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Genome-wide association for heifer reproduction and calf performance traits in beef cattle

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Abstract

The aim of this study was to identify SNP markers that associate with variation in beef heifer reproduction and performance of their calves. A genome-wide association study was performed by means of the Generalized Quasi-Likelihood Score (GQLS) method using heifer genotypes from the BovineSNP50 BeadChip and estimated breeding values for pre-breeding body weight (PBW), pregnancy rate (PR), calving difficulty (CD), age at first calving (AFC), calf birth weight (BWT), calf weaning weight (WWT) and calf pre-weaning average daily gain (ADG). Data consisted of 785 replacement heifers from three research herds namely; Brandon Research Centre, Brandon, University of Alberta Roy Berg Kinsella Ranch, Kinsella and Lacombe Research Centre, Lacombe, Canada. After applying a false discovery rate correction at a 5% significance level, a total of 4, 3, 3, 9, 6, 2, and 1 SNPs were significantly associated with PBW, PR, CD, AFC, BWT, WWT and ADG, respectively. These SNPs were located on chromosomes 1, 5 - 7, 9, 13 - 16, 19 - 21, 24, 25 and 27 - 29. Chromosomes 1, 5 and 24 had SNPs with pleiotropic effects. New significant SNPs that impact functional traits were detected, many of which have not been previously reported. The results of this study support quantitative genetic studies related to the inheritance of these traits, and provides new knowledge regarding beef cattle quantitative trait loci effects. The identification of these SNPs also provides a starting point to identify genes affecting heifer reproduction traits and performance of their calves (BWT, WWT and ADG), improve the understanding of the biology underlying these traits, and for potential use in marker- and genome-assisted selection and management.

Keyword: GWAS, SNP, allele substitution, genotypic effect, heifer reproduction, beef cattle

Introduction

The development of replacement heifers for breeding is a significant expense for most cow-calf operations in the beef cattle industry. It is estimated to cost between \$1000 to \$1500 to develop a heifer from weaning to breeding and in most cases a mature cow may not reach optimum performance until their 5th year. Consequently, producers are careful in the selection of replacement heifers as this affects the genetic progress and profitability of their herds. Although, sorting of the female calf crop and selection of replacement heifers can begin before weaning, breeders can make a more informed decision in selecting fertile and productive heifers if additional information is available early in life. The fertility of replacement heifers and their postpartum performance are important traits that minimise the non-productive period and help to maintain a seasonal calving pattern. However, improvement of reproduction traits in beef cattle using traditional selection practices has had limited success due to low heritability and delayed expression of traits in the life of the animal (MacNeil et al. 2006).

Recent advances in molecular genetic technology have developed massive genotype information in the form of single nucleotide polymorphism (SNP) panels for animal breeding. Currently, different SNP panel densities are commercially available for cattle (Affymetrix 2013; Illumina 2013) ranging from a few thousands to more than three million SNPs (Khatkar et al. 2012). This is useful for genome-wide association studies (GWAS) to identify chromosomal regions and SNP markers associated with quantitative trait loci and could clarify inheritance of complex traits (Maher 2008). In addition, if SNP markers are identified, they could be used in marker-assisted management programs or for genetic improvement of livestock through genomic

selection (Meuwissen et al. 2001), by focusing on relevant SNP markers rather than whole genome predictions.

Snelling et al. (2012) identified strategies by which SNP markers can advance our knowledge of factors influencing female reproduction in beef cattle and aid selection of replacement heifers. These authors also suggested that building a test panel with fewer markers associated with genes involved in reproductive processes would be a cost effective approach for the beef industry. Presently, a limited number of studies have found significant SNPs from the BovineSNP50 BeadChip (Illumina, San Diego, CA) affecting female reproduction in beef (Hawken et al. 2012; Saatchi et al. 2014) and dairy (Olsen et al. 2009; Sahana et al. 2011; Wu et al. 2014) cattle. In addition, Snelling et al. (2010) and Buzanskas et al. (2014) found several chromosomal regions and SNP markers associated with birth and weaning weights of beef cattle. Thus, the aim of this study was to identify SNP markers that associate with variation in beef heifer reproduction and performance of their calves.

Materials and methods

Animals

Data from a total of 785 crossbred replacement heifers in their first calving, born between 2004 and 2012 were collated from three research herds in Canada, including 120 from Brandon Research Centre, Brandon, MB, 201 from the University of Alberta Roy Berg Kinsella Ranch, Kinsella, AB and 464 from the Lacombe Research Centre, Lacombe, AB. The foundation breeds for Brandon and Lacombe herds were largely made up of Angus, Charolais and Hereford while at Kinsella, the beef-dairy hybrids were heavily influenced by Angus and Hereford breeds with

infusion of Holstein. Each heifer was weighed at pre-breeding, pre-calving and at pregnancy evaluation. Dam and sire breed group and age along with calf birth and weaning date and weight, calf condition, calving ease and calf sex were recorded.

Genotyping and quality control

Blood samples were collected from each replacement heifer by jugular venipuncture for DNA extraction. Samples were collected into evacuated tubes containing EDTA (Vacutainer, Beckton Dickinson and Co., Franklin Lakes, NJ) and refrigerated at 4 °C until DNA extraction using the QiagenDNeasy 96 blood and tissue kit (Qiagen Sciences, Germantown, MD). Scoring of marker genotypes was performed using BovineSNP50 BeadChip (Illumina, San Diego, CA) and was completed at Delta Genomics, Edmonton, AB, Canada. The SNPs with call rate less than 90%, minor allele frequency (MAF) less than 2%, heterozygous excess greater than 15% and significant ($P < 0.05$) Hardy-Weinberg Equilibrium deviation were removed, resulting in a total of 38,116 SNPs used for analysis. In addition, animals with call rate less than 90% were also removed and only autosomal SNPs with known genome position according to the UMD_3.1 bovine assembly map (Zimin et al. 2009) were used.

Detection and correction for population stratification

To detect and correct for possible population stratification in the data set, a classical multidimensional scaling (MDS) approach was used to derive co-ordinates for each individual from a matrix of dissimilarity based on identity by state (IBS) using PLINK v1.07 (<http://pngu.mgh.harvard.edu/purcell/plink/>) (Purcell et al. 2007). To account for population stratification in the association analysis, animals were put into clusters using pairwise population

concordance test (PPC) offered by PLINK. These clusters were fitted in the mixed model equation for estimating breeding values as a classification factor.

Traits and data analyses

The reproduction traits studied were pre-breeding body weight (PBW) expressed in kg, age at first calving (AFC) expressed in days, pregnancy rate (PR) scored as 1 for pregnant and 0 for not pregnant, calving difficulty (CD) scored as 1 for no assistance and easy pull and 0 for hard pull and caesarian. Although, any assistance is less desirable than heifers calving on their own, easy pull was scored the same as no assistance in this study because the few scores recorded for easy pull were a calving intervention of convenience (it may not have been needed if heifers were given more time to calve). The performance traits included calf birth weight (BWT) expressed in kg, calf weaning weight expressed in kg, and pre-weaning average daily gain (ADG) expressed in kg/d; all of which were treated as traits of the heifer's calf, thereafter. Pedigree records were available and consisted of 8,255 individuals, 553 sires and 3,234 dams.

The estimated breeding values (EBV) were obtained for each trait by means of single trait analysis implemented in the ASreml software (Gilmour et al. 2009) using an animal model that included the additive genetic and residual random effects, fixed effects of contemporary group (herd of origin and birth year), month of birth, population cluster, calf sex, covariates of dam age and weaning age as appropriate for the analysed trait. Random maternal effects were not included for BWT, WWT and ADG as these traits were analysed as a trait of the heifer. The random heifer effects represent the total maternal effects including maternal environment and maternal genetic contribution to the calf's growth potential. Calf sex and BWT were included as fixed effects for CD and AFC while covariate of age at pregnancy determination was included

for PR. Fixed effects were included in the animal model if found to be significantly associated with the analysed trait. All traits were analysed assuming normality and linearity of observation except for PR and CD which were analysed as a binomial distribution using the logistic link function available in ASreml.

Genome-wide association study

Association analysis was carried out by means of the Generalized Quasi-Likelihood Score method (GQLS) developed by Feng et al. (2011) and implemented in the SLEUTH software provided by Dr. Mehdi Sargolzaei (personal communication, 2014). In this method, logistic regression was used to associate the EBVs for each trait treated as covariates with genotypes treated as a response variable. Genotypes in the AB format were coded as 0, 1 and 2 for number of the “B” alleles observed. Analyses were conducted for one SNP at a time. Details of the mathematical theory and assumptions of GQLS can be found in Feng et al. (2011) and Buzanskas et al. (2014). However, this methodology could not estimate SNP effects simultaneously in the analysis. Another step described below has to be carried out to obtain the effects of each significant SNP.

The false discovery rate (FDR) was used to correct for multiple testing (Benjamin and Hochberg 1995). A maximum threshold of 5% FDR was applied to each chromosome using the chromosome-wise correction. A 5% FDR was chosen to eliminate the potential for a false positive or negative test in the identification of significant SNPs. The nominal *P*-value for each significant SNP was also reported.

Estimation of allele substitution and genotypic effects

Single regression analyses was carried out to estimate the allele substitution effects for each significant SNP identified in the GQLS analyses. For each trait, the EBV was regressed against the number of copies of “B” alleles (0, 1, 2) of the significant SNP fitted as a covariate. Similarly, a genotypic model that adjusts for non-genetic fixed effects for each analysed trait plus the genotype of the significant SNPs fitted as a classification factor was applied using the raw phenotypes as the response variable. Subsequently, the least square means for each genotypic class was predicted. All analyses were conducted in R statistical software using default programs where applicable (Ihaka and Gentleman 1996).

Results and discussion

Descriptive statistics, heritability and population stratification

The descriptive statistics and heritability estimates for heifer reproduction traits and performance of their calves are given in Table 1. Crossbred heifers on average, weighed 392 kg at breeding and conceived and calved at 386 and 716 days of age, respectively, with minimal assistance in their first calving. Resulting calves weighed about 38 kg and 240 kg at birth and weaning, respectively, growing at a rate of 0.98 kg/d. The heritability estimates for the studied traits were low to moderate and agreed with previous findings, as described by Cammack et al. (2009).

Figure 1 displays the population structure identified by the first 3 dimensions of a classical MDS. The figure shows that sub-populations exist among the studied heifers from the three research herds. Animals from Kinsella Research Ranch tend to form clusters among themselves (green color in Figure 1) while animals from Lacombe Research Center and Brandon Research Center tend to form clusters according to the proportions of ancestral breeds such as

Angus, Charolais or Hereford present. Further evidence to support this finding is shown in Figure 2 which depicts the proportion of the various breeds in each of the studied animals. Angus breed (red color) was greatly represented in over 98% of the studied population. This is followed by Charolais (green color) and Hereford (blue color). The Kinsella hybrids which appear in orange color are heavily influenced by Angus and Hereford breeds with the known infusion of Holstein. The PPC test result from PLINK generated 97 clusters or groups based on breed ancestry which were used to account for population stratification in the subsequent analysis.

Distribution of significant SNPs associated with studied traits

The number of SNPs evaluated in each *Bos taurus* autosome (BTA) and the distribution of chromosome-wise significant SNPs is shown in Table 2. At the level of 5% FDR, the number of significant SNPs found to be associated with the studied traits were 4 for PBW, 3 for PR, 3 for CD, 9 for AFC, 6 for BWT, 2 for WWT and 1 for ADG. These SNPs were located on chromosomes 1, 5 - 7, 9, 13 - 16, 19 - 21, 24, 25 and 27 - 29. Chromosomes 1, 5 and 24 had significant SNPs with pleiotropic effects. Table 3 shows the result of the GQLS, single regression and genotypic model analyses including the accession number for the significant SNPs, position on the chromosome, allele substitution effects and least square means for the observed genotypes. The allele substitution effects estimated for all the identified SNPs using a single regression model and EBV as dependent variable were highly significant ($P < 0.00001$). In addition, the least square means predicted from raw phenotypes and observed genotypes of significant SNPs are shown.

Pre-breeding body weight (PBW)

The SNPs rs110332219, rs111011302, rs41641007 and rs109853761 located on BTAs 6, 13, 19 and 19, respectively, were significantly ($P < 0.00001$) associated with PBW (Table 3). Allele substitution effects for these SNPs were found to have an impact on PBW of between 3.17 kg and 5.27 kg. As a consequence, heifers having two copies of the positive effect alleles for any of the associated significant SNPs were heavier at breeding than heifers that had two copies of negative effect alleles, thus, these SNPs can be included in a selection program to identify replacement heifers with potential for a higher PBW at breeding, if desired. Previous studies have found a low to moderate and positive genetic correlation between body weight and conception rate in cattle (Bourdon and Brinks 1982; MacNeil et al. 1984; Boligon and Albuquerque 2011; Santana et al. 2012). None of the SNPs identified for PBW were associated with other traits.

Pregnancy rate (PR)

The rate of pregnancy in replacement heifers was found to have low heritability (0.14 ± 0.099), which suggest that environmental factors largely control the expression of this trait. However, the present study identified three significant SNPs on different chromosomes that have significant ($P < 0.00001$) association with PR (Table 3). These SNPs, rs43161706, rs43587346 and rs43724154, showed a significant effect on log odds of EBV for PR. The result of the least square analysis showed that replacement heifers with two copies of positive effect alleles had a greater likelihood of conception (86 to 100%) in their first breeding season compared to their contemporaries that have two copies of the negative effect alleles (58 to 80% chance). Previous studies have identified numerous SNPs that are associated with variation in different pregnancy indicators in beef cattle (Cole et al. 2011; Hawkens et al. 2012; Wu et al. 2014) and mapped

them to several cattle BTAs including those observed in this study. Greater lifetime production has been associated with heifers that conceive early in their first breeding season (Lesmeister et al. 1973), thus, an effort to increase conception rate of young heifers using genomics would be beneficial to the beef cattle industry.

Calving difficulty (CD)

The incidence of calving difficulty is often observed among young heifers with calves having heavier birth weight (Basarab et al. 1993a,b; Sheldon and Dobson 2003). Three SNPs rs43428256, rs41624797 and rs41813617 located on different chromosomes were found to be significantly ($P < 0.00001$) associated with CD in young heifers (Table 3). These SNPs also had a significant impact on log odds of EBV for CD. Young heifers having two copies of the negative effect alleles showed a decrease in the likelihood for normal calving and thus, a likely increase in the incidence of dystocia. The identified SNPs were in the same chromosomal region as previously found SNPs having strong association with direct and maternal calving ease in beef cattle (Saatchi et al. 2014). Olsen et al. (2009) found a strong association of calving ease with SNPs on several chromosomes including BTA5 observed here. Further, SNP rs43428256 identified for CD in this study was on the same chromosome with SNP rs109575263 observed to be significantly associated with BWT of calves. These two SNPs were about 112.9 megabase pairs apart. The presence of a quantitative trait loci (QTLs) affecting BWT and CD suggests that calf BWT must be monitored in order to avoid problems with dystocia in young heifers (Mee 2008; Olsen et al. 2009).

Age at first calving (AFC)

The highest number of significant SNPs identified in this study were associated with AFC (Table 2). Two of these SNPs were on BTA24 and three were on BTA25 and BTA29. The heritability of AFC ranges from 0.01 to 0.27 in beef cattle (Cammack et al. 2009) and is highly genetically correlated with age at subsequent calving as well as calving interval (Gutiérrez et al. 2002). As a consequence, AFC is often used to evaluate heifer fertility. Heifers having two copies of alleles with a negative allele substitution effects for SNPs rs108946497, rs109311252 and rs42164599 were found to calve between 708 – 716 days of age which is 11 days earlier than their contemporaries (718 – 728 days) with two copies of positive effect alleles. Despite short calving intervals, a later AFC is associated with a decrease in lifetime productivity of the beef cows (Gutiérrez et al. 2002).

In addition, SNP rs108946497 which was found to be associated with AFC was located on the same BTA24 with another SNP rs108969215 identified to be associated with BWT. These SNPs are approximately 22.7 kilobase pairs apart. Further, heifers homozygous for the significant SNP alleles associated with early AFC tended to be homozygous for SNP alleles associated with lower BWT. Mujibi and Crews (2009) reported a high and positive genetic correlation between BWT and gestation length in Charolais cattle. Therefore, this finding suggests that first-calf heifers having two copies of negative alleles for SNP rs108946497 are likely to produce calves with lighter BWT and show a lower incidence of dystocia, although, it is likely that AFC is confounded by the gestation length of individual heifers which warrants further investigation.

Birth weight (BWT), weaning weight (WWT) and pre-weaning average daily gain (ADG)

In the model for genetic evaluation of BWT, WWT and ADG, the maternal component is often included to account for maternal genetic effects. This type of model reflects the complex nature of these traits because BWT, WWT and ADG of a calf are not only a function of their inherent growth ability but also a function of the maternal heritability and other environmental factors associated with the dam. As with the direct effects, maternal effects are also passed on from generation to generation and accumulate in a population. In this study, BWT, WWT and ADG were treated as traits of the dam primarily to account for the total maternal effect on the calf and to identify SNPs in the maternal genome that may associate with variation in BWT, WWT and ADG. For BWT, six SNPs including SNPs rs42792088, rs42792101, rs108969215, rs109575263, rs110600672 and rs110786757 were identified to be significantly ($P < 0.00001$) associated with the trait (Table 3). Two of the these SNPs, rs42792088 and rs42792101, were located on BTA27 at approximately 21.0 kilobase pairs apart and are likely to be associated with the same underlying QTL controlling BWT of calves.

The occurrence of SNPs rs109575263 and rs108969215 located on BTA5 and BTA24 with other SNPs associated with CD and AFC, have been described above. Such pleiotropic effects can bring about correlated response in the associated traits. Contrary to the results of this study, Snelling et al. (2010) and Buzanskas et al. (2014) identified SNPs associated with BWT on BTA4, BTA6 and BTA9 based on using the calf's genotype rather than the maternal genotype. However, Snelling et al. (2010) also performed GWAS on dam genotype but significant association with BWT were not observed. In addition, significant associations between SNP and traits are highly dependent on the datasets used (particularly if different breeds and breed combinations are involved). Thus, it is likely that differences are simply a sampling issue.

Two SNPs were found to be significantly associated with WWT and ADG. On BTA1, a pleiotropic effect for SNP rs29016259 was found between WWT and ADG which corroborates the high and positive genetic correlation that is known to exist between the two traits (Kennedy and Henderson 1975). This is very likely because of the relationship between WWT and computation of ADG and suggests that maternal genes that act on WWT also act on ADG. Previously, Snelling et al. (2010) and Buzanskas et al. (2014) observed pleiotropic effects for BWT, WWT and yearling weight of beef cattle in a GWAS. In general, our findings agree with expected results from quantitative genetic studies where high correlation exist between direct and maternal genetic effects for BWT, WWT and ADG, and demonstrate genetic correlations of moderate to high magnitude between reproduction traits and performance of calves (BWT, WWT and ADG).

Conclusion

The Generalized Quasi-Likelihood Score method was used to perform a genome-wide association study on Canadian crossbred replacement heifers. Chromosomal regions were identified to be associated with seven heifer reproduction traits and performance of their calves. New significant SNPs that impact functional traits were identified, many of which have not been previously reported. The results of this study support quantitative genetic studies related to the inheritance of these traits and should contribute to the process of building consensus beef cattle QTL effects. The identification of these SNPs also provides a starting point to identify genes affecting beef heifer reproduction and performance of their calves (BWT, WWT and ADG) for use in marker- and genome-assisted selection and management.

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Table 1. The number of records (n), descriptive statistics and heritability estimates for pre-breeding body weight (PBW), pregnancy rate (PR), calving difficulty (CD), age at first calving (AFC), calf birth weight (BWT), calf weaning weight (WWT) and calf pre-weaning average daily gain (ADG) of crossbred replacement heifers

Trait	n	Mean	Standard-Deviation	Minimum	Maximum	h^2	SE
PBW, kg [†]	728	392.0	42.23	293.0	519.4	0.43	0.119
PR [*]	734	0.82	0.38	0	1	0.14	0.099
CD [*]	543	0.94	0.24	0	1	0.32	0.174
AFC, d	538	716.5	20.79	666.0	791.0	0.13	0.130
BWT, kg	543	37.5	4.87	15.9	56.3	0.43	0.180
WWT, kg	501	239.8	39.64	117.5	362.9	0.24	0.189
ADG, kg/d	501	0.98	0.15	0.40	1.37	0.42	0.194

[†]Average age of heifer at pre-breeding weigh day was 386 days

^{*}Pregnancy rate was analysed as a binary response where 1 is pregnant and 0 not pregnant after the breeding season. Similarly, calving difficulty was classified as 1 for normal calving and easy pull and 0 for hard pull and caesarean.

Note: BWT, WWT and ADG were treated as traits of the heifer

Table 2. Number of SNPs evaluated for each *Bos taurus* autosome (BTA), BTA length in mega base pair (Mbp), and number of chromosome-wise significant SNPs* for pre-breeding body weight (PBW), pregnancy rate (PR), calving difficulty (CD), age at first calving (AFC), calf birth weight (BWT), calf weaning weight (WWT) and calf pre-weaning average daily gain (ADG), respectively

BTA	BTA length (Mbp)	SNPs evaluated	PBW	PR	CD	AFC	BWT	WWT	ADG
1	158.09	2562	0	0	0	0	0	1	1
2	136.51	2068	0	0	0	0	0	0	0
3	121.14	1875	0	0	0	0	0	0	0
4	120.39	1856	0	0	0	0	0	0	0
5	121.08	1579	0	0	1	0	1	0	0
6	119.19	1893	1	0	0	0	0	0	0
7	112.36	1671	0	0	0	1	0	0	0
8	113.01	1811	0	0	0	0	0	0	0
9	105.46	1510	0	1	0	0	0	0	0
10	103.25	1643	0	0	0	0	0	0	0
11	107.18	1631	0	0	0	0	0	0	0
12	90.94	1243	0	0	0	0	0	0	0
13	83.84	1334	1	0	0	0	0	0	0
14	83.15	1355	0	0	1	0	0	0	0
15	84.17	1265	0	0	0	0	0	1	0
16	81.25	1200	0	0	1	0	0	0	0
17	74.89	1177	0	0	0	0	0	0	0
18	65.29	965	0	0	0	0	0	0	0
19	63.47	1000	2	0	0	0	0	0	0
20	71.59	1139	0	1	0	0	0	0	0
21	71.10	1037	0	1	0	0	0	0	0
22	61.22	942	0	0	0	0	0	0	0
23	52.07	790	0	0	0	0	0	0	0
24	62.04	884	0	0	0	2	1	0	0
25	42.71	722	0	0	0	3	0	0	0
26	50.95	797	0	0	0	0	0	0	0
27	45.27	707	0	0	0	0	3	0	0
28	46.15	707	0	0	0	0	1	0	0
29	51.10	753	0	0	0	3	0	0	0
Total	2498.88	38116	4	3	3	9	6	2	1

*Total number of significant SNPs after multiple testing correction at a 5% false discovery rate

Table 3. Identities, positions and effects of the significantly associated SNPs obtained by the Generalised Quasi-Likelihood Score method (GQLS) for pre-breeding body weight (PBW), pregnancy rate (PR), calving difficulty (CD), age at first calving (AFC), calf birth weight (BWT), calf weaning weight (WWT) and calf pre-weaning average daily gain (ADG) in crossbred heifers

Trait	SNP Reference	BTA	Position (bp)	MAF	P-value	FDR	Allele substitution effect [†]	Least square mean of genotypes as fixed effects			
								AA	AB	BB	Sig.
PBW, kg	rs110332219	6	36986502	0.39 (B)	1.95E-07	5	5.27±0.98	384.9±3.09	393.1±2.91	402.9±3.94	***
PBW	rs111011302	13	68688618	0.29 (B)	1.66E-05	5	3.17±1.07	387.4±2.94	393.0±3.01	405.5±4.97	***
PBW	rs41641007	19	26848767	0.21 (B)	5.51E-05	5	-3.92±1.20	395.4±2.80	386.2±3.27	372.7±6.41	***
PBW	rs109853761	19	27529791	0.30 (A)	5.16E-05	5	4.76±1.02	377.8±4.66	390.4±3.02	397.1±3.03	***
PR	rs43587346	9	23069571	0.09 (B)	1.56E-05	5	0.05±0.02	0.80±0.02	0.90±0.03	1.00±0.00 [†]	*
PR	rs43161706	20	54329189	0.17 (B)	4.07E-05	5	-0.06±0.01	0.86±0.01	0.76±0.03	0.62±0.10	**
PR	rs43724154	21	18410412	0.19 (A)	8.65E-06	1	0.08±0.01	0.58±0.09	0.76±0.03	0.86±0.02	***
CD	rs43428256	5	7002633	0.08 (B)	7.83E-06	5	-0.07±0.02	0.96±0.01	0.91±0.03	0.92±0.10	*
CD	rs41624797	14	3956956	0.05 (A)	1.96E-05	5	0.10±0.02	1.00±0.00 [‡]	0.87±0.05	0.96±0.01	**
CD	rs41813617	16	61668331	0.16 (A)	2.47E-06	1	0.06±0.01	0.85±0.08	0.91±0.03	0.98±0.01	*
AFC, d	rs41655238	7	63901946	0.05 (A)	1.23E-05	5	-0.23±0.17	711.1±11.79	723.3±2.43	715.7±0.83	**
AFC	rs108946497	24	32517935	0.23 (B)	3.26E-05	5	-0.49±0.08	718.3±0.99	714.6±1.25	707.8±3.32	**
AFC	rs110549926	24	60745394	0.40 (B)	0.000101	5	0.27±0.07	713.0±1.32	717.7±1.07	719.1±1.81	*
AFC	rs109227898	25	22617891	0.36 (B)	7.74E-05	5	0.25±0.08	714.6±1.24	717.2±1.09	719.0±2.06	**
AFC	rs110912921	25	26468456	0.04 (A)	8.01E-05	5	0.32±0.19	681.6±16.55	709.8±2.64	717.2±0.82	*
AFC	rs109311252	25	27736138	0.15 (A)	0.000147	5	-0.39±0.10	728.2±4.48	717.07±1.58	715.9±0.88	**
AFC	rs43092370	29	13651466	0.37 (A)	0.000112	5	0.36±0.07	714.0±1.87	714.4±1.12	719.9±1.23	**
AFC	rs42164599	29	13695183	0.35 (B)	0.000157	5	-0.37±0.07	719.6±1.21	714.4±1.13	714.0±1.97	***
AFC	rs42095209	29	23903248	0.38 (A)	8.81E-05	5	0.33±0.07	712.2±1.91	714.3±1.12	720.4±1.19	*
BWT, kg	rs109575263	5	119853322	0.15 (B)	2.79E-06	1	-0.42±0.10	38.0±0.24	37.2±0.39	35.1±1.13	***
BWT	rs108969215	24	54893494	0.07 (B)	2.36E-06	1	-0.72±0.14	37.9±0.22	37.1±0.52	29.9±1.92	**
BWT	rs110600672	27	37389551	0.14 (B)	9.00E-05	5	-0.32±0.11	38.1±0.24	36.9±0.38	34.6±1.30	***
BWT	rs42792088	27	45347958	0.48 (A)	4.44E-05	5	0.27±0.07	36.1±0.39	37.8±0.28	38.8±0.35	***
BWT	rs42792101	27	45368987	0.48 (A)	4.44E-05	5	0.27±0.07	36.1±0.39	37.8±0.28	38.8±0.35	**
BWT	rs110786757	28	3710024	0.06 (A)	1.05E-05	1	0.45±0.16	30.8±2.15	37.7±0.59	37.8±0.22	**
WWT, kg	rs29016259	1	24302207	0.34 (B)	7.71E-06	5	0.82±0.25	237.0±1.72	244.3±1.60	244.5±3.25	***
WWT	rs41630261	15	32983903	0.28 (B)	3.55E-05	5	0.92±0.26	238.4±1.56	242.7±1.69	253.6±3.80	***
ADG, kg/d	rs29016259	1	24302207	0.33 (B)	4.29E-06	5	0.01±0.00	0.96±0.01	1.00±0.01	1.04±0.02	***

BTA = *Bos taurus* autosome; bp = base pairs; MAF = Minor allele frequency; FDR = False discovery rate; Sig. = Significance of least square means; * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$; †Coefficient of single locus regression of significant SNPs on estimated breeding value ($P < 0.00001$); ‡ Genotype observed for pregnant cattle only; was not observed; §Genotype observed for no assistance and easy pull only

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Figure 1. Population structure identified by first 3 dimensions of a classical multi-dimensional scaling approach for heifers contributed by the three research centers. Blue for Brandon Research Center, red for Lacombe Research Center and green for Roy Berg Kinsella Research Ranch. See online version for figure in colour

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Figure 2. Distribution of breed proportions in the studied animals. Angus is red, Hereford is blue, Charolais is green and Kinsella hybrid is orange. The Kinsella hybrid is a beef-dairy hybrid heavily influenced by Angus and Hereford breeds with infusion of Holstein. See online version for the color version of the figure

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Population structure identified by first 3 dimensions of a classical multi-dimensional scaling approach for heifers contributed by the three research centers. Blue for Brandon Research Center, red for Lacombe Research Center and green for Roy Berg Kinsella Research Ranch. See online version for figure in colour 317x238mm (96 x 96 DPI)

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