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Administration of estradiol benzoate before insemination could skew secondary sex ratio toward males in Holstein dairy cows

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ABSTRACT

The present study was conducted to investigate the effect of estradiol benzoate administration before insemination on secondary sex ratio (proportion of male calves at birth) in Holstein dairy cows. Cows (n = 1,647) were randomly assigned to 2 experimental groups by parity over a 1-yr period. Cows in the control group (n = 827; 232 primiparous and 595 multiparous cows) received 2 administrations of $PGF_{2\alpha}$ (500 μg) 14 d apart, started at 30 to 35 d postpartum. Twelve d after the second $PGF_{2\alpha}$ injection, cows received GnRH (100 μg), followed by administration of $PGF_{2\alpha}$ 7 d later. Cows in the treatment group (n = 820; 238 primiparous and 582 multiparous cows) received the same hormonal administrations as the cows in the control group. Additionally, cows in the treatment group received estradiol benzoate (1 mg) 1 d after the third $PGF_{2\alpha}$ injection. Estrus detection by visual observation was started 1 d after the third $PGF_{2\alpha}$ injection and after estradiol administration in the control (for 6 d) and treatment (for 36 h) groups, respectively. Artificial insemination was carried out 12 h after observation of standing estrus. Exposure of cows to heat stress at conception was determined based on temperature-humidity index. Estrus detection rate was lower in primiparous than in multiparous cows ($P < 0.05$), but conception rate was higher in primiparous vs multiparous cows ($P < 0.05$). Estradiol administration improved estrus detection rate and fertility ($P < 0.05$); moreover, it increased secondary sex ratio (adjusted odds ratio: 1.645; $P = 0.017$). Exposure to heat stress diminished heat detection rate and fertility ($P < 0.05$), and altered secondary sex ratio toward males (adjusted odds ratio: 2.863; $P = 0.012$). In conclusion, the present study revealed that estradiol administration before insemination could improve fertility and increase the probability of calves being male in Holstein dairy cows. Moreover, the results showed that cows exposed to heat stress around conception had diminished fertility and increased secondary sex ratio.

1. Introduction

Although it is generally assumed that mammals produce equal proportions of male and female offspring, different factors have been indicated to alter the proportion of sexes [1,2]. The 2 main factors hypothesized to influence the sex of offspring in mammals are maternal body condition [3] and maternal dominance [4]. The former, ie, mothers with better body condition are more likely to give birth to sons than daughters [3], has been attributed to maternal glucose concentration [5] and alters the sex ratio of offspring postconceptionally because glucose adversely impacts the development of female embryos [6–8]. Whereas the latter, ie, more dominant mothers are more likely to produce sons than daughters [4], has been attributed to maternal testosterone concentration [9,10] and influences the sex ratio of offspring preconceptionally as oocytes originated from

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Table 1
Ingredients and nutrient composition of the diet fed to the dairy cows.

Item	
Ingredients (% of dry matter)	
Alfalfa	20.00
Corn silage	20.00
Beet pulp	1.50
Ground corn grain	15.95
Ground barley grain	15.95
Soy bean meal	9.95
Canola meal	4.43
Cotton seed	1.35
Corn gluten meal	2.88
Wheat bran	1.08
Fish meal	1.51
Fat supplement	1.91
Sodium bicarbonate	1.07
Sodium chloride	0.16
Dicalcium phosphate	0.16
Calcium carbonate	0.80
Magnesium oxide	0.30
Vitamin supplement	0.50
Mineral supplement	0.50
Nutrient composition	
Metabolizable energy (Mcal/kg)	2.56
Crude protein (%)	17.30
Neutral detergent fiber (%)	32.10
Calcium (%)	0.90
Phosphorus (%)	0.45

the follicles with high intrafollicular testosterone subsequently produced a male-biased sex ratio [11,12].

In ruminants, numerous factors have been reported to affect the sex of offspring, including parity of dam [13,14], maternal hormonal profile [2,10], stress [15], season or climatic parameters [13,14,16], maternal body condition [17–19], nutrition at conception [1,20], time of insemination relative to the onset of estrus [16,21–23], site of semen deposition [24], and sire [14,25,26].

Reduced reproductive efficiency has been a ubiquitous problem of farms with high-producing dairy cows, leading to the development of different synchronization of ovulation protocols. Synchronization of ovulation necessitates management of ovarian follicular wave emergence, corpus luteum regression, and ovulation induction [27]. Estradiol, administered 24 h after $PGF_{2\alpha}$ treatment, has been observed to induce an LH surge in dairy cows [28] and could efficiently replace GnRH in the Ovsynch protocol for induction of ovulation of the preovulatory follicle [29]. Further, comparing reproductive performance of estradiol-treated and untreated cows, Cerri et al [30] revealed that incorporation of estradiol in the synchronization of ovulation protocol could enhance estrus detection, conception, and pregnancy rates in dairy cows.

Table 2
Secondary sex ratio in primiparous and multiparous cows at first service postpartum in years 2006 to 2010.

Year	Secondary sex ratio (%)
2006	52.3 (551/1,053)
2007	51.5 (522/1,013)
2008	50.5 (524/1,038)
2009	51.1 (587/1,149)
2010	49.7 (565/1,136)
Total	51.0 (2,749/5,389)

Incubation with high concentrations of estradiol has been reported to result in male-biased in vitro-produced embryos in murine [31]. On the other hand, Perret [32] observed greater late-follicular estradiol concentrations in urine of lemurs with female-biased litters than those with male-biased litters. Effect of estradiol administration on sex ratio of offspring has not been investigated in the bovine, however. Given that the sex of a calf is of economic significance, ie, increased females in the dairy industry, as production of more heifers could enhance milk production efficiency and genetic progress [33], the present study was conducted to elucidate the effect of estradiol administration before insemination on secondary sex ratio (proportion of male calves at birth) in Holstein dairy cows.

2. Materials and methods

2.1. Location and animals

This study was carried out at a commercial dairy farm with 2,700 Holstein dairy cows, which was located in the north of Iran (latitude: 36°42′ N; longitude: 53°10′ E; altitude: −8 m) with a warm temperate climate. Cows were housed in open shed barns and fed 3 times daily with a total mixed diet formulated to meet or exceed requirements for lactation (Table 1) [34]. The cows were milked thrice daily and the herd's average milk production was 11,346 kg per lactation. Data associated with secondary

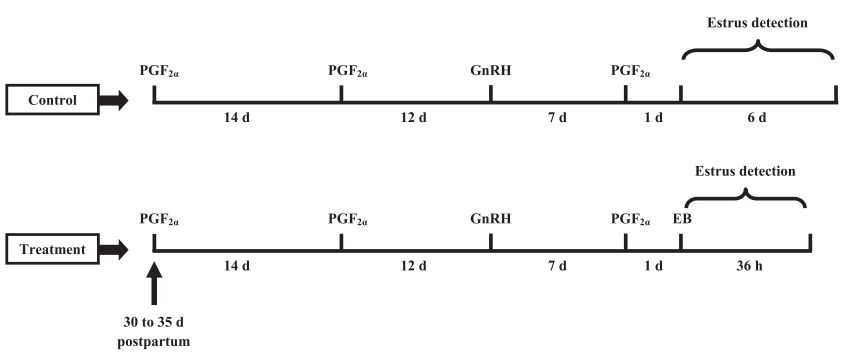

Fig. 1. Experimental design of the study. EB, estradiol benzoate.