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Regulation of FSH secretion in broiler breeders

Regulação de secreção de FSH em reprodutores de frango

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Abstract

Synthesis and secretion of LH and FSH takes place in the gonadotropes of the pituitary gland (adenohypophysis). The hypothalamus, in turn, controls the secretion of gonadotropins by the pulsatile secretion of gonadotropin releasing hormone. Therefore, the neuroendocrine linkage of the hypothalamic-pituitary-gonadal axis provides an integrated system responsible for proper reproductive performance, including gamete development and sex steroids secretion. In broilers there is a lack of synchrony between LH and FSH pulses, and gonadotrophs reside in separate cells within the pituitary gland, suggesting that in the adult male fowl LH and FSH secretion are regulated independently. Modern male broiler breeders (selected for increased growth rate) fail to maintain testis size and semen production after 25 weeks of age. Testis size closely follows changes in FSH concentrations, suggesting that circulating FSH levels are a very good indicator of testis function in mature broiler breeder. Although cGnRH-I is believed to be the prime regulator of gonadotropin release in male birds, the role of other GnRH isoforms in the regulation of FSH remains unclear. Therefore, there is critical need to further evaluate the actual regulation of FSH, and the underlying effect FSH on testicular function in mature broilers.

Keywords: broilers, FSH, GnRH, LH, reproduction.

Resumo

A síntese e secreção de LH e FSH acontece nos gonadotropos da glândula pituitária (adenohipófise). O hipotálamo, por sua vez, controla a secreção de gonadotropinas pela secreção pulsátil do hormônio liberador de gonadotropina. Portanto, a ligação neuroendócrina do eixo hipotálamo-pituitária-gonodal fornece um sistema integrado responsável pelo desempenho reprodutor correto, incluindo desenvolvimento de gametas e secreção de esteroides sexuais. Em frangos há uma falta de sincronia entre pulsos LH e FSH, e gonadotropos residem em células separadas na glândula pituitária, sugerindo que a secreção de LH e FSH do macho adulto é regulada de forma independente. Reprodutores modernos de frango macho (selecionados para taxa de crescimento aumentada) falham na manutenção do tamanho dos testículos e produção de sêmen após 25 semanas de idade. Apesar de que se acredita que o cGnRH-I é o regulador primário da liberação de gonadotropina em pássaros macho, a função de outras isoformas de GnRH na regulação de FSH não é clara. Portanto, há uma necessidade crítica de se avaliar melhor a real regulação de FSH, e o efeito basal do FSH na função testicular em frangos maduros.

***Palavras-chave**: frangos, FSH, GnRH, LH, reprodução.*

Introduction

Selection for increased growth rate and yield over the past 60 years has dramatically altered the "physiology" of the modem broiler breeder chicken. As a consequence, and due to the negative correlation between growth and reproductive fitness, broilers have experienced a tremendous reduction in reproductive capacity (Richards et al., 2010). This period of genetic selection has resulted in a reduction in the number of eggs produced by females (Etches, 1996), and a reduction in the quality and numbers of viable sperm produced by males (Kirby et al., 1998). Other consequences of this selection include the requirement for quantitative feed restriction of breeders (both male and female) and a significant reduction in the reproductive "life-expectancy" of breeders. In commercial flocks, it is very common for fertility to begin to decline immediately following "peak" egg production. As a result, producers are frequently bringing new, younger, males into their flock, a process known as spiking, to rescue fertility. Based on our observations, we propose that a primary cause for the decline in fertility observed in post-peak production flocks is the loss of the robust pattern of episodic release of follicle stimulating hormone (FSH) by the pituitary gland (Vizcarra et al., 2004).

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Role of FSH in reproductive performance

As might be expected, there are marked effects of FSH on gonadal functioning in poultry. For instance, FSH can stimulate proliferation of granulosa cells (Davis et al., 2001) and is required for the long term culture of granulosa cells (Hattori et al., 1986). Moreover, FSH plays an important role in the differentiation of granulosa cells; stimulating progesterone production and expression of steroidogenic acute regulatory protein and P450 side chain cleavage in granulosa cells from small yellow follicles (Johnson et al., 2002). In mammals, FSH signaling is essential for initiation and maintenance of spermatogenesis and high production of spermatozoa due to intimate contacts between Sertoli cells and differentiating spermatogonia (Dierich et al., 1998). There is evidence that a similar situation exists in poultry (Kuenzel, 2015; Vizcarra et al., 2015).

Specific radioimmunoassays for chicken and/or other avian FSH have been developed (Follett, 1976; Krishnan et al., 1993). Despite this, and the sequencing of chicken FSH sub-unit cDNA (Shen and Yu, 2002) and gene (Yang et al. [GenBank Accession number AF467082]), there is a general paucity of information regarding FSH synthesis and release in birds, including chickens.

In broiler breeders we found that males fail to maintain testis size and semen production after 25 weeks of age (Fig. 1). Note that testis size closely follows changes in FSH levels, increasing in mass with increased FSH and decreasing in mass with reduced FSH (Vizcarra et al., 2010). The cause for concern associated with declining testis weights is shown in Fig. 2. Testis weights in the fowl are highly correlated to daily sperm production (DSP), with an R^2 of 0.89. Thus, there is a direct and easily measurable relationship between testis size and reproductive status. Further, we have shown that the efficiency of spermatogenesis (DSP/gram of testis) reaches a peak at about 9 grams, and that those testes less than 9 grams producing fewer sperm per gram of testis per day are more likely to produce abnormal sperm. Circulating FSH levels are a very good indicator of testis function in mature (28 weeks of age and older) broiler breeder males (Fig. 3). Based on this experiment, with a regression R^2 of 0.90, we have concluded that FSH is critical, not just in the initiation of spermatogenesis at puberty, but also for the long term maintenance of quantitatively normal spermatogenesis in mature males, a critical component for the maintenance of adequate fertility levels. In contrast, concentrations of Luteinizing Hormone (LH) explained only 35% of the variation of testis weight. FSH and LH are secreted in asynchronous pulses, with pulse frequencies of 0.33 and 0.54 per hour, respectively (Vizcarra et al., 2004). The relationship between reproductive status and the pulsatility of FSH and LH has received only limited attention at this time. However, the patterns of LH and testosterone, sampled at 10 min intervals for 12 h, were evaluated in a low FSH male (baseline FSH <6.7ng/ml with no observed pulses and left testis weight of 6.3g) and from a normal male (baseline FSH >10.5ng/ml with 4 FSH pulses and a left testis weight of l8.5 g). This showed firstly, a loss of pulsatile testosterone secretion in response to LH pulses in the low FSH male; and secondly, a significant decline in both the baseline and amplitude of LH pulses, with no apparent reduction in LH pulse frequency in the absence of FSH pulses (Vizcarra et al., 2004).

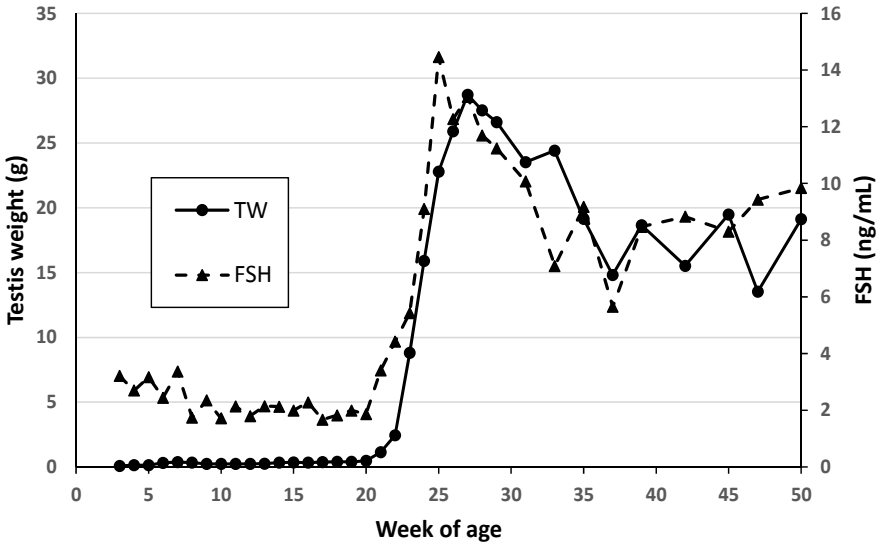

Figure 1. Testis weight (TW) and concentrations of FSH in male broiler breeders from 3 to 50 weeks of age. Each week 30 birds were sampled. Adapted from Vizcarra et al. (2010).

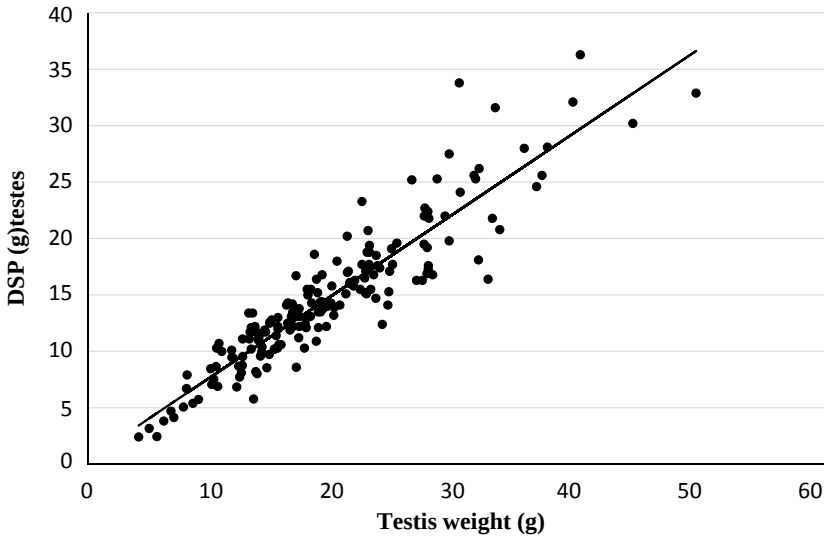

Figure 2. Relationship between daily sperm production (DSP) and testis weight in male broiler breeders. A total of 2,500 birds were sampled. Adapted from Vizcarra et al. (2010).

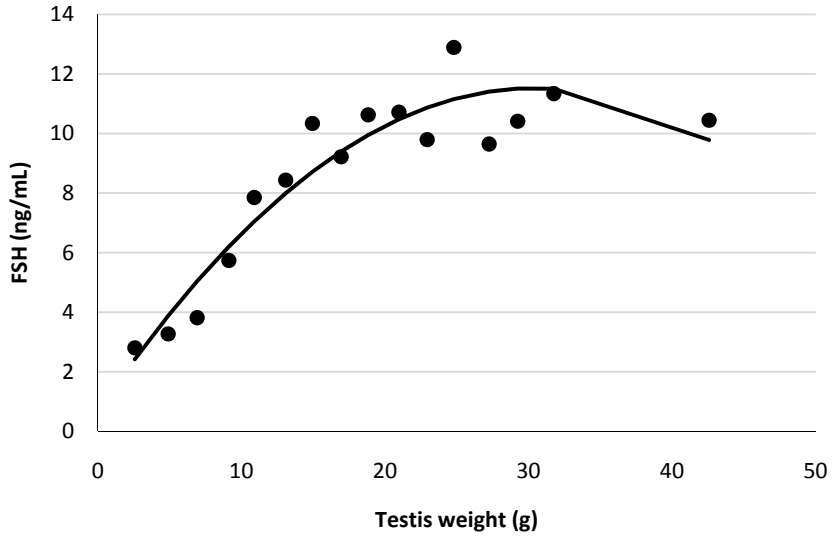

Figure 3. Relationship between FSH concentrations and testis weight in male broiler breeders. A total of 2,500 birds were sampled. Adapted from Vizcarra et al. (2010).

Hypothalamic control of FSH synthesis and release

Gonadotropin-releasing hormone (GnRH) is a decapeptide that plays a fundamental role in the release of gonadotropins from the pituitary gland, and is the primary hormone that regulates reproduction. The nomenclature used to distinguish different GnRH isoforms between mammalian and non-mammalian species have been described using a variety of phylogenetic and genomic synteny analyses (Kim et al., 2011; Millar et al., 2004; Roch et al., 2011; Tostivint, 2011). Chicken GnRH-I (King and Millar, 1982; Miyamoto et al., 1982) differs in only one amino acid compared with the mammalian form (cGnRH-I; [Gln8]-GnRH); whereas chicken GnRH-II (Miyamoto et al., 1984) differs in three amino acids (cGnRH-II; [His5-Trp7-Tyr8]-GnRH). The coordination of gonadotropin secretion via cGnRH is also modulated by the interaction of the GnRH peptide with its receptors. The Gonadotropin-releasing-hormone receptors (GnRHR) have the characteristic feature of a classical seven-transmembrane, G-protein-coupled receptor (Millar, 2003, 2005; Neill, 2002). Four vertebrate GnRHR lineages have been proposed using genome synteny and phylogenetic analyses; nonmammalian type I, nonmammalian type II, nonmammalian type III/mammalian type II, and mammalian type I (Kim et al., 2011).

Two GnRHRs have been identified in chickens (McFarlane et al., 2011; Shimizu and Bedecarrats, 2006). The non-mammalian type I receptor is predominantly express in the chicken pituitary gland (Joseph et al., 2009; Shimizu and Bedecarrats, 2006); whereas the non-mammalian type II receptor is not only expressed in the

in the pituitary and the brain, but also in the in the gonads and other tissues (Sun et al., 2001).

The affinity of the cGnRH-I peptide to the non-mammalian type II receptor is higher than the affinity of cGnRH-II peptide to the same receptor (Sun et al., 2001). The non-mammalian type I receptor in the chicken pituitary is differentially expressed with respect to the reproductive status, and is associated with the control of gonadotropin secretion (McFarlane et al., 2011).

In avian species only indirect measurements of the GnRH pulse generator is available by measuring plasma LH concentrations in frequent samples or in pituitary extracts (Chou and Johnson, 1987; Sharp and Gow, 1983; Wilson and Sharp, 1975). In addition, the episodic nature of LH and FSH was evaluated in unrestrained male broiler breeders using serial blood sampling (Vizcarra et al., 2004). Gonadotropin secretion in males is characterized by a pulsatile pattern with LH pulses being more frequent and having greater amplitude than FSH pulses. In chickens, LH- and FSH-containing gonadotrophs reside in separate cells within the pituitary gland (Proudman et al., 1999). We observed that there was a lack of synchrony between the episodic release of LH and FSH. Only 23% of the LH pulses were associated with FSH episodes, suggesting that in the adult male fowl LH and FSH secretion are regulated independently (Vizcarra et al., 2004).

Active immunization against cGnRH-I and cGnRH-II in adult broiler breeder males was associated with a differential response on the ability to produce an immune reaction (Vizcarra et al., 2000). Titers were increased in cGnRH-I but not in cGnRH-II treated birds compared with BSA immunized males (Fig. 4). Concentrations of LH and FSH in frequent samples were not affected by treatment; however, testis weight was significantly decreased in cGnRH-I birds compared with the other treatments (Fig. 5). Taken together, these data, and data from other laboratories (Dawson and Sharp, 2007; Katz et al., 1990; Sharp et al., 1990; Stevenson et al., 2012; Ubuka and Bentley, 2009), support the idea that cGnRH-I is the prime regulator of gonadotropin release in male birds.

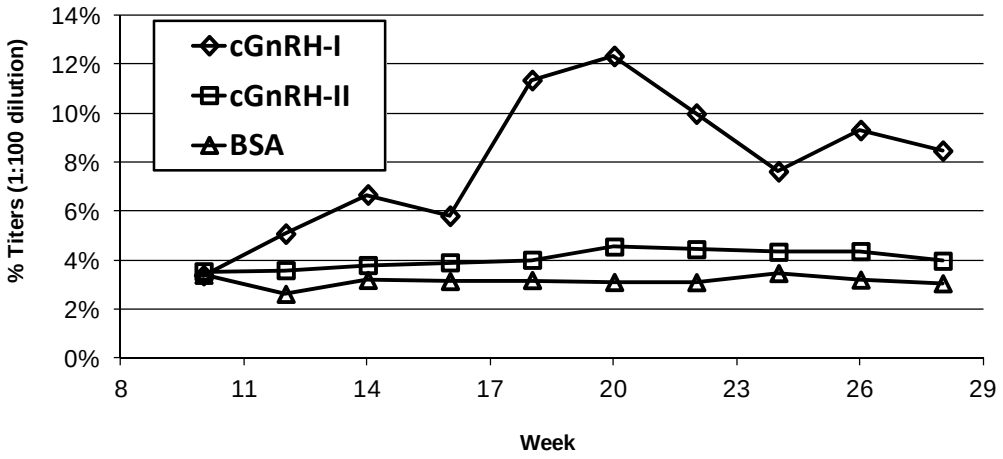

Figure 4. Antibody titers (^{125}I-cGnRH bound, %) of male broiler breeders immunized against cGnRH-I, cGnRH-II, and BSA. Titers were increased ($P < 0.05$) in cGnRH-I but not in cGnRH-II treated birds compared with BSA immunized males. Adapted from Vizcarra et al. (2000).

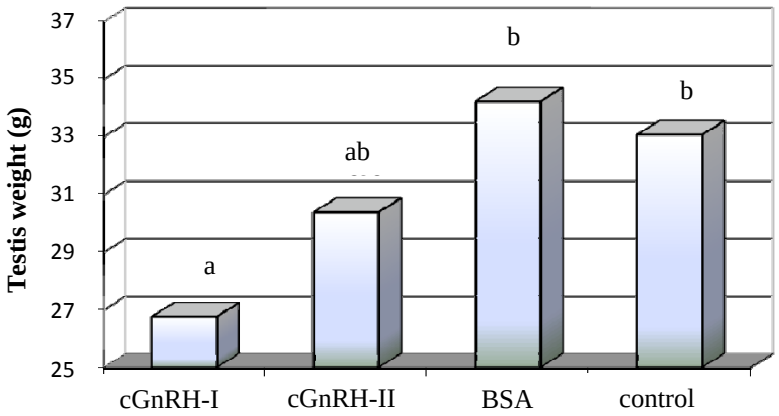

Figure 5. Testis weight of male broiler breeders immunized against cGnRH-I, cGnRH-II, BSA, and not immunized (Control) birds. Different letters indicate significant differences ($P < 0.05$). Adapted from Vizcarra et al. (2000).

Conclusions

FSH is a heterodimeric glycoprotein hormone produced in the pituitary that has profound effects on male and female reproductive function. The episodic release of FSH is readily apparent in the circulation of cannulated and unrestrained males. The dynamic pattern of hormone release appears to be necessary for the initiation and maintenance of spermatogenesis in the fowl. However, the actual regulation of FSH secretion remains unclear, as the factors responsible for the physiological control of synthesis and release of this hormone have not been revealed.

The fowl provides a unique model for studying the differential regulation of gonadotropin secretion in that FSH and LH appear to be produced by separate cells in the pituitary, and both are released in a robustly pulsatile fashion. Based on the fact that FSH and LH pulses are asynchronous, the role of the GnRH family in regulating FSH remains unresolved. That FSH is a critical component of the reproductive control scheme in males is of little doubt, the control of its synthesis and secretion, while elusive to date, deserves further attention.

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