Genome-wide association studies using BayesC and estimation of genetic parameters for perinatal sucking reflex in Brown Swiss calves

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Summary

The intake of a sufficient amount of colostrum during the first few hours *post partum* is essential for new born calves. Brown Swiss calves show an absence of a healthy sucking behavior more frequently than other dairy breeds. In an earlier study low to moderate heritabilities were estimated for sucking reflex in Italian Brown Swiss calves. In our study phenotypes of 9,307 South-German Brown Swiss calves were recorded *post partum* with sucking reflex encoded in 4 categories (no reflex, weak reflex, normal reflex and strong reflex). 50K SNP chip genotypes were collected from 3,072 calves.777K SNP chip genotype data from 192 calves was available and was used for imputation of 50K SNP chip genotypes. Variance components were estimated using sire threshold models and genome-wide association analyses (GWAS) were carried out with high-density genotype data using single-marker models or Bayesian multi-marker models. Low heritabilites were estimated between 0.08 and 0.12, depending on the trait coding. GWAS results of single-marker models showed significant SNPs on BTA 1, 6, 7, 9, 10, 11, 13, 16, 19, 22 and 26, pointing to a polygenic nature of sucking reflex. Possible candidate genes could be found on BTA 6, 19 and 22. Bayesian GWAS results identified a window on BTA 19 explaining 10 % of the genetic variance.

Keywords: Brown Swiss, sucking reflex, genetic parameters, GWAS

Introduction

Brown Swiss calves show difficulties in their sucking behavior shortly after birth more frequently than other dairy breeds (Maltecca *et al*., 2007; Rossoni *et al*., 2005). The absence of a healthy sucking behavior leads to sick and weak calves and in worst cases to an increase in mortality (Hadorn *et al.*, 1997). It also includes a higher workload for the farmer, because more time for drenching each calf with a weak sucking ability is needed. Just *et al.* (2017, submitted) investigated the importance of a healthy sucking reflex for the Brown Swiss breeders in the southwest of Germany and found a high interest in the invention of an effective breeding strategy against the weak sucking reflex in the Brown Swiss calves.

Maltecca *et al.*, (2007) revealed that 10 % of Italian Brown Swiss calves had sucking difficulties. They estimated heritabilities between 0.08 and 0.26, depending on the model and data recording; therefore, breeding against a weak sucking behavior could be successful. The aim of the present study was to estimate genetic parameters and to perform genomewide association analyses for sucking behavior of new-born calves in German Brown Swiss population.

Material and methods

Data sampling

Phenotypes of Brown Swiss calves were recorded between June 2015 and July 2016 on 224 farms in the southwest of Germany including as traits of interest calving process, size of the calf and vigor shortly after birth, duration between first attempt to suck and successful sucking and the target trait was sucking reflex. Only herds with more than 50 calves in the data and at least one calf in every trait level were taken into account for variance component estimation so that 3,850 animals from 52 herds and 176 sires were left in the data set. Bovine 50K SNP chip genotypes were available for 3,072 calves and high-density genotypes (777K) for 192 calves were collected, which were used to impute all genotypes up to 777K. After quality control high-density genotypes for 2,561 calves with 545,287 SNPs were left in the data.

Statistical analyses

Genomic heritabilities were first estimated using reml function in GCTA (Yang *et al*., 2011) and prevalence function for the binary data to transform the estimate of variance from the observed scale to that on the underlying scale. Fixed effects for estimation were sex, herd and season. Variance components were further estimated by sire threshold models in the R-package MCMCglmm (Hadfield, 2010). Explanatory variables included as fixed effects in the model were herd, half-year, gestation length linear and gestation length squared. Sucking reflex was encoded in three different ways (Table 2). The MCMC sampling algorithm was performed with 250,000 iterations with a burn-in period of 10,000 and thinning interval of 100.

Single-marker GWAS was conducted in GCTA (Yang *et al.*, 2011) using the implemented mlma-loco function with pre-correction of the phenotypes for fixed effects sex, herd and season. The significance level was set at p 5 x .

BayesC multi-marker GWAS was conducted in R package BayesDSamples (Wellmann and Bennewitz, 2012). Prior assumptions were based on variance component estimates from GCTA, and degrees of freedom were either 2.5 or 4. The Gibbs sampling algorithm was performed with 50,000 sampling iterations with a burn-in period of 25,000 iterations. Sliding windows were calculated for sizes of 0.25 Mb, 0.5 Mb, 1 Mb and 5 Mb. Every 25th sample was drawn from the posterior distribution to calculate the window posterior probability of association (WPPA) (Fernando and Garrick, 2013).

Results and Discussion

We found an incidence of 7 % non-sucking calves in the examined Brown Swiss population and also 14 % of the calves showing a weak sucking reflex. There was no difference between the sexes.

Table 1: Incidence of sucking reflex for genotype(g) and phenotype(p) data

sucking reflex	male (g)	female (g)	sum (g)	male (p)	female (p)	sum (p)
none	117	303	420	175	198	373

weak	12	455	467	241	270	511
normal	10	1310	1320	1235	1213	2448
strong	10	352	354	251	267	518
sum	141	2420	2561	1902	1948	3850

The high incidence in the population underlines the importance of the examined trait and the problems it implies for the farmers in southern Germany.

Variance components and heritabilities

We estimated heritabilities between 0.07 and 0.12 in the genotype and phenotype data, comparable to other fitness-related traits (Table 2).

Table 2: Encoding for trait definition of sucking reflex, estimated heritabilities () with 95 % confidence interval (l-95% CI, u-95% CI) and estimated genomic heritabilies () with standard errors ()

Model	Encoding of sucking reflex		l-95% CI	u-95% CI	()
Model 1	1 (no), 2 (weak), 3 (normal), 4 (strong)	0.12	0.012	0.261	0.07 (0.02)
Model 2	1 (no), 0 (weak, normal, strong)	0.10	0.015	0.223	0.10 (0.05)
Model 3	1 (no, weak) , 0 (normal, strong)	0.08	0.011	0.178	0.09 (0.04)

GWAS results

Single-marker analyses showed different trait associated SNPs for each tested model (Table 3). Model 1 revealed 24 SNPs with effects above the significance level, located on BTA 1, 6, 7, 13, 19, 22 and 26. When using model 2 and 3 we also found SNPs on BTA 9, 10 and 16.

Table 3: number of significant SNPs (p < for all models

Model	Number of significant SNPs	chromosome
Model 1	24	1, 6, 7, 13, 19, 22 ,26
Model 2	10	1, 6, 9, 16
Model 3	7	10, 19, 22

WPPA results of multi-marker analyses displayed the clearest outputs when using 2.5 degrees of freedom for BayesC sampling and a window size of 1 Mb.

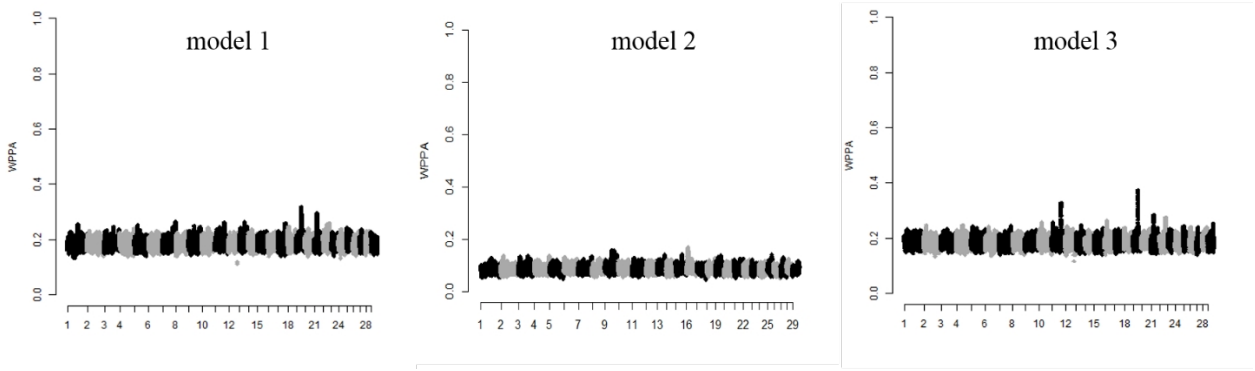

Figure 1: WPPAs for model 1, model 2 and model 3 with window size of 1Mb

As shown in Figure 1 the windows with the largest estimated effects could be found on BTA 19 in model 1 as well as in model 3. Observing model 1 we found a window in the region between 39,191,368 and 41,964,506 bp on BTA 19 which explains 10 % of the genetic variance.

Results of the conducted GWAS clearly indicate the polygenic nature of the sucking reflex. Due to these findings it becomes clear that the breeding process must be interpreted as a long term project and that the invention of genetic tests is not an option.

Conclusion

Results of GWAS identified the polygenic nature of the sucking reflex in Brown Swiss calves. Future breeding strategies cannot rely on the invention of a genetic test to solve the problem of the absent sucking reflex. Instead, genomic selection and consequent recording of phenotype data could provide a solution to reduce the proportion of non-sucking calves in the South-German Brown Swiss breed.

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