INTRODUCTION

Scrapie is a fatal progressive degenerative disorder of the central nervous system that occurs as a natural infection in sheep and goats. It is transmissible experimentally to various species, including mice, but the disease has not been produced in all species that have been tested, such as, rabbits and guinea pigs. There are many strains of the agent that causes scrapie, and their molecular structure is probably outside the range for conventional viruses. Consequently, they display very high resistance to inactivation by a wide range of physical and chemical treatments. Although scrapie agents remain infectious after treatment with very large doses of 254 nm UV irradiation, this finding does not necessarily exclude nucleic acids as the informational molecule in scrapie, as many have assumed. It is possible that nucleic acids can be protected chemically, or repaired, in ways not yet known. Because of these uncertainties many workers have preferred to use the operational term “agent” rather than “virus.”

The many unusual properties have prompted various hypotheses about the nature of these agents, though none of them can now accommodate all the findings. An unfortunate corollary has been the uncritical acceptance of various unlikely “findings” that would quickly be recognized as experimental errors, if conventional microorganisms were involved. The only agreed remnant of these hypotheses is that most of the infectivity, detectable by present methods in brain homogenates from mice with advanced disease, accompanies cell membranes, and it is assumed that this association is present in the living animal. It is unknown whether this applies to most of the agent earlier in incubation or in tissues other than brain. Neither the form of this association with membranes nor its significance, if any, for agent replication or protection is known.

Infectivity assays in whole animals are the only available tests for the presence of the agent: There are no immunologic, electron microscopic, or tissue culture findings on which to base in vitro tests. Many attempts have
been made to grow tissues (usually brain) from affected animals in which the agent could be shown to be replicating, but there has only been satisfactory evidence of success in one case (2).

**AGENT REPLICATION**

**Agent Replication during Incubation in Mice**

The importance of the lymphoreticular system (LRS) in the early pathogenesis of scrapie first became apparent from a time-sequence study involving 13 organs (14). The same general pattern appears to be followed by various agent strains, at least in the most widely used mouse strains, but the different host/agent combinations vary widely in the absolute time intervals involved.

The sequence of events follows. The earliest rise in titer occurs in organs of the LRS, such as the spleen, after either intracerebral (i.c.) or extraneural injection. Agent titer rises at a later stage in other tissues, such as the lungs, the intestines, and the uterus, but only later, if the extraneural injection in the spinal cord and brain led to death after a relatively short clinical course. It is unknown how the agent is transported from the site of injection to the LRS and, with peripheral injections, eventually from the LRS to the brain. No convincing "viremic" phase has been detected, with the exception of the brief one occurring immediately after i.c. or intravenous injection. Presently, it is reasonable to assume that rise of titer in an organ indicates that replication is occurring there. In the case of an i.c. injection, the extraneural events seem to be irrelevant to the course of the disease because the agent that remains in the brain starts to replicate there much sooner than it would have after extraneural injection, though still not as soon as it does in the spleen. It is possible that the agent that has received some types of treatment (e.g., heating) (7, 13) may not do so, but with ordinary inocula, there are several reasons for concluding that the agent replicates in the brain after an i.c. injection: Incubation is shortest after i.c. injection, even when sterile i.c. injection-trauma accompany intraperitoneal (i.p.) injection of the agent; splenectomy (or genetic asplenia) has no effect after an i.c. injection but increases incubation after an i.p. injection (8, 18); the effective titer of an inoculum is higher by the i.c. than the i.p. route (Fig. 1.1); neonatal and young mice are easier to infect by the i.c. than the i.p. route (27); the suppression of susceptibility with large doses of steroids does not apply to i.c. injections.

Replication in the spleen can occur fairly quickly. It is possible that there is a short delay of a day or so before replication commences in the spleen in the quicker host/agent combinations (e.g., ME7 agent in C57BL mice, with 160-day incubation period after an i.c. injection of $10^5$ LD$_{50}$ units). But this then proceeds with a doubling time of about 2 days, and the process is complete 4 to 5 weeks later. Afterward, there is a titer plateau phase for the remaining 17 weeks before death. The amount of agent dur-