illustrated in Fig. 1b was only 15 minutes. Survival times for the sciatic nerve ranged from 1.5 to more than 4 hours over all seasons (11 experiments). An increase of intensity decreased the survival time (Fig. 1b). If failure is exclusively the result of the lowering of the action potential below a critical level, little change in threshold would be expected. A decrease in threshold averaging 15% in 10 experiments (Fig. 1a) was found. The extreme rise obtained with single fibers(1) was absent. Continuous irradiation leads to a progressive depolarization of the treated region. Six identical results were obtained; one is illustrated in Fig. 2a. During irradiation the supernormal period disappears and the relative refractory period becomes extensive (5 experiments) as shown in Fig. 2b. Cysteine (0.02 M), at pH 7, calcium chloride (0.02 M) and visible light (2000 watts) were ineffective in retarding the fall in action potential. Both short and long periods of illumination were equally ineffective in producing photoreactivation.

*Discussion.* Ultraviolet appears to affect nerve in much the same way as several metabolic inhibitors. Cyanide and nitrogen, for example, cause a fall in resting potential with little or no change in threshold(2). Ultraviolet differs from the effect of methylfluoracetate(3) in that the latter affects the threshold strikingly while leaving the resting potential unaltered. It appears probable that a major effect of ultraviolet on nerve is the result of the inhibition of one or several respiratory enzymes. Since the magnitude of the threshold change is slight, little direct effect on the membrane seems involved.

*Summary.* Ultraviolet irradiation of nerve at 2537A leads to a fall in action potential, a depolarization and a prolongation of the relative refractory period. A slight fall in threshold was observed.

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## Effect of Hypophysectomy on Pigmentation and Ascorbic Acid Excretion in Black Rats.* (19326)

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It was previously observed that adrenalectomy in black rats was associated with a marked increase in the deposition of melanin in the hair follicles and bulbs in these animals(1). This report is concerned with the effect of hypophysectomy on pigmentation in black rats.

*Experimental procedure.* Male and female black rats of the Long-Evans strain, bred in our laboratories, were hypophysectomized when the animals were between 50 and 60 days of age, by the Hormone Assay Laboratories in Chicago. The weight of the males ranged from 125 to 150 g and of the females from 100 to 125 g. After hypophysectomy, the rats were fed a normal experimental diet supplemented with small amounts of canned dog food or evaporated milk. The fur on the back was shaved and the degree of pigmentation and hair growth recorded. A number of animals were placed in metabolism cages and the urine was collected for 24-hour periods. Ascorbic acid was determined by the method of Rowe and Kuether(2). At the conclusion of the experiments, the rats were sacrificed and the adrenal glands were removed, weighed, and prepared for pathological study.

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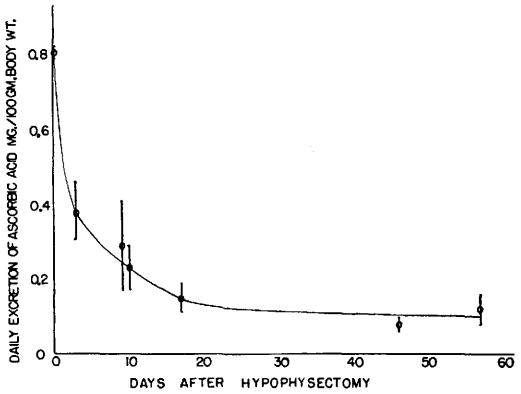


FIG. 1. Daily excretion of ascorbic acid by hypophysectomized rats is in mg of ascorbic acid per 100 g body weight. Vertical lines indicate the standard errors.

*Results. Pigmentation.* Seven or 8 days after hypophysectomy, a bluish pigmentation was visible through the thin overlying cutis of the black hypophysectomized rats, similar to the pigmentation previously observed in black rats following adrenalectomy(1). The pigmentation was not as uniformly intense as that observed in the adrenalectomized rat, but the deep blue coloration due to increased melanin deposition was clearly visible through the thin overlying cutis. This coloration appeared 7 or 8 days following hypophysectomy and reached the maximum at 15 to 17 days. After 20 days, the pigmentation gradually faded to a grayish color.

*Ascorbic acid excretion.* The data on ascorbic acid excretion are summarized in Fig. 1. Prior to hypophysectomy, the rats excreted 0.80 ± 0.14 mg of ascorbic acid per 100 g of body weight in 24 hours. Following hypophysectomy, the ascorbic acid excretion decreased, reaching a level of about 0.1 mg per 100 g of body weight by 50 days after hypophysectomy. We previously reported that adrenalectomized rats on identical diets excrete approximately 0.25 mg of ascorbic acid per 100 g of body weight(3).

*Adrenal weights.* The mean weights of the adrenals of normal male and female rats 60 days of age and of hypophysectomized male and female rats are shown in Table I. Without exception, the adrenals of the hypophysectomized rats were found to be smaller than the adrenals of normal 60 day old rats, the age at which the experimental rats were hypophysectomized. Pathological study revealed marked atrophy of the adrenal cortex of the hypophysectomized rats.

*Discussion.* The development of intense pigmentation in the hair apparatus of black rats following hypophysectomy supports the suggestion previously made that the increased deposition of melanin following adrenalectomy is related to the absence of the steroid hormones of the adrenal cortex(1,4). The observations that pigmentation in adrenalectomized black rats was inhibited by the daily injection of DCA tends to support this suggestion(4). The decrease in the excretion of ascorbic acid which occurred in both adrenalectomized(3) and hypophysectomized rats may be related to the increased deposition of melanin in these animals. It has been reported that the administration of ascorbic acid will correct a disorder in the metabolism of tyrosine and phenyl alanine that occurs in scorbutic guinea pigs(5) and in human infants(6).

Obviously, the decreased excretion of ascorbic acid by hypophysectomized and adrenalectomized rats is only one factor which may contribute to the increased pigmentation in these animals, as the increased pigmentation fades after four weeks, while the decreased excretion of ascorbic acid continues throughout the life of the animal.

*Summary.* Pigmentation was observed in

TABLE I.

| | No. of rats | Age, days | Avg wt of rats, g ± S. E. | Absolute wt in mg ± S. E. | Wt in mg/100 g body wt ± S. E. |
|---|---|---|---|---|---|
| Intact ♂ | 8 | 60 | 129 ± 9 | 20 ± .8 | 15.6 ± .8 |
| ♀ | 6 | 60 | 110 ± 5 | 22 ± .4 | 20.2 ± .8 |
| Hypophysectomized ♂ | 11 | 111-185 | 133 ± 7 | 8.9 ± .6 | 6.9 ± .6 |
| ♀ | 6 | 111-185 | 112 ± 3 | 9.3 ± .4 | 8.3 ± .4 |

black rats following hypophysectomy, similar to the pigmentation occurring in black rats following adrenalectomy. The adrenals of the hypophysectomized rats were smaller than normal, with atrophic cortices. Excretion of ascorbic acid decreased to low levels following hypophysectomy. The increase in pigmentation is believed to be related to the derangement of the steroid hormones of the adrenal cortex of the hypophysectomized rat.

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## *In Vitro* Effect of Aureomycin, Terramycin, and Chloramphenicol on Typhus Rickettsiae.* (19327)

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In a previous communication(1) from this laboratory it was reported that purified typhus rickettsiae oxidize glutamate, and that the rate of this oxidation is proportional to the concentration of viable rickettsiae, as determined by their toxicity for white mice. It was further shown(2) that the antibiotics effective in rickettsial infections, aureomycin, terramycin, and chloramphenicol, do not reduce the toxicity for white mice when tested *in vitro* for one hour at concentrations of 10 and 30 μg per ml. The present paper deals with the effect of these antibiotics in concentrations of 10-300 μg per ml on the rate of glutamate oxidation and the toxicity for mice of purified suspensions of epidemic and murine typhus rickettsiae.

*Materials and methods. Antibiotics.* Aureomycin hydrochloride (twice recrystallized, Lederle), terramycin hydrochloride (Pfizer), and chloramphenicol (synthetic, Parke, Davis)† were used. In some experiments, terramycin sterile base (Pfizer), and chloramphenicol (pure crystalline) were used; however, no differences between the two preparations of these drugs were observed. The antibiotics were dissolved in a salt solution of the following composition: KCl 0.126 M; NaCl 0.0018 M; $KH_2PO_4$ 0.0012 M; $Na_2HPO_4$ 0.0109 M (solution K 7.5)(3); the pH was adjusted to 7.5 by addition of KOH, and dilutions were made with solution K 7.5. *Rickettsial suspensions* (the Wilmington strain of *R. mooseri* and the Breinl strain of *R. prowazeki*) were prepared from frozen infected yolk sac pools as described previously (1,3). The precipitate from the final high speed centrifugation was resuspended in either sucrose-P (sucrose 0.225 M; $KH_2PO_4$ 0.0016 M; $K_2HPO_4$ 0.0086 M, pH 7.5)(3), or 6% bovine serum albumin powder, Armour's fraction V, in solution K 7.5, pH 7.5. In all experiments, 1 ml of the suspending fluid was used for each 2 g of original infected yolk sac. *The rate of oxygen uptake* was measured by the Warburg method at 34.5°C. Flasks of two capacities, 3 ml and 8 ml, both

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