Effect of anaesthesia on growth hormone secretion in the domestic fowl (Gallus domesticus)

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Summary. Chloral hydrate, Equithesin, Althesin, Rompun, Sagatal and ether anaesthesia lowered the concentration of plasma growth hormone in immature cockerels. Handling and serial bleeding of the conscious controls had no effect on the growth hormone concentration.

To facilitate handling and bleeding procedures, anaesthetised animals are often used for physiological studies. However, in mammals it is well established that anaesthesia has a marked effect on many endocrine systems[1-5]. In particular the secretion of plasma growth hormone may be higher[6-8] or lower[8,9] than in conscious counterparts, depending on the species and anaesthetic used. In contrast, in birds very little is known of the effects of anaesthesia, and studies on growth hormone physiology have been hindered due to the lack of adequate assay systems capable of measuring its plasma concentrations. However, the recent development of a sensitive and specific radioimmunoassay for chicken growth hormone has initiated studies on growth hormone secretion in conscious and anaesthetised domestic fowl[10,11], although the influence of anaesthesia has not been assessed. The aim of the present communication was, therefore, to investigate the effects of a number of commonly used anaesthetics on plasma growth hormone levels in the domestic fowl.

Materials and methods. All the birds used in this study were 6-week-old cockerels (Thornber 909's). Birds were bled from the wing vein by venipuncture and 1–1.5 ml of blood taken immediately before, 30, 60 and 90 min after the i.m. injection of test substances. The anaesthetics used were Equithesin[12] 3 ml/kg b.wt, Sagatal (sodium pentobarbitone, May and Baker Ltd) 30 mg (0.5 ml)/kg, Rompun (2% 2-(2,6-xylidino)-5,6-dihydro-4H-1,3 thiazine hydrochloride, Bayer Agrochem Ltd) 4 ml/kg, Althesin (0.9% alphaxalone, 0.3% alphadolone acetate, Glaxo Laboratories Ltd) 6 ml/kg and Chloral hydrate (Chas. F. Thackray Ltd) 300 mg/kg. All anaesthetics except Althesin were injected in a total volume of 4 ml/kg and dilutions and control injections were made with 0.9% saline. The doses of the anaesthetics used were the minimum required to achieve anaesthesia throughout the period of study and in the case of Rompun and Althesin were considerably higher than those required in mammals. In a second experiment a group of cockerels were anaesthetized with diethyl ether vapour and maintained under anaesthesia for 80 min. A similar untreated group served as controls. 1 ml blood samples were taken before, 5, 10, 20, 40 and 80 min after treatment. The concentration of plasma growth hormone was determined by

Table 1. Effect of 5 anaesthetics on plasma growth hormone levels in 6-week-old cockerels

Treatment	Time after treatment (min) 30	60	90
Saline	120.3± 9.0 (5)	108.6± 4.4 (5)	125.3±11.4 (5)
Sagatal	45.4±10.6*+(5)	57.9±14.0*+(5)	55.0±12.2*+(5)
Equithesin	37.0± 7.5*+(5)	37.6± 8.3*+(5)	35.4± 5.7*+(5)
Rompun	36.8± 9.3*+(4)	22.5± 6.5*+(4)	27.2± 9.4*+(4)
Althesin	18.5± 6.3*+(4)	31.4±10.8*+(4)	28.8± 7.9*+(4)
Chloral Hydrate	24.1± 9.7*+(4)	19.4± 5.5*+(4)	19.8±10.5*+(4)

Figures refer to the plasma growth hormone concentration, expressed as a percentage mean ± SEM (number of cockerels) of the pretreatment level. *Significantly different from the saline controls. $p < 0.01$; +significantly different from the pretreatment levels, $p < 0.05$.

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Table 2. Effect of ether anaesthesia on the levels of plasma growth hormone in 6-week-old cockerels

Treatment	Time after treatment (min) 5	10	20	40	80
Saline	106.7 ± 14.8 (4)	102.2 ± 3.7 (4)	93.3 ± 12.6 (4)	107.9 ± 8.8 (4)	92.6 ± 23.5 (4)
Ether	22.4 ± 5.0**+ (4)	18.8 ± 1.8**+ (4)	23.3 ± 3.5**+ (4)	24.4 ± 7.1**+ (4)	20.0 ± 1.9*+ (4)

Figures refer to the plasma growth hormone concentration, expressed as a percentage mean ± SEM (number of cockerels) of the pre treatment level. Significantly different from the saline controls, *$p < 0.05$, **$p < 0.002$; significantly different from the pre-treatment levels, + $p < 0.001$.

radioimmunoassay[10] and the results expressed as a percentage of the pretreatment level because of variations in basal concentrations. Statistical differences were determined by Student's t-test.

Results and discussion. It can be seen from table 1 that injection and serial bleeding of conscious birds had very little effect on the levels of plasma growth hormone. Handling and bleeding has been shown to increase[13] or decrease[9] the plasma growth hormone levels in mammals although not in the rabbit[14]. In contrast, in this study anaesthesia resulted in a marked depression of the plasma growth hormone levels (table 1). The concentration of plasma growth hormone was significantly lower than the pretreatment levels in all anaesthetized birds 30, 60 and 90 min after treatment. Throughout the period of study the levels of plasma growth hormone in these birds were also significantly different from the conscious controls. There were no significant differences in the levels of plasma growth hormone in birds treated with different anaesthetics. The effects of Althesin, Rompun and Chloral hydrate on plasma growth hormone levels have not been reported in other animals. However, whereas sodium pentobarbitone is known to be a provocative stimulus of growth hormone secretion in the rat[8], Sagatal and Equithesin (sodium pentobarbitone based) had the opposite effect in these experiments. The effect of ether anaesthesia can be seen in table 2. The concentration of plasma growth hormone was significantly lower than the pretreatment levels in anaesthetized birds 5, 10, 20, 40 and 80 min after treatment. Moreover, serial bleeding of the control birds had no effect on the plasma growth hormone levels and were significantly higher than their anaesthetized counterparts. Similar results of ether anaesthesia have been seen in the rat and mouse[8, 9] although in primates ether treatment elevates the concentration of plasma growth hormone[6].

The results of this investigation clearly demonstrate that several commonly used anaesthetics have a profound and consistent effect on the secretion of plasma growth hormone. Furthermore, physiological studies in the domestic fowl may be affected by the use of anaesthetics.

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Mathematical model of pituitary thyrotropic function

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Summary. A nonlinear differential equation is used to develop a mathematical model describing the time course of thyrotropin (TSH) concentration in human plasma after thyroliberin (TRH) stimulation. The application of the model to real data shows that pituitary responsiveness to TRH is highest in euthyroidism, reduced in primary hypothyroidism, and lowest in hyperthyroidism.

The secretion by the anterior pituitary gland of thyrotropin (TSH) is stimulated by thyroliberin (TRH) originating from the hypothalamus, and is inhibited by triiodothyronine and thyroxine, which feed back from the thyroid gland. The thyrotropic secretory capacity of the pituitary is tested by the increase of plasma TSH after injection of TRH. The dynamics of TRH and TSH together with triiodothyronine and thyroxine in plasma can serve as data to formulate a mathematical model of thyrotropic secretion in man.

Origin of biological data. 20 healthy male and female volunteers (group A), 12 hyperthyroid patients (group B) and 7 patients with primary hypothyroidism (group C) were diagnosed by clinical signs and by a TRH test: About 8.00 a.m. samples of venous blood were withdrawn for concentration measurements of basal TSH in plasma (h_0), of total plasma triiodothyronine (T_3), of total plasma thyroxine (T_4) by radioimmune assay, and for estimation of relative serum binding capacity (R) for radioactive triiodothyronine by equilibrium distribution in presence of resin. Thereafter, at time t = 0, 400 µg of TRH (r_0) were injected i.v. as a bolus in order to stimulate TSH secretion. In general the reactive peak level of plasma TSH is reached between 20 and 35 min after TRH injection. Hence t = 20 min was usually chosen as a second time point to measure TSH in plasma h(t).

Mathematical description. The feedback inhibition of TSH in the thyrotropic cells of the pituitary is brought about by a moiety of triiodothyronine and of thyroxine, the intracellular concentration of which (x_3, x_4) is nearly equal to that of the free plasma fraction of T_3 and T_4 not bound to plasma proteins. The respective concentrations of the free fractions we call F_3 and F_4.

The bound fraction of T_3 is set equal to the bound fraction p of radioactive triiodothyronine of patient serum. A normal standard serum yielding a bound fraction of $p_0 = 0.7$ served as control for measurements of R. With $R = p/p_0$, $R < 1/p_0 = 1.42$, T_3 in ng/dl, and c as proportionality factor we define

$$x_3 = T_3 - pT_3 = T_3(1 - p_0R) \approx cF_3. \quad (1)$$

As T_4 binds reversibly to plasma proteins, mainly to thyroxine-binding globulin (TBG), the law of mass action is applicable as soon as equilibrium is obtained. With [TBG] and [TBG-T_4] standing for concentration of TBG and TBG-thyroxine complex respectively, and K_1 = const., we have

$$F_4 = \frac{K_1[\text{TBG-}T_4]}{[\text{TBG}]}.$$

Normally 99.97% of total plasma T_4 is bound to TBG. Therefore [TBG-T_4] is essentially equal to T_4. Furthermore free [TBG] is approximately proportional to $p = p_0R$. With [TBG] $\approx K_2p_0R$ we can write:

$$F_4 \approx \frac{K_1T_4}{K_2p_0R}.$$

With $K_3 = K_2p_0/K_1$ we define (T_4 in µg/dl):

$$x_4 = \frac{T_4}{R} \approx K_3F_4. \quad (2)$$