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Effect of hCG vs GnRH at the beginning of the OVSYNCH plus on conception rates in acyclic Murrah buffalo heifers (*Bubalus bubalis*)

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Abstract

The peripubertal anestrus Murrah buffalo heifers (*Bubalus bubalis*) were selected at random from herd and the animals were randomly divided into two groups and received either “Ovsynch-Plus” (Group I; n = 13) or “Modified Ovsynch-Plus” (Group II; n = 13) treatment. All the treated females were inseminated at fixed time on day 9 and 10. Pregnancy was diagnosed by ultrasonography on day 28 after insemination. In the present study conception rate in the Ovsynch-Plus and Modified Ovsynch-Plus treated heifers were 30.76% and 15.38%, respectively. The overall conception rate observed was 23.1%.

Keywords: Murrah, anestrus, ovsynch-plus, modified ovsynch-plus, ultrasonography

1. Introduction

India is richest among all nations of world in term of livestock resources and these resources play an important role in uplifting the socio economic status of country's rural population. On the basis of utility and production potential, the buffalo is so-called — The Black Gold (Acharya and Bhat, 1988) ^[1]. However, low reproductive efficiency in buffalo poses a major economic constraint towards profitable dairy farming (Kumar *et al.*, 2009) ^[10] and this low reproductive efficiency has been attributed to some inherent problems like late maturity, silent estrus, poor expression of estrus and inactive ovaries (Cockrill 1980, Madan 1988) ^[3, 11].

Various therapeutic agents including hormonal and non-hormonal compounds have been used extensively for the restoration of cyclicity in anestrus cattle and buffalo by several workers with varying degree of success. GnRH-based protocol such as Ovsynch (Pursley, Mee, & Wiltbank, 1995) ^[15] may be used for inducing fertile and synchronized oestrus in anestrus buffaloes. But, Ovsynch protocol was primarily designed for oestrus and ovulation synchronization in cyclic animals, but this protocol showed reduced fertility to the TAI protocol in acyclic condition (Gümen, Guenther, & Wiltbank, 2003) ^[6].

Incorporation of eCG in synchronization protocol has either improved ovarian follicular response and fertility in *Bostaurus* (Mussard, Burke, Behlke, Gasser, & Day, 2007) ^[13] and buffaloes (Murugavel, Antoine, Raju, & López-Gatius, 2009) ^[12] during low-breeding season or had no effect on follicular response and pregnancy rate in cattle (Souza *et al.*, 2009) ^[16] and buffalo (Neglia *et al.*, 2003) ^[14]. Keskin *et al.*, (2010) ^[8] by using Ovsynch protocol and replacing 1st GnRH (10 g) with 1500 IU hCG administration in cyclic cattle, observed higher conception rate in GnRH treated than hCG group. But no such study is there which had studied the effect of replacing 1stGnRH of Ovsynch protocol with hCG, thus present study is intended to compare fertility response after Ovsynch plus and modified Ovsynch plus protocol (first GnRH is replaced with hCG) in anestrus buffalo heifers.

2. Material and Methods

2.1 Study location and animals

The present study was conducted at ICAR-Central Institute for Research on Buffaloes, Hisar on 26 peri-pubertal anestrus Murrah buffalo heifers (*Bubalus bubalis*). All these heifers were between age groups of 30-36 months weighing 300-340 kg body weight having good body condition of 2.75-3.0.

2.2 Housing, feeding and management

Experimental buffalo heifers were housed in half-walled pucca sheds with asbestos sheet roof.

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Heifers were let loose daily for pasture grazing for 3-4 hr. In addition, the animals were stall-fed with adequate quantity of green fodder and wheat-straw. The concentrate feed, supplemented with mineral mixture and common salt prepared in the institute, was also provided to the animals. Drinking water was available *ad libitum*. Animals were protected from the inclement weather and given water shower bath daily during summer season.

2.3 Schedule for treatment

All buffaloes heifer before to start of treatment were screened twice at 6–10 days interval for monitoring their cyclic status using transrectal ultrasonography. The absence of CL in the ovary on both occasions was categorized as anestrus buffaloes heifer.

Total of 26 buffalo heifers were randomly divided into two groups *viz.* Ovsynch Plus and Modified Ovsynch plus group. In Ovsynch Plus group, buffaloes ($n = 13$) were administered 500 IU eCG (2.5 ml, Folligon®, Intervet) intramuscularly 3 days prior (day-3) to Ovsynch protocol {Ovsynch protocol consisting of 16 µg GnRH agonist (Buserelin acetate, Receptal, Intervet) intramuscularly on day 0 and 9 with 750 µg PGF_{2α} (Tiaprost, Iliren, Intervet) on day 7}. (Fig.1) In the second group (Modified Ovsynch plus, $n = 13$), Ovsynch was initiated (day 0) with hCG (3000 I.U., i.m., Chorulon, Intervet). All the experimental buffalo heifers were inseminated at a fixed time with a good quality frozen-thawed semen on day 9 (day of 2nd GnRH injection) and again on day 10. (Fig. 2).

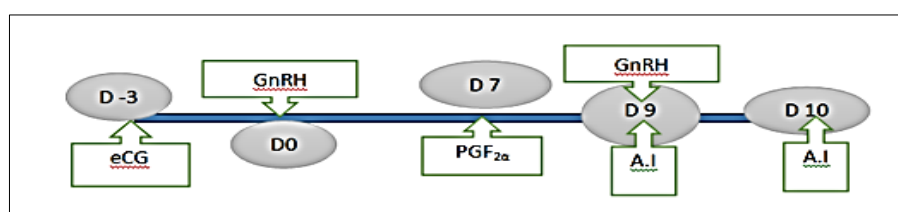


Fig 1: Ovsynch-plus protocol

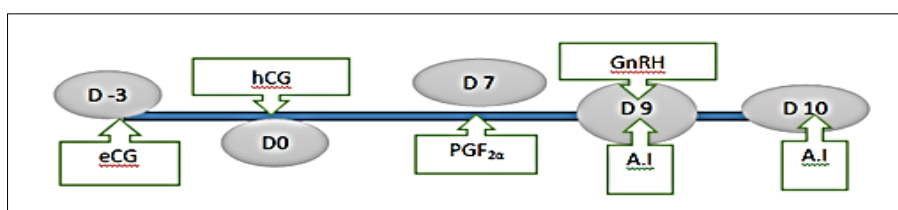


Fig 2: Modified Ovsynch-plus protocol

2.3.3 Pregnancy diagnosis

Animals were scanned with a real time B-mode ultrasound scanner (Just Vision 200–Model SSA-320A, Toshiba, Japan) equipped with a convex array multi frequency transducer using frequency of 7.0 MHz 28 days post A.I. for diagnosis of pregnancy.

2.3.4. Statistical analysis

The data obtained in the present study were subjected to Analysis of Variance and students T-test analysis (Snedecor and Cochran, 1994) to draw scientific inferences. Characteristics associated with ovarian follicular development

were examined with General Linear Model Procedures in the statistical software package for Windows Version 9.0.1 (SPSS Inc. Chicago, IL).

3. Results and Discussion

Conception rate was evaluated at 28 days post-insemination by confirmation of pregnancy by ultrasonography. A lesser number of treated females of both the groups became pregnant. In the current study the conception rate in the Ovsynch-Plus and Modified Ovsynch-Plus treated heifers were 30.76% and 15.38%, respectively. The overall conception rate observed in the present study was 23.1%.

Table 1: Conception rate in treated heifers

Sr. No.	Group	Total animal	No. of pregnant	Percentage (%)
1.	Ovsynch-Plus	13	4	30.76
2.	Modified Ovsynch-Plus	13	2	15.38

The low conception rate was observed in modified ovsynch protocol (15.38%) as compared to ovsynch protocol (30.76%). Replacement of GnRH by hCG did not affect the overall outcome of the Ovsynch plus. In previous studies, treatment with hCG induced accessory corpus luteum, increased plasma progesterone level, and improved conception rate in lactating dairy and beef cattle [Geary TW, Salverson RR, Whittier JC, 2009] [5]. In early studies, hCG was used to replace the first and second GnRH of Ovsynch in lactating dairy and beef cows [De Rensis F, Marconi P, Capelli T, Gatti F, Facciolo F, Franzini S, Scaramuzzi RJ, 2002] [4]. However, despite the fact that usage of hCG in the Ovsynch increased cumulative conception rate in lactating dairy cows, there was no effect on conception rate in cold season [De Rensis F, Marconi P, Capelli T, Gatti F, Facciolo F, Franzini S, Scaramuzzi RJ, 2002] [4]. In the case of the beef cows, hCG was used in TAI protocol (CO-Synch) and these studies showed that hCG was not a suitable replacement for GnRH to synchronize ovulation when using the TAI protocol [Geary TW, Salverson RR, Whittier JC, 2009] [5]. However the overall conception rate observed in the present study was 23.1% which was superior to 9.5% reported

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by Karen and Darwish (2010) and 6.9% Baruselli *et al.*, (2003) ^[2] when applied timed-AI after using Ovsynch protocol during non-breeding season in buffaloes.

In conclusion, replacement of GnRH by hCG in Ovsynch plus did not increase conception rates in anestrus buffalo heifer. Thus, hCG is not a suitable replacement for the first GnRH of in Ovsynch plus protocol. However, a study is warranted in large number of animals for some concrete recommendation.

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