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118. Divergent genetic selections for social attractiveness or tolerance toward humans in sheep

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Abstract

Including behavioural traits in genetic selection could be an advantageous strategy for improving adaptation and welfare of farm animals to extensive farming systems in an agroecological perspective. The aim of the present study was to investigate in sheep the efficiency of an early selection for social attractiveness or tolerance to humans. Four divergent lines were created according to the reactivity of lambs to social separation or to human presence. Lambs were individually exposed just after weaning to two behavioural tests. The arena test aims at assessing the motivation of the lambs towards conspecifics and the corridor test aims at assessing the reactivity of the lambs to an approaching shepherd. Several behaviours were measured including vocalizations and flight distance from a human used as selection criteria. After two generations of selection, the divergent selection led to highly significant line differences for social attractiveness or tolerance to humans, respectively 1.8 and 1.0 genetic standard deviation between high and low lines. Such divergent selections also affected additional behaviours expressed in the tests.

Introduction

Genetic selection including behavioural traits could be an advantageous strategy for improving robustness and welfare of farm animals in various farming conditions by minimizing unsuitable responses to changes in their social and physical environment, limiting an excessive fear of humans and improving sociability (Mignon-Grasteau *et al.*, 2005). Farm animals are social and gregarious, and relational behaviours are essential for ensuring social cohesion, social facilitation, offspring survival and docility toward humans. Breed differences and genetic variation within breed have been reported in sheep for early social behaviours and found to be heritable, and associated with some QTL, suggesting such behaviours could be selected (Boissy *et al.*, 2005; Beausoleil *et al.*, 2012; Hazard *et al.*, 2014; Cloete *et al.*, 2020). In addition, such early social reactivity of lambs was identified as a robust trait (Hazard *et al.*, 2016). However, up to now, any study has demonstrated the feasibility, efficiency and effects of genetic selection for early reactivity to social separation or towards a human in domestic sheep. Thus, the aim of the present study was to investigate in sheep the efficiency of an early genetic selection for social attractiveness or tolerance to humans using experimental divergent lines.

Materials & methods

Animals and management. The experimental animals were Romane meat sheep, reared at the INRAE experimental farm of La Fage (Saint-Jean Saint-Paul, Causse du Larzac, France) exclusively outdoors under harsh environmental conditions. The flock comprised about 250 reproductive females reared on 280 ha of rangelands. The farming system and management characteristics have been described previously by Gonzalez *et al.* (Gonzalez-Garcia *et al.*, 2014). Ewes lambed for the first time at 2 years of age and all the lambs were born outdoor in the spring.

Behavioural tests and responses. Each year during 7 years, in average 300 female and male lambs were individually exposed just after weaning to two behavioural tests. The arena test (AT) consisted of two successive phases evaluating: (1) reactivity to social isolation (AT1); (2) the motivation of the lamb towards conspecifics in presence of a motionless human (AT2). The arena test pen consisted in an unfamiliar enclosure virtually divided into 7 zones as described in detail by Ligout *et al.* (Ligout *et al.*, 2011). The corridor test (CT) consisted of two successive phases evaluating: (1) reactivity to social isolation (CT1) and; (2) reactivity to an approaching human (CT2). The test pen consisted in a closed, wide rectangular circuit and has been described in detail by Boissy *et al.* (Boissy *et al.*, 2005). Several behaviours were measured: vocalizations (i.e. frequency of high-pitched bleats), locomotion (i.e. number of virtual zones crossed), vigilance posture (i.e. time spent by the animal motionless), the proximity score (i.e. weighting of time spent in virtual zones, a high score indicated a high duration spent close to conspecifics and a human). The mean flight distance (DIST) separating the human and the lamb and the time during which the human saw the lamb (SEEN) were measured in CT2.

Statistical handling. Two synthetic variables were constructed using the first component of each principal component analysis (PCA) (PRINCOMP procedure in the SAS® software). A PCA was performed on vocalizations across the different phases of behavioural tests (AT1, AT2 and CT1) and the resulting synthetic variable (HBLEAT) was used as indicator of the reactivity to social isolation. A PCA was performed on DIST and SEEN variables and the resulting synthetic variable (HUMAPPRO) was used as indicator of the reactivity to an approaching human. For divergent selection, individual estimated breeding values (EBV) were calculated for HBLEAT and HUMAPPRO with univariate analyses under an animal mixed model using the REML methodology implemented in the ASREML software (Gilmour *et al.*, 2009). In order to constitute the divergent lines, extreme animals were chosen as sires (in average 10 sires per generation and line) or dams (in average 70 dams per generation and line) at each generation according to their high or low EBV for HBLEAT or HUMAPPRO while intermediary for HUMAPPRO or HBLEAT, respectively. Selection intensities were in average 10 and 45% for sires and dams, respectively. Two generations of selected animals were produced (in average 270 female and male lambs per generation and line). Linear models (proc GLM, SAS Institute Inc.) were applied to test whether the divergent selections on behavioural traits had a significant effect on the recorded traits considering the adequate fixed effects in the analyses. The linear model used was $y_{ijklm} = \mu + \text{line}_i + \text{sex}_j + \text{lsr}_k + \text{age}_l + \text{year}_m + e_{ijklm}$; where y_{ijklm} was the behavioural observations of lambs; μ was the overall mean of the population; line, sex, lsr, age and year were the fixed effects of the behavioural line i , the sex j of the lambs, the litter size born and reared k , age l of the dam, and year m of the experiment, respectively.

Results & discussion

For gregarious species such as sheep, vocalization and locomotion behaviours in response to social separation are interpreted as an active way to re-establish social links (Boissy *et al.*, 2005; Beausoleil *et al.*, 2012). The divergence between low (S-) and high (S+) lines selected on social attractiveness reached 1.8 genetic standard deviation after two generations of selection (Figure 1). The divergence between low (H-) and high (H+) lines selected on tolerance toward humans reached 1.0 genetic standard deviation after two generations of selection. Responses to selection obtained were consistent with those expected considering the heritabilities previously estimated for both traits ($h^2=0.5\pm0.06$ for social attractiveness and 0.15 ± 0.04 for tolerance toward humans) and the intensity of selection applied. As expected, the divergent lines for tolerance toward humans were intermediary for social attractiveness compared to S- and S+ lines even if a slight difference was observed between H+ and H- lines for the social criterion (Figure 1). A high difference was observed between S- and S+ lines for tolerance toward humans while no difference was expected between these lines for this criterion. Such difference could be linked to the low positive genetic correlation previously reported between criteria for social attractiveness and tolerance toward humans

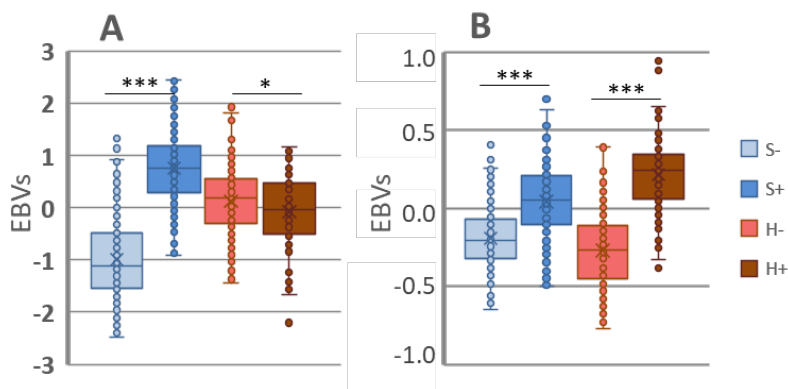


Figure 1. Estimated breeding values for social attractiveness (A) and tolerance toward humans (B) in the four divergent lines after two generations of selection. S- (n=279) and S+ (n=314) lines, low and high social attractiveness, respectively; H- (n=233) and H+ (n=149) lines, low and high tolerance toward humans, respectively. Pairwise comparison between Least Squares Means for line effect: *, and *** for P-value<0.05 and 0.001, respectively.

($r_g=0.25\pm 0.11$) (Hazard *et al.*, 2016). This difference may also be due to the selection criterion that involved measurement of reactivity to social separation in presence of a human (i.e. vocalizations in AT2). Thus, conflict behaviour between the social motivation toward conspecifics and avoidance of a motionless human included in the selection criterion may thus have resulted in differences between S- and S+ lines for both social attractiveness and tolerance toward humans.

Effects of divergent selections on the different behaviours expressed in response to social separation with or without human presence in arena and corridor tests were also investigated (Table 1). Selection for social attractiveness resulted in a two-fold higher expression of vocalizations in response to social isolation without or with human presence. This selection for social attractiveness also resulted in a higher locomotor activity in response to visual isolation from conspecifics (i.e. AT1 phase) as well as a higher proximity score with conspecifics in presence of a human (i.e. AT2 phase). Thus, selection for social attractiveness based on vocalizations not only affected vocalization behaviour but also locomotor activity in a context

Table 1. Behaviours in the four divergent lines after two generations of selection.

Behaviours	Test	S- ¹	S+	Line ²	H-	H+	Line
Vocalizations	AT1	8.0	16.2	***	13.6	11.9	***
Vocalisations	AT2	3.0	7.6	***	5.7	5.0	NS
Locomotion	AT1	23.3	27.2	***	26.5	24.0	*
Locomotion	AT2	6.6	7.2	NS	7.2	4.9	**
Vigilance	AT1	18.6	18.2	NS	17.7	19.3	NS
Proximity	AT2	23.9	27.9	*	25.0	29.1	*
Flight distance	CT2	5.5	5.3	*	5.6	5.1	***

¹ S- and S+ lines, low and high reactivity to social separation, respectively; H- and H+ lines, low and high tolerance toward a human, respectively. AT1, arena test 1 (visual social isolation); AT2, arena test 2 (social isolation in presence of a human); CT2, corridor test 2 (reactivity to an approaching human); Vocalizations: frequency; Locomotion: number; Vigilance: duration in sec; Proximity: duration in sec; Flight distance: meter.

² Line effect: *, ** and *** for P-value<0.05, 0.01 and 0.001, respectively.

specific way. In one hand, more social lambs (S+) increased vocalizations and locomotion behaviours in response to visual separation from conspecifics (i.e. AT1 phase) probably to search actively for them and re-establish social links. On the other hand, when lambs were separated from conspecifics by a grid barrier and a human (i.e. AT2 phase), S+ lambs showed higher levels of vocalizations and proximity while they expressed quite similar level of locomotion compared with S- lambs. This suggested that locomotor activity of S+ lambs was more frequently observed in virtual zones close to conspecifics and the human in the second phase of arena test. As expected, the flight distance of lambs from an approaching human decreased with the selection for tolerance toward humans (Table 1). This selection also resulted in a slight decrease in vocalization and locomotion behaviours in response to social isolation as well as an increase in the proximity score toward conspecifics in presence of a human. We hypothesized that genetic selection for lambs more tolerant toward humans also reduced the overall reactivity to social separation and exposure to humans, may be by reducing overall stress responses.

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