

of the hemagglutinating factor upon erythrocytes of the burned host are not clear. They may, however, provide an experimental approach to the study of hematologic complications of the burn syndrome.

Summary. Thermal injury is associated in the rat with the development of an agglutinin directed against syngeneic erythrocytes. This hemagglutinin is present in the *lymphatic* but *not* in the *blood* circulation of the burned host, and has the characteristics of an immunoglobulin.

1. Holman, E., Surg. Gyn. & Obst., 1924, v38, 100.
2. Kay, G. D., Ann. N. Y. Acad. Sci., 1957, v64, 767.
3. Rapaport, F. T., Converse, J. M., Horn, L., Ballantyne, D. L., Mulholland, J. H., Ann. Surg., 1964, v159, 390.
4. Rapaport, F. T., Converse, J. M., Proc. 2nd International Conference on Burns Research, Univ.

of Edinburgh, 1965, in press.

5. Atherton, S., Merrill, N., McCarthy, M. D., Fed. Proc., 1960, v19, 195.
6. McCarthy, M. D., McCarthy, M. D., Ann. Surg., 1963, v157, 446.
7. ———, Proc. Soc. Exp. Biol. and Med., 1964, v117, 693.
8. Horn, L., Malm, O. J., Report to the Committee on Shock and Trauma, Nat. Acad. Science, Nat. Research Council, 1958.
9. Bollman, J. L., Cain, J. C., Grindlay, J. H., J. Lab. Clin. Med., 1948, v33, 1349.
10. Gowans, J. L., Brit. J. Exp. Pathol., 1957, v38, 67.
11. Deutsch, H. F., Morton, J. I., Science, 1957, v125, 600.
12. Scheidegger, J. J., Int. Arch. Allergy & Appl. Immunol., 1955, v7, 103.
13. Milgrom, F., Wozniczko, G., Dudziak, Z., Schweiz Z. f. Alleg. Path. u. Bakt., 1957, v20, 373.

Received August 25, 1966. P.S.E.B.M., 1966, v123.

Elongation of the Interpubic Ligament in the Little Brown Bat (*Myotis lucifugus*).* (31642)

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Separation of the pubic symphysis during late pregnancy and after treatment with relaxin in an estrogenized animal has been described in only 5 species: the guinea pig(1), the mouse (*Mus musculus*)(2), the deer-mouse (*Peromyscus maniculatus* sp. *bairdii* and *Peromyscus maniculatus* sp. *gracilis*)(3), and the Skomer bank vole (*Clethrionomys skomerensis*)(4). This phenomenon occurs also in the monkey (*Macaca mulatta*) although it differs from the above species in requiring pretreatment with progesterone in addition to estrogen(5,6).

The present study is concerned with the elongation of the pubic ligament of the little brown bat (*Myotis lucifugus*) during pregnancy and after treatment with relaxin.

Materials and methods. Female little brown bats (*Myotis lucifugus*) were obtained from

Ray's Cave, Greene County, Indiana, and Grotto Cave, Monroe County, Indiana. These hibernating animals were collected in 2 groups, one group in late November, 1965, the second group in early April, 1966. The bats were maintained in the laboratory at 92°F, housed 2 per cage and fed 10-20 mealworms daily. A constant supply of water was available within the cages. Bats collected in April, 1966, that were thought to be pregnant were observed daily. Immediately after delivery or abortion the bats were killed and their pubic symphyses examined.

The non-pregnant bats were primed with a single dose of 5 µg estradiol cyclopentylpropionate (ECP) administered into the pectoral muscles. Seven days later the bats received a single injection of relaxin† in-

* Aided in part by grant HD-02068 from Nat. Inst. Health and in part by a grant from Purdue Research Foundation.

† Obtained as Releasin® through the courtesy of Dr. R. L. Kroc, Warner-Chilcott Laboratories, N. J. The preparation used (W1164A Lot 05339) had an activity of 180 guinea pig units (GPU) per mg.

TABLE I. Effect of Estradiol Cyclopentylpropionate (ECP), Relaxin and Pregnancy on Interpubic Ligament of the Little Brown Bat (*Myotis lucifugus*).

Treatment	Dose	No. of bats	Mean body wt, g	Mean pubic separation in mm \pm S.E.
Controls	—	4	7.3	.52 \pm .097
ECP	5 μ g	5	7.2	.78 \pm .114
ECP & Relaxin	5 μ g 5 GPU*	6	7.7	1.03 \pm .056
ECP & Relaxin	5 μ g 10 GPU	6	7.7	1.25 \pm .100
Pregnant at parturition	—	4	7.8	2.01 \pm .077

* Guinea pig units.

tramuscularly. The estrogen was dissolved in peanut oil and the relaxin in 1% Benzopurpurin 4B. A volume of 0.1 ml was used in both instances. Twenty-four hours after injection of the relaxin the bats were killed with ether. The abdominal cavity was opened and the pubic symphysis exposed. Excess tissue was dissected away and the length of interpubic ligament was measured by a calibrated ocular micrometer under a dissecting microscope(7). Non-treated, non-pregnant control bats and bats treated with 5 μ g estradiol cyclopentylpropionate only were examined in the same way.

Results. Microscopic examination of the interpubic ligaments of the various experimental bats indicated some gross differences in the ligament and adjacent pubes. The control bats showed a very narrow separation of the pubic symphysis with a short rigid ligament. The bats treated with estradiol cyclopentylpropionate only had a longer and wider ligament than the controls. The ligament was of a rigid consistency with a thickening in the center. Definite resorption of the medial ends of the pubes, especially at the symphyseal tips, was noted. The interpubic ligaments that were observed immediately after parturition were similar in gross structure to those of bats subjected to estrogen-relaxin treatment. In addition to pubic resorption a flexible ligament of greater length was observed. The ligament exhibited the characteristics of loose, thin, fibrous connective tissue in contrast to the dense fibrous connective tissue of the two control groups. The only grossly observable difference between the interpubic ligament of the pregnant bats and of the estrogen relaxin

treated bats was the greater length of the ligaments in the pregnant group.

The data obtained from the measurements of the length of the interpubic ligament indicate a significant increase from 0.52 mm to 2.01 mm at time of delivery (Table I). A slight increase in the non-pregnant animal to 0.78 mm was obtained with estrogen alone but the combination of estrogen and relaxin caused a highly significant increase to a mean length of 1.03 mm with 5 GPU of relaxin and 1.25 mm with 10 GPU of relaxin.

Discussion. These results indicate the elongation of the interpubic ligament during pregnancy in the little brown bat (*Myotis lucifugus*). It is highly likely that this phenomenon is under the hormonal control of estrogen and relaxin as described for other species(8). It is of interest to note that this phenomenon has been previously described for only 2 orders of mammals, *i.e.*, rodents and primates. The current results indicate that the Chiroptera may be added to the above two orders. However, this does not mean that all members of the order Chiroptera will show interpubic elongation. Indeed among the Rodentia, neither the rat nor hamster show elongation of interpubic ligament(4).

Zarrow and Wilson(4) state that the members of the Rodentia exhibiting this property have narrow pelvis due to ecological reasons. Hence, adaptative changes in the pelvic canal were necessary to insure delivery of the young. Similarly, the narrow pelvis of the bat can be regarded as an adaptation to flight and the corresponding changes in the birth canal may be viewed as a case of convergent evolution. It may be that the size of animal

in relation to the size of young is also a factor to be considered.

The present results demonstrate that estrogen alone is inadequate to produce a substantial pubic separation and that relaxin plays a necessary role in interpubic ligament formation. It can also be concluded that the action of relaxin and estrogen on the interpubic ligament is similar in this species to that described in the mouse. Hall(9), Crelin(10) and Steinetz *et al*(11) suggest that relaxin in an estrogen environment may activate the catabolic enzyme systems of the osteoclasts and chondrocytes, thus accounting for the resorption of bone and for the swelling and transformation of hyaline cartilage to fibrocartilage. From the gross microscopic observations of the bat pubic ligament, it can be tentatively concluded that the proposed mechanism of action of relaxin appears to explain the observed results in this species.

Summary. A significant elongation of the interpubic ligament occurs in the pregnant little brown bat (*Myotis lucifugus*). Examined immediately after delivery, the mean ligament length measured 2.01 as compared with 0.52 mm in the non-pregnant bat. Treatment of non-pregnant female bats with estradiol induced a mean pubic separation of 0.78 mm, while treatment with ECP and

relaxin induced a mean separation of 1.03 and 1.25 mm with 5 and 10 GPU of relaxin, respectively. It is concluded that separation of the pubic symphysis with the elongation of the interpubic ligament occurs during pregnancy in the little brown bat and after treatment with ECP and relaxin.

1. Hisaw, F. L., Proc. Soc. Exp. Biol. and Med., 1926, v23, 661.
2. Hall, K., Newton, W. H., J. Physiol., 1947, v106, 18.
3. Zarrow, M. X., Eleftheriou, B. E., Whitecotten, G. L., King, J. A., Gen. Comp. Endocrinol., 1961, v1, 386.
4. Zarrow, M. X., Wilson, E. D., J. Endocrinol., 1963, v28, 103.
5. Hartman, C. G., Strauss, W. L., Am. J. Obstet. & Gynec., 1939, v37, 498.
6. Hisaw, F. L., Jr., Hisaw, F. L., *ibid.*, 1964, v89, 141.
7. Steinetz, B. G., Beach, V. L., Kroc, R. L., Stasilli, N. R., Nussbaum, R. E., Nemith, P. J., Dun, R. K., Endocrinology, 1960, v67, 102.
8. Hisaw, F. L., Zarrow, M. X., Vit. and Horm., 1951, v8, 151.
9. Hall, K., J. Endocrinol., 1956, v13, 384.
10. Crelin, E. S., Anat. Rec., 1963, v146, 149.
11. Steinetz, B. G., Manning, J. P., Butler, M., Beach, V., Endocrinology, 1965, v76, 876.

Received August 25, 1966. P.S.E.B.M., 1966, v123.

Influence of *E. coli* Endotoxin on Serotonin Contractions of the Rabbit Aortic Strip.* (31643)

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The mechanisms by which bacterial endotoxins disrupt peripheral vascular function remain largely conjectural(1). Among the explanations advanced for the shock-like

collapse of the individual following endotoxin are a release of mediators such as histamine, serotonin, catecholamines, etc.(1), a direct biphasic action on vascular smooth muscle(2), an increased responsiveness to catecholamines(3), and a depressing action on myocardial muscle(2). Data based on *in vitro* experiments to support a direct action of bacterial endotoxins on smooth muscle are for the most part inconclusive(1).

The present report describes experiments with isolated strips of aortic smooth muscle

* This work was supported in part by a grant from Office of the Surgeon General, Dept. of the Army, Contract DA-49-007-MD-993.

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