



UNIVERSIDAD
POLITECNICA
DE VALENCIA



MÁSTER INTERUNIVERSITARIO EN MEJORA GENÉTICA
ANIMAL Y BIOTECNOLOGÍA DE LA REPRODUCCIÓN

Divergent selection for residual variance of
litter size in rabbits

Tesis de Máster

Valencia, Julio 2013

Marina Martínez Álvaro

Director:

Agustín Blasco



ACKNOWLEDGEMENTS

Me gustaría agradecer a Agustín Blasco por el tiempo que me ha dedicado y por las oportunidades que me ofrece, y su empeño en hacerme pensar por mi misma y en cuestionarme todas las cosas. También a Noelia Ibáñez por su tiempo dedicado en mi visita a Lérida, y a María José Argente y Mari Luz García por su colaboración en esta tesis.

Quería agradecer a mis compañeros Cris, Vero y Luis su apoyo en el trabajo del día a día, y por hacerme este camino más fácil.

Por último, a mi familia y a mis amigos por estar conmigo siempre.

CONTENT

ABSTRACT	7
RESUMEN	8
RESUM	9
LITERATURE REVIEW	10
1. Residual variance and its interest	10
2. Estimation of genetic variability of residual variance.....	11
2.1. Analysis of data bases	11
2.2. Problems of heteroscedastic models	13
3. The modeling of the residual variance	14
4. Relationship between mean and variance	16
5. Selection experiments on residual variance	17
OBJECTIVES	19
ARTICLE 1: DIVERGENT SELECTION FOR RESIDUAL VARIANCE OF LITTER SIZE IN RABBITS.....	20
Abstract	21
Introduction	21
Materials and methods	23
Results and discussion.....	26
Conclusions.....	36
Acknowledgements	36
References	37
ARTICLE 2: GENETIC TRENDS IN A DIVERGENT SELECTION EXPERIMENT BY RESIDUAL VARIANCE OF LITTER SIZE IN RABBITS.	40
Abstract	41
Introduction	41
Material and methods.....	42
Results and discussion.....	45
Conclusions.....	49
References	49
GENERAL DISCUSSION AND CONCLUSIONS	52
REFERENCES.....	53

INDEX OF TABLES

ARTICLE 1

Table 1. Mean, median, standard deviation (Sd) and coefficient of variation (CV) of residual variance of litter size precorrecting data (V_e), residual variance of litter size without pre correction (V_r), residual standard deviation of litter size (Sd_e) and litter size (LS).....	26
Table 2. Weighted selection differential for residual variance of litter size precorrected for year-season and lactation status (V_e) in each generation, measured as kits ²	28
Table 3. Features of the estimated marginal posterior distributions of the differences in V_e between the high and the low lines selected for residual variance of litter size using precorrected data.....	29
Table 4. Features of the estimated marginal posterior distributions of the differences in Sd_e between the high and the low lines selected for residual variance of litter size using precorrected data.....	31
Table 5. Features of the estimated marginal posterior distributions of the differences in V_r between the high and the low lines selected for residual variance of litter size using precorrected data.....	33
Table 6. Features of the estimated marginal posterior distributions of the differences in LS between the high and the low lines selected for residual variance of litter size using precorrected data	35

ARTICLE 2

Table 1. Heritabilities for traits V_e , V_r and LS	49
--	----

INDEX OF FIGURES

ARTICLE 1

- Figure 1.** Data distribution by quartiles of (A) residual variance without precorrecting data (V_r) and (B) residual variance precorrecting data (V_e), with litter size average of all parities of each female..... 27
- Figure 2.** Means and standard deviations of the estimated marginal posterior distributions of each line in each generation for residual variance of litter size using precorrected data (V_e); H:high line; L:low line; Base:Base generation; Gen: generation. 28
- Figure 3.** Histograms of the (A) residual variance of litter size data (V_e) and for the (B) residual standard deviation of litter size data (Sd_e). 30
- Figure 4.** Means and standard deviations of the marginal posterior distribution of each line in each generation for residual standard deviation of litter size (Sd_e). H:high line; L: low line; Base:Base generation; Gen: generation..... 30
- Figure 5.** Means and standard deviations of the estimated marginal posterior distributions of each line in each generation for residual variance of litter size data without precorrection (V_r); H:high line; L:low line; Base:Base generation; Gen: generation. 32
- Figure 6.** Means and standard deviations of the estimated marginal posterior distributions of each line in each generation for litter size (LS); H:high line; L:low line; Base:Base generation; Gen:generation..... 34

ARTICLE 2

Figure 1. Means and standard deviations of marginal posterior distributions of the responses obtained in each line and generation for residual variance of litter size, precorrecting data (V_e). H: high line. L: low line..... 46

Figure 2. Means and standard deviations of marginal posterior distributions of the responses obtained in each line and generation for residual variance of litter size, without precorrecting data (V_r). H: high line. L: low line. 47

Figure 3. Means and standard deviations of marginal posterior distributions of the responses obtained in each line and generation for litter size (LS). H: high line. L: low line. 48

ABSTRACT

The aim of this study is to analyze the genetics of residual variance of litter size. For this purpose, a divergent selection experiment by residual variance in rabbits has been performed during five generations. The selection criterion was the residual variance of litter size (V_e) estimated as the variance of litter size within doe, precorrecting litter size data by generation and lactation status effect, to avoid the possible interference of systematic effects on the variance. The residual variance of litter size was estimated without pre-correction as well (V_r). Bayesian methods were used to carry out the analyses. At the end of the experiment, V_e and V_r were 37% and 38% higher in line H than in line L, respectively. Differences between lines were 1.21 kits² and 1.19 kits² and reached at least 0.87 kits² and 0.86 kits² with a probability of 80%, for V_e and V_r , respectively. The selection response on V_e was asymmetrical, showing the high line a greater response. The estimated genetic means for V_e and V_r in the fifth generation were 0.45 kits² and 0.47 kits² in the high line and -0.24 kits² and -0.26 kits² in the low lines, respectively. V_r was highly correlated with V_e , thus pre-correction had almost no effect on the response. There was a negative correlated response on litter size (LS). In generation five, the phenotypic difference between high and low line for LS was -0.63 kits, and reached at least -0.43 kits with a probability of 80%. Litter size was 7% higher in L line than in H line. A negative indirect response was found for LS in the homogeneous line, which showed an estimated genetic mean in the fifth generation of 0.66 kits. Estimates of heritability for V_e and V_r were low, 0.06 and 0.05 respectively, thus responses could be due to the high variability of the traits. To summarize, there is new experimental evidence for the residual variance being partly under genetic control. In addition, decreasing the residual variance of litter size is accompanied by an increase of litter size.

Keywords: residual variance, rabbits, litter size.

RESUMEN

El objetivo de este estudio es analizar la genética de la varianza residual del tamaño de camada. Para ello se llevó a cabo un experimento de selección divergente por varianza residual en conejos, a lo largo de cinco generaciones. El criterio de selección fue la varianza residual del tamaño de camada (V_e) estimada como la varianza del tamaño de camada dentro de hembra, precorrigiendo los datos de tamaño de camada por los efectos de generación y estado de lactación para evitar posibles interferencias de los efectos sistemáticos en la varianza. Se ha calculado también la varianza residual del tamaño de camada sin precorregir los datos (V_r). Los análisis se han llevado a cabo con metodología bayesiana. Al final del experimento V_e y V_r fueron un 37% y un 38% superiores en la línea seleccionada para baja variabilidad respecto de la línea seleccionada para alta variabilidad, respectivamente. Las diferencias entre líneas fueron de 1.21 gazapos² y 1.19 gazapos² y alcanzaron, al menos, 0.87 gazapos² y 0.86 gazapos² con una probabilidad del 80%, en V_e y V_r , respectivamente. La respuesta a la selección de V_e fue asimétrica, mostrando la línea heterogénea una mayor respuesta. Las medias genéticas estimadas para V_e y V_r en la quinta generación fueron de 0.45 gazapos² y 0.47 gazapos² en la línea de alta variabilidad y de -0.24 gazapos² y -0.26 gazapos² en la línea de baja variabilidad, respectivamente. V_r y V_e mostraron estar altamente correlacionados, así que la precorrección de los datos prácticamente no afectó a la respuesta. Hubo una respuesta correlacionada en tamaño de camada (LS). En la quinta generación, la diferencia fenotípica entre las líneas de alta y baja variabilidad fue de -0.63 gazapos, y alcanzó, al menos, -0.43 gazapos con una probabilidad del 80%. LS fue un 7% más alto en la línea homogénea que en la heterogénea. Se observó una respuesta indirecta correlacionada para LS en la línea homogénea, que mostró una media genética estimada de 0.66 gazapos. Las estimas de heredabilidad para V_e y V_r fueron bajas, de 0.06 y 0.05 respectivamente, por tanto las respuestas obtenidas pueden deberse a la alta variabilidad de los caracteres. En conclusión, se han encontrado evidencias experimentales de la existencia de una componente genética en la varianza residual del tamaño de camada. Además, la selección para disminuir la varianza residual del tamaño de camada va acompañada de un incremento en el tamaño de camada.

Palabras clave: varianza residual, conejos, tamaño de camada.

RESUM

L'objectiu d'aquest estudi és analitzar la genètica de la variància residual del tamany de la ventrada. Per a açò es va dur a terme un experiment de selecció divergent per variància residual en conills, al llarg de cinc generacions. El criteri de selecció va ser la variància residual del tamany de la ventrada (V_e) estimada com la variància del tamany de la ventrada dins de femella, precorregint les dades de tamany de la ventrada pels efectes de generació i estat de lactació per a evitar possibles interferències dels efectes sistemàtics en la variància. S'ha calculat també la variància residual del tamany de la ventrada sense precorregir les dades (V_r). Els anàlisis s'han dut a terme amb metodologia bayesiana. Al final de l'experiment V_r i V_e van ser un 37% i un 38% superiors en la línia seleccionada per a baixa variabilitat respecte de la línia seleccionada per a alta variabilitat, respectivament. Les diferències entre línies van ser de 1.21 gazapos² i 1.19 gazapos² i van aconseguir, almenys, 0.87 gazapos² i 0.86 gazapos² amb una probabilitat del 80%, en V_e i V_r , respectivament. La resposta a la selecció de V_e va ser asimètrica, mostrant la línia heterogènia una major resposta. Les mitjanes genètiques estimades per a V_e i V_r en la cinquena generació van ser de 0.45 gazapos² i 0.47 gazapos² en la línia d'alta variabilitat i de -0.24 gazapos² i -0.26 gazapos² en la línia de baixa variabilitat, respectivament. V_r i V_e van mostrar estar altament correlacionats, així que la precorrecció de les dades pràcticament no va afectar a la resposta. Va haver-hi una resposta correlacionada en tamany de la ventrada (LS). En la cinquena generació, la diferència fenotípica entre les línies d'alta i baixa variabilitat va ser de -0.63 gazapos, i va aconseguir, almenys, -0.43 gazapos amb una probabilitat del 80%. LS va ser un 7% més alt en la línia homogènia que en l'heterogènia. Es va observar una resposta indirecta correlacionada per a LS en la línia homogènia, que va mostrar una mitjana genètica estimada de 0.66 gazapos. Les estimes de heretabilitat per a V_e i V_r van ser baixes, de 0.06 i 0.05 respectivament, per tant les respostes obtingudes poden deure's a l'alta variabilitat dels caràcters. En conclusió, s'han trobat evidències experimentals de l'existència d'una component genètica en la variància residual del tamany de la ventrada. A més, la selecció per a disminuir la variància residual del tamany de la ventrada va acompanyada d'un increment en el tamany de la ventrada.

Paraules clau: variància residual, conills, tamany de la ventrada.

LITERATURE REVIEW

1. Residual variance and its interest

Observed variability of traits is composed of genetic and environmental variability. Environmental variability can be reduced by improving the management and the environmental conditions of the farm and correcting data by environmental effects. After correcting data by environmental effects, the remaining environmental variance is the residual variance caused by non-controlled random effects. Residual variability has become a point of interest both in animal production and evolutionary biology.

Animal production has different interests in reducing the variance; however is interesting to reduce the variability of the traits but without reducing its possibilities for improving the mean. Uniformity of animal products is more economically efficient. In sheep, ewes lambing twins represent the economic optimum since lambings with more lambs are frequently not viable (SanCristóbal *et al.*, 2001). In prolific species, uniformity of litters facilitates management by reducing adoptions. Homogeneity of birth weight within litter in rabbits may be related with a higher viability of the young rabbits, higher litter size at weaning and less variability of weight at weaning (Bolet *et al.*, 2008). Optimal weights and ages at slaughtering for pigs, broilers and lambs are demanded, and the profits of the breeders depend on their ability to send large homogenous groups to the slaughterhouse. A primary concern of beef producers is to improve uniformity of production traits such as carcass weight, fat deposition and carcass composition, because the price paid to breeders is based in this criteria. Optimal characteristics of meat, such as, pH 24 h after slaughter are requested as well (Larzur *et al.*, 2006).

Reducing the residual variance can augment the heritability of the selected traits. Reproductive traits, particularly litter size, are characterized by low heritabilities that make their genetic improvement a slow and costly process. Furthermore, the estimates of the litter size heritabilities do not correspond to the low success obtained in selection, and they are usually inflated with respect to the realized heritabilities (Blasco, 1996). With a reduction of the residual variance, genetic progress by generation would be higher due to an increase of accuracy in selection. Several

models have been proposed to explain that stabilizing selection can result in an increase in the heritability (Gavrilets and Hastings, 1994).

Other interest is to improve the welfare of animals. García *et al.* (2012) used biomarkers of health status and welfare to compare the health status and welfare of does with high variability of litter size and does with low variability of litter size, in rabbits. They found that does with high variability of litter size have poorer health status than does with low variability.

Residual variance has also interest in evolutionary biology. Stabilizing selection is the most common mechanism of action in natural selection. Stabilizing selection favouring an optimum phenotype depletes variation in quantitative traits (Falconer and Mackay, 1996). Moreover, if a genetic component is involved in the residual variance, it would tend to be reduced. However, residual variance has been maintained along the time, thus it could be possible that residual variance is not under genetic control. Several theories try to explain this fact. Slaktin and Lande (1976) showed, that if the optimum phenotype changes over time due to the permanent changing environment, phenotypic (and residual) variance is maintained. Zhang and Hill (2008) proposed equilibrium between mutation and stabilizing selection that maintains the level of the residual variation, which implies that residual variance would be under genetic control.

2. Estimation of genetic variability of residual variance

2.1. Analyses of data bases

Several authors have analysed data bases with the aim of giving evidences that there are genes controlling residual variance.

An easy way to find evidence is to analyse the standard deviation of a trait. Damgaard *et al.* (2003) used a pig dataset of weight at birth and at 3 weeks. The traits analysed were standard deviations and means of weight at birth and at 3 weeks of age regarded as maternal traits. They found genetic variance for the standard deviations of weight at birth and at 3 weeks. However, the results may be biased

because they did not correct the birth weight and the birth at 3 weeks by litter size effect, which affects their standard deviation.

Analyses of datasets using heteroscedastic models are the most common analyses made to find evidence of a genetic component of residual variance. Several authors applied multiplicative heteroscedastic models to find a genetic component in the residual variance. SanCristóbal *et al.* (2001) analyzed data of litter size in Lacaune sheep and found polygenes controlling the residual variance. Sorensen and Waagepetersen (2003) used pig litter size data to test four normal mixed models with different levels of heterogeneity in the residual variance; all criteria for the assessment of the models favored the one with genetic and permanent effects at the level of residual variance. Ros *et al.* (2004) provided evidence that residual variance for adult weight in the snail *H. aspersa* was partly under genetic control; the posterior mean of genetic variance of residual variance was 0.29. Gutiérrez *et al.* (2006) estimated genetic parameters associated with residual variance of litter size, litter birth weight and individual mean birth weight in mice and obtained genetic variances for all of them, although their values were low. Ibáñez-Escriche *et al.* (2008a) used data from a selection experiment for weight gain in mice to compare homoscedastic and heteroscedastic models, and found evidence that the heterogeneity of residual variance has a genetic effect, thus making selection possible. Ibáñez-Escriche *et al.* (2008c) studied data from uterine capacity in rabbits, and compared the fitness of two different models with different levels of complexity of residual variance; the one postulating that residual variance was heterogeneous and partly under genetic control showed a better adjustment and the posterior mean of genetic variance of residual variance was 0.16. Ibáñez-Escriche *et al.* 2008b gave evidence of genetic variation at the level of residual variance in slaughter weight pig data. Neves *et al.* (2012) and Fina *et al.* (2013) found a genetic variance component of residual variance in weight traits, in Nellore beef cattle and Bruna dels Pirineus beef cattle, respectively.

Likelihood or bayesian approaches were used in the estimation procedures of these models. Recently, Ibáñez-Escriche *et al.* (2010) designed software to analyze heteroscedastic models in a Bayesian approach. However, due to the complexity of resolving these heteroscedastic models, some authors decided to solve them in a two-step approach. In the first step, an animal model was fitted to the phenotypic

data. In the second step, a logarithmic function of estimated residuals for each phenotypic record obtained in the first step was fitted with a different animal model. By this approach, evidence of genetic variance in residual variation of body weight and body conformation in broiler chickens was given by Wolc *et al.* (2009). Using this procedure there was evidence of genetic variability for residual variance of weight traits in Nellore beef cattle as well (Neves *et al.* 2012).

Most of this evidence is indirect because it comes from data bases analyses and not from experiments designed to find the genetic part of the variance. Moreover, there is a discussion about the complexity degree of the models employed to analyze these data bases, and how they would affect the results if they are not robust. We will examine this in the next paragraph.

2.2. Problems of heteroscedastic models

Models considering heterogeneity at the level of residual variance carry on several problems; they depend on a large number of parameters and they are not robust. Heteroscedastic models fit better than homoscedastic models. A better fit of the heterogeneous variance model can be due to its large number of parameters, making it flexible to account for features of the data, but not providing decisive evidence. A better fitting does not mean a better predicting ability; too complex models can fit simultaneously the biological phenomena plus part of the residuals of the model. Cross validation methods are more appropriate to find the prediction ability of a model.

Furthermore, these models can lead to distorted inferences due to, among other reasons, confusion between genetic and environmental effects, the choice of the wrong functional relationship between mean and variance, the wrong choice of the sampling model of the data, or the wrong scale of measurement (Sorensen, 2010). For instance, the lack of normality of residues dramatically change the results: Ros *et al.* (2004) log-transformed snail data to avoid asymmetry in residuals; then the correlation between genetic effects affecting the mean and the variance, changed from positive to negative. Ye Yang *et al.* (2011) compared genetic parameters of residual variance in an untransformed scale with the same data after a Box-Cox

transformation finding that the coefficient of correlation between genetic effects of the trait and its residual variance changed sign compared to the results in the untransformed scale, in data bases of both pigs and rabbits.

On the other hand, as these models depend on a large number of parameters, a high amount of data are needed for accurate estimates of all parameters. More difficulties of heteroscedastic models occur when ignoring fixed effects that should not affect the mean but affect the variance. There could exist major or medium genes influencing the variance, or dominance or epistasis phenomena that are not taken into account.

3. The modeling of the residual variance

Quoting Sorensen (2010), “Given a genotype, the observed phenotype differs from its expectation. This deviation, which is the focus of the modeling exercise in heteroscedastic models, may include a genetic component.” Different genotypes differ in both mean and residual variance. Statistical modeling and estimation procedures have been developed to model the residual variance. Heteroscedastic models have the following structure:

$$y_{ij} = \mu + a_i + f(\mu^* + a_i^*)\varepsilon_{ij}$$

$$(y_{ij} | \mu, a_i, \mu^*, a_i^*) \sim N(\mu + a_i, V_e)$$

Being y_{ij} the data j of the individual i , μ the population mean of the trait and a_i the genetic effects of the individual i involving the mean. Residues are modeled as a function, where μ^* is the average residual variance of the trait, a_i^* the genetic effects of the individual i affecting the residual variance and ε_{ij} a random variable distributed as $\varepsilon_{ij} \sim N(0, 1)$.

Genetic effects are distributed as:

$$(\mathbf{a} | \sigma_a^2, \sigma_{a^*}^2, \rho_{aa^*}) \sim N(\mathbf{0}, \mathbf{G} \otimes \mathbf{A})$$

Where \mathbf{a} includes a_i and a_i^* of each individual, and \otimes denotes the Kronecker product between the relationship matrix \mathbf{A} and the genetic effects variance-covariance matrix \mathbf{G} , that looks like:

$$\mathbf{G} = \begin{pmatrix} \sigma_a^2 & \rho_{aa^*}\sigma_a\sigma_{a^*} \\ \rho_{aa^*}\sigma_a\sigma_{a^*} & \sigma_{a^*}^2 \end{pmatrix}$$

Where σ_a^2 and $\sigma_{a^*}^2$ are the genetic variance components affecting mean and variance respectively, and ρ_{aa^*} is the genetic correlation between genetic effects at the level of the mean and variance. Residues are normally distributed, with mean 0 and variance:

$$V_e = V(y_i | \mu, a_i, \mu^*, a_i^*) = [f(\mu^* + a_i^*)]^2 V(\varepsilon_{ij}) = [f(\mu^* + a_i^*)]^2$$

SanCristóbal *et al.* (1998) proposed a multiplicative heteroscedastic model, considering logarithmic relationship between variance and parameters. In the model, the residuals were modeled as:

$$f(\mu^* + a_i^*) = \sqrt{\exp(\mu^* + a_i^*)}$$

Variables are the same as previously defined. The logarithm of the residual variance was:

$$\log V_e = \log(\sqrt{\exp(\mu^* + a_i^*)})^2 = \mu^* + a_i^*$$

Thus, the logarithm of the residual variance and the effects of the mean of the trait are linearly correlated.

Another model was proposed by Mulder *et al.* (2007, 2008). In the model, the residuals were modeled as:

$$f(\mu^* + a_i^*) = \sqrt{\exp(\mu^*) + a_i^*}$$

Variables are the same as previously defined. The residual variance was:

$$V_e = (\sqrt{\exp(\mu^*) + a_i^*})^2 = \exp(\mu^*) + a_i^*$$

In this model, mean and variance are linearly related.

All previous models can be easily generalized for fixed effects instead μ^* , and permanent effects can be simply included in the residual variance.

Nonetheless, these models carry several problems, and can lead to distorted inferences. Difficulties are commented in the previous paragraph. No comparison between which of the proposed heteroscedastic models predicts better has been published.

4. Relationship between mean and variance

Reduction of residual variance can affect the mean of the trait if they are correlated. Several authors have estimated the genetic correlation between the genetic effects of the mean and the genetic effects of the residual variance, using heteroscedastic models.

No genetic correlation was found by several authors. In rabbits, Bolet *et al.* (2008) selected by within litter variability of birth weight but no effect was shown in the mean of the trait. In pigs, Ibáñez-Escriche *et al.* (2008b) did not find correlation between mean and residual variance of slaughter weight.

Other authors found positive genetic correlations between mean and residual variance. In *Helix aspersa* snails, Ros *et al.* (2004) provided a strong indication of a positive genetic correlation between mean and residual variation of adult body weight. In broiler chickens, Wolc *et al.* (2009) estimated positive genetic correlations between means and residual variances for body weight and body conformation of 34 days. Fina *et al.* (2013) estimated a positive genetic correlation between mean and residual variance of birth weight in Bruna dels Pirineus beef cattle.

Different results were shown by Sorensen and Waagepetersen (2003), who detected a negative genetic correlation between mean and residual variance in pig litter size data, and it was confirmed by Felleki *et al.* (2012) with the same dataset using other methodologies. Gutiérrez *et al.* 2006 used mice data to estimate the mean-residual variance genetic correlation in litter size, litter weight and mean individual weight at birth; results were -0.929, -0.815 and 0.696 respectively, this last being positive. Ibáñez-Escriche *et al.* (2008a) estimated the genetic correlation between mean and residual variance of weight at 21 days, weight at 42 days, and weight gain between 21 and 42 days, in mice, and they were between -0.19 and -0.38. Ibáñez-Escriche *et al.* (2008c) estimated the correlation between the genetic effects of the mean and

those on the residual variance in uterine capacity in rabbits, and it was -0.74. Rowe *et al.* (2006) estimated the genetic correlation between effects on progeny mean and residual variance for body weight in broiler chickens, and it was small and negative (-0.1). Mulder *et al.* (2009) found genetic correlations between body weight in broiler chickens and its residual variance of -0.41 in females, and -0.45 in males.

Nevertheless, as commented in previous paragraphs, these correlations were obtained analyzing heteroscedastics models, complex and not robust, and as said before, it is being questioned whether they are an artifact of the model (Ye Yang *et al.* 2011).

5. Selection experiments on residual variance

Few evidence for the existence of a genetic component in residual variance comes from selection experiments.

Garreau *et al.* (2008) performed a divergent selection experiment based on residual variance of birth weight in rabbits. The selection criterion was the predicted genetic value of the model proposed by SanCristóbal, in which the logarithm of the residual variance has environmental and genetic components (SanCristóbal *et al.* 1998). The model was analyzed in two steps. In the first step, an animal model was fitted to the birth weight data. In the second step, the logarithm of the square of estimated residual values obtained in the first step was fitted with a different animal model. A high response was found in the first generation, but no changes were found in the following four generations; nevertheless it should be noted that these estimates were model dependent. Moreover, no consistent results were observed in the correlated responses in standard deviation of birth weight, standard deviation of weaning weight, individual birth weight and individual weaning weight. In the work of Bolet *et al.* (2008) for the same experiment, equally difficult was to interpret the difference between lines in stillbirth rate and birth-weaning mortality.

Other experiment has been made by Pun *et al.* (2012) in environmental variability of birth weight in mice. They used an inbred line (supposing not having genetic variability) to evaluate the base generation animals for being the parents of the first generation. To evaluate the progeny of first generation, they mated males of the

selected line with females of the inbred line. They observed response in the first generation, but no response was observed in the rest of generations. They conclude that residual variability of birth weight seemed to be partially under individual genetic control and further but not totally under maternal control, thus response was not observed in the cross between males from the selected line and females from the inbred line.

Larzur *et al.* (2006) realized a divergent experiment with the purpose of achieving homogeneity in the ultimate pH value in pigs. The animals were selected on their genetic value estimated according to the method described by SanCristóbal *et al.* (1998). Boars were chosen with the additional constraint of maintaining a constant average of the ultimate pH. No response was clearly observed, since the number of animals measured within each generation was low, and it was difficult to conclude the validity of the results. The experiment was not successful and did not cast light in the alleged genetic base under residual variance.

Several authors agree that an experimental process should be a better way to approach the genetic performance of residual variance (Bodin *et al.*, 2002; Ros *et al.*, 2004; Bolet *et al.*, 2008; Sorensen, 2010). Until now, no direct evidence of genetic control of residual variance has been obtained. The objective of our experiment is to carry out a divergent selection on residual variance of litter size.

OBJECTIVES

The objective of this study is to find evidence that residual variance of litter size is under genetic control, and consequently can be considered as a selection criterion. For the first time, a direct selection experiment by residual variance has been performed to examine the genetics of residual variance.

This thesis is composed by two articles. Article 1 will be modified and submitted to a journal. Article 2 was previously submitted to the XV Jornadas sobre Producción Animal of AIDA (Zaragoza, May 2013), and it has been translated and modified for showing in this thesis. It will be submitted to a journal after small modifications.

ARTICLE 1: DIVERGENT SELECTION FOR RESIDUAL VARIANCE OF LITTER SIZE IN RABBITS

To be submitted to a journal.

DIVERGENT SELECTION FOR RESIDUAL VARIANCE OF LITTER SIZE IN RABBITS

ABSTRACT

A divergent selection experiment for residual variance of litter size was carried out in rabbits during five generations. The selection criterion was the residual variance of litter size (V_e) estimated as the variance of litter size within doe, after precorrecting litter size for generation and lactation status. The traits analyzed were V_e , residual standard deviation of litter size (Sd_e), residual variance of litter size without precorrecting data (V_r) and litter size (LS). Bayesian methods were used to analyze the response to selection. Results confirm the existence of genes controlling V_e . In the last generation residual variance was 37% higher in line H than in line L, and they differed at least in 0.87 kits² with a probability of 80%. In terms of V_r line H was 38% higher than L line in the fifth generation, and the difference between lines was at least 0.86 kits² with a probability of 80%, thus precorrecting data had almost no effect on the response. A negative indirect response was found for LS , homogenous does having higher LS . In the last generation line L was 7% higher than line H. Difference between lines in the last generation was at least -0.43 kits with a probability of 80%.

Keywords: residual variance, rabbits, litter size.

INTRODUCTION

Reproductive traits, particularly litter size, are characterized by low heritabilities that make their genetic improvement a slow and costly process. Furthermore, the estimates of the litter size heritabilities do not correspond to the low success obtained in selection, as they are usually inflated respect with the realized heritabilities (Blasco, 1996). Reducing residual variance, the genetic progress by generation should be higher due to an increase of accuracy in selection. Moreover, homogeneity is interesting for other productive reasons. In prolific species, uniformity facilitates handling and reduces adoptions. In sheep, ewes lambing twins represent the

economic optimum (SanCristóbal *et al.*, 2001). Homogeneity of birth weight within litter in rabbits is related with a higher viability of the kits (Bolet *et al.*, 2008). Slaughterhouses demand large homogeneous groups of pigs, broilers and lambs in an optimal weight and age. Optimal characteristics of meat such as pH 24 hours after slaughter are requested as well (Larzul *et al.*, 2006). In addition, animals with low residual variance are alleged to be animals which are more resistant to diseases or stress, with a better welfare status (García *et al.*, 2012).

In evolutionary biology, there is an increasing interest in understanding the mechanism regulating residual variance. Stabilizing selection favoring an optimum phenotype depletes variation in quantitative traits (Falconer and Mackay, 1996). Moreover, if a genetic component is involved in the residual variance, it would tend to be reduced. However, residual variance has been maintained along the time, thus it could be possible that residual variance has not a genetic component. These arguments have been discussed by Slaktin and Lande (1976), Zhang and Hill (2008) and Hill and Mulder (2010).

Nevertheless, there is some evidence that residual variance is under genetic control. Most of this evidence is indirect because it comes from data bases analyses and not from experiments designed to find the genetic part of the variance (SanCristóbal *et al.*, 2001; Sorensen and Waagepetersen, 2003; Ros *et al.*, 2004; Gutiérrez *et al.*, 2006; Ibáñez-Escriche *et al.*, 2008a,b,c; Wolc *et al.*, 2009 and Neves *et al.*, 2012). The models employed to analyze these data bases depend on a large number of parameters and they are not robust. For instance, Ye Yang *et al.* (2011) investigated how the scale of measurement affects inferences under heteroscedastic models; they compared genetic parameters of residual variance in an untransformed scale with the same data after a Box-Cox transformation, finding that the coefficient of correlation between genetic effects of the trait and its residual variance changed sign compared to the results in the untransformed scale.

Other evidence of the existence of a genetic component in residual variance comes from selection experiments. Garreau *et al.* (2008) performed a divergent selection experiment based on residual variance of birth weight in rabbits; but selection criterion and the estimates of the responses were dependent on the complex models quoted before.

Until now, no direct evidence of genetic control of residual variance has been offered. The objective of our experiment is to carry out a divergent selection on residual variance of litter size. Our selection criterion is the variance of the phenotypic records of litter size of the does. Then, residual variance is analyzed directly as an observed trait.

MATERIALS AND METHODS

Animals

An experiment of divergent selection was carried out during five generations. Each divergent line (high line (H) and low line (L)) had approximately 125 females and 25 males per generation. Data came from a total of 7285 parities from 1591 does. The animals of the base population were chosen at random between offspring of an F2 population derived from divergent lines selected for uterine capacity. The selection pressure on females was approximately 30% in each line. Males were chosen within sire families in order to avoid an increase of inbreeding.

The average number of parities per doe was 4.6. Rabbits used in this study were bred at the farm of the Universidad Miguel Hernández of Elche. Reproduction was organized in discrete generations. The does were first mated at 18 weeks of age, thereafter 10 d after parturition. They were under a constant photoperiod of 16:8 h and controlled ventilation.

Selection criterion

Selection was based on residual variance of litter size (V_e). This variance was calculated as the variance of litter size within doe, after precorrecting litter size for year-season and lactation status, with the following model:

$$LS_{ij} = YS_i + L_j + e_{ij} \quad (1)$$

Where LS_{ij} is the litter size of doe; YS is the effect of year-season with twenty three levels; L is the effect of lactation status with three levels (nulliparous, lactating and nonlactating does with more than one parity) and e is the residual. The systematic effects were estimated by least square means. Litter size data have been precorrected to avoid systematic effects that could affect the variance. Nevertheless,

residual variance of litter size without pre correction was also calculated (V_r). Variances V_r and V_e were calculated using the minimum quadratic risk estimator:

$$\frac{1}{n+1} \sum_{i=1}^n (x_i - \bar{x})^2$$

Where x_i is the litter size (or precorrected litter size) of parity i of a doe, and n is the total number of parities of the doe (n varying from 2 to 12).

Statistical analysis

The traits analyzed were: V_e , V_r , residual standard deviation of litter size (Sd_e) calculated as the square root of V_e , and litter size (LS) as number of total born. A Bayesian statistical analysis was performed.

Both V_e and V_r were analyzed with the model:

$$\mathbf{V}_e = \mathbf{X}\mathbf{b} + \varepsilon \quad (2)$$

Residual variance was assumed to be conditionally distributed as follows:

$$(\mathbf{V}_e | \mathbf{b}, \sigma_\varepsilon^2, \mathbf{K}) \sim N(\mathbf{X}\mathbf{b}, \mathbf{K}\sigma_\varepsilon^2)$$

Where \mathbf{b} is a vector that only included the line-generation effect (with eleven levels; base generation and high and low lines for each of the five generations), \mathbf{X} is the known incidence matrix, σ_ε^2 is the residual variance and \mathbf{K} is a diagonal matrix used for weighting the residuals according to the number of parities of each doe. The element in the diagonal was:

$$\frac{2(n-1)}{(n+1)^2}$$

Being n the number of parities of each doe. This weight comes from obtaining the variance of a minimum quadratic risk estimator of the variance (Searle, 1982). Sd_e was analyzed with the same model as V_e and V_r , using the identity matrix \mathbf{I} instead of \mathbf{K} . Bounded uniform priors were used to represent vague previous knowledge of \mathbf{b} and σ_ε^2 .

The model for LS was:

$$LS = Xb + Wp + \varepsilon$$

Litter size was assumed to be conditionally distributed as follows:

$$(LS|b, p, \sigma_\varepsilon^2) \sim N(Xb + Wp, I\sigma_\varepsilon^2)$$

$$p \sim N(\mathbf{0}, \sigma_p^2)$$

Where vector b included the effects of line-generation, year-season and lactation status, p includes the permanent effects, σ_p^2 and σ_ε^2 are the variances of permanent effects and residuals, respectively, X and W are the known incidence matrices and I is an identity matrix. The priors for b and for variances σ_ε^2 and σ_p^2 were bounded uniform.

Features of the marginal posterior distributions of differences between lines were estimated using Gibbs sampling. Results corresponding to models are based on MCMC runs consisting of 60,000 iterations, with a burn-in period of 10,000. Only one every 10 samples were saved for inferences. Convergence was tested using the Z criterion of Geweke and Monte Carlo sampling errors were computed using time-series procedures (Sorensen and Gianola, 2002). Software was designed to solve the models for V_e and V_r analyses, and the Rabbit programme was used to solve the model for Sd_e and LS analyses (Rabbit, 2012).

The parameters obtained from the marginal posterior distributions of the differences between lines were: the median of the marginal posterior distribution of the difference (D_{H-L}) in each generation; the highest posterior density region at 95% ($HPD_{95\%}$); the probability of the lines being different (probability of the difference between H and L lines being greater than zero when this difference is greater than zero, or the probability of the difference being lower than zero when this difference is lower than zero) (P) and the guaranteed value of a difference with a probability of 80% (limit of the interval $[k, +\infty)$ when the difference is greater than zero or the limit of the interval $(-\infty, k]$ when the difference is lower than zero, that is the minimum or maximum value that the variable can take, with a probability of 80%. Frequently, comparisons are carried out based on the differences between means, which in most cases is equivalent to use a guaranteed value of 50%. We used guaranteed values at 80% of probability which are more strictly.

RESULTS AND DISCUSSION

Table 1 summarizes the descriptive features of the traits in the base population. Both V_e and V_r showed high coefficients of variation, partially due to being asymmetric and heavy-tailed to the right. Notice that V_e and V_r means are similar, implying that precorrection has scarce effect in LS . Concerning to LS , mean and standard deviation were in accordance with other studies (Blasco *et al.*, 1993).

Table 1. Mean, median, standard deviation (Sd) and coefficient of variation (CV) of residual variance of litter size precorrecting data (V_e), residual variance of litter size without precorrection (V_r), residual standard deviation of litter size (Sd_e) and litter size (LS).

	Mean	Median	Sd	CV
V_e	3.73	2.72	3.36	0.90
V_r	3.96	3.13	3.55	0.90
Sd_e	1.74	1.65	0.84	0.48
LS	8.71	9	3.01	0.35

In all Bayesian analysis, Monte Carlo standard errors were small and lack of convergence was not detected by the Geweke test. Marginal posteriors distributions were approximately normal.

Figure 1A displays the plot of mean and variance of litter size without precorrecting data of each doe by quartiles. First quartile shows that V_r increases with the litter size average of the doe due to a scale effect, but fourth quartile illustrates that when the litter size average of the doe is high, V_r of the doe is low. Does with high litter size average should have a high litter size in all parities, thus their V_r should be small.

However, when we get away from the extremes, we found does with medium litter size average that are small, medium or highly variable. The same conclusions can be awarded to V_e (Figure 1B).

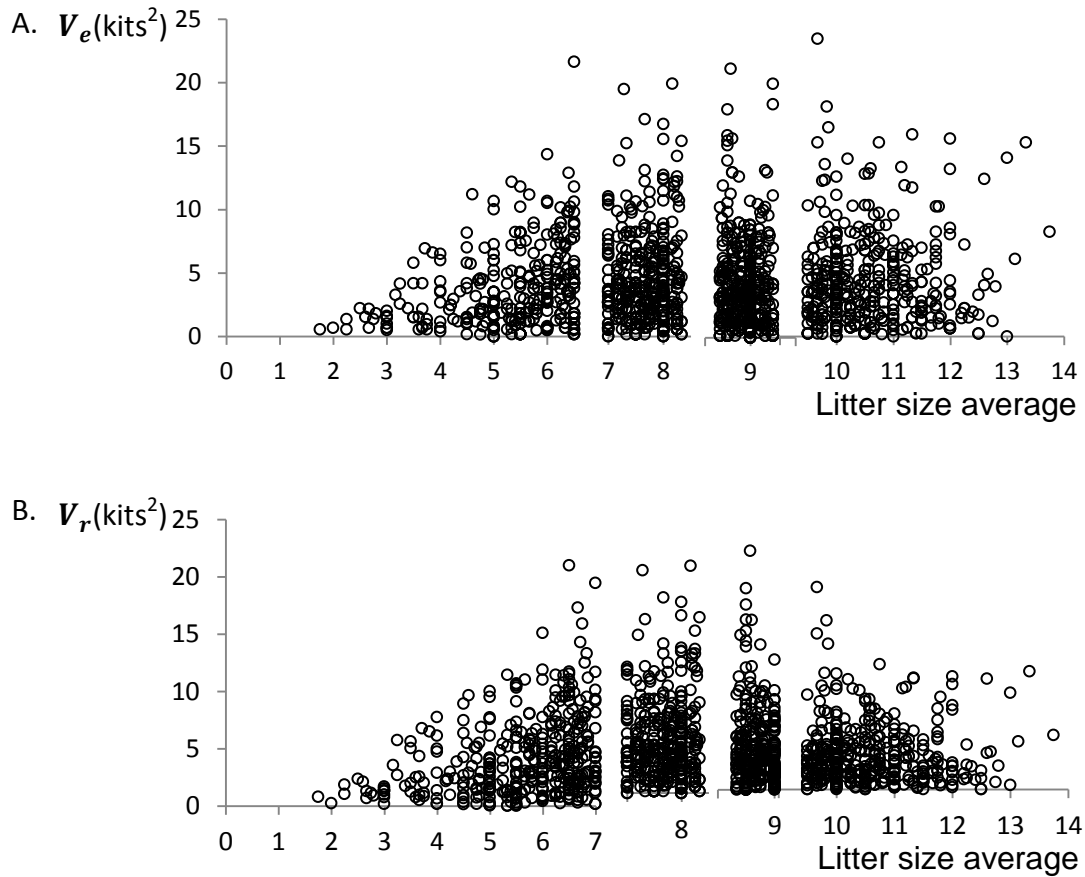


Figure 1. Data distribution by quartiles of (A) residual variance without precorrecting data (V_r) and (B) residual variance precorrecting data (V_e), with litter size average of all parities of each female.

Response to selection is plotted in Figure 2. Means and standard deviations of the estimated marginal posterior distributions for each line and generation are represented for V_e . A high response was obtained in the first generation, no appreciable response was obtained in the second generation, and a lower response was observed during the third, fourth and fifth generations, according to the selection differential applied, showed in Table 2. In divergent selection experiments, number of animals in the base generation approximately doubles the number of animals in the rest of lines, thus, a higher selection pressure can be applied, and the selection

differential is greater. In the last generation, V_e of line H was 37% higher than line L. Changes in the response are related to changes in the selection pressure and sampling errors.

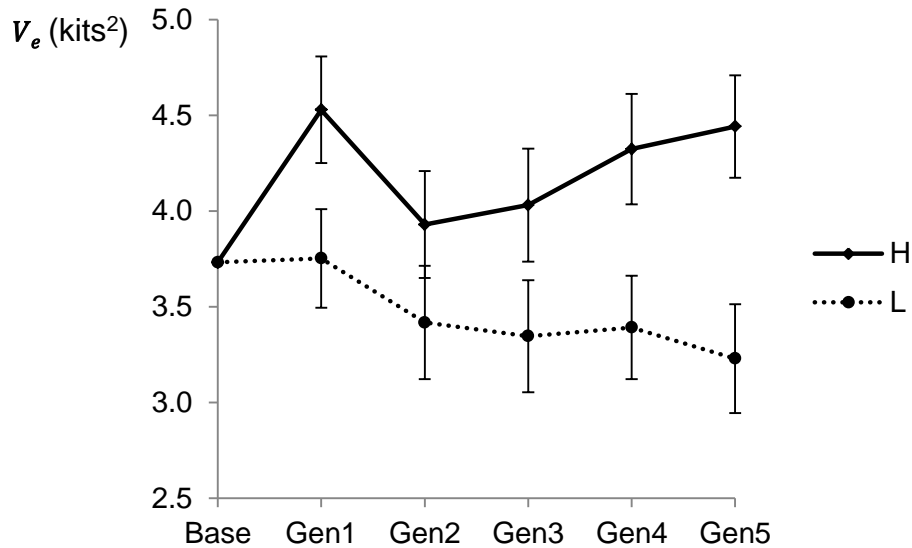


Figure 2. Means and standard deviations of the estimated marginal posterior distributions of each line in each generation for residual variance of litter size using precorrected data (V_e); H: high line; L: low line; Base: Base generation; Gen: generation.

Table 2. Weighted selection differential for residual variance of litter size precorrected for year-season and lactation status (V_e) in each generation, measured as kits².

Generation	High line	Low line
0	2.5	1.6
1	1.5	0.3
2	1.6	0.3
3	2.2	0.5
4	1.8	0.3

Table 3 shows that both lines at least differed in 0.87 kits² in generation five, with a probability of 80% ($k_{80\%}$). Notice, that despite of zero being inside the highest posterior density at 95% of probability (HPD_{95%}) of generations 2 and 3, the probability of the difference being greater than zero (P) is 0.90 and 0.95, respectively.

Table 3. Features of the estimated marginal posterior distributions of the differences in V_e between the high and the low lines selected for residual variance of litter size using precorrected data.

Generation	D_{H-L}	HPD _{95%}		P	$k_{80\%}$
1	0.78	0.03	1.50	0.98	0.45
2	0.51	-0.31	1.29	0.90	0.17
3	0.68	-0.14	1.47	0.95	0.33
4	0.93	0.17	1.73	0.99	0.61
5	1.21	0.44	1.96	1.00	0.87

D_{H-L} , median of the marginal posterior distribution of the difference between high and low lines, measured as kits².

HPD_{95%}, highest posterior density region at 95% of probability, measured as kits².

P , probability of the difference being greater than zero.

$k_{80\%}$, guaranteed value (limit of the interval $[k, +\infty)$ at 80% of probability, measured as kits²).

We assumed that V_e and V_r were normally distributed, which is not the case (Figure 3A). Nevertheless marginal posterior distributions tend to normality with enough data. A squared root normalizing transformation was used, obtaining Sd_e (Figure 3B), and the same conclusions on the response to selection can be drawn.

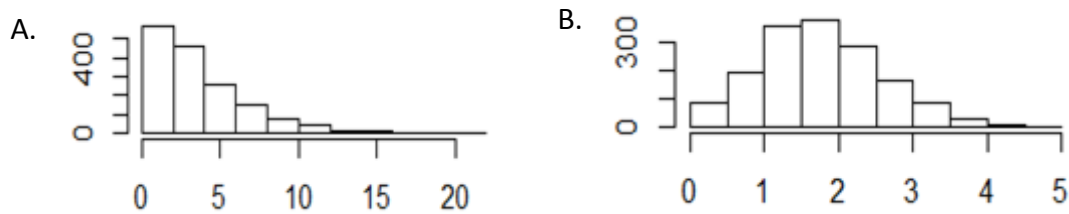


Figure 3. Histograms of the (A) residual variance of litter size data (V_e) and for the (B) residual standard deviation of litter size data (Sd_e).

Figure 4 plots the means and standard deviations of the estimated marginal posterior distributions of the line-generation effect for Sd_e . As before in V_e , it is shown that high response was obtained in the first generation and a lower response was observed during the fourth and fifth generations.

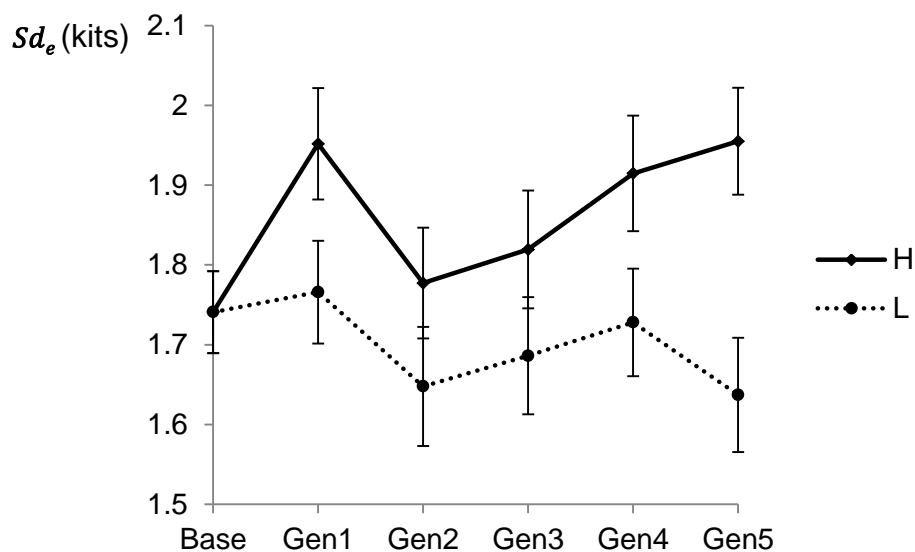


Figure 4. Means and standard deviations of the marginal posterior distribution of each line in each generation for residual standard deviation of litter size (Sd_e). H: high line; L: low line; Base: Base generation; Gen: generation.

Table 4 shows the features of the estimated marginal posterior distributions of the differences between H and L lines for Sd_e . The differences between lines in terms of Sd_e , in the last generation showed a guaranteed value at 80% ($k_{80\%}$) of 0.24 kits.

Table 4. Features of the estimated marginal posterior distributions of the differences in Sd_e between the high and the low lines selected for residual variance of litter size using precorrected data.

Generation	D_{H-L}	HPD _{95%}		P	$k_{80\%}$
1	0.19	0.00	0.37	0.98	0.11
2	0.13	-0.08	0.32	0.90	0.04
3	0.13	-0.07	0.33	0.91	0.04
4	0.19	-0.02	0.37	0.97	0.11
5	0.32	0.12	0.51	1.00	0.24

D_{H-L} , median of the marginal posterior distribution of the difference between high and low lines, measured as kits.

HPD_{95%}, highest posterior density region at 95% of probability, measured as kits.

P , probability of the difference being greater than zero.

$k_{80\%}$, guaranteed value (limit of the interval $[k, +\infty)$ at 80% of probability, measured as kits).

We assumed that precorrecting LS by systematic effects does not affect the genetic component of V_e . Besides, V_e was calculated with estimated residuals that should be correlated to each other. In order to draw conclusions independent on the model (equation 1), we decided to use V_r to examine how results were distorted by the pre-correction. Figure 5 plots the means and standard deviations of the estimated marginal posterior distributions of the line-generation effect for V_r . As before in V_e , high response was obtained in the first generation, no appreciable response was obtained in the second generation, and a lower response was observed during the

third, fourth and fifth generations. In generation five, V_r was 38% higher in line H than in line L.

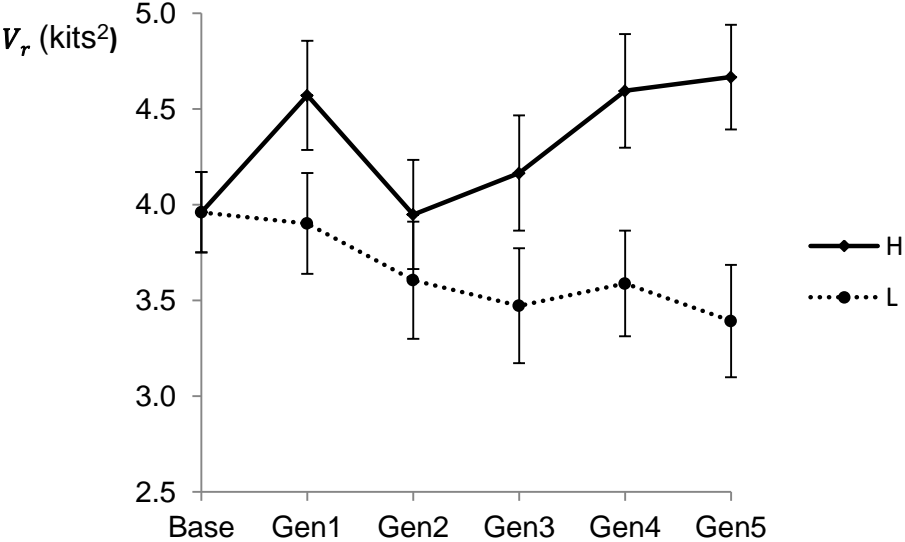


Figure 5. Means and standard deviations of the estimated marginal posterior distributions of each line in each generation for residual variance of litter size data without precorrection (V_r); H: high line; L: low line; Base: Base generation; Gen: generation.

Tables 5 shows the features of the estimated marginal posterior distributions of the differences between H and L line for V_r . As in V_e , difference between H and L line in the last generation was 0.86 with a probability of 80%. The mean of the marginal posterior distribution of the phenotypic correlation estimate between V_r and V_e was 0.97, thus the precorrection of the data has almost no effect.

Table 5. Features of the estimated marginal posterior distributions of the differences in V_r between the high and the low lines selected for residual variance of litter size using precorrected data.

Generation	D_{H-L}	HPD _{95%}		P	$k_{80\%}$
1	0.67	-0.09	1.41	0.96	0.34
2	0.35	-0.52	1.11	0.80	0.00
3	0.69	-0.16	1.50	0.95	0.33
4	1.01	0.17	1.77	0.99	0.67
5	1.19	0.40	1.96	1.00	0.86

D_{H-L} , median of the marginal posterior distribution of the difference between high and low lines, measured as kits².

HPD_{95%}, highest posterior density region at 95% of probability, measured as kits².

P , probability of the difference being greater than zero.

$k_{80\%}$, guaranteed value at 80% (limit of the interval $[k, +\infty)$ of probability, measured as kits²).

It is the first time that direct selection by residual variance of litter size is carried out, being treated as an observed trait. Results lead us to think that V_e is under genetic control, which would allow selection to improve litter size homogeneity in rabbits. Other experiment of selection by residual variance was carried out by Garreau *et al.* (2008). They performed a selection experiment by residual variance of birth weight within litter on rabbits. The selection criterion was the predicted genetic value of the model proposed by SanCristóbal in which the logarithm of the residual variance has environmental and genetic components (SanCristóbal *et al.*, 1998). The model was analyzed in two steps. In the first step, an animal model was fitted to the birth weight data. In the second step, the logarithm of the square of estimated residual values obtained in first step was fitted with a different animal model. A high response was found in the first generation, but no changes were found in the following four generations; nevertheless it should be noted that these estimates and the selection criterion were model dependent.

Heteroscedastic models had been largely employed to analyze data bases, in order to estimate genetic parameters that show evidence of a genetic factor under residual variance (SanCristóbal *et al.*, 2001 in litter size in sheep; Sorensen and Wagepatersen, 2003 in litter size in pig; Ros *et al.*, 2004 in adult weight in snails; Gutiérrez *et al.*, 2006 in litter size and litter weight in mice; Rowe *et al.*, 2006 and Wolc *et al.*, 2009 in body weight in broiler chickens; Ibáñez- Escriche *et al.*, 2008c, in uterine capacity in rabbits; Ibáñez- Escriche *et al.*, 2008b in slaughter weight in pigs). As well, Neves *et al.* (2012) analyzed log squared residuals associated with each observation according to an animal model, and suggested that residual variance could be decreased by selection, in production traits in beef cattle. Nevertheless, these models are not robust. For instance, transforming data by Box-Cox leads to different results in the estimate of the genetic correlation between litter size and its residual variance (Ye yang *et al.*, 2011).

Figure 6 plots the means and standard deviations of the estimated marginal posterior distributions of the line-generation effect for LS . Direct selection for V_e had an indirect negative response in LS . In the last generation, LS was 7% higher in line L than in line H.

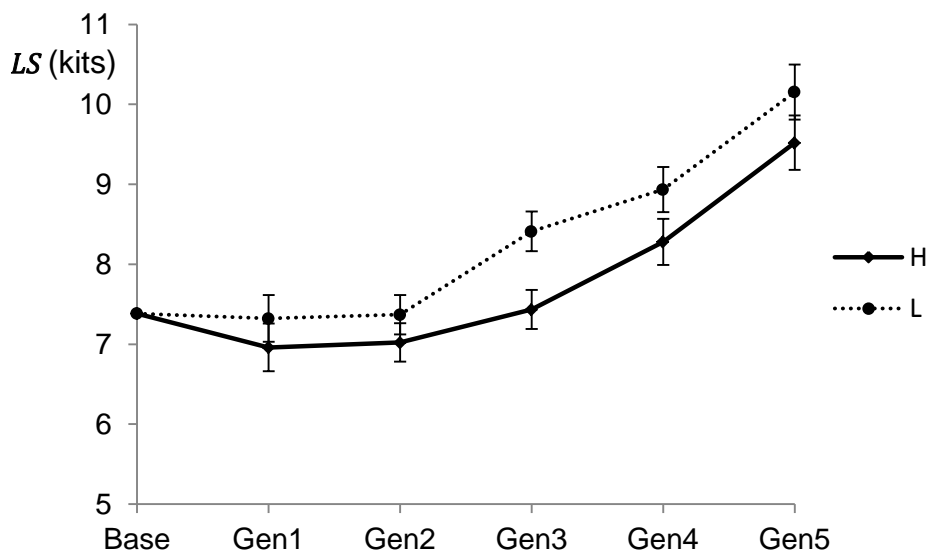


Figure 6. Means and standard deviations of the estimated marginal posterior distributions of each line in each generation for litter size (LS); H: high line; L: low line; Base: Base generation; Gen: generation.

Table 6 shows the features of the estimated marginal posterior distributions of the differences between H and L line for *LS*. At the end of the fifth generation, the difference between high and low lines was at least -0.43 kits with a probability of 80 % ($k_{80\%}$).

Table 6. Features of the estimated marginal posterior distributions of the differences in *LS* between the high and the low lines selected for residual variance of litter size using precorrected data.

Generation	D_{H-L}	$HPD_{95\%}$		P	$k_{80\%}$
1	-0.36	-0.79	0.07	0.95	-0.18
2	-0.35	-0.81	0.11	0.93	-0.15
3	-0.98	-1.43	-0.49	1.00	-0.77
4	-0.65	-1.09	-0.19	1.00	-0.45
5	-0.63	-1.09	-0.18	1.00	-0.43

D_{H-L} , median of the marginal posterior distribution of the difference between high and low lines, measured as kits.

$HPD_{95\%}$, highest posterior density region at 95% of probability, measured as kits.

P , probability of the difference being lower than zero.

$k_{80\%}$, guaranteed value (limit of the interval $(-\infty, k]$ at 80% of probability, measured as kits).

The line selected for low V_e showed a higher *LS* in all generations. The homogeneous does presented higher litter size than the heterogeneous line. This may be due to does presenting less environmental variance, should be more resistant to diseases and stress (García *et al.*, 2012), thus, these environmental factors affect less their production level. The correlation between the mean and the variance of a trait has been the goal of several studies, with different results. A negative correlation between the mean of a trait and its residual variance was detected by : Sorensen and Waagepetersen (2003) in litter size in pigs and confirmed by Felleki *et al.* (2012);

Gutiérrez *et al.* (2006) in litter size and litter weight at birth in mice.; Ibáñez-Escriche *et al.* (2008a) for weight gain in mice; Ibáñez-Escriche *et al.* (2008c) in uterine capacity in rabbits; Mulder *et al.* (2009) for body weight in broiler chicken and Rowe *et al.* (2005) analyzing 35-day body weights of broiler chickens. By contrast, Bolet *et al.* (2008) observed no effect on the mean weight of the young rabbits selecting by homogeneity of birth weight. Ibáñez-Escriche *et al.* (2008b) detected an absence of correlation, or small negative correlation between mean and variance genetic effects affecting slaughter weight in pigs. Finally, Ros *et al.* (2004) provide a strong indication of a positive correlation between mean and environmental variation of adult body weight in snails, and Wolc *et al.* (2009) gauged positive correlations between body weight and body conformation in broiler chickens. Nevertheless, these results have been obtained analyzing heteroscedastics models, complex and not robust, and all these correlations have been questioned as artifacts of the model (Ye Yang *et al.*, 2011). These last authors obtained that the genetic correlation coefficient, which was negative when was estimated with untransformed data, changed to positive when was estimated with transformed data (data were transformed to correct the lack of normality of residues). In our case, the correlation between LS and V_e was negative, and the observation of Ye Yang *et al.* (2011) cannot be applied.

CONCLUSIONS

It is the first time that direct selection by residual variance of litter size is carried out, treating residual variance as an observed trait. An experimental evidence lead us to think that residual variance of litter size is under genetic control, and is feasible to reduce the residual variance by selection. A negative indirect response was found in the mean on the trait, the homogeneous does showing a higher litter size than the heterogeneous does.

ACKNOWLEDGEMENTS

Special thanks for the cooperation of Raquel Muelas Domingo in the experiment.

REFERENCES

- Blasco, A., Bidanel, J.P., Bolet G., Haley C., and Santacreu, M.A. 1993. The genetics of prenatal survival of pigs and rabbits: a review. *Livestock production Science* 37 : 1-21.
- Blasco, A. 1996. Genetics of litter size and does fertility in the rabbit. *6th World Rabbit Congress, Toulouse (France) 2*: 219-227.
- Bolet, G., Garreau, H., Hurtaud, J., Saleil, G., Esparbié, J., Falieres, J., Theau-Clément, M. and Bodin, L. 2008. Canalising selection on within litter variability of birth weight in rabbits: responses to selection and characteristics of the uterus of the does. *8th World Rabbit Congress- Verona (Italy)*.
- Falconer, D.S. and Mackay T.F.C. 1996. *Introduction to quantitative genetics*. 4th ed. Edinburgh Gate, Harlow, U.K. Addison Wesley Longman Limited.
- Felleki, M., Lee, D., Lee, Y., Gilmour, A.R. and Rönnegard, L. 2012. Estimation of breeding values for mean and dispersion, their variance and correlation using double hierarchical generalized linear models. *Gen. Res. Camb.* 94: 307-317.
- García, M.L., Argente, M.J., Muelas, R., Birlanga, V. and Blasco, A. 2012. Effect of divergent selection for residual variance of litter size on health status and welfare. *10th World Rabbit Congress-Sharm El Sheikh-Egypt*.
- Garreau, H., Bolet, G., Larzul, C. Robert-Graine, C., Saleil, G., SanCristóbal, M. and Bodin, L. 2008. Results of four generations of a canalising selection for rabbit birth weight. *Livestock Science* 118: 55-62.
- Gutiérrez, J.P., Nieto, B., Piqueras, P., Ibáñez, N. and 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. and Mulder, H.A. 2010. Genetic analysis of environmental variation.» *Genet. Res. Camb.* 92: 381-395.
- Ibáñez-Escriche, N., Moreno, A., Nieto, B., Piqueras, P., Salgado C. and Gutiérrez 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., Varona, L., Sorensen, D. and Noguera J.L. 2008b. A study of heterogeneity of environmental variance for slaughter weight in pigs. *Animal* 2 :19-26.

- Ibáñez-Escriche, N., Sorensen D. and Blasco., A. 2008c. A study of enviromental variance genetic control for uterine capacity in rabbits. *9th World Rabbit Congress- Verona(Italy)*.
- Larzul, C., Le Roy, P., Tribout, T.,Gogue, J. and SanCristóbal, M. 2006.Canalizing selection on ultimate pH in pigs: consequences on meat quality.*8th World Rabbit Congress on Genetics Applied to Livestock Production, Belo Horizonte, MG, Brasil*.
- Mulder, H.A., Hill, W.G., Vereijken, A. and Veerkamp, R.F. 2009. Estimation of genetic variation in residual variance in female and male broiler chickens. *Animal* 3: 1673-1680.
- Neves, H.H.R., Carneiro, R. and Queiroz S.A. 2012. Genetic and environmental heterogeneity of residual variance of weight traits in Nellore beef cattle. *Genet. Sel. Evol.* 44: 19.
- Rabbit programme. 2012. <http://www.dcam.upv.es/dcia/ablasco/Publi.htm>
- Ros, M., Sorensen, D., Waagepetersen, R., Dupon-Nivet, M., SanCristóbal, M., Bonnet, J.C. and Mallard, J. 2004. Evidence for genetic control of adult weight plasticity in the snail *Helix aspersa*.*Genetics* 168: 2089-2097.
- Rowe, S.J., White, I.M.S., Avedano, S and Hill, W.G. 2006.Genetic heterogeneity of residual variance in broiler chickens. *Genet. Sel. Evol* 38: 617-635.
- SanCristóbal, M., Bodin, L., Elsen, J.M. and Chevalet, C. 2001. Genetic components of litter size variability in sheep. *Genet. Sel. Evol* 33: 249-271.
- SanCristóbal, M., Elsen, J.M., Bodin, L., and 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.
- Searle, R. 1982.*Matrix algebra useful for statistics*. Canada: John Wiley and Sons, Ind.
- Slatkin, M. and Lande, R. 1976. Niche width in a fluctuating environment-density independent model. *American Naturalist* 110: 31-55.
- Sorensen, D. and Gianola, D. 2002. *Likelihood, Bayesian and MCMC Methods in Quantitative Genetics*. New York, USA: Springer.
- Sorensen, D. and Waagepetersen, R. 2003. Normal linear models with genetically structured residual variance heterogeneity: a case study. *Genet. Res. Camb.* 82 : 207-222.

- Wolc, A., White, I.M.S., Avedano, S. and Hill, W.G. 2009. Genetic variability in residual variation of body weight and conformation scores in broiler chickens. *Poultry Science* 88: 1156-1161.
- Ye Yang, Ole F. Christensen, Daniel Sorensen. 2011. Analysis of a genetically structured variance heterogeneity model using the Box-Cox transformation. *Gen. Res. Cam.* 93 : 33-46.
- Zhang, X. and Hill, G. 2008. Mutation-Selection balance for environmental variance. *The American Naturalist* 171: 394-399.

**ARTICLE 2: GENETIC TRENDS IN A DIVERGENT SELECTION
EXPERIMENT BY RESIDUAL VARIANCE OF LITTER SIZE IN
RABBITS.**

Previously submitted to the XV Jornadas sobre Producción Animal of AIDA (Zaragoza, May 2013). This version has been translated and modified, and it will be submitted to a journal after small modifications.

GENETIC TRENDS IN A DIVERGENT SELECTION EXPERIMENT BY RESIDUAL VARIANCE OF LITTER SIZE IN RABBITS.

ABSTRACT

A divergent selection experiment for residual variance of litter size was carried out in rabbits. The selection criterion was the residual variance of litter size (V_e), after precorrecting litter size for the effects of generation and lactation status. Analyzed traits were: V_e , residual variance of litter size without precorrection (V_r), residual standard deviation of litter size with precorrected data (Sd_e) or without precorrection (Sd_r), and litter size (LS). Bayesian methods were used to analyze the response obtained in the first five generations. The response obtained on V_e was asymmetrical, showing the high line a greater response. The estimated genetic means for V_e and V_r in the fifth generation were 0.45 kits² and 0.47 kits² in the high line and -0.24 kits² and -0.26 kits² in the low line, respectively, thus precorrection had almost no effect on the response. An asymmetric negative correlated response was observed in litter size (LS) for homogeneous line, which showed an estimated genetic mean in the fifth generation of 0.66 kits, and no response was observed in heterogeneous line. Estimates of heritability for V_e and V_r were low, 0.06 and 0.05 respectively, thus responses could be due to the high variability of the traits.

Keywords: Residual variances, rabbits, litter size.

INTRODUCTION

Several studies proposed that residual variance can be under genetic control (SanCristóbal *et al.*, (2001) in litter size in sheeps; Sorensen and Waagepetersen, (2003) in litter size in pigs; Bolet *et al.*, (2008) in birth weight in rabbits; Ibáñez-Escriche *et al.*, (2008) in uterine capacity in rabbits; Gutiérrez *et al.*, (2006) in birth weight in mice; Ros *et al.*, (2004) in adult weight in snails; Wolc *et al.*, (2009) in body weight and conformation in broiler chickens; Neves *et al.*, (2012) in weight traits in beef cattle). Nevertheless, these studies are based in models that depend on a large number of parameters. Moreover, the models are not robust. For instance, even a

slight lack of normality in residuals can lead to highly different results (Ye Yang *et al.*, 2011). Direct selection by residual variance would help to resolve this question because it does not depend (or is less depending) on the modeling of the trait. For this purpose, a divergent selection experiment of direct selection on residual variance of litter size is performed. A limitation of divergent selection experiments is that response can be asymmetrical. In the first article of this thesis, response was estimated as the difference between the means of the lines, and the response in each line was not estimated. Moreover, the precision of the estimation of the response by differences between lines is lower than the response obtained by estimating genetic means applying an animal model. The aim of this study is to estimate the selection response of residual variance of litter size in a divergent selection experiment in rabbits using an animal model.

MATERIAL AND METHODS

Data came from 1591 does of a divergent selection experiment by residual variance of litter size (V_e), estimated as the variance of litter size within doe. Litter size data were precorrected by year-season and lactation status. Nevertheless, residual variance without precorrection was also calculated (V_r). Variances V_r and V_e were calculated using the minimum quadratic risk estimator:

$$\frac{1}{n+1} \sum_{i=1}^n (x_i - \bar{x})^2$$

Where x_i is the litter size (or precorrected litter size) of parity i of a doe, and n is the total number of parities of the doe (n varying from 2 to 12). Animals were bred at the farm of Universidad Miguel Hernández de Elche. Each line had 125 females and 25 males per generation, approximately. The average number of parities per doe was 4.6. The selection pressure on females was around 30% in each line. Males were chosen within sire families in order to avoid an inbreeding increase. The traits analyzed were: V_e , V_r , residual standard deviation of litter size with precorrected data (Sd_e) or without precorrection (Sd_r), both estimated as the square root of their respective variances and litter size (LS). A Bayesian statistical analysis was performed.

To estimate genetic trends of V_e , the following univariate model was fitted:

$$V_{e_i} = \mu + a_i + \varepsilon_i$$

Where V_{e_i} is the residual variance of litter size of doe i , μ is the mean of the trait, a is the genetic value of the doe and ε is the residual of the model. No environmental factors were included in the model because the variances are calculated with data that are taken along a large period of time. V_e was assumed to be conditionally distributed as follows:

$$(V_e | \mu, a, \sigma_\varepsilon^2) \sim N(\mu + Z\mathbf{a}, K\sigma_\varepsilon^2)$$

Where μ is a vector that only included the mean of the trait and \mathbf{a} is the vector of genetic effects. As residual variance of litter size of each doe was not calculated with the same number of data for all does, it was considered that residues of the model were normally distributed with mean 0 and variance $K\sigma_\varepsilon^2$, where σ_ε^2 is the variance of the residues of the model and K is a diagonal weight matrix with the following element in the diagonal:

$$\frac{2(n-1)}{(n+1)^2}$$

Being n the number of parities of each doe. This weight comes from obtaining the variance of a minimum quadratic risk estimator of the variance (Searle, 1982). Genetic effects were assumed to be normally distributed, as follows:

$$\mathbf{a} \sim N(\mathbf{0}, A\sigma_a^2)$$

Where A is the known genetic relationship matrix and σ_a^2 is the genetic variance. Trait V_r was analyzed with the same model. Analyses of Sd_e and Sd_r , and the estimates of heritabilities of V_e and V_r were also carried out with the same model, but with the identity matrix I instead of K .

To estimate genetic trends of LS , the following univariate model was fitted:

$$LS_{ijk} = YS_i + L_j + a_k + p_k + \varepsilon_{ijk}$$

Where LS_{ijkl} is the litter size of doe k , YS is the effect of year-season (23 levels), L is the lactation status (3 levels: nulliparous, lactating and non-lactating females), a includes the genetic value of the doe, p includes the common permanent effects and ε is the residual of the model. Litter size was assumed to be conditionally distributed as follows:

$$(LS|b, a, p, \sigma_{\varepsilon}^2) \sim N(Xb + Za + Wp, I\sigma_{\varepsilon}^2)$$

Where b is a vector that included year-season and lactation status effects, a is the vector that included the genetic effects, p is the vector of common permanent effects and σ_{ε}^2 is the variance of the residues of the model. Genetic effects were assumed to be normally distributed, as follows:

$$a \sim N(\mathbf{0}, A\sigma_a^2)$$

Where A is the known genetic relationship matrix and σ_a^2 is the genetic variance. Permanent effects were assumed to be normally distributed as:

$$p \sim N(\mathbf{0}, I\sigma_p^2)$$

Where σ_p^2 is the permanent effects variance.

Bounded uniform priors were employed for all effects and variances. Features of the marginal posterior distributions were estimated using Gibbs sampling. Results corresponding to models were based on MCMC runs consisting of 1,000,000 iterations, with a burn-in period of 200,000. Only one every 100 samples were saved for inferences, in order to avoid high correlations between consecutive samples. Convergence was tested using Z criterion of Geweke and Monte Carlo sampling errors were computed using time-series procedures (Sorensen y Gianola, 2002). Software was designed to solve the models for V_e and V_r analyses, programme TM by Legarra *et al.* (2008) was used to solve the model for Sd_e , Sd_r and LS analyses and the Rabbit programme was used for inferences in the MCMC chains (Rabbit, 2012).

RESULTS AND DISCUSSION

All Monte Carlo standard errors were small and lack of convergence was not detected by the Geweke test.

Response to selection was estimated as the mean of the genetic values of the animals in each line and generation. Figure 1 plots the features of the marginal posterior distributions of the response on V_e in each generation. Response on V_e was found in every generation, and it was asymmetrical, showing the high line a greater response. The estimated genetic means for V_e in the fifth generation were 0.45 kits² in the high line and -0.24 kits² in the low line. This asymmetric response could be partially attributed to natural selection favoring animals more resistant to diseases and stress, implying more homogeneous animals. Some evidence in this direction was provided by García *et al.* (2012) that found an association between concentration of biomarkers of health status and variability of litter size in rabbits. Does with higher litter size variability had poorer health status than does with lower litter size variability.

There are other experiments designed to find evidences of a genetic component in residual variance. Garreau *et al.* (2008) performed a selection experiment by residual variance of birth weight within litter on rabbits. The selection criterion was the predicted genetic value of the model proposed by SanCristóbal in which the logarithm of the residual variance has environmental and genetic components (SanCristóbal *et al.*, 1998). As commented previously in the literature review and in article 1 of this thesis, no consistent results were obtained. Only in the first generation was observed response, but the estimates of response were dependent on complex models.

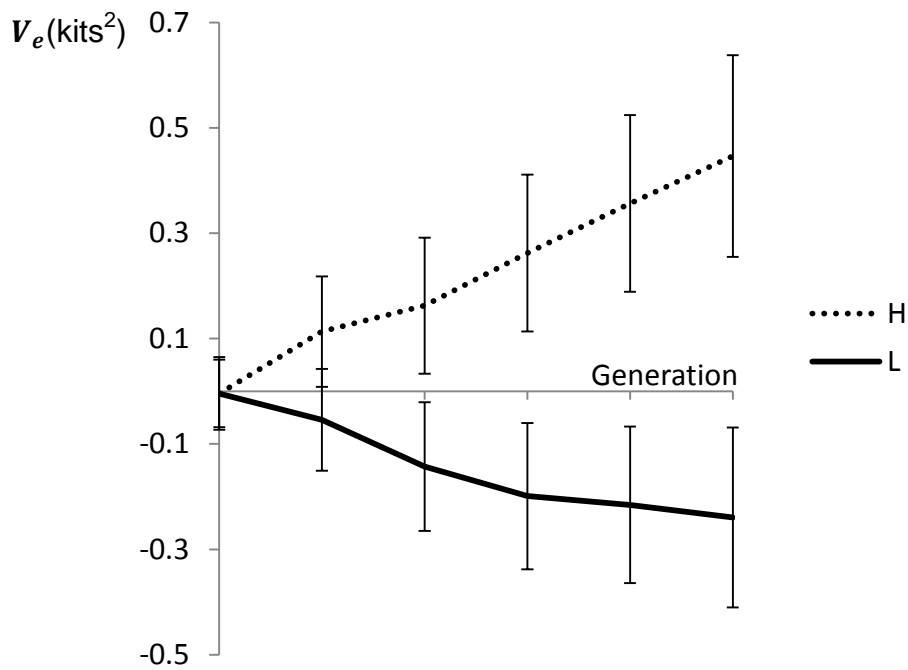


Figure 1. Means and standard deviations of marginal posterior distributions of the responses obtained in each line and generation for residual variance of litter size, precorrecting data (V_e). H: high line. L: low line.

Figure 2 shows the features of the marginal posterior distributions of the responses on V_r obtained in each generation and line. The estimated genetic means for V_r in the fifth generation were 0.47 kits² in the high line and -0.26 kits² in the low line. Comparing responses obtained for V_e and V_r it is noticed that both are closely correlated, thus it is concluded that the precorrection of the data has almost no effects. This is in accordance with previous conclusions in article 1 of this thesis.

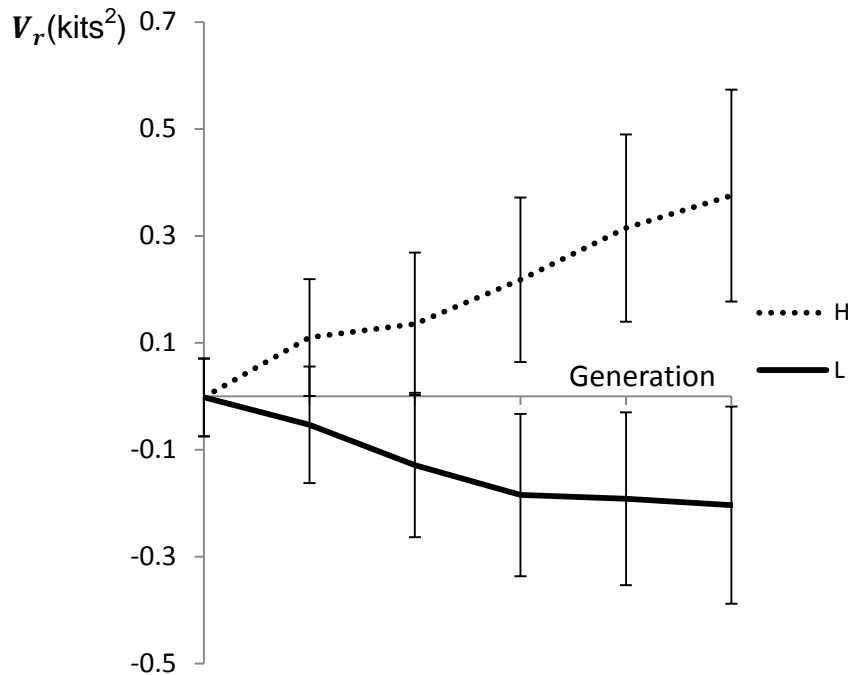


Figure 2. Means and standard deviations of marginal posterior distributions of the responses obtained in each line and generation for residual variance of litter size, without precorrecting data (V_r). H: high line. L: low line.

We assumed that V_e and V_r were normally distributed, which is not the case (see Figure 3 of article 1 of this thesis). Nevertheless marginal posterior distributions tend to normality with enough data. A squared root normalizing transformation was used obtaining Sd_e and Sd_r respectively, and the same conclusions on the response to selection can be drawn.

Figure 3 shows the features of the marginal posterior distributions of the responses on LS obtained in each generation and line. A negative indirect response was found for LS in the homogeneous line, which showed an estimated genetic mean in the fifth generation of 0.66 kits. Litter size increased in the heterogeneous line in a more irregular way. This is coherent with the conclusions obtained in the first article of this thesis. This may be due to the homogeneous does being more resistant to diseases and stress (García *et al.*, 2012) thus their litter size is less penalized. In our experiment, homogenous does had greater litter sizes in almost all their parities. Litter size homogeneity can also occur when LS has a small average, but is not our

case. Heterogeneous does had heterogeneous litter sizes, which could partially explain the lack of correlated response in LS when selecting by high variance of litter size.

Ibáñez-Escriche *et al.* (2008) estimated a negative genetic correlation between uterine capacity and its residual variance in rabbits, but Ye Yang *et al.* (2011), making a Box-Cox transformation to normalize the residues, observed that changed sign. These results have been discussed as an artifact of the model that as said before, is not robust. In our case, results agree with the existence of a negative genetic correlation between the mean and the variance, and we are not affected by the observation of Ye Yang *et al.* (2011).

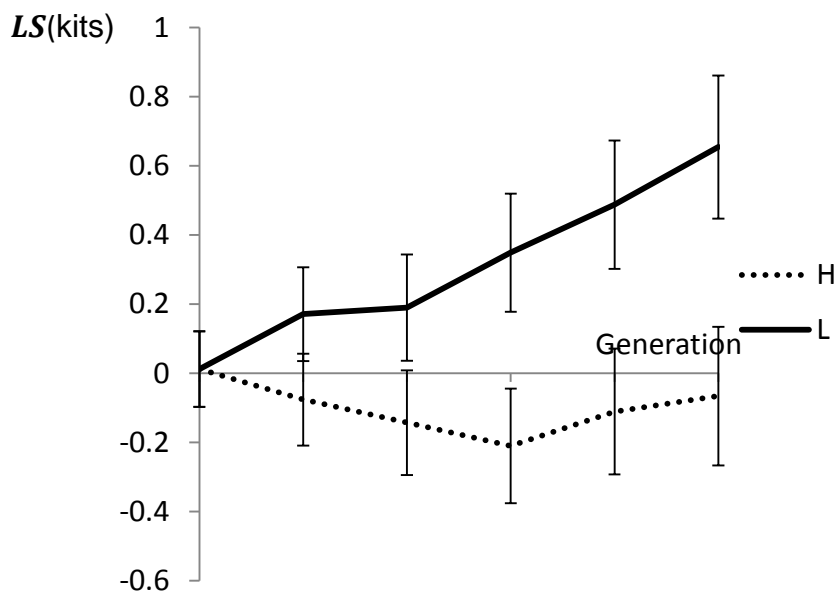


Figure 3. Means and standard deviations of marginal posterior distributions of the responses obtained in each line and generation for litter size (LS). H: high line. L: low line.

Table 1 shows the features of the marginal posterior distributions of the heritabilities for the analyzed traits. Estimates of heritability for V_e and V_r were low, 0.06 and 0.05 respectively, thus responses in V_e and V_r should be due to the high variability of the traits (see Table 1 of article 1 of this thesis). Low estimates of heritabilities were also found by other authors in variability traits. In mice, Gutiérrez *et al.* (2006) estimated

low heritabilities for residual variability of litter size and litter weight. In pigs, Damgaard *et al.* (2003) estimated the heritability for within-litter standard deviation in birth weight and weight at 3 weeks and they were low, 0.08 and 0.06 respectively. Heritability for *LS* was 0.12 and was in accordance with the litter size heritabilities found in the literature (Blasco *et al.*, 1993).

Table 1. Heritabilities for traits V_e , V_r and *LS*.

	M	HPD(95%)
V_e	0.06	[0.01, 0.11]
V_r	0.05	[0.01,0.10]
<i>LS</i>	0.12	[0.08,0.17]

V_e : residual variance of litter size precorrecting data. V_r : residual variance of litter size without precorrecting data. *LS*: litter size. M: median of the marginal posterior distribution. HPD(95%): highest posterior density region at 95% of probability.

CONCLUSIONS

An asymmetric response was observed after selecting directly by residual variance of litter size, showing the heterogeneous line a greater response. An asymmetric negative correlated response in litter size was observed when selecting by homogeneity of litter size. From a productive point of view, is interesting that homogeneity is accompanied by an increase of litter size.

REFERENCES

Blasco, A., Bidanel, J.P., Bolet G., Haley C., and Santacreu, M.A. 1993. The genetics of prenatal survival of pigs and rabbits: a review. *Livestock production Science* 37 : 1-21.

- Bolet, G., Garreau, H., Hurtaud, J., Saleil, G., Esparbié, J., Falieres, J., Theau-Clément, M. and Bodin, L. 2008. Canalising selection on within litter variability of birth weight in rabbits: responses to selection and characteristics of the uterus of the does. *8th World Rabbit Congress- Verona(Italy)*.
- Damgaard, L.H., Rydhmer, L., Lovendahl, P. and Grandison, K. 2003. Genetic parameters for within-litter variation in piglet birth weight and change in within-litter variation during suckling. *Journal of Animal Science* 81: 604-610.
- García, M.L., Argente, M.J., Muelas, R., Birlanga, V. and Blasco, A. 2012. Effect of divergent selection for residual variance of litter size on health status and welfare. *10th World Rabbit Congress-Sharm El Sheikh-Egypt*.
- Garreau, H., Bolet, G., Larzul, C. Robert-Graine, C., Saleil, G., SanCristóbal, M. and Bodin, L. 2008. Results of four generations of a canalising selection for rabbit birth weight. *Livestock Science* 118: 55-62.
- Gutiérrez, J.P., Nieto, B., Piqueras, P., Ibáñez, N. and 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.
- Ibáñez-Escriche, N., Sorensen D. and Blasco., A. 2008. A study of enviromental variance genetic control for uterine capacity in rabbits. *9th World Rabbit Congress- Verona(Italy)*.
- Legarra, A., L. Varona, and E. López de Maturana. 2008. TM Threshold Model. Available at <http://snp.toulouse.inra.fr/~alegarra/manualtm.pdf> [accessed Aug. 3, 2011; verified January 7, 2013]. GenoToul Bioinformatics, Toulouse, France.
- Neves, H.H.R., Carneiro, R. and Queiroz S.A. 2012. Genetic and environmental heterogeneity of residual variance of weight traits in Nellore beef cattle. *Genet. Sel. Evol.* 44: 19.
- Rabbit programme. 2012. <http://www.dcam.upv.es/dcia/ablasco/Publi.htm>
- Ros, M., Sorensen, D., Waagepetersen, R., Dupon-Nivet, M., SanCristóbal, M., Bonnet, J.C. and Mallard, J. 2004. Evidence for genetic control of adult weight plasticity in the snail *Helix aspersa*. *Genetics* 168: 2089-2097.
- SanCristóbal, M., Bodin, L., Elsen, J.M. and Chevalet, C. 2001. Genetic components of litter size variability in sheep. *Genet. Sel. Evol* 33: 249-271.
- SanCristóbal, M., Elsen, J.M., Bodin, L., and 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.

- Searle, R. 1982. *Matrix algebra useful for statistics*. Canada: John Wiley and Sons.
- Sorensen, D. and Gianola, D. 2002. *Likelihood, Bayesian and MCMC Methods in Quantitative Genetics*. New York, USA: Springer.
- Sorensen, D. and Waagepetersen, R. 2003. Normal linear models with genetically structured residual variance heterogeneity: a case study. *Genet. Res. Camb.* 82 : 207-222.
- Wolc, A., White, I.M.S., Avedano, S. and Hill, W.G. 2009. Genetic variability in residual variation of body weight and conformation scores in broiler chickens. *Poultry Science* 88: 1156-1161.
- Ye Yang, Ole F. Christensen, Daniel Sorensen. 2011. Analysis of a genetically structured variance heterogeneity model using the Box-Cox transformation. *Gen. Res. Cam.* 93 : 33-46.

GENERAL DISCUSSION AND CONCLUSIONS

Direct selection by residual variance of litter size has been carried out for the first time, treating residual variance as an observed trait. Rabbit is a good model for the studies since they have a short generation interval, their litter size is similar to pigs, and litter size can be measured several times in the same animal.

The obtained response is an experimental evidence of residual variance of litter size being under genetic control. It is feasible to reduce the residual variance by selection. Response was asymmetrical, showing the heterogeneous line a greater response. This asymmetry could be partially attributed to natural selection selecting by resistance to diseases and stress, implying more homogeneous animals.

Although residual variance it is not normally distributed, results in the response are not affected by using models that assume normality of the traits.

The pre correction of the litter data to calculate the selection criterion residual variance of litter size had almost no effect.

An asymmetric negative indirect response was found in litter size, homogeneous does showing a higher litter size. From a productive point of view, is interesting that homogeneity of litter size is accompanied by an increase of litter size. This may be due to more resistance to diseases and stress of homogeneous does thus they litter size is less penalized.

Conclusions obtained in articles 1 and 2 were in coherence:

1. Experimental evidence of a genetic component under residual variance of litter size is given. It was obtained an asymmetric response, showing the heterogeneous line a greater response.
2. An asymmetric negative correlated response was obtained in litter size, showing the homogeneous does higher litter sizes.

REFERENCES

- Blasco, A. 1996. Genetics of litter size and does fertility in the rabbit. *6th World Rabbit Congress, Toulouse (France) 2*: 219-227.
- Bodin, L., Robert-Granié, C., Larzul, C., Allain, D., Bolet, G., Elsen, J.M., Garreau, H., de Rochambeau, H., Ros, M. and SanCristobal, M. 2002. Twelve remarks on canalisation in livestock production. *7th World Congress on Genetics Applied to Livestock Production (2002)*.
- Bolet, G., Garreau, H., Hurtaud, J., Saleil, G., Esparbié, J., Falières, J., Theau-Clément, M. and Bodin, L. 2008. Canalising selection on within litter variability of birth weight in rabbits: responses to selection and characteristics of the uterus of the does. *8th World Rabbit Congress-Verona (Italy)*.
- Damgaard, L.H., Rydhmer, L., Lovendahl, P. and Grandison, K. 2003. Genetic parameters for within-litter variation in piglet birth weight and change in within-litter variation during suckling. *Journal of Animal Science* 81: 604-610.
- Falconer, D.S. and Mackay T.F.C. 1996. *Introduction to quantitative genetics*. 4th ed. Edinburgh Gate, Harlow, U.K. Addison Wesley Longman Limited.
- Felleki, M., Lee, D., Lee, Y., Gilmour, A.R. and Rönnegard, L. 2012. Estimation of breeding values for mean and dispersion, their variance and correlation using double hierarchical generalized linear models. *Gen. Res. Camb.* 94: 307-317.
- Fina, M., Ibáñez-Escriche, N., Piedrafita, J. and Casellas, J. 2013. Canalization analysis of birth weight in Bruna dels Pirineus beef cattle. *Journal of Animal Science* 91:3070-3078; published ahead of print May 8, 2013, doi:10.2527/jas.2012-5675
- García, M.L., Argente, M.J., Muelas, R., Birlanga, V. and Blasco, A. 2012. Effect of divergent selection for residual variance of litter size on health status and welfare. *10th World Rabbit Congress-Sharm El Sheikh-Egypt*.
- Garreau, H., Bolet, G., Larzul, C., Robert-Graine, C., Saleil, G., SanCristóbal, M. and Bodin, L. 2008. Results of four generations of a canalising selection for rabbit birth weight. *Livestock Science* 118: 55-62.

- Gavrilets, S. and Hastings, A. 1994. A quantitative-genetic model for selection on developmental noise. *Evolution* 48: 1478-1486.
- Gutiérrez, J.P., Nieto, B., Piqueras, P., Ibáñez, N. and 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.
- Ibáñez-Escriche, N., García, M. and Sorensen, D. 2010. GSEVM v.2: MCMC software to analyze genetically structured environmental variance models. *J. Anim. Breed. Genet.* 127:249-251.
- Ibáñez-Escriche, N., Moreno, A., Nieto, B., Piqueras, P., Salgado C. and Gutiérrez 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., Varona, L., Sorensen, D. and Noguera J.L. 2008b. A study of heterogeneity of environmental variance for slaughter weight in pigs. *Animal* 2 :19-26.
- Ibáñez-Escriche, N., Sorensen D. and Blasco., A. 2008c. A study of environmental variance genetic control for uterine capacity in rabbits. *9th World Rabbit Congress- Verona(Italy)*.
- Larzul, C., Le Roy, P., Tribout, T., Gogue, J. and SanCristóbal, M. 2006. Canalizing selection on ultimate pH in pigs: consequences on meat quality. *8th World Rabbit Congress on Genetics Applied to Livestock Production, Belo Horizonte, MG, Brasil*.
- Mulder, H.A., Bijma, P. and Hill, W.G. Prediction of breeding values and selection responses with genetic heterogeneity of environmental variance. *Genetics* 175: 1895-1910.
- Mulder, H.A., Hill, W.G., Vereijken, A. and Veerkamp, R.F. 2009. Estimation of genetic variation in residual variance in female and male broiler chickens. *Animal* 3: 1673-1680.
- Mulder, H.A., Bijma, P. and Hill, W.G. 2008. Selection for uniformity in livestock by exploiting genetic heterogeneity of residual variance. *Genet. Sel. Evol.* 40: 37-59.

- Neves, H.H.R., Carneiro, R. and Queiroz S.A. 2012. Genetic and environmental heterogeneity of residual variance of weight traits in Nelore beef cattle. *Genet. Sel. Evol.* 44: 19.
- Pun, A., Cervantes, B., Nieto, C., Salgado M.A., Perez-Cabal, N., Ibáñez-Escriche, N., and Gutiérrez J.P. 2012. Genetic parameters for birthweight environmental variability in mice.» *Journal of Animal Breeding and Genetics*. Article first published online : 19 DEC 2012, DOI: 10.1111/jbg.12021.
- Ros, M., Sorensen, D., Waagepetersen, R., Dupon-Nivet, M., SanCristóbal, M., Bonnet, J.C. and Mallard, J. 2004. Evidence for genetic control of adult weight plasticity in the snail *Helix aspersa*. *Genetics* 168: 2089-2097.
- Rowe, S.J., White, I.M.S., Avedano, S and Hill, W.G. 2006. Genetic heterogeneity of residual variance in broiler chickens. *Genet. Sel. Evol* 38: 617-635.
- SanCristóbal, M., Bodin, L., Elsen, J.M. and Chevalet, C. 2001. Genetic components of litter size variability in sheep. *Genet. Sel. Evol* 33: 249-271.
- SanCristóbal, M., Elsen, J.M., Bodin, L., and 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.
- Slatkin, M. and Lande, R. 1976. Niche width in a fluctuating environment-density independent model. *American Naturalist* 110: 31-55.
- Sorensen, D. 2010. The genetics of environmental variation. *9th World Congress on Genetics applied to Livestock Production- Leipzig(Alemania)*.
- Sorensen, D. and Waagepetersen, R. 2003. Normal linear models with genetically structured residual variance heterogeneity: a case study. *Genet. Res. Camb.* 82 : 207-222.
- Wolc, A., White, I.M.S., Avedano, S. and Hill, W.G. 2009. Genetic variability in residual variation of body weight and conformation scores in broiler chickens. *Poultry Science* 88: 1156-1161.
- Ye Yang, Ole F. Christensen, Daniel Sorensen. 2011. Analysis of a genetically structured variance heterogeneity model using the Box-Cox transformation. *Gen. Res. Cam.* 93 : 33-46.

Zhang, X. and Hill, G. 2008. Mutation-Selection balance for environmental variance. *The American Naturalist* 171: 394-399.