

SOMATOTROPHIC AND THYROID HORMONES AROUND THE ONSET OF LAY IN BROILER BREEDERS UNDER DIFFERENT CONDITIONS

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Somatotrophic and thyroid hormones were determined around the onset of reproduction in broiler breeders reared in two different housing systems [dark, close-sided house (CH) and conventional, open-sided house (OH)]. In both groups age-related changes were obvious for thyroxine (T₄), growth hormone (GH) and insulin-like growth factor (IGF-1); levels of T₄ decreased, especially between 24 and 28 weeks in both groups; concomitantly GH sharply increased over the same period. A transient peak in triiodothyronine (T₃) occurred between 25 and 27 weeks. The effect of housing was only present after the onset of lay. Between weeks 27–28 and the end of the period studied, the CH group showed higher levels of GH and T₃ but lower T₄ levels as compared to the OH group. A significant increase in GH after onset of lay, without any significant rise in T₃ or in IGF-I, could point to a relative insensitivity to high plasma GH levels. Changes at GH receptor level, together with an increased pituitary GH secretion and/or decreased GH turnover may be expected. This may indicate that hypothalamo-pituitary changes at the onset of lay not only imply changes of gonadotrophic cell function, but also other hormonal axes. The relatively decrease in T₄ without changes in T₃, may point to a decrease in the activity of the thyrotropic axis.

Key words: Hormones, broiler breeders, reproduction, environment, growth hormone, thyroid hormones

In the literature, age-related changes in endocrine parameters before and after the onset of lay have been described mainly for laying hens. Changes in LH around puberty were described by Sharp (1975), Williams and Sharp (1977), while for thyroid hormones these age-related changes were described by Senior

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(1974), Williams and Sharp (1977), Muray et al. (1980), Tanabe et al. (1981), and Decuypere et al. (1985).

Long-term studies on changes of hormones of the thyrotropic axis during the onset of egg-laying in a layer strain were realised by Kühn et al. (1982).

Although a somewhat older but extensive literature exists about the developmental changes of the above-mentioned hormones, also as indicators of sexual development in laying hens, little is known about these hormones in broiler breeder females.

Quantitative food restriction to control body weight gain has become the standard procedure to improve the reproduction efficiency of broiler breeders (Costa, 1981). However, a recent study has described the influence of food restriction on plasma concentrations of growth hormone (GH), insulin-like growth factor I (IGF-I), triiodothyronine (T₃) and thyroxine (T₄) in the period before sexual maturity in three groups of broiler breeders fed different quantities of food (Bruggeman et al., 1997, 1998, 1999). To our knowledge, no studies exist that describe both metabolic hormones as well as hormones of the reproductive axis in long-term studies spanning the prepubertal period and the onset as well as peak of lay.

In mammals, several studies indicate that food intake and its associated effects on metabolic rate may be the triggering mechanism for changes in reproductive function. Fluctuations in plasma hormones may provide a signal that link metabolic status to the activation of the reproduction system.

Since the degree of restriction in broiler breeders changes with approaching sexual maturity and further decreases as the birds come into lay, this may interfere with age-related changes in hormonal profiles.

Moreover, a common practice in tropical countries is sometimes changing from open, environmentally less controlled housing systems to an alternative with controlled ambient conditions as for temperature, humidity and light. This variable was taken in account to study and link the changing hormonal profiles with the differential effects of the housing conditions on body weight changes, age at start of laying and performance.

Materials and methods

The experiment was carried out between September and December 1996, during the springtime in South America, and lasted 15 weeks. As experimental unit a flock of broiler breeders selected for chest size (*musculus pectoralis*) and body weight was used. During the rearing period, the pullets were kept in a dark, close-sided house, with full control of light, temperature and air humidity. At 19 weeks of age, half of the population of 1600 females, mated with 160 males, was housed in a dark, close-sided house (CH) and the other half in a conventional,

open-sided house (OH). The CH consisted of a closed installation, excluding penetration of external light, with intensity, time of illumination, and temperature/humidity of air totally under control. The OH was composed of a conventional installation, with open screens at both lateral sides, where the birds were displayed to light and natural ventilation during the day and night. The feed and sanitary programs, as well as the amount of feed given were in accordance with the recommendation of the industry. The composition of the diets were 16% CP and 2850 kcal of ME/kg, and were made on the basis of corn and soy meal. All birds were fed daily around 8:00 a.m.

The photoperiod was 12 hours of light, until the 19th week of age of the birds. From that week the photoperiod was increased until 17 hours of light and 7 hours of dark were attained at 29 weeks of age, in both environments. In the OH birds were raised in a period of crescent natural light (spring season in the southern hemisphere). For CH the light intensity was kept around 22–25 lumens/square metres, measured at the level of the birds' head. The temperature was $\pm 22^{\circ}\text{C}$ and the relative air humidity varied between 60 and 80%. For OH, light was provided until 17 hours of light a day, at the same intensity as in CH; temperature and humidity were closely correlated with the outside environment.

Blood samples from 20 females were collected weekly in both groups, around 10:00 am, from 19 until 33 weeks of age. Blood was taken from the brachial vein with a heparinised syringe. After centrifugation, the plasma was kept frozen at -20°C until analysis. Individual body weight of each female was recorded.

Triiodothyronine (T_3) analysis was performed by radioimmunoassay (RIA), using a commercially available antibody for T_3 (Mallinckrodt Diagnostica, Dietzenbach, Germany) in combination with a specific tracer (Amersham International, Slough, England), following the methodology proposed by Huybrechts et al. (1989). The intra-assay coefficient of variation was 3.2%. Thyroxine concentrations (T_4) were analysed by RIA using I^{125} , iodine from Amersham International, Slough, England, and antibody against T_4 , produced in rabbit from Mallinckrodt Diagnostica, Dietzenbach, Germany. The antibody showed 0.16% cross-reaction with T_3 (Huybrechts et al., 1989). The intra-assay coefficient of variation was 4.5%. The analysis of growth hormone (GH) was performed by a homologous radioimmunoassay (RIA), developed and validated for chicken by Berghman et al. (1988). The intra-assay coefficient of variation was 4.2%. All measurements of Insulin-like Growth Factor (IGF-I) were done by heterologous RIA, validated for chicken by Huybrechts et al. (1985). The intra-assay coefficient of variation was 6.3%.

The experiment was performed in a factorial arrangement of 15×2 (15 ages of birds \times 2 types of housing) with 20 replications. Data were subjected to statistical analysis using the GLM procedure of SAS[®] (1998) and the means were compared by Tukey test with 5% of probability.

Results

The body weight curves of both groups are given in Fig. 1 (expressed as the body weight at the end of each week of the experimental period). Body weight increased more rapidly before than after the onset of lay under both housing conditions, and levelled off between 26 and 33 weeks of age. Before the onset of lay, the OH birds had a higher mean body weight compared to CH chickens, but after the onset of lay (24–25 weeks) the situation was reversed. This resulted in a significant age–housing interaction effect ($P < 0.01$).

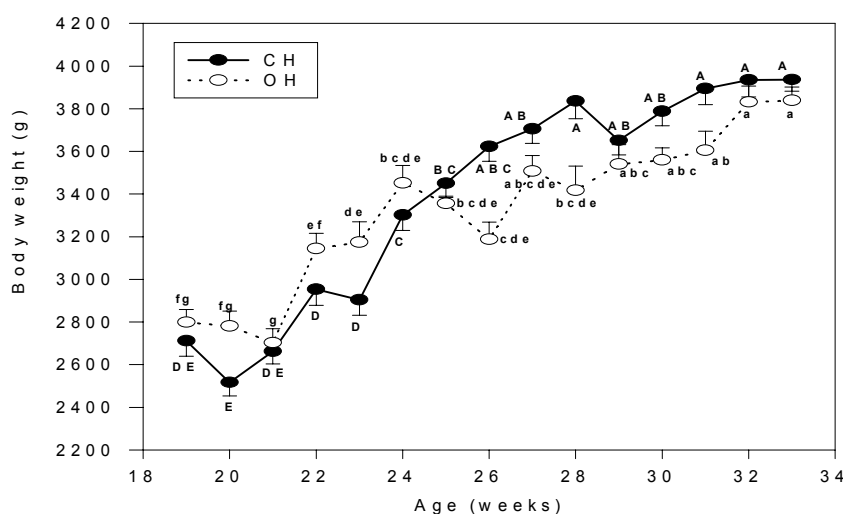


Fig. 1. Body weight (means \pm SEM) of broiler breeders ($n = 20$) raised in dark, close-sided house (CH) and conventional, open-sided house (OH) from 19 to 33 weeks of age. Significant age differences within a group are shown by different letters ($P < 0.05$). SEM smaller than symbol are not shown

Birds housed in OH reached sexual maturity and onset of lay (5% egg production level) about one week earlier (at 24 weeks) than the CH group (at 25 weeks) (Fig. 2). Furthermore, the OH group showed higher egg production until week 31, while from the 32nd week onwards the CH group started to have a higher production level.

In both groups age-related changes were obvious for T_4 , GH and IGF-1 (Figs 3, 5 and 6, respectively); levels of T_4 decreased especially between 24 and 28 weeks in both groups, concomitantly the GH levels sharply increased over the same period. A transient peak in T_3 (Fig. 4) occurred between 25 and 27 weeks. Between weeks 27–28 and the end of the period studied, the CH group showed higher levels of GH and T_3 but lower T_4 levels as compared to the OH group. In both groups, IGF-I concentrations started to increase from 20 weeks onwards, peaked at 26 weeks and decreased thereafter.

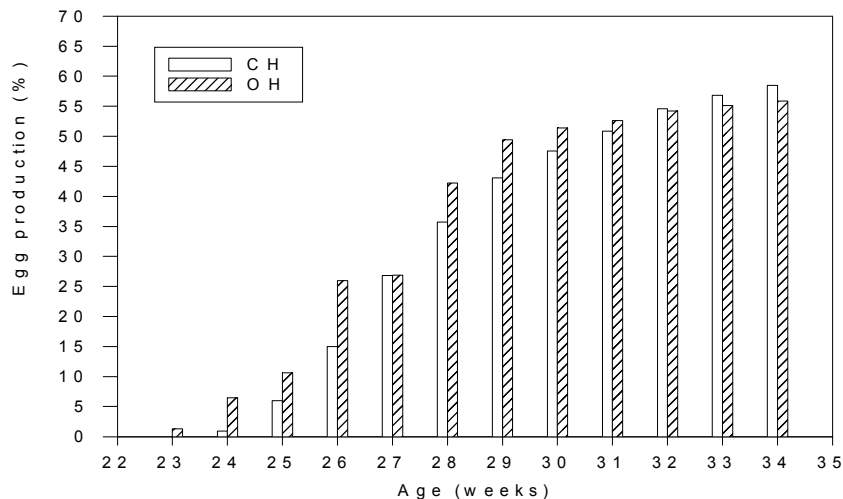


Fig. 2. Egg production percentage, by hen housed per week, of broiler breeders raised in dark, close-sided house (CH) and conventional, open-sided house (OH)

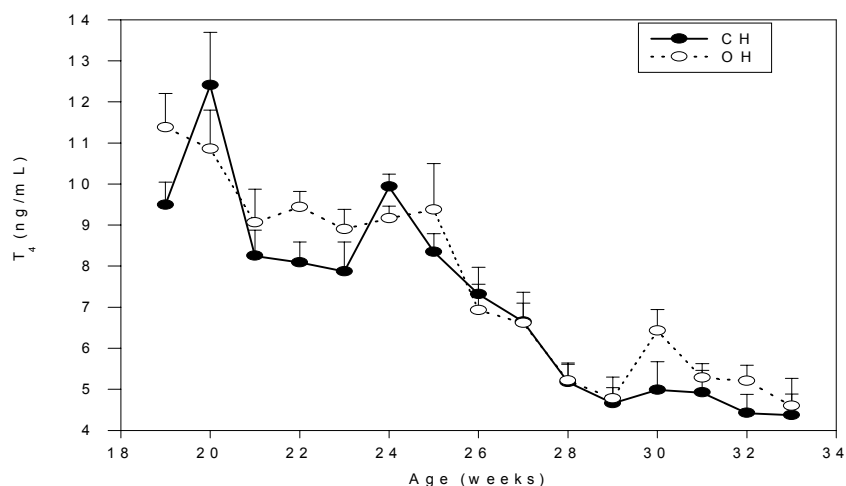


Fig. 3. Plasma concentrations of T₄ (mean \pm SEM) in broiler breeders (n = 20) raised in dark, close-sided house (CH) and conventional, open-sided house (OH), from 19 to 33 weeks of age. SEM smaller than the symbol are not shown

Correlation analysis of the mean plasma hormone levels pooled over age and housing systems revealed negative correlation of plasma T₄ levels ($r = -0.92$) with GH and with IGF-I levels ($r = -0.46$). Plasma T₃ levels were negatively correlated ($r = -0.31$) with GH and positively correlated with T₄ ($r = +0.40$) and with IGF-I ($r = +0.70$). Finally, GH was positively correlated with IGF-I ($r = +0.39$). However, the only significant ($P < 0.05$) correlation was between T₄ and GH.

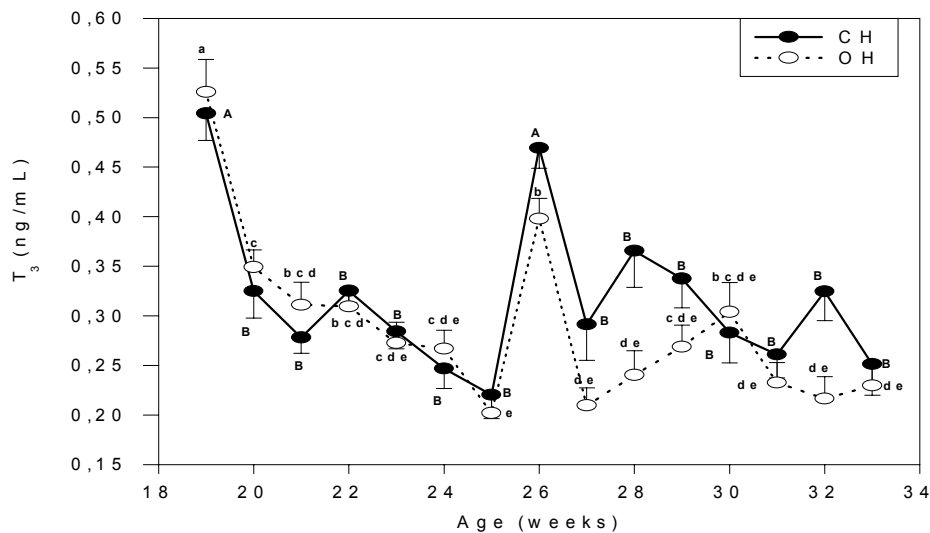


Fig. 4. Plasma concentrations of T₃ (mean ± SEM) in broiler breeders (n = 20) raised in dark, close-sided house (CH) and conventional, open-sided house (OH), from 19 to 33 weeks of age. Significant group differences within age are shown by different letters (P < 0.05). SEM smaller than the symbol are not shown

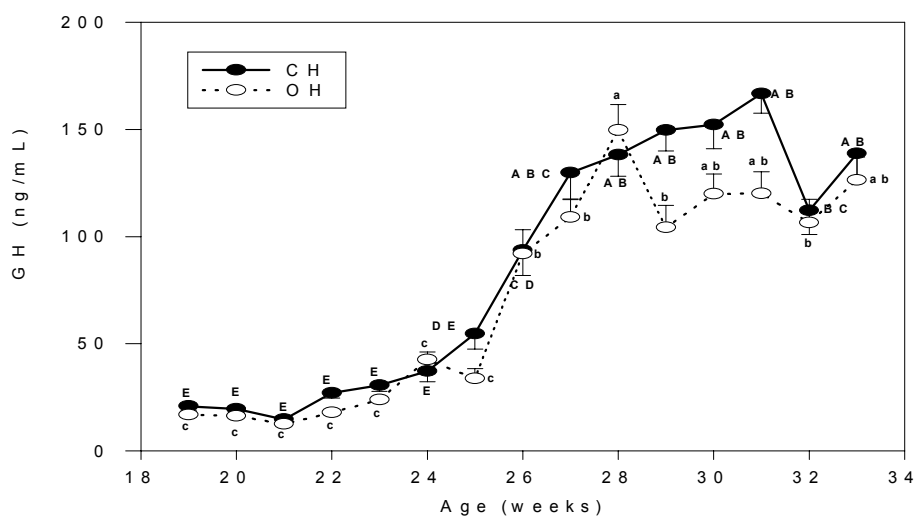


Fig. 5. Plasma concentrations of GH (mean ± SEM) in broiler breeders (n = 20) raised in dark, close-sided house (CH) and conventional, open-sided house (OH), from 19 to 33 weeks of age. Significant group differences within age are shown by different letters (P < 0.05). SEM smaller than the symbol are not shown

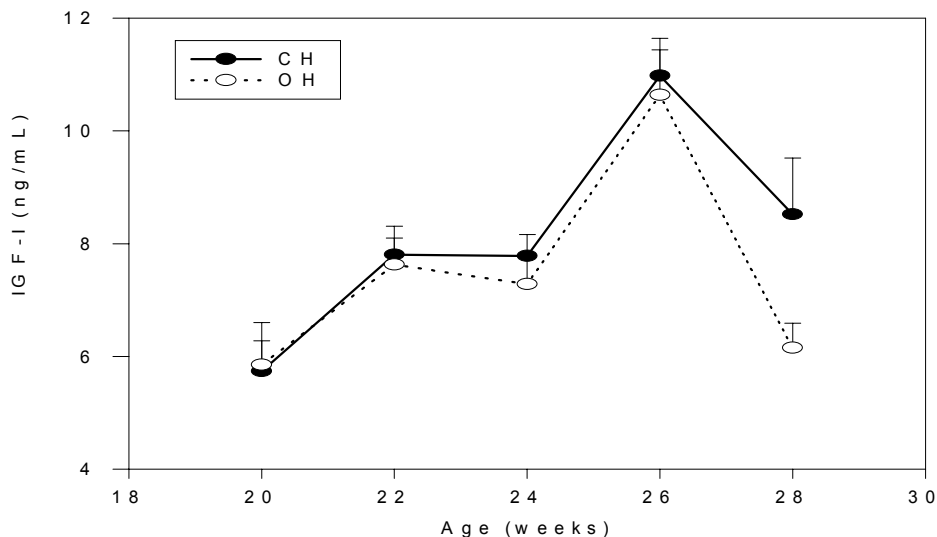


Fig. 6. Plasma concentrations of IGF-I (mean \pm SEM) of broiler breeders ($n = 10$) raised in dark, close-sided house (CH) and conventional, open-sided house (OH), from 19 to 33 weeks of age. SEM smaller than the symbol are not shown

Discussion

The higher body weight (BW) in the OH group before the onset of lay may have stimulated the earlier start of lay in this group since the onset of lay is dependent on age as well as on BW and body condition; the effect of BW is probably more important in feed-restricted birds. However, Yu et al. (1992) stressed that controlling feed intake and growth from 4 to 18 weeks is more important in determining age at sexual maturity than feed intake and changes in body composition from week 18 until the onset of lay.

Age-related changes in GH, T_4 , T_3 and IGF-I over the period from 19 until 33 weeks of age, over the transient period of the onset of lay, complete the earlier work of Bruggeman et al. (1997) who followed these metabolic hormones from 2 to 22 weeks in feed-restricted and *ad libitum* fed broiler breeders. The absolute levels of all these hormones in both the study of Bruggeman et al. (1997) and this study at 19–22 weeks of age were in the same order of magnitude, although T_4 was slightly higher (± 20 ng/mL) in the study of Bruggeman compared to the present results (± 10 ng/mL) at 20–22 weeks of age, while for T_3 values of 1 ng/mL and 0.5 ng/mL were found by Bruggeman and in this study, respectively.

The age-related increasing trend in T_4 and decreasing GH levels from 2 to 22 weeks as found by Bruggeman et al. (1997, 1999) was shown to be reversed during the transition period when birds are coming into lay; GH was rising again to levels of 100–150 ng/mL, like levels found in young animals, while T_4 was

decreasing again to pre-puberty levels of 4–6 ng/mL. These age-dependent inverse changes are reflected in the significant negative correlation of 0.92 between T_4 and GH. The decreasing trend in T_3 in growing feed-restricted broiler breeders as found by Bruggeman et al. (1997) is, however, not reversed, but apart from a transient peak at 26 weeks of age in both groups, T_3 levels remained low around 0.2–0.4 ng/mL. On the other hand, IGF-I showed a slightly increasing trend during this period of onset of lay, but this increase was not followed over the entire period.

The stimulatory effect of IGF-I on the proliferation of granulosa and theca cells, as well as on enhancing progesterone production by granulosa cells and androstenedione production by theca cells *in vitro* is well documented (Roberts et al., 1994; Williams et al., 1994; Onagbesan and Peddie, 1995; Onagbesan et al., 1999). Moreover, *in vivo* supplementation of IGF-I changed *in vitro* LH sensitivity of the granulosa cells, implicating a role of IGF-I on follicular maturation (Huybrechts et al., 1993) as was also suggested by Hocking et al. (1994).

Around the same age of 26 weeks, IGF-I levels also reached high values. These rises in T_3 and IGF-I occurred around the start of egg production in both groups. The mechanism whereby these differences in plasma hormone levels of the somatotrophic and thyroid axis interfere with the development and regulation of the reproductive system remains totally speculative. However, from *in vitro* studies it is clear that T_3 and IGF-I are involved in the steroidogenesis of ovarian cells. The LH-stimulated P_4 production by granulosa cells in T_3 -treated animals was increased as shown by Vanmontfort et al. (1993).

The mutual relationships between GH, T_4 and T_3 as pointed out by several authors (Darras et al., 1995; Buyse and Decuypere, 1999; Buyse et al., 2000) after short-term food restriction or food deprivation, were also found in this study when observing the differences between OH and CH groups during the period after the onset of lay (28–33 weeks). A lower GH was concomitant with a higher T_4 and lower T_3 in the OH group and can be explained by the main reducing effect of GH on 5D-III deiodinase activity in birds (Darras et al., 1995; Vasilatos-Younken et al., 1999).

However, a very significant (6-fold) increase in GH after the onset of lay without any significant rise in either T_3 or IGF-I could point to a relative insensitivity to high GH plasma levels, and this is corroborated by the weak, nonsignificant correlations between these plasma parameters. Therefore changes at the hepatic GH-receptor level, together with an increased pituitary GH secretion and/or decreased GH turnover may be expected. This may indicate that hypothalamo-pituitary changes at the onset of lay not only imply changes of gonadotrophic cell function but also of other hormonal axes. The relatively abrupt decrease in T_4 without apparent changes in T_3 also may point to a decrease in the activity of the thyrotrophic axis.

Teleologically, the suggested antigonadal effect of thyroid hormones in laying hens (Verheyen et al., 1986; Dewil et al., 1991) may be brought in connection with the generally lower thyroid activity after the onset of lay, although the mechanism is poorly understood and may involve interaction with hormones of the gonadotrophic axis.

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