



Estrus Synchronization in Ugandan Cattle Herds

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ABSTRACT

This survey aimed to assess the breeding practices and pregnancy rates of cattle on farms that utilized artificial insemination (AI) after estrus synchronization in Uganda. Management and breeding data were collected from 297 farms using a semi-structured questionnaire and supported by breeding record reviews and AI technician interviews. Kruskal-Wallis tests were used to determine associations between breeding practices and pregnancy rates using the R software. Breed categories on the farms were exotic dairy and their crosses (64.7%), exotic beef and their crosses (15.1%), and indigenous breeds of Ankole longhorn and shorthorn Zebu (20.2%). Main breeding methods on the farms were natural mating (53.3%), AI after natural estrus (34.7%), and AI after estrus synchronization (11.9%). Estrus synchronization protocols used on the farms were Ovsynch (42.4%), 7-day Co-synch + progesterone-releasing intravaginal device (P4ID) (32.3%), Prostaglandin F_{2α} (PG) single injection (23.4%), and 7-day Co-synch (2.4%). Mean pregnancy rates to AI after synchronization were not significantly different ($P>0.05$) among protocols and were 33.00%, 42.65%, 36.87%, and 41.33% for 7-day Co-synch, 7-day Co-synch+P4ID, Ovsynch, and PG single injection protocols, respectively. Mean pregnancy rates were 29.9%, 40.4%, and 42.7% for indigenous breeds, exotic beef/crosses, and exotic dairy/crosses, respectively, and significantly lower in indigenous breeds than exotic dairy/crosses ($P_{\text{adj}}<0.001$) and exotic beef breeds/crosses ($P_{\text{adj}}=0.004$). Pregnancy rates were also significantly lower on farms where the main breeding method was natural mating than AI on natural heat ($P_{\text{adj}}=0.03$) and AI after synchronization ($P_{\text{adj}}=0.02$). Ovsynch, 7-day Co-synch+P4ID, PG single injection and 7-day Co-synch were the estrus synchronization protocols used in Uganda. Estrus synchronization was more common in dairy exotic cattle than in beef exotic or indigenous cattle. Pregnancy rates with synchronized AI were lower in *B. indicus* than *B. taurus* herds. Improvements in management and design of controlled reproductive studies are required to enhance the success of reproductive technologies in *B. indicus* cattle.

Keywords: Artificial Insemination, *Bos indicus*, Estrus Synchronization, Reproductive Performance.

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INTRODUCTION

For centuries, farmers worldwide have practiced animal genetic improvement through selective breeding. Crossbreeding of *B. indicus* cattle with *B.*

taurus has been proven to improve animal productivity in tropical countries by combining the climate and disease adaptation of the *B. indicus* with the high productivity traits of *B. taurus* breeds (Galukande *et*

al., 2013). To sustainably introduce high-grade *animal* genetics from all over the world into the resource-constrained tropical farming systems in Africa, artificial insemination (AI) technology is the most suitable alternative because it is efficient, affordable and takes advantage of heterosis (Leil *et al.*, 2020). The traditional AI based on natural heat is unreliable for achieving optimal reproductive performance due to the high incidences of anestrus, silent and split estrus, and management failures in heat detection (Othman *et al.*, 2023). However, when AI is combined with estrus synchronization, the herd genetic improvement rate is optimized by removing the major barriers, such as laborious estrus detection, thus enabling the recruitment and breeding of a large number of cows at once (Islam, 2011; Lamb *et al.*, 2016).

Several estrus synchronization protocols have been used in cattle breeding. These range from prostaglandin F2 α (PG) or its analogues, given as a single injection (PG single injection) or twice, 10-14 days apart; progesterone, either in feed or as P4ID (Islam, 2011) and a combination of the two (Lucy *et al.*, 2001). Others include progesterone-GnRH protocols such as the Ovsynch, Co-synch (Kesler and Constantaras, 2004) and Double Ovsynch protocols (Souza *et al.*, 2008). There are also Co-synch+P4ID protocols, such as the 7-day Co-synch+P4ID protocol and 5-day Co-synch+P4ID protocols, including Bee synch I and Bee synch II protocols (Bridges *et al.*, 2014; Bonacker *et al.*, 2020; Williams and Stanko, 2020).

Recent studies have shown that the pregnancy rate of inseminated cows varied inconsistently across estrus synchronization protocols and among different cattle breeds. Most of the protocols produced acceptable results in *B. taurus* (Bridges *et al.*, 2008; Lamb and Mercadante, 2016) but not in *B. indicus* cows (Saldarriaga *et al.*, 2007; Zuluaga *et al.*, 2010). These findings could be attributed to differences in reproductive endocrinology and physiology between *B. indicus* and *B. taurus* cattle. (Bó *et al.*, 2003; Landaeta-Hernández *et al.*, 2002).

In Uganda, AI in cattle was introduced in 1959, but its use has mainly been limited to dairy farms in peri-urban areas (Mugisha *et al.*, 2014). Moreover, six decades later, over 77% of the national cattle herd is still comprised of indigenous *B. indicus* breeds (Uganda Bureau of Statistics [UBOS], 2021). *Bos indicus* cattle are characterized by low milk yields, slow growth rates, and low carcass weights (Endris, 2017; Mekonnen *et al.*, 2020). Consequently, Uganda's livestock production contributes only 4.3% to the gross domestic product despite more than half (58%) of the country's households depending on livestock for their livelihood (Food and Agriculture Organization [FAO], 2019).

To address this gap, the government of Uganda introduced countrywide cattle crossbreeding program in 2019, which involves mass AI based on estrus synchronization. However, to date, information is lacking on the characteristics and reproductive performance of cattle bred by AI after estrus synchronization in Uganda, yet there exist inherent differences in effectiveness between estrus synchronization protocols. Furthermore, the effectiveness of a single synchronization protocol may vary depending on cattle breed and management factors. This study therefore aimed to evaluate the reproductive performance of cattle bred through AI following estrus synchronization in Uganda. Specifically, it aimed to determine the estrus synchronization protocols in use, their pregnancy rates and determinants in Uganda. Information on the effectiveness of protocols in the Ugandan cattle breeds and management context, if available, would significantly guide in the design and implementation of successful breeding programs.

MATERIALS AND METHODS

Study area and design

A cross-sectional survey was conducted from June 2022 to May 2023, covering 25 districts across the four geographical regions of Uganda, namely: Northern - Gulu, Oyam, Lira, Kole, Nwoya, Pader, Adjumani, Moyo, and Arua; Eastern – Serere, Ngora, Kumi, Katakwi, Jinja, and Kamuli; Central – Kampala, Wakiso, Mukono, and Buikwe; and Western – Mbarara, Kiruhura, Ibanda, Kabarole, Bunyangabo, and Kibale. This design was chosen to account for potential regional variations in cattle breeds, climate, management systems and farm practices that could influence reproductive outcomes.

Sample size determination and sampling technique

The target population consisted of all cattle farms in the four regions that had implemented AI after estrus synchronization in the previous two years. The sampling frame was derived from government records, which identified a total (N) of 800 farms that met the criterion. A minimum sample size of 267 farms was obtained using the formula (Naing, 2003): $n = N / (1 + N(e)^2)$, where: n = required sample size (number of farms); N = estimated total number of cattle farms that utilized estrus synchronization and AI; and e = desired sampling error (0.05). However, data was collected from a total of 297 farms to account for potential non-response and to enhance the robustness of the data. A proportionate stratified random sampling technique was employed to select the farms, where allocation of sample size to each region was based on the proportion of eligible farms in that region relative to the total population size (N=800). The AI technicians

considered for interview were those who had conducted the estrus synchronization and AI on the selected farms.

Data collection method

Data were obtained from both primary and secondary sources. A semi-structured questionnaire was designed, pre-tested, and administered to respondents on the selected farms following prior informed consent by trained research assistants. The data collected from the questionnaire included the production system, breeds on the farm, main breeding method, number of cows/heifers served by AI after estrus synchronization, and the resultant number of pregnancies. Furthermore, the farm AI technicians were followed up to capture data on the type of estrus synchronization protocol used. Secondary data was obtained from farm breeding records, which provided data on the number of cattle synchronized and inseminated and the corresponding pregnancy rates. The pregnancy rates were calculated for each farm as the percentage of the pregnant proportion of cows/heifers that were synchronized and inseminated that were confirmed pregnant by per-rectal examination around 90 days after the AI (Masho et al., 2024): $\text{Pregnancy rate} = \frac{\text{number of cattle pregnant}}{\text{number of cattle inseminated}} \times 100$.

Statistical analyses

Data were analysed using R version 4.4.2 (Team, 2010). Descriptive statistics were generated using the 'BarChart' and 'PieChart' functions in the 'lessR' (Gerbing et al., 2025) and 'lattice' packages in R (Sarkar, 2021). Since the conception rate data did not meet the assumption of normality, Kruskal-Wallis tests at a 95% confidence level and $\alpha=0.05$, were employed to determine if there were statistically significant differences in the median values of conception rates across the different categories of the independent variables, and where a significant Kruskal-Wallis result was obtained, Dunn's tests were performed for pairwise comparisons between categories of the variable. The 'qqbetweenstats' function in the 'ggstatsplot' package in R (Patil, 2021) was used to execute the Kruskal-Wallis test and where significant differences were detected, this function automatically conducted post-hoc comparisons at once. For example, the test to determine if there were significant differences in conception rates between synchronization protocols was: `ggbetweenstats(data= data, x=Protocol, y=conception rate, type="nonparametric")`. Tables, bar charts and a combination of violin plot, box plot and jittered data points were used to summarize the statistics.

RESULTS

Cattle production system, breed, and herd size

Data was collected from a total of 297 farms of which 113 (38.0%) were in the northern region, 88 (29.6%) in the Eastern region, 67 (22.6%) in Western

and 29 (9.8%) in the Central part of Uganda. The breed categories on the farms were: exotic dairy and their crosses with local breeds, 64.7% (189/292); exotic beef and their crosses with local breeds, 15.1% (44/292); and indigenous breeds, 20.2% (59/292). The distribution of cattle breed categories by region is presented in Fig.1.

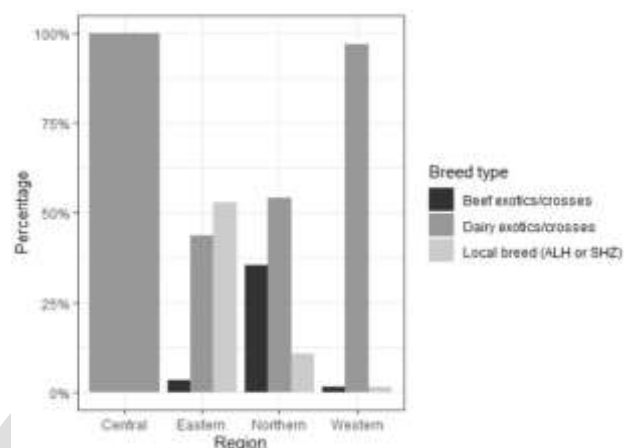


Fig.1: Regional distribution of cattle breed categories on farms that utilized estrus synchronization and AI. All sampled farms in Central and the majority in Western regions had exotic dairy/crosses, while those in the Eastern had mainly indigenous breeds and exotic dairy/crosses. Northern region farms exotic dairy/crosses, exotic beef/crosses, and indigenous breeds in that order.

The production systems of the studied farms were: extensive (43.5%; 128/294), semi-intensive (40.8%; 120/294), and intensive management system (15.6%; 46/294). On 54.5% (156/286) of farms, herds comprised 30 or fewer heads of cattle (small herds), while 17.5% (50/286) of the farms had 31 - 50 heads of cattle (medium herds), and 28.0% (80/286) farms had herds of more than 50 heads of cattle (large herds). The distribution of herd size by region is shown in Fig.2.

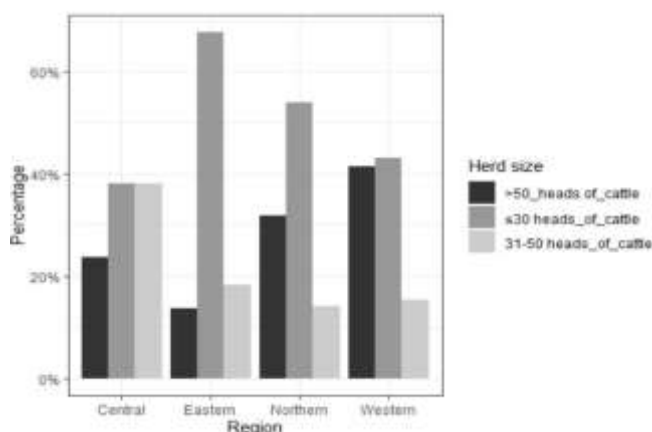


Fig.2: Regional variation of herd size on farms that utilized estrus synchronization and AI. Eastern and Northern regions had similar proportions of farms with small and medium herd sizes. The largest herds of more than 50 heads of cattle are Northern followed by the Western and Eastern regions, respectively. Central region farms had the lowest number of farms studied, comprising all three categories of herd sizes in similar proportions.

Breeding methods on the farms

Data on breeding methods used on the farms were collected from a total of 285 farms. Of those, 152 (53.33%) reported natural mating as their main breeding method, while 99 (34.74%) reported mainly using AI on natural heat, and only 34 (11.92%) were using AI after estrus synchronization as their main breeding method. The distribution of main breeding methods on cattle farms stratified by location is presented in **Fig.3**. Natural mating was the main breeding method on majority of farms in the Northern and Eastern regions, while AI on natural heat was the predominant breeding method in central Uganda. In the Western region, all three breeding methods were utilized in similar proportions.

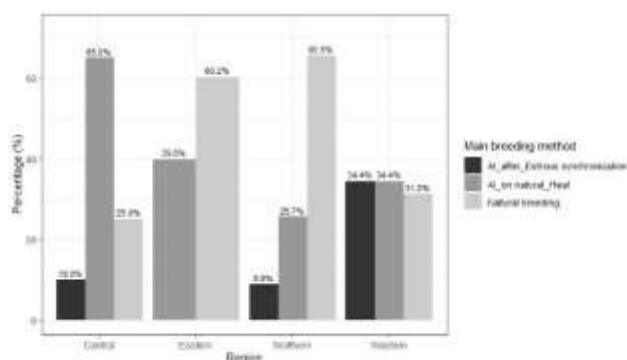


Fig. 3: Main breeding method on the farms studied. Whereas natural mating was the most common breeding method on most Eastern and Northern region farms, AI after natural heat was predominant on farms in the Central region, while the three breeding methods were used at similar proportions on farms in Western Uganda. Breeding by AI with or without estrus synchronization was the main breeding method for the majority of Western region farms. In the Eastern region, no farm considered AI after estrus synchronization as the main breeding method, while only a few farms in Central and Northern regions had considered AI after estrus synchronization as the main breeding method.

Time of AI after estrus detection

The time at which AI was conducted with reference to the first observation of estrus signs by farmers was variable. For 14.7% (37/252) of the farms, inseminations were conducted within 7 h of observation of estrus, while for 36.1% (91/230) of the farms, AI was conducted 8-12 h after observation of estrus, and between 12 – 24 h for 31.0% (78/252) of the farms. A substantial number of farmers (18.3%; 46/230) reported incidences where no technician responded to their insemination requests.

Estrus synchronization protocols used on the farms

The estrus synchronization protocols used on the farms were: Ovsynch, 42.4% (126/297); 7-day Co-synch+P4ID, 32.3% (96/297); PG single injection, 23.4% (68/297); and 7-day Co-synch, 2.4% (7/297). The use of these four protocols varied with location and

herd size, cattle breed, and production system, as presented in **Table 1**.

In the Northern region, the PG single injection protocol was the predominant method in use, while the 7-day Cosynch+P4ID protocol was predominant in the Western and Central regions, and the Ovsynch was predominant in the farms of the Eastern region. There was a tendency towards the more extensive application of 7-day Cosynch+P4ID and Ovsynch on extensive and semi-intensive farms. The majority of the indigenous cattle herds were synchronized using the Ovsynch protocol.

Farmers' perspectives on reasons for adaptation and challenges of estrus synchronization and AI.

The reasons from a total of 277 farmers for breeding using estrus synchronization and AI were: to increase pregnancy rates (89.9%), to eliminate heat detection (55.9%), and to facilitate batch management of calves and dams (28.2%) while the challenges faced with this practice were: high cost of synchronization hormones (69.6%) and low conception rates from estrus synchronization and AI (59.6%). The other challenges identified were difficulty in accessing hormones (18.1%), and difficulty in segregating breeding bulls on farms (7.2%).

Pregnancy rate of cows bred after estrus synchronization

A total of 6,171 heifers and cows received AI after estrus synchronization from 282 farms. Of those, 2,298 became pregnant, giving an overall pregnancy rate of 37.24%. Herd level pregnancy rates of cows/heifers following estrus synchronization and AI varied widely, from 0.00% to 68.75%, with a mean of $39.74 \pm 0.96\%$. The mean herd pregnancy rates were not significantly different across protocols ($P=0.13$) and were: 7-day Co-synch, $33 \pm 6.30\%$; 7-day Co-synch+P4, $42.65 \pm 1.56\%$; Ovsynch, $36.87 \pm 1.66\%$; and PG single injection, $41.33 \pm 1.60\%$ (**Fig.4**).

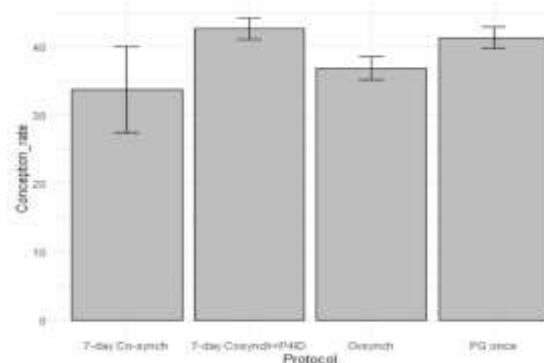


Fig. 4: Distribution of mean conception rate by protocol. The mean conception rate was highest for the 7-day Cosynch+P4ID protocol, followed by the PGF once (PG single injection) and the Ovsynch protocols, while the 7-day Cosynch protocol had the lowest conception rate.

Factors influencing the pregnancy rate of artificially inseminated cattle after estrus synchronization.

As presented in Table 2, the type of production system, herd size, and type of estrus synchronization protocols used did not have a significant influence on pregnancy rate in cows/heifers bred after estrus synchronization. However, the breed of cattle and the main breeding method had significant effects on pregnancy rate. Post hoc pairwise comparisons revealed that farms of

indigenous breeds had significantly lower pregnancy rate than farms of exotic dairy breeds and their crosses (P.adj<0.001) or exotic beef breeds and their crosses (P.adj=0.004) in Fig. 5. Farms that relied predominantly on natural mating had significantly lower pregnancy rates after synchronization and AI than farms where the main breeding method was AI after synchronization (P.adj=0.02) or AI after natural estrus (P.adj=0.03) in Fig.6.

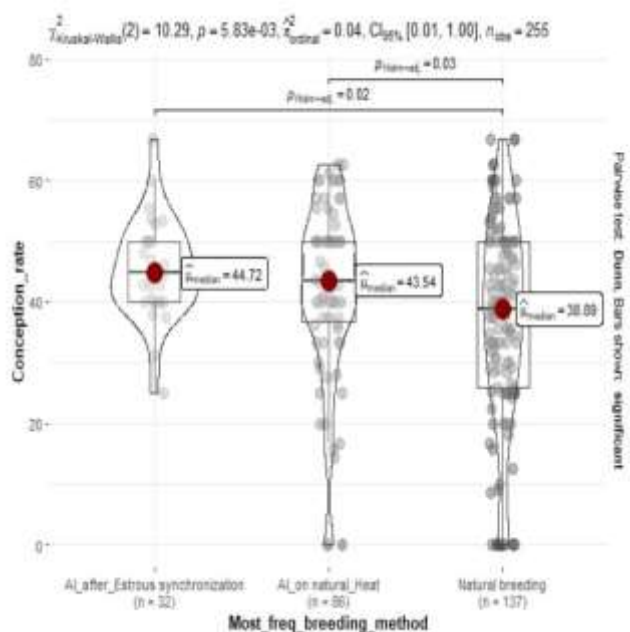


Fig.5: Influence of cattle breed on conception rates after estrus synchronization. The conception rate synchronized cattle was significantly lower for farms of local (indigenous) breeds than for farms of exotic dairy breeds/crosses or exotic beef breeds/crosses. However, there were no significant differences in conception rate between farms of exotic dairy and exotic beef breeds. Adjustment method for pairwise comparisons =Holm.

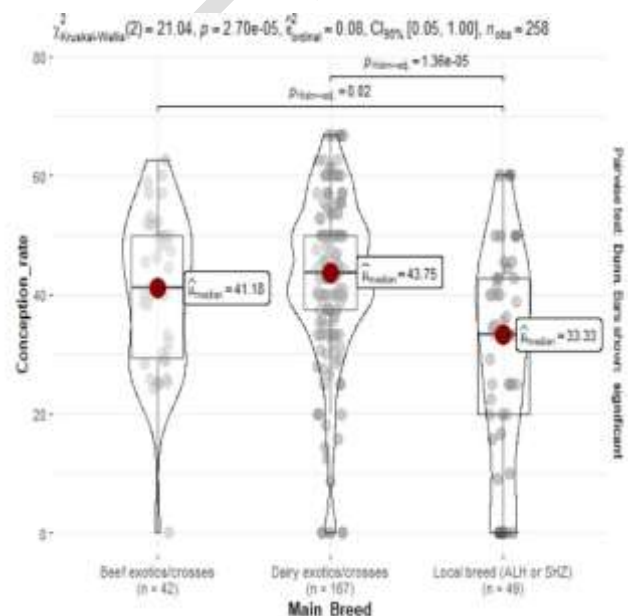


Fig.6: Influence of main breeding method on conception rate after estrus synchronization.

Mean conception rates of synchronized cattle were highest for farms whose main breeding method was AI after estrus synchronization, and this was followed by AI on natural heats, and least for farms whose main breeding method was natural breeding. There were significant differences in conception rates to AI after synchronization, between farms whose main breeding method was synchronization and farms whose main breeding method was natural mating. Similarly, the conception rate to AI after synchronization was significantly higher for farms whose main breeding method was AI on natural heats, compared to farms whose main breeding method was natural mating. There was no difference in conception rate to AI after synchronization between farms whose main breeding method was AI after natural heats and AI after synchronization. Adjustment method for pairwise comparisons =Holm.

Table 1: Variation of estrus synchronization protocols with farm characteristics.

Variables	Estrus Synchronization Protocol				Total
	7-day Co-synch	7-day Cosynch+P4	Ovsynch	PG single	
Region of Uganda					
Northern	0	31	25	57	113
Eastern	3	8	72	5	88
Western	4	34	24	5	67
Central	0	23	5	1	29
Production system					
Extensive	0	31	66	31	128
Intensive	5	30	10	1	46
Semi-intensive	2	35	49	34	120
Breed category					
Exotic beef/crosses	0	16	7	21	44
Exotic dairy/crosses	7	74	72	36	189
Indigenous breeds	0	2	47	10	59
Herd size					
<30 heads of cattle	4	42	69	41	156
30≤49 heads of cattle	1	18	24	7	50
≥50 heads of cattle	2	26	32	20	80

Table 2: Influence of farm factors on pregnancy rate of synchronized cattle.

Farm characteristic	n	Mean	SD	Median	DF	Statistic	P-value	Test
Estrus synchronization protocol								
7-day Co-synch	7	33.75	16.69	40.00	3	5.70	0.13	KW
7-day Co-synch+P4ID	84	42.64	14.38	43.54				
Ovsynch	107	36.87	17.17	40.00				
PG single injection	65	41.33	12.96	41.17				
Breed category								
Exotic beef/crosses	42	40.39	12.98	41.17	2	21.04	<0.001	KW
Exotic dairy/crosses	167	42.66	14.12	43.75				
Local breed	49	29.88	17.99	33.33				
Production system on the farms								
Extensive	116	37.37	17.00	40.00	2	4.75	0.09	KW
Semi-intensive	111	41.56	14.03	43.75				
Intensive	33	41.94	14.29	40.91				
Herd size								
≤30 heads of cattle	133	39.29	17.21	41.17	2	0.11	0.95	KW
31-50 heads of cattle	47	39.44	15.06	42.85				
>50 heads of cattle	77	40.57	12.10	40.74				
Main breeding method								
Natural mating	137	36.77	16.77	38.89	2	10.29	0.005	KW
AI after natural estrus	86	41.68	14.49	43.54				
AI after synchronization	32	45.41	8.08	44.72				

KW = Kruskal-Wallis's test; df = degrees of freedom. Whereas there were no significant differences in mean pregnancy rates of synchronized cattle across production systems, herd size, and estrus synchronization protocol, pregnancy rate varied significantly across levels of farm location, cattle breed category, and main breeding method on the farm.

DISCUSSION

Our finding that the majority of the farms that utilize estrus synchronization were comprised of exotic dairy and crosses is in agreement with the report of another study (**Mugisha *et al.*, 2014**) which indicated that the majority of cattle farms that utilized AI were dairy farms concentrated around peri-urban areas. Indeed, all the farms we surveyed in the Central region that had exotic dairy breeds and their crosses were in districts surrounding Kampala City. However, a considerable number of farms comprised of local cattle breeds in the current study had used synchronized AI, especially in the Eastern and Northern parts of Uganda. We attribute the increasing trend of synchronized AI in local breeds to the increase in adoption of cattle genetic improvement through crossbreeding, championed by the National Animal Genetic Resources Center and Data Bank, through its community-based animal breeding outreach program.

The low level of utilization of estrus synchronization and AI on beef farms realized in this study was not surprising, since a report from another study indicated that AI was concentrated on dairy farms in urban areas of Uganda (**Mugisha *et al.*, 2014**). We recommend increased adoption of synchronized AI in beef cattle in Uganda since this technology has been proven to have the potential of boosting productivity in beef cattle enterprises through the use of semen from superior bulls, resulting in faster growth rates and heavier weaning weights, ultimately boosting the efficiency of beef enterprises (**Bó and Baruselli, 2014; Bó *et al.*, 2016**).

The finding that the majority of the farms that utilized estrus synchronization were comprised of small herds of 30 or fewer heads of cattle agrees with the report on dairy farming systems in Greece that commercial AI is more profitable in small herds than in large herds (**Valergakis *et al.*, 2007**). Small-sized cattle farmers in Uganda seem to have realized the economic merits of using AI rather than keeping one or two breeding bulls on the farm. Keeping bulls for natural breeding comes with extra feeding and management costs and missed breeding opportunities due to bull infertility that could go undetected for protracted periods. The fertility of a bull depends on its breeding soundness (**Fordyce *et al.*, 2006; Siddiqui *et al.*, 2008; Latif *et al.*, 2009; Diskin and Kenny, 2014**).

The low level of utilization of AI in large cattle herds recorded in our study could be due to the perceived difficulty of AI implementation, including estrus detection and segregation of bulls in large herds compared to small herds (**Mugisha *et al.*, 2014**). Although this perspective could be true for natural mating versus conventional AI, fixed-time estrus

synchronization is designed for breeding a large number of cattle at reduced costs, taking advantage of large numbers of open cows, and the prefixed insemination time eliminates the burden of cumbersome estrus detection. The farmers' challenges revealed by this study, including the high cost of estrus synchronization and AI, as well as associated low pregnancy rates, should be addressed through cost subsidization and development of locally tailored effective estrus synchronization protocols.

Natural mating was the predominant breeding method on the majority of the surveyed farms, and even where synchronization and AI on natural heats were the main breeding methods, the use of natural mating persisted. This was not surprising since even in areas where AI had been successful, natural mating was reported to be widespread (**BonDurant, 2005**). The persistence of natural breeding presents a significant challenge to AI after estrus synchronization, since the presence of a breeding bull will cause a constant disruption of the breeding program by mounting synchronized and receptive cows before AI. In addition, the use of estrus synchronization in our study was less in herds of indigenous cattle breeds or exotic beef breeds than on farms with exotic dairy breeds or their crosses. This finding agrees with the reports of over 80% (**Mburu *et al.*, 2011**) and 90% (**Mugisha *et al.*, 2014**) levels of utilization of natural breeding among cattle farmers in Uganda. This widespread practice of natural breeding in favour of AI could be responsible for the country's consistently poor livestock productivity, as it might be responsible for poor genetic gains due to inbreeding. Farmers should adopt estrus synchronization and AI since these technologies are known to accelerate genetic and productivity improvement by shortening the generational interval and enabling high-intensity selection (**Rodriguez-Martinez, 2012**).

Like in our study, the order of frequency of use of breeding methods on cattle farms in Jimma, Ethiopia, was natural mating, followed by AI after natural estrus and lastly, AI after estrus synchronization (**Seid *et al.*, 2017**). However, AI was the predominant breeding method in some parts of Ethiopia. The order of breeding methods used in cattle in the Andracha, Ethiopia was reported to be AI after estrus synchronization (71.7%), AI after natural heat (28.8%), and natural mating (2.5%) (**Gebremichael, 2015**). The wide use of AI after natural heat on farms in Central Uganda could be attributed to being predominantly zero-grazing farms, comprised of small herds with 30 or fewer heads of cattle. On such farms, estrus detection could be easier due to the small herd sizes and the presence of improved access to AI services (**Mugisha *et al.*, 2014**).

In Western Uganda, increased use of estrus synchronization as the main breeding method on farms could be attributed to farms being comprised predominantly of exotic dairy breeds and the large herd sizes that could have made estrus detection difficult. In Northern and Eastern Uganda, the high proportion of farms that used natural mating more frequently than other breeding methods could be attributed to a lack of access to AI services, high AI service costs, and low pregnancy rates from AI. In Uganda, there was a contrast between farmers' practice and preference for breeding method; while the farmers' preference for natural breeding was only at 63.6%, over 80% of the farmers practiced natural breeding (Mugisha *et al.*, 2014). The discrepancy between farmers' breeding method preference and practice could be attributed to the identified challenges of the high cost of estrus synchronization and AI services, low pregnancy rates of AI, and difficulty in accessing AI services. This presents an opportunity for increased adoption of synchronized AI in Uganda, contingent upon addressing these identified challenges.

Ovsynch was the most widely used estrus synchronization protocol, followed by 7-day Co-synch+P4ID and PG single injection protocols, whereas the 7-day Co-synch protocol was the least utilized among cattle farms in Uganda. The 7-day Co-synch protocol differs from Ovsynch in only one aspect: FTAI is conducted at the time of administration of the second GnRH dose on Day 9. Given its similarity to the Ovsynch protocol in terms of hormones used and their timings, and its comparative advantage of fewer farm visits, the low level of utilization of the 7-day Co-synch protocol could be attributed to a lack of knowledge about it. The popularity of the Ovsynch protocol could be attributed to knowledge and the cost of associated hormones. Although Ovsynch involves four farm visits for its implementation, only two hormones, GnRH and PG, are involved, making it cheaper than the 7-day Co-synch+P4ID protocol, which involves only three farm visits but the extra hormone, P4ID, which might have been expensive and unaffordable for many farmers. It is noteworthy that the high cost of hormones was among the major challenges affecting the adoption of synchronized AI identified by farmers in this study. Although the 7-day Co-synch+P4ID protocol was reported to be more frequently used in this study relative to PG single injection and 7-day Co-synch, this could be attributed to the community-based breeding program implemented by the government. This program provided estrus synchronization hormones at no cost. The PG single injection protocol was used mainly on farms in Northern Uganda, possibly due to the high cost of hormones.

Our study revealed a generally low pregnancy rate that varied widely across farms. This finding is similar to that of another study, which reported pregnancy rates ranging from 0% to 50% in dairy cows under extensive management in Western Uganda, although only the Ovsynch protocol was used in their study (Kwon *et al.*, 2017). Pregnancy rate to AI was also reported to be low in dairy cattle herds in peri-urban areas of Kampala, based on farmer responses and review of farm breeding records (Eklundh, 2013). The low pregnancy rates reported in our study could be attributed to poor cattle BCS and suboptimal nutrition prevalent on tropical farms (Robinson *et al.*, 2006; Ayres *et al.*, 2009). The wide variation in pregnancy rate between farms in our study could be due to differences in cattle nutrition, age, parity, timing of AI, accuracy of estrus detection, season of breeding, semen quality and handling, disease and stress across farms (Belay *et al.*, 2016; Bigirwa *et al.*, 2019; Duro, 2022). Improvement in these farm factors could lead to an increase in pregnancy rates, since the heritability of conception rate in cattle is very low (Bigirwa *et al.*, 2019; Getabalew *et al.*, 2019).

This study found no significant differences in pregnancy rates between protocols, which is in agreement with the findings in other studies (Arndt *et al.*, 2009; Colazo *et al.*, 2015; Monteiro Jr *et al.*, 2015). However, pregnancy rates were reported to significantly vary between protocols (Yan *et al.*, 2016). In another study, pregnancy rates with protocols that involve P4ID implants were reported to be significantly lower than those without P4ID implants (Parr *et al.*, 2014). In our study, while there were no significant differences in pregnancy rates between protocols, it is noteworthy that the pregnancy rates were generally low. This could indicate a generally poor performance of the available estrus synchronization protocols in Ugandan cattle and highlight the need for the development of effective breeding protocols.

The pregnancy rates obtained within the indigenous cattle breeds were significantly lower than in exotic beef and dairy breeds and their crosses. This finding revealed a similar trend to the 68.4% conception rate in *B. taurus* x *B. indicus* crossbreeds versus a 53.3% rate in *B. indicus* cows reported in one study (Getabalew *et al.*, 2019). Similar breed differences in pregnancy rates of AI in estrus-synchronized cattle were also reported in other studies (Miah *et al.*, 2004; Woldu *et al.*, 2011). Our study was a survey that relied on farm records and did not have control over many variables that could be important in animal reproduction. Therefore, the low pregnancy rates obtained in *B. indicus* compared to *B. taurus* and *B. taurus* x *B. indicus* crossbreeds in our study could be attributed to factors such as cattle nutrition, health, climate, semen quality

and technician factors. *Bos taurus* and *B. taurus* x *B. indicus* crosses are known to be given better management, such as housing and nutrition, compared to *B. indicus* cattle herds (Kaziboni *et al.*, 2004; Woldu *et al.*, 2011). Poor BCS and suboptimal nutrition are known to characterize *B. indicus* farming systems in the tropics, contributing to decreased pregnancy rates (Robinson *et al.*, 2006; Ayres *et al.*, 2009, 2014; Nishimura *et al.*, 2018). However, significant physiological differences, including in the duration of estrus, the interval between estrus onset and ovulation, and the incidences of anestrus, silent estrus, and split estrus, are also known to exist between *B. taurus* and *B. indicus* cattle types (Bó *et al.*, 2003; Holland *et al.*, 2012). These could be responsible for the differences in pregnancy rates between the cattle types in our study. Since reproductive physiology is the basis for the design of estrus synchronization protocols, the significantly lower fertility recorded in the *B. indicus* compared to *B. taurus* cattle using the protocols in this study justifies the urgent need for novel estrus synchronization protocols that are based on the reproductive physiology of the *B. indicus* cattle.

Farmers in this study identified lowering breeding costs and increasing pregnancy rates as the major motivations for adopting estrus synchronization. This same study demonstrated low reproductive performance with the available estrus synchronization protocols. There is therefore an impending risk of reduced adoption of estrus synchronization technologies, unless significant gains are made in improving reproductive performance. Improvements in general animal management, AI implementation, and deployment of effective estrus synchronization protocols are thus urgent measures required to mitigate the reversal of the small genetic gains already made in the national cattle herd.

A noteworthy limitation of this current study is its cross-sectional design and reliance on retrospective data. Important potential confounders such as nutrition, body condition score, health status, semen quality and handling, and season could not be accounted for due to a lack of adequate records. This affects the generalizability of the findings from this study. Future studies aimed at assessing the effects of estrus synchronization protocols on reproductive performance of cattle should be controlled and prospective in design and take into consideration these potential confounders.

CONCLUSIONS

Currently, four estrus synchronization protocols—Ovsynch, 7-day Co-synch, 7-day Co-synch+P4ID, and PG single injection—are the common estrus synchronization protocols applied to facilitate AI

on Ugandan cattle farms. The majority of the farms that utilize AI after estrus synchronization are dairy farms, and there is a wide use of natural mating on cattle farms that have adopted artificial insemination, which could present a significant challenge to cattle genetic improvement using breeding technologies. The pregnancy rates due to estrus synchronization and AI are lower in *B. indicus* herds compared to *B. taurus* and *B. taurus* x *B. indicus* crossbreeds. Improvements in the management of cattle herds, including optimizing their nutrition and health, should accompany any new reproductive technology to improve success. Prospective controlled studies, designed to take into account the potential effects of cattle management and technician factors, are required to facilitate a better understanding of the effects of different synchronization protocols on the reproductive performance of *B. indicus* cattle in Uganda.

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Conflict of interest

The authors declare that they have no competing interests

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