

Breeding and Genetics: Application and methods—Dairy II

748 The “it factor” for long-lived, high-producing dairy cows.

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Holstein cows that dairyman love live a long time and produce lots of milk. What is unique about these high-producing, long-lived cows? Obviously, these cows have received good management and avoided major health issues. Are the 50K genetics of these elite cows different from other Holsteins? Elite cows were defined phenotypically as having produced over 68,039 kg (150,000 lb) of milk during their lifetime and were classified as very good (VG) or excellent (EX). Elite cows were born in the decade before 2008. For a control, females born in the decade before 2008 with a 50K Holstein genome evaluation were chosen. Control females either had not produced 68,039 kg of milk during their lifetime or were not classified VG or EX. Genomes (50K or 77K) were available on 823 elite cows and 1,589 control females. Defining elite or control as binomial allowed detection of almost 200 markers that were different in allele frequency between elite cows and control females. The most significant chi-squared for differences in allele frequency between elite cows and control females identified a marker on chromosome 5, which had the largest difference in minor allele frequency of 0.17 between elite and control groups. Basing significance on chi-squared $-\log_{10} P$ of 8.000 as a threshold, 199 markers were significant and were distributed across all bovine chromosomes. Minor allele frequencies of elite cows were greater for 140 of these markers and minor allele frequencies of control females were greater for 59 markers. As interpretation, minor allele frequencies define uniqueness. The uniqueness of elite cows was supported by more positive changes in minor allele frequencies for the elite cows. A single “it factor” is insufficient to identify uniqueness of elite cows, but many markers are candidates to contribute to the uniqueness. Because allele frequency differences were found across all chromosomes, balance continues to be important in striving to increase the number of elite cows in the breed.

Key Words: genome, milk production, longevity

749 Identification of gene networks underlying dystocia in dairy cattle.

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Dystocia is a trait with high impact in the dairy industry. Among its risk factors are calf weight, gestation length, breed and conformation. Biological networks have been proposed to capture the genetic architecture of complex traits, where GWAS show limitations. Our objective was to identify gene networks in Brown Swiss (BS), Holstein (HO) and Jersey (JE) cattle related to dystocia. De-regressed PTA (dPTA) for calving ease, gestation length, stature, strength and rump width of 8780 HO, 505 BS, and 1818 JE bulls were used in the analysis. A total of 45188 genotypes were available for all bulls. A single trait Bayes B GWAS was performed within breed with $\pi = 0.9$. The proportion of genetic variance (PV_g) explained by each SNP was $(2pq\tilde{a}^2)/\sum^{45188}(2pq\tilde{a}^2)$, with \tilde{a} = posterior mean of the allelic effect. SNP with $VP_g \geq 75$ th percentile of the sample were ruled significant. Relevant SNP (rSNP) were defined as: significant in all traits, significant in all functional traits, or significant in all type traits. An association weight matrix (AWM) was constructed with rSNP in rows and traits in columns. Cells of the AWM corresponded

to rSNP normalized effect size. These were mapped to genes with a 5' or 3' maximum distance of 2500 bp, rows in the AWM were indexed with genes. Genes were used to search for enriched functional annotation (FDR ≤ 0.15 HO, JE; FDR ≤ 0.3 BS). AWM row-wise partial correlations were computed. Significant correlations were interpreted as gene-gene interactions, resulting in a gene network. Networks included 1454 (BS), 1272 (HO) and 1455 (JE) genes. Their number of connections ranged between 1 and 15 (BS), 80 (HO), 13 (JE). A total of 13 (BS), 152 (HO), 108 (JE) genes in the networks were within reported dystocia QTL. Top enriched terms were cell adhesion (HO, JE), regulation of purine nucleotide metabolic process (BS). Most connected genes in the networks, enriching GO terms and within dystocia QTL were: FLOT1 (BS, 9 interactions), RASA1 (HO, 77) and ADRBK2 (JE, 12). Integrating knowledge from annotation tools to identify the functional biology of dystocia in dairy cattle can potentially improve genomic predictions that could result in increasing profitability of the dairy industry.

Key Words: dystocia, gene network, dairy cattle

750 Distribution of runs of homozygosity and its association with inbreeding depression in United States and Australia Jersey cattle.

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Differences in environment, management practices or selection objectives have led to a variety of choices being made in the use of dairy sires between countries. This may result in variation in selection intensity across the genome and could result in detectable differences in patterns of genome-level homozygosity between populations and consequently affect inbreeding depression differently across populations. The objective of the study was to characterize the frequency of homozygosity and its relationship with regions associated with inbreeding depression in Jersey dairy cattle from the United States (US) and Australia (AU). Genotyped cows with phenotypes on milk, fat and protein yield ($n = 6,751$ US; $n = 3,974$ AU) and calving interval ($n = 5,816$ US; $n = 3,905$ AU) were utilized in a 2-stage analysis. A run of homozygosity statistic (ROH4Mb), counting the frequency of a SNP being in a ROH of at least 4 Mb, was calculated across the genome. In the first stage residuals were obtained from a model that accounted for the additive genetic as well as fixed effects. In the second stage these residuals were regressed on ROH4Mb using a single marker regression model or a machine-learning tree based regression algorithm (gradient boosted machine). The relationship between ROH4Mb and the SNP effect of a region for each trait was further characterized based on sliding window (500kb) direct genomic value (DGV) derived from a Bayesian LASSO analysis. The ROH4Mb effects were estimated by regressing residuals from the 2-stage approach on ROH4Mb and SNP effects estimated by regressing residual deviations from a model including only fixed effects on SNP markers. Genomic regions across multiple traits were found to be associated with ROH4Mb on BTA13, BTA23 and BTA25 for the US population and BTA3, BTA7, BTA17 for the AU population. Furthermore, multiple potential epistatic interactions were characterized. Lastly, the covariance between ROH4Mb and the SNP effect of

a region depended on the genome region, with positive covariances in some regions

Key Words: run-of-homozygosity, genome-wide association study, inbreeding depression

751 The effect of *DGATI* polymorphism on milk production and fat, protein, and mineral composition of dairy cattle. Henk Bovenhuis*¹, Marleen Vikser¹, Nina Poulsen², Jakob Sehested³, Hein van Valenberg⁴, Johan van Arendonk¹, Lotte Bach Larsen², and Bart Buitenhuis⁵, ¹*Animal Breeding and Genomics Centre, Wageningen University, Wageningen, the Netherlands*, ²*Department of Food Science, Aarhus University, Tjele, Denmark*, ³*Department of Animal Science, Aarhus University, Tjele, Denmark*, ⁴*Dairy Science and Technology Group, Wageningen University, Wageningen, the Netherlands*, ⁵*Center for Quantitative Genetics and Genomics, Department of Molecular Biology and Genetics, Aarhus University, Tjele, Denmark*.

Since the identification of the diacylglycerol O-acyltransferase 1 (*DGATI*) K232A polymorphism, many studies reported associations between this polymorphism and milk production traits but only a few investigated effects on detailed milk composition. In the current study data collected as part of the Dutch and Danish-Swedish Milk Genomic Initiatives were used to estimate the effect of *DGATI* polymorphism on milk fat, protein, and mineral composition in Holstein and Jersey cattle. The use of data from different breeds allows identifying associations of *DGATI* that are consistent across populations. Results showed that effects of *DGATI* K232A polymorphism on especially milk yield, fat% and protein% are not constant during lactation. Effects of *DGATI* were small in early lactation and they gradually increased until d 50 to 150 in lactation. Analyses of the effects of *DGATI* polymorphism on detailed milk fat, milk protein, and mineral composition showed that *DGATI* has major effects on fat- and mineral composition of milk. There is also evidence for effects on milk protein composition but these effects seem to be more subtle. Part of the effects of the *DGATI* polymorphism on milk composition can be explained by effects on de novo fatty acid synthesis and on excretion of water (dilution effect). For example, the total amount of Ca, P, and Zn excreted in milk of cows with different *DGATI* K232A genotypes is identical; however, the milk volume differs and therefore contents of Ca, P, and Zn differed between *DGATI* genotypes. A similar "dilution effect" can be observed when studying effects on fatty acids: there were no significant effects of *DGATI* on the yields of C18:1 *cis*-9, CLA *cis*-9,*trans*-11, C18:2 *cis*-9,12 and C18:3 *cis*-9,12,15; however, there were highly significant effects of *DGATI* on these fatty acids when expressed as w/w%.

Key Words: *DGATI*, mineral, fatty acid

752 Variation in milk fat globule size in Canadian dairy cattle and its prediction using mid-infrared spectroscopy. Allison Fleming*¹, Astrid Koeck¹, Flavio Schenkel¹, Milena Corredig^{2,3}, Mehdi Sargolzaei^{1,4}, Bonnie Mallard⁵, R. Ayesha Ali⁶, Saranya Gunasegaram¹, and Filippo Miglior^{1,7}, ¹*CGIL, University of Guelph, Guelph, ON, Canada*, ²*Gay Lea Foods, Mississauga ON, Canada*, ³*Dept of Food Science, University of Guelph, Guelph, ON, Canada*, ⁴*Semex, Guelph, ON, Canada*, ⁵*Dept of Pathobiology, OVC, University of Guelph, Guelph, ON, Canada*, ⁶*Dept of Mathematics and Statistics, University of Guelph, Guelph, ON, Canada*, ⁷*Canadian Dairy Network, Guelph, ON, Canada*.

Milk fat globule (MFG) size in bovine milk is a trait of interest for selection due to its influence on the composition and nutritional quality of the milk as well as its technological properties. However, large-scale phenotyping is currently impractical. For a given fat content, milk with smaller MFG will have more membrane material, which has been proposed nutraceutical. The objective of this study was to examine the variation in MFG size and the potential of its prediction using mid-infrared spectral data. A total of 1,689 milk samples from 343 Canadian cows representing 4 dairy breeds from 44 herds across the provinces of Ontario, Alberta, and Quebec were collected during routine milk testing. Samples were divided and a certain portion was sent to a Canadian DHI laboratory and their spectral data were recorded using FOSS MIR machines. The MIR data for each sample contained 1,060 data points in the infrared range from 900 to 5,000 cm⁻¹. The additional portion was analyzed for mean MFG size using integrated light scattering and reported as both a volume moment mean and surface moment mean. Mean values (±SD) for volume and surface moment means were 4.16 ± 0.51 μm and 3.49 ± 0.34 μm, respectively. The average MFG size was positively correlated with the percent fat in the sample (volume moment mean, r = 0.30; surface moment mean, r = 0.32). MFG size records were combined with their spectral data, and outliers and non-informative regions of the spectrum were removed. Records were randomly assigned to either the training or validation sets. Partial least squares regression method was utilized to predict MFG size. For volume moment mean, an R²_{cv} of 0.52 and R²_v of 0.41 were found. Surface moment mean equations had an R²_{cv} of 0.55 and R²_v of 0.52. At this time MFG size cannot be accurately quantified through MIR prediction, but it may be possible to identify milk samples with either small or large MFG.

Key Words: milk fat globule, dairy cattle, mid-infrared spectroscopy

753 Including different groups of genotyped females for genomic prediction in the Nordic Jersey population. Hongding Gao*¹, Per Madsen¹, Ulrik S. Nielsen², Gert P. Aamand³, and Just Jensen¹, ¹*Center for Quantitative Genetics and Genomics, Department of Molecular Biology and Genetics, Aarhus University, Tjele, Denmark*, ²*Knowledge Centre For Agriculture, Aarhus N, Denmark*, ³*Nordic Cattle Genetic Evaluation, Aarhus N, Denmark*.

Including genotyped females in the reference population (RP) is an obvious way to increase RP but caution is needed because of potential preferential treatment of the genotyped cows and lower reliabilities of phenotypes compared with proven bulls. Denmark, Finland and Sweden have implemented a female genotyping project with voluntary genotyping of entire herds using low-density chip (LD project). The objective of the present study was to examine the effect of adding different sources of genotyped females to RP for Nordic Jersey. Five scenarios for building RP were considered: (1) bulls only; (2) bulls with females from LD project; (3) bulls with females from LD project plus non-LD project females genotyped before their first calving date; (4) bulls with females from LD project plus non-LD project females genotyped after their first calving date; (5) bulls with all genotyped females included. Genomically enhanced breeding values (GEBV) were predicted for 8 traits in the Nordic Total Merit (NTM) index through a genomic BLUP (GBLUP) model using deregressed proofs (DRP). The validation population (VP) was formed by a cut-off using birth year of 2005 based on the genotyped bulls with DRPs. Average gain in reliability over the 8 traits ranged from 1.8% to 4.5% points compared with the scenario with only bulls in RP (scenario 1). Adding all the genotyped females in the RP achieved highest gain in reliability (scenario 5), followed by scenario 3, scenario 2 and scenario 4. The mean reliability of scenario 3 was 0.5% points higher than scenario 2 due to a slightly larger size

of RP, and a decrease of 1.1% points in mean reliability were observed when including the extra 143 genotypes cows in scenario 4 compared with scenario 2. The mean reliabilities of scenario 2 and 3 were 1.6 and 1.1% points lower than of scenario 5. All scenarios led to inflated GEBVs since the regression coefficients are below 1. However, scenario 2 and scenario 3 led to less bias of genomic predictions than scenario 5 with the mean regression coefficients closer to 1. The results suggest adding unselected females in the RP significantly improve the reliabilities and tend to reduce the prediction bias compared with adding selectively genotyped females.

Key Words: genotyped female, reliability, prediction bias

754 Estimation of genetic parameters for metabolic disease traits and their predictors in Canadian Holsteins. Astrid Koeck^{*1}, Janusz Jamrozik^{1,2}, Gerrit J. Kistemaker², Flavio S. Schenkel¹, Robert K. Moore⁴, Daniel M. Lefebvre⁴, David F. Kelton³, and Filippo Miglior^{1,2}, ¹CGIL, Dept. of Animal and Poultry Science, Guelph, ON, Canada, ²Canadian Dairy Network, Guelph, ON, Canada, ³Department of Population Medicine, Ontario Veterinary College, Guelph, ON, Canada, ⁴Valacta, Québec, QC, Canada.

The objective of this study was to estimate genetic parameters for metabolic diseases and their main predictors in Canadian Holsteins. Records from first to fifth lactation were considered for ketosis (KET), displaced abomasum (DA), milk fever (MF), fat to protein ratio (F:P) and milk β -hydroxybutyrate (BHBA), whereas for body condition score (BCS) only records from first lactation cows were available. Binary disease traits (0 = no case, 1 = at least one case), F:P and milk BHBA were treated as different traits in first and later lactations. Records for MF in first lactation were not considered in the present study as the disease frequency was near zero and a preliminary analysis revealed a heritability of zero. Bivariate and multivariate linear sire models were fitted using AI-REML. Heritability for metabolic disease traits ranged from 0.011 to 0.047. Higher heritabilities were found for BCS, F:P and milk BHBA, with estimates ranging from 0.10 to 0.22. First-lactation KET was strongly correlated with DA (0.76) and milk BHBA (0.75), whereas lower genetic correlations were found with BCS and F:P (-0.54 and 0.37, respectively). Displaced abomasum in first lactation was moderately correlated with BCS (-0.40) and F:P (0.19). Similar genetic correlation estimates were estimated in higher lactation cows. Milk fever, which was only evaluated in higher lactation cows, was moderately correlated with KET (0.39) and milk BHBA (0.33). Genetic correlations of disease traits between first and later lactations were relatively high (0.79 for KET and 0.86 for DA).

Key Words: metabolic disease, predictor, genetic correlation

755 A genomic-wide association study on development of hyperketonemia in periparturient Holstein dairy cows. Francisco A. Leal Yepes^{*1}, Heather J. Huson¹, Sabine Mann², Jessica A. A. McArt², Luciano Caixeta¹, Thomas R. Overton¹, Joseph J. Wakshlag², and Daryl V. Nysdam², ¹College of Agriculture and Life Sciences, Cornell University, Ithaca, NY, ²College of Veterinary Medicine, Cornell University, Ithaca, NY.

The objective was to detect cows with elevated postpartum nonesterified fatty acids (NEFA) and β -hydroxybutyrate (BHBA) concentrations, with or without concurrent hyperketonemia, and identify genomic regions associated with development of hyperketonemia in periparturient Holstein cows. The study population consisted of cows from 2 different trials: In the first study, 63 cows (parity ≥ 2) were enrolled and blood

was collected from 3 to 16 d in milk. In the second study, 84 cows (parity ≥ 2) were enrolled and sampled from -21 d to +21 d relative to calving. Blood samples were tested for NEFA and BHBA concentration. Hyperketonemia was defined as a BHBA concentration ≥ 1.2 mmol/L. All BHBA and NEFA measurements were grouped using incremental area under the curve (AUC) to identify individuals with the most variation. Holstein cows were genotyped on the Illumina Bovine High-density (777K) Beadchip. Quality control filtering produced ($n = 522,231$) single-nucleotide polymorphism (SNP). A genomic wide association study was performed to establish correlation between low frequency SNP ($<5\%$) and development of hyperketonemia using Golden Helix software. The linear regression $R^2 = 0.21$ suggested a low strength correlation between BHBA AUC and NEFA AUC concentration. Although a small sample size, given that these cows were managed under similar conditions, multiple SNP associated with high concentrations of BHBA were found (Table 1). These results might improve genetic selection criteria to identify high-risk animals and develop preventative measures to decrease hyperketonemia development.

Table 1 (Abstr. 755). Regions and candidate genes associated with development of hyperketonemia

Index	Chr.	Region start (bp)	Region end (bp)	$-\text{Log}_{10}$ (P-value)	Genes
1	2	12,122,028	12,128,393	5.086193951	<i>RRAGA</i> <i>PECR</i> ; <i>IGFBP2</i> and <i>IGFBP5</i>
2	2	104,958,840	104,958,840	5.410113588	<i>LRP8</i> ; <i>CPT2</i> and <i>SCP2</i>
3	3	93,634,769	93,634,769	5.031371132	<i>CHD2</i>
6	21	14,153,070	14,153,070	5.111968697	<i>SV2B</i>
7	21	16,112,290	17,017,717	6.451923226	

Key Words: hyperketonemia, SNP, gene

756 Evaluation of survival in the first year after calving across years and seasons. Mathijs L. van Pelt^{*1,2} and Roel F. Veerkamp¹, ¹Wageningen UR Livestock Research, Animal Breeding and Genomics Centre, Wageningen, the Netherlands, ²CRV BV, Arnhem, the Netherlands.

Longevity of dairy cattle is an important trait from an economic and welfare perspective. Dairy cows are culled for various reasons and therefore the trait definition of longevity might have changed over time. Also, culling is likely to be affected by the end of the quota year, and therefore a seasonal effect is expected. Therefore the objective was to evaluate if longevity is a different trait between years or seasons of first calving, utilizing genetic links through common bulls. Survival was defined as survival until 12 mo after first calving. The data set comprised 524,529 animals that could have lived until 12 mo after the first calving, with the first calving between 1988 and 2011. Multiple trait models were used with survival defined in 3 8-year blocks (1988–1995, 1996–2003, and 2004–2011) or 4 seasons of first calving (Jan-Mar, Apr-Jun, Jul-Sep, and Oct-Dec). The mean survival was 87.8% in the total data set, but increased with more recent calving years. The lowest survival rate was in 1991 with 82.9% surviving the first 12 mo and highest survival rate was in 2007 (92.2%). Survival rate also varied between calving months, with highest survival rate in September with 89.2% and lowest survival rate in March with 86.3%. Age of first calving showed an optimum for survival for heifers calved between 22 and 26 mo of age, with survival rates of 88.4% or higher. Survival rate decreases 0.75% for each extra month calving after 26 mo. Genetic correlations between 1988 and 1995 and 2004–200 was lowest (0.67 ± 0.15), and suggests survival

has changed over years. Genetic correlations between all seasons were higher than 0.90, and although there appears a phenotypic effect of the end of the quota year on survival, genetically there is no effect of season of calving on survival until 12 mo after first calving.

Key Words: longevity, survival, genetic correlation

757 Genetic and genomic analysis of superovulatory response in Canadian Holsteins. Cindy Jaton*^{1,2}, Astrid Koeck¹, Mehdi Sargolzaei^{1,3}, Christopher A. Price⁴, Flavio S. Schenkel¹, and Filippo Miglior^{1,5}, ¹*Centre for Genetic Improvement of Livestock, University of Guelph, Guelph, ON, Canada*, ²*Centre d'insémination artificielle du Québec, St-Hyacinthe, QC, Canada*, ³*Semex Alliance, Guelph, ON, Canada*, ⁴*Université de Montréal, Faculté de Médecine Vétérinaire, St-Hyacinthe, QC, Canada*, ⁵*Canadian Dairy Network, Guelph, ON, Canada*.

Superovulation of dairy cattle is frequently used in Canada. The cost of this protocol is high, and so is the variability of the outcome. Knowing the superovulatory potential of a donor cow could influence the breeder's decision to superovulate it or not. The ultimate objective of this study was to identify chromosome regions associated with superovulatory response in Canadian Holsteins. Data were provided by Holstein Canada and contained the total number of embryos and the number of viable embryos from every successful flushing performed across Canada. After editing, 137,446 records of superovulation done between 1992 and 2014 were considered for the analysis. A univariate repeatability animal model analysis was performed for both total number of embryos and number of viable embryos, yielding heritability estimates (SE) of 0.18 (0.01) and 0.14 (0.01), respectively. Breeding values were estimated for 54,463 cows, and 3,513 sires. Only estimated breeding values of animals that were genotyped with a SNP panel denser than 3K and having a reliability higher than 40% were considered ($n = 5,122$) for further analyses. All lower density genotypes were imputed to 50K using FImpute software. A genome-wide association study was carried out using a single SNP regression method, which also fits a polygenic background effect. Results were similar for the 2 highly genetically correlated ($r_g = 0.94$) traits considered in this study. A major significant peak on chromosome 11 was detected, suggesting the presence of an important candidate gene in this region.

Key Words: superovulatory response, heritability, GWAS

759 Genetic parameters of individual hoof lesions in Canadian Holsteins. Francesca Malchiodi*¹, Astrid Koeck¹, Núria Chapinal², Mehdi Sargolzaei^{1,3}, Allison Fleming¹, David F. Kelton⁴, Flavio S. Schenkel¹, and Filippo Miglior^{1,5}, ¹*Centre for Genetic Improvement of Livestock, University of Guelph, Guelph, ON, Canada*, ²*Animal Welfare Group, University of British Columbia, Vancouver, BC, Canada*, ³*Semex Alliance, Guelph, ON, Canada*, ⁴*Department of Population Medicine, Ontario Veterinary College, Guelph, ON, Canada*, ⁵*Canadian Dairy Network, Guelph, ON, Canada*.

Hoof lesions in dairy herds represent a painful condition for the cow and pose a financial loss for farmers, due to the costs associated with treating lesions, as well as to decreased cow performance. The objective of this study was to estimate genetic parameters for individual hoof lesions in Canadian Holsteins. Data were recorded by 26 hoof trimmers serving 365 herds located in Alberta, British Columbia and Ontario, and trained to use a rugged touch-screen computerized lesion recording system. A total of 108,032 hoof-trimming records from 53,654 cows were collected between 2009 and 2012. Hoof lesions included in the analysis were digital dermatitis, interdigital dermatitis, interdigital hyperplasia, sole hemorrhage, sole ulcer, toe ulcer, and white line lesion. All variables were analyzed as binary traits, as the presence or the absence of the lesions, and as categorical variables, using a severity score from 1 to 3. Only the first hoof-trimming session of each lactation was included in the analyses. When considering the presence or absence of the lesions, heritabilities (SE) for digital dermatitis, interdigital dermatitis, interdigital hyperplasia, sole hemorrhage, sole ulcer, toe ulcer, and white line lesion were 0.053 (0.005), 0.011 (0.002), 0.025 (0.004), 0.012 (0.002), 0.031 (0.004), 0.004 (0.001), and 0.012 (0.002), respectively. Similar results were found when severity was considered. The estimated breeding values between these 2 set of variables showed correlations ranging from 0.88 to 0.97. The reliability of EBV was slightly higher when severity was analyzed. Infection lesions showed moderate genetic correlations with interdigital hyperplasia and low negative correlations with horn lesions. Among horn lesions, moderate to high genetic correlations were found.

Key Words: hoof lesion, dairy cattle