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Effect of uterine size on fertility of lactating dairy cows

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ABSTRACT

There are multiple reasons for reduced fertility in lactating dairy cows. We hypothesized that one cause of reduced fertility could be the overall size of the reproductive tract, particularly the uterus, given well-established uterine functions in many aspects of the reproductive process. Thus, the objectives of this study were to evaluate the variability in uterine size in primiparous and multiparous dairy cows and to analyze whether there was an association between uterine size and fertility, particularly within a given parity. Lactating Holstein dairy cows (n = 704) were synchronized to receive timed artificial insemination (TAI) on Day 81 ± 3 of lactation by using the Double-Ovsynch protocol (GnRH-7d-PGF-3d-GnRH-7d-GnRH-7d-PGF-56h-GnRH-16h-TAI). At the time of the last injection of PGF, uterine diameter was determined at the greater curvature using ultrasound, uterine length was determined by rectal palpation, and uterine volume was calculated from these two measurements. Blood samples were also taken to measure progesterone to assure synchronization of all cows used in the final analysis (n = 616; primiparous, n = 289; multiparous, n = 327). Primiparous cows had greater percentage pregnant/AI (P/AI) compared to multiparous cows (49.8% vs. 39.1% at 67 days of pregnancy diagnosis, P = 0.009). Diameter, length, and volume of the uterus were larger in multiparous than in primiparous cows (P < 0.001). For multiparous cows, uterine diameter and volume were smaller in cows that became pregnant compared to cows that were not pregnant to the TAI with a similar tendency observed in primiparous cows. Logistic regression and quartile analysis also showed that as uterine volume increased, there was decreased P/AI in either primiparous or multiparous cows. Thus, there is a negative association between uterine size and fertility in lactating dairy cows with a larger uterus associated with reduced fertility, particularly for multiparous cows.

1. Introduction

Reproductive efficiency in lactating dairy cows is not optimal due to multiple causes [1]. Some of these causes have been extensively studied including anovulation [2,3], problems with detection of estrus [4], reduced fertilization [5,6], degeneration of the embryo during the first week after artificial insemination (AI) [6–8], inadequate rescue of the CL between Days 17 and 24 of pregnancy [9,10], embryonic mortality (Days 27–60; [11,12]), and later fetal losses [13,14]. The practical conditions and physiology or pathophysiology that give rise to these reproductive obstacles are becoming better defined with clear roles for altered hormonal concentrations, particularly due to increased steroid metabolism [15], altered metabolic environment and changes in body condition [16–19], disease-related problems reducing reproductive efficiency [20], and altered oocyte competence [21] perhaps related to

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increased follicle size [22] or reduced circulating progesterone (P4) concentrations [23]. In addition to these factors, the role of the reproductive tract, particularly the uterus, in the reproductive problems of lactating dairy cows has been the subject of substantial research [24–26]. It is clear that clinical metritis and clinical or subclinical endometritis can reduce fertility in lactating dairy cows [27]. However, the roles of other anatomical or physiological alterations in the uterus on fertility of lactating dairy cows have not yet been fully explored. In our on-going fertility trials, we noticed substantial variation in size of the uterus in lactating dairy cows, particularly between parities and breeds, and were intrigued with the possibility that uterine size might be related to fertility of lactating dairy cows.

Uterine size has been previously evaluated in multiple species. In humans, an increase in uterine size [28] or umbilical cord length [29], as a measure of uterine size, has been related to increasing parity or age but has not yet been clearly associated with fertility. In swine, uterine capacity was increased with age of the sow, whereas uterine length and weight have been positively related to uterine capacity as measured by the number of fetuses [30]. Early studies in beef cattle [31] reported the dramatic decrease in external diameter of the previously gravid uterine horn from 14.0 cm to 3.9 cm between Days 7 and 28 after calving, termed uterine involution. This process was reported to be complete by Day 28 after calving [31]. In addition, diameter of the uterine horns, measured postmortem, was highly correlated ($r = 0.94$) with uterine weight [31]. Similarly, in healthy dairy cattle, uterine involution was complete by 28 days postpartum as assessed by rectal palpation using a 6-point reproductive tract scoring system [32]. In cows with uterine disease, the uterine horns were larger during the early postpartum period but had decreased to a similar size as cows without uterine disease by Days 18 (determined by ultrasound) or 25 (determined by rectal palpation) after calving with uterine involution completed by Day 25 after calving in both groups [33]. Earlier uterine involution was reported when follicular development was limited either by using implants of deslorelin, a GnRH analog [34], or transvaginal follicular aspiration [35]. Thus, research on uterine size in cattle has been primarily related to timing of uterine involution and presence of uterine disease. However, a preliminary report [36] indicated that the overall size of the reproductive tract near the time of AI may be related to fertility of dairy cattle. The mentioned report used a simple 3-point scale (1: small uterus; 3: large uterus) to evaluate size of the reproductive tract and found that primiparous cows had the lowest percentage of cows with a large uterus (4.73%), whereas older cows had a greater percentage (12.94%) of cows with a larger uterus. Of particular interest, cows with a reproductive tract score of 3 (large uterus) had reduced conception rates (24.41%) compared to cows with scores 1 (38.65%) or 2 (34.26%). The results of this study were confounded due to intermixing of parities in the analysis of the effect of uterine score with fertility. Nevertheless, this preliminary result provides a practical rationale for a more detailed study of the relationship between uterine size and fertility in lactating dairy cows.

Physiologically, it seems logical that a larger uterine size in dairy cattle could be associated with reduced fertility. First, a larger reproductive tract could reduce fertilization. Failure of sperm transport in the female reproductive tract accounts for most fertilization failure [37]. Consistent with the idea that a large uterus could reduce fertilization, there have been improvements in fertilization with deep-horn insemination in some [38–40] but not all studies [41–48]. A second physiologic mechanism linking uterine size with fertility could be reductions in rescue of the CL during early pregnancy in cows with a larger uterus. Early studies showed that hysterectomy prolonged the life span of the CL when performed completely but not if part of the uterine tissue remained after surgery [49]. Similarly, elegant studies in pregnant ewes [50] reported that isolation of the embryo from exposure to even a small portion of the uterus, by using sutures, led to CL regression. This result shows the need for a local action of the embryo on all parts of the ipsilateral uterine horn to produce efficient rescue of the CL during pregnancy. It is now well known that interferon-tau is the primary signal that modulates the cross talk between the embryo and uterine endometrium [51], although other molecules may also be involved [52,53]. Theoretically, lack of exposure of the entire uterine horn to interferon-tau could lead to inappropriate CL regression in a pregnant cow with a larger uterus. Thus, previous physiological research also provides further rational for evaluating the association of uterine size with fertility in lactating dairy cows.

Therefore, the objective of this research was to test the hypotheses that (1) uterine size is greater in multiparous than in primiparous lactating Holstein cows and (2) within each parity, the number of pregnancies per artificial insemination (P/AI) would decrease with increases in uterine size. Previous studies have measured uterine size: at slaughter [31], using rectal palpation with simple reproductive tract scoring systems, 1 to 6 [54] or 1 to 3 [36], or with a single evaluation of uterine diameter by ultrasound [55]. In this study, we decided against using a categorical classification of uterine size and instead used transrectal ultrasound to determine diameter of each of the uterine horns and hand palpation to determine uterine length to allow calculation of uterine volume. In addition, to have all cows inseminated near the same time postpartum ($\pm$3 days) and to provide a uniform hormonal environment and size of the ovulatory follicle, the Double-Ovsynch protocol [56,57] was used in all cows. Thus, fertility in this study was defined as the percentage of cows that became pregnant to a fixed-time artificial insemination (FTAI) after a hormonal treatment program that provides highly synchronized patterns of ovarian dynamics, circulating reproductive hormones, and time of ovulation. No attempt was made to relate uterine size to the numerous other aspects of fertility, such as expression of estrus and fertility after AI to an estrus, which may be critical for reproductive efficiency in many commercial dairy operations.

2. Materials and methods

2.1. Animals and management

The study was conducted on a commercial dairy farm in Southeast Wisconsin (Nehls Brothers Farms, Juneau, WI, USA) between September and February 2013. Cows on this