and TEM 4T. Hycase can be replaced by an amino acid mixture, with a slightly lower plane of growth. An interchangeable requirement for pantethine, pantetheine or coenzyme A was demonstrated. Calcium pantothenate had very low activity and panthenol was inactive. Biotin and vitamin B_{12} stimulated acid production during growth.

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The Electroencephalogram in Experimental Allergic Encephalomyelitis in the Lewis Rat. (30970)

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Experimental allergic encephalomyelitis (EAE) is a distinct, reproducible disease that can be induced in a variety of animal species by injection of nervous tissue incorporated with Freund's adjuvant(1). Although considerable variability has been noted, electroencephalographic (EEG) abnormalities and convulsions have been reported to occur in all of the species so examined: monkey(2), rabbit(3), and guinea pig(4). In this communication we will report the results of a study on the effects of EAE on the EEG of the Lewis rat.

Methods. Electrodes were chronically implanted in 11 male Lewis strain rats (Microbiological Associates, Bethesda, Md., weighing 150-200 g) under sodium pentobarbital anesthesia (35 mg/kg). The electrodes consisted of stainless steel screws driven into the skull over the frontal, temporal and occipital cortex unilaterally on the left side. The screws were wired to a Winchester miniature socket and encased in acrylic cement. Bipolar EEG recordings were obtained using a Grass model III-D electroencephalograph. Recordings were obtained with the animals unrestrained in a 30 by 10 by 10 cm metal box to which the rats had previously been acclimated. A control record was obtained from each rat on day 1. On day 2, a 40% w/v homogenate of isologous spinal cord in distilled water containing 0.5% phenol was emulsified with an equal volume of Freund's complete adjuvant (4 mg/ml killed tubercle bacilli). A single dose of 0.05 ml of the encephalitogenic emulsion was then injected into the right hind foot pad(5). Recordings were obtained on days 4, 6, 7, 8, 11, 12, 13, 14, 15, 17, 19 and 20 after injection of emulsion. This included the period before, during, and after the paralysis observed in EAE. All animals were examined and weighed on the days when EEG records were obtained.

Results. Animals continued to gain weight for a period of 8 days after injection of emulsion but then lost weight rapidly over the next 7 days (Fig. 1). From day 12 to day 17 all animals showed pronounced flaccid hind-

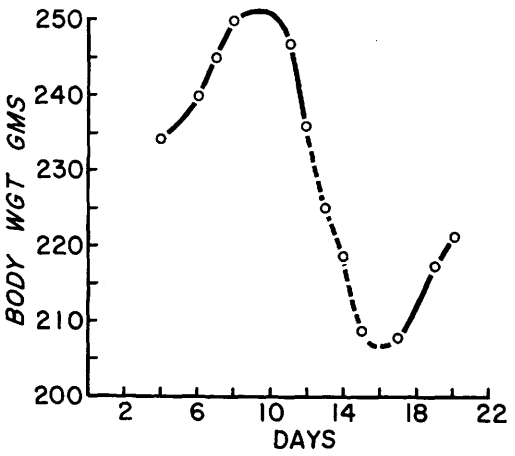

FIG. 1. Rat mean body weights during EAE. Dotted portion indicates period during which all animals showed hind-limb paralysis. Number of days after injection of the encephalitogenic emulsion are indicated on abscissa. Open circles indicate days on which body weights and EEG recordings were obtained.

limb paralysis, fecal impaction, and urinary incontinence. Complete recovery from paralysis, accompanied by a gain in body weight, occurred in all animals by day 19.

No difference could be detected in the EEG records obtained before, during, and after the induction of paralysis. During all 3 periods, the EEG records most commonly showed a mixed pattern with no clearly dominant frequency, although occasionally a pattern showing pronounced high frequency activity accompanied by behavioral signs of increased alertness was obtained. A typical EEG tracing obtained from a paralyzed rat is shown in Fig. 2.

Discussion. Previous studies using monkeys, rabbits and guinea pigs in which EEG abnormalities have been reported(2,3,4) indicated that convulsions often occurred during EAE. In a study of Wistar rats it was found that EAE increased central nervous system excitability as indicated by a decreased threshold for pentylenetetrazol (Metrazol)-induced convulsions(5). No mention was made in this report of "spontaneous" convulsions during EAE in these rats. In previous studies in this laboratory, EAE has been produced in over 300 Lewis rats: convulsions have never been observed in these experiments. This study indicates that in the highly inbred Lewis rat given isologous spinal cord emulsion, EEG abnormalities and convulsions do not necessarily accompany the paralysis observed during EAE.

It has been stated that the lesions observed in EAE are found more often in the subcortical rather than cortical regions in the monkey(2). It is possible that in the rat the cortical lesions occur even less often than in the monkey and therefore EEG abnormalities are not found in this animal. It should be recognized, however, that in general organisms lower on the phylogenetic scale are more resistant to procedures which produce convulsions than are those species which are advanced and have a more elaborately developed central nervous system(6).

Summary. Electroencephalograms were recorded from Lewis rats before, during, and after the paralysis seen during EAE. In contrast to studies in other species during EAE, no convulsions and no EEG abnormalities were observed.

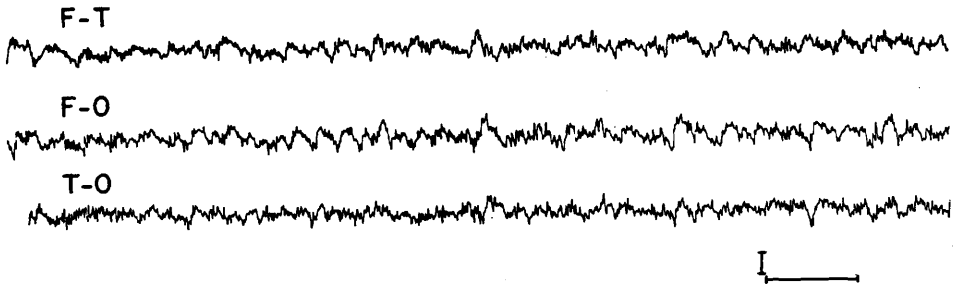

FIG. 2. EEG tracing obtained during EAE induced paralysis. Bipolar recordings: F-T, frontal temporal leads; F-O, frontal occipital leads; T-O, temporal occipital leads. Calibration: vertical bar, 200 μV; horizontal bar, one sec.

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Erythopoiesis During Pregnancy and Lactation in the Mouse. II. Role of Erythropoietin.* (30971)

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An increase of total blood volume and red cell volume in lactating rats has been demonstrated by Bond(1). During lactation in mice, plasma volume and red cell volume increased(2) and their plasma had increased erythropoietic activity(3). While prolactin has been found to stimulate erythropoiesis in polycythemic mice(3), in postpartum nonlactating mice(2), and in orchidectomized male mice(3), other hormones active during lactation have not shown an erythropoietic effect(3,4).

Since erythropoietin is a known potent erythropoietic stimulant, the relation of this hormone to the erythropoietic effect of prolactin was investigated to determine if prolactin acted directly on the bone marrow or through stimulation of ESF production.

Materials and methods. To determine how rapidly erythropoiesis was suppressed following exposure to hyperoxia, the incorporation of Fe59 into erythrocytes was determined in normal female mice exposed to 60% O2-40% N2 environment for 1, 2 and 5 days. Twenty-four hours prior to their removal from this environment, 0.5 μc of Fe59 C13 was injected into the tail vein and immediately upon removal, blood was obtained by cardiac puncture. The 24-hour incorporation of Fe59 into erythrocytes of these groups of mice was compared to that of normal unexposed female mice.

Since hyperoxia abolishes the secretion of erythropoietin(5), groups of normal female mice and of postpartum lactating mice, 0-12 hours from birth were placed with their newborn in a 60% O2-40% N2 environment for periods of 5, 10 and 15 days. Two similar groups of mice were simultaneously exposed to hyperoxia and injected daily with 250 μg of prolactin† for 15 days. Upon removal from the hyperoxia, the RCV was determined as described previously(2) in all mice and compared with the values obtained in normal females, in 15-day lactating mice in normal atmosphere, in normal female mice injected daily with 250 μg of prolactin for 15 days, and in normal mice and nonlactating postpartum mice exposed to 10% O2 environment for 15 days.

To determine whether hyperoxia would influence the plasma erythropoietic activity of lactating mice, plasma was collected as previously described(3) on the 1st, 5th and 10th postpartum days from lactating mice exposed from the time of parturition to hyperoxia. For comparison, plasma was collected from normal female mice exposed to *hypoxia* (10% O2-90% N2) for 24, 48 and 72 hours. The erythropoietic activity of these plasmas was

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