THE EFFECTS OF CORTISONE AND ADRENOCORTICOTROPHIC HORMONE ON POLIOMYELITIS AND ON OTHER VIRUS INFECTIONS

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There is growing evidence that under certain circumstances cortisone reduces the resistance of animals to bacterial infections; thus the infection becomes more intense after cortisone in experimental tuberculosis, syphilis, and pneumococcal septicæmia. There is also evidence that cortisone has a similar action in some virus infections. Schwartzmann[1] found that cortisone had an accelerating action on poliomyelitis in mice when the virus was inoculated intracerebrally. In addition, golden hamsters, *Mesocricetus auratus*, which normally are highly resistant to intracerebral inoculation with Lansing types of poliomyelitis virus, were found to become highly susceptible to this route of infection when MEF1 poliomyelitis virus was injected intracerebrally.

Experiments on Poliomyelitis in Mice

Experiment 1. 30 mice were inoculated intracerebrally with 0·03 ml. of a suspension containing approximately 100 LD50 doses of MEF1 poliomyelitis virus. 10 mice were left as controls: a second group of 10 mice was given 5 mg. of cortisone acetate 2 hours, and again 19 hours, after the intracerebral injection of virus: a third group of 10 mice received 5 mg. of ACTH intramuscularly 2 hours before being injected with the virus; 5 mg. of cortisone acetate was then injected intramuscularly 2 hours after the injection of the virus and the same dose was repeated 19 hours after the injection of virus. The average period from inoculation of virus to death is shown in Table I.

TABLE I

Number of mice	Treatment	Average period from inoculation to death days	Percentage mortality
10	Control	12·3 ± 3·2	90
10	ACTH and Cortisone	6·4 ± 2·6	100
10	Cortisone	7·5 ± 2·2	100

Experiment 2. A similar experiment was carried out with 30 mice, 15 mice being used as control and 15 receiving cortisone alone in the dosage as before. The results were similar: the controls died in an average of 14·2 days while the cortisone-treated mice died in an average of 6·2 days. One curious fact was that whereas the cortisone was invariably injected intramuscularly into the hind leg, paralysis almost always occurred in the front legs, thus showing that the paralysis was not due to the trauma occasioned by the cortisone iniection.

Experiment 3. In this experiment ACTH alone was used. 40 mice were inoculated intracerebrally with 0·03 ml. of approximately 50 LD50 doses of MEF1 poliomyelitis virus in a suspension of mouse cord in saline solution. 20 mice were given 5 mg. of ACTH intramuscularly 2 hours before being injected with the poliomyelitis virus and again with the same

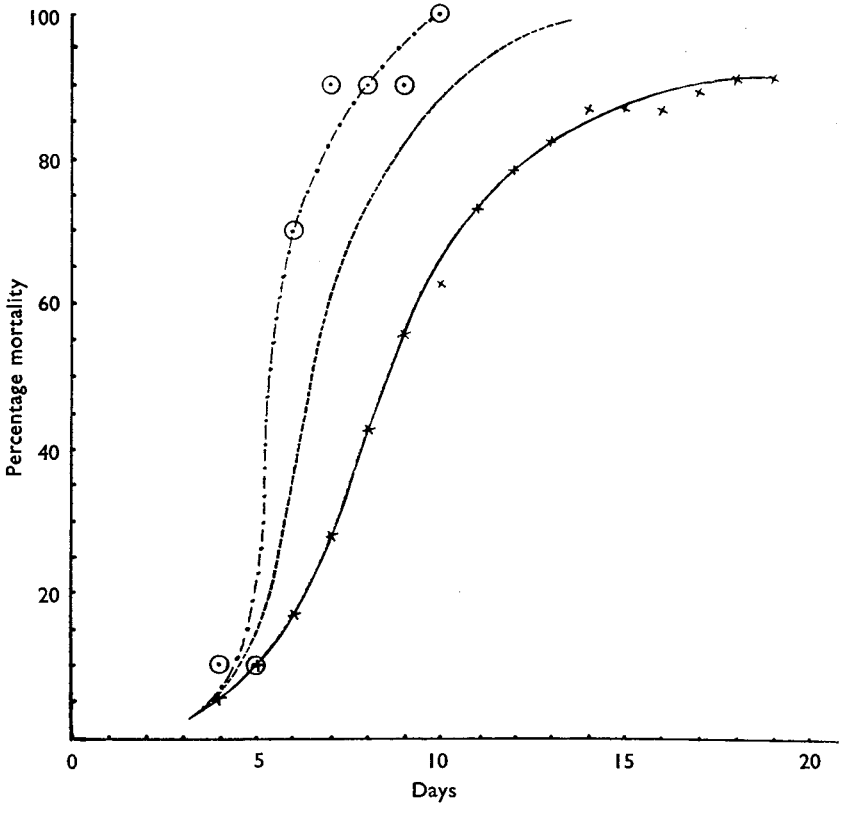

FIG. 1. Percentage mortality in days after inoculation of MEF1 poliomyelitis virus to:

———— normal mice.
– – – – – – mice treated with cortisone.
– · – · – · – mice treated with cortisone and ACTH.

dose 22 hours after injection of the virus. The average period from inoculation to death was 10·2 days in the control mice and 9·8 days in the ACTH-treated mice. Of the ACTH-treated mice half were dead by the seventh day after infection, whereas of the control mice half were dead only by the tenth day after infection. Thereafter the ACTH-treated mice showed a slight retardation and 2 mice survived the observation period of 25 days. The effect of ACTH on the poliomyelitis infection was not therefore very noticeable.

Experiment 4. The experimental results described suggested that cortisone in some way reduced the resistance of the central nervous system

to poliomyelitis virus once it had gained entrance to the central nervous system. This reduced resistance might be due to a change in the nervous system whereby paralysis and death occurred in the presence of a smaller amount of virus than was required to produce paralysis in the normal mouse or to the more rapid multiplication of virus in the spinal cord.

In order to throw light on this point 6 mice were each injected intramuscularly in the right leg with 5·0 mg. of cortisone in 0·5 ml. of saline solution: 6 control mice were similarly given an intramuscular injection of 0·5 ml. of saline solution. 2 hours later the 6 cortisone-treated and the 6 control mice were injected with 0·03 ml. of 50 MLD50 of MEF1 poliomyelitis virus in the form of a saline suspension of mouse cord. 18 hours later on the following day the cortisone-treated group were again injected with 5·0 mg. of cortisone in 0·5 ml. of saline solution and the control mice received 0·5 ml. of saline solution. 24 hours after the original injection of cortisone 3 mice from the cortisone-infected and 3 from the saline-treated mice were killed. Their cervical and lumbar cords were dissected out. The 3 cervical cords were weighed together and ground up to form a 1 in 10 suspension in physiological saline solution: the 3 lumbar cords were similarly treated. Dilutions of the 1 in 10 suspensions in saline solution were diluted so as to provide suspensions of 1 in 100, 1 in 200, and 1 in 400. Batches of 4 mice were then inoculated intracerebrally with 0·03 ml. of each dilution of cervical cord and of each dilution of lumbar cord. 48 hours after the original injection of cortisone the remaining 3 mice in each batch were killed and suspensions of their cervical and lumbar cords were prepared as before, batches of mice receiving 0·03 ml. of each dilution as before. The period from intracerebral inoculation to death is shown in Table II: mice were observed for 25 days; S signifies survival for this period.

TABLE II

PERIOD OF SURVIVAL OF MICE IN DAYS AFTER INTRACEREBRAL INOCULATION OF CORDS FROM CORTISONE-TREATED AND FROM NORMAL MICE INFECTED WITH POLIOMYELITIS (MEF1 STRAIN)

Dilution of cord suspension	Cords from cortisone-treated mice killed after		Cords from saline-treated mice killed after	
	24 hours	48 hours	24 hours	48 hours
Cervical cord:				
1 in 10 ..	9, 14, 15, 16	9, 10, 10, 16	S, S, S, S	22, S, S, S
1 in 100 ..	4, 9, 13, 14	6, 10, 11, 20	S, S, S, S	S, S, S, S
1 in 200 ..	16, 20, 20, S	9, 11, 11, 13	S, S, S, S	S, S, S, S
1 in 400 ..	13, 17, S, S	11, 11, 13, S	S, S, S, S	S, S, S, S
Lumbar cord:				
1 in 10 ..	22, S, S, S	6, 15, 13, S	S, S, S, S	S, S, S, S
1 in 100 ..	S, S, S, S	S, S, S, S	S, S, S, S	S, S, S, S
1 in 200 ..	S, S, S, S	S, S, S, S	S, S, S, S	S, S, S, S
1 in 400 ..	S, S, S, S	S, S, S, S	S, S, S, S	S, S, S, S

S = Survival.

The evidence thus shows that poliomyelitis virus was present in cervical cords even 24 hours after intracerebral inoculation of suspensions of cervical cord from mice inoculated with MEF1 poliomyelitis and treated with cortisone whereas no virus was present in the cervical cords of mice similarly infected with MEF1 virus but injected intramuscularly with

saline solution only. This difference between cortisone-treated and saline-treated mice is even more marked when the cervical and lumbar cords were removed from mice 48 hours after injection of MEF1 virus.

The evidence thus suggests that cortisone increases the rate of multiplication of poliomyelitis virus in the spinal cord of mice. Cultures from the heart blood and brains of cortisone-treated mice showed no evidence of bacterial infection.

Experiments in mice using 5-pregnene-3-β-ol-20-one, in the same dosage as with cortisone, showed no effect in accelerating the development of poliomyelitis.

Experiments on Poliomyelitis in Golden Hamsters

Even more striking than the effect of cortisone on MEF1 poliomyelitis virus infection in mice were the results obtained by Schwartzman[1] in the golden hamster (*Mesocricetus auratus*). The experiments here recorded were carried out with the Lansing strain of poliomyelitis. This strain in mice is normally rather less active than the MEF1 virus and the intervals between inoculation and death of a batch of mice show a more irregular distribution. From time to time batches of golden hamsters have been inoculated intracerebrally with the Lansing strain of poliomyelitis virus: no symptoms have been seen and the animals have survived.

10 half-grown golden hamsters were inoculated intracerebrally with 0·05 ml. of a 1 in 20 suspension in saline solution of mouse cord infected with the Lansing strain of poliomyelitis virus. 5 of the hamsters were injected intramuscularly in the hind leg with 5 mg. of cortisone acetate in 0·5 ml. of physiological saline solution: the same dose of cortisone was injected intramuscularly 3 hours after the intracerebral injection of the poliomyelitis virus. 5 control hamsters were given intramuscular injections of physiological saline solution. Of the control hamsters 1 died within 3 hours as a result of the shock of inoculation. Of the cortisone-treated hamsters, 1 became paralysed after 3 days, 1 after 8 days, 1 after 11 days and 1 after 28 days. The fifth hamster survived. The control hamsters were observed for 35 days without symptoms. Just as was described by Schwartzman, the cortisone-treated hamsters at the same time as they developed paralysis were seen to have hunched backs, ruffled fur and conjunctivitis. As in the case of mice, paralysis first appeared in the front legs.

Cultures of heart blood from the paralysed hamsters were bacteriologically sterile. In order to be quite certain that an encephalo-myocarditis virus was not being carried over together with the Lansing poliomyelitis virus suspensions 1 in 5, 1 in 50 and 1 in 200 of the spinal cords from paralysed mice were mixed with normal rabbit serum and with rabbit serum containing immune bodies against Columbia SK virus. Mice were then injected intracerebrally with 0·03 ml. of the various suspensions. There was no difference in the time at which the two series of mice developed paralysis and died: in other words the Columbia SK immune serum had no action on the virus present in the cord suspensions which was therefore not a virus of the encephalo-myocarditis group.

One point of considerable interest was in the histological appearances of the cords from hamsters which had developed paralysis as a result of cortisone treatment. The anterior horn cells in the cervical cord especially showed widespread necrosis but there was remarkably little round cell or neuroglial reaction in the neighbourhood of the degenerate neurones. As no material was available for comparison from normal hamsters inoculated with the Lansing virus 6 young hamsters were injected intracerebrally with 0·05 ml. of a 1 in 10 suspension in saline of spinal cord from mice paralysed by the MEF1 virus. 2 of these hamsters became paralysed in 18 and 24 days after inoculation: histological examination of their spinal cords showed a much more intense reaction round the anterior horn cells.

Experiments with Other Viruses

A small number of experiments were carried out with other viruses to determine the effects of cortisone.

Rift Valley Fever Virus. The viscerotropic strain of this virus when inoculated intraperitoneally into mice causes diffuse necrosis of the liver. 2 experiments were carried out. 20 mice were injected intramuscularly with 5 mg. of cortisone acetate: 2 hours later these 20 mice and 24 controls were inoculated intraperitoneally with 0·1 ml. of dilutions of mouse blood infected with Rift Valley fever virus. The cortisone-treated mice were given a further intramuscular injection of 5 mg. of cortisone 3 hours after the intraperitoneal injection of the virus. The period from inoculation to death is shown in Table III.

TABLE III

MICE INOCULATED INTRAPERITONEALLY WITH RIFT VALLEY FEVER VIRUS

Dilution of blood	Time in days from infection to death	
	Control mice	Cortisone-treated mice
10^{-1}	2, 2, 2, 2	2, 2, 2, 2
10^{-4}	3, 3, 4, 5	2, 2, 2, 2
10^{-5}	3, 3, 4, 5	2, 2, 2, 2
10^{-6}	3, 4, 4, 5	2, 2, 2, 2
10^{-7}	S, S, S, S	3, 3, 3, 3
10^{-8}	S, S, S, S	5, 5, 6, S

S = Survival.

It will be seen that the infection in the cortisone-treated mice was much more rapid than in the control mice. Histological examination of the livers of the cortisone-treated mice, dying in 48 hours after an injection of 0·1 ml. of blood diluted 10^{-6} showed a diffuse liver necrosis with margination of the nuclear chromatin and acidophilic intranuclear inclusions entirely comparable to that seen in mice inoculated with infected blood diluted 10^{-1}.

Coxsackie Virus. Evidence has been obtained that young adult mice which show only a very slight multiplication of Coxsackie Virus in the muscles, when injected with cortisone exhibit a much more intense production of virus.

Encephalo-myocarditis Viruses. The Columbia SK Virus when inoculated intraperitoneally into mice gives rise to viræmia followed by the

production of encephalitis and myocarditis. When cortisone-treated and control mice are inoculated intraperitoneally with dilutions of Columbia SK virus it is again found that the cortisone-treated mice exhibit a rapid infection after inoculation with dilutions of infected mouse brain which kill normal mice only after a delay of 4 to 5 days.

Discussion

The evidence obtained with poliomyelitis viruses of the Lansing type, with Rift Valley fever virus, with Coxsackie viruses, and with encephalomyocarditis virus (Columbia SK and Senger Viruses) all points to the fact that cortisone and ACTH permit a more rapid multiplication of these viruses. How exactly this increased virus multiplication occurs is not known although there is a suggestion that the tissue reaction of the host to infection is in some way inhibited. This effect on the tissue reactivity is not specific to any one tissue; Kass *et al.*[2] found that in mice infected with influenza virus (a dilution of 10^{-5} intranasally) and treated with cortisone, 10 of 10 mice died in an average of 6·5 days: only 6 of 10 control mice died in an average of 8·5 days. In the survivors after 12 days the lung lesions were not extensive. Kligman *et al.*[3] similarly found that when 5 mg. of cortisone was injected daily into guinea-pigs for 2 days and vaccinia virus was then inoculated intradermally the lesions were more intense in the cortisone-treated animals than in normal guinea-pigs.

Conclusions

1. Treatment of mice infected with poliomyelitis virus by cortisone early in the infection results in a more rapid onset of paralysis and death than in control mice untreated by cortisone.

2. The evidence suggests that there is more rapid multiplication of virus in the cervical cord of cortisone-treated mice.

3. Injections of ACTH early in infection produce early paralysis and death in some mice but the effect is less marked than with cortisone.

4. Golden hamsters infected with Lansing poliomyelitis are highly resistant but if cortisone is injected at the time of infection the majority develop paralysis and die. This infection is due to poliomyelitis virus and not to an intercurrent infection.

5. There is evidence that cortisone increases the rate of multiplication and hence the rapidity and severity of symptoms in mice infected with the viruses of Rift Valley fever, encephalomyocarditis (Columbia SK) and Coxsackie (type A).

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