



Increased exposure to yolk testosterone has feminizing effects in chickens, *Gallus gallus domesticus*

B. Riedstra*, K. A. Pfannkuche, T. G. G. Groothuis

Behavioural Biology, Centre for Life Sciences, University of Groningen, Groningen, The Netherlands

ARTICLE INFO

Article history:

Received 11 September 2012
 Initial acceptance 25 October 2012
 Final acceptance 20 December 2012
 Available online 5 March 2013
 MS. number: 12-00700

Keywords:

behavioural lateralization
 comb colour
 domestic chicken
Gallus gallus domesticus
 long-term effect
 maternal testosterone
 prenatal testosterone exposure
 rank order
 short-term effect
 testosterone level

Competing for food by altricial and semiprecocial bird nestlings is a behaviour well known for its sensitivity to maternal androgens during prenatal development. Whether a similar effect is present in precocial species that do not beg is less well known. We therefore increased yolk testosterone levels within the physiological range at the onset of incubation to study its effects on food competition behaviour in the domestic chicken, *Gallus gallus domesticus*. We found an increase in competitiveness in testosterone-treated male domestic chicks, raising their level to that of the females. This is in line with the decrease in circulating plasma levels of males in the direction of the levels in females, and the overall decrease in androgen receptor densities after prenatal treatment as found previously. Hormones are known to have long-lasting organizing effects on behaviour and to affect sexual differentiation in vertebrates. Although research into hormone-mediated maternal effects has been productive, only a few studies describe (the ambiguous) effects into adulthood. Therefore we followed our animals into adulthood and recorded androgen-dependent social behaviour and secondary sexual characteristics, body mass and circulating plasma testosterone levels and checked whether these variables were treatment dependent. Treatment had a near significant effect on comb colour (both brightness and chroma). Again treatment caused a shift towards a more female-like phenotype. This suggests that, in contrast to earlier suggestions, maternal androgens may interact with (but not disrupt) sexual differentiation of brain and behaviour and the development of secondary sexual characteristics.

© 2013 The Association for the Study of Animal Behaviour. Published by Elsevier Ltd. All rights reserved.

In many vertebrate species embryos are exposed to steroids of maternal origin (for birds, fish, reptiles and mammals see e.g. Schwabl 1993; McCormick 1999; Bowden et al. 2000; Drea 2011, respectively). Such exposure can organize phenotypic differences within and between the sexes and affects behaviour, physiology and the organization of the brain (Hews & Moore 1995; Gil 2003; Strasser & Schwabl 2004; Groothuis et al. 2005; Daisley et al. 2005; Cohen-Bendahan et al. 2005; Rogers 2006; Eising et al. 2006; Dloniak et al. 2006; Weinstock 2008; Pfannkuche et al. 2009). Exposing offspring to hormones during early development is generally interpreted as a way for mothers to adjust their offspring's phenotype to the prevailing environmental conditions to maximize fitness (Groothuis et al. 2005).

Much research on these hormone-mediated maternal effects makes use of bird species, because bird embryos develop outside the mothers' body in a sealed environment that allows for easy measuring and manipulation of prenatal exposure to steroid

hormones. Effects of varying levels of maternal testosterone (T) in egg yolk have received most attention. This focus on T is caused by the finding that egg yolks contain maternally derived androgens (Schwabl 1993) that vary systematically within and between clutches (Groothuis et al. 2005; Gil 2008; von Engelhardt & Groothuis 2011).

Prenatal exposure to T is important since increased yolk T levels can affect important traits in altricial and semiprecocial chicks such as competition, for example begging for parental food provisioning and aggressive behaviours, growth, immunity and survival (Schwabl 1996; Strasser et al. 1998; Eising et al. 2003; Groothuis et al. 2005; von Engelhardt et al. 2006; Rubolini et al. 2006b; Muller et al. 2006, 2009a, b; Gil 2008; von Engelhardt & Groothuis 2011). Several of these effects of yolk T are sex specific; for example in zebra finches, *Taeniopygia guttata*, yolk T affected female but not male begging and growth (von Engelhardt et al. 2006) and in the domestic canary, *Serinus canaria domestica*, it increased post-hatching growth in females, but decreased it in males (Muller et al. 2008, 2009b).

Begging behaviour in altricial and semiprecocial species in particular has received much attention. Such chicks are dependent on their parents for food and compete with siblings for parental

* Correspondence: B. Riedstra, Behavioural Biology, Centre for Life Sciences, University of Groningen, Nijenborgh 7, 9747 AG Groningen, The Netherlands.
 E-mail address: B.J.Riedstra@rug.nl (B. Riedstra).

provisioning by begging. Increased yolk T levels stimulate begging behaviour in several altricial and semiprecocial species (Schwabl 1996; Eising & Groothuis 2003; von Engelhardt et al. 2004; Rubolini et al. 2006b). Although yolk androgens can affect behaviour or morphology in precocial chicks too (Daisley et al. 2005; Okuliarova et al. 2007), the effect on sibling competition has been mentioned (without data being presented) in only one study (see Groothuis et al. 2005). This lack of studies is probably related to the erroneous assumption that precocial chicks do not rely on parental provisioning and the fact that fowl species are studied in laboratory conditions without the parents being present. However, fowl mothers, including the domestic chicken, *Gallus gallus domesticus*, perform tid-biting displays (Domm 1927) by which they indicate to offspring the location of highly prized food items. This is usually followed by chicks trying to outcompete each other for this item. Therefore the first aim of this study was to test the effect of elevated yolk T on sibling competition in the precocial domestic chick. Based on Groothuis et al. (2005) we predicted that increased exposure to T would increase competitiveness, especially in male chicks.

Prenatal exposure to androgens can also have long-term organizing effects on behaviour and phenotypic traits (Nelson 1995). This is intriguing from both a proximate and an ultimate perspective: First, our knowledge of organizing effects of prenatal exposure to androgens on brain and behaviour in avian species comes mainly from studies on sexual differentiation (Cohen-Bendahan et al. 2005). Recent evidence from egg injection studies using dosages within the physiological range indicates that maternal androgens may affect the phenotype via different pathways from sexual differentiation of the gonads, the reproductive tract, and brain and behaviour (see Carere & Balthazart 2007 and Groothuis & Schwabl 2008 for a discussion on this topic). Second, such prenatal programming suggests that mothers may adjust the offspring's phenotype not only to early life conditions but also to conditions further in the future. Alternatively, maternal modification of the offspring's adult phenotype may be the outcome of a natural selection process on 'adjusted' chick phenotypes.

Studies on any long-term effects of prenatal exposure to steroids on behaviour and phenotypic traits, however, are scarce and present ambiguous results. In almost adult house sparrows, *Passer domesticus*, the size of the black bib, an important sexually selected trait, was increased by prenatal exposure to T (Strasser & Schwabl 2004). Likewise, in 10-month-old black-headed gulls, *Larus ridibundus*, the development of the black mask was accelerated (Eising et al. 2006). However, the effect on the sparrows could not be replicated (Partecke & Schwabl 2008). Moreover, there were no long-term effects on such traits in adult European starlings, *Sturnus vulgaris* (Muller & Eens 2009), a negative effect on spur length in adult ring-necked pheasants, *Phasianus colchicus* (Rubolini et al. 2006a) and delayed song development in adult canaries (Muller et al. 2008). In almost adult canaries, house sparrows and black-headed gulls aggression, competitiveness and dominance were increased (Eising & Groothuis 2003; Strasser & Schwabl 2004; Schwabl 1993; Partecke & Schwabl 2008), but the effect in the canary could not be replicated in adult birds (Muller et al. 2008). We therefore not only tested the effect of increased yolk T level on early sibling competition but also followed these birds into early adulthood. To this end we injected eggs before incubation either with T dissolved in sesame oil or with sesame oil only, taking care that the manipulation was within the physiological range of this species.

Recently it was shown that the same experimental manipulation resulted in a decrease in plasma T levels and androgen receptor densities in the hypothalamic area in 2-week-old chicks (Pfannkuche et al. 2011). The latter finding is especially interesting because, although hormone-mediated effects on behaviour have

been extensively researched, much about the underlying mechanisms is as yet unknown. One possibility for how the organizing effects of hormones may come about is via an alteration of the hypothalamus–pituitary–gonadal axis, which would affect endogenous hormone production later in life. Although in quail, *Coturnix japonica*, chicks the effect on plasma T levels was not statistically significant (Daisley et al. 2005), prenatal exposure increased plasma T levels significantly in 3-week-old nestlings of the spotted starling, *Sturnus unicolor* (Muller et al. 2007). A long-term study in the house sparrow, however, did not show an effect of prenatal exposure to androgens on plasma T levels in adulthood (Partecke & Schwabl 2008). Given these results the decrease in T production and sensitivity in the domestic chick is intriguing. We therefore investigated whether the decrease in plasma T levels found in 2-week-old chickens was still present after the onset of reproduction.

Since circulating plasma levels in adult chickens are positively associated with winning fights and social position (Ligon et al. 1990; Johnsen & Zuk 1995), we predicted that elevation of yolk T would result, at least in males, in a lower social position later in life. We also predicted that this would coincide with less pronounced secondary sexual characteristics such as smaller and less red combs and wattles since these are under the influence of circulating T (Ligon et al. 1990; Johnsen & Zuk 1995; Zuk et al. 1995; Verhulst et al. 1999).

METHODS

Treatment and Housing

Freshly laid eggs of laying hens were purchased from a commercial breeder (Verbeek Lunteren, The Netherlands) and incubated at the Zoological Laboratory in Haren (University of Groningen, The Netherlands). We used the LSL strain, a commercially available white leghorn type, which is selected on productivity traits. There is no reason to assume that prenatal exposure to hormones would exert other general effects in this strain more than in other strains. It is relatively easy to keep and there are no large behavioural differences apparent compared to other commercial strains or junglefowl, *Gallus gallus*. The LSL allows for easy neonatal phenotypic sexing of the animals on the basis of feather growth.

Eggs received an intrayolk injection of 75 ng crystalline T dissolved in 100 μ l sesame oil or 100 μ l sesame oil (C) only. This 75 ng is approximately two times the standard deviation of T measured in a subset of eggs of the same batch of eggs (Groothuis et al. 2005; Pfannkuche et al. 2011). Injecting was facilitated by drilling a small hole in the egg shell which, after injection, was sealed with candle wax. Eggs were incubated at 37.5 °C and 60% humidity, and turned automatically three times a day. At day 18 eggs were reallocated to hatching trays within the same incubator. After hatching in individual compartments, sexing and individual marking, chicks were kept in 12 groups of eight (two chicks of each sex * treatment combination). After 6 weeks a subset of animals of both treatments and sexes was killed for brain analyses (Pfannkuche et al. 2011) and the rest were kept in single-sex and mixed-treatment groups in two large outdoor roofed aviaries (approximately 10 \times 20 m and 3.5 m high). In addition to a metal wing clip received early in life, males and females were colour marked (black, blue or green) on the feathers (head, neck, back or a combination of two regions) with a permanent marker at the onset of the observations to enable individual recognition. To decrease the chance of observer bias we took the following measures: (1) colours or coloured regions were not specific for sex or treatment, and within but not between groups colour markings were unique to an individual; (2) there was no information about individual treatment in the housing or

Download English Version:

<https://daneshyari.com/en/article/10970658>

Download Persian Version:

<https://daneshyari.com/article/10970658>

[Daneshyari.com](https://daneshyari.com)