Global Veterinaria 18 (2): 146-150, 2017
ISSN 1992-6197

DOI: 10.5829/idosi.gv.2017.146.150

Persistent Corpus Luteum in a 9 Year-Old Afrikaner Cow: A Case Report

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Abstract: An Afrikaner beef cow was presented for reproductive tract assessment. Records showed that the cow was last served by a bull during an oestrus two years earlier and appeared to be pregnant ever since. Failure to deliver this pregnancy led the farm management to request an investigation into why an animal with an impeccable calving history of five calves and was in good body condition was failing to deliver this particular 'pregnancy'. Rectal examination revealed a soft rubbery structure on the right ovary, a doughy consistency in the uterus and a tight cervix whose lumen contained a structure which felt twig-like. Examination of the ovaries after slaughter confirmed small cysts on both ovaries, a large corpus luteum occupying almost 75% of the right ovary, pyometra and that the twig-like structure in the cervical lumen was an inspissated and crystalized mucous plug. These findings supported diagnosis of a persistent corpus luteum due to pyometra.

Key words: Post-partum anoestrus · Pyometra · Progesterone · Estradiol

INTRODUCTION

Persistent or retained corpus luteum (CL) is a condition that occurs when the CL does not undergo regression after 20 days of its formation in a non-pregnant cow [1, 2]. The condition is characterised by the presence of a large CL on the ovary, failure to return to heat, a persistently high level of progesterone in blood [3, 4] and pseudo-pregnancy [5].

Various studies reported persistent CL prevalence ranging from 11% [6], 20% [7], to 32.69% [8]. The later workers showed that persistent CL was higher in spring and winter than in autumn and summer. They also observed that there was a negative correlation between parity and the prevalence of CL. Other workers [9], however, reported conflicting positive correlation between the prevalence of persistent CL and parity of cows. Mukasa-Mugerwa, [3], reported that persistent CL occurs more on the right (5.1%) than on the left ovary (1.2%) and the condition appears to have a genetic predisposition.

The pathogenesis of retained CL is complicated and not yet fully understood but it is believed that the final pathway involves an altered uterine environment that results in diminished secretion of luteolytic prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$) and increased secretion of luteotrophic PGE_2 by the uterine wall [6, 10].

The formation, growth, maintenance and demise of the CL in cattle is governed by several anatomical and hormonal factors [6]. Anatomically, the CL of the bovine is formed by granulosa as well as the theca cells of the Graafian follicle soon after ovulation. After its formation, the CL begins to grow and attains a diameter of 18 to 20mm within 8 days. If the animal becomes pregnant from successful mating during the preceding oestrus, further growth occurs until it is 20 to 25 mm. If the animal does not fall pregnant, the CL begins to undergo reduction in size between days 18 to 20 and it disappears by day 22. The corpus albicans is its only visible remnant thereafter [6].

Physiologically, the formation and regression of the CL is controlled by 2 switch mechanisms that involve the uterus and the pituitary gland [6]. A pituitary luteotropin identical to prolactin or luteinizing hormone (LH) switches the ovary "on" at the time of ovulation and initiates the formation of the CL. The uterine endometrium, however, produces PGF2α to switch the system "off" at the end of the oestrus cycle if the animal does not receive a signal of

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pregnancy in the form of conceptus secretory proteins or interferon (IFN) tau [6]. This switching "off" process starts the demise of the CL.

The production of progesterone by a functional CL "locks" the resumption of the next oestrus cycle by negatively feeding back on pituitary production of LH even though waves of follicular maturation may continue to occur. This "lock" results in anoestrus which continues as long as there are insufficient quantities of LH, oestradiol, or follicle stimulating hormone (FSH). Some researchers have proposed that a gonadotropin releasing hormone (GnRH) and an LH surge are simultaneously required to "kick-start" the next cycle following regression of the current CL [11].

Postpartum anoestrus allows anatomical and physiological recuperation of the uterus from the rigours of pregnancy and parturition [8, 12]. This anoestrus is associated with decreasing levels of progesterone and oestradiol, infrequent and low levels of LH pulses in the absence of progesterone [10]. The low levels of oestradiol and progesterone allow for transient and recurrent increases in FSH. These FSH increases "kick-start" the growth of the first postpartum wave of ovarian follicles.

Retained CL has been associated with uterine infections (including pyometra), high milk production (post-partum), prolonged non-steroidal anti-inflammatory drug (NSAID) therapy and glucocorticoid therapy [13]. Lack of uterine involution and uterine infections have been reported to result in diminished endometrial production of PGF2α [11]. Separate reports suggested metritis, abnormal vaginal discharge, retained placenta, parity and early return to oestrus as risk factors that prolonged luteal phases and concluded that the uterus was more instrumental than with the ovary in pathogenesis of persistent CL [7].

The postpartum uterus is usually contaminated with bacteria that ascend through the open cervix resulting in endometritis, metritis and pyometra [14, 15]. In these inflammatory conditions, normal regeneration of endometrial cells which should produce PGF2α is limited by cells undergoing necrosis, inflammatory cells, granulation tissue, or fibrous connective tissue [8, 14, 15]. Consequently, the CL continues to exist unperturbed. The toxic damage to the endometrium caused by endometritis, metritis and pyometra which occurs postpartum may be worsened by dystocia and retained foetal membranes thus resulting in retention of the CL [6]. The high levels of progesterone from the persistent CL worsen the situation by decreasing the immune response to infection.

Furthermore, the growth of bacteria especially *Escherichia coli* (*E. coli*) leads to increased production of lipopolysaccharides (LPS). These LPS have a direct negative effect on endometrial secretion of PGF2α [7, 15]. The inflammatory response of the uterus to LPS also results in increased levels of PGE_2 which is luteotrophic. LPS may also have an indirect effect on CL retention through disruption of the gonadotropin surge-generating mechanism's ability to respond to a rise in oestradiol levels [16].

Accordingly, uterine distension results in lengthening of the oestrus cycle especially if it occurs during the luteal phase [17]. Accumulation of pus in the uterus (pyometra) results in uterine distension depending on the quantity of pus produced. However, it is not known whether a persistent CL causes pyometra or if it is the result of pyometra [18]. It was reported that pyometra, the presence of high levels of progesterone and a closed cervix results in pseudo-pregnancy [6].

Crowe [10] suggested that, for oestrus cycles to resume, cows should calve down in optimal body condition (body condition score; BCS; 2.75– 3.0) with post-partum body condition loss restricted to <0.5 BCS units.

The diagnosis of persistent CL in cattle can be achieved by history, rectal palpation, progesterone assay, ultrasound, trans-vaginal biopsy, endometrial biopsy and post-mortem examination [6, 7, 11, 19]. When a cycling cow is served by a bull or artificially inseminated and does not return to heat at least 42 days after service, the animal is presumed pregnant. However, if the pregnancy lasts beyond the normal gestation period, a persistent CL is suspected [13]. It is normal for a pregnancy diagnosis by rectal palpation or ultrasound [11] to be performed in commercial herds. Persistent CL can be suspected if a CL is palpated in the absence of a conceptus in the uterus. Other studies assayed progesterone to determine the presence of persistent corpora lutea [16, 19].

According to Mukasa-Mugerwa [3], treatment of persistent CL requires the injection of $PGF_{2\alpha}$ alone or in combination with GnRH 48 to 56 hours later to kick start the oestrus cycle. Many authors agree that a single injection of PGF2α is the best way to resolve persistent corpora lutea [6, 11, 12, 19]. Lashari and Tasawar [6] successfully treated 85% of all persistent CL cases with the first injection of PGF2α, a majority of which returned to normal fertility. Strüve *et al.* [19], obtained a 100% resolution of persistent CL when they used PGF2α alone. Mukasa-Mugerwa [3] has also suggested manual enucleation of the CL per rectum as an alternative method of treatment.

Case Presentation: A 9-year-old Afrikaner cow with a body condition score of 5 was presented to the veterinary team at University of Namibia's Neudamm Campus farm after breeding and non-return to estrus for 2 years and was now a candidate for culling.

On physical examination, the cow was over-conditioned but otherwise healthy. Rectal palpation results by 2 different veterinarians were inconsistent. The 1st veterinarian reported palpation of hard, 10 to 25 cm cysts on the right ovary and concluded cystic ovarian disease. The 2nd veterinarian reported a distended uterus with a doughy consistency. This veterinarian also reported an enlarged right ovary and a 'twig-like' hardening deep within the cervical lumen. Failure to reconcile the conclusions of the two veterinarians led to no suggested treatment. Instead, slaughter of the animal was suggested for reasons of salvage.

The cow was slaughtered for meat. Gross inspection of the carcass and "pluck" revealed that the carcass was over-conditioned with a back fat cover of over 3 cm. The carcass was otherwise normal and there were no obvious lesions save for those of the reproductive tract.

A veterinarian pathologist was requested and was granted permission to remove the reproductive tract for thorough examination.

On further gross post mortem examination, the right ovary (Plate B) was about 5 cm (roughly 2.5 times larger) than the left ovary (Plate A). The right ovary had one large yellow/orange body (assumed to be *CL*) that occupies 75% of the ovary. The rest of the left ovary showed small cysts of about 2 mm in diameter that were strewn all over the surface of an ovary that contained a clear follicular fluid. Some of the cysts showed a gritty surface upon incision. The surface of the left ovary also revealed numerous follicles containing clear fluid (Plate A).

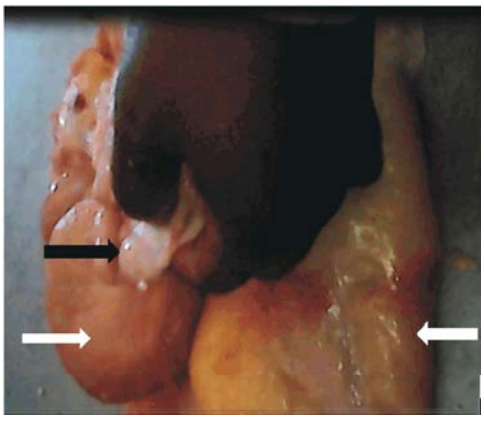

Plate A: Normal left ovary (black arrow) and the uterine horns distended by pus (white arrows)

Plate B: Right ovary incised through the centre showing a yellowish corpus luteum (white arrow) occupying more than 75 % of the ovary

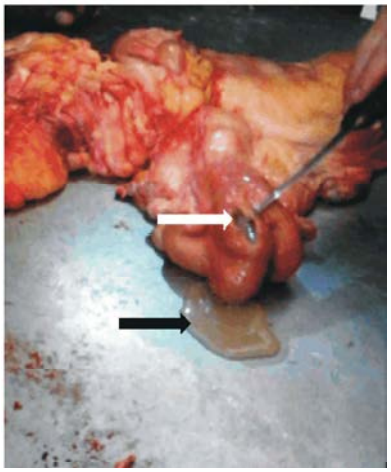

Plate C: Pus (black arrow) coming out of incision of the left uterine horn (white arrow)

A hard twig like material could be felt through the wall of the cervix. Upon opening this hardening turned out to be hard, transparent, brittle, crystalline, inspissated material occupying the lumen of the cervix following through the curves of the lumen. The uterus was slightly distended (Plate A) by a yellowish brown, viscous and malodorous uterine secretions measuring 200 ml (Plate C).

DISCUSSION

A history of prolonged non-return to service, ballottement indicating pregnancy (high body condition) and presence of a large Cl, on the right ovary, closed cervix and pyometra at post mortem examination, all confirmed a typical case of persistent CL. Persistent CL has not been previously reported in Namibia.

Persistent CL is a fairly common post-partum condition in cattle and occurs in about 11-20% of cows that give birth [6, 7]. Our finding of the persistent CL on the right ovary is in agreement with findings by Mukasa-Mugerwa [3] which reported that the condition occurred more often (5.1%) on the right ovary than the left (2.1%). In addition, the current condition's occurrence in summer when the cow was 7 years old fits the profile of persistent corpus luteum reported by others [3, 8].

The diagnosis of ovarian cyst by one of the veterinarians in our team was not entirely unexpected. Post-mortem examination of this persistent CL showed that it was somewhat flatulent and soft in the centre. Wiltbank *et al*. [11], previously pointed out the difficulty in distinguishing follicular from luteal cysts by rectal palpation. In addition, there were a number of emergent cysts on the surface of both ovaries to further confuse findings on rectal palpation.

It is difficult to conclude if the pus in the uterus was the cause or result of the retained or persistent corpus luteum in this case. Previous authors [18] have failed to clearly explain the cause/effect relationship of this situation. The presence of pus in the uterus may either suggest that the uterine environment was contaminated by bacteria before or after the closure of the cervix. It has been suggested that bacterial contamination of the uterine environment may either precede conception thus compromising the ability of the uterus to support the growing embryo and eventually leading to embryonic mortality [4]. Death and decomposition of the embryo would then attract neutrophils into the lumen of the uterus, resulting in pyometra as confirmed post-mortem.

Accumulation of pus in the uterus usually results in distension of the uterine wall, leading to further tightening of the cervix thus simulating pregnancy [6]. This scenario probably created a vicious cycle in which distension of the wall of the uterus by accumulating pus would have been read as advancing pregnancy by the endometrium. Meanwhile, the mucus plug in the cervical lumen was in place for too long and thus turned crystalline and hardened upon dehydration thus producing the confusing hard and gritty consistency in the cervix upon rectal palpation. After failure of regression, the CL persisted and continued to secrete progesterone. Increasing and persistent levels of progesterone resulted in the continued tightening and closure of cervix.

Conflicts of Interest: The authors declare that they have no financial or personal relationships which may have inappropriately influenced them in writing this article.

ACKNOWLEDGMENTS

The authors wish to thank the farm management of the permission to publish this case. We are also grateful to the University of Namibia Research and Publications Office (RPO) of the University Research and Publications Committee (URPC) for funding the publication of this manuscript.

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