Journal of
Animal Breeding and Genetics

J. Anim. Breed. Genet. ISSN 0931-2668

ORIGINAL ARTICLE

# Genetic control of the environmental variance for birth weight in seven generations of a divergent selection experiment in mice

N. Formoso-Rafferty[1], I. Cervantes[1], N. Ibáñez-Escriche[2] & J.P. Gutiérrez[1]

1 Departamento de Producción Animal, Facultad de Veterinaria, UCM, Madrid, Spain
2 Genètica i Millora Animal – Centre IRTA_Lleida, Lleida, Spain

**Keywords**
Birth weight; canalization; environmental variability; mice; selection experiment.

**Correspondence**
J.P. Gutiérrez, Department of Animal Production, Faculty of Veterinary, UCM, Avda, Puerta del Hierro s/n, E-28040 Madrid, Spain.
Tel:/Fax: +34913943767;
E-mail: gutgar@vet.ucm.es

Received: 1 April 2015;
accepted: 1 June 2015

## Summary

Data from seven generations of a divergent selection experiment designed for environmental variability of birth weight were analysed to estimate genetic parameters and to explore signs of selection response. A total of 10 783 birth weight records from 638 females and 1127 litters in combination with 10 007 pedigree records were used. Each record of birth weight was assigned to the mother of the pup in a heteroscedastic model, and after seven generations of selection, evidence of success in the selection process was shown. A Bayesian analysis showed that success of the selection process started from the first generation for birth weight and from the second generation for its environmental variability. Genetic parameters were estimated across generations. However, only from the third generation onwards were the records useful to consider the results to be reliable. The results showed a consistent positive and low genetic correlation between the birth weight trait and its environmental variability, which could allow an independent selection process. This study has demonstrated that the genetic control of the birth weight environmental variability is possible in mice. Nevertheless, before the results are applied directly in farm animals, it would be worth confirming any other implications on other important traits, such as robustness, longevity and welfare.

## Introduction

The aim of the genetic selection in animal breeding has traditionally been to increase (or decrease) the mean of the productive traits. Selection for robustness, that is less sensitivity to environmental effects as indicated by a low variation, benefits the welfare of animals (Mormede & Terenina 2012), which is one of the main targets of selection. Published studies show that by reducing the variability, this could lead to reduced mortality in pigs (Högberg & Rydhmer 2000) and increased animal welfare (Damgaard *et al.* 2003). Furthermore, homogeneous animal production would decrease the cost of handling and the production of animals that should result in better profits for the farm (Bolet *et al.* 2007). Hence, the growing interest in the genetic control of environmental variability. On the other hand, birth weight is also a very important trait in some livestock species like rabbit (Bodin *et al.* 2010) or pig (Berard *et al.* 2008) in which birth weight heterogeneity within the litter causes competition and reduces survivability (Damgaard *et al.* 2003; Garreau *et al.* 2008). The possibility of selection against environmental variability is called canalization or stabilizing selection and was addressed a long time ago (Waddington 1942). A genetic background affecting the variability of a trait, which is different from that controlling the trait mean, would enable a genetic selection on the variability of a trait by reducing it and reach homogeneity of the trait (Scheiner & Lyman 1991). One way to achieve this could be using the GSEVM software (Ibáñez-Escriche *et al.* 2010) that fits the model developed by SanCristobal-Gaudy *et al.* (1998), which can simultaneously determine the

doi:10.1111/jbg.12174

genetic parameters for the mean and for the environmental variability and their correspondent breeding values.

Although several authors have added to the knowledge on canalization in several species (SanCristobal *et al.* 2001; Sorensen & Waagepetersen 2003; Ros *et al.* 2004; Ibáñez-Escriche *et al.* 2008b), there are few properly designed selection experiments for environmental variability (Gutiérrez *et al.* 2006; Ibáñez-Escriche *et al.* 2008a). Pun *et al.* (2013) designed a genetic divergent selection experiment in mouse to assay by selecting the environmental variability of the birth weight. However, this experiment failed, because as the authors argued, the trait was attributed to the individual when it should have been assigned to the mother (Pun *et al.* 2013). Moreover, they also identified some anomalous results such as an extreme genetic correlation between the birth trait and its environmental variability, or a too high value for the additive genetic variance of the environmental variability as first warned by Hill & Mulder (2010). Hill & Mulder (2010) reported that typically, the heritability of the environmental variance was under 10%, and its genetic coefficient of variation (CV) was typically 20% or more. According to them, the parameter estimates obtained by Pun *et al.* (2013) would largely be out of the range found for all the heritability estimates and all the coefficients of variation found in the literature.

Consequently, a new divergent selection experiment was designed to check the conclusions reached by Pun *et al.* (2013) that environmental variability of birth weight in mice must be selected via the dam.

Hence, the aim of this paper was to verify whether selection for environmental variability of birth weight could be successful while selecting the genetics of environmental variability attributed to the mother instead of the individual. Genetic parameters and breeding values were estimated using a model including random dam and litter effects. Selection response was assessed by estimating genetic and phenotypic trends of birth weight and its variability in the first seven generations of divergent selection for environmental variability.

## Materials and methods

### Experimental population

The origin of the experiment population was the same as that used for other selection experiments (Fernández and Toro 1999; Gutiérrez *et al.* 2006; Ibáñez-Escriche *et al.* 2008a; Moreno *et al.* 2012; Pun *et al.* 2013). It was started from a created mouse population originating from a balanced genetic contribution of three inbred mice lines: BALB/c, C57BL and CBA. The three-way crossed population was maintained in panmixia for 40 generations ensuring high levels of both genetic and phenotypic variability.

A divergent selection experiment was designed and performed. The selection criterion of the animals was the predicted breeding value for birth weight environmental variability (PBVv) associated with their mother. Thus, each dam had multiple records. Pregnant females were checked every 24 h during the birth period, and the newborns were weighed and individually identified within the first 24 h after birth.

From the panmictic population described above, a total of 64 males and females were randomly selected to be mated, one male to one female having two litters to evaluate the mothers for the birth weight environmental variability of their offspring.

Pup weight of pups and their environmental variability were considered as maternal traits, and a genetic evaluation was then carried out (some details of the genetic evaluation are included below). An initial selection was carried out to set up the low and high environmental variability lines (abbreviated as low and high lines across the paper) as follows. First, dams were ranked according to their PBVv and then, animals to be mated were selected among their offspring. Thus, to establish the lines, four males and four females were sequentially selected, while available, among the offspring of the best (lowest or highest PBVv), respectively, for the low and the high lines) dams, to complete 43 males and 43 females to be mated within each line. A restriction of no sharing grandparents was imposed on animals to be mated. The resulting mating design was called the standard solution. Instead, an improved solution was thereafter defined by implementing a weighted selection. To achieve this, first the mean coancestry of the 86 selected animals of the standard solution was computed and a simulated annealing algorithm (Fernández & Toro 1999) was used to reach an optimal solution. Then, the improved mating design was established by maximizing the mean genetic breeding value of the progeny by not restricting the size of the offspring to be selected from each mating. Mean coancestry of the improved solution was restricted to be equal or lower than the mean coancestry obtained in the standard solution defined above. The mating design from the improved solution was actually performed. This process was identically followed within lines from the second generation onwards, and all the processes were repeated for seven additional generations. A scheme of the experimental design is shown in Figure 1.

The selection intensities for each generation and line are shown in Table 1. The final selection intensity was computed by $i = ((\bar{x}_s - \bar{x}_c)/\sigma_c)$, where $\sigma_c$ is the standard deviation (SD) of the predicted breeding values (PBVvs) of the candidates for selection, and $\bar{x}_s$ and $\bar{x}_c$ are the mean PBVv of the selected and candidate animals, respectively. The theoretical selection intensity for the generation establishing the lines was 1.49, corresponding to approximately 17% of proportion selected (the best 10 of 60 mates). The theoretical selection intensity across lines was taken to select a proportion of approximately 25% (selection intensity of 1.27), selecting the best 10 of 40 mates within each line. Taking into account the empirical fertility, 64 (in the founder generation) and 43 (the rest of generations) were set as the number of females to be mated to finally account, respectively, with approximately 60 and 40 available mates giving births. The actual selection intensity was not achieved due to several factors such as reproductive performance, sex ratio within litter, fraternal mating restriction, PBVvs distribution and efficacy of weighed selection.

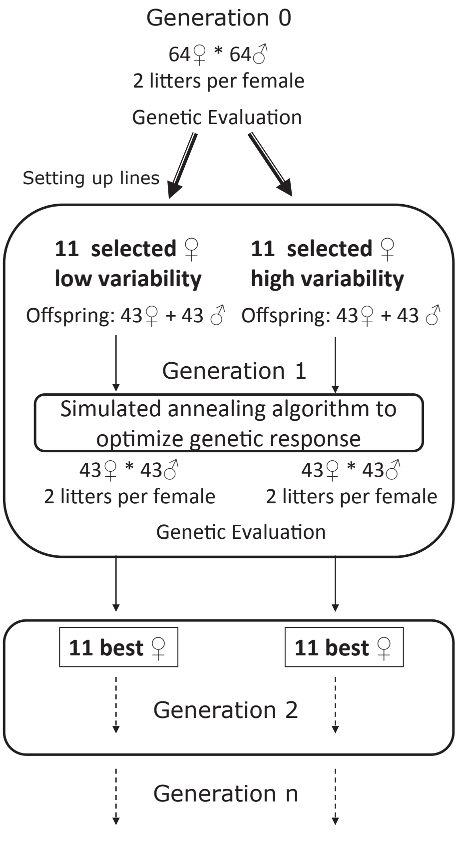

**Figure 1** Scheme of the experiment.

**Table 1** Selection intensity (*i*) and equivalent proportion selected (%) in both high and low lines in all generations selected for environmental birth weight variability

| | High | | Low | |
|---|---|---|---|---|
| Generation | *i* | % | *i* | % |
| Initial | 1.9308 | 7 | −1.3699 | 21 |
| 1 | 1.2446 | 26 | −1.1892 | 29 |
| 2 | 1.1596 | 30 | −1.2447 | 26 |
| 3 | 1.2774 | 25 | −1.3255 | 23 |
| 4 | 1.2280 | 27 | −1.2012 | 28 |
| 5 | 1.2351 | 27 | −1.0773 | 34 |
| 6 | 1.0930 | 34 | −1.2530 | 26 |
| 7 | 1.0792 | 34 | −0.8907 | 44 |

The data of individual birth weight (BW) obtained from all the litters and the whole pedigree that included five generations of the panmictic population were used to evaluate the selected progenitors. Table 2 shows the number of females and litters within line cumulated and per generation along the experiment, and the number of performance and pedigree records, mothers and litters cumulated and per generation. The final evaluation data set contained a total of 10 783 records of BW from 1127 litters of 638 females. The mean ± SD for the litter size (newborns) was 8.84 ± 2.81 and for the BW (g) 1.57 ± 0.21. The total number of individuals included in the analysed pedigree was 10 007.

### Genetic evaluation

The heteroscedastic model, developed by SanCristobal-Gaudy *et al.* (1998), was used for this study. It is assumed in the model that the environmental variance is heterogeneous and partially under genetic control:

$$y_i = \mathbf{x}_i\mathbf{b} + \mathbf{z}_i\mathbf{a} + \mathbf{w}_i\mathbf{c} + e^{1/2(\mathbf{x}_i\mathbf{b}^* + \mathbf{z}_i\mathbf{a}^* + \mathbf{w}_i\mathbf{c}^*)}\varepsilon_i,$$

where $y_i$ is the BW of the $i$ individual; * indicates the parameters associated with environmental variance; **b** and **b*** are the vectors of the systematic effects; **a** and **a*** are the vectors of the direct additive genetic effect of the mother; and **c** and **c*** are the vectors of the litter effect; and $\mathbf{x}_i$, $\mathbf{z}_i$ and $\mathbf{w}_i$ are the incidence vectors for systematic, additive genetic and litter effects, respectively. And finally, $\varepsilon_i \sim N(0,1)$. It must be noted that as defined, the direct genetic effects **a** and **a*** are maternal effects that also include half of the direct genetic effect of the pup. The **c** and **c*** vectors are strictly fitting the litter effect unlike models in which

**Table 2** Number of females and litters with offspring in low and high environmental variability lines, BW records, pedigree records, females and litters available for the analyses across generations and cumulated

| | Initial | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
|---|---|---|---|---|---|---|---|---|
| Females in low line | 62 (Initial) | 42 | 40 | 41 | 42 | 43 | 42 | 40 |
| Females in high line | | 41 | 39 | 43 | 42 | 39 | 40 | 42 |
| Litters in low line | 123 (Initial) | 75 | 69 | 68 | 73 | 73 | 73 | 71 |
| Litters in high line | | 72 | 64 | 73 | 72 | 76 | 73 | 73 |
| Records | 1256 | 1348 | 1266 | 1404 | 1387 | 1344 | 1417 | 1361 |
| Pedigree | 586 | 1256 | 1348 | 1266 | 1404 | 1387 | 1345 | 1415 |
| Females | 62 | 83 | 79 | 84 | 84 | 82 | 82 | 82 |
| Litters | 123 | 147 | 133 | 141 | 145 | 149 | 146 | 143 |
| Cumulated records | 1256 | 2604 | 3870 | 5274 | 6661 | 8005 | 9422 | 10 783 |
| Cumulated pedigree | 586 | 1842 | 3190 | 4456 | 5860 | 7247 | 8592 | 10 007 |
| Cumulated females | 62 | 145 | 224 | 308 | 392 | 474 | 556 | 638 |
| Cumulated litters | 123 | 270 | 403 | 544 | 689 | 838 | 984 | 1127 |

the trait is attributed to the pup, where the litter effects are assumed to be also fitting most of the maternal effect as observed by Ibáñez-Escriche *et al.* (2008a). Both direct and maternal genetic effects were not simultaneously fitted given that there is no software available to solve such a complex heterocedastic model.

The genetic effects **a** and **a*** are distributed together and are assumed to be Gaussian:

$$\begin{pmatrix}\mathbf{a}\\ \mathbf{a}^*\end{pmatrix} \sim N\left(\begin{bmatrix}\mathbf{0}\\ \mathbf{0}\end{bmatrix}, \begin{bmatrix}\sigma_{\mathrm{a}}^2 & \rho\sigma_{\mathrm{a}}\sigma_{\mathrm{a}^*}\\ \rho\sigma_{\mathrm{a}}\sigma_{\mathrm{a}^*} & \sigma_{\mathrm{a}^*}^2\end{bmatrix}\otimes \boldsymbol{A}\right),$$

where $\boldsymbol{A}$ is the additive genetic relationship matrix; $\sigma_{\mathrm{a}}^2$ is the additive genetic variance of the trait; $\sigma_{\mathrm{a}^*}^2$ is the additive genetic variance affecting the environmental variance of the trait; $\rho$ is the coefficient of genetic correlation; and $\otimes$ denotes the Kronecker product.

The vectors **c** and **c*** are also assumed to be independent, with $\mathbf{c} \sim N(\mathbf{0}, \boldsymbol{I_c}\sigma_{\mathrm{c}}^2)$ and $\mathbf{c}^* \sim N(\mathbf{0}, \boldsymbol{I_c}\sigma_{\mathrm{c}^*}^2)$, where $\boldsymbol{I_c}$ is the identity matrix of equal order to the number of litters, and $\sigma_{\mathrm{c}}^2$ and $\sigma_{\mathrm{c}^*}^2$ are the litter effect variances affecting, respectively, the BW mean and its environmental variability (Ibáñez-Escriche *et al.* 2008a). Covariance between litter effects affecting the trait and its variability was not fitted mainly because there is no software available to solve such a complex model.

The model applied included generation (eight levels), litter size (from 2 to 17, 16 levels), sex (male, female, unknown) and parity number (two levels) as systematic effects in **b** and **b***, and the litter (1127 levels) and additive genetic effect (10 007 levels) as random effects besides the residual effect. For each generation, the genetic parameters were estimated and the genetic evaluation was carried out using the accumulated available information at that point. The model was solved using the GSEVM program (Ibáñez-Escriche *et al.* 2010).

### Selection response

Genetic trends of BW trait and its environmental variability were analysed by averaging the predicted breeding values for the trait (PBVs) and for its environmental variability (PBVvs) within line and generation and plotting them against generation.

To obtain descriptive statistics for the phenotypic trends, BW and other within-litter traits such as mean birth weight (MBW), birth weight variance (VAR), birth weight SD and birth weight CV were recorded and analysed under the following model:

$$y_i = \mathbf{x}_i\mathbf{b} + e_i,$$

in which $y_i$ is either BW, MBW, VAR, SD or CV. Firstly, a model including line*generation (15 levels), litter size (16 levels) and, in the case of BW, also sex (three levels) as systematic effects in **b** was fitted (Model Ve). The same model but with only line*generation as systematic effect was fitted to study how correction for other systematic effects can impact the final estimates (model Vr). Marginal posterior distributions of the difference between solutions for different lines but same generation were drawn to infer probabilities of response. The model was solved using the TM program (Legarra 2008) slightly modified to fit this model.

## Results

### Genetic parameter estimates

The mean and SDs of the marginal posterior distributions for the estimated genetic parameters across generations are shown in Table 3. These estimates were performed sequentially for each generation as the data were recorded using all the available information at that point. The parameters became stable only after the

**Table 3** Mean and standard deviation (in brackets) of the marginal posterior distribution for the BW genetic parameters estimated initially and across generations 1–7

| | Initial | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
|---|---|---|---|---|---|---|---|---|
| $\sigma_a^2$ | 0.0569 (0.0118) | 0.0323 (0.0048) | 0.0252 (0.0033) | 0.0059 (0.0015) | 0.0057 (0.0013) | 0.0055 (0.0011) | 0.0052 (0.0010) | 0.0052 (0.0009) |
| $\sigma_c^2$ | 0.0398 (0.0646) | 0.0252 (0.0028) | 0.0210 (0.0019) | 0.0132 (0.0014) | 0.0125 (0.0011) | 0.0120 (0.0010) | 0.0110 (0.0008) | 0.0103 (0.0007) |
| $\sigma_{a^*}^2$ | 0.1902 (0.0691) | 0.1872 (0.0604) | 0.1405 (0.0361) | 0.0621 (0.0288) | 0.0577 (0.0211) | 0.0508 (0.0179) | 0.0480 (0.0157) | 0.0566 (0.0160) |
| $\sigma_{c^*}^2$ | 0.3044 (0.0774) | 0.3386 (0.0630) | 0.2745 (0.0451) | 0.2947 (0.0414) | 0.2754 (0.0334) | 0.2760 (0.0300) | 0.2825 (0.0278) | 0.2634 (0.0251) |
| $\rho_{a,a^*}$ | 0.7283 (0.2647) | 0.4643 (0.2887) | 0.1482 (0.2958) | 0.2181 (0.2265) | 0.2371 (0.2027) | 0.2422 (0.1872) | 0.3181 (0.1766) | 0.2607 (0.1518) |

$\sigma_a^2$ and $\sigma_{a^*}^2$ are the additive genetic variance affecting, respectively, the BW mean and its variation; $\sigma_c^2$ and $\sigma_{c^*}^2$ are the litter effect variances affecting, respectively, the BW mean and its variation; and $\rho_{a,a^*}$ is the coefficient of genetic correlation.

third generation as a consequence of the limited information. The final estimates for the last generation show more importance to the litter effect (0.0103 for BW and 0.2634 for its variability) than the additive genetic effect (0.0052 for BW and 0.0566 for its variability), both on the mean trait (twofold) and its variability (four- or fivefold). Also, dispersion of the marginal posterior distributions was much lower for the parameters concerning the mean of trait than those corresponding to its environmental variability, particularly as generations increased. Regarding variability-genetic parameters, and according to the expression developed by Hill & Mulder (2010), heritability of the variability resulted in 0.008 and genetic CV was 0.22. These parameters are within the range of estimates given in the review by Hill & Mulder (2010).

### Genetic trends

Evolution of mean breeding values of PBVs and PBVvs within line and generation are shown in Figure 2. A fairly linear divergent evolution appears for PBVvs as the logical consequence of the selection was carried out with this criterion. Also, a divergent genetic evolution emerged as the consequence of the estimated positive genetic correlation between BW and its environmental variability (0.26) which implied an expected correlated response in the mean of the trait. However, the observed trend for BW appears asymmetric and remained roughly stable in the low line.

### Phenotypic trends

Raw evolution of BW, VAR, SD and CV was inspected by simply averaging each of these traits within line and generation, and plotting the averages against generation (Figure 3). Divergence was observed from the beginning of the experiment even though it was almost imperceptible for VAR and SD, as opposed to CV, when setting up the lines. There was a great divergence in the following generation and keeping the divergence across generations up to the last one in which a new strong divergence emerged. The mean

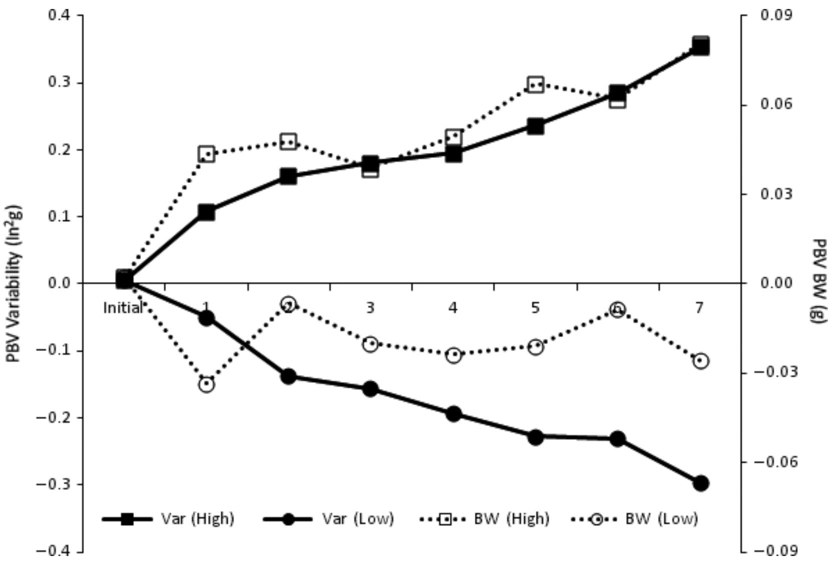

**Figure 2** Mean predicted breeding values for variability of birth weight (PBV*) and for BW (PBV) across generations.

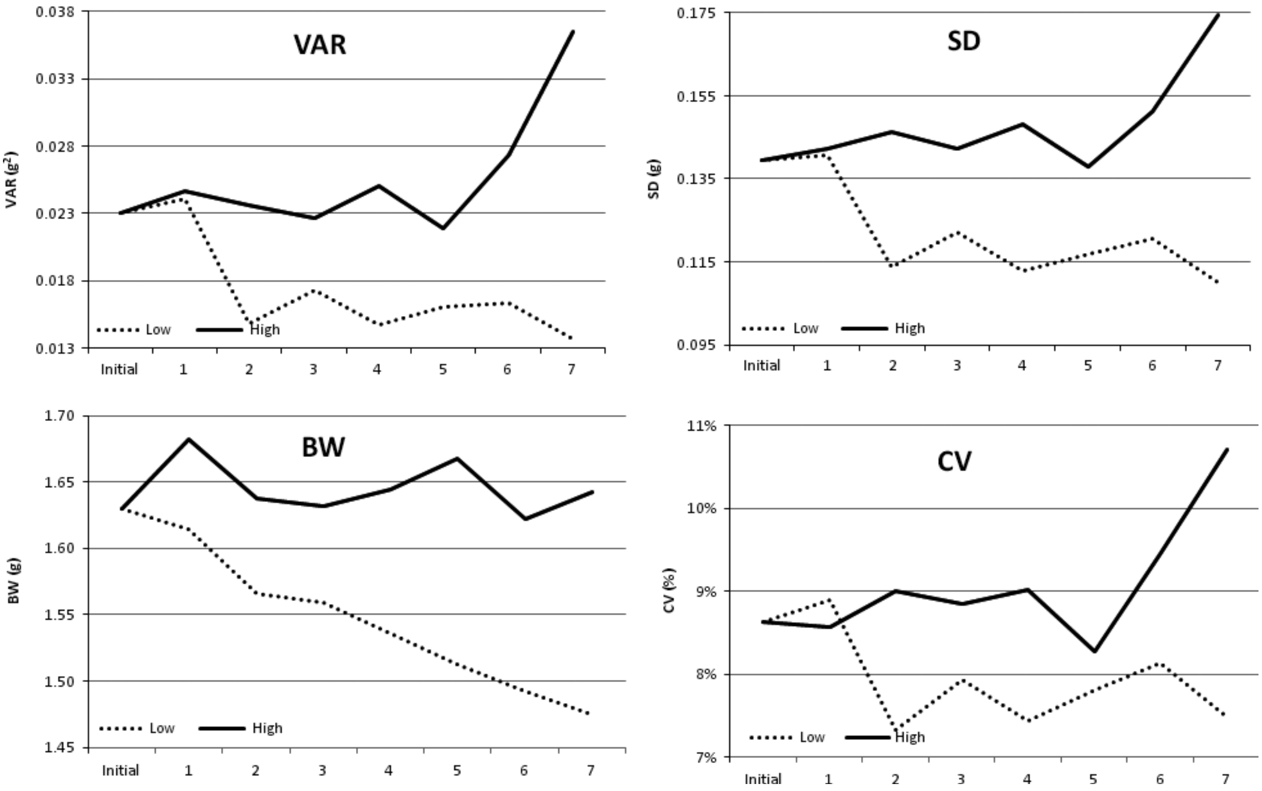

**Figure 3** Phenotypic trends of mean variance of birth weight (VAR), mean standard deviation of birth weight (SD), mean birth weight (BW) and coefficient of variation of birth weight (CV) across seven generations of selection.

values at the last generation were, respectively, for the high and the low lines, 1.64 and 1.47 for BW, 0.037 and 0.014 for VAR, 0.17 and 0.11 for SD, and 0.107 and 0.075 for CV 1. Particularly striking was that VAR in the high line was 2.67 times that of the low line. An appreciable response was also observed by ordering the litters of the last generation based on their variance and marking the position of those belonging to the different lines (Figure 4). As expected, most of the litters were ranked properly according to its association with the high or the low line. BW diverged from the beginning and during all the generations in a reasonable correlated way as expected from the positive genetic correlation.

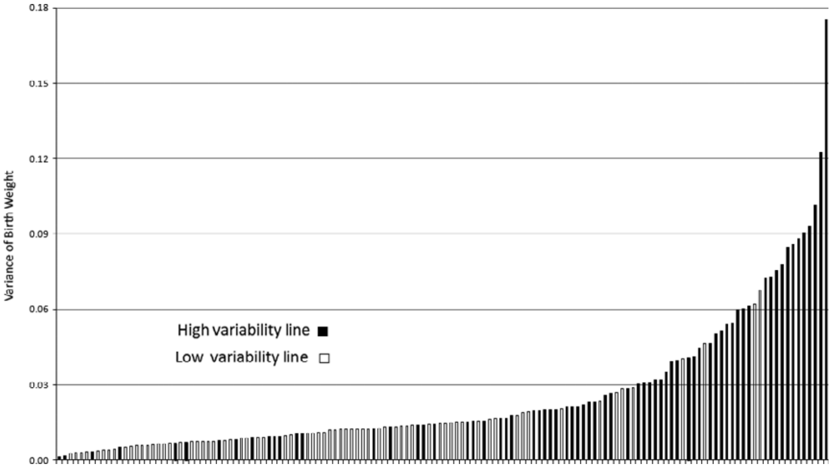

**Figure 4** Litters of the seventh generation of selection of both high and low variability lines ranked according to the variance of the birth weight.

### Selection responses

Table 4 includes information about the marginal posterior distribution of the difference between lines of each generation, considering VAR, SD, CV, BW and MBW traits, both, when considering or not, models also fitting sex (only for BW), parity and litter size.

The traits addressing the mean of the trait (BW and MBW) seemed to respond in the first generation showing a probability of the difference between lines higher than 98%. The probability of the difference in SD between lines higher than zero became higher than 97% from the second generation, and this probability was 100% for the VAR and CV traits in the last generation. At the seventh generation, VAR was 167%, SD 59%, BW 11% and CV 43% higher in the high line than in the low line (values extracted from Figure 3).

## Discussion

In this paper, we present genetic parameters and trends for BW in mice, and mainly for environmental variance, in a divergent selection experiment for BW environmental variability when the trait is attributed to the mother of the pup. The experiment for the present study was designed on the basis of the conclusions arrived at from a previous experiment by Pun

**Table 4** Mean (*D*), standard deviation (SD), 95% highest posterior density intervals (HPD95%) and probability of $D > 0$ (*P*%) of the marginal posterior distribution of the difference between lines per generation (G) for variance of birth weight (VAR), standard deviation of birth weight (SD), coefficient of variation of birth weight (CV), mean (MBW) and individual birth weight (BW), from models fitting also sex (for BW), parity and litter size as systematic effects (Ve) or not (Vr)

| G | Ve | *D* | SD | $HPD_{95\%}$ | | *P* | Vr | *D* | SD | $HPD_{95\%}$ | | *P* |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | VAR | −0.002 | 0.004 | −0.010 | 0.006 | 33 | VAR | −0.002 | 0.004 | −0.010 | 0.006 | 31 |
| | SD | 0.003 | 0.009 | −0.015 | 0.021 | 63 | SD | 0.002 | 0.009 | −0.016 | 0.020 | 56 |
| | CV | −0.002 | 0.006 | −0.014 | 0.009 | 34 | CV | −0.003 | 0.006 | −0.015 | 0.008 | 28 |
| | BW | 0.075 | 0.010 | 0.055 | 0.095 | 100 | BW | 0.069 | 0.011 | 0.048 | 0.091 | 100 |
| | MBW | 0.072 | 0.031 | 0.011 | 0.133 | 99 | MBW | 0.063 | 0.034 | −0.003 | 0.130 | 97 |
| 2 | VAR | 0.004 | 0.004 | −0.004 | 0.012 | 84 | VAR | 0.005 | 0.004 | −0.003 | 0.014 | 89 |
| | SD | 0.032 | 0.010 | 0.012 | 0.050 | 100 | SD | 0.033 | 0.010 | 0.013 | 0.052 | 100 |
| | CV | 0.017 | 0.006 | 0.005 | 0.029 | 100 | CV | 0.017 | 0.006 | 0.005 | 0.029 | 100 |
| | BW | 0.060 | 0.011 | 0.039 | 0.080 | 100 | BW | 0.073 | 0.011 | 0.051 | 0.095 | 100 |
| | MBW | 0.058 | 0.033 | −0.007 | 0.122 | 96 | MBW | 0.080 | 0.036 | 0.010 | 0.148 | 99 |
| 3 | VAR | 0.005 | 0.004 | −0.003 | 0.013 | 87 | VAR | 0.003 | 0.004 | −0.005 | 0.011 | 77 |
| | SD | 0.019 | 0.010 | 0.000 | 0.037 | 97 | SD | 0.020 | 0.009 | 0.002 | 0.038 | 98 |
| | CV | 0.009 | 0.006 | −0.002 | 0.021 | 94 | CV | 0.009 | 0.006 | −0.003 | 0.021 | 94 |
| | BW | 0.059 | 0.010 | 0.039 | 0.078 | 100 | BW | 0.075 | 0.011 | 0.054 | 0.096 | 100 |
| | MBW | 0.104 | 0.032 | 0.041 | 0.166 | 100 | MBW | 0.127 | 0.034 | 0.059 | 0.194 | 100 |
| 4 | VAR | 0.007 | 0.004 | −0.001 | 0.015 | 97 | VAR | 0.009 | 0.004 | 0.001 | 0.017 | 99 |
| | SD | 0.033 | 0.009 | 0.014 | 0.051 | 100 | SD | 0.034 | 0.009 | 0.016 | 0.052 | 100 |
| | CV | 0.014 | 0.006 | 0.003 | 0.026 | 99 | CV | 0.014 | 0.006 | 0.002 | 0.026 | 99 |
| | BW | 0.086 | 0.010 | 0.066 | 0.106 | 100 | BW | 0.103 | 0.011 | 0.081 | 0.125 | 100 |
| | MBW | 0.058 | 0.032 | −0.005 | 0.119 | 96 | MBW | 0.085 | 0.034 | 0.020 | 0.152 | 99 |
| 5 | VAR | 0.002 | 0.004 | −0.006 | 0.010 | 69 | VAR | 0.005 | 0.004 | −0.004 | 0.013 | 87 |
| | SD | 0.019 | 0.009 | 0.001 | 0.037 | 98 | SD | 0.021 | 0.009 | 0.003 | 0.039 | 99 |
| | CV | 0.005 | 0.006 | −0.007 | 0.016 | 79 | CV | 0.005 | 0.006 | −0.007 | 0.016 | 79 |
| | BW | 0.116 | 0.010 | 0.096 | 0.136 | 100 | BW | 0.141 | 0.011 | 0.120 | 0.163 | 100 |
| | MBW | 0.138 | 0.031 | 0.076 | 0.200 | 100 | MBW | 0.159 | 0.033 | 0.095 | 0.225 | 100 |
| 6 | VAR | 0.009 | 0.004 | 0.001 | 0.017 | 98 | VAR | 0.013 | 0.004 | 0.005 | 0.020 | 100 |
| | SD | 0.025 | 0.009 | 0.006 | 0.044 | 100 | SD | 0.029 | 0.009 | 0.011 | 0.047 | 100 |
| | CV | 0.014 | 0.006 | 0.002 | 0.026 | 99 | CV | 0.014 | 0.006 | 0.002 | 0.025 | 99 |
| | BW | 0.049 | 0.010 | 0.029 | 0.069 | 100 | BW | 0.099 | 0.011 | 0.078 | 0.121 | 100 |
| | MBW | 0.052 | 0.032 | −0.012 | 0.115 | 95 | MBW | 0.122 | 0.034 | 0.054 | 0.188 | 100 |
| 7 | VAR | 0.020 | 0.004 | 0.012 | 0.028 | 100 | VAR | 0.023 | 0.004 | 0.015 | 0.032 | 100 |
| | SD | 0.064 | 0.009 | 0.046 | 0.083 | 100 | SD | 0.064 | 0.009 | 0.046 | 0.083 | 100 |
| | CV | 0.035 | 0.006 | 0.023 | 0.046 | 100 | CV | 0.032 | 0.006 | 0.020 | 0.044 | 100 |
| | BW | 0.115 | 0.010 | 0.095 | 0.135 | 100 | BW | 0.143 | 0.011 | 0.122 | 0.165 | 100 |
| | MBW | 0.137 | 0.032 | 0.073 | 0.199 | 100 | MBW | 0.198 | 0.034 | 0.130 | 0.265 | 100 |

*et al.* (2013), showing that the BW environmental variability should be considered as a maternal trait. Pun *et al.* (2013) showed that, if the BW was assigned to the individual, the additive genetic variance of the environmental variability and the genetic correlation between the trait and its environmental variability would be estimated out of the expected range for them according to the revision by Hill & Mulder (2010). But these parameters would be correctly estimated if the BW and its environmental variability were attributed to the mother. Furthermore, selection accuracy is likely to be higher in the parental generations (dams) than in the offspring generation individuals, given that the former generations include their own information on the trait, information on their ascendants and that on their descendants. Besides, there are many records attributed to the same dam. This is likely to make selection of dams more effective compared to the selection of the offspring. The content of this paper centres on analysing how the experiment evolved after seven generations of divergent selection.

The absolute values of the additive genetic variance for the trait assigned to the mother resulted in half of that estimated by Pun *et al.* (2013) in a mice population with a common origin while litter variance became similar. Regarding parameters concerning variability, genetic variance was much smaller while the litter component was similar. The mean of the marginal posterior distribution of the genetic correlation between BW and its environmental variability resulted in 0.26, lower than that of 0.48 estimated by Pun *et al.* (2013) for the same trait under the same model and for a population with a common origin. The fairly low magnitude of this correlation was a satisfying result because a high correlation, either positive or negative, would imply that PBVvs was strongly influenced by PBVs as these predictions include less noise. Note that this genetic correlation was estimated to be 0.73 when splitting the lines and that only from the second generation, it approached lower magnitudes.

On the other hand, one must be careful about the relatively high SD of its marginal posterior distribution. Therefore, it is important to note that PBVs and PBVvs in each generation depended on the genetic parameters estimates and these seemed not to be reliable up to the third generation, at which time they became stable. Gutiérrez *et al.* (2006) also found extreme genetic correlation (0.97) between the MBW and its environmental variability also considering the trait as a maternal effect. This estimate is in fact incomparable given that working on MBW leads to studying the environmental variability between litters, whereas when this model is applied for individual birth weight what the analysis is accounting for is the within-litter variability (Pun *et al.* 2013). In addition to the genetic correlation, another identified checkpoint is the magnitude of the additive genetic component regarding environmental variability (Hill & Mulder 2010; Pun *et al.* 2013). Genetic CV of the environmental variability can be approximated by the squared root of $\sigma^2_{a^*}$ (Hill & Mulder 2010). This parameter was from 0.19 in the first generation to become stable around 0.05 in the fourth to seventh generation. While this value is the lowest when compared with that reviewed by Hill & Mulder (2010), it seems simply to provide information about a low genetic variance of the environmental variability with its consequent low expected response to genetic selection.

After a first inspection of the evolution of the experiment, the genetic (Figure 2) and phenotypic (Figure 3) trends and the differences between lines after seven generations led to the conclusion that genetic control of the environmental variance for birth weight was possible by artificial selection in mice. As an example, two animals with extreme weight belonging to the same litter are shown in Figure 5.

Damgaard *et al.* (2003) also concluded that in pigs, the maternal genetic variance and heritability found for within-litter SD in BW indicated that genetic improvement of this trait by selective breeding was possible.

Closer inspection of Figure 3 enabled us to unravel some particular issues. For example, in the initial generation when lines split, the success in the divergent selection for environmental variability seemed

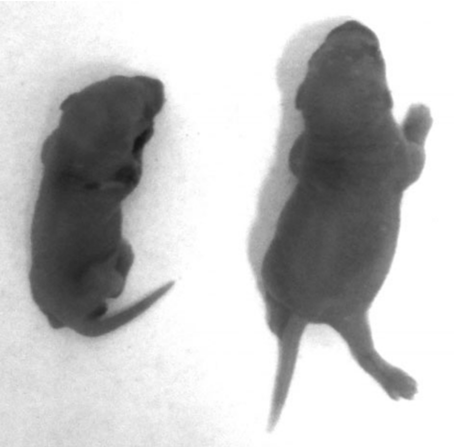

**Figure 5** Example of mice born with extreme weights (1.15 and 2.06 g) in the same litter of the high variability line.

too weak. But there was a clear divergence for BW, originating an unwanted response in CV. As postulated above, this seemed to have been due to the high estimated genetic correlation at this point of the experiment (0.73), leading most of the weight used by the model to imply that PBVvs were coming from their correlated PBV. This is because modelling the environmental variance is subject to much more noise than when modelling for the mean of the trait. For example, the same variance can appear in a litter of eight pups when all of them differ by 0.1 g, or when all of them have identical weight except one with 0.7 g less. The first scenario is a clear variable litter, and the second scenario is a homogeneous litter with the birth of a runt. The reason for the variance of the second litter is probably not related with the fitted maternal influence. However, this mother will be discarded in the low line and selected in the high line, with the aggravating weighted selection that will imply selecting as many as possible individuals from its litter.

Another example of noise is that originating from the litter size. Higher litter sizes are linked to smaller variance, and this is expected to be partially corrected by fitting the corresponding systematic effect in the model. But there is another influence of litter size. If a female tended to give high variability litters, and if the litter size is small, all animals can be of a similar size by chance, but if the litter size was larger, the animals will tend to be born with different weights.

Even though the first generation was with the strongest selection intensity, this effort was really applied on the mean of the trait and not on the environmental variability as desired. Unfortunately, this genetic correlation, as the other genetic parameters, only became stable, and therefore reliable, in the third generation, when the number of records approached 4000. Given this observation, and in the light of the results by Pun *et al.* (2013), if, when using the model by SanCristobal-Gaudy *et al.* (1998), the genetic correlation between the trait and its variability was extreme, the use of the genetic value for the variability will achieve a result in response to the mean of the trait, but the result for variability will be doubtful.

The evolution of the difference of SD between lines across generations was 0.003, 0.032, 0.019, 0.033, 0.019, 0.025 and 0.064 that seem to be a premature result but keeping the divergence for five additional generations finally to arrive at a pronounced increase in the divergence (Figure 3). A result that seems to be clear but it is also necessary to have many more generations to establish whether this result was at the initial stage of the experiment, or whether this was a continuous result that was obtained for each generation as Figure 2 seems to demonstrate. After four generations of selection for birth weight environmental variability in rabbits, Garreau *et al.* (2008) also showed a strong divergence at the generation splitting the lines but without a posterior increase in the divergence. Argente *et al.* (2012) reported successful divergent selection after five generations for residual variance of litter size, but the maximum difference between lines appeared at the third generation and then decreased again to remain two-thirds of that value for two additional generations.

Anyway, nature is probably much more complex than the pure additive model fitted to analyse the variability, and it seems that this particular model somehow fails in this type of trait. In fact, when thinking of the possible causes of the efficacy of the selection process, multiple causes are at play such as those related to the morphology of the uterus (Bolet *et al.* 2007), litter size (Argente *et al.* 2012), survivability (Mesa *et al.* 2006) and many more other causes.

On the other hand, underlying mechanisms of genetic heterogeneity are still unclear. Rönnegård & Valdar (2011, 2012) demonstrated the presence of major genes controlling the phenotypic variance on simulated and real F2 intercrosses, referring to these as vQTL. They also illustrated the connection between vQTL and QTL involved in epistasis, explaining how these concepts overlap. Environmental variability should therefore be called residual variability given that the fitted model operates on the residual and some non-environmental underlying genetic mechanisms could be involved.

Taking into account that selecting to modify the environmental variability seems to be feasible, an important point is to clarify whether such selection is beneficial, whether it is interesting to increase or decrease the variability and what applications and implications are involved. In general, homogeneity has been associated with productivity. Bolet *et al.* (2007) affirmed that reducing the heterogeneity might be useful for the rabbit industry, as it would induce lower mortality, as a result of the loss of the weakest animals. These authors cite other references who suggest that heterogeneous litters would be more prone to diseases that infect the other pups in the litter (Poignier *et al.* 2000). However, grading birth weight and fostering methods would improve preweaning survival (Perrier 2003). Survivability is also a key trait in animal production that might be affected by differences in variability. Also, losses from birth to weaning were moderately genetically linked with an increase in the within-litter variability in birth weight of pigs (Damgaard *et al.* 2003; Wolf *et al.* 2008). In addition, selecting for the capacity of sows to give birth to homogeneous litters might be advantageous for piglet growth and litter homogeneity at weaning (Damgaard *et al.* 2003). Moreover, in the successful divergent selection experiment for BW in rabbits by Bolet *et al.* (2007), a favourable correlated response for litter size at weaning and for survival from birth to weaning was also observed, but differences between lines in litter size at birth were inappreciable. Other authors have also reported higher mortality on heterogenous litters than on homogenous ones in rabbits (Poigner *et al.* 2000) and in pigs (Mesa *et al.* 2006). Argente *et al.* (2012) found that environmental variance of litter size appeared to be negatively correlated with litter size after three generations of divergent selection in rabbits, showing that the relationship between homogeneity and litter size must to be studied in each case.

A final point worth mentioning refers to the relationship between homogeneity and robustness. This term is usually controversial given that the meaning of robustness in the animal genetic context can point to two different concepts. The first considers the robustness as an ability to maintain a global production level in stressful environments. This is has been partially explained because it is mainly related to the role of the corticotrop axis. The second meaning defines the robustness as the ability to maintain the expression of a given trait with a low variability in different environments (Bodin *et al.* 2010). Obviously, the second definition of the term is closely linked to the selection aimed at reducing the variability, a concept called canalization. The relationship between the homogeneity and the robustness in the first cited meaning remains to our knowledge unexplored. Another interesting topic is the animal welfare. García *et al.* (2012) have found evidence of a relationship between homogeneity and animal welfare by measuring reactive protein, haptoglobin and amyloid A in does within both lines of the divergent selection experiment for variability of litter size in rabbits (Argente *et al.* 2012), but results were inconclusive.

To conclude, even though satisfactory results were obtained in this experiment, the small limitation already reported by Pun *et al.* (2013) cannot be ignored. The BW trait seems to be partially under individual genetic control but not only under maternal control. Unfortunately, we do not have the software that can solve the model by SanCristobal-Gaudy *et al.* (1998) with both direct and maternal genetic effects thus affecting the mean and the variability of the trait.

Genetic control of the birth weight environmental variability has been shown to be possible in mice. Before applying our results directly on farm animals, it would be worthwhile to confirm any implications on other traits, such as robustness, longevity and welfare.

## Acknowledgements

This manuscript was partially funded by a grant from the Spanish Government (AGL2008-00794). The experiment will be continued with partial funding of Feed-agene and a grant from MEC- INIA (RTA2014-00015-C02-01).

## References

Argente M.J., García M.L., Muelas R., Blasco A. (2012) Divergent selection for residual variance of litter size. In: 10th World Rabbit Congress, Sharm El-Sheikh, Egypt, pp. 97–101.

Berard J., Kreuzer M., Bee G. (2008) Effect of litter size and birth weight on growth, carcass and pork quality, and their relationship to postmortem proteolysis. *J. Anim. Sci.*, **86**, 2357–2368.

Bodin L., Bolet G., Garcia M., Garreau H., Larzul C., David I. (2010) Robustesse et canalisation, vision de généticiens. *INRA Prod. Anim.*, **23**, 11–22.

Bolet G., Gaffeau H., Joly T., Theau-Clement M., Faheres J., Hurtaud J., Bodin L. (2007) Genetic homogenisation of birth weight in rabbits: indirect selection response for uterine horn characteristics. *Livest. Sci.*, **111**, 28–32.

Damgaard L., Rydhmer L., Lovendahl P., Grandinson K. (2003) Genetic parameters for within-litter variation in piglet birth weight and change in within-litter variation during suckling. *J. Anim. Sci.*, **81**, 604–610.

Fernández J., Toro M. (1999) The use of mathematical programming to control inbreeding in selection schemes. *J. Anim. Breed. Genet.*, **116**, 447–466.

García M.L., Argente M.J., Muelas R., Birlanga V., Blasco A. (2012) Effect of divergent selection for residual variance of litter size on health status and welfare. In: 10th World Rabbit Congress, Sharm El- Sheikh, Egypt, pp. 103–106.

Garreau H., Bolet G., Larzul C., Robert-Granie C., Saleil G., SanCristobal M., Bodin L. (2008) Results of four generations of a canalising selection for rabbit birth weight. *Livest. Sci.*, **119**, 55–62.

Gutiérrez J.P., Nieto B., Piqueras P., Ibañez N., Salgado C. (2006) Genetic parameters for canalisation analysis of litter size and litter weight traits at birth in mice. *Genet. Sel. Evol.*, **38**, 445–462.

Hill W.G., Mulder H.A. (2010) Genetic analysis of environmental variation. *Genet. Res.*, **92**, 381–395.

Högberg A., Rydhmer L. (2000) A genetic study of piglet growth and survival. *Acta Agric. Scand. Sect. A Anim. Sci.*, **50**, 300–303.

Ibáñez-Escriche N., Sorensen D., Waagepetersen R., Blasco A. (2008b) Selection for environmental variation: a statistical analysis and power. *Genetics*, **180**, 209–226.

Ibáñez-Escriche N., Moreno A., Nieto B., Piqueras P., Salgado C., Gutierrez J.P. (2008a) Genetic parameters related to environmental variability of weight traits in a selection experiment for weight gain in mice; signs of correlated canalised response. *Genet. Sel. Evol.*, **40**, 279–293.

Ibáñez-Escriche N., García M., Sorensen D. (2010) GSEVM v. 2: MCMC software to analyze genetically structured environmental variance models. *J. Anim. Breed. Genet.*, **127**, 249–251.

Legarra A. (2008) TM Threshold Model (available at: http://acteon.webs.upv.es/; last accessed 3 September 2014).

Mesa H., Safranski T.J., Cammack K.M., Weaber R.L., Lamberson W.R. (2006) Genetic and phenotypic relationships of farrowing and weaning survival to birth and placental weights in pigs. *J. Anim. Sci.*, **84**, 32–40.

Moreno A., Ibáñez-Escriche N., García-Ballesteros S., Salgado C., Nieto B., Gutiérrez J.P. (2012) Correlated genetic trend in the environmental variability of weight traits in mice. *Livest. Sci.*, **148**, 189–195.

Mormede P., Terenina E. (2012) Molecular genetics of the adrenocortical axis and breeding for robustness. *Domest. Anim. Endocrinol.*, **43**, 116–131.

Perrier G. (2003) Influence de l'homogénéité de la portée sur la croissance et la viabilité des lapereaux de faible poids à la naissance. In: 10èmes Journées de la recherche cunicole, Paris, France, pp. 119–122.

Poigner J., Szendrö Zs., Levai A., Radnai I., Biro-Nemeth E. (2000) Effect of birth weight and litter size on growth and mortality in rabbit. *World Rabbit Sci.*, **8**, 103–109.

Pun A., Cervantes I., Nieto B., Salgado C., Pérez-Cabal M.A., Ibáñez-Escriche N., Gutiérrez J.P. (2013) Genetic parameters for birth weight environmental variability in mice. *J. Anim. Breed. Genet.*, **130**, 404–414.

Rönnegård L., Valdar W. (2011) Detecting major genetic *loci* controlling phenotypic variability in experimental crosses. *Genetics*, **188**, 435–447.

Rönnegård L., Valdar W. (2012) Recent developments in statistical methods for detecting genetic loci affecting phenotypic variability. *BMC Genet.*, **13**, 63–447.

Ros M., Sorensen D., Waagepetersen R., Dupont-Nivet M., SanCristobal M., Bonnet J.C., Mallard J. (2004) Evidence for genetic control of adult weight plasticity in the snail *Helix aspersa*. *Genetics*, **168**, 2089–2097.

SanCristobal M., Bodin L., Elsen J.M., Chevalet C. (2001) Genetic components of litter size variability in sheep. *Genet. Sel. Evol.*, **33**, 249–271.

SanCristobal-Gaudy M., Elsen J., Bodin L., Chevalet C. (1998) Prediction of the response to a selection for canalisation of a continuous trait in animal breeding. *Genet. Sel. Evol.*, **30**, 423–451.

Scheiner S.M., Lyman R.F. (1991) The genetics of phenotypic plasticity. II. Response to selection. *J. Evol. Biol.*, **4**, 23–50.

Sorensen D., Waagepetersen R. (2003) Normal linear models with genetically structured residual variance heterogeneity: a case study. *Genet. Res.*, **82**, 207–222.

Waddington C.H. (1942) Canalization of development and the inheritance of acquired characters. *Nature*, **150**, 563–565.

Wolf J., Žakova E., Groeneveld E. (2008) Within-litter variation of birth weight in hyperprolific Czech Large White sows and its relation to litter size traits, stillborn piglets and losses until weaning. *Livest. Sci.*, **115**, 195–205.