PREGNANCY RATES IN PHILIPPINE SWAMP BUFFALOES (CARABAOS) FOLLOWING CLITORAL STIMULATION DURING TIMED INSEMINATIONS

S. S. Capitan[1], V. G. Momongan, A. R. Obsioma and A. N. del Barrio

Institute of Animal Science, University of the Philippines at Los Baños College, Laguna 4031, Philippines

Summary

One hundred sixty three (163) animals were used in 2 × 2 and 3 × 2 factorial experiment in randomized complete block design (RCBD) to determine the effect of clitoral stimulation during timed inseminations on the fertility of Philippine swamp buffaloes (carabaos). There were 3 separate trials conducted with two treatment groups per trial; control and with 30 second clitoral stimulation after each insemination. Parity, tone of uterus and site of semen deposition were also used as criteria in subdividing the main groups.

The pregnancy rates of animals that received clitoral stimulation were significantly ($p < 0.05$ or $p < 0.01$) higher than those of control for both caraheifers (52.98 vs 31.41%) and caracows (52.68 vs 27.07%); under all uterine conditions: tone 1 (53.33 vs 31.75%), tone 2 (35.83 vs 22.82%) and tone 3 (75.65 vs 42.22%); and in both site of semen placement, cervix (43.99 vs 22.85%) and uterus (60.92% vs 37.81%). Fertility was also significantly ($p < 0.05$) higher when semen was deposited in the uterus (49.37%) than in the cervix (33.42%). Interaction effect was insignificant.

Clitoral stimulation should be utilized as a routine procedure following artificial insemination in carabaos.

(**Key Words**: Pregnancy Rate, Fertility, Clitoral Stimulation, Heat Synchronized Carabaos)

Introduction

There are various factors identified that affect the success of artificial insemination (A.I.) as an important breeding tool. These include the quality of semen used, the time of breeding, the length of time to complete insemination, the relative skill of the inseminator, the health status of the animals to be bred, and others. Inspite of the almost complete understanding of these different factors that influence the success of the technique, pregnancy rates obtained from A.I. are relatively low compared to natural breeding. This is more pronounced among heat synchronized animals. A critical investigation of the physiological effects of management of females at insemination time may help improve reproductive efficiency.

Uterine motility increased in cows exposed to the presence of bull, nuzzling by the bull, noncopulatory mounting and mounting with copulation and ejaculation (Van Demark and Hays, 1952). Likewise, an increase in intramammary pressure during massage of the vulva and cervix, and natural mating has been reported by Hays and VanDemark (1953a). The same workers (1953b) showed that oxytocin caused increased uterine activity at all stages of the estrous cycle. Results of their work indicate the involvement of a neural pathway with the release of oxytocin from stimulation of the reproductive tract at mating. The study of Robertson and Rakha (1965) showed that chlorpromazine, a neural blocking agent, blocked ovulation in sheep when administered within 2 hours after the onset of estrus. These data suggest that a neural connection between gonadotropin release and ovulation exist, at least in the species they used.

The work of Cooper et al. (1985) showed that a 30 second clitoral stimulation caused an immediate single uterine contraction, which could be repeated at 2.5 minute intervals. Nevertheless, they were unable to show any change in oxytocin concentration during intensive blood sampling following clitoral massage. They suggested a neural rather than hormonal mediation of uterine response. Earlier, Randel et al. (1975) and Short et al. (1979) reported that manual clitoral massage

[1]Address reprint requests to Dr. S. S. Capitan Institute of Animal Science, University of the Philippines at Los Baños College, Laguna 4031, Philippines.

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significantly increased pregnancy rates in cows but not in heifers.

No study along this line has been done in Philippine swamp buffaloes (carabaos), the focus of nationwide massive upgrading programs utilizing estrus synchronization and A.I. This study was undertaken to determine whether a 30 second stimulation of clitoris following timed inseminations would increase conception rate in heat synchronized carabaos.

Materials and Methods

The carabaos of Philippine Carabao Research and Development Center (PCRDC) cooperators in the various villages of Lucban, Quezon were utilized in this experiment. A total of 163 carabaos were used in three (3) separate trials. For estrus synchronization, the animals were treated with double regimen injection of prostaglandin analogue (Oestrophane) administered intramuscularly at the rate of 2 ml (0.50 mg) per animal and given 11 days apart. At predetermined time the animals were bred artificially and divided into two (2) treatment groups, namely:

Group 1 = control, no stimulation

Group 2 = 30 second clitoral stimulation after each insemination

Clitoral stimulation was done by placing the forefinger and thumb on the lower half portion of the vulva and then applying the pressure for 30 seconds. The clitoris being situated on the ventral commisure of the vulva is thus stimulated during the process.

Each group was later further subdivided according to parity, tone of the uterus at insemination time and site of semen deposition. Tone of the uterus was gauged according to rigidity, i.e. tone 1-slightly rigid, tone 2-rigid, tone 3-very rigid.

Inseminators were randomly alloted to all the experimental animals bred. Pregnancy diagnosis was done two to three months after breeding.

The data was analyzed using the analysis of variance (ANOVA) technique for a 2 × 2 and 3 × 2 factorial experiment in a randomized complete block design.

Results and Discussion

Data on pregnancy rates as affected by clitoral stimulation and parity are presented in table 1a. The effect of stimulation is positive ($p < 0.05$) for both caraheifers (52.98 vs 31.41%) and caracows (52.68 vs 27.07%). These results contradict the work reported earlier in cattle (Randel et al., 1975; Short et al., 1979) that clitoral stimulation increased pregnancy rate only in primaparous and multiparous and not in nulliparous animals. The earlier postulate that heifers seemed to become uncomfortable because it generally takes a longer time to inseminate them is not tenable from the results of the present study. On the contrary, it may be theorized that both subgroups received sufficient stimulation that made them to respond well to the treatment. The effect of parity on pregnancy rate is insignificant as further revealed by statistical analysis (table 1b).

Table 2a shows the effect of clitoral stimulation on pregnancy rate of carabaos inseminated at varying tone of the uterus. Analysis of variance (table 2b) indicated significant ($p < 0.05$) effect of stimulation under all conditions of the uterus the animals were bred: tone 1 (53.33 vs 31.75%), tone 2 (35.83 vs 22.82%) and tone 3 (75.65 vs 42.22%). Average pregnancy rates as affected by

TABLE 1a. EFFECT OF CLITORAL STIMULATION ON PREGNANCY RATE (%) OF CARAHEIFERS AND CARACOWS

Parity	Treatment Group	Trial I	Trial II	Trial III	Mean
Caraheifer	Control	11.11 (1/9)	28.57 (4/14)	54.55 (6/11)	31.41
	Clitoral stimulation	37.50 (3/8)	50.00 (5/10)	71.43 (5/7)	52.98
Caracow	Control	41.66 (5/12)	11.76 (2/17)	27.78 (5/18)	27.07
	Clitoral stimulation	58.33 (7/12)	35.71 (10/28)	64.71 (11/17)	52.68
Mean		37.15	31.33	54.61	

Figures in parentheses represent number of animals pregnant out of number of animals inseminated.

TABLE 1b. ANALYSIS OF VARIANCE

SV	DF	MS	F-Value	Prob
Trial	2	587.43	2.70	0.145
Parity (A)	1	16.15	0.07	
Treatment (B)	1	1669.46	7.67*	0.032
A × B	1	12.28	0.06	
Error	6	217.78		

* Significant ($p < 0.05$).

TABLE 2a. EFFECT OF CLITORAL STIMULATION ON PREGNANCY RATE (%) OF CARABAOS INSEMINATED AT VARYING TONE OF UTERUS

Tone of uterus	Treatment Group	Trial I	Trial II	Trial III	Mean
Tone 1	Control	66.67 (2/3)	0 (0/4)	28.57 (2/7)	31.75
	Clitoral stimulation	60.00 (3/5)	0 (0/2)	100.00 (1/1)	53.33
Tone 2	Control	14.29 (2/14)	16.67 (2/12)	37.50 (6/16)	22.82
	Clitoral stimulation	27.27 (3/11)	23.08 (3/13)	57.14 (8/14)	35.83
Tone 3	Control	50.00 (2/4)	26.67 (4/15)	50.00 (3/6)	42.22
	Clitoral stimulation	100.00 (4/4)	52.17 (12/23)	77.78 (7/9)	76.65
Mean		53.04	19.77	58.50	

Figures in parentheses represent number of animals pregnant out of number of animals inseminated.

TABLE 2b. ANALYSIS OF VARIANCE

SV	DF	MS	F-Value	Prob
Trial	2	2637.20	5.79*	0.021
Tone of uterus (A)	2	1366.85	3.00	0.095
Treatment (B)	1	2382.11	5.23*	0.045
A × B	2	174.28	0.38	
Error	10			

* Significant ($p < 0.05$).

tone of the uterus were not statistically different from each other. This observation deviates from the otherwise expected effect of uterine tone on the success rate of artificial insemination. It is, therefore, worthwhile to consider the effect that clitoral stimulation has in improving the pregnancy rate irrespective of the uterine changes.

Response to clitoral stimulation of carabaos inseminated in the cervix or in the uterus indicated significant effects to pregnancy rate of both factors (table 3a and 3b). Stimulation significantly ($p < 0.01$) improve fertility for both site of semen placement, cervix (43.99 vs 22.85%) and uterus (60.92 vs 37.81%). Likewise, fertility is significantly ($p < 0.05$) higher when semen is deposited in the uterus (49.37%) than in the cervix (33.92%). Interaction effect was, however, not significant.

Manual clitoral stimulation has been shown to hasten the LH surge and shorten interval from estrus to ovulation (Randel et al., 1973) and also improve artificial insemination pregnancy rates in cows (Randel et al., 1975). The results of the present work, however, do not permit the determination of the mechanism involved in the improvement of fertility of animals that received stimulation following artificial insemination. As suggested by Randel et al. (1973), when cows are inseminated earlier than schedule in relation to ovulation time, the stimulation would hasten ovulation. Sperm transport mechanism may be

TABLE 3a. EFFECT OF CLITORAL STIMULATION ON PREGNANCY RATE (%) OF CARABAOS INSEMINATED AT DIFFERENT SITES OF SEMEN DEPOSITON

Site of semen deposition	Treatment Group	Trial I	Trial II	Trial III	Mean	
Cervix	Control	27.78 (5/18)	10.00 (2/20)	30.77 (4/13)	22.85	> 33.42
	Clitoral stimulation	38.46 (5/13)	36.36 (4/11)	57.14 (4/7)	43.99	
Uterus	Control	33.33 (1/3)	36.36 (4/11)	43.75 (7/16)	37.81	> 49.37
	Clitoral stimulation	71.43 (5/7)	40.74 (11/27)	70.59 (12/17)	60.92	
Mean		42.75	30.87	50.56		

Figures in parentheses represent number of animals pregnant out of number of animals inseminated.

TABLE 3b. ANALYSIS OF VARIANCE

SV	DF	MS	F-Value	Prob
Trial	2	393.52	5.97*	0.037
Site of semen deposition (A)	1	763.05	11.58*	0.014
Treatment (B)	1	1468.10	22.27**	0.003
A × B	1	2.91	0.04	
Error	6	395.49		

* Significant ($p < 0.05$)
** Significant ($p < 0.01$).

the one involved and be the focus of greatest action as massage of the reproductive tract has been shown to affect uterine motility (VanDemark and Hays, 1952; Hays and VanDemark, 1953a). Whatever may be the exact mechanism of action involved, the results of this experiment produced an inexpensive and practical technology of improving artificial breeding pregnancy rate, and may justify the recommendation that clitoral stimulation be utilized as a routine procedure following artificial insemination in carabaos.

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