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Improvement of Pregnancy Rate in Dairy Cows Using GnRH Analog

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ABSTRACT

The study was done to calibrate the efficacy of GnRH (Gonadotropin Releasing Hormone) analog on pregnancy rate administered before the time of AI (Artificial Insemination) in dairy cows. Total 90 cows were selected and separated randomly into two groups. GnRH analog received group was administered i/m (intramuscularly) in the neck muscle with GnRH analog one hour before the time of AI. The dose of GnRH analog administration was 300 µg and we used Ovurelin® which contains GnRH analog as gonadorelin acetate. Overall 73.33 % pregnancy rate was found in GnRH analog received group besides 57.78 % pregnancy rate was in control group and having significance effect (P<0.05). The study revealed that using GnRH analog before the time of AI increased the pregnancy rate in dairy cows.

Keywords: *Pregnancy rate, dairy cows, GnRH analog and AI.*

INTRODUCTION

Reproductive capability in dairy cows has lowered during the last few eras because of many causes including estrus detection technique, improper timing of AI and delayed ovulation etc. Other cause contributing to low pregnancy rates is embryonic death. Along with many possible etiologies, decrease plasmaprogesterone concentration has been relatedto early embryonic death [1]. Some articles have been shown the effect of GnRH analog on reproduction cases in dairy cows when administered on the day of AI [2], early luteal phase [18]or mid luteal phases [3].

The estrus cycle of cows takes about 21 days (18-24 days). The highest time of the cycle is the estrus and the ovulation [4,7]. These processes are controlled through the neuronal-endocrinal system. The absence or the delay of the ovulation is one of the causes which downer the conception of the dairy cows [5]. Now the relations between system hypatalmo -hypofisar - ovaries are well known [6].Onthe estrus, the mature follicle grows up and about 12 hours after it ovulates [7].

There are many causes which are affecting reproduction in dairy cows in addition with high milk production, postpartum disorders and low body condition score (BCS) [8].GnRH is a protein hormone secreted by hypothalamus. It controls the production and release of LH and FSH those are originating from the pituitary gland. These two hormones control follicular growth, ovulation and corpus luteum (CL). GnRH analog have been used to control reproductive function in dairy cows [9]. GnRH and its various analogs treated at the time of AI are the familiar treatments in various management programs for dairy herds, given by veterinarians [10].Economy of dairy farming largely depends on a good pregnancy rate after AI or natural service. The one year calving interval was advantages for maximum milk production per cows per year with good economic benefit[11]. The main obstacle of profitable dairy farm was low pregnancy rate [12].

So the above condition, main problem shown is low pregnancy rate using AI. This was the major problem that needs a solution to increase pregnancy rate in dairy cows[13]. According to the various study, major reproductive problems were, delayed ovulation, anovulation, anestrus, cystic ovaries and early embryonic death which cause cows to show repeated estrus cycle and low pregnancy rate [13]. Many methods were taken to treat reproductive problems associated

with such hormonal imbalances. One of these techniques was the use of synthetic GnRH analog that has been used to modify reproductive performance of healthy dairy cows which has been tested widely. Synthetic buserelin acetate has an added advantage of improved half life in the blood circulation, greater stability to enzymatic degradation, increased receptor affinity, prolonged biological potency and less antigenic than the other molecules for several use [14]. Exogenous administration of these hormones at different period of estrus of cow may lead to treat disease condition or cysts in the ovary or may rectify the hormonal insufficiency to these cows [29]. The GnRH analog had been used as treatment for pregnancy rates, early embryonic death and treatment for cystic ovaries [15].

Effective improvement of the pregnancy rate after administration of GnRH analog has been attributed to the prevention of an ovulation failure or a small variation in the time between the onset of estrus and ovulation by increased preovulatory LH surge [16, 17]. Enhancement of pregnancy rate through administration of GnRH analog on present estrus and before the time of AI of dairy cows enabled the changed of breeding policy. Production of more calves per year on herds will change the demand of milk and meat production. In addition improvement of pregnancy rate using GnRH analog will help in reduce cost involve in breeding policy. No study was occurs to calibrate the GnRH analog administration before the time of AI for improvement of pregnancy rate in dairy cows. Howeverthis study was performed with the aim of improving the pregnancy rate in dairy cowsusing GnRH analog before the time of AI on treated groups as compared to control. Thus this study proved that using GnRH analog before the time of AI increased the pregnancy rate in dairy cows.

MATERIALS AND METHODS

The study was conducted at Rajshahi district in Bangladesh. Ninety cows were selected and those are corssbred that is Holstein Friesian with Local (HFxL).Their coat color varies from red, grey, white, black or a mixture of them in different proportion. For the identification of the Holestein Friesian crossbred we used the phenotypic characters that were color, no hump and chute, size etc. To achieve the results of pregnancy rate in cattle the influencing factors were classified according to the age, parity, body condition score, milk yield.

The method or techniques were classified into following main two groups-

- GnRH analog received group; n= 45
- Control group; n=45

The dose of GnRH analog was 300 µg(Ovurelin® Renata Ltd.).GnRH analog was injected intramuscularly in every treated cow one hour before the time of AI.Again, the age of these cows were determined from birth register and examined by teeth and reading the corneal ring. After confirmation of age of these cows we have taken 4-5 years old dairy cows. Parity of these cows was determined by the information from farmer and observation of corneal ring of cows. After determined of parity of these cows we have taken 2^{nd}-3^{rd} parity of dairy cows.The scoring of body condition involved a manual assessment of the thickness of fat covered and prominence of bone at the tail head and loin area. The tail head score by feeling for the amount of fat around the tail head and the prominence of the pelvic bones. The loin scored by feeling the horizontal and vertical projection of the vertebrae and the amount of fat in-between. The body condition score (BCS) varied from 1.00-5.00, according to five scale point system outline by Wildman [34]. In this scale emaciated cow were scored 1.00; thin cows 2.00; medium or average cows 3.00; fat or good cows 4.00 and very fat or obese cows were scored 5.00. We have taken only Good (BCS 4) dairy cows. Milk yield of those the cows were 10-more Liters per day.

These all 90 animals were maintained under almost same routine feeding and management conditions. The routine feeding consisted of available green fodder plus concentrate ration according to the daily milk yield. Vaccination was carried out as per vaccination schedule. Estrus detection of cows was carried out intensively twice daily by experienced herd persons. Only those animals were selected and recorded in heat which stood still while being mounted by other female cows. The animals were also observed for behavioural symptoms like frequent urination, bellowing, raised tail, restlessness and licking of external genitalia. Different visible external changes like vulvular edema and absence of wrinkles on vulvular lips, vaginal hyperaemia, wetness and mucus discharge were also observed.

Farmers were made aware about mucosal discharges from vulva, restlessness, frequent

urination; stand to be mounted and raised tails during estrus exhibition. As soon as such symptoms were observed, they informed to make phones to local inseminator immediately and included in the study. Based on the above criterion; selection process of animals for the study was carried out and assigned randomly to control and treatment groups.

The GnRH analog received group (n=45) was inseminated along with intramuscular injection of 300 µg (3ml) GnRH analog. Control group, (n=45) receive no treatments except single AI.Frozen semen from Friesian bulls was used and evenly allotted to the cows in those the groups. The semen was stored in liquid nitrogen at temperature of –196ºC. The semen was thawed at temperature 37–40ºC in water bath for 10–15 seconds and after washing the external genitalia with water the cows were inseminated. Sterilized Insemination Gun with disposable plastic sheath was used for insemination of the entire animal. All the hygienic measures were adopted to check the possibility of infection at the time of artificial in semination. The duration of study was six months from the start of experiment (from February 2017 to August 2017).Animals which did not return to estrus after 30 days post insemination were examined per rectum 90 days post insemination for the diagnosis of pregnancy. The cows were considered as pregnant if were able to palpate the fetal membrane, amniotic vesicles, and fetus otherwise the cows were classified as negative.

Study variables were collected from dairy cows owners by asking breed type, insemination and service history, parity, management condition, milk yield, time and day of estrus exhibition and previous condition of dairy cows. During this study period, pregnancy diagnosis was made through rectal palpation and positive and negative results were recorded. Some data were collected from farm records about animals' previous milk yield, clinical cases, origin of the animal and breeding type. The experimental study was of a randomized type in which data was collected from treated and controlled study animals. The raw data was sorted and computed and statistically analyzed to calculate the effect of using GnRH analog hormonal drugs on the estrous cycle in relation to GnRH analog administration, age, parity, body condition score, milk yield. Collected data were compiled by Microsoft Excel 2007 and then Statistical Package for Social Science (SPSS) software 20.0 version.

Result and Discussion

From the 90 dairy cows included in this study, 59 cows were found to be pregnant. A total of 33 cows from the GnRH analog received group were found as pregnant with the pregnancy rate of 73.33 %. In control group, from the 45; 26 cows were pregnant with the 57.78 % as indicated in Table 1.

Effects of GnRH on Conception Rate in Dairy Cows

The improvement of pregnancy rate by using GnRH analog is shown table 1 and figure 1. It was shown that the proportion of pregnant number and percentage share were tabulated 33& 73.33 % and 26 & 57.78 % in GnRH analog received and control groups respectively. Since the calculated value of the test was higher than the tabulated value, so researcher may reject the null hypothesis that means there was significant effect on pregnancy rate in relation to GnRH analog administration at the time of artificial insemination in dairy cows.

Table1.*Effects of GnRH analog administration on conception rate in dairy cows.*

Group	Number of cows	Number of pregnant cows	Percentage of pregnancy rate
GnRH analog received	45	33[a]	73.33 %
Control	45	26[b]	57.78%

[a,b] *The values of the superscript within the same column differed significantly from each other (P<0.05).*

It was hypothesized that GnRH analog (Ovurelin® as gonadorel in acetate) treatment at the time of artificial insemination increases pregnancy rate. This work is almost same to the Raoyos said that pregnancy rate for hormone treatment after AI in cows to be 70 % compared to 50 % in control cows. Mann and Lamming [1]revealed on the treatment of cows with 10µg GnRH analog Buserelin (Receptal; Hoechst) 12 days after insemination. Stevenson [16]informed that cows and heifer treated with GnRH analog at times of AI pregnancy rate 25% more in treated than control cow and heifer. Our results are in agreement with the findings that the use of GnRH analog at the time of AI improves pregnancy rate in dairy cows [2].

It was previously thought and that using GnRH analog on heat period might prevent ovulation failure or reduce the time interval to ovulation [19]. Besides, our study differed from the other

researcher's observation [6] who informed that GnRH analog treatment had no effect on conception rate if administered at the time of artificial insemination. An extreme delay in ovulation may affect fertilization and also delay the establishment of luteal function [20] and reduce fertility. Thus, if GnRH analog treatment were given at estrus period, failure or delay of ovulation might be prevented and pregnancy rate might improve. On the other hand, if GnRH analog is administered at the time of AI, especially when detection of estrus is not carried out frequently, the administration is likely, in most cases, to be rather late in estrus, with unknown effects on the timing of ovulation, and a prospective decline in plasma progesterone levels in the subsequent luteal phase [21].

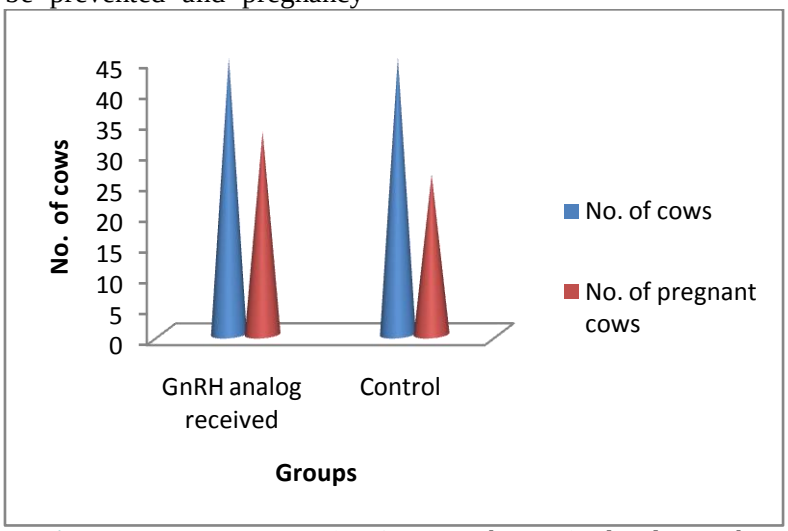

Figure1.*Pregnancy rate using GnRH analogreceived and control.*

The improvement in pregnancy due to GnRH analog treatment in the present study has two possible explanations. Firstly, the positive effect of GnRH analog at the time of AI is mediated by the improved ovulation rate [22]. Cows treated with GnRH analog have a LH surge, which is maximum two hours after treatment [23] and causes a fourfold increase in plasma LH concentration after 2-2.5 h [24]. Since the preovulatory surge of LH normally occurs about 6 h after onset of estrus [25], treatment with GnRH analog at insemination may have induced a secondary surge of LH before or after the spontaneous preovulatory surge of LH. That added increment of LH may be helpful to the events associated with pregnancy rate.

Secondly, progesterone is a vital hormone during early pregnancy that nourishes embryo development and controls the luteolytic mechanism [1]. A single GnRH analog dose at AI can improve subsequent plasma progesterone concentration [17] which is related to hypertrophy and hyperplasia of the luteal cells. GnRH analog injection at the time of estrus causes LH surge and following ovulation LH increases blood flow to ovaries, causing ovarian hyperemia. Therefore, CL formation occurs rapidly and progesterone production increases significantly [26]. In various studies, progesterone concentration has increased, decreased or remained unchanged during the luteal phase, following GnRH analog administration [10, 21, 27, 28].

The improvement in the conception rate with the use of GnRH analog during luteal phase has been attributed to the fact that GnRH analog on day 5 induces ovulation of the first wave dominant follicle, thus forming an accessory CL (corpus luteum) and increasing progesterone production early in the cycle. This increase in progesterone secretion caused by GnRH analog may facilitate embryonic development [1]. Exogenous administration of GnRH analog could initiate the endogenous increase in progesterone [30] via increasing numbers and sizes of CLs following luteal phase administration of GnRH analog[31]. The results of the present study in which the use of GnRH analog during the time of AI improved the pregnancy rate in the dairy cows are also in agreement with the earlier findings [18].

Pregnancy rate was higher in cows with high BCS than in cows with low BCS [32]. The higher response to GnRH analog in cows with high BCS, found in the latter study, was attributed to an earlier postpartum resumption of cyclicity. Butler [33] suggested that the low fertility of the dairy cow under negative energy balance is associated with low estrogen and LH secretion, which could be due to low glucose,

insulin and IGFI secretion, and that part of the low-fertility syndrome could be related to low secretion of progesterone as well. In our study we used only good cows having body condition score 4 in the case of improving pregnancy rate using GnRH analog.

The findings of our study showed that the use of GnRH analog therapy with the time of AI improved in pregnancy rate in dairy cows. The scored pregnancy rates were 73.33 % and 57.78 % in GnRH analog received and control groups respectively. Use of GnRH analog at the time of AI increases the spontaneous LH peak, prevents delays in ovulation, and induces postovulatory progesterone. Thus GnRH analog has the potential to improvepregnancy rate. In conclusion our study revealed that administration of GnRH analog at the time of artificial insemination improved the pregnancy rate in dairy cows.

CONCLUSION

The experiment was conducted to calibrate the effects of GnRH analogadministration before the time of AI on pregnancy rate in dairy cows. The findings of our study showed that the use of GnRH analog along with AI improved the pregnancy rate in dairy cows. There were significant effects of GnRH analog on improvement of pregnancy rate in GnRH analog received dairy cows group.

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