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IS THERE A RELATIONSHIP BETWEEN TEMPERAMENT AND INTERNAL PARASITE RESISTANCE IN MERINOS?

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SUMMARY

It has been suggested that Merino sheep with flighty temperament are more susceptible to internal parasite infection. This hypothesis was further tested in this study using data from the Sheep Genomics Falkiner Memorial Field Station Flock. Genotypic and phenotypic parameters and heritabilities for temperament, measured as flight time and agitation score, and internal parasite resistance, measured as worm egg counts from *Haemonchus contortus* and *Trichostrongylus colubriformis* challenges were estimated. Heritabilities for the traits examined were moderate with the exception of flight time, which was low (h^2_{flight} = 0.07 ± 0.04). The heritability of agitation score was estimated at 0.21 ± 0.05. Worm egg count heritabilities ranged from 0.13 to 0.30, and were lowest in the *T. colubriformis* challenge. Genetic correlations between worm egg counts and agitation score were generally moderately negative (r_g = -0.16 to -0.21); the exception was the first count from the *T. colubriformis* challenge. Genetic correlations of worm egg counts with flight time were lower than with agitation score, the exception was Twec2 which was higher (r_g = -0.30). All genetic correlations were associated with high standard errors. Our results suggest that animals with faster flight times and/or low agitation score may have higher WEC scores following a nematode challenge. Further research is needed to validate the existence of such as relationship.

INTRODUCTION

It is well documented that stress, in particular chronic stress, can influence the immune response in humans. It has also been demonstrated that more disturbed and anxious individuals exhibit delayed, weaker or shorter immune responses resulting in increased susceptibility to disease (Glaser and Kiecolt-Glaser 2005). Some evidence exists that temperament and susceptibility to internal nematodes are related in sheep; animals selected for resistance to nematodes recorded lower agitation readings than a control group in an isolation box test (Radzikowska *et al.* 1999). Infection of Merino ewes with *Haemonchus contortus* (*H. contortus*) has also been found to alter subsequent performance in an Arena test (Fell *et al.* 1991). Both these studies only explored phenotypic aspects of the relationship.

The aim of this study was to determine both phenotypic and genetic correlations between temperament and internal parasite resistance traits in Merino sheep. It was hypothesised that animals with low flight times and/or high agitation scores will stress more easily and thus have higher worm egg counts (WEC).

MATERIALS AND METHODS

Experimental Design. Data was collected on approximately 2500 Merino lambs, born in 2005 and 2006 in the Sheep Genomics Falkiner Memorial Field Station (FMFS) Flock at Deniliquin, NSW. Animals were from 11 sire groups. Temperament data was recorded after weaning at approximately 4 months of age as flight time and agitation score. Flight time was measured as the time taken for an animal to travel 1.7m upon release from a weigh crate after a confinement period of 15 seconds. Animals were also confined in an isolation box for 30 seconds. The box was raised off the ground and a meter was positioned underneath the box. The number of movements and vocalisations within the box were recorded by a purpose-built meter over this period to give an agitation score (Lee 2006). WEC scores were obtained from challenges with approximately 20,000 *Trichostrongylus colubriformis* larvae (*T. columbriformis*) and 8,000 *H. contortus* larvae at approximately 5 and 7 months of age respectively. Animals were drenched prior to infection with larvae. Count 1 occurred approximately 4 weeks after infection date and count 2, 5 weeks after infection date. Birth date, dam ID, birth type and rearing type were known only for lambs born in 2006.

Statistical Analysis. Genotypic and phenotypic parameters and heritabilities were estimated using ASReml software (Gilmour *et al.* 2002). Dam pedigree was unknown and 50% of animals did not have dam ID recorded. Therefore a genomic relationship matrix (GRM), based on 48,263 SNP was used instead of a pedigree based relationship matrix. SNP with minor allele frequencies of less than 0.01 were not included when computing the GRM. SNP information was known for 1892 animals. These animals were used in the analysis. The number of records retained for analysis varied between traits and are summarised in Table 1. Animals which did not have sire, year of birth or sex recorded were removed from the data. Animals with weaning weights below 10kg were also removed from the data. WEC scores were cube root transformed and agitation score was square root transformed. Transformed WEC scores were used to calculate mean WEC score. The following fixed effects were tested and fitted if significant ($P<0.05$): year, sex of animal nested within year, technician measuring WEC nested within year, sampling group and weaning weight.

Table 1. Number of records retained for univariate analysis and the minimum number of records retained for bivariate analysis

No. records	Hwec1	Hwec2	Hwec mean	Twec1	Twec2	Twec mean	Agit	Flight
Univariate	1547	1499	1677	1678	1733	1692	1473	1620
Bivariate	1473	1473	1408	1495	1484	1408	1507	1507

* Hwec1, Hwec2 = Worm egg count (WEC) 1 & 2 from *H. contortus* challenge, Hwec mean = Mean WEC from Haemonchus challenge. Twec1, Twec2 & Twec mean = measurements from *T. colubriformis* challenge, Agit = agitation score from isolation box test, Flight = flight time

RESULTS AND DISCUSSION

The traits explored in this study are summarised in Table 2. In both challenges, mean WEC score was higher at the second collection date, though not significantly so. Mean flight time was 0.78 seconds with a standard deviation of 0.28 seconds. Mean agitation score was 7.02 with standard deviation of 1.91.

Table 2. Mean, standard deviation, genetic (Va) and phenotypic (Vp) variances*

	Hwec1	Hwec2	Hwec mean	Twec1	Twec2	Twec mean	Agit	Flight
Mean	16.76	18.52	17.55	11.59	12.32	11.95	7.02	0.78
	(7.61)	(7.14)	(6.93)	(3.17)	(3.38)	(2.98)	(1.91)	(0.28)
Va	11.60	11.86	9.89	1.16	1.14	0.93	0.69	0.01
Vp	39.75	39.02	33.27	7.20	8.75	6.25	3.34	0.07

*where data were transformed for analysis, reported values are on transformed data

WEC scores from the *H. contortus* challenge were moderately heritable (h^2_{Hwec1}= 0.29 ± 0.06 and h^2_{Hwec2}= 0.30 ± 0.07; Table 3.). WEC scores from the *T. colubriformis* challenge had lower heritabilities than the *H. contortus* challenge (h^2_{Twec1}= 0.16 ± 0.05, h^2_{Twec2}= 0.13 ± 0.05). Agitation score was moderately heritable (h^2_{agit} = 0.21 ± 0.05), whilst flight time had a low heritability (h^2_{flight} = 0.07 ± 0.04). This is similar to the findings of Lennon *et al.* (2009) who reported heritabilities of 0.20 ± 0.05 for agitation score and 0.12 ± 0.05 for flight time.

All genetic correlations were associated with high standard errors and can thus only be interpreted as an indication of the existence of a relationship between internal parasite resistance and temperament. In this study, *H. contortus* WEC counts were negatively genetically correlated with agitation score ($r_{g\ Hwec1}$= -0.21 ± 0.17, $r_{g\ Hwec2}$= -0.16 ± 0.18). Thus, animals with lower agitation scores may have high *H. contortus* WEC scores which is evidence against the hypothesis and the results found by Radzikowska *et al.* (1997). This seems somewhat counterintuitive, however uncertainty remains as to what aspect of temperament is being measured in the isolation box test. *T. colubriformis* WEC count 2 was also negatively genetically correlated with agitation score (r_g = -0.18 ± 0.22). This differs from Blache & Ferguson (2005) who found a positive correlation between post-weaning faecal egg count and agitation score (r_g = 0.22 ± 0.10). However in that study, WEC scores were from mixed species natural challenge, and the analysis included data on progeny from maternal and terminal as well as Merino sires.

Table 3. Estimates of heritability, genetic and phenotypic correlations*

	Hwec1	Hwec2	Hwec mean	Twec1	Twec2	Twec mean	Agit	Flight
Hwec1	**0.29**	0.75	0.94	0.06			-0.07	0.04
	(0.06)	(0.01)	(0.00)	(0.03)	X	X	(0.03)	(0.03)
Hwec2	0.99	**0.30**	0.93		0.10		-0.06	0.03
	(0.02)	**(0.07)**	(0.00)	X	(0.03)	X	(0.03)	(0.03)
Hwec	1.00	1.00	**0.30**			0.09	-0.06	0.03
mean	(0.01)	(0.01)	**(0.07)**	X	X	(0.03)	(0.03)	(0.03)
Twec1	0.62			**0.16**	0.59	0.88	-0.04	0.00
	(0.19)	X	X	**(0.05)**	(0.02)	(0.01)	(0.03)	(0.03)
Twec2		0.73		0.96	**0.13**	0.91	-0.05	0.00
	X	(0.18)	X	(0.06)	**(0.05)**	(0.00)	(0.03)	(0.03)
Twec			0.63	0.99	0.99	**0.15**	-0.06	0.00
mean	X	X	(0.20)	(0.02)	(0.02)	**(0.05)**	(0.03)	(0.03)
Agit	-0.21	-0.16	-0.21	-0.05	-0.18	-0.11	**0.21**	0.01
	(0.17)	(0.18)	(0.18)	(0.22)	(0.22)	(0.22)	**(0.05)**	(0.02)
Flight	-0.10	-0.13	-0.12	-0.02	-0.30	-0.18	0.20	**0.07**
	(0.26)	(0.27)	(0.27)	(0.31)	(0.28)	(0.30)	(0.26)	**(0.04)**

* Heritability in bold on the diagonal, phenotypic correlations displayed above the diagonal and genetic correlations below the diagonal. Standard errors given in brackets. X indicates untested correlation.

In this study, no genetic correlation was found between Twec1 and agitation score or flight time ($r_{g\ agit}$ = -0.05 ± 0.22 and $r_{g\ flight}$ = -0.02 ± 0.31) . The highest correlation was between Twec2 and flight time (r_g = -0.30 ± 0.28). This might suggest that later measurements of WEC in *T. columbriformis* challenges are a more useful indication of the animal's ability to mount an immune response following the challenge. Moderate negative genetic correlations were found between flight time and all other WEC scores. This suggests that more flighty animals may have poorer immune responses leading to reduced resistance to internal parasites. Earlier studies support the existence of a relationship; animals selected for resistance to nematodes recorded lower agitation scores in an isolation box test (Radzikowska *et al.* 1999). In a different study an association between infection with nematodes and performance in an Arena test has been found (Fell *et al.* 1991).

The genetic correlation between agitation score and flight time was positive ($r_g = 0.20 \pm 0.26$), which disagrees with Lennon *et al.* (2009) who reported a negative correlation. However, FMFS animals were all temperament tested shortly after weaning whereas Lennon *et al.* (2009) conducted temperament tests on mature ewes. A study by Blache and Ferguson (2005) also reported a negative genetic correlation between the two traits. Although weaners were tested in their study, age at measurement varied and not all tests were conducted before the animals reached 12 months of age. We found the phenotypic correlation between these two temperament tests to be close to zero, which agrees with the value of 0.04 found by Blache and Ferguson (2005). We found generally phenotypic correlations between traits measured to be close to zero with the exception of measurements within the *H. contortus* challenge and measurements within the *T. colubriformis* challenge which were high.

CONCLUSIONS

Our findings indicate a genetic relationship exists between temperament and internal parasite resistance but the estimates were associated high standard errors. The correlations reported here indicate that animals with faster flight time and lower agitation score may have compromised immune responses resulting in higher WEC scores following a nematode challenge. Further research is needed to validate the existence of a relationship between temperament and internal parasite resistance. Measuring individual animal WEC scores is time consuming and can be cost prohibitive. Should further work confirm the existence of a relationship between internal parasite resistance and temperament, traits such as flight time may become available as another easily measurable and affordable indicator of internal parasite resistance.

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