**ABSTRACT.** Breeding livestock for disease resistance is an attractive approach to increase animal health, longevity and profitability. Identifying dairy cattle that are more resistant to disease is now possible using High Immune Response™ (HIR) technology. Antibody-mediated immune response (AMIR), one heritable component of HIR™, was examined in a population of 1044 AMIR-tested Holsteins to investigate the effect of sire on AMIR performance of their offspring. In this population, 254 sire-offspring pairs were identified and groups of 50 high- and 50 low-responder offspring and their sires were compared. Mean AMIR phenotype in high responder offspring was 2 SD higher than in the low group. Sires of the high responder group had mean AMIR EBV 1 SD higher than sires of the low responder group (p <0.0001). These results suggest that bulls with higher EBV for AMIR, sire offspring with higher antibody responses (higher AMIR phenotype).

**Keywords:** High immune response; Holstein; Estimated breeding value; Offspring performance

**Introduction**

The immune system is a complex network of tissues, cells and molecules critical to the maintenance of animal health. This system is controlled by more than 3,000 genes in mammals and its performance can be affected by various genetic and epigenetic interactions (Breuer et al. (2013); Paibomesai et al. (2013); Wilkie and Mallard (1999)).

Quantifying the effective performance of the immune system against disease is complicated by the wide range of pathogens operating in diverse environments. Nonetheless, two key components of the adaptive immune system, antibody- (AMIR) and cell-mediated immune response (CMIR) are critical to the control of extracellular and intracellular pathogens, respectively (Thompson-Crispi et al. (2012a)).

Recently, a novel method called the High Immune Response (HIR)™ technology has been successfully applied to measure the performance of the adaptive immune system of dairy cattle and classify animals into 3 classifications - high, average and low immune responders. Studies have shown that these traits are moderately to highly heritable (Wagter et al. (2000); Thompson-Crispi et al. (2012a)) and that high responder animals are more resistant to various diseases (Thompson-Crispi et al. (2012b)). Additionally, production traits are not negatively affected by immune response performance (Thompson-Crispi et al. (2012a)).

Recently, the Semex Alliance company has utilized the HIR™ technology to identify sires that exhibit robust and balanced AMIR and CMIR. This new line of sires, marketed under the Immunity+™, is those with the highest EBV for both immune response (IR) traits.

Previous studies have demonstrated that cows with the highest AMIR are half as likely to get clinical mastitis as those with low AMIR (Thompson-Crispi et al. (2013)). Given the economic importance of this disease, AMIR was the focus of the current study. The association of sires’ EBVs with their offsprings' AMIR phenotypes was investigated.

**Materials and Methods**

**Animals:** The data set consisted of 1,044 IR-phenotyped Holstein bulls (n=763) and cows (n=281). Holstein bulls were housed and immune response tested within one of the 3 Semex Alliance bull testing facilities (EastGen, WestGen or Centre d'Insémination Artificielle de Québec [CIAQ]). Holstein cows were housed and immune response tested within the Elora Research Facility of the University of Guelph. A total of 254 sire-offspring IR-tested pairs were identified. The sire-offspring pair group consisted of 51 unique sires and their 187 male and 67 female offspring. Two groups of high and low responders, each consisting of 50 sire-offspring pairs, were selected based on AMIR performance of the offspring. The high responder group consisted of 14 cows and 36 bulls from 29 unique sires. The low responder group consisted of 19 cows and 31 bulls from 21 unique sires. These animals were all older than 2 months of age on the test date and no clinical symptoms of disease were observed during the immune response testing procedure.

**Antibody-mediated Immune Response:** Animals in this study were immune response phenotyped using the HIR™ method (US Patent #7,258,858; Wagter-Lesperance and Mallard 2007) Briefly, serum samples were collected at day 0 of test and cattle were then immunized with a type 2 test antigen. Serum samples were collected 14 days after immunization, aliquoted and stored frozen until antibody was quantified by ELISA as described previously (Thompson-Crispi et al. (2012a)).
Phenotypic Analysis: Optical densities (OD) from ELISA assays were log transformed to normalize their distribution. The data were analyzed using SAS PROC GLM (SAS 9.1.3, SAS Institute, Cary, NC). Normality of residuals was tested using Shapiro-Wilk statistics. The statistical model was:

\[ y_{ijkl} = \mu + h_i + p_j + m_k + hp_{ij} + \beta_1 \times a_l + \beta_2 \times d_l + e_{ijkl} \]  

Where \( y_{ijkl} = \log_{10} \) of OD at day 14 for the \( i^{th} \) animal; \( h_i = \) fixed effect of the \( i^{th} \) housing facility; \( p_j = \) fixed effect of the \( j^{th} \) phase of testing; \( m_k = \) fixed effect of the \( k^{th} \) pregnancy status (heifer, pregnant cow, non-pregnant cow and bull); \( hp_{ij} = \) interaction effect of the \( i^{th} \) phase and the \( j^{th} \) housing facility; \( \beta_1 = \) linear coefficient of the fixed regression on age \((a_i)\) of the \( l^{th} \) animal (in months); \( \beta_2 = \) linear coefficient of the fixed regression on \( \log_{10} \) of OD at day 0 \((d_l)\) of the \( l^{th} \) animal; \( e_{ijkl} = \) random residual effect.

Standardized Z-values of the animals’ residual effects in model [1] (adjusted \( \log_{10} \) of OD at day 14) were calculated and animals with the 50 highest and 50 lowest Z-values were categorized into high responder and low responder groups, respectively.

Genetic Analysis: To estimate (co)variance components and breeding values for AMIR, the same statistical model [1] as for the phenotypic analysis was used, but with the addition of the animal random additive genetic effect \((u_i)\). The covariance between animals was modeled by the additive genetic relationship matrix, using the pedigree information extracted from the Canadian Dairy Network database, including a total of 23,913 records. The genetic analysis was carried out using ASReml 3.0 software, which uses a restricted maximum likelihood algorithm for estimation of (co)variance components. Heritability value was calculated as:

\[ h^2 = \frac{\sigma^2_u}{\sigma^2_u + \sigma^2_e} \]

Where \( \sigma^2_u \) is the additive genetic variance and \( \sigma^2_e \) is the random residual variance.

Finally, Sires’ EBVs of high responder and low responder animals were standardized to Z-values for comparison between low and high responder groups. Differences in mean phenotype or EBV between groups were tested using Students’ t-test (independent, two-tailed).

Results and Discussion

The estimated heritability of AMIR in this population was 0.50 (SE= 0.06). The mean AMIR phenotype \( Z \)-value of the high responder offspring was +1.26 and the mean AMIR phenotype \( Z \)-value of the low responder offspring was -1.39 (Figure 1). This difference was statistically significant (\( p < 0.0001 \)). The mean EBV \( Z \)-values among the sires of high responder offspring was +0.27 and the mean was -0.81 among sires of low responder offspring (Figure 1). The difference between the EBVs of sires of low responder and high responder offspring was statistically significant (\( p < 0.0001 \)). In addition, the linear regression of the adjusted phenotypes of offspring on the sires’ EBV was positive (\( b = 0.81 \)) and significant (\( p < 0.0001 \)) (Figure 2).

![Figure 1. Mean standardized adjusted offspring phenotypes and mean sire EBVs in high responder and low responder groups. * indicates significant differences (p <0.0001).](image)

![Figure 2. Regression analysis of standardized adjusted offspring phenotypes (y axis) on sire EBVs (x axis).](image)

Immune response function is affected by environmental elements (e.g. management) as well as pathogens. The heritability of immune response traits varies depending on the immune parameter measured, the species and the size of the studied populations (Emam et. al. 2014). The HIR™ technology was developed to quantify immune responses in dairy cattle. Since it is based on breeding values, it minimizes environmental and pathogen effects and therefore represents the most likely estimate of genetic potential for immune response. The high heritability of AMIR in this study aligns with estimates of heritability of AMIR in other studies using HIR in different populations (Thompson-Crispi et al. 2012a)).
The significant association between sires' EBV and offspring phenotype shows the possibility of genetic improvement for AMIR in dairy Holstein through sire selection. The contribution of genetic potential of sires for AMIR is similar to some other production traits, such as milk production (Wiggans et al. (2011)). Therefore, it may be that the trend of genetic improvement for AMIR by selective breeding will be similar to that for milk production.

Conclusion

The positive association between sires’ EBVs and their offsprings' phenotypes for AMIR emphasizes the possibility of breeding for higher AMIR in Holstein dairy cattle. Since AMIR is associated with resistance to diseases, such as mastitis, disease occurrence can be decreased by using bulls with superior immune responses.

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Literature Cited