TABLE I.
Comparison of Systolic Blood Pressure Readings by 3 Methods in Heated Normal Rats and in Priscol Treated Rats Without Heating.

| | Rats heated 10 min. at 40°-41°C | | | | Priscol treated | | |
|---|---|---|---|---|---|---|---|
| Rat | Plethysmograph | Ammeter | Headphones | | Plethysmograph | Ammeter | Headphones |
| 1 | 108 | 112 | | * | 90 | | 104 |
| | 112 | 110 | | | 90 | | 94 |
| | 112 | 114 | | | 88 | | 92 |
| | 104 | | 114 | | 90 | 110 | |
| | 106 | | 108 | | 100 | 106 | |
| | 110 | | 116 | | 98 | 102 | |
| 2 | 98 | | 102 | * | 100 | 112 | |
| | 98 | | 98 | | 98 | 104 | |
| | 96 | | 96 | | 90 | 108 | |
| | 90 | 90 | | | 96 | | 102 |
| | 90 | 90 | | | 94 | | 102 |
| | 88 | 88 | | | 90 | | 98 |
| 3 | 116 | | 122 | † | 70 | | 88 |
| | 114 | | 118 | | 78 | | 92 |
| | 114 | | 120 | | 84 | | 104 |
| | 118 | 122 | | | 90 | 94 | |
| | 116 | 116 | | | 86 | 96 | |
| | 112 | 116 | | | 82 | 90 | |
| 4 | 84 | 88 | | † | 80 | 94 | |
| | 82 | 84 | | | 90 | 90 | |
| | 84 | 84 | | | 86 | 92 | |
| | 78 | | 82 | | 82 | | 96 |
| | 80 | | 84 | | 84 | | 96 |
| | 78 | | 82 | | 80 | | 94 |

* 3 mg/kg Priscol I.M. † 4 mg/kg Priscol I.M.

plethysmograph. This greater sensitivity was observed during the periods of minimum vasodilatation before and after the maximum effect. Frequently readings taken by the ammeter circuit and the acoustical indicator coincided. However, the acoustical indicator gave readings that averaged 1.25 mm Hg above those of the ammeter circuit when a series of 100 readings by each device was compared.

*Conclusion.* The acoustical modification of the Skeggs and Leonards apparatus described facilitates determination of the blood pressure of rats but can be used only after vasodilatation produced by heat or drugs. Priscol produces vasodilatation sufficient for repeated blood pressure determination during a 9- or 10-hour period.

We wish to express appreciation to A. A. Foster for directing our attention to the electrical circuit herein described and to L. T. Skeggs and J. R. Leonards for one of their electrical end point devices.

## 16132
## Relaxin Content of Blood, Urine and Other Tissues of Pregnant and Postpartum Guinea Pigs.*

M. X. ZARROW.† (Introduced by Frederick L. Hisaw.)
*From the Biological Laboratories, Harvard University, Cambridge, Mass.*

Relaxin was first detected in the blood of pregnant rabbits and guinea pigs (Hisaw[1]) and was subsequently found in the blood of pregnant sows, dogs, cats, mares and women (Hisaw;[2] Brouha and Simonnet;[3] Pommerenke;[4] Abramson, Hurwitt and Lesnick[5]). Separation of the pubic bones which occurs

* Aided in part by a grant from the Donner Foundation to Prof. Frederick L. Hisaw.

† Present address, Department of Pharmacology, Yale University School of Medicine, New Haven, Conn.

1 Hisaw, F. L., PROC. SOC. EXP. BIOL. AND MED., 1926, **23**, 661.

during the normal gestation in the guinea pig has been described by a number of investigators and this reaction, as induced experimentally by relaxin, has been used extensively in the assay of this hormone. Nevertheless, except for the rabbit (Marder and Money[6]) no attempt has been made to determine the amount of relaxin present throughout the various stages of pregnancy. Therefore, it was felt desirable to study quantitatively the concentration of relaxin in the blood, urine, uterus and placenta of guinea pigs at different times of pregnancy and after delivery of the young.

*Methods.* Adult female guinea pigs weighing from 600 to 800 g were mated and pregnancy dated from the morning on which the presence of vaginal plugs was noted. The average length of pregnancy was considered to be 68 days (Young and Blandau[7]). Blood was obtained from the animals by cardiac puncture at definite intervals after mating and the blood serum from a group of guinea pigs of a similar length of pregnancy pooled in order to obtain a sufficient quantity to carry out relaxin determinations. Urine samples were secured from a number of guinea pigs for a 24-hour period prior to obtaining the blood and dialyzed against running tap water. Placentæ and uteri were removed from animals on the 56th and 63rd day of pregnancy, ground up and extracted twice with 3% HCl. The extracts were kept in the cold at a pH of approximately 0.5 and prior to injection were brought to pH 7 with 10% NaOH.

The assays for relaxin content were performed according to the method of Abramowitz *et al.*[8] except that the guinea pigs were pretreated with one $\mu$g of estradiol daily for 3 days. Each sample was tested at several dilutions in order to abtain a concentration that gave a 67% response. The guinea pigs used for the relaxin determinations were part of a large colony maintained solely for assay purposes. In addition to the animals employed in the assay a total of 80 guinea pigs were used in this investigation, 57 for the study of pregnancy and 23 following parturition. The relaxin content of the blood serum was determined at 15, 21, 28, 35, 42, 49, 56, and 63 days after mating and at 2, 6, 24, and 48 hours postpartum. Twenty-four-hour samples of urine were obtained 42, 56, and 63 days after mating and the uterine and placental tissues were removed at 56 and 63 days of pregnancy.

*Results.* Relaxin assays on the 15th day of pregnancy were negative at a level of 10 ml of serum. However, a positive response was obtained by the 21st day indicating approximately 0.25 G.P. units per ml. (Fig. 1). Between the 21st day and the 28th day the concentration of relaxin was doubled, reaching a maximum value of 0.5 G.P. units per ml of serum. This level was maintained for 4 weeks and was followed by a drop to 0.33 G.P. units per ml by the 63rd day of pregnancy. Shortly after parturition a second and more precipitous decrease took place so that in the 48-hour postpartum guinea pig relaxin was no longer detectable in 10 ml of blood serum.

The urine showed an activity of 0.5 G.P. units of relaxin per ml at 42 and 56 days of pregnancy, and approximately 0.25 G.P. units per ml of urine on the 63rd day. It is of interest to note that the drop in the relaxin concentration of the urine occurred at the same time as that in the blood serum. The total volume of urine for a 24-hour period varied from 10 to 20 ml so that the guinea pigs excreted in their urine from 5 to 10 G.P. units of relaxin daily between the 28th and 56th days of pregnancy.

Approximately 10 G.P. units of relaxin per gram of fresh tissue was found in the uterus of the guinea pigs on the 56th day of pregnancy whereas the placenta showed a

2 Hisaw, F. L., *Physiol. Zool.*, 1929, **2**, 59.

3 Brouha, L., and Simonnet, H., *Compt. rend. Soc. de biol.*, 1928, **99**, 1769.

4 Pommerenke, W. T., *Am. J. Obst. and Gynecol.*, 1934, **27**, 708.

5 Abramson, D., Hurwitt, D., and Lesnick, G., *Surg. Gynec. and Obst.*, 1937, **65**, 335.

6 Marder, S. N., and Money, W. L., *Endocrinol.*, 1944, **34**, 115.

7 Young, W. C., and Blandau, R. J., *Science*, 1936, **84**, 270.

8 Abramowitz, A. A., Money, W. L., Zarrow, M. X., Talmage, R. V. N., Kleinholz, L. H., and Hisaw, F. L., *Endocrinol.*, 1944, **34**, 103.

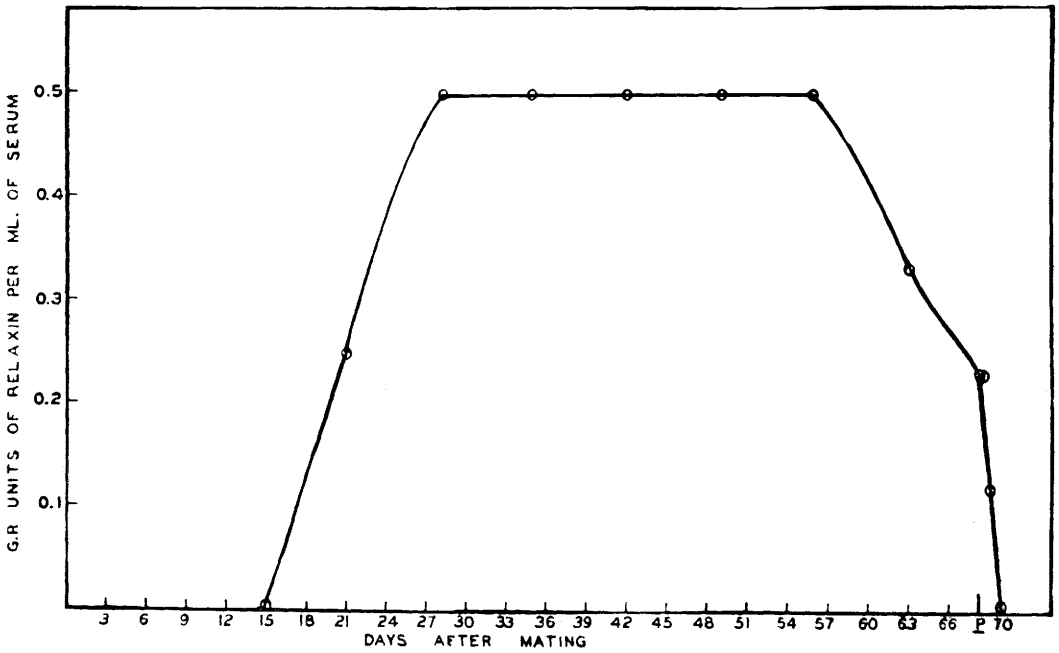


FIG. 1.

The relaxin content of the blood serum of pregnant and postpartum guinea pigs. Parturition (P) occurred 68 days after mating.

relaxin content of 5 G.P. units per gram. On the 63rd day of pregnancy the uterus still contained 10 G.P. units of relaxin per gram of fresh tissue while the placenta had only 2.5 G.P. units per gram.

*Discussion.* A series of 20 guinea pigs were palpated daily starting with the first day of pregnancy in order to determine the time at which relaxation of the symphysis pubis normally occurred. Relaxation was first detected on about the 20th day (range, 18 to 25 days), which compared favorably with the time at which relaxin first appears in the blood of the pregnant animals (Fig. 1.) It should be further pointed out that these two events may also be correlated with the time during which the placenta undergoes its most rapid development (Marshall[9]). There is a similar correlation in the rabbit between the development of the placenta and the appearance of relaxin in the blood (Marder and Money[6]). The maximum concentration of relaxin found in the blood of the pregnant guinea pig was 0.5 G.P. units per ml of serum which is one-twentieth the concentration found in the pregnant rabbit.

The fall in the relaxin content of the blood serum of the guinea pig previous to parturition is significantly different from the situation in the pregnant rabbit. In the rabbit the relaxin concentration is maintained at a plateau until parturition, at which time a precipitous drop occurs (Marder and Money[6]). However, there is some evidence which indicates that the situation in the pregnant woman may be similar to that in the guinea pig. It has been found that the amount of relaxin in the blood serum of pregnant women is considerably greater during the first half of pregnancy than during the 8th month (Pommerenke,[4] Abramson, Hurwitt, and Lesnick[5]). Thus, it would appear that in both the guinea pig and woman, unlike the rabbit, changes take place prior to parturition resulting in a decreased production of relaxin.

From the results obtained on the relaxin content of the uterus and placenta of the guinea pig at 56 and 63 days of pregnancy it would appear that the drop in the relaxin concentration of the blood serum may be due to a decreased output by the placenta. However it is not possible to account for this

9 Marshall, F. H. A., 1922, The Physiology of Reproduction. Longmans, Green and Co., London.

with certainty and the problem is further complicated by the fact that a species difference may exist with regard to the part of the female reproductive tract that may serve as a source for relaxin.

*Summary.* Relaxin is found in the blood of guinea pigs on about the 21st day of pregnancy, at which time relaxation of the symphysis pubis may first be detected by palpation. The concentration of relaxin reaches a peak of 0.5 G.P. units per ml of blood serum on the 28th day and is maintained until the 63rd day when the concentration falls to 0.33 G.P. units per ml. Immediately after parturition a precipitous decline of the relaxin content of the blood serum takes place. The urine shows a concentration of 0.5 G.P. units of relaxin per ml at 42 and 56 days of pregnancy with a drop to 0.25 G.P. units by the 63rd day. The uterus contains about 10 G.P. units per gram of tissue on the 56th and 63rd day of pregnancy whereas the placenta shows 5 G.P. units per gram of tissue on the 56th day and 2.5 G.P. units on the 63rd day. It seems probable that the drop in serum relaxin in the latter part of pregnancy may be due to a decreased output from the placenta.

## 16133 P

## Enhancement of Effect of Chorionic Gonadotrophin on Ovarian Hyperemia in the Rat by Pituitary Extract.

A. E. RAKOFF AND PAUL H. FRIED.*

*From the Department of Obstetrics and Gynecology, and the Hospital Laboratories, Jefferson Medical College, Philadelphia.*

The earliest demonstrable effect of chorionic gonadotrophin on the ovary of rats is the appearance of hyperemia.[1] This reaction has been used with varying degrees of success for the rapid diagnosis of pregnancy. Attempts to enhance the effect of chorionic gonadotrophin on ovarian hyperemia led to our present use of a pituitary extract for this purpose. This was based on the well-known synergistic effect of certain anterior pituitary extracts in augmenting the ovarian response when used in conjunction with chorionic gonadotrophin.[2] The effective component of such pituitary extracts is not known. The earlier claims[3] that such "synergists" represent a distinct gonadotrophin no longer seems likely in view of later work[4] indicating that similar effects can be obtained with follicle-stimulating gonadotrophins (F.S.H.). The pituitary synergist† employed in these studies is reported to be chiefly F.S.H., with traces of luteinizing principle.[5] Prolactin was not demonstrable.

*Methods and Results.* The 28- to 32-day-old, 50 to 60 g immature albino female rats used were of a mixed strain of unknown identity. Chorionic gonadotrophin, the pitui-

* Schering Fellow in Gynecologic and Obstetric Endocrinology.

1 Zondek, B., and Sulman, F., *Vitamins and Hormones*, 1945, vol. 3, p. 328, Academic Press, N. Y.

2 a. Evans, H. M., Meyer, K., and Simpson, M. E., PROC. SOC. EXP. BIOL. AND MED., 1931, **28**, 845.
b. Evans, H. M., Meyer, K., and Simpson, M. E., *Am. J. Physiol.*, 1932, **100**, 141.
c. Fevold, H. L., Hisaw, F. L., Hellbaum, A. A., and Hertz, R., *Am. J. Physiol.*, 1933, **104**, 710.
d. Fevold, H. L., Hisaw, F. L., and Hertz, R., PROC. SOC. EXP. BIOL. AND MED., 1933, **30**, 914.
e. Leonard, S. L., PROC. SOC. EXP. BIOL. AND MED., 1932, **30**, 403.
f. Leonard, S. L., and Smith, P. E., *Am. J. Physiol.*, 1934, **108**, 22.
g. Smith, P. E., and Leonard, S. L., PROC. SOC. EXP. BIOL. AND MED., 1933, **30**, 1248.
h. Collip, J. B., Selye, H., Anderson, E. M., Thomson, D. L., *J.A.M.A.*, 1933, **101**, 1553.

3 a. Evans, H. M., Simpson, M. E., and Austin, P. R., *J. Exp. Med.*, 1933, **57**, 897.
b. Evans, H. M., Simpson, M. E., and Austin, P. R., *J. Exp. Med.*, 1933, **58**, 545.
c. Evans, H. M., Pencharz, R. I., and Simpson, M. E., *Endocrinology*, 1934, **18**, 601, 607.