

Fertility response on administration of insulin in repeat breeding crossbred cows

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Recently, insulin has been used as a new approach to modulate reproduction in livestock (Selvaraju *et al.* 2002, Kharche *et al.* 2003a,b and Shukla *et al.* 2005). Exogenous insulin administration in bovine enhances steroidogenesis and folliculogenesis (Simpson *et al.* 1994). Also, the low insulin concentration might limit the responsiveness of ovary to endogenous gonadotropin secretion thus affecting ovulation (Butler and Smith 1989). Direct effect of IGF-1 and/ insulin on the follicles and corpus luteum resulting increased peripheral concentration of progesterone (Lucy *et al.* 1993). The insulin regulates GnRH/ pulsatile LH secretions (Tanaka *et al.* 2000). Such information is meager in repeat breeder cows. Thus, the present experiment was designed to study the effect of exogenous insulin and GnRH on fertility response in repeat breeding cows.

Animals and experimental design: The study was conducted on 18 repeat breeder crossbred cows maintained at Livestock farm, Jabalpur. Animals were randomly divided into 3 groups, each comprising 6 animals (n=6). White-side test was carried out in cervical mucus to screen out subclinical cases of infectious genitalia. All animals were inseminated at morning and evening using good quality frozen semen and treatments were given just after second dose of artificial insemination (AI). Animals of group 1 were treated with long acting bovine insulin @ 0.25 IU/kg b.wt., S/C, OD for consecutive 3 days, group 2 with buseralin acetate @10 µg as a single intramuscular injection. Animals of group 3 were kept as untreated control. All the inseminated experimental animals were observed for return to estrus after AI and treatment. Pregnancy diagnosis was carried out at 60 days post AI. Serum progesterone assay was carried out at estrus

(day 0), day 7th, 14th and 21st post estrus. The data were analyzed statistically for the mean and standard error. Comparison of progesterone concentration at different days was done using CRD and student's test (Snedecor and Cochran 1994).

Fertility response: Higher overall conception was found in animals of insulin treatment (50% -group1) as compared to GnRH and control groups (16.67% each in groups 1 and 3). Results of overall conception in insulin treated groups were comparable to the study of Kharche *et al.* (2003a). They reported 57.14 and 53.33% overall conception, respectively, in insulin treated and untreated control animals in the consecutive three services. However, the conception at first service was similar (16.67%) in each group. The conception at second service was higher (40%) in insulin group whereas, none conceived in GnRH alone and control group. Kharche *et al.* (2003a) also reported 21.42 and 28.57% conception, respectively, at first and second services in insulin treatment groups as compared to 26.66 and 20% in untreated control.

Better conception was also reported on insulin administration during diestrus in repeat breeding cattle (Selvaraju *et al.* 2002, Kharche *et al.* 2003 b). This may be due to luteotrophic effect of insulin in ovary. Insulin may stimulate IGF-I secretion, a potent ovarian and uterine growth factor mediating the reproductive function (Pawshie 1998). *In vitro* studies revealed better early embryonic development with use of insulin and IGF-I (Totey *et al.* 1996). Insulin enhances growth and proliferation of granulosa, theca and luteal cells present in the ovary, thus enhances folliculogenesis (Simpson *et al.* 1994). Insulin along with IGF-I enhances growth and proliferation of theca cells leading to production of progesterone (Stewart *et al.* 1995). Insulin enhanced pulsatile LH secretion acting through CNS a pre-requisite for maturation of dominant follicle was reported (Tanaka *et al.* 2000). The overall conception in GnRH administered group (16.67%) was comparable to findings of Mee *et al.* (1990) and Shams-Esfandabadi *et al.* (2006).

Serum progesterone profile: The mean serum progesterone (P₄) concentration is presented in Table 1. Analysis of data

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Table 1. Serum progesterone profile (ng/ml)

Groups	Reproductive status	Day-0 (At estrus)	Day-7th post estrus	Day-14th post estrus	Day-21st post estrus
G1(Insulin)	Pregnant	0.64±0.19 ^a	6.67±0.39 ^b	9.23±0.18 ^c	10.70±0.23 ^{cA}
	Non-pregnant	1.13±0.42 ^a	4.74±1.37 ^b	7.88±0.70 ^c	1.04±0.08 ^{aB}
G2(GnRH)	Pregnant	1.19±0.00 ^a	5.11±0.00 ^b	6.84±0.00 ^b	8.17±0.00 ^{bA}
	Non-pregnant	0.98±0.40 ^a	4.17±0.98 ^b	5.75±1.28 ^b	1.07±0.28 ^{aB}
G3(Control)	Pregnant	0.91±0.00 ^a	3.93±0.00 ^b	8.41±0.00 ^b	8.42±0.00 ^{bA}
	Non-pregnant	1.36±0.26 ^a	4.06±0.75 ^b	6.28±0.95 ^b	1.31±0.25 ^{aB}

Mean values bearing different superscript (A, B) in a column differ significantly within the group ($P < 0.05$), means bearing different superscript (a, b, c) in a row differ significantly ($P < 0.05$).

revealed that P_4 concentration at estrus was nonsignificantly higher in non-pregnant as compared to pregnant animals ($P > 0.05$). The results of higher P_4 concentration on day of estrus are comparable to the findings of Bedi *et al.* (2007) and Vijayrajan *et al.* (2007). Bedi *et al.* (2007) reported 60% repeat breeder cows with suprabasal concentration of P_4 (0.303 ± 0.022 ng/ml) on the day of standing estrus. Repeat breeder animals that had supra-basal P_4 concentration on day of estrus also had significant lower concentration on day 5 to 7 post insemination than the remaining repeat breeder and normal animals.

The serum P_4 concentration on day 14th and 21st was significantly higher in insulin treated pregnant animals (G1 vs G2 and G3) as compared to day 7th post estrus ($P < 0.05$). Our results of higher P_4 concentration in luteal phase after insulin treatment are in accordance with the findings of Selvaraju *et al.* (2002) and Shukla *et al.* (2005) indicating beneficial effect of insulin on steroidogenesis. Direct effect of insulin on follicle and corpus luteum resulting in increase peripheral progesterone concentration has also been reported by Lucy *et al.* (1993). Selvaraju *et al.* (2002) also reported increase concentration of progesterone following administration of insulin in cow. The increase progesterone concentration in the present study on day 7th, 14th and 21st post estrus would explain the better conception obtained in insulin treated animal.

SUMMARY

The present study found trends of beneficial effect of insulin on fertility response in repeat breeder cows. Also, supra-basal concentration of progesterone at estrus may be used as a marker for prediction of repeat breeding in cows.

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