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Section I

Physiology and Pathophysiology of Follicular Recruitment and Selection

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Abstract

Successful reproduction in females requires the coordinated activity of the hypothalamic-pituitary-ovarian (HPO) axis. An understanding of the physiological control mechanisms which govern the HPO axis during the spontaneous menstrual cycle is essential in order to devise successful strategies for the pharmacological manipulation ovarian function in clinical practice as well as to understand the pathophysiological mechanisms responsible for altered ovarian function that occurs in endocrine disorders such as polycystic ovarian syndrome (PCOS). The goal of this chapter is to summarize the current knowledge of the regulation of ovarian function during the normal menstrual cycle and to apply this information to devise novel strategies for ovulation induction and for the understanding of endocrine disorders which adversely affect ovarian function.

Key words: Follicle Stimulating Hormone (FSH), Luteinizing Hormone (LH), FSH threshold, Follicle selection

The HPO axis in the control of the menstrual cycle

It is well established that successful menstrual cycles require a functional HPO axis which consists of: i) the production of GnRH by the hypothalamus and its transport and release into the hypothalamic-hypophyseal portal vessels, ii) the GnRH-mediated stimulation of

FSH and LH secretion by gonadotrophs within the anterior pituitary, iii) the interactions of blood-borne FSH and LH with their receptors on target cells within the ovary and iv) the reciprocal negative feedback actions of ovarian steroids on GnRH secretion by the hypothalamus and on FSH and LH secretion by the pituitary gonadotrophs (1, 2).

GnRH secretion by the hypothalamus and its transport to the anterior pituitary gland is absolutely essential for the production of normal menstrual cycles. Experimental perturbation of GnRH signaling in monkeys by lesioning of GnRH-producing neurons in the hypothalamus (3), by disrupting the transit of GnRH to the pituitary gland by sectioning of the hypothalamic-pituitary portal vessels (4) or by disrupting GnRH signaling at the pituitary with GnRH agonists or antagonists causes amenorrhea (5, 6). Likewise, genetic mutations or other disorders in humans that disrupt normal GnRH secretion cause amenorrhea (7). However, although GnRH production and release is absolutely required for normal ovarian function, it does not appear that regulation of GnRH secretion is essential for normal menstrual cycles. Thus, in monkeys rendered anovulatory by lesions of the hypothalamus or by disruption of the hypothalamic-hypophyseal portal vessels, normal menstrual cycles can be re-established by the administration of a fixed amount of exogenous GnRH in a pulsatile manner with an unvarying one pulse per hr frequency (8). Likewise in humans with hypothalamic amenorrhea, administration of GnRH at a unvarying frequency of one pule per ninety minutes is sufficient to produce normal menstrual cycles and establish pregnancies (9). Collectively, these observations indicate that the major site of regulation of FSH and LH secretion, hence menstrual cyclicity, is at the level of the pituitary gland.

Characteristics of follicular development

As illustrated in Figure 1, the maturation of a preovulatory follicle is the culmination of a lengthy process in which the growth of dormant primordial follicles is initiated when the granulosa cells begin to proliferate and form preantral follicles. Granulosa cell proliferation

continues and the number of granulosa cell layers increase as the preantral follicle continues to grow. After the preantral follicle attains six-seven granulosa cell layers, the theca interna layer becomes evident and the antral cavity begins to past beyond the early antral stage and death (atresia) of the follicle ensues. Although the follicular phase of the normal menstrual cycle typically lasts 14 days, it has been estimated that the length of time required for the growth of a follicle from the primordial stage to the large preantral stage takes in excess of 150 days (10). Thus, a follicle which will ovulate in any one menstrual cycle will have actually initiated growth at least five menstrual cycles earlier.

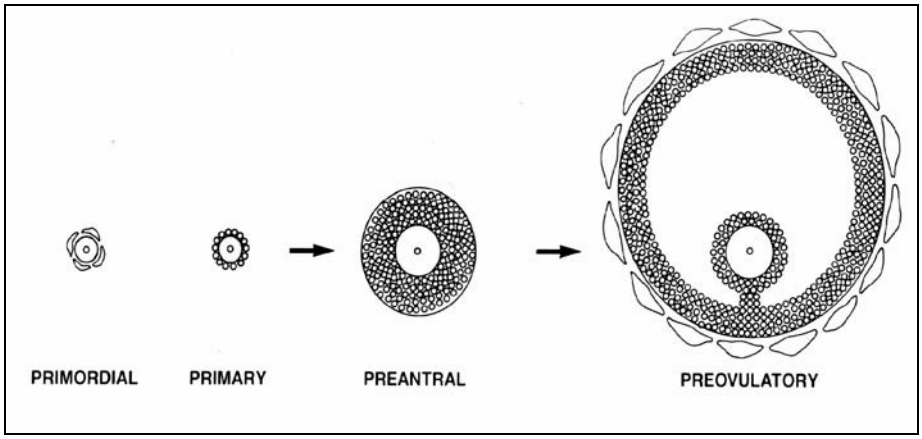

Figure 1. Regulation of follicular development. The early stages of follicular development from the primordial to the large preantral stages do not appear to require cyclic fluctuations in the secretion of FSH and LH. The role of FSH is to stimulate the formation of a large preovulatory follicle which, because of its FSH-dependent maturation, is capable of ovulation and forming a corpus luteum in response to the midcycle surge of LH.

In primates, preantral and early antral follicles are present in ovaries throughout the follicular as well as the luteal phase and even prior to the onset of puberty (11, 12). It is generally accepted that the early stages of follicular growth from the primordial stage to the early antral stage is independent of the pituitary gonadotropins FSH and LH. In non-human primates, autoradiographical studies have shown

that preantral follicles possess FSH receptors, but not LH receptors, similar to that previously show in rats (13, 14). Because early antral follicles are FSH responsive and are present throughout the menstrual cycle as a product of the continual supply of preantral follicles from the primordial pool, it is generally accepted that process of preantral folliculogenesis serves to provide a continuously available source of growing follicles for final maturation to the preovulatory stage when provided with the appropriate hormonal support, which, in primates, begins upon the onset of puberty.

The roles of FSH and LH in ovarian follicular maturation

Studies using autoradiography demonstrated that receptors for FSH were confined specifically to the granulosa cells of follicles while LH receptors were uniformly present on theca cells of maturing follicles (13). This observation was somewhat perplexing because it was well established that LH is the principal hormone involved in the transformation of the follicle into a corpus luteum yet granulosa cells, the precursor cells of the corpus luteum, did not possess LH receptors. The observations by Channing and Kammerman (15) that LH receptors on granulosa cells isolated from small follicles but are present on granulosa cells removed from large follicles demonstrated that the appearance of the LH receptor on granulosa cells is associated with advanced follicle development. The studies of Zeleznik et al (13) were the first demonstrate that the appearance of LH receptors on granulosa cells was the result of FSH stimulation. It is now generally accepted that FSH is the principal (if not the sole) hormone involved in the initiation of preovulatory follicle development. As a result of FSH stimulation, granulosa cells acquire LH receptors which render the follicle responsive to LH and able to ovulate and form a corpus luteum in response to the midcycle LH surge. In addition, FSH stimulation also results in the induction of the aromatase enzyme in granulosa cells (16). This enzyme, which is the rate-limiting step in estradiol biosynthesis, allows the follicle to metabolize androgens (androstenedione and testosterone), produced by the theca cells under LH stimulation, into estrogen. The progressive rise in serum estrogen

levels during the mid through late follicular phase of the menstrual cycle is the result of the acquisition of aromatase by the developing follicle.

Follicular maturation and the selection of the preovulatory follicle

During the follicular phase of the menstrual cycle of humans and non-human primates, a single follicle usually matures and releases its oocyte for possible fertilization and establishment of pregnancy. In simple terms, the process of follicle selection must involve a mechanism by which the presence of a maturing follicle inhibits the maturation of other follicles without compromising its own development. Studies in both monkeys (17) and humans (18) have shown that the absolute plasma concentration of FSH is critical for both the initiation of preovulatory follicle development at the beginning of the menstrual cycle as well as for the selection process itself. Figure 2A summarizes the current views on the role of FSH on the initiation of follicle development and follicle selection. During the luteal phase of the menstrual cycle serum concentrations of FSH are maintained below threshold levels and follicular development does not proceed beyond the early antral stages (19, 20). Upon the regression of the corpus luteum, feedback inhibition of gonadotropin secretion is relieved, FSH secretion increases and plasma concentrations of FSH rise above the threshold necessary to initiate preovulatory follicular development. As a direct consequence of FSH stimulation, aromatase is induced in the granulosa cells of the FSH stimulated follicle and plasma concentrations of estradiol increase. The increase in serum estradiol levels suppresses FSH secretion below threshold levels and the maturation of other lesser mature follicles is curtailed.

Thus, follicle selection in humans and subhuman primates involves a process by which the presence of a maturing follicle inhibits the maturation of other follicles by suppressing FSH secretion such that plasma FSH concentrations fall below that necessary to maintain the growth of less mature follicles. This finding indicates that there must be specific functional changes in the FSH-stimulated follicle that protects it from the fall in circulating FSH concentrations. As noted