the activity in the maternal plasma was approximately the same as that on day 21. Similar results were obtained after injection of radioactive estradiol-17 $\beta$  (figure, B). As we did not directly measure radioactive corticosterone and estradiol-17 $\beta$  in the plasma, but measured total radioactivity, it is not possible to say to what extent the injected radioactive steroids had been metabolized in the mother and/or the placenta. Nevertheless, the essential point is that the placental transfer of steroids declines toward the end of gestation in the rat. It has been shown that the plasma estrogen concentration in pregnant rats is elevated markedly on day 21 as compared with that on day 16 of gestation 12. Therefore, there may be an explanation that, because of the rise of plasma estrogen concentration toward the end of gestation, some dilution of the injected labeled steroid could occur, which would result in an apparent decrease of the transfer rate. However, data regarding the difference of plasma estrogen concentration between day 21 and day 22 of gestation are

still lacking and it is not known when the concentration begins to rise.

In relation to this, the plasma corticosterone concentration in pregnant rats on day 22 has been reported to be less than that on day 21 <sup>13</sup>. Therefore, the dilution of injected labelled steroid is unlikely, at least as regards corticosterone, although plasma estrogen should be assayed during these 2 days. On the contrary, there may be another possibility that, if the total amount of circulating corticosterone or estrogen including labelled steroids decreases from day 21 to day 22, the absolute amount transported to the fetus would decrease proportionately, resulting in the apparent decrease of the placental transfer rate.

- 12 H. B. Waynforth, G. S. Pope and Z. D. Hosking, J. Reprod. Fert. 28, 191 (1972).
- 13 A. Cohen, Hormone Metab. Res. 5, 66 (1973).

## Effects of adenohypophysectomy on cAMP levels in chick embryo thyroid, adrenal and spleen1

R. Ferrand, J.-F. Renaud and G. H. Le Douarin

Laboratoire de Physiologie animale et cellulaire de l'Université de Nantes, 38, Boulevard Michelet, Boite postale 1044, F-44037 Nantes-cédex (France), 4 July 1977

Summary. cAMP levels in thyroid and adrenal are higher in 17-day-old decapitated chick embryos than in controls, while spleen cAMP is not modified.

embryos.

In chick embryo, partial decapitation results in various defects which are, totally or partially, corrected by embryonic adenohypophysis grafts. Therefore, decapitation may be considered endocrinologically equivalent to adenohypophysectomy (see Betz² for review). Complete embryonic development of endocrine glands such as thyroid and adrenal³-5, and non-endocrine organs, e.g. spleen 6, is pars distalis-hormone-dependent. Our study was designed to investigate whether embryonic hypophysectomy affects the adenosine 3′,5′-cyclic monophosphate (cAMP) levels in these organs.

Material and methods. Chick embryos were decapitated at the 12–15-somite stage after removal of 5 ml albumen. In control series, the same amount of albumen was removed. The organs were studied at the 17th day. Organs were quickly taken out and homogenized in ice-cooled 6% trichloracetic acid. Pools of thyroids and adrenals (10–14 pairs) and individual spleens were frozen in liquid nitrogen

- 1 This work was supported by a grant from D. G. R. S. T.
- 2 T. W. Betz, Acta endocr. 79, suppl. 198 (1975).
- 3 N. W. Fugo, J. exp. Zool. 85, 271 (1940).
- 4 T. W. Betz, Gen. comp. Endocr. 9, 172 (1967).
- 5 J. F. Case, Ann. N. Y. Acad. Sci. 55, 147 (1952).
- 6 T. W. Betz, J. Embryol. exp. Morph. 24, 357 (1970).
- 7 A. G. Gilman, Proc. nat. Acad. Sci. USA 67, 305 (1970).
- 8 R. C. Thommes, R. L. Vieth and S. Levasseur, Gen. comp. Endocr. 31, 29 (1977).
- 9 P. M. Wise and B. E. Frye, J. exp. Zool. 185, 277 (1973).

cAMP values are expressed in pmoles per mg of protein as means  $\pm$  SE of duplicates from a number of determinations indicated in parentheses

	Thyroid	Adrenal	Spleen
Decapitated embryos Controls	a) 22.72±4.45 (8) 7.68±1.21 (7)	b) 18.18±3.17 (5) 8.60±1.22 (5)	11.30±2.73 (9) 10.76±1.96 (8)

a) and b) differ significantly from the control,  $\rm p < 0.01$  and 0.05 respectively.

and kept at -30 °C until assay. cAMP levels were measured by isotopic dilution method 7, using kits obtained from Boehringer Mannheim. Protein content was determined by the method of Lowry.

Results. As shown in the table, thyroid and adrenal cAMP levels were higher in operated embryos than in controls. In contrast, embryonic hypophysectomy did not modify spleen cAMP level.

Discussion. In chick embryo, the differentiation of thyroid and adrenal is not affected by hypophysectomy until about 11-12-day-stage and during the last days of this period hormonal secretions occur as shown for thyroxine by Thommes et al.8 and for corticosterone by Wise and Frye<sup>9</sup>. In further development, a functional adenohypophysial-target gland axis is required to allow complete differentiation and normal physiology of thyroid and adrenal. In differentiated state, it is generally accepted that the effects of thyrotropin and corticotropin on target glands are mediated through stimulation of the adenylcyclase-cAMP system, resulting in an increase of the cAMP level. So, it could have been expected that lack of 'first hormonal messenger' would result in a low cAMP level in thyroid and adrenal of decapitated chick embryos. Opposite results were obtained for both thyroid and adrenal. A hypothesis can be proposed to explain this discrepancy: high cAMP levels could be related to the incomplete state of development of these glands in 17-dayold decapitated embryos. Moreover, the question arises whether these high cAMP levels are responsible for the basal hormonal secretion by thyroid and adrenal in decapitated chick embryos, this secretion remaining at the level observed before the function of the glands is controlled by adenohypophysis. Work is in progress to check these hypothesis. Adenohypophysectomy enhances spleen growth and modifies histogenesis. It is likely that this effect results from an indirect mechanism in which corticoids are involved. This may explain that spleen cAMP level is not different in normal and decapitated