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Effect of mastitis on luteal function and pregnancy rates in buffaloes

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ABSTRACT

The aim of this study was to investigate the effects of mastitis on CL development and function and pregnancy rate in buffaloes. Sixty-six buffaloes (*Bubalus bubalus*) reared in a commercial farm at El-Beheira governorate, north of Egypt were used in this study. According to the visual observation of milk, physical examination of the udder and actual somatic cell count in milk, buffalo cows were divided into three groups: without mastitis (W), n = 23; subclinical mastitis (SC), n = 18; and clinical mastitis (C), n = 25. All buffalo cows were synchronized by double dose of $PGF2_{\alpha}$ (11-day interval) and inseminated by frozen-thawed semen of fertile bull. Mean CL diameter was ultrasonically examined on Days 5, 9, 12, 16, 21, and 25 after artificial insemination (AI). Blood samples were taken on the days of ultrasonography for progesterone (P4) assay. Results indicated that pregnancy rates were lower ($P < 0.05$) in C (28.00%) and SC (55.56%) compared with W (69.57%) on Day 25 after first AI. Pregnancy rates reduced to 60.87%, 44.45%, and 16.00% in W, SC, and C, respectively, at Day 45 after insemination. Thus, the embryonic loss was 8.7%, 11.11%, and 12.00 % in W, SC, and C cows, respectively. Pregnancy rates decreased between 44.32% and 50.51% when mastitis occurred during Day −15 before to Day +30 after AI, compared with 59.22% in the uninfected cows. The diameter of CL was greater ($P < 0.05$) in W than SC and C cows starting at Day 9 postbreeding onward. Likewise, P4 concentrations on Days 9 through 25 after AI were greater ($P < 0.05$) in W cows as compared to SC and C cows. Positive correlations ($P < 0.01$) were found on Days 5, 9, 12, 16, 21, and 25 after AI between CL diameter and P4 concentrations. Similar trend was found among CL diameter, P4 concentrations, and pregnancy rate. Accordingly, incidence of mastitis revealed suppression to both CL diameter and function leading to significant reduction in pregnancy outcome of buffalo cows.

1. Introduction

Buffaloes are recognized to have economic significance among livestock animals in terms of milk and meat yields as well as work purposes [1]. Mastitis is an infection of the mammary gland, which is usually correlated with physical, chemical, and bacteriological changes in the milk and pathologic changes in the glandular tissue of the udder [2]. Bovine mastitis is an important and a persistent infection in the buffalo population culminating in economic losses; drop in milk production, increases in the cost of treatment, and culling process [3,4]. Buffaloes have some characteristics that may contribute to greater risk of mastitis. For example, the udder is more pendulous, and teats are longer in comparison with cattle [5]. Somatic cells count (SCC) is an indicator of both resistance and susceptibility of animals to mastitis and can be used to monitor the level or occurrence of subclinical mastitis in herds or

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individual cows [6]. The SCC also has been used for buffaloes in mastitis diagnosis, and in fact, it seems probable that a SCC greater than 200,000 cells/mL is an indicator of an udder infection [7,8]. Clinical mastitis is manifested by secretion of abnormal milk (i.e., watery milk, presence of flakes in milk, and so forth) and/or inflammation (i.e., redness, swelling, hardness, and so forth) of the mammary gland [9]. Subclinical mastitis (i.e., the asymptomatic inflammation of mammary tissue) is the most common form of mastitis representing 15 to 40 times higher incidence than clinical cases [10]. Subclinical mastitis is a bigger concern than clinical mastitis because it remains undetected without the use of SCC, a measure not available to regular raisers of dairy animals [11].

Luteal size and progesterone (P4) secretion is an important indicator for functional CL in buffaloes [12,13]. The development and function of the CL differs between pregnant and nonpregnant buffalo cows [13]. The rate of CL growth between Days 5 and 10 after artificial insemination (AI) could be a more accurate indicator of CL function, and a predictor of the likelihood of pregnancy in buffalo cows [14]. Also considered important was the increase in CL area between Days 15 and 20 after breeding in pregnant buffalo cows and the decrease in CL area in nonpregnant buffalo cows during same period [13]. The secretion of P4 during early luteal phase is essential for successful establishment of pregnancy [15]. Low plasma P4 concentration during early luteal phase was shown in nonpregnant buffalo cows compared with their pregnant counterparts [16]. Studies in dairy cattle have revealed associations between clinical mastitis and increased odds of abortion [17], abnormal length interservice intervals [18], and failure to become pregnant after a service [19]. Other studies have identified associations between subclinical mastitis as measured by increased individual-cow SCC and increased odds of embryonic loss [20], abortion [21], and failure to become pregnant to first service [21]. Several potential mechanisms have been proposed to explain the effect of mastitis on reproductive performance. These are comprehensively reviewed by Hansen et al. [22], but broadly encompass detrimental impact of inflammatory mediators on ovarian follicular function [23], intrauterine embryonic survival [24], decreased luteal-phase length [18], and the balance of luteolytic versus luteotrophic prostaglandins after conception [25,26]. Besides, possible reason that mastitis has an inhibition effect on gonadotropin secretion leading to reduced gonadotropin support for ovulation, oocyte maturation, folliculogenesis, and luteal function [22]. Mastitis is associated with increased secretion of cytokines that in turn can inhibit secretion of LH and reduces circulating concentrations of P4 [22]. However, virtually all the published information about the risk factors for mastitis refers to dairy breeds of cattle but little information is available for buffaloes. Although a high probability exists that these identified risk factors may also be observed among these species. Therefore, the objectives of this study were to evaluate the effects of mastitis on development and functions of the CL and consequently, its effect on pregnancy rates in buffaloes. It was hypothesized that both clinical and subclinical mastitis might be associated with a reduction in CL functionality leading to pregnancy reduction in buffaloes.

2. Materials and methods

2.1. Animals and management

Sixty-six lactating and cyclic buffalo cows (*Bubalus bubalus*) reared in a commercial farm at El-Beheira governorate, north of Egypt, where 50% of the yard area was sheltered were chosen for this study. Cows were at postpartum (average 181 ± 21.65 days) at the commencement of the experiment. Average body condition score (BCS) of cows according to Lowman et al. [27] was 3.3 ± 0.5 (range: 2.5–4). Cows were fed according to the recommendation of the National Research Council standards for buffaloes [28,29]. Briefly, buffaloes were fed on green fodder (*Trifolium alexandrinum*) with an adequate amount of concentrate mixture (maize or wheat 60%, soybean 25%, wheat bran 10%, rice bran 5%, and common salt 1%). Mineral-balanced mixture blocks and clean drinking water were offered as free choice. Cows were hand milked twice daily, and the average milk yield was 12.65 ± 2.87 kg/day.

2.2. Experimental design

Consistent with the National Mastitis Council recommendation [30] and according to the visual observation of milk, physical examination of the udder and actual SCC in milk, the buffalo cows were randomly divided into three groups. First group cows (without mastitis [W], n = 23) in which milk has a white appearance and free of flakes, clots, and other gross alterations in appearance and has SCC less than 200,000/mL before or after first AI. Second group (subclinical mastitis [SC], n = 18) cows had no clinical changes in milk; however, SCC exceeds 200,000 cells/mL before or after first AI (SCC elevated between 200,000–600,000 cells/mL). Third group (clinical mastitis [C], n = 25) cows had abnormal milk with flakes, clots, and other gross alterations plus physical inflammation of the udder (i.e., swelling, hardness, and so forth) and SCC greater than 600,000/mL before or after first AI. Each of the previously mentioned three groups was further divided into four subgroups of 15 days each according to the time of mastitis diagnosed before first AI: −30 to −16 and −15 to −1 days and after first AI: +1 to +15 and +16 to +30 days. Data from Days −1 to +1 around detected estrus and AI were not shown.

2.3. Estrus synchronization and AI

All buffalo cows were synchronized by the double dose $PGF_{2\alpha}$ at 11 days apart protocol (500 μg intramuscular, Estroplan, Cloprostenol sodium; Parnell Laboratories New Zealand Ltd., New Zealand). Before applying the synchronization protocol, an ultrasound examination was applied on all cows to identify the cyclic and noncyclic ones, on the basis of presence of a CL [31]. Based on the ovarian cyclicity, 66 cows were cyclic. After 72 hours of the second $PGF_{2\alpha}$ injection, signs of induced estrus were monitored by visual observation, teaser bull, and transrectal palpation. Based on the estrus observation, all cows were detected in estrus and had one opportunity to be AI. First AI was carried out during mid-to-late estrus, and subsequent inseminations