35R0029

APPLICATION FOR APPROVAL OF THE USE OF ANIMALS IN RESEARCH AND TEACHING
Principal Investigator: [redacted]
Date: 4/3/97

Page 3, supplement

NOT A FOIA DELETION

VIII. c. Animals will be used in a project involving pain or distress without administration of appropriate anesthetic, analgesic or tranquilizer drugs. (Attach a statement justifying why such drugs will not be used.)

The purpose of the proposed study is to determine the mechanism of latency of herpes simplex virus. The virus is a neurotrophic virus which establishes latency in the sensory nerves (in this case, the trigeminal ganglia) of the infected dermatome, and DNA persists at the site of infection in the dermatome (in this case, the cornea). Treatment or physical manipulation of these tissues may induce reactivation and thus disturb the latent state.

IX. Write a brief description of the procedures to be utilized on the animals in your research project. Indicate any physical or physiological impairment or loss of function occurring to the animal from your procedure, i.e., diabetes, blindness, loss of motor abilities. Use additional paper if necessary and attach the methods and vertebrate animal portions from your grant proposal.

Rabbits will be given primary herpetic infections by the injection of 10^5 pfu of HSV-1 at the midline of the alveolar mucosa. About 30 days later the corneas of rabbits will be infected with HSV-1 by intrastromal injection of 1000 plaque forming units of virus in 20 microliters or an equivalent dilution of mock antigen. Prior to injection the rabbits will be anesthetized with 25 mg/kg ketamine and 5 mg/kg xylazine and 2 drops of 0.5% proparacaine per eye. Herpetic keratitis will be allowed to develop. Thus the animals will experience reduced vision for about 15 days. The keratitis will spontaneously resolve and latency will have been established. Any animals that become blinded will be sacrificed immediately. Our past studies tell us that it will be less than 1% with this virus. Other strains of virus produce blinding complications at much higher frequency. At varying time intervals between 40 and 120 days post infection, rabbits will be sacrificed using euthanasia solution (Beuthanasia-D). Immediately following death, tissues (corneal and trigeminal ganglia) will be taken for laboratory analysis.

Justification for 10 rabbits in Column E

Registration No.: 35-R-0029

35R0029

CONT.

In these studies, the corneas of rabbits will be infected withthe McKrae strain of herpes simplex virus type 1. Herpetic keratitis will be allowed to develop for 7 days prior toinitiating therapy. During this time the rabbits will experience some pain and discomfort. The level of discomfort the animals experience is hard to quantify but they do not rub their eyes excessively, suggesting the discomfort is not severe.

It is necessary to allow the disease to develop untreated in order to mimic the human situation. Patients do not come for therapy until they become symptomatic. By this time, the immune/inflammatory response has been triggered and latent infection of the sensory nerves that serve the infected deratome have been established.

The use of topical or systemic analgesics is not a viable option in these studies becasue HSV is a neurotrophic virus. Any treatment that includes drugs that alter nerve function could affect the outcome of experiments designed to evaluate antiviral/ anti-inflammatory therapy. Similarly, studies of viral latency could be dramatically influenced by neuroactive drugs. Initiation of antiviral therapy on the day of infection or within a day or two of infection would prevent the disease from developing, thus negating the model.

pw [redacted] 4/12/99 [redacted]

(b)(6), (b)(7)(C)