

## THE EFFECT OF HYPOPHYSECTOMY ON THE RESISTANCE OF MICE TO INFECTION WITH POLIOMYELITIS VIRUS.

MARY F. LOCKETT, G. A. H. BUTTLE AND E. M. HOWARD.

*From the Pharmacology Department, The School of Pharmacy,  
University of London.*

Received for publication March 3, 1954.

SHWARTZMAN (1950) was the first to show that cortisone lowers the resistance of mice to poliomyelitis virus. His work was confirmed and extended by Findlay and Howard (1952), who showed that cortisone both reduces the survival time of infected mice and accelerates the spread of the virus along the spinal cord from the site of the intracerebral inoculation. Neither Shwartzman nor Findlay and Howard were able to demonstrate a similar effect when adrenocorticotrophic hormone (ACTH) was substituted for cortisone. This paper reports the results of experiments in which the effect of hypophysectomy on the resistance of mice to poliomyelitis virus was examined.

### MATERIALS AND METHODS.

Male white mice weighing 18–20 g. were used throughout. Hypophysectomy was performed, under bromethol anaesthesia, by a simplification of Smith's rat technique (1930). The control animals underwent mock operation in parallel with those hypophysectomised. The authors present no guarantee that hypophysectomy was complete. However, the heads of all the hypophysectomised animals were removed after death, fixed in formal-saline, and inspected for the absence of recognisable pituitary gland; none was found. Also, as an additional check, the adrenal glands were removed from both control and hypophysectomised mice, fixed and weighed. Fig. 1 shows that there was no overlap between the weights of the glands from the hypophysectomised animals and the controls.

Intracerebral inoculation with poliomyelitis virus, strain M.E.F.1, was performed between 3 and 13 days after the hypophysectomy operation. The inoculum was prepared from a 10 per cent suspension of several infected mouse cords. This suspension was centrifuged at 3,000 r.p.m. for 5 min., the supernatant was diluted in saline to give a concentration equivalent to 1 g. cord/100 ml. saline: 0.03 ml. of this dilution was injected intracerebrally under ether anaesthesia. This dose of virus produced a 90–100 per cent mortality in normal mice.

### RESULTS.

In each of the five experiments performed, the mean survival time of the group of hypophysectomised mice was longer than that of the mock-operated animals; a summary of these results is given in Table I. There was no significant difference in the percentage mortality between the two groups.

When the adrenal glands were weighed, it became apparent that, although there was no correlation between adrenal weight and survival time in the control

TABLE I.—*Resistance of Hypophysectomised Mice to Infection with Poliomyelitis Virus.*

Exp. No.	Mice used.	Survival time in days.		Adrenal weights at death (mg./20 g. mouse).	
		Controls.	Hypophysectomised.	Controls.	Hypophysectomised.
1	29	9.2±0.98 (20)	14.2±1.95 (9)*	Not weighed	Not weighed
3	19	6.9±1.51 (9)	11.4±1.16 (10)*	4.80±0.18 (9)	3.04±1.34 (10)*
5	20	12.6±2.18 (14)	16.7±1.86 (6)	2.34±0.07 (16)	1.55±0.08 (10)*
6	17	6.7±1.49 (6)	10.5±0.97 (11)*	2.43±0.06 (8)	1.36±0.05 (22)*
7	96	11.1±0.59 (49)	15.9±0.98 (47)**	5.06±0.11 (45)	2.43±0.07 (46)**

Percentage survival in control mice = 1.01 per cent.

“ “ “ hypophysectomised mice = 13.3 per cent.

Figures for survival time, and adrenal weight are entered as mean ± standard error (number of animals) correct to two decimal places. In experiments 3 and 7, paired glands from single animals were weighed; in experiments 5 and 6, individual glands were weighed separately. Significant difference between values for control and hypophysectomised animals is indicated by asterisks: one,  $P = <0.05$ ; two,  $P = <0.001$ .

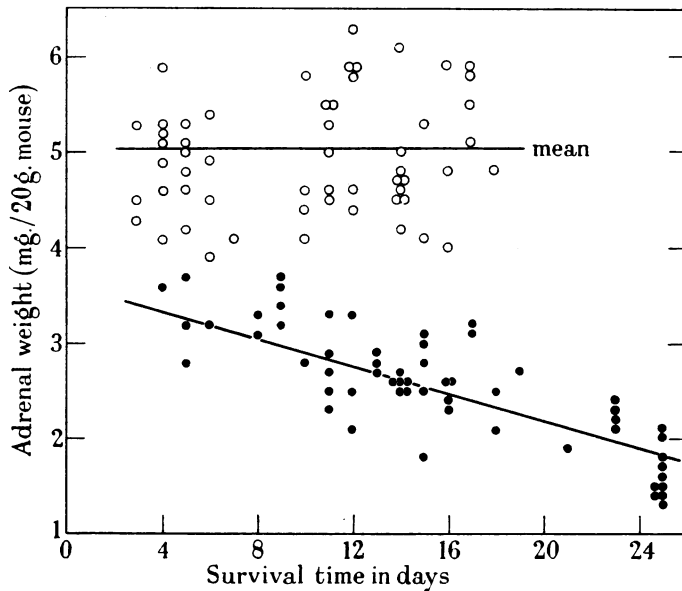


FIG. 1.—Weights of adrenal glands at death related to survival time for control (open circles) and hypophysectomised (closed circles) mice inoculated intracerebrally with a fatal dose of poliomyelitis virus, strain M.E.F.1 at zero time. Each circle represents one mouse. The data are taken from experiments 3 and 7, Table I. In controls there is no correlation between adrenal weight and survival time. In the operated there is correlation ( $P = <0.001$ ;  $r = 0.86$ ).

groups, there was highly significant correlation in this respect in the groups of hypophysectomised animals (Fig. 1). This was probably due to the decrease in the size of the adrenals throughout the experimental period (Fig. 2). It was therefore of interest to determine whether the length of the interval between hypophysectomy and inoculation had any effect on survival time.

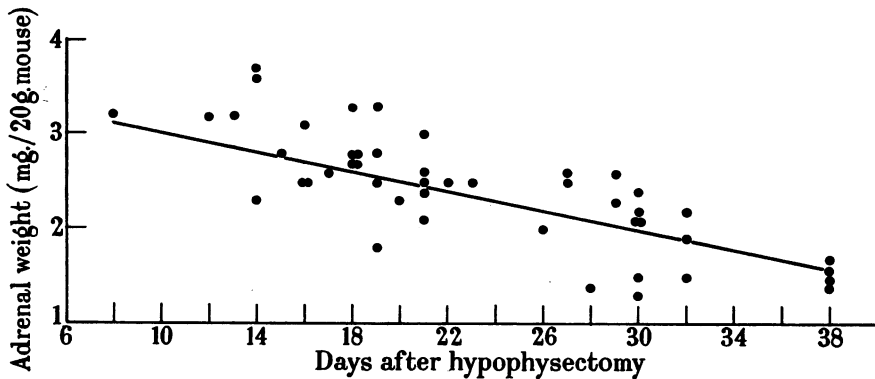


FIG. 2.—Relation of weight of adrenal glands from hypophysectomised mice, dead of poliomyelitis, to time after hypophysectomy. Each circle represents one mouse. The data are from experiment 7, Table I. Correlation between adrenal weight and days post-operative:  $P = <0.001$ ;  $r = 0.64$ .

As the mice used in experiment 7, Table I were hypophysectomised at intervals varying between 3 and 13 days before inoculation, the survival times of these groups of animals have been compared (Table II). The effect of hypophysectomy on survival time was found to be independent of the interval between the operation and inoculation.

TABLE II.—*Survival Times of Mice Inoculated with a Fatal Dose of Poliomyelitis Virus at Various Intervals after Hypophysectomy (Data from Experiment 7, Table I).*

Days between hypophysectomy and inoculation.	Survival times in days mean $\pm$ S.E. (number mice).
Controls .	11.1 $\pm$ 0.59 (49)
3-4 .	15.7 $\pm$ 1.0 (11)
5 .	15.5 $\pm$ 2.1 (11)
7 .	16.3 $\pm$ 1.8 (15)
13 .	16.8 $\pm$ 2.9 (10)

It was thought that the lengthened survival time of the hypophysectomised mice might be due to their inability to respond to the stress of infection by an increased production of adrenocorticotrophic hormone from the anterior pituitary gland. In an attempt to show that normal mice responded to this type of stress by an increased output of this hormone and consequent hypertrophy of the adrenal glands, groups of mice were inoculated intracerebrally with poliomyelitis virus. Similar groups of animals were left uninoculated. Both groups were kept under similar conditions. One uninoculated mouse was killed whenever an infected one died. The adrenal glands of these mice were fixed and weighed. Table III shows that, although there appeared to be a highly significant difference between the adrenal weights of infected and non-infected animals when expressed in terms of body weight, this difference did not exist when the absolute weights

of the adrenals from the two groups were compared. This could be accounted for by the fact that the control mice gained weight during the experimental period whereas the infected animals did not. Thus these results do not provide conclusive evidence of adrenal hypertrophy as a result of infection with the virus.

In the experiments recorded in Table III, correlation was found between the weight of adrenal gland per 20 g. mouse and body weight. Normal mice, in a weight range of 15–31 g., gave a correlation coefficient of  $-0.55$  ( $P = < 0.001$ ,  $N = 33$ ). Infected mice, in a weight range of 10–22 g., gave a correlation coefficient of  $-0.72$  ( $P = < 0.001$ ,  $N = 33$ ).

TABLE III.—*Adrenal Weights of Normal Mice Compared with those of Mice Infected with Poliomyelitis Virus.*

Experiment.	Group.	Adrenal weights.	
		mg./20 g. body weight.	mg./mouse.
I	Normal	$3.10 \pm 0.15$ (12)	$2.55 \pm 0.16$ (12)
	Infected	* $5.51 \pm 0.18$ (12)	$3.71 \pm 0.18$ (12)
II	Normal	$4.38 \pm 0.37$ (9)	$4.94 \pm 0.39$ (9)
	Infected	$4.88 \pm 0.37$ (9)	$3.99 \pm 0.27$ (9)
III	Normal	$3.69 \pm 0.26$ (12)	$3.84 \pm 0.13$ (12)
	Infected	* $6.05 \pm 0.41$ (12)	$4.01 \pm 0.18$ (12)
I, II and III	Normal	$3.66 \pm 0.27$ (33)	$4.04 \pm 0.24$ (33)
	Infected	* $5.08 \pm 0.33$ (33)	$4.09 \pm 0.21$ (33)

Significance ( $P = < 0.01$ ) is indicated by an asterisk.

#### DISCUSSION.

It has already been reported that adrenocorticotrophic hormone does not influence the survival time of mice infected with poliomyelitis virus (M.E.F.1) when only two doses of the hormone are given (Findlay and Howard, 1952). Further work has shown that repeated injections of 0.25 mg. ACTH twice daily from the day of inoculation do not increase the sensitivity, although they do cause adrenal hypertrophy. Cortisone, on the other hand, does increase susceptibility. Similar results have been reported by Kass, Lundgren and Finland (1954) for mice infected with pneumococci and with influenza virus respectively.

It is possible that the increase in survival time after hypophysectomy may be due to the inability of the animal to respond to infection by increasing the output of ACTH, but the lack of other pituitary hormones, possibly growth hormone, may also be responsible.

#### SUMMARY.

Hypophysectomy has been shown to prolong the lives of mice inoculated with poliomyelitis virus.

This increase in survival time of hypophysectomised mice is independent of the time interval between the operation and the inoculation.

The adrenal glands of the hypophysectomised mice decreased in weight throughout the experimental period of 38 days.

It is a pleasure to acknowledge the encouragement and help given by the late Dr. G. M. Findlay. Part of the expense was defrayed by a grant from the Medical Research Council to one of us (G. A. H. B.).

REFERENCES.

- FINDLAY, G. M., AND HOWARD, E. M.—(1952) *J. Pharm. and Pharmacol.*, **4**, 37.  
KASS, E. H., LUNDGREN, M. M., AND FINLAND, M.—(1954) *J. exp. Med.*, **99**, 89.  
SHWARTZMAN, G.—(1950) *Proc. Soc. exp. Biol., N.Y.*, **75**, 835.  
SMITH, P. E.—(1930) *Amer. J. Anat.*, **45**, 205.