Conclusion. Morphologic and histochemical studies of glial reaction during Wallerian degeneration in the dorsal column of the spinal cord have shown that reactive gliosis is independent of alterations of the blood-spinal cord barrier. These data suggest that substances inducing gliosis may pre-exist in normal central nervous system tissue.

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Influence of Pregnancy and Cortisone Treatment on Brown Adipose Tissue in Bats.* (27823)

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The demonstration of rabies virus in the brown adipose tissue of experimentally and naturally infected Chiroptera has led to the hypothesis that this tissue, the so-called hibernating gland, may serve as an extraneural site for viral sequestration in the naturally infected, asymptomatic bat(1,2). Subsequent studies have dealt with the influence of certain stress factors which the bat may encounter or experience in nature (*i.e.*, environmental temperature and the reproductive cycle) on the course of rabies infection in these animals(3,4). Although the frequency with which bats could be experimentally infected with rabies virus was not increased during the gestation period over that observed in the non-gravid animal, the ability to demonstrate viral proliferation in the brown fat of infected gravid animals varied noticeably with the stage of pregnancy(4). During the course of these studies differences were noted in gross appearance of the brown fat lobes of bats in various stages of the gestation period, and the amount of tissue present in animals at time of delivery and in the early postpartum period was depleted. In view of these findings, the present study was undertaken to examine histologically the brown adipose tissue of the Mexican free-tailed bat (*Tadarida b. mexicana*) throughout the reproductive cycle. In addition, since cortisone has been shown to greatly influence the brown fat of hamsters and mice(5) and of rats(6), the response of this tissue in cortisone treated, non-gravid bats is presented.

Methods. Spring-breeding Mexican free-tailed bats,† *Tadarida b. mexicana,* used throughout these studies, were maintained at 29°C ± 2°C (relative humidity 60-70%). Methods for the laboratory care and maintenance of these animals have been described (7).

Gravid bats were netted at progressive intervals in the reproductive cycle between early May and late June. In groups of 25-30 per

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† Bats were obtained from the Blowout Cave, Blanco Co., Texas.

harvest, the animals were anesthetized and exsanguinated. The interscapular brown adipose tissue, an encapsulated, bilobed structure, was extirpated and fixed in formalin for H & E sections. As an index to the approximate stage of pregnancy, fetuses from gravid animals were weighed and measured(4).

Non-gravid bats netted in November received a total of 5 mg of cortisone (Cortone acetate, Merck, in saline suspension) intramuscularly in 2 doses of 2.5 mg each, 24 hours apart. A control group of bats was inoculated with an equal volume of saline. Beginning 48 hours following cortisone treatment, interscapular brown adipose tissue was extirpated at successive intervals from both cortisone-treated and control groups of bats, a total of 20 animals per group for each harvest day. The brown fat lobes from each group were weighed collectively on a Mettler analytical balance and immediately fixed in formalin for histologic examination.

Results. Effect of pregnancy on brown adipose tissue of bats. Microscopic analyses of the brown fat of bats in various stages of the reproductive cycle showed that as pregnancy progressed, the lipid content of the brown fat cells increased proportionally, reaching a maximum late in the gestation period. The gross appearance of brown fat varied from depleted lobes with deep red coloration (early pregnancy) to relatively enlarged lobes of light brown color (mid to late pregnancy). At time of parturition and in the early postpartum period this tissue was dark red and, again, depleted. The brown fat lobes of bats netted 2-3 weeks after the delivery period had already begun to increase in size, indicating that the observed depletion during parturition is of short duration.

Representative sections of brown fat from bats netted in early June (late pregnancy) and in late June (parturition) are shown in Fig. 1 and 2, respectively. Large, coalesced, lipid locules are evident in the cytoplasm and the nucleus is displaced toward the periphery of the cell, indicating a marked hypertrophy due to lipid deposition in the brown fat of bats in the late period (Fig. 1). In sharp contrast, the brown fat of puerperal bats is essentially free of lipid (Fig. 2).

TABLE I. Effect of Cortisone on Weight of Interscapular Brown Adipose Tissue of the Bat *(Tadarida b. mexicana).*

Group	Days following cortisone treatment 2	4	5	6
Cortisone-treated*	53.5†	56.2	63.3	41.5
Control	32.5	30.5	32.0	31.0

* Each bat received 2 intramuscular doses of 2.5 mg cortisone 24 hr apart.

† Avg wt (mg) of interscapular brown fat/bat.

Effect of cortisone on brown adipose tissue of bats. A comparison of the average weight of the brown fat of cortisone-treated and control groups of non-gravid bats is shown in Table I. Two days following treatment with cortisone, the average weight of brown fat per bat is noticeably increased: 53.5 mg as compared to 32.5 mg in the control group. By the 5th day the average weight of brown fat in the cortisone group (63.3 mg) is twice that of the control group (32.0 mg), and 24 hours later has begun to decline sharply (41.5 mg), approaching the level of the control group (31.0 mg). Histologic sections show a marked hypertrophy of lipid locules in the brown fat cells of the cortisone-treated bats, especially in the lobes of animals sacrificed 5 days after receiving the hormone (Fig. 3). Conversely, sections of brown fat from control bats show very little evidence of lipid in the dense cytoplasm (Fig. 4).

Discussion. Marked variations have been observed in the lipid content of the brown adipose tissue of bats during the reproductive cycle. As pregnancy progressed, there was an accumulation of fat droplets which increased in amount in the late period and decreased during and shortly after parturition. Similar results have been reported on utilization of the interscapular brown fat of gravid mice. Rothbard(8) noted an hypertrophy of brown fat in pregnant mice shortly before term and a rapid depletion of lipid in the fat cells just prior to and at time of delivery. In contrast, Teodoru and Grishman(9) observed a gradual depletion of lipid accompanied by weight reduction of the brown fat of hamsters as the gestation period advanced.

That the gravid period is accompanied by profound hormonal alterations, especially in regard to adrenal function, has been estab-

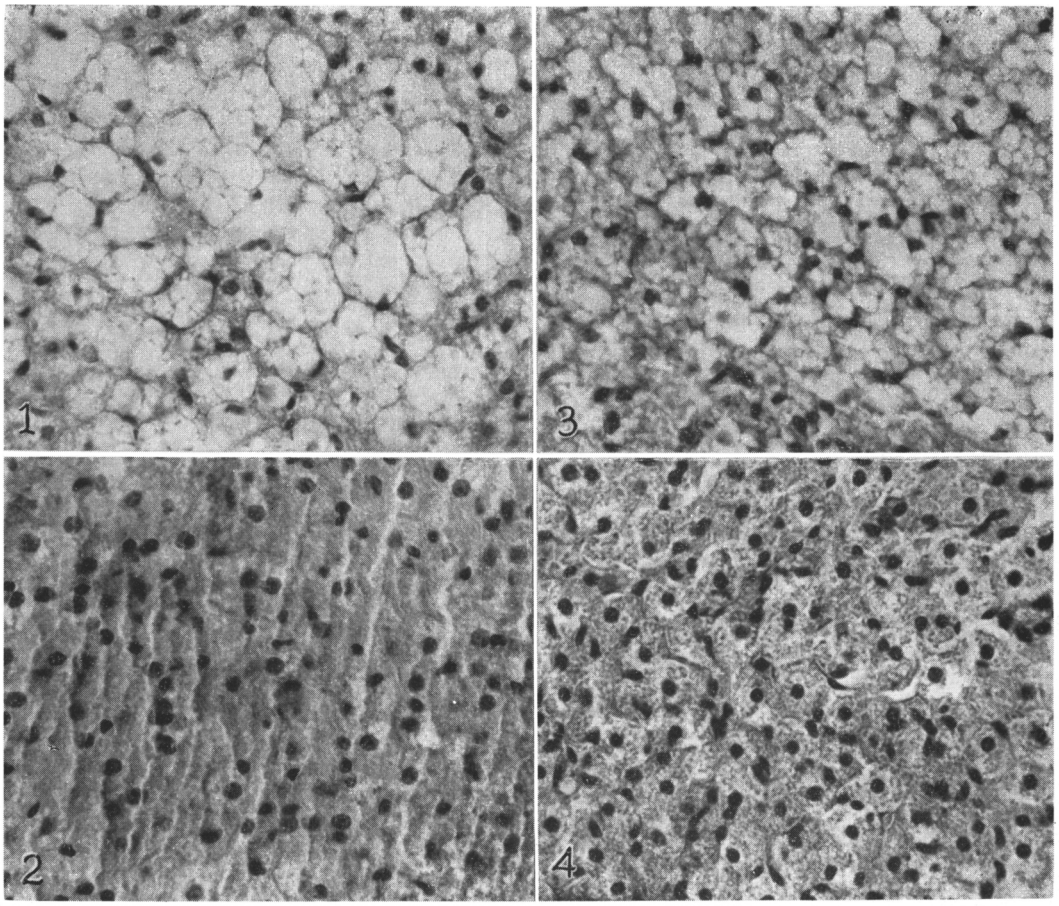

FIG. 1. Representative H & E section of brown fat of gravid bats (late in gestation period). Note large, coalesced, lipid locules in the cytoplasm and displacement of nuclei toward periphery of the cells. ×600.

FIG. 2. Representative section of brown fat of bats sacrificed shortly after parturition. Cells are deplete of lipid. ×600.

FIG. 3. Representative section of brown fat of bats sacrificed 5 days following cortisone treatment. Note increased lipid accumulation similar to Fig. 1. ×600.

FIG. 4. Representative section of brown fat of control bats which received no cortisone. ×600.

lished. The adrenal gland is greatly enlarged, reaching a maximum late in the gestation period in man and many animals, resulting in increased cortical output(10-12). Fawcett and Jones(13) have recorded definite association between adrenal glands and brown fat of mice; diminished adrenocortical function was followed by a depletion of lipid in brown fat, whereas injections of cortical hormones prevented depletion, suggesting that maintenance of the normal complement of lipid in this tissue is directed by the functional integrity of the adrenal cortex. In this regard, cortisone has been shown to induce increased lipid deposition in the brown fat of hamsters and mice(5) and of rats(6). In the present study, the brown fat of cortisone-treated nongravid bats showed a marked increase in lipid deposition, similar to that observed in the brown fat of bats in late pregnancy. Furthermore, cortisone was shown to have a definite though short-lived effect on the weight of brown fat; a significant increase in weight of this tissue in cortisone-treated animals paralleled lipid accumulation.

Thus, it appears that the lipid content of the interscapular brown adipose tissue of *Tadarida b. mexicana* is highly mobile, sub-

ject to profound fluctuations during the gestation period and also in response to cortisone treatment. Although very little is known concerning hormonal function in bats during the reproductive cycle, it is suggested that adrenal activity may have played an important role in directing the observed alterations in lipid content of the brown fat of gravid bats.

Summary. The interscapular brown adipose tissue of the bat (*Tadarida b. mexicana*) undergoes significant fluctuations in lipid content during the reproductive cycle. There is a progressive accumulation of lipid in the brown fat cells, beginning in early pregnancy and reaching a maximum level late in the gestation period. In contrast, during and shortly after parturition the brown adipose tissue is essentially deplete of lipid, while 2 weeks following delivery lipid can again be found in the brown fat cells. Cortisone treatment of non-gravid bats results in increased lipid accumulation in the brown fat similar to that observed in the pregnant animal. A gradual increase in weight of brown fat parallels lipid hypertrophy. The role of adrenal activity in directing alterations in the lipid content of the interscapular brown adipose tissue of bats during the reproductive cycle is discussed.

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Localization of Endotoxin in the Walls of the Peripheral Vascular System During Lethal Endotoxemia.* (27824)

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The principal sign of lethal endotoxemia is peripheral vascular collapse. Routine pathologic methods, however, have failed to demonstrate a lesion and provide an explanation for the shock. We are inquiring whether endotoxin is localized in blood vessel walls where it could exert a direct toxic effect and yet be undetected by conventional methods.

Studies using radioactively labeled endotoxin have not permitted precise localization and have been difficult to interpret. For example, Barnes, Lupfer, and Henry(1) studied in rats the fate of intravenously administered I^{131}-labeled *Sh. flexneri* endotoxin and found most of the radioactivity in the liver. They inferred that the hepatic cell was the biochemical target of endotoxin activity. Braude, Carey and Zelesky(2) in a similar study in rabbits with Cr_{51}-labeled *E. coli* endotoxin

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