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Genetic analysis for quality of frozen embryos produced by Holstein cattle donors in Canada

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ABSTRACT

The number of embryos produced by Holstein donors has been shown to be heritable, so it could be possible to genetically select for this trait to improve the efficiency of the assisted reproductive technology (ART) in dairy cattle. Another important parameter to consider for achieving good results from ART is embryo quality because embryos of good quality have more chance of producing live offspring. The possibility of using genetic selection for increasing the quality of embryo produced from ART has yet to be assessed. The objective of this study was, therefore, to perform a genetic analysis of embryo quality of Holstein donors in Canada using data recorded by Holstein Canada. The data set used was missing quality score data for embryos transferred fresh into a recipient, so the analyses were only performed for frozen embryos. With most traits in the Canadian dairy industry being evaluated with linear models, embryo quality was also evaluated with this class of models. However, considering the categorical nature of embryo quality, a threshold model was also evaluated. Embryo quality data were analyzed with either a univariate linear animal model or a univariate binomial threshold animal model. Genetic parameters estimated from the different models were comparable. A low heritability was found for the donor ($0.04 \pm <0.01$) and the service sire ($0.02 \pm <0.01$), but the repeatability estimate for the donor was higher (0.17), indicating that it was worthwhile to use a repeated records model. Overall, considering the low genetic parameters estimated, slow genetic progress is expected for the quality of frozen embryos produced by Canadian Holstein donors. Rank correlations were calculated between breeding values estimated from different models. High correlations were found between all models, indicating that no substantial re-ranking of the animals is expected from the different models. So, even though a threshold model is better suited for the analysis of categorical data, a linear model could be used for the analysis of embryo quality because it is less computationally demanding.
Key words: embryo quality, Holstein, genetic parameter, breeding value

INTRODUCTION

In dairy cattle, like in other species, the main goal of assisted reproductive technologies (**ART**) is to produce more live offspring from elite animals in a short period of time, aiming to accelerate the genetic gain of a population. To improve ART, 2 important parameters should be considered: the number of embryos produced and the quality of those embryos. In the short term, improving the technical aspects of ART could help produce more embryos of higher quality, but for a long-term improvement, genetic selection might be considered. In a previous study it was shown that it is possible to genetically select Holstein donors for increased embryo production (Jaton et al., 2016a). However, the possibility of genetic selection for improving embryo quality has yet to be assessed.

Embryos of higher quality usually have a higher chance of yielding a healthy offspring after being implanted in a recipient (Bó and Mapletoft, 2013). The birth of the offspring represents the ultimate assessment of embryo quality, but this implies a waiting period of at least 9 mo (Kanka et al., 2012). Therefore, other methods have been developed for evaluation of embryo quality to identify which embryos have a higher likelihood of getting a pregnancy (Van Soom et al., 2003). Nowadays, the most practical and noninvasive approach for choosing the best embryos for transfer is based on the morphology of the embryos (Van Soom et al., 2003; Farin et al., 2007; Kanka et al., 2012). In Canada and around the world, embryo quality is assessed visually by certified practitioners that use the classification system of the International Embryo Technology Society (**IETS**; Merton et al., 2003). To

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increase the uniformity across practitioners worldwide, IETS provides standardized coding systems that can help to assess the quality of the embryo at different stages of development (Bó and Mapletoft, 2013). Some of the factors that are considered for the morphological assessment of an embryo include color, developmental stage, size, compaction, and the presence of cellular debris (Van Soom et al., 2003; Bó and Mapletoft, 2013).

Large data sets of phenotypes measured at embryo recovery are required to perform a genetic analysis of embryo quality. Holstein Canada has such a data set, and the objective of the present study were therefore (1) to characterize the embryo quality data set, and (2) to estimate genetic parameters and EBV for embryo quality using different statistical models.

MATERIALS AND METHODS

Data

The data were provided by Holstein Canada and contained all viable embryos produced in Canada from 1980 to 2016, for a total of 1,334,414 records. This data set was complementary to the flushing data set analyzed in a previous study (Jaton et al., 2016b). Therefore, edits similar to Jaton et al. (2016b) were performed to include the same 150,971 superovulation and ovum pick-up and in vitro production of embryos (**OPU-IVP**) procedures. A total of 1,150,558 viable embryos were left after matching both data sets. These embryos were produced between January 1992 and January 2016 by 59,586 donors across Canada.

Around 36% of retained embryos were transferred fresh into a recipient and 64% were frozen after recovery. Key information was missing for the fresh embryos, namely the quality and the stage of development. Therefore, only frozen embryos were considered for further analysis. Moreover, additional edits were performed to only keep regular embryos, with complete information. Consequently, all embryos that were biopsied, divided, or produced by nuclear transfer were excluded from the data set. Veterinarians assessed the quality of the embryo with a score between 1 and 4, as suggested by IETS. Considering that embryos scored 4 for quality (n = 160) were considered to be dead or degenerating, they were excluded from the data set. Three quality scores were considered: 1 (excellent), 2 (good), and 3 (poor). In addition, considering the low number of embryos of quality 3 (n = 4,932), these embryos were grouped with quality 2 embryos for some analyses. The stage of development of the embryo was scored on a linear 1 to 9 scale, as defined by IETS. A higher score corresponds to a more developed embryo. Embryos with a developmental stage between 2 (2- to 12-cell) and 8 (hatched blastocyst) were included, whereas stages 1 (unfertilized; n = 253) and 9 (expanded hatched blastocyst; n = 54) were excluded. At recovery, the embryos need to be washed with sterile medium and trypsin, which is especially important if the embryos are going to be exported (Farin et al., 2007). Nowadays, it is recommended to wash the embryos 10 times with sterile medium (Farin et al., 2007) and 2 times with trypsin for a total of 12 times washed. In the data set, information about the number of times the embryos were washed was known, but information about the washing solution used was not available. The number of times the embryos were washed was grouped into 4 classes: embryos that were washed less than 10 times, embryos that were washed 10 and 11 times, embryos that were washed 12 times, and embryos that were washed more than 12 times. The data set also contained information about the zona pellucida, which is a spherical layer that surrounds the embryo between the zygote and blastocyst stage (Van Soom et al., 2003; Farin et al., 2007). In order for embryos to be certified for export, the zona pellucida must be intact. Records were excluded if information about the number of times the embryo was washed or the zona pellucida were missing.

The other information available was the service sire used for fertilization of the oocyte, the year and month of embryo recovery, the clinic that recovered the embryo, the age of the donor at recovery, and the type of service, which indicates if embryos were produced using superovulation or OPU-IVP. If superovulation was used, more details were available about the person that performed the insemination, who could be the herd owner or an AI technician. Only clinics with at least 100 viable embryos and only embryos recovered between 1992 and 2015 were considered. The final data set consisted of 714,534 viable embryos produced by 44,584 donors, 3,140 sires (sire of the donor), and 2,255 service sires (sire of the embryo).

A pedigree file containing 221,215 animals was generated by tracing the pedigrees of the donors with records 7 generations back.

Models

In many countries, most traits considered for genetic selection of dairy cattle are continuous and normally distributed so that they are analyzed using a standard linear model (Meijering and Gianola, 1985). However, ordered categorical traits, such as embryo quality, do not follow a normal distribution, so linear models may not be optimal for genetic selection purposes of these traits (Gianola and Foulley, 1983; Harville and Mee,