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Role of the Gonadotropin-Releasing Hormone (GnRH) Receptor in Determination of Ovulation Rate Between Lines of Swine

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Summary and Implications

Litter size plays a major role in the economics of swine production. Even modest increases in average litter size can have considerable effects on overall profitability. Two major components of litter size, 1) ovulation rate, and 2) embryonic survival, have been used in a selection index project ongoing for several generations at the University of Nebraska-Lincoln (UNL). To better understand the mechanisms of one component, ovulation rate, we are investigating the role of the gonadotropin-releasing hormone (GnRH) receptor in determination of this important trait. Although other factors may influence determination of ovulation rate, this receptor has an established physiological importance to the processes comprising ovulation rate, recruitment of follicles and regulation of ovulation. In addition, the GnRH receptor gene is located near a chromosomal marker for ovulation rate in the pig, providing a genetic rationale to study this receptor. Recently, the sequence for the porcine GnRH receptor gene has been determined, allowing comparisons between lines of pigs divergent for ovulation rate. Identification of unique genetic changes in swine strains with increased ovulation rates, such as the Chinese Meishan and the index selection line at UNL, may allow for a better understanding of prolificacy. This critical information may also be utilized to enhance litter size in other lines of pigs and improve efficiency of pig production.

Background

Prolificacy is an important measure of productivity in the swine industry, representing an economically important trait. Two primary components of litter size are ovulation rate and uterine capacity. While uterine capacity is a critical component, it is hard to quantify and difficult to select for. Ovulation rate, on the other hand, can be improved in a number of different ways including nutrition (flushing) and hormonal treatment. However, there is very little known about the mechanisms behind increased ovulation rate.

Ovulation rate is influenced by circulating levels of follicle stimulating hormone (FSH) and luteinizing hormone (LH), also known as the gonadotropins. The production of these hormones is controlled by the reproductive axis (Figure 1), consisting of the hypothalamus, anterior pituitary gland and gonads (ovaries or testes). Specifically, gonadotropin-releasing hormone (GnRH) is released from the hypothalamus and binds to its receptor on gonadotrope cells of the anterior pituitary gland. Upon binding to its receptor, GnRH stimulates the expression of the genes that lead to the production of FSH and LH, as well as the GnRH receptor itself. The secreted gonadotropins then act on the ovaries to recruit follicles (FSH) or induce ovulation (LH). Steroids, produced by the ovaries, such as estrogen and progesterone provide feedback at the level of both the anterior pituitary gland and hypothalamus to regulate subsequent gonadotropin production.

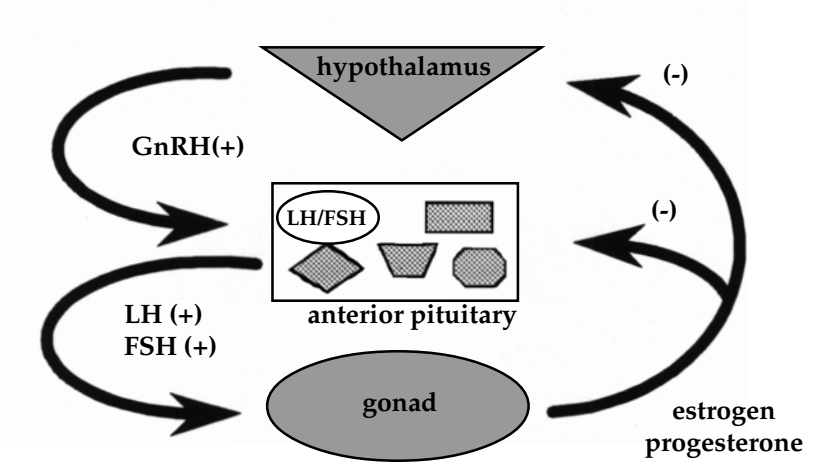

Figure 1. The Reproductive Axis. Gonadotropin-releasing hormone (GnRH) is released from the hypothalamus and binds to its receptor on gonadotrope cells of the anterior pituitary gland. The binding of GnRH to its receptor causes production and secretion of the gonadotropins, follicle-stimulating hormone (FSH) and luteinizing hormone (LH). Both FSH and LH travel through the blood and act on the gonads. Steroid hormones produced by the gonads provide feedback to both the hypothalamus and anterior pituitary gland, regulating further GnRH, LH, and FSH secretion.

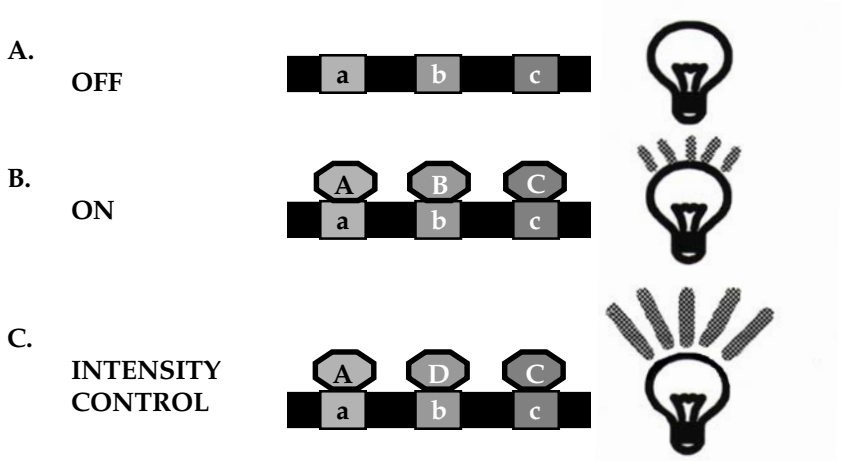

Figure 2. Regulation of gene expression. Inactivated gene (A), activated gene (B), activated gene with increased intensity (C).

Therefore, reproductive function is highly dependent on the interaction of GnRH and its receptor.

Sensitivity or number of GnRH receptors present on the anterior pituitary gland may stimulate higher levels of gonadotropin production in lines of swine with increased ovulation rates. There is evidence that elevated amounts of gonadotropins may contribute to determination of ovulation rate. Previous studies at the University of Nebraska-Lincoln (UNL) indicated that concentrations of both FSH and LH were elevated in lines of swine with increased ovulation rates (17 to 26) when compared to control lines (12 to 16). Levels of FSH could aid in determination of ovulation rate via: 1) enhanced recruitment of follicles during development, 2) decreased follicular atresia (death) during the luteal phase, or 3) increased selection during the beginning of the follicular phase. According to researchers at the Agricultural Research Service of the United States Department of Agriculture (USDA), an FSH surge following ovulation increased small follicle (<4 mm in diameter) populations in pigs, indicating FSH may be involved in early follicular recruitment at the start of the estrous cycle. In contrast, higher ovulation rates may be a result of prolonged exposure to FSH during follicular recruitment. Exposure to increased concentrations of FSH during the estrous cycle may improve both the viability and number of recruited follicles.

Although all of the human and mouse genes have been identified and the information made available, many of the porcine genes remain to be identified. Recently, the entire sequence for the porcine GnRH receptor gene was reported by researchers at the University of Guelph. Upon isolation of the gene, investigators at the USDA Meat Animal Research Center, have determined that it is uniquely located in a similar region as a chromosomal marker for ovulation rate. Therefore, the GnRH receptor gene is both a physiological and positional candidate for genes influencing ovulation rate in swine. This sequence allows for isolation and quantification of GnRH receptor gene expression, so comparisons can be made between lines of pigs with ovulation rate differences. Furthermore, scientists can also look for differences in the regulatory region of the gene between lines of swine with increased ovulation rates and their contemporaries. The regulatory region dictates the activation and intensity of gene expression. Regulation of gene expression can be compared to an adjustable light switch (Figure 2). If the correct components are unavailable, the gene is turned off, or inactivated (Panel A). When those components become available (Panel B), the gene is turned on, or activated. Finally, a different combination of components, such as those produced following hormonal stimulation, can increase the intensity of gene expression (Panel C).

Several differences have been reported within the regulatory region of the GnRH receptor gene between Chinese Meishan and white crossbred pigs by researchers at the University of Guelph. Females of the Chinese Meishan breed have a higher ovulation rate than occidental breeds, resulting in four to five more piglets per litter. Thus, Meishan pigs may harbor genetic differences with the potential to enhance the reproductive performance of white crossbred pigs. Consistent with the Meishan model, researchers at UNL have developed a line of pigs that was selected 11 generations for an index of ovulation rate and embryonic survival and nine more generations for increased litter size. At generation 14 the UNL index selection line of white crossbred pigs ovulated 7.4 more eggs and produced 1.1 more live born piglets per litter than unselected, control animals. Classically, genetic improvements have been accomplished by selection for desirable traits. However, genetic change per generation in each trait may be small. Identification of a gene that could be easily screened and that is correlated with ovulation rate in young animals would be of great interest for selection purposes. Given the physiological relevance and chromosomal location of the GnRH receptor gene, it may represent a potential marker to identify females with a genetic capacity to produce larger litter sizes.

Preliminary Data

Current experiments being carried out in our laboratory seek to identify differences in production of GnRH receptors between lines of swine with improved ovulation rates and standard lines. Of

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interest to us are differences in GnRH receptor gene expression, number of receptors available to bind GnRH and the affinity (attraction) of those receptors to GnRH. Our laboratory has isolated and sequenced approximately 5000 base pairs of the regulatory region for the GnRH receptor gene. Work is now underway to characterize the regulatory region of the GnRH receptor gene and identify segments of this region that promote differences in expression between lines of swine. Finally, we would like to understand how hormones, such as GnRH and estrogen, are involved in the regulation of GnRH receptors.

Implications

Determination of ovulation rate is very important to swine production, as it is a component of litter size. A modest increase in average litter size of 0.2 pigs per litter on a 10,000 sow operation could net a producer nearly $99,000 in additional profit, depending on pork prices. If differences in non- or hormonally-stimulated GnRH receptor gene expression levels between Meishan, Index, and Control lines are determined, a region of the gene may be isolated to provide a genetic test for ovulation rate. Ultimately, the unique genes from individuals with increased ovulation rates could be incorporated into transgenic swine. This would allow the opportunity to increase ovulation rate in any breed or line of pigs, while maintaining the beneficial characteristics of that breed or line. These animals would be very valuable to pork production worldwide.

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