CONGENITAL INFECTIONS WITH REOVIRUS*

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PLATES 1 AND 2

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The problem of slow or chronic virus infections in which cells are not destroyed, but whole virus, or its components replicate in a surviving cell offers a new challenge to the understanding of the infective process. Such virus-cell interactions may exist with or without circulating serum antibody in the host. Reoviruses are not immediately cytocidal, have a prolonged growth cycle in cell culture, and multiply in association with the mitotic apparatus of the affected cell (1–3). The following experiments describe the results of prolonged reovirus infections initiated during intrauterine development.

Materials and Methods

Virus.—A strain of reovirus, type 2 (988) isolated in August of 1960 at the Boston City Hospital from a rectal swab of an 8-yr-old boy with a febrile exanthem was used throughout (4). This strain has been passaged in rhesus kidney tube cultures, and later in L cells. It has been characterized, concentrated, and partially purified (5–11). Ten thousand hemagglutinating units of reovirus 988 were inoculated into 32-oz monolayer bottles of L-48 cells which had been grown in Eagle's medium (12) with twice the minimal essential amounts of amino acids and 10% calf serum. At the time of inoculation a similar medium containing 5% fetal bovine serum which was free of homotypic hemagglutinating inhibiting antibodies (HIA) was exchanged for the growth medium. After a 72 hr incubation period at 37°C, cells from 10 such bottles were removed with a rubber policeman, centrifuged at 600 *g* (4°C) for 15 min, and the supernatant fluid discarded. The remaining pellet of cells was taken up in 10 ml of Earle's saline (BSS), and their cellular membranes disrupted in acetone and dry ice by three cycles of freezing and rapid thawing in warm (37°C) water. After 500 μg/ml of desoxycholate was added to this debris, it was agitated for 120 min in a 25 ml Erlenmeyer flask by means of a magnetic stirrer (4°C). Subsequently, the sediment was separated by centrifugation (600 *g*, 15 min, 4°C).

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The resultant supernatant fluid with partially purified and highly concentrated reovirus had a hemagglutination (HA) titer of 8.1×10^5 hemagglutinating units/ml. It contained approximately 2.4×10^9 TCD_{50}/ml.[1] This stock virus was kept in 5 ml screw capped vials at (−20°C) until used.

Mice, Experimental Infections, and Clinical Observations.—ICR albino Swiss mice were obtained from Roy Rawley Farms (Plymouth, Michigan). Sixty young adult females and the same number of mature male mice were caged together for 24 hr. On the 1st, 3rd, 6th, 10th, and 15th day after possible conception, 10 female mice were inoculated intraperitoneally with 0.1 ml of a dilution of the stock reovirus containing 20,000 HA units. All of the female mice (inoculated and 10 controls) and their offspring were observed daily for 2 months and then every several days. Except for the omission of the 10 control mice, the identical experiment was repeated several months later.

Virology.—After delivery baby mice were sacrificed when ill; at 3 wk, apparently well, congenitally inoculated and control mice were sacrificed by ether anesthesia and autopsied. All tissues, suspensions, and fluids were stored in 5 ml screw capped vials at (−20°C) until used. In certain mice showing eye signs, the entire globe with some adherent extraocular muscle was enucleated as a unit, and processed for virology. Tissues were minced, ground with alundum, and 20% suspensions were made in Eagle's medium. Isolations from tissue or blood and subsequent titrations of these reovirus strains were done in roller tube cultures of rhesus kidney (MKTC)[2] using Eagle's medium with twice the usual concentration of amino acids to which 5% fetal bovine serum and antibiotics (250 units of penicillin and 250 μg of streptomycin per ml) were added. Cell cultures (2 per specimen) were inoculated with 0.1 ml of these tissue suspensions and examined for cytopathic effects every other day for 2 wk. Medium was replenished at 5-day intervals. When typical cytopathic effects (6) were seen, isolates were examined for their ability to hemagglutinate freshly washed type O human red blood cells. Supernatant fluids from cultures not showing microscopic changes at 14 days were also examined for HA before being considered negative.

Virus titrations of tissue suspensions or blood were done using 3 MKTC tubes per dilution. End points were estimated by the 50% method (13).

Serology.—At 3 wk of age surviving congenitally infected mice and their noninfected controls from the first experiment were sacrificed. After 3 months 10 apparently well survivors from the second experiment were killed. After light anesthesia with ether, blood was collected by decapitation or after partial skinning and cutting deeply into the axilla, and collecting the blood that accumulated between the skin and thoracic cage. Serum from each mouse was tested for the presence of HIA to reovirus 988 as follows: 1.0 ml of a 1:10 dilution of serum in Earle's saline was added to 1.0 ml of a 25% suspension of kaolin in balanced salt solution, mixed thoroughly and incubated for 1 hr at 20°C. The kaolin was separated by centrifugation. The test system consisted of 0.2 ml of twofold dilutions of the kaolin washed serum, plus 0.1 ml of reovirus suspension (prepared to contain 16 HA units) plus 0.3 ml of a 0.4% suspension of freshly prepared human type 0 erythrocytes. The agglutinations were read by the pattern method after incubation for 75 min at 20°C. The HA titer of the test virus was determined before use, with virus, serum, and cell controls included routinely.

Pathology.—When mice were sacrificed for virologic study, tissues were immediately fixed in 10% formalin. Transverse sections of brain, lung, heart, liver, kidney, spleen, hind limb, and eye were cut and stained with hematoxylin and eosin. Control mice in a ratio of about 1 to 10 were similarly processed.

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[1] TCD_{50}, that dilution of virus which will cause cytopathic effects in half of the inoculated tissue culture tubes.

[2] Obtained from Microbiological Associates, Bethesda, Maryland.

RESULTS

Findings in Mice Congenitally Infected with Reovirus, Type 2 (988) Epidemiologic (Table I).—Of the 20 female mice inoculated with reoviruses during these two experiments on days 1, 3, 6, 10, and 15 of gestation,[3] 7, 4, 6, 6, and 6 mothers delivered. Two of 10 control mice delivered. The total number of newborn mice followed was 229 with an average litter size of 8 babies. Fourteen suckling mice were eaten and lost to follow-up.[4] Sixty-three mice (27.5%) showed signs of illness or died within the first 2 wk of life.

At 3 wk of age survivors from nine litters (67 mice) of the first experiment were sacrificed for serologic and selected pathologic and virologic study. Of the 114 mice in the second group, 54 (47.4%) showed signs of illness between days 15 and 36 after birth. None of these mice with late illnesses died.

Clinical (Table II).—Early illnesses consisted of lassitude, stunting of growth, and some roughening and loss of sheen of fur. The fur of affected mice did not appear oily, nor was jaundice observed. There were no paralysis, tremors, ataxia, or convulsions. The mode of exodus of the dying animals was not clinically apparent.

Other mice showed no manifestations of early illness, but demonstrated decreased spontaneous activity when compared to controls and a marked retardation of growth between the 15th and 36th days of life (Text-fig. 1). Between days 18 and 34, 3 mice with late illnesses showed signs of involvement of one or both eyes. These were: variable degrees of proptosis, cloudiness of the bulbar conjunctiva, and finally closing of the eyelids (Text-fig. 2). Animals with late illnesses apparently recovered completely.

Early and late illnesses occurred with about equal frequency regardless of the time during intrauterine development when reovirus infection was initiated. No controls showed any of these signs. Several months after their original intraperitoneal inoculation, a few of the brood mothers exhibited abdominal distention. Postmortem examinations demonstrated small bowel obstruction.

Virologic Findings.—Representative tissues from mice with early illnesses were processed for the presence of reovirus, type 2. Autopsies and virus studies were done on mice dying or others were sacrificed when ill as early as 2 hr after birth. About half of these mice died within the first day of life and most of the other deaths occurred within the first week. The lungs and kidneys contained highest titers of virus most frequently, but brain, myocardium, and liver contained virus in some of the mice when there was no viremia indicating primary multiplication in these organs as well (Table III). In addition, hind limbs from 2 of 13 animals studied contained reovirus, but these examinations were done in mice whose blood was virus positive.

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[3] The gestation period of the mouse is 20 to 21 days.

[4] These mice are not included in these data.

At 3 wk of age, 16 apparently healthy congenitally inoculated mice were examined. Again, there was widespread evidence of the presence of reovirus. Viremia was documented in 4 of them, 24 to 42 days after their intrauterine infections. Again, a number of the lungs, kidneys, hearts, livers, and hind limbs contained virus in mice which had no viremia indicating sustained reovirus rep-

TABLE I

Epidemiologic Findings in Mice After Congenital Inoculations With Reovirus, Type 2 (988)

	No. of Mice	Total in group	Per cent
Early illnesses* (day 1 to 14)‡	63	229	27.5
Late illnesses§ (day 15 to 36)	54	114	47.4
No illness to date§ (day 1 to 90)	35	114	30.7

* Fourteen baby mice were eaten by their mothers and are excluded from these data (includes both experiments).

‡ Day, postnatal day on which first signs of illness were observed.

§ Only second experiment included here. In first experiment all well animals were sacrificed at 21 days.

TABLE II

*Clinical Findings in Mice Congenitally Infected With Reoviruses**

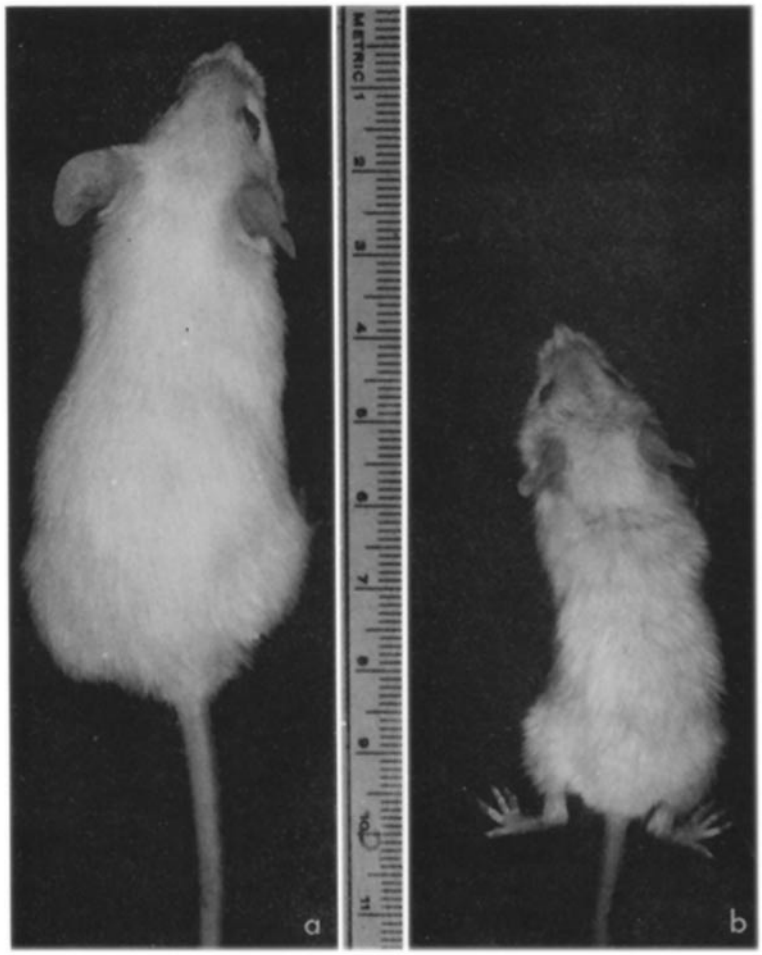
Day of gestation at virus inoculation	Total	No. of Mice		
		Early illness (day 1 to 14 after birth)	Late illness (day 15 to 36 after birth)	Conjunctivitis ± proptosis (day 18 to 34 after birth)
days				
1	34	13	21	1
3	15	11	4	0
6	20	9	11	1
10	14	7	7	0
15	34	23	11	1
Total.....	117	63	54	3

* Mice which were eaten by their mothers or were sacrificed when well at 3 wk are excluded from these data.

lication in these tissues (Table III mouse numbers 16, 17, 20, 21, 23, 27, 28 and 29). Therefore, virologic findings in both the early illnesses and in apparently well animals at 3 wk indicate that reovirus multiplied in many organs and for a protracted period. These general results were not altered by the times of congenital infection.

Two of the congenitally infected mice with conjunctivitis and exophthalmos were examined. Both of them showed clinical signs of late illness, beginning on

day 18 in one, and on day 28 in the second. Involvement of the eye appeared on days 18 and 34, and the animals were sacrificed at these times. In both mice, 10^3 TCD_{50} of reovirus were recovered from each affected eye. Although the



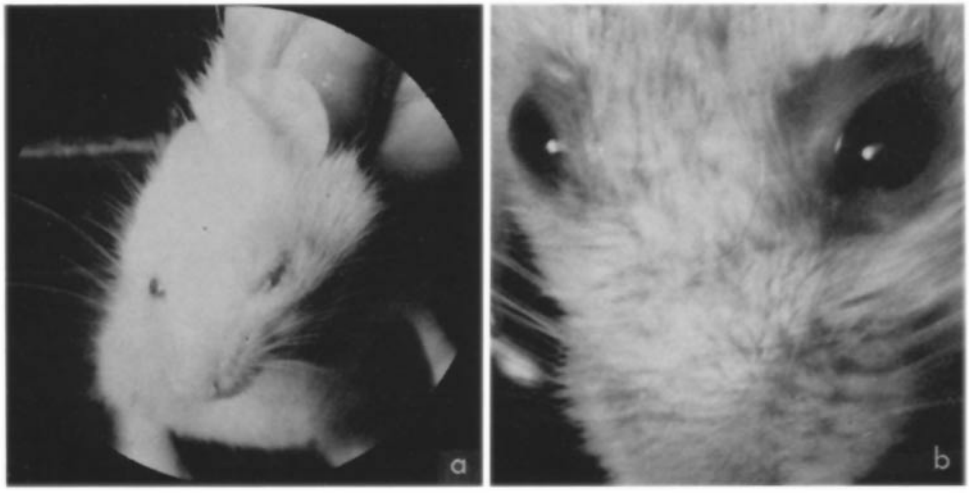
TEXT-FIG. 1. Late illness. This mouse was delivered from a brood mother inoculated with reovirus type 2 on the 15th day of gestation. At the time of this photograph the congenitally infected mouse (*b*), and its control (*a*) are 28 days old. The stunting of growth and alteration in fur of the affected mouse are obvious.

globe was processed as a unit, it is assumed that virus multiplication occurred in the subepidermal conjunctiva and extraocular muscle where abnormal histologic findings were noted. In one mouse other tissues were studied and here,

blood, skeletal muscle, kidney, liver, heart, brain, and lung contained no virus (Table IV).

Serologic Studies.—At 3 wk postpartum, sera from 9 brood mothers and their surviving offspring were examined individually for the presence of HIA to reovirus, type 2 (Text-fig. 3). Similar tests were done on 10 congenitally infected mice when they were 3 months old.

Antibody titers[5] in brood mothers 3 wk after delivery ranged from 12 to 192, with a mean of 65. At 3 wk suckling mice generally had HIA titers which were



TEXT-FIG. 2. Congenitally infected mouse with reovirus, type 2 (988) conjunctivitis (*a*) 34 days after birth, and its uninfected control (*b*).

somewhat lower than those of their mothers. At 3 months of age sera from 10 congenitally infected mice inoculated less than 5 days after conception showed HIA titers of less than 20 indicating that the antibody measured at 3 wk was maternal antibody which had reached the fetus via circulation of the yolk sac before delivery, or by absorption from their mother's milk (14). Other experiments in these mice show that actively formed antibodies are persistent.

Pathologic Findings (*Plate 1, Figs. 1 to 7*).—Marked histologic abnormalities among mice with early illnesses were seen in sections from lung and kidney, although there were changes as well in some hearts, hind limbs, and livers. The brains and spleens were unremarkable. Again, these morphologic findings (as those of virology and serology) did not differ according to the time of development when infection occurred.

The kidneys showed diffuse necrosis of proximal and distal convoluted tubules as well as collecting ducts. The most superficial zone of proximal convoluted tubules of the renal cortex

[5] Antibody titers are expressed as reciprocal numbers.

TABLE III

*Virology in Suckling Mice After Congenital Infections With Reovirus, Type 2**

Virus inoculation at day of gestation	Mouse No.	Reovirus, type 2 recovered from several tissues (TCD_{50}/g)‡							
		Blood	Brain	Lung	Myocardium	Liver	Spleen	Kidney	Skeletal muscle
Early illness									
1	1	Pos.§	Neg.	5.7	3.5	2.5	Neg.	Neg.	Neg.
	2	Pos.	3.5	4.3	Neg.	Neg.	Neg.	5.3	2.5
	3	Neg.	Neg.	2.5	3.5	4.7	Neg.	2.5	Neg.
3	4	Neg.	Neg.	4.3	2.5	Neg.	Neg.	3.3	Neg.
	5	Pos.	Neg.	4.3	Neg.	Neg.	Neg.	3.5	Neg.
	6	Neg.	Neg.	4.3	2.5	Neg.	Neg.	3.3	Neg.
	7	Pos.	Neg.	4.3	Neg.	Neg.	Neg.	3.5	Neg.
	8	Pos.	Neg.	3.3	Neg.	Neg.	Neg.	2.5	Neg.
15	9	Neg.	3.3	Neg.	Neg.	Neg.	Neg.	Neg.	Neg.
	10	Neg.	Neg.	2.5	Neg.	Neg.	Neg.	Neg.	Neg.
	11	Neg.	2.5	Neg.	2.5	Neg.	Neg.	4.17	Neg.
	12	Pos.	3.5	Neg.	Neg.	Neg.	Neg.	Neg.	Neg.
	13	Pos.	Neg.	Neg.	Neg.	2.5	Neg.	3.3	2.5
Mice apparently well (3 wk)									
1	14	Pos.	2.5	2.5	2.5	Neg.	2.5	3.3	2.5
	15	Pos.	Neg.	2.5	3.5	Neg.	2.5	Neg.	Neg.
	16	Neg.	Neg.	2.5	Neg.	2.5	Neg.	Neg.	2.7
	17	Neg.	Neg.	2.5	Neg.	2.5	Neg.	Neg.	3.7
3	18	N.D.‖	Neg.	3.5	Neg.	Neg.	Neg.	Neg.	3.5
	19	N.D.	Neg.	2.5	Neg.	2.5	Neg.	Neg.	2.7
	20	Neg.	Neg.	3.5	Neg.	Neg.	Neg.	3.5	Neg.
	21	Neg.	Neg.	4.3	Neg.	Neg.	Neg.	0	Neg.
6	22	Pos.	Neg.	Neg.	2.5	Neg.	2.5	Neg.	0
	23	Neg.	Neg.	3.3	Neg.	Neg.	Neg.	Neg.	2.5
10	24	Neg.	Neg.	Neg.	Neg.	Neg.	Neg.	Neg.	Neg.
	25	N.D.	Neg.	Neg.	Neg.	Neg.	Neg.	Neg.	Neg.
15	26	Pos.	Neg.	4.5	Neg.	Neg.	Neg.	4.3	2.5
	27	Neg.	4.17	4.17	2.5	Neg.	Neg.	4.17	Neg.
	28	Neg.	Neg.	Neg.	Neg.	Neg.	3.5	Neg.	2.5
	29	Neg.	2.5	3.5	Neg.	Neg.	Neg.	2.7	Neg.

* Only representative mice from each group were examined for the presence and amounts of reovirus in several tissues.

‡ Expressed as logarith to the base, 10.

§ Pos., positive for virus, but not titered.

‖ Not done.

was spared. Changes in epithelial cells of the affected renal tubules ranged from cloudy swelling to frank necrosis of the cytoplasm. In addition, lesser effects such as vacuolization and hyaline droplets in tubular cells were noted. Necrotic epithelial cells were sloughed into the

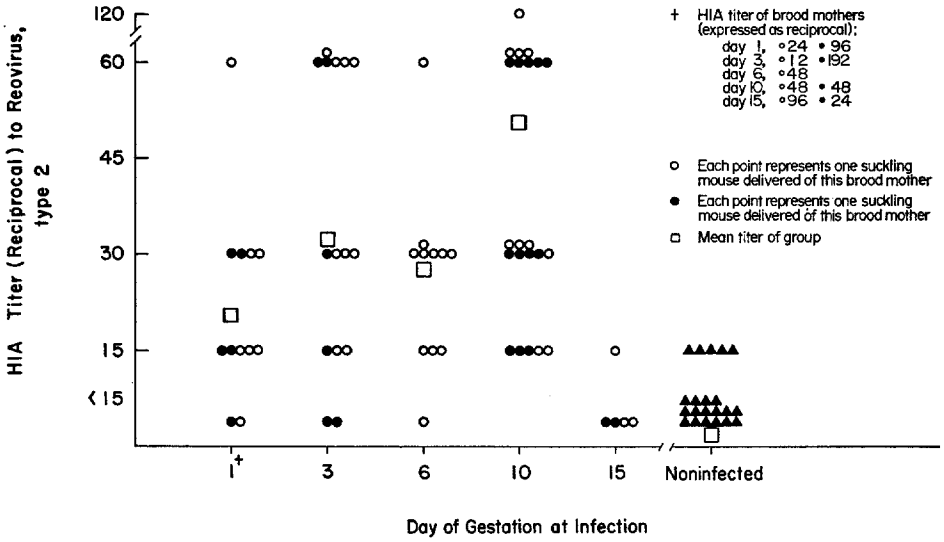
TABLE IV

Findings in Two Congenitally Infected Mice With Reovirus Conjunctivitis and Proptosis

Day of gestation at virus inoculation	Age of Mice (Days)			Virology (TCD_{50})/Eye
	Early Illness (1 to 14 days after birth)	Late Illness (15 to 36 days after birth)	Conjunctivitis ± proptosis (days after birth)	
1	Absent	28	34	10^{3}*
15	Absent	18	18	10^{3}‡

* Blood, skeletal muscle, kidney, liver, heart, brain, and lung did not contain reovirus, type 2.

‡ Virologic examination of other tissues not done in this mouse.



TEXT-FIG. 3. Hemagglutinating inhibiting antibody titers to reovirus, type 2, in 3-wk-old mice after intrauterine inoculations.

tubular lumens. Some necrotic tubules were markedly dilated exhibiting the lesions of "internal hydronephrosis" (Allen). There were no casts, nor was there any evidence of inflammation in the tubules or interstitium of the kidneys. No evidence of tubular regeneration was seen. Glomeruli and blood vessels were normal.

The lungs of these mice with early illnesses showed an interstitial pneumonia. Alveolar septae were thickened and contained an infiltrate of lymphocytes and plasma cells. Occasionally the visceral pleura contained a similar infiltrate and fibrin. The alveoli were clear,

without cellular infiltrate, serum, or fibrin. The epithelium of the alveolar septae, bronchi, and bronchioles was unaffected. There was no sign of secondary bacterial invasion.

Areas of interstitial mononuclear cell infiltrate were seen in sections from hind limbs. There were focal areas of muscle necrosis in the myocardium with sarcolemmal cell proliferation and mononuclear cells (lymphocytes and plasma cells). These were located in the interstitium between myocardial cells and surrounded vessels. Most livers showed striking extramedullary hematopoiesis.

Similar sections of brain, skeletal muscle, lung, liver, kidney, and spleen of 3-wk-old congenitally infected mice containing large quantities of virus showed no pathologic lesions.

Sections of the globe from mice showing reovirus conjunctivitis and proptosis revealed a lymphocytic infiltrate in the subepidermal tissue of the conjunctiva and in the extraocular muscles. The cellular reaction was focal in some areas and diffuse in others. There was no ulceration of the epithelium of the conjunctiva, and the lens, retina, choroid, and ciliary body were normal (Plate 2, Figs. 8 *a* and 8 *b*).

Tissue sections from other organs of mice older than 3 wk were completely normal. All sections of tissue from noninfected control animals were normal.

DISCUSSION

Reovirus infections are common in man, cattle, chimpanzees, monkeys, and mice. Serologic evidence indicates an even wider occurrence in the animal kingdom (4). In man, acute reovirus infections have been implicated in upper (15) and lower respiratory disease (16), rashes (4), and an encephalitic syndrome involving the liver (17, 18). Congenital reovirus infections in animals or man have not been previously described.

Reovirus infections in neonatal mice have been well studied (19–23). "Runting," jaundice, oily hair effect, and ataxia have been described. Hepatitis, pancreatitis, myocarditis, encephalitis, myositis, and pneumonia have been noted. Pneumonia and myocarditis are most severe after neonatal murine infections with reoviruses types 1 and 2, while encephalitis is more marked with reovirus, type 3 (23). However, lesions in the kidneys and those of the eyes observed here with congenital reovirus, type 2 infection have not heretofore been observed.

After intraperitoneal inoculation of pregnant mice with reovirus, type 2, a prolonged intrauterine infection of the developing fetus was produced. Like rubella in man and ferrets (24, 25) and cytomegaloviruses in man (24), reovirus multiplies during the intrauterine hiatus and clinical disease is evident after birth. Like cytomegaloviruses (24), but in contrast to rubella (17), developmental malformations have not been noted here. However, in contrast to rubella and cytomegaloviruses in man (24), but similar to infection in infant mice with lymphocytic choriomeningitis virus (LCM), specific hemagglutinating inhibiting antibodies are not produced.

The only sign of disease in the newborn mouse infected with lymphocytic choriomeningitis virus is a temporary runting with loss of hair, blepharitis,[6] and

[6] The blepharitis of neonatal LCM infection may be completely analogous to the conjunctivitis and proptosis of congenital reovirus infections. Virus studies of eyes in mice with neonatal LCM infection have apparently not been done.

a "jumpy" reactivity (26, 27). The mice had no detectable complement-fixing nor neutralizing antibody in their blood but were immune to challenge, and in spite of their normal appearance, carried high titers of LCM virus in all organs and in their blood.

Chronic reovirus infections in newborn mice have been associated with high and persistent antibody titers (23). In addition, congenitally infected mice inoculated early and late in gestation reported here had significant HIA titers at 3 wk of age. However, after intrauterine infection, by 3 months of age mice did not have serum antibodies suggesting that the immunoglobulins measured earlier were passively acquired and that immunologic tolerance may have occurred (14). Further experiments studying this phenomenon are in progress.

Congenital murine reovirus infections presented several clinical patterns. About a quarter of these rodents had illnesses within the first 2 wk of life which were often fatal. Stunted growth and roughening of fur was evident. Paralysis, ataxia, convulsions, and jaundice were not seen. Although congenitally infected mice had focal lymphocytic infiltrations in the myocardium and skeletal muscle, it is likely that death was either the result of respiratory or renal failure.

Interstitial pneumonia, renal tubular necrosis, and internal hydronephrosis of these early congenital illnesses were marked. The absence of cellular infiltrates in sections from the kidney in the presence of tissue destruction contrasts with other lesions seen here; namely in the lung, heart, and hind limb where there was an infiltrate of lymphocytes and plasma cells. Reovirus was easily demonstrated in all of these tissues, but highest titers appeared in the lungs and kidney where histologic lesions were most severe.

Another half of the congenitally infected mice (Table I) showed later illnesses (days 15 to 36) characterized by decreased activity, growth retardation, and occasional eye signs. In 3 of the 54 mice with late illnesses conjunctivitis and proptosis were noted. A mononuclear infiltrate associated with specific virus replication in the subepidermal conjunctiva and extraocular muscles was observed. In the 34-day-old-mouse with late illness and eye lesions, reovirus, type 2 was multiplying in the globe when the several other organs tested (liver, spleen, brain, heart, hind limb, and blood) did not contain reovirus. These latter organs were normal histologically. Since this mouse was inoculated 24 hr after fertilization, the total period of infection here was 53 days.[7] It appears that there is an especial and prolonged susceptibility of the eye.

Finally, a quarter of these mice remain well. At 3 wk of age virus was still present in high titer in the same organs that it had been found in the early illnesses, but in contrast to mice with early illnesses, these livers, hearts, kidneys, lungs, and limbs were normal histologically. In some of these mice viremia was documented fully 42 days after inoculation. Reoviruses like cytomegalovirus

[7] In neonatal murine reovirus infections, virus has not been found in any tissue after the 42nd postnatal day.

and rubella,[8] some bacteria (e.g. *Treponema pallidum*), and some parasites (e.g. *Toxoplasma gondii*, *Trepanosoma cruzi*) are able to produce congenital infections with consequences in the progeny months after the original infection. Whether such infections may possibly be tumorigenic remains to be determined. An initial isolation of a strain of reovirus, type 3 from a tumour syndrome affecting children in tropical Africa (Burkitt's lymphoma) (28, 29) suggests that further observations of these surviving mice which appear well should continue.

SUMMARY AND CONCLUSIONS

Congenital reovirus, type 2 infections were produced after intraperitoneal inoculations of brood mothers on the 1st, 3rd, 6th, 10th, and 15th day of gestation. The offspring presented with a varied syndrome. About a quarter of a total of over 200 mice showed symptoms within the first 14 days of life; namely, lassitude, retarded growth, and roughening of fur. Some died, apparently of respiratory or renal failure. Post mortem examination showed marked interstitial pneumonia and subcortical renal tubular necrosis. Reovirus was isolated in high titer from the kidney and lungs as well as from blood, hearts, hind limbs, and brains in lesser titer.

At 3 wk of age over 50 apparently well mice were sacrificed, and virologic, serologic, and pathologic study was done. High titers of virus were again found in the kidney, lung, blood, brain, heart, and skeletal muscle, but all tissues appeared normal histologically. Type-specific serum antibody titers in these mice were approximately those of their mothers.

Another half of these mice showed decreased spontaneous activity and growth retardation which appeared between the 15th and 36th days of life. Three of these mice with late illnesses had marked proptosis and conjunctivitis. A subepidermal conjunctival and extraocular muscle lymphocytic infiltrate was observed on section, and reovirus was isolated from these eyes in tissue culture. Again blood, brain, kidney, liver, spleen, myocardium, and skeletal muscle were studied, and were found to be normal histologically and not to contain reovirus. Finally, the rest of the mice remain well to date.

At 3 months of age, 10 of them were sacrificed. All had lost their maternal antibody and contained no reovirus, type 2 hemagglutinating inhibiting antibodies.

No developmental abnormalities were observed.

These data suggest that prolonged reovirus infections may be established by means of congenital inoculation of the developing fetus. Tolerant infection with immune paralysis seems to have been established.

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[8] Experiments similar to those reported here have been done by Dr. F. M. Wilson and one of us (AML). Strain 13 of Coxsackie A9 virus does not produce congenital infections in mice.

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EXPLANATION OF PLATES

PLATE 1

Representative pathologic sections from cross-sections of early illness in mice 1 to 14 days old with congenital reovirus, type 2 (988) infections.

FIG. 1. Normal lung. × 90.

FIG. 2. Interstitial pneumonia. Alveolar septae are thickened by an infiltrate of lymphocytes and plasma cells. The alveolar spaces are uninvolved. × 90.

FIG. 3. Focus of visceral pleuritis in reovirus pneumonia showing a mononuclear cell infiltrate with attached fibrin. × 135.

FIG. 4. Normal kidney. × 90.

FIG. 5. Kidney showing necrosis of proximal and distal convoluted tubules and collecting ducts. Proximal tubules of the outermost area of the renal cortex are normal. The glomeruli are also normal. × 90.

FIG. 6. Kidney with marked "internal hydronephrosis" and sloughing of necrotic epithelial cells into the lumens of the tubules. There are no casts, nor is there any evidence of inflammation. × 135.

FIG. 7. Skeletal muscle, showing an interstitial area with an infiltrate of lymphocytes and plasma cells. × 135.

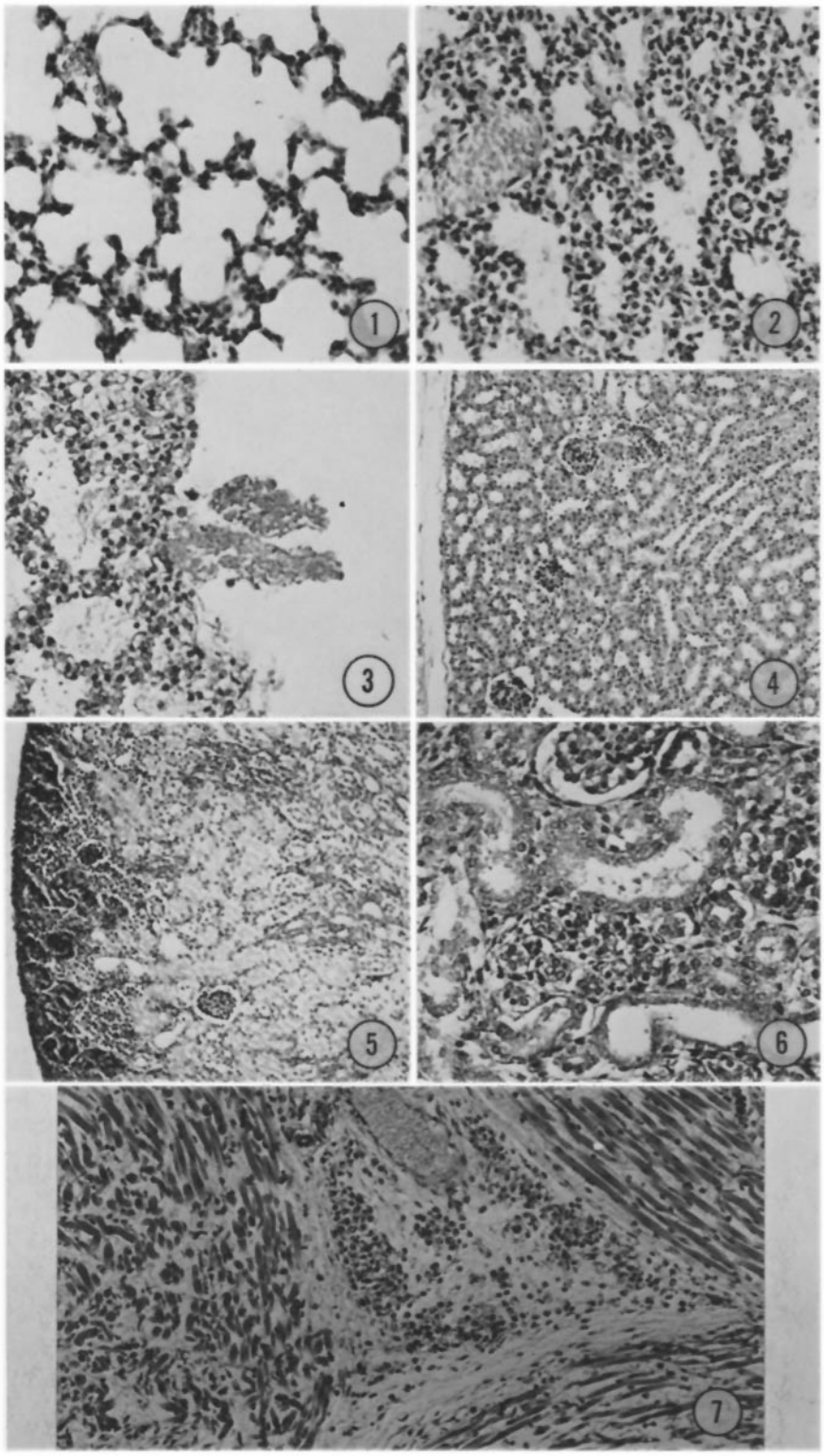

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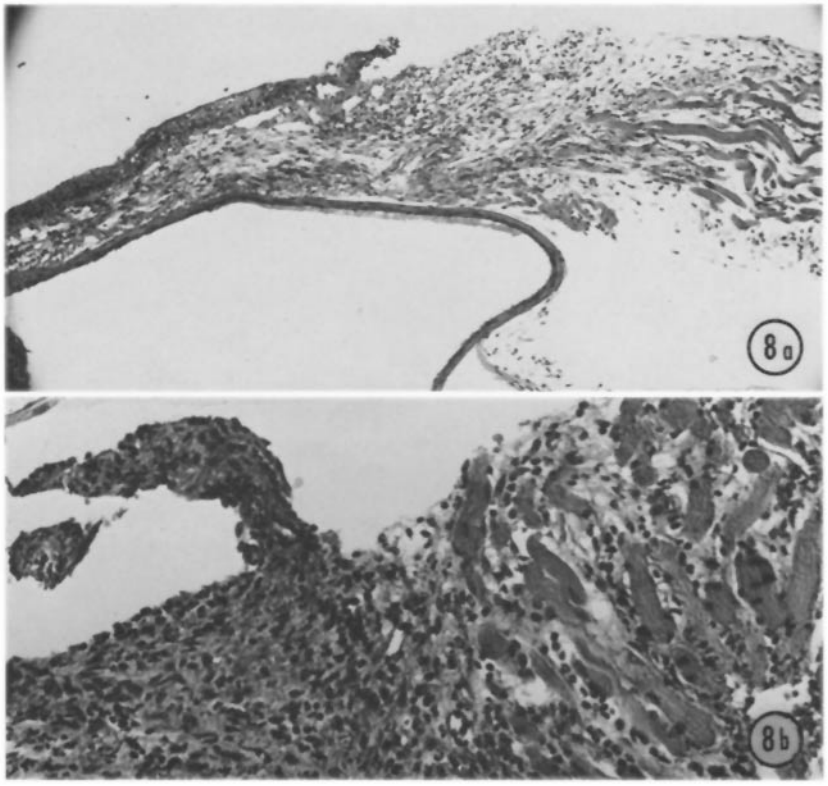
PLATE 2

FIGS. 8 *a* and 8 *b*. Subconjunctival (Fig. 8 *a*) and extraocular muscle (Fig. 8 *b*) lymphocytic infiltrates in a 15-day-old congenitally infected mouse with a late illness. × 225.



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