

Effect of ambient temperature on number of embryos obtained during embryo transfer process

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Abstract: The objective of the project was to evaluate the effect of the ambient temperature on the uterus flushing out during embryo transfer process. It was generally speaking about a comparison of the donors flushed out during the hot weather with the donors flushed out during the cold weather. It had regard to the number of the embryos obtained and to the ambient temperature. All numbers of the embryos obtained by the flushing out from the 33 milking cows were registered during the embryo transfer process. The three groups of the milking cows were measured during the *high ambient temperature* (24°C and higher), the *medium ambient temperature* (from 4.1°C to 23.9°C) and the *low ambient temperature* (4°C and lower) in the heat time. This research has been done on the dairy cow breed, specifically on the Holstein breed. However the results are easily extrapolated to the other dairy breeds. The experiment was focused on the embryos presented in the flushed liquid. All flushing out results were written down to the Excel MS and analyzed by the SPSS Statistics 17.0; exactly by the One-way ANOVA. The embryos obtained were found to fluctuate with the ambient temperature, being the lowest just during the *high ambient temperature*, high during the *medium ambient temperature*, high again at the time of the *low ambient temperature* during the heat phase of the cycle. The *low ambient temperature* had no significant effect to the embryos obtained. As the *high ambient temperature* had significant negative effect to the embryos obtained, it seems to be necessary to protect milking cows against the *high ambient temperature* during the heat time.

Key-Words: - embryo, ambient temperature, milking cow, flushing out

Introduction

Donor cows of particularly fine pedigree are treated with hormones to increase the number of eggs released at the ovulation [1]. The principle of the superovulation is basically simple: to induce more ovulations than the normal rate by giving a gonadotrophin stimulus (at critical moments of follicular development), followed by control of luteolysis, synchronous ovulation, high fertilisation and early embryonic development rates. The preparations to induce superovulation include: equine chorionic gonadotrophin, the follicle stimulating hormone and luteinizing hormone [2]. However the number of embryos obtained by flushing out is different every single time.

There is a question mark regarding the ambient temperature and its effect on the number of embryos flushed out from the donor. The number has the huge influence to the farm economy issue. Holstein cows give 5.26 embryos in average per a flushing out [3] and a low number of embryos obtained causes worst farm economy. As need to have a sufficient economy farmers try to improve the conditions of the flushing out. The target is to obtain as much embryos as possible. Therefore the aim of the project was to evaluate the effect of the ambient temperature on the uterus flushing out during embryo transfer process. Reproduction suffers as a result of heat stress on folliculogenesis and oocyte maturation as well as embryonic development and survival [4].

Under the influence of heat stress, the duration and the intensity of the estrus are reduced. There is a clear decrease in the mobility and other manifestations of estrus [5]. Follicle-stimulating hormone secretion from the pituitary gland does not appear to be impaired in animals exposed to the high ambient temperatures [6]. In contrast a clear reduction of production in the pulse and amplitude of luteinizing hormone release has been observed in cows exposed to heat stress [5]. The decrease in productivity and the negative reproductive effects render periods of heat stress, and those periods following, less profitable for dairy operations, making heat stress abatement practices cost effective and advantageous [4].

Material and Methods

The study was carried out in 33 dairy farms in The Netherlands. In total, 33 cows were included. The different ambient temperatures were regarded because the research was focused on the ambient temperature effect on the embryos obtained during the embryo transfer process. The main research question was: *What is the effect of the ambient temperature on the uterus flushing out during the embryo transfer process?*

Established hypothesis were: *The number of the embryos obtained is not different during the high/medium/low ambient temperature (H0) and the number of the embryos obtained is different during high/medium/low ambient temperature (H1).*

The milking cows for the uterus flushing out process were selected. They were divided according the ambient temperature into the 3 groups with 11 members per a group. The groups were marked by the letters A – C. The level of the high ambient temperature meant $\geq 24^{\circ}\text{C}$ [7]. The medium ambient temperature level was $> 4^{\circ}\text{C}$ to $< 24^{\circ}\text{C}$. The low level of ambient temperature was established on $\leq 4^{\circ}\text{C}$ [8]. The number of embryos obtained was high for > 8 embryos; the medium level was > 4 to < 8 and the low number presented < 4 embryos [3].

The cows in the group A were measured during *the high* ambient temperature. Group B was measured within *the medium* ambient temperature. The data about the group C were collected during *the low* ambient temperature. The group B formed a control group for the groups A and C.

The two veterinarians flushed out the uterus of the donor cows during the embryo transfer process. The experiment was focused on the embryos present in the flushed liquid. The flushed liquid was strained through a sterile sifter. After that it was observed under a microscope by trained assistants. The process of the uterus flushing out was done by the two different veterinarians. However the technical processes were absolutely uniform. Both of them had the similar workflow for the whole process of the uterus flushing out including the same tools used.

All flushing out results were written down to the Excel MS. Data were collected in MS Excel and analyzed by SPSS Statistics 17.0.

Results and Discussion

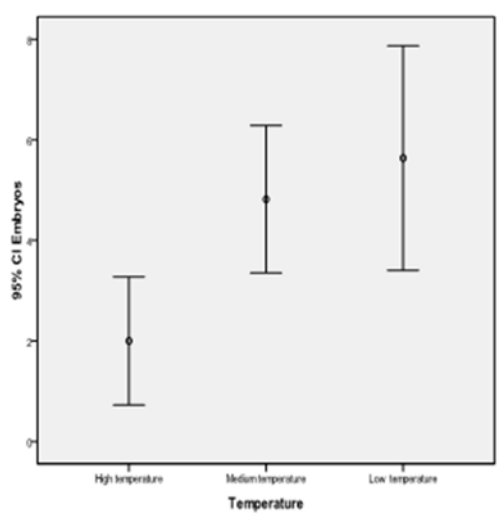
In regards to the project results the milking cows produced more embryos ($P < 0.05$) during the low ambient temperature than the cows during the high ambient temperature. Further the cows produced more embryos ($P < 0.05$) during the medium ambient temperature than the cows during the high ambient temperature. The high temperature had the significant ($P < 0.05$) negative effect to the embryos obtained.

Due to many projects which has been done in this area the negative influence of high ambient temperature was expected. This result is consistent with the data reported by [9] who reported that the production of embryos by the superovulation is often reduced in periods of heat stress (climatic factors that may influence the degree of heat stress include: temperature, humidity, radiation, and wind [10] and the associated reduction in the number of transferable embryos is due to the reduced superovulatory response, lower fertilization rate, and reduced embryo quality. Further is reported that the success of in vitro fertilization procedures is reduced during warm periods of the year and the heat stress can compromise the reproductive events required for the embryo production by decreasing expression of the estrus behavior, altering follicular development, compromising oocyte competence, and inhibiting embryonic development [9]. High ambient temperature reduced embryo quality [11]. The superovulation result is highly dependent on the kind of the gonadotropin used and on the frequency of the application [12]. On the other hand no significant influence on the embryo

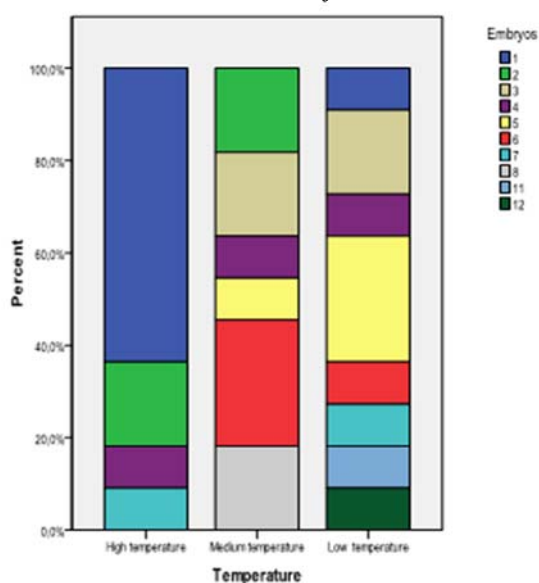
yield could be noted, regarding the day of the cycle on which the treatment is started [11]. Concerning the yield of embryos, the age of the donor has also no significant impact on the results [12].

The low ambient temperature had no significant ($P>0.05$) effect to the embryos obtained. Based on the outcome the zero hypothesis had to be rejected and at the same time the hypothesis number one was accepted. The number of the embryos obtained is different during the *high/medium/low* ambient temperature (H1).

The following (figure 1) shows the distribution of the population in every ambient temperature group (*the high, medium and low*). The every single segment illustrates the range of the distribution.



The other (Figure 2) shows frequency of the distribution. Every single color represents the certain number of the embryos obtained.



Conclusion

The negative influence of the high ambient temperature on the uterus flushing out during the embryo transfer process was expected. The embryos obtained were found to fluctuate with the ambient temperature, being the lowest ($P<0.05$) just during the high ambient temperature (24°C and higher), high during the medium ambient temperature (4.1°C - 23.9°C), high again at the time of the low ambient temperature (4°C and lower) during the heat phase of the cycle. As the high ambient temperature had the significant ($P<0.05$) negative effect to the embryos obtained, it seems to be necessary to protect the milking cows against high ambient temperatures during their heat time. The low air temperature had no significant effect to the embryos obtained. There is no need to protect them during low ambient temperatures. A possible solution could be selecting the most suitable ambient temperatures for the estrus evocation and the following embryo transfer process by cow's keepers. Even though the research has been done on the dairy breed specifically on Holsteins breed the results are easily extrapolated to other dairy breeds.

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References:

- [1] Anonymous, Superovulation (Multiple Ovulation) and Embryo Transfer, EDEXCEL Biotechnology Booklet, Edexcel Foundation [online] www.biopics.co.uk/edexcel/biotechnol/supovu.html (accessed Feb. 02, 2011).
- [2] Renaville R, Burny A, Biotechnology in Animal Husbandry (Focus on Biotechnology), Kluwer Academic Publisher, 2001.
- [3] Lazarevic R, Miscevic B, Influence of different hormonal treatment on number and quality of obtained embryos, *Institute for Animal Husbandry*, Vol.44, No.169, 2001, Special Issue.
- [4] Kennedy BS, Thermoregulation and the Effects of Heat Stress on Dairy Cattle, Production Medicine Graduate Program, 1999.
- [5] Kanitz W, et al., Superovulatin in cattle: practical aspects of gonadotropin

- treatment and insemination, *Reprod Nutr Dev.*, Vol.6, No.42, 2002, pp. 587–599.
- [6] Anonymous, Mechanism of the negative impact of heat stress on reproductive function in cattle, *Partners in reproduction*, [online] <http://www.partners-in-reproduction.com/reproduction-cattle/heat-stress-impact.asp>.
- [7] Broucek J, et al., Effect of high temperature on milk production of cow from free-stall housing with natural ventilation, *Animal Science*, Vol.42, No.4, 2009, pp. 167–173.
- [8] Dragovich D, Effect of low winter temperatures on milk production of dairy cows grazed on farms in a warm temperature climate (Australia), *International Journal of Biometeorology*, Vol. 24, No.2, 1980, pp. 167–173.
- [9] Hansen PJ, et al., Adverse impact of heat stress on embryo production: causes and strategies for mitigation, *Theriogenology*, Vol. 55, No. 1, 2001, pp. 91-103.
- [10] Jordan ER, Effect of Heat Stress on Reproduction, *Journal of Dairy Science*, Vol.86, 2003, pp. 104–114.
- [11] Benyei B, et al., Effect of the El Nino phenomenon on the ovarian responsiveness and embryo production of donor cows, *Acta Vet Hung.*, Vol.51, No.2, 2003, pp. 209–218.
- [12] Martens G, Research into the effect of different dosages and schedules of application of FSH on the results of superovulation in Simmental cows, School of Veterinary Medicine Hannover, Tierärztliche Hochschule, Dissertation, 2004.