mydiae contained phage particles 50 nm in diameter in a crystal lattice array (Fig. 1C).

Organisms in digestive tubule inclusions of soft clams were ribosome-rich undulating rods measuring 300 by 2500 nm (Fig. 1D) characteristic of rickettsiae (*3*). Organisms in gut goblet cell inclusions in oysters were ribosome-rich, round to kidney bean–shaped bodies 250 to 350 nm in cross section and 400 to 1000 nm in length (Fig. 1E) characteristic of mycoplasmas (*4*).

Corroborative fluorescent antibody analyses remain to be carried out on fresh material. If verified, these findings may have far-reaching economic and public health significance. Serious diseases of humans and domestic animals are caused by organisms in all three of these groups, and all the species of bivalves found infected with these organisms are consumed raw. Psittacosis, an avian disease caused by a chlamydia that also infects humans, is known from numerous species of birds (*5, 6*), many of which are permanent or transitory residents of the Chesapeake Bay area. These observations raise the possibility that bivalves may be alternate hosts for zoonotic chlamydial, rickettsial, and mycoplasmal microorganisms. However, the discovery of a chlamydial phage suggests a potential mechanism by which pathogenic chlamydiae may be controlled.

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References and Notes

1. J. Storz, *Chlamydia and Chlamydia-Induced Diseases* (Thomas, Springfield, Ill., 1971).
2. L. A. Page, in *Bergey's Manual of Determinative Bacteriology*, R. E. Buchanan and N. E. Gibbons, Eds. (Williams & Wilkins, Baltimore, 1974), p. 914.
3. J. M. Moulder, in *ibid.*, p. 882.
4. E. A. Freundt, in *ibid.*, p. 929.
5. K. F. Meyer, in *Viral and Rickettsial Infections of Man*, F. L. Horsfall and J. Tamm, Eds. (Lippincott, Philadelphia, 1965), p. 1006.
6. ———, *Am. J. Ophthalmol.* **63**, 1225/199 (1967).
7. We thank G. Ward for collecting some of the hard clams used in this study, J. Hammed and L. Cullen for technical assistance, and P. Schellenger for clerical and editorial assistance. Supported in part by FDA contract 223-74-2129, NIH contract NO1-CB-33874, Department of Commerce contract NOAA 76051402, and the State of Maryland Fisheries Administration project 3-188-R.

22 November 1976

Latent Form of Scrapie Virus: A New Factor in Slow-Virus Disease

Abstract. *Scrapie is an unusual slow-virus disease of sheep which is very much like kuru and Creutzfeldt-Jakob disease, both fatal, slow neurological diseases of man. In mice, scrapie usually has an incubation period of about 6 months. Intraperitoneal inoculation of virus particles into newborn mice caused no disease, and there was no detectable virus replication for 1 year, but high titers of scrapie were present in the spleen and brain at 18 months. Virus replication occurred in mice injected from 4 days after birth by all inoculation routes, whether or not they were injected with scrapie virus on day 0. The results suggest that scrapie virus replicates peripherally only in thymocytes, which are not present in mice until a few days after birth. The latent state suggests that the comparable human diseases could appear in later life as a result of perinatal infection. In some respects these diseases resemble premature senility.*

Scrapie, an old and mysterious neurological disease of sheep, has received great attention as a classical example of slow-virus disease (*1*) and as a replicating agent of unusual smallness and stability that so far defies analysis or definition. It has been described as a subviral entity able to reproduce but possibly lacking the nucleic acid common to all known life forms (*2*), or as an agent comparable to a gene or provirus (*3*).

The importance of scrapie to man lies in the fact that it is the best known example of the spongiform encephalopathies, a group which includes two lethal diseases of the human brain, Creutzfeldt-Jakob disease and kuru, the "occupational" disease of the cannibals of New Guinea (*4*). The recent finding of cerebral amyloid deposits in murine scrapie suggests that there may be a virus cause for the similar amyloid plaques found in aged humans and in the premature senility of Alzheimer's disease and Down's syndrome (*5*). Thus the scrapie agent may offer clues to a cause of human aging.

Our results show that this agent can exist in a latent, undetectable form for many months and then return to its replicating infective state. This makes the provirus view more plausible. Newborn mice showed no disease or virus for about 1 year after they were injected intraperitoneally with scrapie virus, yet at 18 months their tissues contained vast numbers of infectious particles.

Albino Swiss mice of the Nya : NY-LAR (or Albany) strain were collected within 18 hours of birth, and three litters were inoculated intraperitoneally with 0.03 ml of brain tissue from mice infected with scrapie virus. The brain tissue was diluted 10^{-1} in tris-buffered 0.7 percent saline at *p*H 7.2. The inoculum contained $10^{6.1}$ LD_{50} (where LD_{50} is the dose that could kill 50 percent of the population) of scrapie virus (Compton strain) (*6*), determined by intracerebral mouse titration and the Reed and Muench end-point method (*7*). This large dose was used to encourage the possible induction of a state of immune tolerance to the scrapie agent in a manner analogous to that known to occur with lymphocytic choriomeningitis virus (*8*). Three other litters were similarly inoculated with ten times less virus ($10^{5.1}$ LD_{50}); sufficient additional litters were set aside for the same inoculation routine to be carried out on each of the following 5 days. A control group of three litters remained uninoculated. Three other litters that were inoculated with a 10^{-2} dilution of normal mouse brain in the same diluent remained well throughout the experiment (data not shown). In addition, 20 mice each weighing 8 to 10 g were inoculated intraperitoneally with each dose of scrapie virus as a positive control of mortality in adult animals. All male mice were removed from the experiment to eliminate the morbidity and mortality caused by fighting.

The results produced two main types of curves (Fig. 1). The control adult animals and older infants injected with scrapie virus showed a pattern of mortality beginning at 7 to 8 months, with a steep slope going up to 70 to 100 percent mortality by 10 months. The other curves showed a slow progressive increase in mortality until month 18.

Newborn animals responded to intraperitoneal injections of scrapie virus in a way similar to that of uninoculated controls: the expected clinical signs of scrapie and the expected mortality due to the disease were absent. In mice given high doses of virus on days 0 and 1 the signs of scrapie and mortality due to the disease were absent, whereas 18 animals injected on day 2 showed a 66 percent mortality by the tenth month, the incubation period relative to adults thus being extended 2 months. The animals injected

on days 3, 4, and 5 showed typical scrapie disease and death with a 4- to 6-week extension of the incubation period.

Although by 18 months none of the mice inoculated on days 0 or 1 developed definite signs of scrapie disease, they showed a somewhat faster mortality than the uninoculated animals. The deaths were sporadic and unexpected, resulting in carcasses too decayed or cannibalized for postmortem examination. This suggests the activity of another agent in the scrapie inoculum causing disease in infant mice. It may be relevant that others have found more than one agent, separable by various means, in stock scrapie preparations (*9*). The tenfold lower scrapie dose ($10^{5.1}$ LD_{50}) caused similar effects except that scrapie mortality was lower in the group inoculated on days 0, 2, and 3, and the mortality attributed to a contaminating agent in the inoculum was not seen.

It was clear that mice inoculated with scrapie virus in the first 48 hours after birth did not respond with clinical scrapie disease in the manner of adult mice. The effect depended on age and virus dose, and mice inoculated at 4 days of age responded in the adult fashion.

To test the possibility that neonatally inoculated but symptom-free mice had a form of neonatally induced immunological tolerance to the virus or had "inapparent disease," the groups of mice which had received $10^{6.1}$ LD_{50} of scrapie virus on days 0 and 1 were killed at 14 months. The spleens of three mice in each group were harvested, pooled, and ground to a 20 percent suspension in sterile diluent; the brains were similarly treated. The four suspensions were each further diluted 1 : 2, and 0.03 ml was inoculated intracerebrally into each of five mice weighing 8 to 10 g; the sterile diluent was similarly inoculated into five control mice. Within 6 months all of these mice, but no controls, had died of typical scrapie. This surprising result was confirmed by titrating the same brain and spleen suspensions of mice from the same group inoculated on day 0, which had been preserved frozen at −80°C. Tenfold dilutions were used through 10^{-10}. Eight months later the end point of the brain titer had reached 3×10^6 LD_{50}/g and the spleen 3×10^5 LD_{50}/g; the control mice were normal (Table 1, experiment 1).

By 16 months, mice which had been inoculated on days 0, 1, and 2 with scrapie virus included a few animals sick with diarrhea, twitching, and waddling gait. Two of these mice had been inoculated on day 0 (with $10^{6.1}$ LD_{50}); two on day 1 (with $10^{6.1}$ LD_{50}); and one on day 2 (with $10^{5.1}$ LD_{50}). Although the signs were not typically those of scrapie, the brains and spleens of these animals were tested by the above procedure of intracerebral inoculation into normal adult mice. Five control mice were inoculated with diluent. All mice except the controls had developed typical scrapie by 7 months after the inoculation. Three of these sick mice were killed and their brains examined histologically; all had the spongiform lesions typical of those produced by the stock of scrapie virus used. Intracerebral titration of the virus from the mice inoculated with $10^{6.1}$ LD_{50} confirmed high titers of virus at 16 months (Table 1, experiment 2). The experiment was terminated at 18 months with the conclusion that the newborn mice were not susceptible to intraperitoneal injections of scrapie virus.

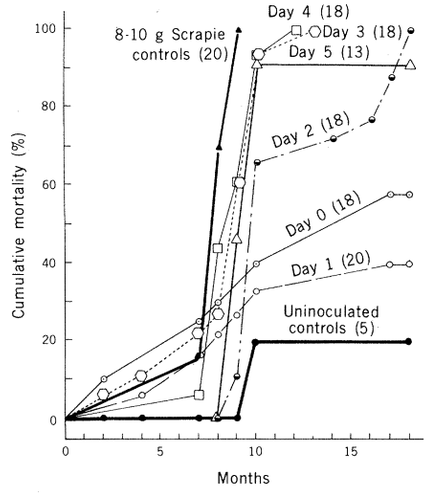

Fig. 1. Cumulative mortality of mice inoculated intraperitoneally with $10^{6.1}$ LD_{50} of scrapie virus on different days during the first 5 days after birth. Scrapie control mice, each weighing 8 to 10 g, were inoculated with the same dose to show the mortality of adult animals. Numbers in parentheses give the number of female animals alive in each group 1 week after inoculation. This interval eliminates the few (one litter) animals neglected and eaten by one mother, and one death at the time of inoculation.

Before it was realized that the neonatally inoculated mice would ultimately produce virus, additional experiments were performed to determine whether such mice were resistant to intracerebral challenge with scrapie virus and whether the virus in neonatal mice was replicating at the same rate as in adult mice. Three groups of two litters each of newborn (less than 24 hours old) mice were used; ten mice were followed for 18 months, the rest were used for other experiments. Groups 1 and 2 were inoculated intraperitoneally with $10^{6.1}$ LD_{50} of scrapie virus; group 3 received the same dilution of normal mouse brain. Two weeks later groups 1 and 3 were inoculated intraperitoneally with the same dose of scrapie, group 3 providing a control of the effect of the scrapie inoculation on 2-week-old mice. After 6 months the mice in groups 1 and 3 began to show signs of scrapie, and by 10 months all were dead from this disease. The mice in group 2 continued to be well through 17 months,

Table 1. Detailed data from scrapie experiments 1 to 3. Except where otherwise indicated, each mouse was inoculated with $10^{6.1}$ LD_{50} of scrapie virus. All inoculations were by the intraperitoneal (IP) or intracerebral (IC) route. The dose used for challenge was also $10^{6.1}$ LD_{50}.

Inoculation			Incubation period (months)	Time to 100 percent death or severe illness (months)	Age when killed (months)	Scrapie titer (LD_{50}/g)	
Age of mouse	Route	Mice tested (No.)				Spleen	Brain
			Experiment 1				
0 day	IP	3			14	3×10^5	3×10^6
			Experiment 2				
0 day	IP	2	? > 16		16	2×10^5	5×10^6
1 day	IP	2			16	*	*
2 days	IP†	1			16	*	*
			Experiment 3				
Group 1							
0 day and 2 weeks‡	IP	10	6	10			
Group 2							
0 day	IP	10	> 17		12 or 9§	0, 0	0
Group 3							
2 weeks‖	IP	10	6	10			

*Virus present, not titrated. †The dose was $10^{5.1}$ ID_{50}. ‡Each mouse was given a second IP inoculation of $10^{6.1}$ LD_{50} at 2 weeks of age. §One mouse was killed at 12 months, three at 9 months. ‖Each mouse at day 0 was given an IP injection of normal mouse brain suspension.

Table 2. Detailed data from scrapie experiments 4 to 6. Each mouse was inoculated with $10^{6.1}$ LD_{50} scrapie virus. All inoculations were by the intraperitoneal (IP) or intracerebral (IC) route. The dose used for challenge was also $10^{6.1}$ LD_{50}.

Inoculation					Challenge		
Age of mouse	Route	Mice tested (No.)	Incubation period (months)	Time to 100 percent death or severe illness (months)	Time from inoculation to challenge (months)	Incubation period (time from challenge to scrapie onset) (months)	Mortality
				Experiment 4			
0 day	IP	10	> 17		9	5	5 out of 5 at 7 months
9 months	IC	5	4	5			
6 weeks	IC	5	6	8			
				Experiment 5			
0 day	IC	16	4	7			
				Experiment 6			
0 day	IP	5			½	7	4 out of 4 at 10 months
0 day	IP	5			6	4 to 5	5 out of 5 at 7 months
0 day	IP	5			9	4 to 5	3 out of 3 at 7 months

with the exception of one which died from an unknown cause (Table 1, experiment 3).

One mouse from group 2 was used at 1 year after injection for brain pathology and spleen titration. The brain was indistinguishable from that of a normal, uninoculated mouse of the same age and strain showing very low levels of vacuolation (about 3 percent of the vacuolation found in mice with terminal scrapie). The mice used for titrating the virus in the spleen remained normal for 10 months. Three other mice from group 2 were killed at 9 months, and their brains, spleens, and blood were subjected to intracerebral titration for scrapie virus. All the animals used for these titrations remained normal through 10 months. Thus in mice that had been inoculated with scrapie when newborn, there was no evidence of scrapie virus for 9 to 12 months (Table 1, experiment 3).

Five of the group 2 survivors were used for intracerebral challenge with $10^{6.1}$ LD_{50} of scrapie virus 9 months after the neonatal inoculation. Five adult mice (9 months old) and five young adults (6 weeks old; 8 to 10 g) that had not received a previous inoculation were similarly inoculated. The group 2 survivors showed symptoms of scrapie 5 months later and were all dead by 7 months after the challenge injection (16 months after the neonatal injection). The 9-month-old controls showed scrapie symptoms at 4 months after the challenge injection and all were dead by 5 months, both end points occurring earlier than with the mice that had received neonatal inoculations of scrapie virus. The 6-week-old mice showed no symptoms until 6 months after the challenge and were dead by the eighth month (Table 2, experiment 4). The longer incubation time for 6-week-old relative to 9-month-old mice is usual for this combination of mouse strain and virus strain. There is some suggestion, however, that the neonatal scrapie inoculation caused a slight prolongation of the incubation period after the intracerebral challenge.

All the neonatal mice that were inoculated with scrapie virus in the above experiment received the virus intraperitoneally. When the experiment was repeated with the neonatal mice receiving intracerebral inoculations, all the mice succumbed to clinical scrapie during the fourth month and were dead by 7 months after the inoculation (Table 2, experiment 5). Thus newborn mice appear to be unresistant to scrapie virus when it is inoculated intracerebrally. Additional tests showed that neonatal mice inoculated intraperitoneally had no scrapie virus in the brain or spleen at 11, 21, or 42 days, or at 3 months. In control mice inoculated intraperitoneally at 10 days with the same dose of virus and tested at the same intervals, scrapie first appeared in the spleen at 42 days and thereafter rapidly increased in titer.

These results showed that mice inoculated intraperitoneally within 24 hours of birth with high doses of scrapie virus contain no detectable virus in their brains or spleens for at least 9 months. Yet they contain virus at 14 months with brain and spleen titers of 3×10^6 and 3×10^5, respectively. When injected into neonatal mice, the virus disappears and remains in a latent form, which after about 1 year begins to replicate in the normal way. It is unclear at present whether the virus causes clinical scrapie in the mice after 14 months, but this outcome appears likely, and a few mice have shown possible early signs of the disease.

Mice inoculated intraperitoneally when newborn were challenged intracerebrally with 10^6 LD_{50} scrapie virus at 2 weeks or at 6 or 9 months. All died of scrapie, with incubation periods of about 7 months following the 2-week challenge and 4 to 5 months following the 6- or 9-month challenge; the mortality was 100 percent at about 10 and 7 months, respectively (Table 2, experiment 6). The incubation period following intracerebral challenge apparently becomes shorter with increasing age at the time of challenge. Thus the latent scrapie infection confers no obvious protection to the brain against scrapie. It is important to stress that control, uninoculated mice were kept in the same room, and sometimes in the same cages, as the infected animals. All mice were tested for scrapie virus after 6 months to 2 years, and there was not one incidence of spontaneous scrapie.

The short neonatal period of insusceptibility to scrapie is similar to the period of tolerance to lymphocytic choriomeningitis (LCM) induced in newborn mice by inoculation of the LCM virus (*10*). However, unlike mice inoculated with LCM, the scrapie-inoculated mice do not carry the virus in a detectable form, and after 4 or 5 days they become fully susceptible to challenge by all routes. A possible explanation of these results could be the need for T cells as a replication site for extraneurally inoculated scrapie virus, since this time period coincides with the acquisition by the infant mouse of a functional cellular immune system consequent upon a maturation of the T cell system (*11*). Scrapie virus is known to replicate most rapidly in the spleen, and splenectomy, before or after scrapie infection, causes prolongation of the incubation period after intraperitoneal but not after intracerebral inoculation (*12*). Immunosuppression by arachis oil or prednisone acetate close to the time of intraperitoneal inoculation of scrapie virus similarly prolongs the incubation period (*13*). Outram *et al.* (*14*) observed pro-

longed incubation periods in mice that had been inoculated intraperitoneally with scrapie when they were born, and we also thought that a developing spleen cell might be needed for replication of the agent.

The hypothesis that after intraperitoneal inoculation the primary site of scrapie replication is strictly limited to the T cells could be readily tested within a year or so by following scrapie replication in T cell–deficient mice, such as the nude strain, both with and without reconstitution of the T cell system. Alternatively, the scrapie permissive spleen cell may be a type other than the T cell, which is susceptible to the same maturation stimulus. In another species, the rhesus monkey, the scrapielike virus of transmissible mink encephalopathy has been found in the brain of a clinically normal animal almost 3 years after it had received oral, intramuscular, and intravenous injections of the virus (*15*). Athymic (nude) mice injected intraperitoneally with hamster-adapted "Chandler" scrapie agent have been found by Marsh (*15*) to exhibit scrapie virus in the spleen for 26 days after injection, the titers being comparable to similarly inoculated normal mice at that time. In any event, careful studies of the existence of this phenomenon in other strains of mice, including the nude, are clearly indicated.

The absence of detectable virus in the animals inoculated at birth, coupled with the normal susceptibility of these animals to challenge, shows that the scrapie virus is present in a nonreplicating latent state for about 1 year. Thereafter, in response to an unknown stimulus, the latent agent becomes active, and detectable scrapie replication begins and appears to follow its usual course to the production of clinical disease. We suggest that latency occurs in the absence of a suitable T cell population when infection is limited to an alternative cell type. During the prolonged latent period, very slow replication occurs until, eventually, there are sufficient virus particles available for the random infection of susceptible T cells. The rate of replication then increases and leads to the neurological disease. This might explain the observed cases of "spontaneous" scrapie in supposedly normal animals (*16*).

These results have powerful implications for man, since scrapie pathogenesis is so similar to that of the human spongiform encephalopathies (SE), including Creutzfeldt-Jakob disease and kuru (*4*). Our results suggest that appropriately timed neonatal or prenatal infection of host species with some strains of SE virus would induce the latent state, with some incubation periods extending as far as old age. In this respect, the parallels which have been drawn between scrapie, Creutzfeldt-Jakob disease, and senility (*5*) imply that variants of the SE group could well cause some of the effects usually regarded as genetically induced old age. If vertical transmission of the agent can occur, as is believed to happen with scrapie in sheep, it would be hard to distinguish the latent SE virus from a gene causing old age. In this view, the earlier conclusion of Parry and others (*17*) that scrapie in sheep is both a hereditary and a transmissible disease seems more conceivable and also may apply to the human situation.

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References and Notes

1. J. Hotchin, in *Progress in Medical Virology*, J. L. Melnick, Ed. (Karger, Basel, 1958–), vol. 18, *Slow Virus Diseases*, J. Hotchin, Ed. (1974), p. 81.
2. _____, in *Monographs in Virology*, J. L. Melnick, Ed. (Karger, Basel, 1968–), vol. 3, *Persistent and Slow Virus Infections* (1971).
3. H. B. Parry, *Heredity* **17**, 75 (1962).
4. D. C. Gajdusek, *Ann. Clin. Res.* **5**, 254 (1973); C. J. Gibbs and D. C. Gajdusek, *J. Clin. Pathol. Suppl.* **6**, 84 (1972); E. Beck, P. M. Daniel, W. B. Mathews, D. L. Stevens, M. P. Alpers, D. M. Asher, D. C. Gajdusek, C. J. Gibbs, *Brain* **92**, 699 (1969).
5. M. E. Bruce and H. Fraser, *Neuropathol. Appl. Neurobiol.* **1**, 189 (1975); A. G. Dickinson, H. Fraser, G. W. Outram, *Nature (London)* **256**, 732 (1975).
6. Kindly supplied by J. Gibbs, National Institute of Neurological Disease and Blindness.
7. L. J. Reed and H. Muench, *Am. J. Hyg.* **27**, 493 (1938).
8. J. Hotchin, *Ann. N.Y. Acad. Sci.* **181**, 159 (1971).
9. P. C. Licursi, P. A. Merz, G. S. Merz, R. I. Carp, *Infect. Immun.* **6**, 370 (1972); R. I. Carp, P. A. Merz, P. C. Licursi, G. S. Merz, *J. Infect. Dis.* **128**, 256 (1973); A. G. Dickinson, D. M. Taylor, H. Fraser, *Nature (London)* **248**, 510 (1974).
10. J. Hotchin, *Cold Spring Harbor Symp. Quant. Biol.* **27**, 479 (1962).
11. M. F. Greares, J. J. T. Owen, M. C. Raff, *T and B Lymphocytes: Origins, Properties and Roles in Immune Response* (American Elsevier, New York, 1973).
12. M. C. Clarke and D. A. Haig, *Res. Vet. Sci.* **12**, 195 (1971); H. Fraser and A. G. Dickinson, *Nature (London)* **226**, 462 (1970).
13. G. W. Outram, A. G. Dickinson, H. Fraser, *ibid.* **249**, 855 (1974); *Lancet* **1975-I**, 198 (1975).
14. _____, *Nature (London)* **241**, 536 (1973).
15. R. F. Marsh, D. Burger, R. Eckroade, G. M. Zu Rhein, R. P. Hanson, *J. Infect. Dis.* **120**, 713 (1969); R. F. Marsh, personal communication.
16. I. H. Pattison and K. M. Jones, *Nature (London)* **218**, 102 (1968); J. M. K. Mackay, *ibid.* **219**, 182 (1968); A. G. Dickinson, *ibid.* **252**, 179 (1974).
17. H. B. Parry, *ibid.* **185**, 441 (1960); *Heredity* **17**, 75 (1962); A. G. Dickinson, J. T. Stamp, C. C. Renwick, *J. Comp. Pathol.* **84**, 19 (1974).

10 June 1976; revised 13 October 1976

Virus-Induced Behavioral Alteration of Mice

Abstract. *Neonatal mice were inoculated intracerebrally with lymphocytic choriomeningitis virus (LCM). These mice developed long-term persistent tolerant infections and when tested at 3.5 to 6.0 months of age they showed significant increases in behavioral latency when subjected to open-field tests, and significant decreases in the current level required to elicit a startle response and in locomotor activity in a running wheel. Comparable results were obtained with mice in which persistent infection was induced at 8 weeks of age and which were tested at 3.5 to 6.0 months of age. It was concluded that mice infected with LCM at birth or as adults exhibited long-lasting behavioral abnormalities.*

That virus infections of the nervous system can cause severe behavioral abnormality is well established. The ability of the rabies virus to induce pathological rage, and the more recent knowledge of the mistaken diagnoses of psychosis in some cases of human slow-virus encephalitis (*1*) have strengthened the concept that chronic virus infection of the brain may cause severe behavioral disorders. We have studied long-term persistent tolerant infection in mice with lymphocytic choriomeningitis (LCM) virus, using mice inoculated with LCM virus either at birth or at 8 weeks of age. Mice injected at birth demonstrate immunological tolerance of the virus in spite of high titers in all organs (*2*); those inoculated later exhibit viremia and are similar in most ways (*3*) to those infected at birth. Preliminary behavioral tests with the mice inoculated at birth (neonatal model) (*4*) suggested that mice persistently infected with LCM virus exhibit subtle behavioral differences when compared with uninfected controls. The results described here confirm and extend the early work and appear to establish a clear role for virus infection in the origin of some behavioral abnormalities.

The mice used were from a colony of the outbred Nya : NYLAR albino strain maintained at this laboratory. All animals used for the neonatal model received within 24 hours of birth an intracerebral inoculation (0.03 ml) of a suspension of brain tissue infected with the LM_4 (*5*) strain of LCM (0.03 ml containing 10^5 ID_{50}, where ID_{50} is the infective dose for 50 percent of the population); controls received 0.03 ml of a comparable dilution of normal mouse