

Rat Anterior Pituitary Nucleic Acid Levels as Function of Age.* (25919)

MELVIN HESS

(With technical assistance of James Bird)

Dept. of Anatomy, University of Pittsburgh School of Medicine, Pa.

During a study of rat anterior pituitary gland nucleic acid levels following adrenalectomy(1), the content of ribonucleic acid (RNA) for control animals varied from 8-12 $\mu\text{g}/\text{mg}$ of tissue. Since animals were not all of the same age, this variability might have been due to pooling of data obtained from rats of different ages. Secondly, the manner by which animals were sacrificed might be a factor influencing nucleic acid levels if RNA content is subject to change with stress. Therefore, this investigation was designed to study RNA content of anterior pituitary tissue of rats of specified age groups, sacrificed by either decapitation or an overdose of ether. In addition, water and collagen contents of the tissue were measured to note their levels as a function of age since fluctuations in these 2 constituents can influence RNA concentration.

Methods. Male rats of Holtzman strain were obtained at various ages: Group I—35-37 days old, Group II—51-59 days old, Group III—70-72 days old, Group IV—93-94 days old, Group V—128-132 days old, and Group VI—one year old. They were maintained on Purina Laboratory Chow pellets and tap water *ad lib.* in temperature-controlled animal quarters. Animals were sacrificed by decapitation or overdose of ether, pituitary glands removed, posterior lobe tissue discarded, and anterior lobes weighed on torsion balance. Tissue collected from each rat was homogenized in cold normal saline to make a 0.6% homogenate. Determination of RNA content from pituitary homogenates followed procedure described by Schneider(2). Water content of pituitaries from each age group was determined by weighing anterior lobe tissue before and after heating to dryness in oven at 100°C until a constant minimum weight was

TABLE I. Anterior Pituitary RNA Content of Various Age Groups Killed by Decapitation or by Overdose of Ether.

Age group	Ant. pituitary RNA content ($\mu\text{g}/\text{mg}$ tissue)	
	Decapitation	Ether
35- 37 days old		$7.1 \pm .2^* (38)$
51- 59	$6.4 \pm .2^* (24)$	$8.8 \pm .4 (24)$
70- 72	$8.5 \pm .6 (10)$	$7.9 \pm .4 (9)$
93- 94	$6.5 \pm .2 (10)$	$6.8 \pm .4 (6)$
128-132	$6.7 \pm .3 (16)$	$7.1 \pm .1 (6)$
1 yr old		$7.4 \pm .6 (17)$

* Stand. error. No. of rats in parentheses.

† Difference from decapitated groups II, IV and V statistically significant ("t" test for significance).

‡ Difference from ether groups I, IV and V and decapitated group II statistically significant.

obtained. A chemical analysis for hydroxyproline was performed† on dried pituitary tissue as an estimate of collagen content(3).

Results. Data obtained for RNA content of anterior pituitary tissue of rats of various ages are summarized in Table I. For the most part, the concentration of RNA in anterior lobe of pituitary gland appears fairly constant.

In the series utilizing decapitation as means of sacrifice, there are no significant differences between Groups II, IV, and V. The figure obtained for rats in Group III killed by decapitation is, however, significantly higher than the remainder of the series.

Among the animals sacrificed by an overdose of ether, only those in Group II demonstrate a significant difference from most other groups. Likewise, it is only this group that shows any difference in RNA content dependent upon manner of sacrifice. Decapitated rats of Group II had $6.4 \pm 0.2 \mu\text{g}$ of RNA/mg of tissue whereas the same age group subjected to ether overdosage had $8.8 \pm 0.4 \mu\text{g}$ of RNA.

The water content of pituitary tissue from

* This investigation supported by research grant from Nat. Inst. of Arthritis, P.H.S., and by Arthritis and Rheumatism Fn.

† The hydroxyproline determinations were performed by Dr. T. B. Rosenthal of this department.

different age groups varied within a narrow range. Samples of tissue lost from 68-76% of wet weights following drying. Likewise, amount of collagen in tissue, calculated from hydroxyproline values, did not vary significantly for various age groups. Amount of collagen in tissue samples of all age groups studied was between 0.5 and 1.5%.

Discussion. The content of RNA in anterior pituitary tissue does not vary markedly from one age group to another. Higher values obtained for rats of Group III sacrificed by decapitation cannot be explained by any alteration in experimental procedure. For most age groups studied, there are no significant differences in RNA values obtained after sacrificing rats by either of the 2 methods. Stress produced by subjecting animals to ether is probably of too short a duration to influence nucleic acid changes. No explanation can be offered to account for the higher RNA values observed in Group II rats sacrificed by ether overdosage compared to the same age group sacrificed by decapitation.

Tissue content of RNA is not influenced by variation of degree of hydration or collagenous connective tissue infiltration from one age group to another. There are no differences in these 2 constituents among various

age groups. Use of hydroxyproline determinations as index of amount of collagen present is reliable. Only collagenous connective tissue contains this amino acid in any appreciable amount(4).

Summary. A study was made of RNA, water, and collagen contents of adeno-hypophyses of rats of various age groups from 35 days to one year old, sacrificed by decapitation or an overdose of ether. There were no significant differences in level of RNA in tissue from one age group to another. Concentration of anterior pituitary nucleic acid was not influenced by manner of sacrifice for most age groups studied. Contents of water and collagen in the tissue were similar in young and old rats.

1. Hess, M., Corrigan, J. J., Jr., Hodak, J. A., *PROC. SOC. EXP. BIOL. AND MED.*, 1959, v100, 763.

2. Schneider, W. C., *Methods in Enzymology*, Vol. III, p680, Edited by S. P. Colowick and N. O. Kaplan, Academic Press, 1957.

3. Stegemann, H., *Hoppe-Seyler's Z. f. physiol. Chemie*, 1958, v311, 41.

4. Tristram, G. R., *The Proteins*, Vol. I, Part A, p222, Edited by H. Neurath and K. Bailey, Academic Press, 1953.

Received June 6, 1960. P.S.E.B.M., 1960, v104.

Dihydrotachysterol (AT 10) and Mammogenesis in Ovary-Thyroid-Parathyroidectomized Rats.* (25920)

R. VON BERSWORDT-WALLRABE[†] AND C. W. TURNER
Dept. of Dairy Husbandry, University of Missouri, Columbia

Continuing study of mammogenesis in ovary-thyroid-parathyroidectomized female rats, influence of estradiol benzoate (EB) and progesterone (P) with thyroxine (T₄) and dihydrotachysterol (AT 10) were compared.

Materials and methods. Sprague-Dawley-Rolfsmeyer rats were ovary-thyroid-parathy-

roidectomized, injected, killed, and DNA of mammary glands determined under standardized conditions described(1). AT 10[‡] was given every second day by stomach tube. Completeness of thyroidectomy checked by I¹³¹ uptake described(1). Group I received 1 µg EB plus 3 mg P. Group II received in addition 20 µg/100 g AT 10. Group III re-

* Contribution from Mo. Agr. Exp. Sta. Journal Series No. 2167. Approved by Director.

[†] Research Fellow, Dept. of Animal Physiology and Animal Nutrition, Univ. of Goettingen, West Germany. Present address: Univ. of Goettingen.

[‡] Kindly supplied by Sterling-Winthrop Research Inst., Rensselaer, N. Y. The commercial solution of AT 10 or dihydrotachysterol (Hytakerol) contains about 1250 µg of active principle per ml.