Table 1. Relationship between *in vitro* Sensitivity of Poliovirus to Guanidine and its Neurovirulence for Monkeys

Viral strain	Guanidine HCl μg/ml. of culture medium		Neurovirulence for the monkeys: c.p.u. injected intramuscularly	No. monkeys with paralysis/ No. monkeys injected	Range of onset of paralysis (days after injection)
	0	1,000			
	c.p.u. detected after 5 days at 37° C*				
CVM†	3×10^7	10^2	10^8	5/5	4–11
			10^6	2/2	7–8
CVM 70 *G*‡	$2{\cdot}5 \times 10^2$	$7{\cdot}5 \times 10^6$	10^8	0·12	—
CVM 70 *G*–20§	$1{\cdot}2 \times 10^7$	$2{\cdot}5 \times 10^5$	10^8	6/6	5–10

* End point method.
† *CVM* = Virulent Mahoney isolated from the spinal cord of a monkey paralysed after the injection of Mahoney polio I and cloned twice.
‡ *CVM* 70 *G* = *CVM* propagated 70 times in human amnion cells (Mascoli-line) in the presence of the following guanidine HCl concentrations (μg/ml.): once, 16; once, 64; twice, 250; twice, 500; twice, 1,000; and 62 times, 2,000.
§ *CVM* 70 *G*–20 = *CVM* 70 *G* propagated 20 times in human amnion cells (Mascoli-line) in the presence of the following guanidine HCl concentrations (μg/ml.): twice, 1,000; twice, 250; once, 125; once, 64; once, 16; and 13 times without guanidine.

Table 1 compares the sensitivity to guanidine *in vitro* with the relative neurovirulence of the different strains used. It appears that the regression of guanidine-dependence *in vitro* is accompanied by a return of the neurovirulence *in vivo*. It is interesting to notice that guanidine-dependent poliovirus retains the cytopathogenic activity *in vitro* provided guanidine is present in the medium.

We may conclude that the guanidine-dependent poliovirus is non-pathogenic for monkeys since the concentration of guanidine required for its growth is much higher than that present in mammalian cells and fluids.

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Posterior Paralysis of Hamsters with Herpes simplex Infection of the Cervix

Little has been reported about specific virus infections of the female genital tract. Most text-books on gynaecology make sparse or no reference to virus infections of the tract except for lymphogranuloma venereum in human beings. As part of an investigation of the role of 'ordinary' viruses in neoplasia of the cervical tract, an attempt was made to develop an experimental model which would enable the interrelationships of viruses and carcinogens to be studied. Herpes simplex virus was selected for initial investigation because of the relatively frequent occurrence of vesicles in the tract of women and because of the latency aspect of its pathogenesis.

The *D.J.* strain of herpes virus (isolated by Dr. G. Waddell from a child with stomatitis) was used in the investigation. Virus pools were prepared in primary rabbit cell cultures. Hamsters (40–60 g) were infected as follows: Vaginal tracts were first swabbed with cotton pledgets on tooth picks. A second swab which had been soaked in the virus fluid (10^7 ID_{50} per ml.) was introduced through a paediatric otoscope speculum and applied directly to the cervix.

Animals were kept under observation for viral growth in the vaginal tract and for clinical and pathological effects. Vaginal exudates were collected daily from the tract using a swab soaked in Hanks's solution. The swabs were placed in vials containing 1 ml. of Eagle's solution with 10 per cent skimmed milk, and then quick-frozen and stored until all samples could be assayed for virus at the same time. Daily samples from pairs of hamsters were pooled and each was titrated in primary rabbit kidney cells using typical cytopathic effects as a measure of the infection.

Six experiments involving 117 infected animals were carried out. Infection in most animals was characterized clinically by a moderate to heavy vaginal exudate which had appeared usually by the third day. At about the fifth to the seventh day many of the hamsters showed a posterior paralysis involving both hind limbs. Such animals appeared alert and continued to eat for several days. A few died and some even recovered, but most were killed for further investigations: in all 48 hamsters (41 per cent) showed paralysis. The results of several experiments are summarized in Table 1.

Table 1. Effects on Hamsters of Herpes simplex Virus administered by way of the Vaginal Tract

Exp. No.	No. hamsters	No. with vagino-cervicitis	No. with posterior paralysis	No. dead
6	20	19	6	8
22	12	11	8	8
23	10	10	6	5
Cont.*	26	3†	0	0

* Controls were inoculated with Eagle's medium.
† Inflammatory response probably resulted from inoculation technique.

Preliminary histopathological findings include a cervicitis and vaginitis characterized by mononuclear cell thickenings of the sub-epithelial layers. In some animals epithelial microabscesses with some cells containing intranuclear inclusions were noted. Spinal cords of the few paralysed animals examined had lesions of viral myelitis involving primarily the grey matter. Lesions were not found in the brains of these animals.

Assays of vaginal washings indicated a period of low yield of virus shortly after inoculation with increasing amounts until day 5 when maximum yields of about 10^6 TCD_{50} per ml. washing fluid were obtained. This was followed by a tapering off until day 12 when virus could no longer be demonstrated. Virus was also isolated specifically from the cervix of some animals by explanting tissues and growing them on 'Millipore' filters; however, the results of this will be presented in a separate report.

The observation of special interest was the development of posterior paralysis in many of the animals with vaginocervicitis. This raises the question of how the virus reached the central nervous system from the tract. The posterior involvement suggests that spread of virus was by means of local neural routes. Paralysed animals did not show signs of encephalitis. It is likely, however, that once the virus had reached the spinal cord, it spread throughout the central nervous system, thus accounting for the death. As yet investigations of nervous tissue for virus growth and spread have not been made.

Johnson[1] recently studied virus pathways to the nervous system of mice given herpes by several non-neural routes. His work indicated that, depending on the route of administration, the virus can enter the central nervous system by either haematogenous or neural routes. Mice given virus subcutaneously or by way of the foot pad developed posterior paralysis. As determined by immunofluorescence techniques, the virus apparently spread along nerves by infection of endoneural cells (Schwann cells and fibroblasts). A similar pathway is suggested in the present investigation.

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