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ASSESSING GENETIC VARIABILITY IN FLYSTRIKE RESISTANCE ACROSS EXPRESSION LEVELS IN AUSTRALIAN MERINO SHEEP

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SUMMARY

This work aimed to provide guidance to breeders on the level of flystrike expression required to achieve effective selection and to develop genomic reference data for future prediction. Genetic parameters and breeding values were compared across different expression thresholds (1% to 10%) using data sourced from 21 research and industry breeder flocks. Among the thresholds examined (1% to 10%), a 2.5% threshold was adequate for excluding data with no genetic variation for breech strike while retaining sufficient data for analysis. Previously, a 5% threshold was considered effective for capturing data with adequate variation, primarily from research flocks with minimal flystrike prevention practices. Lowering this threshold to 2.5% enables greater breeder participation by making it more achievable to obtain information for selection purposes. As flystrike resistance has proven to be heritable, breeding for genetic resistance offers a sustainable, long-term solution, helping to reduce flystrike. Additionally, this change supports a broader breeding approach to reduce flystrike, potentially minimising reliance on chemical treatments and management practices.

INTRODUCTION

The Australian sheep industry faces significant financial and animal welfare challenges due to flystrike in sheep, a common and debilitating infection (Shephard *et al.* 2022). Research has demonstrated flystrike resistance is heritable, making genetic resistance a promising and sustainable solution for long-term control through breeding programs (Smith *et al.* 2009; Dehnavi *et al.* 2023). Establishing a well-designed sheep reference population is essential for developing genomically enhanced Australian Sheep Breeding Values (ASBVs; Brown *et al.* 2010), particularly given the varying incidence rates of flystrike across different environments (Bird-Gardiner *et al.* 2013; Greeff *et al.* 2014; Smith *et al.* 2009). This study aims to estimate genetic parameters using data collected from a range of Australian Merino sheep populations, including research and seedstock flocks, with documented cases of flystrike across varying expression levels. The findings aim to guide breeders on the adequate level of flystrike expression required to facilitate the effective selection and development of a reference population. The Flystrike Genomics Reference Flock project (funded by Australian Wool Innovation) is a three-year project aimed at increasing the phenotypic and genomic information needed to establish a flystrike ASBV.

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MATERIALS AND METHODS

Data. The dataset comprised 39,998 breech strike (BRS) records (1/0) from young sheep (up to yearling age), including 20,806 ewes and 19,192 wethers and rams, alongside 26,136 genotypes with 61,998 markers on these animals. This dataset was collected from 21 research and industry seedstock flocks. The pedigree consisted of 79,484 animals from 6,805 sires and 36,597 dams. Data filtering was applied across a range of expression thresholds ranging from 1% to 10% per flock, year of birth and sex (Table 1). A minimum of 20 animals per contemporary group (defined as flock, year of birth and sex) were included. At each expression threshold, the data was partitioned into two subsets: The *Excluded* subset, consisting of records below the threshold level due to low incidence rates within each of the flock-year of birth-sex groups, and the *Included* subset, containing records where the incidence rate was equal to or greater than the threshold levels.

Statistical Analyses. Analyses were conducted using single-step REML in BLUPF90 software (Misztal *et al.* 2014). Initial bivariate analyses of subsets (*Excluded* and *Included*) revealed non-significant genetic correlations, particularly at lower thresholds (data not shown). Consequently, separate univariate analyses were conducted for each of the *Excluded* and *Included* subsets at each threshold level of expression. BRS records (1/0) were standardised (mean=0, SD=1) for strike incidence rate within the flock, year of birth, and sex to account for heterogeneous variance (Dehnavi *et al.* 2024b). Linear mixed models included contemporary groups (flock, year of birth, and sex) and the birth-rear type interaction as fixed effects, with the animal's direct genetic effect fitted as a random effect. Variance components estimated from single-step REML analysis were used to compute breeding values.

Genotype data were incorporated using the H matrix in single-step genomic best linear unbiased prediction (Misztal *et al.* 2014). Heritability estimates were obtained for each subset across the different threshold levels. Because of the non-significant genetic correlation between *Excluded* and *Included* subsets (data not shown), Pearson correlations were calculated to compare research breeding values (RBVs) of common sires and their progeny between the two subsets, contrasting RBVs derived from the *Excluded* data with those obtained from the *Included* subset (Figure 1).

Table 1. Summary of the number of sires (Nsire) and their progeny count (Nprog) categorised by expression threshold (Exc: *Excluded*, Inc: *Included*). The proportion of sires in common across subsets and their respective progeny counts (Link%) are provided in parentheses

Count\ Threshold	1%	2%	2.5%	3%	4%	5%	8%	10%
NsireExc	348	523	597	653	718	756	825	880
(Link%)	(32%)	(43%)	(46%)	(43%)	(34%)	(28%)	(27%)	(27%)
NsireInc	760	698	676	625	520	454	392	351
(Link%)	(15%)	(32%)	(41%)	(45%)	(46%)	(47%)	(56%)	(67%)
NprogExc	4,358	4,898	6,064	4,256	4,615	4,333	5,009	5,084
(Link%)	(39%)	(35%)	(39%)	(23%)	(21%)	(18%)	(18%)	(16%)
NprogInc	4,093	6,925	7,949	6,342	6,345	5,270	5,503	4,725
(Link%)	(15%)	(28%)	(34%)	(31%)	(38%)	(37%)	(53%)	(60%)

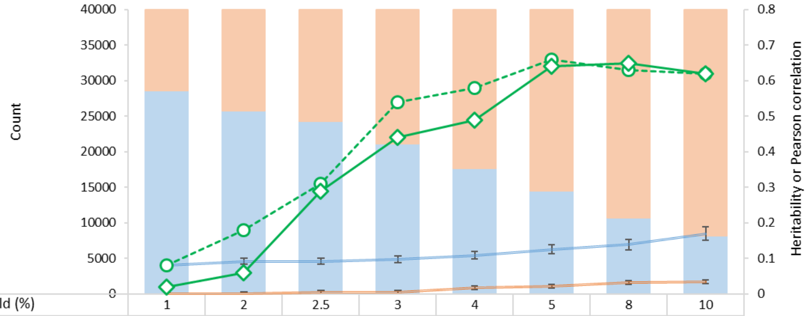
RESULTS AND DISCUSSION

As the threshold increased from 1% to 10%, a significant reduction in the number of records available for the *Included* subset was observed. The number of sires in the *Excluded* subset (NsireExc) rises steadily, peaking at 880 sires at the 10% threshold. Conversely, the number of sires in the *Included* subset (NsireInc) decreases with increasing threshold levels, while their linkage percentage exhibits an inverse pattern, rising from 15% linkage at the 1% threshold to 67% linkage at the 10% threshold. These patterns highlight the trade-off between data quantity and the genetic linkage captured between the two subsets (Table 1).

Figure 1 illustrates the number of records in the *Excluded* (Exc) and *Included* (Inc) subsets across various expression thresholds, along with relevant heritability estimates (h^2). The heritability for the entire unfiltered dataset was low at 0.05 ± 0.01. However, as the thresholds increased, heritability estimates for the high-incidence subset (*Included*; Inc) improved significantly, reaching a maximum of 0.17 ± 0.02 at the 10% threshold. This suggests that applying higher expression thresholds enhances heritability but reduces the number of animals in the analysis.

These heritability estimates are lower than those reported in previous studies, such as 0.32 ± 0.11 (Smith *et al.* 2009) and 0.57 ± 0.13 (Greeff *et al.* 2014) for breech strike using different models and data transformation. However, Greeff *et al.* (2021) later reported a heritability of 0.07 ± 0.01 using a linear model on logarithm-transformed breech strike counts. These differences may stem from variations in trait definition, modelling approaches, scaling and environmental and management factors influencing trait expression. Notably, none of the earlier studies applied thresholds on expression levels.

Higher incidence rates are often associated with increased heritability on the liability scale when running a threshold model (Dahlqwist *et al.* 2019). This aligns with the observed trend in this study, where applying higher expression thresholds led to greater heritability estimates. This occurs because a greater proportion of individuals exceeding the expression threshold leads to higher observable genetic variance. However, this relationship is complex, as heritability estimates are influenced by both the proportion of individuals exceeding the threshold and the underlying genetic variance, which may not increase linearly with incidence (Dahlqwist *et al.* 2019).



Threshold (%)	1	2	2.5	3	4	5	8	10
🟧 N (Exc)	11,403	14,184	15,681	18,837	22,350	25,519	29,263	31,845
🟦 N (Inc)	28,595	25,814	24,317	21,161	17,648	14,479	10,735	8,153
— h2 (Exc)	0.00	0.00	0.00	0.01	0.02	0.02	0.03	0.03
— h2 (Inc)	0.08	0.09	0.09	0.10	0.11	0.13	0.14	0.17
--○-- RBVs' cor common prog	0.08	0.18	0.31	0.54	0.58	0.66	0.63	0.62
—◇— RBVs' cor common sire	0.02	0.06	0.29	0.44	0.49	0.64	0.65	0.62

Figure 1. Number of records (orange and blue bars) and heritability (double orange and blue lines) in *Excluded* (Exc) and *Included* (Inc) subsets. The subsets represent different levels of expression thresholds (1%, 2%, 2.5%, 3%, 4%, 5%, 8%, and 10%). The Pearson correlation was assessed between the RBVs of common sires and their progeny (RBVs' corr common prog or sire) derived from *Excluded* data with those derived from *Included* data (solid and dotted green lines)

Figure 1 also highlights the comparison of breeding values of common sires and their progeny between the two subsets (solid and dotted green lines). A comparison was conducted between the RBVs of common sires and their respective progeny across the two (*Excluded* and *Included*) subsets. At lower thresholds, the weaker pedigree linkage between the subsets, and the low heritability resulted in more diverse RBV predictions, as indicated by the lower Pearson correlation (solid and dotted green lines in Figure 1). Increasing the threshold levels strengthened the pedigree linkage, improving the Pearson correlation of RBVs for common individuals between the two subsets.

Although heritability estimates remain adequate within the *Included* subset (Inc; 0.08 to 0.09), the low linkage and reduced correlation between RBVs of common sires at these thresholds (1% to 2%) highlight the importance of incorporating high-incidence records. While a threshold of 5% appears reasonable (Dehnavi *et al.* 2024a), lowering the threshold to 2.5% is a viable compromise to enable a great level of participation. Below this level, heritability and genetic variance are minimal (with phenotypic variance approaching 1), indicating that excluding the low-incidence subset (*Excluded*) would preserve genetic information and allow for accurate selection decisions based on genetic merit. This adjustment balances the number of excluded records, prediction accuracy, and genetic variation while encouraging breeders to actively use breeding values for selection for flystrike resistance.

CONCLUSION

Identifying the most appropriate threshold for flystrike expression remains an important consideration to ensure the most accurate breeding values are provided to breeders and enable future genomic selection. The selected threshold impacts both heritability estimates and the correlations of predicted breeding values significantly. Higher thresholds improve both heritability estimates and the accuracy of breeding value predictions in sheep selection programs. However, to encourage breeders to participate effectively in direct selection for flystrike, a threshold of 2.5% appears optimal, as it excludes records with minimal genetic variation while maintaining selection efficiency for more breeders. To ensure broader applicability, this approach will be evaluated further for body strike in young sheep and records of both breech and body flystrike across later ages.

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REFERENCES

Bird-Gardiner T.L., Brown D.J., Smith J. and Mortimer S.I. (2013) *Proc. Assoc. Advmnt. Anim. Breed. Genet.* **20**: 183.

Brown D.J., Swan A.A. and Gill J.S. (2010) *Anim. Prod. Sci.* **50**: 1060.

Dahlqwist E., Magnusson, P.K.E., Pawitan Y. and Sjölander, A (2019) *Hum. Genet.* **138**: 425.

Dehnavi E., Swan A.A. and Brown D.J. (2024a) 'Milestone report' AWI ON-00775.

Dehnavi E., Swan A.A., Burbidge G. and Brown D.J. (2024b) *Proc. Aus. Assoc. Anim. Sci.* **35:** 70.

Dehnavi E., Swan A.A., Ramsay A.M.M., Bird-Gardiner T.L. and Brown D.J. (2023) *Proc. Assoc. Advmnt. Anim. Breed. Genet.* **25**: 354.

Greeff J.C., Karlsson L.J.E. and Schlink A.C. (2014) *Anim. Prod. Sci.* **54**: 125.

Greeff J.C., Schlink A.C. and Karlsson L.J.E. (2021) *Anim. Prod. Sci.* **61**:1893.

Misztal I., Tsuruta S., Lourenco D.A.L., Aguilar I., Legarra A. and Vitezica Z. (2014) 'Manual for BLUPF90 family of programs'.

Shephard, R., Webb Ware, J., Bloomfield B. and Niethe, G. (2022) Final report of project B.AHE.0327, Meat and Livestock Australia Ltd.

Smith J.L., Brewer H.G. and Dyall T. (2009) *Proc. Assoc. Advmnt. Anim. Breed. Genet.* **18**: 334.