



## Bellwether Magazine

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**T**ight profit margins are the norm in agricultural industry, and any factor decreasing production can make the difference between realizing a profit or just breaking even. Farmers raising pigs for slaughter expect a sow to produce six or more litters over her lifetime. But 8 to 10 percent of sows have a poor production record due to cystic ovaries. Initially, these animals have normal first or second litters and then, instead of showing an increase in litter size, the number of piglets decreases; the animal exhibits irregular estrus or no estrus at all. The animal is culled from the herd and the farmer begins anew with a replacement, hoping that it will not develop cystic ovaries. The disorder, curable with hormone therapy in cows, is not treatable in pigs.

"Cystic ovary disease is seen in one form or another in all mammals," said Dr. Gbolagade O. Babalola of the University of Pennsylvania School of Veterinary Medicine. "It is generally believed that cysts arise in the swine ovaries following failure of the follicles to rupture and ovulate, and that this failure is probably due to inappropriate gonadotropin stimulation." The interactions of hormones during the reproductive cycle are complex, and Dr. Babalola is studying the levels of ten steroid hormones in the follicles of pigs as part of the effort to determine the mechanism causing cystic ovary disease.

The follicle houses the female gamete and nourishes it through the production of follicular steroids and other factors. These growth factors, steroids and other compounds, in turn, are necessary for normal development and maturation of the ovarian follicles. Follicles go through several stages prior to maturation and ovulation when the ovum is released. Following ovulation, the follicle becomes a corpus luteum, a gland which produces mainly progesterone, a hormone needed to maintain pregnancy. The stages of follicle maturation are controlled by hormones; some of these are produced by the pituitary gland and others originate in the ovarian follicles. A complete interplay of hormonal action exists between the ovarian and pituitary hormones.

Dr. Babalola is studying the steroid hormone production capacity of ovarian follicles. "As a necessary first step in studying cystic follicles, we had to establish normal hormonal levels in healthy follicles," Dr. Babalola said. "It was basic research but necessary to identify any hormonal abnormalities in the cystic follicles." Pigs, during each reproductive cycle, produce a number of ova. It is not unusual to find cystic and healthy follicles in the same animal.

Previously, steroid hormone levels in healthy and cystic swine ovaries have not been completely characterized. In Dr. Babalola's project, 10 steroid hormones present in the ovarian follicles of sows were measured by multiple, simultaneous radioimmunoassay techniques. The hormones were: pregnenolone, progesterone,  $20\alpha$ -dihydroprogesterone,  $17\alpha$ -hydroxyprogesterone, androstenedione,  $5\alpha$ -dihydrotestosterone, dehydroepiandrosterone, testosterone, estrone, and estradiol- $17\beta$ . These steroids represent three major groups of gonadal (reproductive organ) steroids—progestins, androgens, and estrogens.

"We obtained the ovaries from pigs slaughtered at a Philadelphia slaughterhouse. They were removed from the carcass within one hour of slaughter, cooled and taken back to the laboratory for examination." Ovaries from animals which had cycled at least once were studied to determine the normal levels of ten steroid hormones in healthy follicles. "We examined the ovaries and assigned them to functional phases of the estrous cycle," he said. "It was divided into the luteal and follicular phases, each subdivided into early, mid, and late stages, and ovulatory stage ovaries. The size of normal follicles was determined and then the follicular fluid was aspirated and pooled for each animal. The fluid was then prepared for radioimmunoassay and analyzed for the ten steroids."

Dr. Babalola found that the progesterone concentration in follicles in the early follicular phase was significantly higher than in those in the mid or late luteal stages. He also found that other progestins were present in significantly higher concentrations in the late follicular stages than in the mid luteal or the mid follicular stages. Androgens, another group of steroid hormones, were low during the early stages of the ovarian cycle, but during the mid and late follicular stages, there was a dramatic increase in the level of three androgens. Estrogens showed the same pattern as androgens.

"We found significantly lower levels of all androgens and estrogens and lower levels of progestins in ovulatory follicles, but the progestin decrease was less dramatic than that of androgens and estrogens. At ovulation, steroids declined significantly. Progestin levels declined 45 to 70 percent, and androgens and estrogens were reduced by more than 90 percent. It was



Cystic follicle vs. normal follicle.

Normal ovary

Cystic ovary.

also found that  $20\alpha$ -dihydroprogesterone did not decrease; rather, it was significantly elevated. This aspect of our finding is of importance as it is of potential applicability as ovulation predictor."

Once data were compiled for steroid hormone levels in healthy follicles, the same process was followed to collect this information for cystic follicles. "We found a distinct steroidogenic aberration in the cystic ovarian follicles," he said. "This was characterized by excessive levels of progesterone and a profound deficiency of androgens and estrogens. Although progesterone was the major progestin found in all follicles, it only accounted for 20 to 50 percent of all steroids measured in healthy follicles. In contrast, 97 percent of the steroid concentrations in cystic follicles was progesterone. Furthermore, the sum of all androgens and estrogens made up less than 0.2 percent of all steroids measured in the cystic follicles, a proportion which was found to be highly significantly different when compared to the 10 percent found within the low level control small follicles."

Dr. Babalola explained that various steroid hormones are formed in follicles by enzyme activity. "It appears that the enzymes transforming progesterone into

androgens and estrogens are defective in cystic follicles." At this point, it is not known whether these enzyme defects lead to the development of cysts or whether the cysts cause this condition. Further studies are needed. Currently, Dr. Babalola is measuring these enzymes in the tissues of cystic follicles, and it appears that the enzyme deficiency exists in the tissues, also.

Dr. Babalola's studies are supported by a USDA Formula Fund. Dr. Babalola has a great interest in reproduction. He received his veterinary degree from the University of Ibadan, Nigeria, in 1978 and began teaching at that university in 1980 in the Department of Surgery and Reproduction in the Faculty of Veterinary Medicine. In 1983, he came to Penn to study surgery. He is now a member of the Graduate Group of Comparative Medical Sciences at the University and is working toward his Ph.D. His project on the study of porcine cystic ovaries is under the direction of Dr. Bernard H. Shapiro, associate professor of biochemistry and biology.

"This work has relevance for me. Pork is an important source of animal protein in many parts of the world," he said. "It is a major food source in the southern part of my country. Perhaps we can find the cause of the disease and then develop a treatment to prevent the losses incurred by this disorder." When Dr. Babalola completes his studies, he will be the first veterinarian from Nigeria to receive a Ph.D. degree from the University of Pennsylvania.