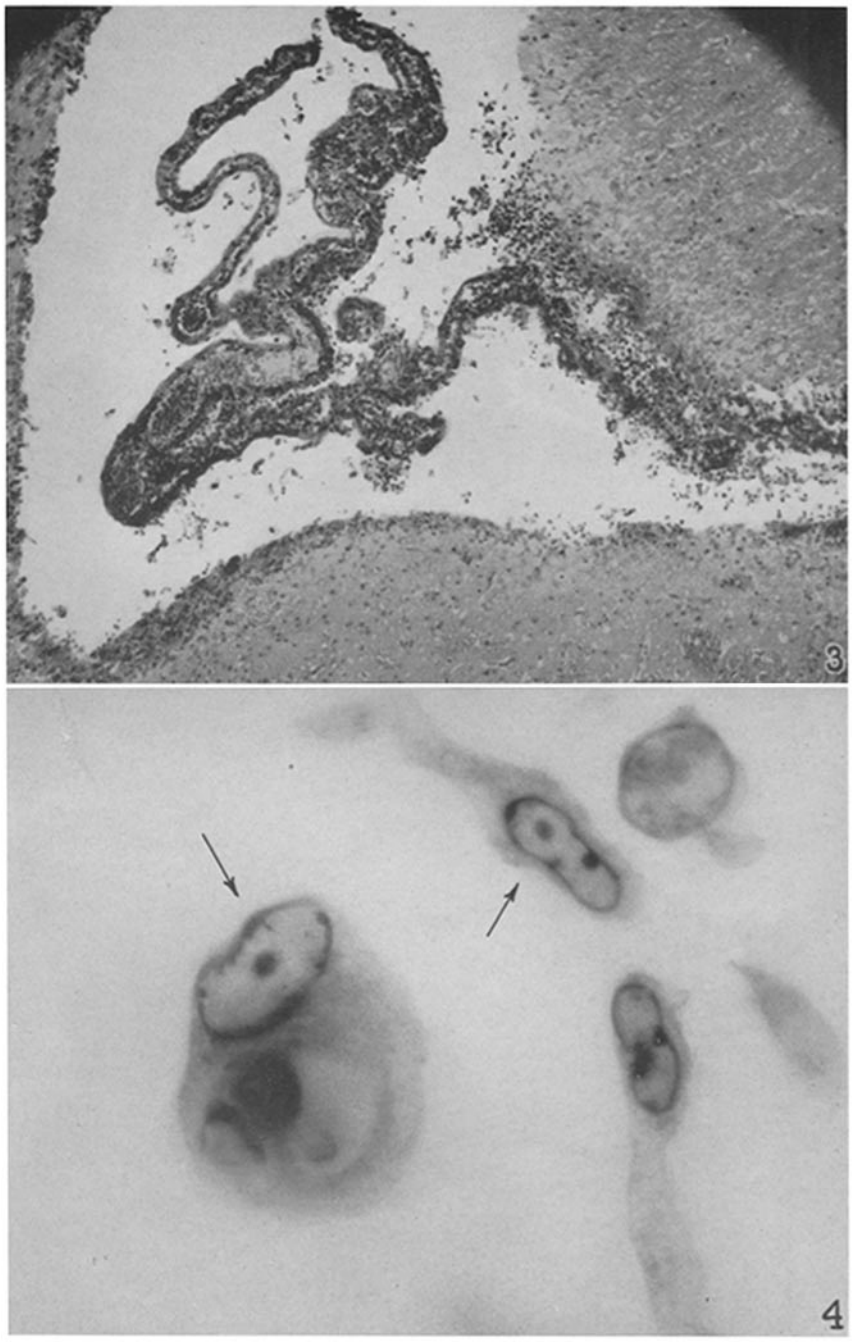
FIG. 4. Guinea pig, intracerebral inoculation. A meningeal cell and an adventitial cell of a small meningeal blood vessel bearing eosinophilic intranuclear bodies. Eosin and methylene blue. ×2310.





Photographed by J. A. Carlile

(Traub: Choriomeningitis virus in a mouse colony)



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