Proc. Assoc. Advmt. Anim. Breed. Genet. 19:347-350

USE OF AUTOMATIC FEEDER DATA FOR ACTIVITY STUDIES IN SHEEP

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SUMMARY

Genetic studies are still limited for behavioural traits since most of them are difficult to measure and/or evaluate. Knowledge of the genetic background of behaviour traits may help to improve animal welfare and husbandry, and contributes to a better understanding of changes during domestication. In the present study the genetic background of feeding activity was analysed in an intensively managed sheep population using 209 sheep, kept in pens with automatic feeders. Daily records of frequency and duration of feeder visits, comparing the period in the feeder and the period eating, were summarized and used in a genome-wide association study. Animals were genotyped using the ovine 50k SNP array. After Bonferroni adjustment markers on 16 chromosomes were significantly ($P < 0.05$) associated with duration of frequency of feeder visits. Especially markers on chromosomes 7, 17 and 19 showed association with three or four different time measures. This study is the first genome-wide association study using time measures from automatic feeder experiments in sheep. Future studies are needed to verify these findings and analyse the data by comparison of animals showing similar patterns of feeding activity.

INTRODUCTION

Little is known about the extent to which differences in feeding behaviour affect feed intake in sheep. The development of models for the study of feeding-related behaviour in sheep might help to overcome productivity restrains due to a mismatch of supplementing feed and nutritional needs of the animals. Electronic feeding stations are commonly used in pig production systems to measure feed intake and growth and strategies to reduce measurement errors associated with such systems were shown previously (Bruininx *et al*. 2001, Zumbach *et al.* 2010). It has yet to be proven if the application of a robust regression procedure as shown in Zumbach et al. (2010) will also fail to exclude abnormal growth curves in sheep. Furthermore, inaccurate data resulting from malfunctions of the feeder were also the basis for data excluding in the study of Bruininx *et al*. (2001), who applied simple exclusion levels to eliminate such data. If similar elimination strategies can be applied to time measures from automatic feeders, this could provide an alternative basis for behaviour using data from automatic feeder. Time measurements of automatic feeders will provide information of time spend in feeders and time eating, furthermore the frequency of feeder visits could provide information about the flock structure. In such a way automatic feeder systems will allow a precise measurement of individual behavioural/activity characteristics in a cost effective way compared with traditional methods such as observation and video analysis (Hyun and Ellis 2001; Desnoyers *et al*. 2009).

This study examines time measurements from automatic feeders to investigate the activity of sheep under feedlot conditions. Genetic regions associated with duration of eating and stay in the feeder as well as frequency of feeder visits were compared to test if these measurements show similar patterns of genetic association.

MATERIALS AND METHODS

A total of 209 Awassi-Merino wethers from three different cohorts/experiments were kept for 12 to 18 week feeding periods in a feedlot with 10 automatic feeders. The feedlot was located at the University Sydney research farm 'Mayfarm' at Camden, New South Wales, Australia. Animals

aged between 1 and 3 years were part of an Awassi-Merino gene-mapping population (Raadsma *et al.* 2009). Animals were fed *ad libitum* using a commercial paddock lamb finisher (Weston Animal Nutrition Company) and had unrestricted access to low quality straw roughage. The automatic feeders recorded date and time the animals entered the feeder, duration of eating and length of stay in the feeder as well as body weight via electronic ear tags.

After initial exclusion of data points associated with erroneous recording such as negative body weights, feed intakes and time measures and non existing electronic tags, further data restriction were body weight >190 kg (> 5 SD of mean observed body weights), time measures > 1 hour in the feeder and corresponding feed intake records were deleted, since it was found that such observations showed an overlap with consecutive feeder visits (date and time of the next sheep entering the feeder). Only a small amount of the raw data (2.8 %) was deleted during this procedure. A total of 6,500, 5,822 and 5,516 record were then available from the three experiments. Further analyses were performed for all animals together. Analyses and data restrictions were performed using *R* (version 2.12.0) (R team).

After the editing of the data, three time measures, time in the feeder, time in the feeder and eating and time in the feeder without eating, were derived for each feeder visit, as well as the total time per day for each activity. The computerised feeder-data also allowed a recording of the frequency of feeder visits for each animal, which in future studies may be used as a possible indicator of the flock hierarchy.

Animals used for the experiment were genotyped using the ovine 50kb SNP array. Markers/ genotypes not within the quality control requirements (minor allele frequency >5%, call rate >95%, inheritance of paternal/ maternal alleles) were excluded. A genome-wide association test was performed to identify association between the time measures, frequency of feeder visits and the genetic markers across all chromosomes using *PLINK* (Purcell *et al.* 2007). Associations tested using *PLINK* were deemed as signification if exceeded $P < 0.01$ before and $P < 0.05$ after Bonferroni single-step adjustment.

RESULTS AND DISCUSSION

After initial elimination of extreme values and data with obvious data logging errors, a total of 17,838 observations were finally used for this study. Each animal entered any of the feeder stations between 1 and 432 times (average 71.2) per day. The average duration of each feeder visit was longer (0.49 minutes) compared to the duration of time during which feed was consumed (0.17 minutes), consequently animals spent almost double the time in the feeder without eating (0.32 minutes) (Table 1).

Table 1. Overview of the average (mean), standard deviation (SD), minimum (min) and maximum (max) duration (in minutes) of each feeder visit and of all feeder visits per day

Trait	mean	SD	min	max
Time eating per day	10.09	4.19	0.03	74
Time eating per feeder visit	0.17	0.07	0.03	0.72
Time in the feeder per day	31.25	16.49	0.05	178
Time in the feeder per feeder visit	0.49	0.23	0.05	2.9
Time in the feeder without eating per day	21.15	14.16	0	149
Time in the feeder without eating per feeder visit	0.32	0.19	0	2.6

The average total time the animals spend in the feeder was 31.25 minutes per day but showed a very large degree of variation (CV 52%). We are not aware of any comparable results in other experiments using automatic feeders, but we expect the variation of the different time measures, if

interpreted at the level of the animal, may reflect the social structure of the flock. But it needs to be validated against observational records if animals higher in the rank tend to occupy the feeder for longer periods, or if animals lower in the flock hierarchy try to hide in the feeder or utilise it at times when the majority of the flock is not eating and sub-ordinate animal get access to the station.

The genome-wide association (GWAS) revealed overlapping regions of statistical significance in the genome between the activity measures (frequency of feeder visit, time eating per day and time eating per feeder visit, time in the feeder without eating per day and time in the feeder without eating per feeder visit) (Figure 1).

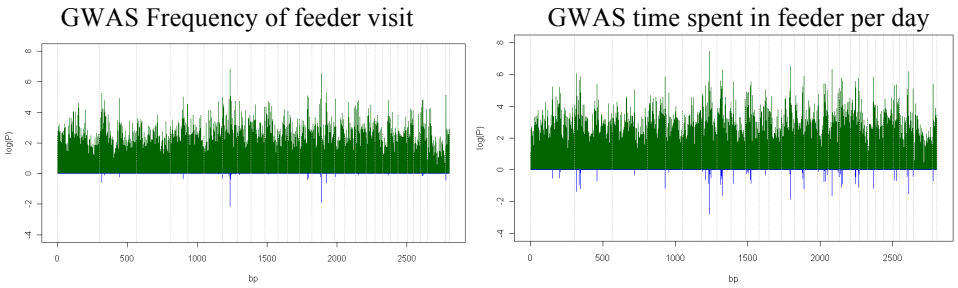

Figure 1. Results of the genome-wide association study (GWAS) for frequency of feeder visits (left graph) and time spend in the feeder per day (right graph). Shown are the $\log_{10}$ transformed *P*-values for unadjusted (green) and Bonferroni adjusted (blue) results, grey lines indicate the chromosomes 1 to X and unassigned SNPs.

Significant ($P < 0.01$) SNP associations were identified on all chromosomes for all traits except time in the feeder per feeder visit. The results were further adjusted (Bonferroni) and we found that none of the markers was significantly ($P < 0.05$) associated with the time in the feeder per feeder visit. Among the other feeding activity traits, between 1 and 11 markers were significantly associated (Table 2). Most of the associations were identified on chromosomes 7, 17 and 19.

Table 2. Overview of the regions showing significant ($P \leq 0.05$) association (Bonferroni adjusted) with the feeding activity traits

	Chromosome																									
Trait	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	X
Time eating /visit	0	0	0	3	0	0	1	0	0	0	0	0	0	0	0	1	1	0	1	0	0	0	0	0	0	0	1
Time eating /day	0	0	0	0	0	0	0	1	0	1	0	0	0	1	0	0	0	0	2	0	0	0	0	0	0	0	0
Visit frequency	0	0	0	0	0	0	1	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0
Time in feeder /day	0	1	0	0	0	0	1	1	0	0	0	0	1	0	0	0	1	0	0	0	0	0	0	0	0	1	0
Time in feeder /visit	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Time no cons* /day	1	0	0	1	0	0	3	0	0	1	0	0	1	0	0	0	1	1	1	0	0	0	0	1	0	0	0
Time no cons* /visit	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Significant traits	1	1	0	2	0	0	4	2	0	2	1	0	2	2	0	1	3	1	3	0	0	0	0	1	0	1	1

*time in the feeder without eating

Among the few studies aiming to detect genetic associations with behaviour in livestock, three genome scans in cattle revealed quantitative trait loci (QTL) for temperament on 21 different chromosomes (Hiendleder *et al*. 2003, Schmutz *et al*. 2001, Gutierrez-Gil *et al*. 2008). The type 4

dopamine receptor (*DRD4*), a gene already well recognized as a candidate for behaviour, mapped to the bovine chromosome 29. Other candidate genes identified in studies using cattle included cannabinoid receptor (*CNR1*) on bovine chromosome 9, and *DRD2* on chromosome 15. *CNR1* is possibly a positional candidate gene in our study, since we identified significant associated markers for the feeding activity on ovine chromosome 8, which is a comparative chromosome to bovine chromosome 9. However, we could neither identify significant associated markers on ovine chromosome 15 or 21, which are comparative to the bovine chromosome 15 and 29. However results before Bonferroni correction did show some significant associations on these chromosomes, we might need to change the adjustment of the results to a less stringent correction or increase the power of the study by inclusion of more animals to detect these smaller effects. Further studies are now required to unravel the genetic architecture of complex traits associated with feeding activity and behaviour in sheep.

CONCLUSIONS

To our knowledge this is the first genome-wide-association study in sheep for feeding behaviour. We have demonstrated the possibility of using data from automatic feeders to analyse the feeding activity of sheep. Using data obtained from automatic feeders for behavioural studies in livestock is advantageous in terms of time and cost compared with manual observations and other electronic equipment used for behaviour measurements. This study also demonstrates the feasibility of using data derived from automatic feeder experiments to undertake genome-wide association studies for feeding activity.

REFERENCES

Bruininx E.M., van der Peet-Schwering C.M., Schrama J.W., den Hartog L.A., Everts H. and Beynen A.C. (2001) *J. Anim. Physiol. Anim. Nutr.* **85**:81.

Desnoyers M., Bechet G., Duvaux-Ponter C., Morand-Fehr P., and Giger-Reverdin S. (2009) *Small Rum. Res.* **83**: 58-63.

Gutierrez-Gil B., Ball N., Burton D., Haskell M., Williams J. L. and Wiener P. (2008) *J. Hered.* **99**(6):629-638.

Hiendleder S., Thomson H., Reinsch N., Bennewitz J., Leyhe-Horn B., Looft C., Xu N., Medjugorac I., Russ I., Kuehn C., Brockmann G. A., Bluemel J., Brening B., Reinhardt F., Reents R., Averdunk G., Schwerin M., Foerster M., Kalm E. and Erhardt G. (2003): *J. Hered.* **94**(6):496-506.

Hyun Y. and Ellis M. (2001) *J. Anim. Sci.* **79**: 803-810.

Purcell S., Neale B., Todd-Brown K., Thomas L., Ferreira M.A.R., Bender D., Maller J., Sklar P., de Bakker P.I.W., Daly M.J. and Sham P.C. (2007) *Am. J. Hum. Genet.* **81**.

R Development Core Team (2008) R Foundation for Statistical Computing V, Austria http://www.R-project.org/.

Raadsma H.W., Thomson P.C., Zenger K.R., Lam M.K., Jonas E., Cavanagh C., Jones M., Attard G., Palmer D. and Nicholas F.W. (2009) *Genet. Sel. Evol.* **41**.

Schmutz S.M., Stookey J. M., Winkelman-Sim D. C., Waltz C. S., Plante Y. and Buchanan F. C. (2001): *J. Hered*. **92**:290.

Zumbach B., Misztal I., Chen C.Y., Tsuruta S., Łukaszewicz M., Herring W.O., Culbertson M. (2010) *J. Anim. Breed. Genet*. **127**:93-9.