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Influence of Restraint Facility on Semen Placement Site and Pregnancy Rates Following Artificial Insemination in Beef Cattle

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Abstract: The effects of restraint facility on site of semen placement and pregnancy rate following artificial insemination was investigated using twenty beef cattle. The cows had body condition score between 5.0 and 6.0 and they were divided into two equal groups 'A' (restrained with chute) and 'B' (hand-held with rope). Estrus was synchronized in the cows using the *Ovsynch* protocol. Pregnancy was evaluated using the 90 days post insemination non-return rate and confirmed with the aid of a rectal ultrasound. The result showed that cows in group 'B' were more aggressive and exhibited vigorous body movement during the insemination procedure. Also, none of the cows in group 'A' had semen placed in the vagina (V), posterior cervix (PC) and middle cervix (MC), while 70%, 20% and 10% had semen placed in the uterine body (UB), left uterine horn (LUH) and right uterine horn (RUH) with the following pregnancy rates: 85.7%, 0% and 100%, respectively. Similarly, for cows in group 'B', none had semen placed in the V, LUH and RUH, while 10%, 60% and 30% had semen placed in the PC, MC and UB with the following pregnancy rates: 0%, 50% and 66.7%, respectively. Since the goal in artificial insemination is to place semen as close as possible to the site of fertilization, the results indicate that the more efficient restraint such as reduces cow aggression facilitates correct semen placement, thereby enhancing the pregnancy rate and *vice versa*.

Key words: Restraint Facility · Semen Placement Site · Cattle Artificial Insemination · Pregnancy Rate

INTRODUCTION

Pregnancy per artificial insemination is the ratio of female animals that are pregnant relative to the number of animals inseminated. In other words, it is the product of the rates of estrus detection (or synchronization) and conception [1]. Although, with programs intended for timed artificial insemination, the synchronization rate may not be evaluated, it is however reasonable to presume that both the pregnancy and conception rates could have been better if cognizance was paid to the synchronization rate. Numerous factors/sub-factors have been known to affect/determine the percentage pregnancies in cattle breeding programs. Among these are four broad ones *i.e.* percentage herd detected in heat [2], herd fertility level [3], semen fertility level [4] and inseminator efficiency [5]. Common indicators of inseminator efficiency have bordered on: semen handling ability, pistolette loading efficiency and proficiency in manipulative procedures to place/deposit semen in the uterine body [3]. As much as these attributes are germane to inseminator efficiency, they may be no far more important in contributing to high pregnancy rates than the efficiency of cattle restraint facility. Apart from conferring comfort on the cows to be inseminated, excellent restraint facility adds value to the insemination process in that it assures the inseminator of adequate protection during the procedure. The inseminator therefore entertains no fears and hence, goes through the entire process with calm and certainty. Furthermore, the cervix acts as a major obstacle to sperm transport, hence, success in artificial insemination requires that semen is placed anterior to it [6].

This report evaluated the influence of restraint facility on pregnancy rates (%) following artificial insemination in a cattle breeding station located in Oyo State, Nigeria and stresses the fact that it affects the efficiency of the inseminator. Perception of relative

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protection is paramount both to success of the insemination as well as the inseminator who cannot 'stick a dose of semen' just at any point, along the reproductive tract.

MATERIALS AND METHODS

Study Location: The study was carried out at a small scale beef cattle station located in the Ibarapa zone, Oyo State, Nigeria. The station houses a little over 100 females cattle, of the *Bunaji* breed.

Animals and Experimental Procedure: Twenty cows, with body condition scores between 5.0 and 6.0 [7], were selected randomly and divided into two equal groups *i.e.* 'A' and 'B'. Cows in both groups were synchronized using the established *Ovsynch* protocol [8]. Briefly, this protocol consist of injecting a gonadotropin on day 0, a prostaglandin on day 7 and a gonadotropin on day 9. For a single cow, the total volume of drugs administered (intramuscular) were: 200 mcg (in two equal volumes) gonadorelin acetate (Gonabreed®, Parnell, Australia Pty. Ltd.) and 500 mcg cloprostenol sodium (Estrumate®, Intervet, Germany). The treated cows were then inseminated between 16 and 24 hours after the second gonadotropin injection [3]. Group 'A' cows were inseminated using a standard chute. Cows in group 'B' were restrained with the aid of a rope tied around both horns and thrown in-between the hind limbs to form a loop just superior to the hock joint and held firmly by the handler. Both groups of cow were synchronized such that group 'A' were ready for insemination a day after group 'B' were inseminated. Semen from the same bull which was being preserved in semen tank was used for the insemination.

The entire protocol, starting from pre-synchronization to post-insemination was carefully carried out in line with established guidelines [3]. Briefly, these included: identification of the animals to be inseminated, semen handling *i.e.* storage, thawing and loading, as well as the actual procedure of insemination. To place the semen in the uterine horns, the pistolette tip was guided into the particular uterine horn, until some form of resistance is felt, then the plunger is gently depressed, after which the pistolette is withdrawn.

Data Collection and Analysis: The region of the reproductive tract at which semen was deposited following single insemination in each cow was recorded. The temperament of the cows during the insemination process was noted either as 'aggressive' or 'docile'. The 60-day non return to estrus [9], as well as rectal ultrasound (Animal Profi 2- Draminski, Poland) imaging at 90-day post insemination were used to identify cows that had conceived. Data were summarized using descriptive statistics [10].

Guide on Research Conduct: The study was conducted under strict adherence to the principles of the care and use of farm animals in research, teaching and testing of the Canadian Council on Animal Use.

RESULTS

The result (in Table (1) showed that semen was not deposited in the vagina (V), posterior cervix (PC) and middle cervix (MC) of cows in group 'A'. In the same group 'A', seven (7), two (2) and one (1) cow(s) had semen deposited in the uterine body (UB), left uterine horn (LUH) and right uterine horn (RUH), respectively. Similarly, semen was not deposited in the V, LUH and RUH of cows in group 'B', while one (1), six (6) and three (3) cows had semen deposited in the PC, MC and UB, respectively.

The pregnancy rates (%) observed in the cows in which semen was deposited were as follows: Group 'A'- 85.7 (UB), 0 (LUH) and 100 (RUH); Group 'B'- 0 (PC), 50 (MC) and 66.7 (UB).

Plate 1 shows the characteristic sonogram of the cows identified as pregnant in the study. The sonogram shows the umbilicus, placentomes, fetus and allantoic fluid, at 90-day post insemination.

Table 1: Site of semen deposition and pregnancy rates (%) in the cows.

Portion of tract where semen was deposited	Group 'A' (%)n = 10	Group 'B' (%)n = 10
Vagina (V)	0 (0%)	0 (0%)
Posterior cervix (PC)	0 (0%)	1 (0%)
Middle cervix (MC)	0 (0%)	6 (50%)
Uterine body (UB)	7 (85.7%)	3 (66.7%)
Left Uterine horn (LUH)	2 (0%)	0 (0%)
Left Uterine horn (LUH)	2 (0%)	0 (0%)
Right Uterine horn (RUH)	1 (100%)	0 (0%)

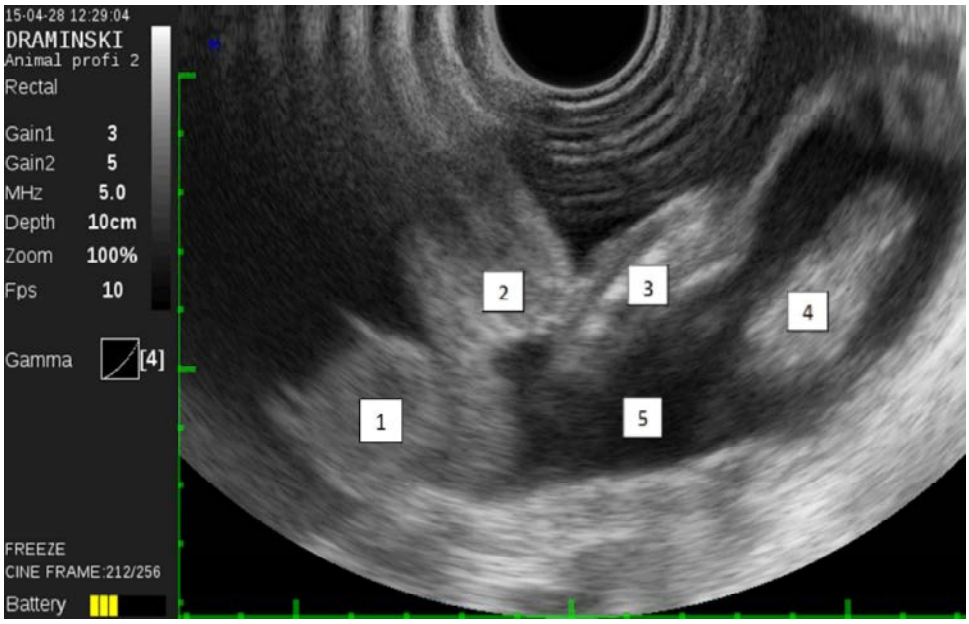

Plate 1: Characteristic sonogram of a representative pregnant cow at 90-day post insemination showing 1: Umbilicus, 2 & 4: Placentomes, 3: Fetus, 5: Allantoic fluid. (Mag. x1).

Most of the cows in group 'B' were aggressive. They attempted to injure, by kicking and vigorous body movements, the inseminator severally had to abruptly halt the procedure, only to continue after the cow calms down. The cows in group 'A' were relatively docile.

DISCUSSION

Under the conditions of this study, the results indicate that restraint facility affected the site of semen placement during artificial insemination. This is obvious in that 100% of the cows in group 'A' had inseminate placed in the uterine body and the horns while, 30%, 60% and 10% of the cows in group 'B' had the inseminate placed in the uterine body, middle and posterior cervices, respectively. According to Mufti *et al.* [11], faulty insemination techniques, with respect to poor semen placement, constitute a major factor responsible for low conception rates in cattle. Although, conflicting reports exist as to the relative advantage, on pregnancy rates, of placing semen in the uterine horns [12, 13], some authors [11], reported a 10.62% higher conception rate with semen placed in the body of the uterus over that at the middle cervix. The cervix has been identified as a major obstacle to sperm transport [14]. Hence, attempts during artificial insemination has always been to place semen as close as possible to the oviductal ampullary-isthmus junction, where fertilization takes place [15]. This way, the reduced number of spermatozoa used in artificial insemination are made to bypass biological obstacles and the distance, compared with natural mating, is reduced to facilitate fertilization [6, 16]. Since the goal of artificial insemination is to place semen further in the tract, the result of the present study suggest that restraint facility affected cows in group 'B' as only three (*i.e.* 30%) had uterine body placement. Also, none of the cows in group 'B' had uterine horn placement. It appears from the result of this study that the further semen can be placed in the reproductive tract depends on the level of restraint provided. This is further supported by the observation in the study where none of the cows in group 'A' had semen placed posterior to the uterine body. The implication of these is that all things being equal, pregnancy rates should be higher in group 'A'.

The pregnancy rate that ensued from the exercise is perhaps interesting and informative. For cows in group 'B', the single cow in which semen was placed in the posterior cervix was not found to be pregnant at both 60 and 90 days, post insemination. Placing semen in the posterior cervix is not recommended nor desirable during artificial insemination, as this would reduce chances of conception, for obvious reasons. It had been reported [17], that following natural mating, only very few selected sperm cells eventually make it up to the site of fertilization, therefore, the farther the inseminate is deposited, the lower the chances of conception [17]. The pregnancy rate (50%) obtained from cervical semen placement showed that only half of the cows were positive for pregnancy.

This rate, following single insemination is a bit better compared with common findings in recent times [18], but a bit lower compared with older reports [19]. Earlier explanations of the cervix acting as obstacle as well as the distance to site of fertilization may also account for the average rate of pregnancy obtained. Apart from physical hindrances to spermatozoa movement, other contributors to the difficulty of the sperm to traverse the cervix include: level of hydration of vagina mucus [20], microarchitecture of the mucus, especially around the cervical border [21] and the ability to initiate immune response against the sperm [22]. The factors responsible for the difference in pregnancy rates following uterine body semen placement in the two groups are not clear. However, aggression and vigorous body movements are capable of producing stress in livestock. Endocrinal evidence that stress exerts negative influence on reproduction has been reported [23]. Although, the influence and duration of exposing animals to stress, on pregnancy rate has not been studied exhaustively [24], the author thinks that if other variables determining pregnancy following artificial insemination are constant, there is a likelihood that inadequacy of restraint facility, such as allowed for vigorous body movements and show of aggressive behaviour in group 'B' cows, may contribute to this difference. Again, in group 'B', it is worthy to note that although, double the number of cows that had uterine body insemination had semen placed in the middle cervix, yet the pregnancy rate in the earlier was higher than in the latter. This finding is similar to earlier observation [11] and may only add to established fact that the cervix constitute a barrier to sperm transport. The percentage difference obtained on pregnancy rate between cervical and uterine body insemination (*i.e.* 16.7%) in group 'B' is however higher than the 10% earlier reported [25]. This earlier report and those of other later researchers prompted scientists to investigate advancing the site of semen placement towards the site of fertilization [16]. What was responsible for the zero pregnancy rate following semen placement in the left uterine horn was not clear. However, the right ovary [26], as well as the right uterine horn [27], are bigger and more active physiologically than their counterparts. According to Larsson and Larsson [28], higher activity of the right side of the reproductive tract facilitates sperm transport, thereby increasing frequency of pregnancy on the right side. These facts may account for the pregnancy rates observed in the two groups of cows in this study in which semen was placed either in the left or right uterine horns. The ovaries were not palpated prior to insemination. Only a single cow had semen placed in its right uterine horn, the pregnancy rate obtained therefore may require further investigation before it can be assigned more clinical significance. However, comparing with earlier reports on *cornual* insemination, the present result supports the observation that the rates of ovulation and pregnancy of the right side of the reproductive tract are greater than of the left side [29].

CONCLUSION

In conclusion, good restraint facility is required during artificial insemination in order to place semen closer to the site of fertilization and thereby improve the pregnancy rate and *vice versa*.

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