Epistasis for Founder-Specific Inbreeding Depression in Rabbits

JOAQUIM CASELLAS, DOLORS VIDAL-ROQUETA, EVA FLORES, DOLORS CASELLAS-VIDAL, MARÇAL LLACH-VILA, ROSER SALGAS-FINA, AND PERE CASELLAS-MOLAS

From the Departament de Ciència Animal i dels Aliments, Universitat Autònoma de Barcelona, Bellaterra, Spain (Casellas); Can Quim Maret farm, Sant Gregori, Spain (Vidal-Roqueta, Llach-Vila, Salgas-Fina and Casellas-Molas); Departament de Biologia, Universitat de Girona, Girona, Spain (Flores); and Servei de Pediatria, Hospital Universitari Josep Trueta, Girona, Spain (Casellas-Vidal).

Address correspondence to Joaquim Casellas at the address above, or e-mail: joaquim.casellas@uab.cat.

Abstract

Inbreeding depression is a topic of main interest in experimental and domestic species, although previous studies simplified this genetically complex effect to the linear (or quadratic) regression coefficient linked to the inbreeding coefficient of each individual or, in more recent studies, to founder-specific inbreeding coefficients. Going beyond generalizing to these traditional scenarios, our research focused on the analysis of gene-by-gene interactions leading to epistasis for inbreeding depression effects. Under a Bayesian context, inbreeding depression effects were evaluated for weaning weight (WW) in a commercial rabbit population founded from 4 bucks and 1 doe (MARET population). Founder-specific inbreeding depression effects for the 4 bucks ranged between $-1.1$ and $3.8$ g for each 1% inbreeding. More interestingly, 2 epistatic interactions between the partial inbreeding coefficient of 2 bucks were also significant and negative, showing a $-1.9$ and $-1.0$ g reduction on WW.

These results provide the first evidence of epistatic inbreeding depression phenomena in domestic species, emphasizing the complexity of the genetic architecture in mammals.

Key words: epistasis, inbreeding, MARET rabbit, weaning weight

Two alleles at a single autosomal locus are identical-by-descent (IBD) if they are identical copies of the same allele in some ancestor from an earlier generation (Cotterman 1940; Malécot 1941, 1946, 1948a, 1948b). Following Wright (1922), the correlation between uniting gametes (i.e., IBD probability) for a given individual can be calculated from pedigree data by the tabular method, leading to the inbreeding coefficient. This biological phenomenon results from the mating of related individuals and can be directly linked with a decrease in the heterozygosity (Wright 1933). Moreover, IBD has relevant implications at the phenotypic level where IBD probabilities were related to reduced fitness in multiple species (Sittmann et al. 1966; Swindell and Bouzet 2006; Norberg and Sørensen 2007) due to the hidden (i.e., recessive) genetic load carried by a population. This harmful effect of increasing IBD probabilities is known as inbreeding depression (Hedrick and Kalinowski 2000; Charlesworth and Willis 2009), and its consequences were first documented by Koelreuter (1766), Knight (1799) and Darwin (1876) in both plant and animal species.

When the recessive genetic load is unevenly distributed among founder genomes or when founder lines are exposed to variable amounts of selection, the descendants of different founders may be differentially affected by inbreeding depression (Man et al. 2003; Gulisija et al. 2006; Casellas et al. 2009). This heterogeneity can be modeled by founder-specific partial inbreeding coefficients (Lacy et al. 1996), where the inbreeding depression inherent to each founder is straightforwardly accounted for in the model as an independent regression coefficient (Rodriga´ñez et al. 1998). Although whole inbreeding depression tends to cause harmful influences on fitness traits (Charlesworth and Willis 2009), founder-specific estimates revealed a wide range of neutral and negative (and even positive) effects of inbreeding in domestic livestock (Rodriga´ñez et al. 1998; Gulisija et al. 2006; Casellas et al. 2008). These results allowed for a more accurate characterization of the deleterious genetic background of each founder, providing new insights to appropriately manage inbreeding in livestock species.

Models accounting for founder-specific inbreeding depression provide a reliable approximation to the genetic mechanisms underlying inbreeding depression at an individual level (Gulisija et al. 2006; Casellas et al. 2008, 2009). Nevertheless, these approaches neglect any epistatic interaction between founder-specific deleterious genetic loads.
Epistasis, the interaction between genes affecting quantitative trait variation (Cockerham 1954) has been demonstrated by Fisher’s (1918) partition of genetic variance into additive, dominance and epistatic components. Epistasis controlling quantitative traits has been reported in multiple species (Brim and Cockerham 1961; Stuber and Moll 1971; Barendse et al. 2007; Leamy et al. 2008; Noguera et al. 2010), and the contribution of epistasis on the total phenotypic variance was suggested as highly relevant (Cheverud and Routman 1995). Within the context of inbreeding depression, epistatic interactions have not been evaluated, although they must be considered as a potential genetic phenomenon linked with the uneven distribution (i.e., genetic variability) of the deleterious genetic load among founder genomes (Rodríguez et al. 1998; Gulisija et al. 2006; Casellas et al. 2009).

The objective of this manuscript was to reparameterize the founder-specific inbreeding depression term in order to accommodate epistatic interactions, as well as to evaluate the potential contribution of epistatic inbreeding depression on quantitative traits. Note that this research will contribute first insights about epistatic mechanisms modulating the inbreeding depression phenomenon in domestic livestock. The new parameterization for inbreeding-related effects has been studied on the weaning weight (WW) of an inbred population of domestic rabbits (Oryctolagus cuniculus) founded from a small sample of individuals (4 bucks and 1 doe).

### Materials and Methods

#### Field Data Source

The Multiple Autochthonous Rabbit Eco-Types (MARET) population was founded in 1992 by the acquisition of 1 doe (F1; Vilafraser, Spain) and 1 buck (F2; Salt, Spain) from 2 autochthonous rabbit populations without known relationships among them. After that, 3 more unrelated bucks were acquired on 1996 (F3; Vilafraser, Spain), 1998 (F4; Girona, Spain) and 1999 (F5; Sant Gregori, Spain), and they contributed to the formation of this rabbit population. Note that the founder doe and 1 buck (F3) came from the same farm, although with a time lag larger than 3 years. After F5, MARET population was closed to additional foreign contributions with all subsequent buck and doe replacements generated from purebred MARET animals. This rabbit population was maintained under standard farm conditions to produce rabbits for the slaughterhouse.

All reproductive (buck, doe, date of mating, birth, and weaning) and productive data (number of pups born alive and dead and number of pups weaned) were recorded since 1992, with 2629 pups born from 349 litters produced by 16 bucks and 60 does. Since August 2002, all rabbits were weighted at weaning (31.0 ± 0.2 days; mean ± standard error [SE]). The analyses focused on the WW of 902 rabbits from 142 litters (606.4 ± 5.1 g). See Table 1 for a detailed description of the data set.

### Analytical model

Take Henderson’s (1973) linear mixed model as starting point

\[
y = X_1b + X_2d + Z_1p + Z_2a + e,
\]

where \( y \) was the vector of phenotypic data affected by systematic (\( b \)), inbreeding depression (\( d \)), and permanent (\( p \)) and genetic (\( a \)) effects, \( X_1, X_2, Z_1, \) and \( Z_2 \) were appropriate incidence matrices and \( e \) stored residual terms. More specifically, \( a \) accounted for the infinitesimal polygenic additive effect (Balmer 1980) linked to each animal included in the pedigree file, whereas \( p \) accounted for the permanent environmental contribution of the dam to their offspring during the suckling period. Note that dominance genetic effects were not modeled due to the restrictions inherent to the inverse of the dominance relationship matrix under inbreeding (Smith 1984; Schaeffer et al. 1989); these dominance (or recessive) effect must accumulate in \( X_3d \), \( p \), and \( e \), although they can also contribute in \( a \) (Fuerst and Sölkner 1994; Miglior et al. 1995). Systematic effects included the sex of the rabbit (male or female), number of pups born alive with 6 levels (1–4 pups, 5 or 6 pups, 7 pups, 8 pups, 9 pups, and 10 or more pups), and year of birth with 8 levels (from 2002 to 2009). \( X_3d \) modeled founder-specific inbreeding depression effects and their epistatic interactions on the basis of the appropriate partial inbreeding coefficients (Lacy et al. 1996; Lacy 1997), although different

### Table 1  Summary of the WW data set provided by the MARET rabbit population

<table>
<thead>
<tr>
<th>Year</th>
<th>Breeding individuals</th>
<th>Parturitions</th>
<th>Pups</th>
<th>Weaning weight (g) ± SE</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Bucks</td>
<td>Does</td>
<td>Parturitions</td>
<td>Born</td>
</tr>
<tr>
<td>2002</td>
<td>2</td>
<td>7</td>
<td>10</td>
<td>92</td>
</tr>
<tr>
<td>2003</td>
<td>3</td>
<td>8</td>
<td>21</td>
<td>164</td>
</tr>
<tr>
<td>2004</td>
<td>3</td>
<td>7</td>
<td>16</td>
<td>134</td>
</tr>
<tr>
<td>2005</td>
<td>2</td>
<td>6</td>
<td>13</td>
<td>103</td>
</tr>
<tr>
<td>2006</td>
<td>3</td>
<td>8</td>
<td>21</td>
<td>149</td>
</tr>
<tr>
<td>2007</td>
<td>4</td>
<td>9</td>
<td>24</td>
<td>172</td>
</tr>
<tr>
<td>2008</td>
<td>3</td>
<td>8</td>
<td>20</td>
<td>189</td>
</tr>
<tr>
<td>2009</td>
<td>3</td>
<td>8</td>
<td>17</td>
<td>126</td>
</tr>
<tr>
<td>Overall</td>
<td>9</td>
<td>30</td>
<td>142</td>
<td>1129</td>
</tr>
</tbody>
</table>
structures were assumed during model comparison (see the Model Selection).

Following Gulisija et al. (2006), the inbreeding coefficient (Wright 1922; Malécot 1948) can be partitioned into partial inbreeding coefficients, that is, inbreeding coefficients attributed to specific founders. The partial inbreeding coefficient for the $i$th individual linked to the $j$th founder ($F_{ij}$) is defined as the IBD probability at any autosomal locus by alleles coming from the $j$th founder (Lacy et al. 1996). Additionally, the overall probability of identity-by-descent can be obtained as the sum of the IBD probabilities contributed by all possible founders (Gulisija et al. 2006). In order to estimate the inbreeding depression effect specific to the $j$th founder ($\pi_j$, element of vector $\mathbf{d}$), the $m$th column of $\mathbf{X}_2$ stored the appropriate $F_{i,k}$ coefficients. On the basis of dominance-by-dominance (i.e., recessive-by-recessive) Cockerham’s (1954) epistatic modeling, the interaction effect between founders $j$ and $k$ can be estimated by storing the appropriate $F_{i,j}F_{i,k}$ values into the $m$th column of $\mathbf{X}_2$.

Bayesian Analysis

Under a standard Bayesian development, the joint posterior probability of all unknown parameters in the model can be written as

$$p(\mathbf{b}, \mathbf{d}, \mathbf{p}, \mathbf{a}, \sigma_a^2, \sigma_s^2, \sigma_e^2 | \gamma) \propto p(\mathbf{y} | \mathbf{b}, \mathbf{d}, \mathbf{p}, \mathbf{a}, \sigma_e^2)$$

$$p(\mathbf{b})p(\mathbf{d})p(\sigma_a^2)p(\sigma_s^2)p(\sigma_e^2) \times p(\mathbf{a} | \mathbf{A}, \sigma_a^2)p(\sigma_a^2)p(\sigma_s^2),$$

where $\sigma_a^2$, $\sigma_s^2$, and $\sigma_e^2$ were permanent environmental additive genetic and residual variances, respectively. Note that $\mathbf{A}$ was the numerator relationship matrix and its inverse, required for the proper construction of the mixed model equations (Henderson 1973), was constructed by Quaas (1976). The Bayesian likelihood was modeled under a multivariate normal distribution

$$p(\mathbf{y} | \mathbf{b}, \mathbf{d}, \mathbf{p}, \mathbf{a}, \sigma_e^2) \sim \text{MVN}(\mathbf{X}\mathbf{b} + \mathbf{X}_2\mathbf{d} + \mathbf{Z}_1\mathbf{p} + \mathbf{Z}_2\mathbf{a}, \mathbf{I}, \sigma_e^2),$$

$I$, being an identity matrix with dimensions equal to the number of elements in vector $\mathbf{y}$. Multivariate normal a priori normal distributions were assumed for permanent environmental and additive genetic effects

$$p(\mathbf{b} | \sigma_e^2) \sim \text{MVN}(0, \mathbf{I}, \sigma_e^2),$$

$$p(\mathbf{a} | \sigma_a^2, \mathbf{A}, \sigma_s^2) \sim \text{MVN}(0, \mathbf{A}, \sigma_a^2).$$

Unbounded uniform flat priors were assumed for $\mathbf{b}$ and $\mathbf{d}$, whereas a priori distributions for $\sigma_a^2$, $\sigma_s^2$, and $\sigma_e^2$ were uniform between 0 and $+\infty$. Autocorrelated samples from the marginal posterior density of all unknowns in model were obtained by Markov chain Monte Carlo sampling (Geman and Geman, 1984; Gelfand and Smith, 1990). For each model, a Monte Carlo Markov chain of 550 000 rounds was launched and the first 50 000 iterations were discarded as burn-in (Raftery and Lewis, 1992). Given the autocorrelation between successive samples, a total of 10 000 samples of model parameters were saved from each chain with a lag interval of 50 iterations. These samples were used to calculate the posterior distributions of all unknown parameters in the model.

The analyses of the Bayesian mixed linear models were performed by ad hoc software developed by the authors.

Model Selection

Although this population was created from 5 founders, only 4 of them had a relevant contribution in terms of inbreeding (F2–F5; Table 2). Founder F1 originated very low levels of inbreeding (0.20–1.05%; Table 2) and was excluded from the analysis to avoid biases due to its scarce informativeness. Inbreeding depression effects included the direct contribution of founders F2–F5 ($f_2, f_3, f_4,$ and $f_5$) as well as 10 epistatic combinations defined as the product between 2 partial inbreeding coefficients from the same or different founders (i.e., $f_2f_3$, $f_2f_4$, $f_2f_5$, $f_3f_4$, $f_3f_5$, $f_4f_5$, $f_2f_3f_4$, $f_2f_3f_5$, and $f_2f_4f_5$). In order to determine the statistical relevance of these inbreeding-related effects, a stepwise-like approach (Hocking 1976) was adopted by using forward selection of new inbreeding-related effects under an iterative process. This stepwise approach evolved by several rounds that started with a base model without inbreeding-related effects ($\mathbf{d} = 0$; Model 0). At each round, the statistical performance of the base model proceeding from the previous iteration was compared with several competing models generated by the independent inclusion of the remaining inbreeding-related effects to the base model. Model performance was evaluated by the deviance information criterion (DIC; Spiegelhalter et al. 2002). Note that reductions of 3 or more DIC units were considered as statistically relevant (Spiegelhalter et al. 2002, 2003). The competing model with the lowest DIC was assumed as the base model for the next round. Alternatively, the stepwise selection procedure finished when the base model obtained a DIC smaller or less than 3 DIC units larger than the competing model with the smallest DIC, assuming its combination of inbreeding-related effects as the preferred one (Model $F_{\text{partial}}$).

In order to compare this model with a more traditional analysis of inbreeding depression, an alternative expansion of Model 0 with the linear and quadratic effects of the whole inbreeding coefficient ($F_1 + F_2 + F_3 + F_4 + F_5$) was also evaluated in terms of DIC and inbreeding depression estimates (Model $F_{\text{overall}}$).

Table 2 Founder individuals contributing to the MARET rabbit population and their inbreeding contributions

<table>
<thead>
<tr>
<th>Founder</th>
<th>Year of acquisition</th>
<th>Average inbreeding, %</th>
<th>Minimum ± SE</th>
<th>Maximum</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>1992</td>
<td>0.20</td>
<td>0.61 ± 0.01</td>
<td>1.05</td>
</tr>
<tr>
<td>F2</td>
<td>1992</td>
<td>2.73</td>
<td>7.20 ± 0.10</td>
<td>11.89</td>
</tr>
<tr>
<td>F3</td>
<td>1996</td>
<td>1.56</td>
<td>8.30 ± 0.15</td>
<td>19.75</td>
</tr>
<tr>
<td>F4</td>
<td>1998</td>
<td>1.17</td>
<td>5.03 ± 0.14</td>
<td>11.12</td>
</tr>
<tr>
<td>F5</td>
<td>1999</td>
<td>5.08</td>
<td>12.42 ± 0.19</td>
<td>20.31</td>
</tr>
<tr>
<td>Overall</td>
<td>1992</td>
<td>4.49</td>
<td>29.76 ± 0.51</td>
<td>53.83</td>
</tr>
</tbody>
</table>
Results

Covariate Selection and Model Comparison

Model 0 was the starting point of the stepwise approach, a hierarchical mixed linear model accounting for systematic (sex, litter size, and year of birth), permanent environmental and additive genetic effects, although excluding inbreeding-related effects \( (d = 0) \). This model obtained a DIC estimate of 11 299.7. The first 4 stepwise rounds sequentially selected direct inbreeding depression effects from founders FO2, FO4, FO3, and FO5, with DIC values of 11 262.2, 11 241.8, 11 230.1, and 11 218.5, respectively. Note that the DIC statistic provided statistical evidences of increasing model performances at each round, with DIC reductions between 37.5 (round 1) and 11.6 units (round 4; Table 3).

Fifth and sixth stepwise rounds selected the interaction effect \( f_2 f_5 \) (DIC = 11 196.0) and \( f_4 f_5 \) (DIC = 11 192.2) and provided statistical evidences of epistatic interactions between founders for inbreeding depression effects on WW in rabbits. Model \( F_{\text{partial}} \) accounted for 4 direct founder-specific inbreeding depression effects \( (f_2, f_3, f_4, \text{and } f_5) \) and 2 epistatic interactions involving founders FO3, FO4, and FO5 \( (f_2 f_3 \text{ and } f_4 f_5) \). It is important to highlight that within-founder quadratic effects \( (f_2 f_5, f_3 f_5, f_4 f_4, \text{and } f_5 f_5) \) were not selected during the stepwise-like approach (Table 3).

In order to characterize the statistical divergence of Model \( F_{\text{partial}} \) in front of more classical parameterizations, Model \( F_{\text{overall}} \) with the linear and quadratic effects of the overall inbreeding coefficient was also evaluated. Its DIC value was 11 228.2, 36.0 DIC units larger than the value obtained under Model \( F_{\text{partial}} \). Moreover, this DIC was clearly larger (+9.7) than DIC value calculated at the end of the fourth stepwise round. Note that these 9.7 DIC units difference supported the advantage of using partial inbreeding coefficients instead of whole inbreeding coefficients, even when epistatic interaction terms were not accounted for by the model.

Inbreeding-Related Estimates and Variance Components

Although Model \( F_{\text{overall}} \) was clearly discarded by the DIC statistic, both linear and quadratic inbreeding-related regression coefficients were useful to provide a rough idea about the overall inbreeding depression effect in this population and to compare statistical performances of Model \( F_{\text{overall}} \) and Model \( F_{\text{partial}} \). The modal estimate for the linear effect reported a −5.5 g of WW reduction for each 1% increase of inbreeding, whereas the quadratic effect showed a modal estimate of 0.07 g². In both cases, the highest posterior density region at 95% (HPD95) excluded the null estimate, suggesting that the DIC penalization was

<table>
<thead>
<tr>
<th>Table 3</th>
<th>Stepwise rounds for model selection. Models were compared in terms of DIC (Spiegelhalter et al. 2002)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Stepwise rounds</td>
</tr>
<tr>
<td></td>
<td>1</td>
</tr>
<tr>
<td>Base model</td>
<td>Effects</td>
</tr>
<tr>
<td>DIC</td>
<td>11 299.7</td>
</tr>
<tr>
<td>Tested effect ( f_2 )</td>
<td>11 262.2</td>
</tr>
<tr>
<td>( f_3 )</td>
<td>11 272.0</td>
</tr>
<tr>
<td>( f_4 )</td>
<td>11 281.5</td>
</tr>
<tr>
<td>( f_5 )</td>
<td>11 285.2</td>
</tr>
<tr>
<td>( f_2 + f_5 )</td>
<td>11 285.4</td>
</tr>
<tr>
<td>( f_2 + f_3 )</td>
<td>11 284.2</td>
</tr>
<tr>
<td>( f_2 + f_4 )</td>
<td>11 287.1</td>
</tr>
<tr>
<td>( f_3 + f_5 )</td>
<td>11 283.4</td>
</tr>
<tr>
<td>( f_3 + f_4 )</td>
<td>11 288.4</td>
</tr>
<tr>
<td>( f_4 + f_5 )</td>
<td>11 276.6</td>
</tr>
<tr>
<td>( f_5 + f_4 )</td>
<td>11 286.0</td>
</tr>
<tr>
<td>( f_5 + f_3 )</td>
<td>11 279.1</td>
</tr>
<tr>
<td>( f_3 + f_5 )</td>
<td>11 285.5</td>
</tr>
<tr>
<td>Selected effect ( f_5 )</td>
<td>11 282.5</td>
</tr>
<tr>
<td>DIC difference ( d )</td>
<td>−37.5</td>
</tr>
</tbody>
</table>

\( a \) Reference model obtained from the previous stepwise iteration. This model accounted for 3 systematic effects (sex, litter size, and year of birth), 2 random sources of variation (permanent environmental and additive genetic effects), and the inbreeding-related effects selected during the previous stepwise rounds.

\( b \) Available effects included the direct inbreeding depression effect of founders FO2 \( (j_0) \), FO3 \( (j_0) \), FO4 \( (j_0) \), and FO5 \( (j_0) \), as well as 10 pairwise interactions between founders \( (f_2 \times f_3, f_2 \times f_4, f_2 \times f_5, f_3 \times f_4, f_3 \times f_5, f_4 \times f_5, f_3 \times f_4, f_4 \times f_5, f_5 \times f_3, \text{and } f_5 \times f_4) \).

\( c \) Inbreeding-related effect from the model with the smallest DIC value.

\( d \) DIC difference between the base model and the competing model with the smallest DIC value.
not due to a non-significant contribution of the linear or quadratic effect but a better fit of inbreeding depression under partial inbreeding coefficients.

Focusing on Model $F_{\text{partial}}$, the direct contribution of each founder to inbreeding depression was heterogeneous. Modal estimates ranged from $-81.1$ (FO2) to $38.5$ g (FO3) for each 1% of partial inbreeding, and all HPD95 excluded the null estimate (Table 4). As shown in Figure 1, differences between predicted inbreeding depression effect from Model $F_{\text{overall}}$ and Model $F_{\text{partial}}$ were substantial. Nevertheless, inbreeding depression was not restricted to the direct contribution of each founder as some significant epistatic interactions between founders FO3, FO4, and FO5 were revealed (see Table 4).

Despite a moderate residual variance (Table 3), both additive genetic and permanent environmental random sources of variation remarkably influenced WW in MARET rabbits under Model $F_{\text{partial}}$. After accounting for systematic effects (i.e., sex of the rabbit, litter size, year of birth, and inbreeding-related effects), the modal estimate for the additive genetic variance was large, providing a heritability ($h^2$) of 0.31 (Table 3). Permanent environmental variance represented another relevant source of variability, accounting for the 44% of the phenotypic variance. This random effect accounted for the maternal ability of each dam and suggested a large degree of heterogeneity among does.

**Discussion**

**Brief Overview on MARET Population**

The MARET population contributed a valuable data set for the analysis of the within-founder additive genetic variability holding on rabbit WW. Although MARET’s growth performance could be anticipated as moderate due to the non-selected origin of its founders and the accumulation of inbreeding depression effects (Table 2), the average WW was comparable to the pre-weaning growth performance of several commercial rabbit lines (Feki et al. 1996; Dalle Zotte and Ouhayoun 1998). Indeed, the values shown in Table 1 fell within the wide range of larger (Prayaga and Eady 2003; Gondret et al. 2005) and smaller (McNitt and Lukefahr 1993; Argente et al. 1999) WWs reported in current commercial production systems. This phenotypic performance could be surprising for an inbred population with non-commercial rustic origins, although it must be interpreted within the context of a moderate reproductive performance averaging of 7.26 pups born per litter and 5.88 pups weaned per litter (Table 1); they probably allow for an enhanced pre-weaning growth (Rommers et al. 2001). Both litter size and inbreeding depression effects were accounted for in our analyses of WW.

WW in MARET rabbits was significantly affected by 2 random sources of variation, the permanent environmental effect characterized by the doe and the additive genetic effect inherent to each individual. The infinitesimal additive genetic effect provided a modal estimate for heritability of 0.31 (HPD95: 0.17–0.47). This heritability agreed with the value reported by Argente et al. (1999; 0.24) and was larger than the estimates provided by Estany et al. (1992; 0.15), Ferraz and Eler (1994; 0.11–0.14), and Rochambeau et al. (1994; 0.08–0.09). Nevertheless, this value was linked to the founder generation, and thus, genetic diversity must be smaller in the last generations of this population due to the increasing inbreeding. On the other hand, the modal estimate for the permanent environmental effect accounted for 44% of the phenotypic variance, a percentage similar to the 1 provided by Estany et al. (1992; 0.15), Ferraz and Eler (1994; 0.11–0.14), and Rochambeau et al. (1994; 0.08–0.09). Nevertheless, this value was linked to the founder generation, and thus, genetic diversity must be smaller in the last generations of this population due to the increasing inbreeding. On the other hand, the modal estimate for the permanent environmental effect accounted for 44% of the phenotypic variance, a percentage similar to the 1 provided by Argente et al. (1999). This estimate prevented additional estimates of maternal ability on rabbit WW although some dominance genetic effects and dam-specific inbreeding depression effects could also be accumulated in $p$. Unfortunately, the small number of breeding females (30 does; Table 1) contributing to this

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**Table 4** Variance components and inbreeding-related estimates for WW in the MARET population

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Mode</th>
<th>HPD95</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\sigma^2_{\text{g}}$</td>
<td>12 855.2</td>
<td>5951.7–24 040.6</td>
</tr>
<tr>
<td>$\sigma^2_{\text{e}}$</td>
<td>17 782.5</td>
<td>9856.5–28 311.3</td>
</tr>
<tr>
<td>$\sigma^2_{\text{r}}$</td>
<td>9516.3</td>
<td>6207.1–12 918.8</td>
</tr>
<tr>
<td>$\rho_{\text{g}}$</td>
<td>0.31</td>
<td>0.17–0.47</td>
</tr>
<tr>
<td>$\gamma^2$</td>
<td>0.44</td>
<td>0.29–0.59</td>
</tr>
</tbody>
</table>

Inbreeding-related effects:

- $f_2$: $-81.1$ to $-100.7$ (empty dots) and $-58.7$ (black dots) for MARET rabbits
- $f_3$: $38.3$ to $27.0$ (empty dots) and $47.8$ (black dots)
- $f_6$: $24.0$ to $7.3$ (empty dots) and $37.5$ (black dots)
- $f_8$: $27.4$ to $16.4$ (empty dots) and $41.3$ (black dots)
- $f_3 \times f_6$: $-1.9$ to $-3.2$ (empty dots) and $-0.9$ (black dots)
- $f_4 \times f_5$: $-1.0$ to $-2.2$ (empty dots) and $-0.1$ (black dots)

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$^a$ $\sigma^2_{\text{g}}$: additive genetic variance; $\sigma^2_{\text{e}}$: permanent environmental variance; $\sigma^2_{\text{r}}$: residual variance; $\rho_{\text{g}} = \sigma^2_{\text{g}}/(\sigma^2_{\text{g}} + \sigma^2_{\text{e}} + \sigma^2_{\text{r}})$; and $\gamma^2 = \sigma^2_{\text{g}}/(\sigma^2_{\text{g}} + \sigma^2_{\text{e}} + \sigma^2_{\text{r}})$.

$^b$ Inbreeding depression effects due to the direct influence of founder $i$ ($a_i$) and the epistatic interaction between founders $i$ and $j$ ($a_i \times a_j$). Regression coefficients were provided on the basis of a 0% (non-inbred individual) to 100% (fully inbred individual) scale of inbreeding.
study prevented for additional estimates about maternal inbreeding-depression effects, although they were revealed as potential source of inbreeding-related effects in other rabbit populations (Nørberg and Sørensen 2007).

Accounting for Inbreeding Depression Effects

This study focused on the study of the genetic architecture of inbreeding depression in rabbits. Taking the founder-specific inbreeding depression parameterization as starting point (Rodrigáñez et al. 1998; Gulisija et al. 2006), the bidimensional interaction between founder-specific partial inbreeding coefficients opened an alternative research field where recessive-by-recessive epistasis played a primary role. The classical parameterization of inbreeding depression assumed a linear (or polynomial) relation between inbreeding and the reduction in fitness traits, based on the assumption that the genetic load was homogeneously distributed across founders and founder lines were exposed to the same selection intensity. Previous studies discarded this simplified scenario and provided evidences supporting a heterogeneous contribution from different founder genomes to inbreeding depression in swine (Rodrigáñez 1998; Casellas et al. 2008), cattle (Gulisija et al. 2006), and sheep (Casellas et al. 2009). Our analyses have provided additional evidences in this line, not only favoring a founder-specific description of inbreeding depression but reporting the first evidence of between-founders epistatic interactions for inbreeding depression effects (Table 4).

The stepwise-like approach conducted in this study supposed the analysis of 78 nested models, allowing for the sequential selection of several inbreeding depression effects on rabbit WW. DIC differences between successive stepwise rounds (−11.6 to −37.5 units) were large enough to assure the statistical relevance of each new term in the model. Founder-specific inbreeding depression effects revealed a high degree of heterogeneity reporting both negative (FO2) and positive (FO3, FO4, and FO5) linear effects on WW. This wide range of inbreeding depression effects agreed with previous studies in other species (Casellas et al. 2008, 2009) and suggested that, although the effect of inbreeding tends to be harmful for fitness traits, neutral or even positive effects cannot be discarded in animals (Wilson 1948; Ballou 1997). More interestingly, the combined effect of partial inbreeding coefficients from founders FO3 and FO5 and from founders FO4 and FO5 led to significant negative recessive-by-recessive epistatic contributions to the WW of MARET rabbits. Note that our inbreeding depression estimates showed moderate HPD95 due to the limited number of rabbits contributing to the data set, suggesting that we must be cautious with our modal estimates and their absolute contribution to the WW of MARET rabbits. Nevertheless, these inbreeding depression estimates were the first evidence of epistasis within the context of inbreeding depression in mammals and recommended an accurate re-examination of this topic to properly account for an increasing degree of complexity linked to the genetic architecture of inbreeding effects. It is important to highlight that estimates shown in Table 4 were specific to MARET rabbits and to the 4 founder individuals contributing current population. They suggested the evidence of founder-specific and epistatic inbreeding depression in mammals, although current estimates cannot be directly extrapolated to other populations.

As pointed out by Crow and Kimura (1970), non-linear inbreeding depression demonstrated by Model \( F_{overall} \) can result from dominant-by-dominant epistatic interactions. This agreed with results obtained under Model \( F_{partial} \) where 2 interaction terms, i.e., \( f_2f_5 \) and \( f_3f_5 \), significantly contributed to the overall inbreeding depression for rabbit WW, even when none of the founder-specific quadratic effects (i.e., \( f_2^2, f_3^2, f_4^2, \) and \( f_5^2 \)) reached statistical relevance. Moreover, our analyses cannot completely discard the contribution of other between- and within-founder interaction terms, given that the moderate to small range of founder-specific inbreeding coefficients in the MARET population (Table 2) could be limiting the ability of the mixed linear model to capture non-linear processes. The joint contribution of all inbreeding-related estimates could not be easily characterized due to the multidimensional structure of inbreeding depression under this detailed parameterization, although some general ideas can be summarized from Figure 1. Starting from modal estimates reported in Table 4, this figure plotted the predicted inbreeding depression effect for each rabbit included in our data set. The dispersion pattern suggested that 1) inbreeding depression followed a rough negative trend with increasing inbreeding coefficients, 2) positive effects of inbreeding depression were not rare, even for intermediate inbreeding coefficients, and 3) inbreeding depression was heterogeneous, allowing for a wide range of estimates within the same overall inbreeding coefficient. These attributes provided an additional flexibility to Model \( F_{partial} \) relative to Model \( F_{overall} \). Note that the simplification of inbreeding depression to the linear and quadratic effects of the overall inbreeding coefficient forced the predicted inbreeding depression to a curve with negative slope (Figure 1), agreeing with the overall negative trend suggested by Model \( F_{partial} \).

This study was an endeavor to explore the structure of inbreeding depression in mammals, providing a more detailed description of this composite effect on fitness traits. Although epistasis phenomena were previously reported in rabbits (Iraqi et al. 2006) and other species (Barendse et al. 2007; Leamy et al. 2008; Nogueira et al. 2010) in a broad sense, the demonstration of epistatic interactions between the hidden (recessive) genetic load of different founder genomes highlighted the complexity of the genetic architecture in mammals. Moreover, the consideration of founder-specific inbreeding depression effects and epistatic interactions would have a relevant impact in the inbreeding management within current selection and conservation programs of livestock.

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