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REGULAR ARTICLE

EFFECT OF BODY CONDITION AND SEASON ON THE YIELD AND QUALITY OF CATTLE EMBRYOS

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ABSTRACT

Unsatisfactory reproductive performance in dairy cows has been associated with environmental influences, such as season, chronic and acute changes in dietary intake and body composition. These factors can affect fertility especially ovarian function and yield and quality of oocytes and embryos. In our study the cow ´s body condition affected the overall embryo recovery rate (proportion of collected embryos to palpated corpora lutea). The significantly higher number of embryos was collected from cows with BCS 2.5- 2.75 (68.32 % embryo recovery rate) and 3.0- 3.25 (63.30 %) compared to the cows with BCS 2.0-2.25 (53.33%) and 3.5-4.0 (47.87%; P<0.01) and compared to the cows with BCS 4.5-5.0 (21.43%; P<0.001). On the other hand, highest percentage of transferable embryos was yielded in the BCS 3.5-4.0. Also the season significantly affected embryo recovery rate. The significantly higher (P<0.001) percentage of embryos was recovered during spring months (59.60% recovery rate) compared to summer months (37.04%) and slightly increased again during the autumn (48.30%; P>0.05). On the contrary, the yield of transferable embryos was higher (P<0.05) during the autumn months (78.94%) compared to spring (58.38%) or summer (60.00%) months. In conclusion, body condition and season may affect the yield and quality of bovine embryos. Higher embryo yield was recorded in average BCS (2.5-3.25) cows, whilst most transferable embryos were obtained in the higher BCS (3.5-4.0). Our results indicate that the best season for collection of transferable bovine embryos is autumn.

Key words: cow, body condition score, season, embryo yield

INTRODUCTION

Fertility is one of the most complex measures of reproduction, being influenced by genes and environment. However, although these two components act in concert, they synergistically mask the contribution of the other thus confounding selection strategies for fertility and, ultimately, affecting reproductive performance.

Body condition score (BCS) is a subjective visual and tactile measure of body condition and temporal changes in BCS which is used to monitor nutritional and health status of high producing cows during their productive cycle (**Berry *et al.*, 2007**). It has been correlated with reproductive performance, both phenotypically (**Buckley *et al.*, 2003**) and genetically (**Berry *et al.*, 2003**) and supports the premise that nutritional status affects reproductive function.

Yield and quality of oocytes and embryos upon superovulation reflects potential fertility of dairy cows. The success of ovarian response to superovulatory treatment is dependent on factors inherent to each individual animal, the breed being used, season of the year and nutritional status (**Ammoun *et al.*, 2006**). According to **Siddiqui *et al.* (2002)**, cows with BCS 2.5-3 are likely to respond better to superovulation treatment than those with BCS 4-5, because cows with higher BCS are more likely to acquire ovarian cysts and less ovulations.

Body condition can affect yield and quality of oocytes and embryo in different ways. Diet has been positively correlated with the growth rate and size of the ovulatory follicle (**Bossis *et al.*, 2000; Armstrong *et al.*, 2001**). A relationship between BCS and the ovary, whereby both follicle number and oocyte quality (proportional of normal oocytes), may be adversely affected by a low BCS (**Dominguez, 1995**). Additionally, the cows with low BCS (BCS 1 to 2) had fewer normal oocytes than cows with higher scores (BCS 3 to 5). This fact may lead to a smaller pool of normal follicles what reduces the probability of normal fertility. Oocyte quality and embryo development may by negatively affected also by a high nutrition diet (**Yaakub *et al.*, 1999**).

Additionally, physiological status of cows (lactating cows or non-lactating heifers) has a significant effect on embryo quality (**Walsh *et al.*, 2011**). Embryos recovered from non-lactating heifers were of higher quality compared to lactating cows (**Leroy *et al.*, 2005; Sartori *et al.*, 2010; Rizos *et al.*, 2005**).

It is known that reproductive performance of dairy (**Sartori, 2002**) and beef (**Wolfenson *et al.*, 2000**) cattle can be influenced by season of the year. It is reduced during hot summer months what can affect follicular growth, corpus luteum function, expression of estrous behavior, superovulatory response, quality of embryos and fertility (**Barati *et al.*, 2006**).

The aim of our study was to evaluate the effect of the body condition and season on the yield and quality of bovine in vivo recovered embryos.

MATERIAL AND METHODS

Animals

A total of 56 Holstein- Friesian cows classified according to a 5-point scale of BCS (Edmonson et al., 1989) were used in the experiment. The estrus of the cows was synchronized by the injection of a PGF_{2alfa} analogue (i.m.; 2 ml of Oestrophan, Lečiva Prague). Estrous detection was performed every 12 h for 3 days starting 24 h after the PGF_{2alfa} analogue injection (D 0 = day of standing estrus). The cows were superovulated by application of porcine FSH (Pluset, Minitube, SRN) twice daily during 3 days at 7.00 and 19.00 hours (given in a decreasing dosage rate) started at day 10 or day 11 of the oestrous cycle. Insemination and reinsemination were performed with frozen-thawed semen at 12 and 24 h after the standing estrus detection.

Embryo recovery and evaluation

Embryo recovery was performed on 6-7-th day after the first insemination by a standard non-surgical technique to flush out the uterine horns. Uterine flushing was conducted with A complete flush solution (Bioniche, USA) using a silicone two-way Foley catheter

(Minitüb GmbH, Tiefenbach, Germany). Flushed ova/ embryos were transferred to the holding medium (TCM 199 with GlutaMAX, Gibco) and assessed using a stereomicroscope. The embryos were evaluated according to their stage of development as a transferable (i.e., morulas, blastocysts) and non-transferable (i.e., unfertilized and fragmented).

Statistical analysis

The experiments were performed in six replicates. The data presented are cumulative values of all experiments. The differences between groups were evaluated using Chi-square test.

RESULTS AND DISCUSSION

The embryos in the present study were obtained within the period from march 2011 to november 2011. Totally 226 embryos from 56 Holstein-Friesian cows with BCS from 2.0 to 5.0 were recovered and evaluated.

In our study BCS affected the overall embryo recovery rate (proportion of yielded embryos to palpated corpora lutea - CL; Table 1). The higher number ($P<0.01$) of embryos was collected from the cows in the BCS 2.5-2.75 and 3.0-3.25 (68.32 and 63.30 % embryo recovery rate, respectively) compared to the cows with BCS 2.0- 2.25 (53.33%) and 3.5-4.0 (46.80 %) and significant higher ($P<0.001$) compared to the cows with BCS 4.5-5.0 (21.43 %). On the other hand, highest proportion of transferable embryos was yielded in the BCS 3.5-4.0 cows (79.54%).

Table 1 Embryo recovery rate in relation to BCS

BCS	No. CL	No. fragmented/ unfertilized embryos	No. morulas	No. blastocysts	No. totally flushed	Embryo recovery %	Transferable from collected %
2.0-2.25	105	20	24	12	56	53,33^{b}	64.29
2.5-2.75	101	28	37	4	69	68.32^{a}	59.42^{e}
3.0- 3.25	109	24	32	13	69	63.30^{a}	65.22
3.5-4.0	94	9	30	5	44	46.80^{b}	79.54^{d}
4.5-5.0	14	1	2	0	3	21.43^{c}	66.66

^{a}vs ^{b} and ^{d}vs^{e} $P<0.01$, ^{a}vs^{c} $P<0.001$

Evaluation of the effect of season of the year showed that the season affects embryo recovery rate.The highest number of embryos was collected during spring months (56.60% recovery rate). It was significantly higher (P<0.001) compared to summer months, when the embryo recovery rate decreased to 37.04%. The proportion of collected embryos slightly increased again during the autumn (48.30%; P>0.05) compared to summer months. On the other hand, the percentage of transferable (good and excelent quality morulas and blastocysts) embryos was higher (79.54%; P<0.05) during the autumn (Table 2), compared to spring (58.38%) and summer (78.94%) months.

Table 2. Embryo recovery rate in relation to season

Season	No. Cows	BCS	No. CL	No. fragmented/ unfertilized embryos	No. morulas	No. blastocysts	No. totally flushed	Embryo recovery %	Transferable (from recovered) n/%
Spring	27	2.69	250	62	69	18	149	59,60[a]	87/58.38[c]
Summer	10	2.53	54	8	9	3	20	37.04[b]	12/60,00[c]
Autumn	19	3.20	118	12	35	10	57	48.30	45/78.94[d]

[a] vs [b] P<0.001, [c]vs[d] P<0.05

Environmental influences such as nutritional management, temperature and seasonality interfere with reproductive efficiency, follicular development, and oocyte quality, and consequently, fertility (**Armstrong *et al.*, 2001; Webb *et al.*, 2004**).

In the past years especially the effects of diets and body condition score on oocyte quality and embryo development were intensively discussed. Data from several species showed that pre-mating diets that improve oocyte maturity are also associated with improvements of embryo survival. For cows, either positive (**Nolan *et al.*, 1998; Boland *et al.*, 2001**), negative (**Yaakub *et al.*, 1999; Armstrong *et al.*, 2001**) or no effects (**Tripp *et al.*, 2000**) of plane of nutrition (high or low energy diets) on oocyte quality, fertilization rate and early embryonic development have been reported. Reduced oocyte and embryo quality are acknowledged as major factors in the widely described low conception rates and in the high prevalence of embryonal mortality (**Leroy *et al.*, 2005**). It was postulated that acute changes in dietary energy intake influenced not only developmental competence of the oocytes but also morphology (**Armstrong *et al.*, 2001; Boland *et al.*, 2001; O´Callaghan *et al.*, 2000; Mc Evoy *et al.*, 1995**).

In our study the body condition score of cows affected the overall embryo recovery rate. The higher number of embryos (proportion of collected embryos to palpated corpora lutea) was collected from cows with the middle BCS (2.5 -3.25) compared to the cows with lower (2.0- 2.25) or higher (3.5-4.0) BCS and significantly higher compared to the cows with the highest BCS (4.5-5.0).

Fertility is a multi-factorial trait and its deterioration can be caused not only by BCS, and also by many other non-genetic effects, such as season, heat stress, breed, age and individual differences between animals (**Thatcher *et al*., 2010**).

The decreased fertility in dairy cows may be associated with the elevated ambient temperature during the summer months. For instance, summer elevation of temperature compromised ovarian follicular dynamics (**Badinga *et al*.,1993**), the ability of the dominant follicle to exert dominance (**Wolfenson *et al*., 1995**) and induced follicular codominance (**Sartori *et al*., 2004; de S.Torres-Júnior *et al*., 2008**) and decreased estrous cycle length, and the oocyte's competence to develop into a blastocyst. It was shown that oocytes harvested from Holstein cows during summer exhibited decreased ability to develop to the blastocyst stage after in vitro fertilization when compared with oocytes harvested during winter (**Rocha *et al*., 1998; Al-Katanani *et al*., 2002**). Moreover, exposure of Holstein heifers to high temperature between the onset of estrus and insemination increased the proportion of abnormal and developmentally retarded embryos as compared with heifers maintained at thermoneutrality (**Putney *et al*., 1989**).

In our study there was a significant higher embryo recovery rate during the spring season compared to summer season, when the embryo recovery rate decreased. The proportion of embryos collected during the autumn season was again increased. On the other hand, the percentage of transferable (good and excelent quality morulas and blastocysts) embryos was higher during the autumn compared to spring and summer months. It is reasonable to hypothesize, that the low fertility of cows during the summer months in our study may be associated with a decreased oocyte competence due to elevated temperatue.

On the other hand **Barati *et al*. (2006)** showed that season of the year did not affect superovulatory responses (total number of CL, unovulated follicles, ova/embryo, transferable and nontransferable embryos) of Sistaini cattle, although there was the numerical increase in transferable embryos during summer (summer: 4.6 ± 0.95 versus winter: 2.4 ±0.87) compared to winter months. Also **Randel *et al*. (1994)** showed that there was not significant influence of the season on the number of transferable embryos in Bos indicus donor cows.

CONCLUSION

In conclusion, body condition and season may affect the yield and quality of bovine embryos. Higher embryo yield was recorded in average BCS (2.5-3.25) cows, whilst most transferable embryos were obtained in the higher BCS (3.5-4.0). Our results indicate that the best season for collection of transferable bovine embryos is autumn.

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