

Relationship between serum gonadotropins and pituitary immunoreactive gonadotropins and steroid receptors during the first FSH increase of the estrous cycle and following steroid treatment in heifers

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Abstract

The objectives were to determine the effects of (i) time during the first FSH increase of the estrous cycle (time-course study) and (ii) exogenous steroid treatment (steroid feedback study) on the relationship between circulating serum gonadotropins, and the proportions of pituitary cells immunoreactive for gonadotropins and steroid receptors during the estrous cycle in heifers. Pituitaries were collected from heifers ($n=40$) slaughtered at 13 h ($n=8$), 30 h ($n=24$) and 66 h ($n=8$) after estrous onset, corresponding to before, during and after the first FSH increase of the estrous cycle. Heifers slaughtered during the FSH increase (at 30 h) either received no treatment ($n=8$), or were treated ($n=16$) with estradiol benzoate and/or progesterone before slaughter. During the time-course study, the proportion of pituitary cells immunoreactive for FSH increased ($P<0.05$) during the first transient FSH increase reflecting serum concentrations. The proportion of pituitary cells immunoreactive for LH was unaltered, a reflection of serum LH concentrations. The proportion of estrogen receptors (ER)- α , but not ER- β , was decreased ($P<0.05$) at 30 h compared with at either 13 or 66 h. During the steroid feedback study, exogenous progesterone with or without estradiol suppressed ($P<0.05$) the proportions of pituitary cells immunoreactive for gonadotropins, serum FSH concentrations and LH pulse frequency. Steroid treatment did not alter the proportion of pituitary cells positive for estrogen receptors (α and β). While progesterone receptors (PR) were not detected in the anterior pituitary by immunohistochemistry during the early estrous cycle or in response to steroid treatment, quantitative real-

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time PCR revealed that mRNA for progesterone receptors was expressed at very low levels. The expression of pituitary PR mRNA was decreased ($P < 0.05$) at 30 and 66 h compared with 13 h, and was suppressed ($P < 0.05$) following steroid treatments. Alterations in pituitary steroid receptors are implicated in the differential regulation of gonadotropin secretion during the first transient FSH rise, but not in response to exogenous steroids. The time-course study and steroid feedback responses support the hypothesis that LH pulse frequency is tightly linked to regulation of GnRH pulse frequency. Serum FSH is regulated by its own synthesis, as reflected by pituitary FSH content and perhaps by alterations in pituitary sensitivity to circulating steroids by changes in steroid receptor content.

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1. Introduction

Gonadotropin-releasing hormone (GnRH) regulates the synthesis and release of the gonadotropic hormones, luteinizing hormone (LH) and follicle-stimulating hormone (FSH) from gonadotrope cells of the anterior pituitary (Wilson et al., 1990). However, dissociation in the pattern of FSH and LH secretion occurs during the estrous cycle of sheep (Pant et al., 1977) and cattle (Sunderland et al., 1994). Recurrent increases in FSH concentrations occur throughout the estrous cycle, associated with emergence of follicular waves (Adams et al., 1992a; Sunderland et al., 1994; Crowe et al., 1997). Furthermore, the LH pulse frequency varies during the cycle independent of FSH (Cupp et al., 1995). Based on histological studies, the bovine pituitary contains approximately 11.5% of cells that are immunohistochemically positive for LH, while only 5.4% are positive for FSH (Bastings et al., 1991). Whether the proportions of FSH or LH immunoreactive cells are differentially altered during the early estrous cycle, reflecting serum gonadotropin concentrations, or in response to treatment with steroids *in vivo* remain unclear.

Many factors are implicated in the differential modulation of LH and FSH secretion (Chappell et al., 1983; Findlay and Clarke, 1987; Price, 1991). Steroids affect their response via receptors and alterations in the concentrations of these receptors may be a means by which the pituitary alters its sensitivity to the effects of circulating estradiol, accounting for discordant patterns in the secretion of FSH and LH. Two types of estrogen receptors (ER), implicated in reproductive processes, have been isolated, ER- α (Toft and Gorski, 1966) and ER- β (Kuiper et al., 1996). The amount of ER- β in gonadotrope cells is low compared with that of ER- α (Mitchner et al., 1998; Shughrue et al., 1998). The literature suggests that the numbers of pituitary ER are altered during the estrous cycle (Nett et al., 1987; Childs et al., 2001). Progesterone receptors (PR) are present in gonadotrope cells in primates (Press and Greene, 1988; Sprangers et al., 1990), humans (Couzinet et al., 1999), rats (Spona et al., 1979) and cattle (Jacobs and Smith, 1981). However, the studies to date that have characterized steroid receptors and gonadotropin content in the pituitary of cattle have not related the sampling times to the stage of the FSH transient increases that stimulate follicle wave emergence. The aim of these experiments was to determine the effect of endocrine status during the first FSH increase of the estrous cycle and exogenous steroid treatment on the relationship between circulating serum gonadotropins (FSH: experiment 1 and pulsatile LH: experiment 2), and the proportions of pituitary cells immunoreactive for gonadotropins and steroid receptors during the early estrous cycle in heifers (experiment 1).

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