Analyses for the Presence of a Major Gene Affecting Uterine Capacity in Unilaterally Ovariectomized Rabbits

M. J. Argente,*,†,[1] A. Blasco,‡ J. A. Ortega,‡ C. S. Haley§ and P. M. Visscher†

*Universidad Miguel Hernández, Departamento de Tecnología Agroalimentaria, División de Producción Animal, 03312 Orihuela, Spain, ‡Universidad Politécnica de Valencia, Departamento de Ciencia Animal, 46071 Valencia, Spain, §Roslin Institute, Roslin, Midlothian, Edinburgh EH25 9PS, United Kingdom and †Institute of Cell, Animal and Population Biology, Ashworth Laboratories, University of Edinburgh, Edinburgh EH9 3JT, United Kingdom

Manuscript received July 8, 2002
Accepted for publication December 6, 2002

ABSTRACT

The presence of a major gene for uterine capacity (UC), ovulation rate (OR), number of implanted embryos (IE), embryo survival (ES), fetal survival (FS), and prenatal survival (PS) was investigated in a population of rabbits divergently selected for UC for 10 generations. Selection was performed on estimated breeding values for UC up to four parities. UC was estimated as litter size in the remaining overcrowded horn of unilaterally ovariectomized does. OR and IE were counted by means of laparoscopy. Bartlett's test, Fain's test, and a complex segregation analysis using Bayesian methods were used to test for the presence of a major gene. All three tests showed that the data appeared consistent with the presence of a major gene affecting UC and IE. The results of the complex segregation analysis suggested the presence of a major gene with large effect on IE and ES ($a > 1\sigma_p$), at high frequency ($p = 0.70$ and 0.68, respectively), and with a large contribution to the total variance ($R_g = 0.39$ and 0.47, respectively); and the presence of a major gene with moderate effect on each of OR, FS, PS, and UC. The results suggest that the studied reproductive traits are determined genetically by at least one gene of large effect.

SELECTION on litter size has had only limited success in mice, rabbits, and pigs, because of the low heritability and the sex-limited expression. Selection on uterine capacity has been proposed as an alternative method to improve litter size (Bennet and Leymaster 1989, in pigs; Clutter *et al.* 1990, in mice; and Blasco *et al.* 1994, in rabbits). Uterine capacity has been defined as the maximum number of fetuses that the dam is able to support at birth when ovulation rate is not a limiting factor (Christenson *et al.* 1987). Unilateral ovariectomy (ULO) in rabbits doubles the ovulation rate in the remaining ovary and the adjacent uterine horn is crowded with embryos. Therefore, observed litter size in ULO rabbit females has been used as an estimator of uterine capacity (Blasco *et al.* 1994).

Major genes or quantitative trait loci (QTL) have been detected for litter size in pigs (Rothschild *et al.* 1996; Janss *et al.* 1997; Wilkie *et al.* 1999) and for ovulation rate (Cassady *et al.* 2001) and uterine capacity (Rohrer *et al.* 1999, in pigs; Messer *et al.* 1999, in mice). The inclusion of major gene information could improve efficiency of selection schemes and would improve understanding of the biology of reproductive traits. In rabbits, the ovulation rate, the number of implanted embryos, and litter size can be measured in the same gestation by laparoscopy without affecting litter size (Santacreu *et al.* 1990), which is not possible in other polytocous mammals. Implantation is an important trait in relation to litter size, because 20–40% of shed ova do not achieve implantation (see review in rabbits and pigs; Blasco *et al.* 1993). There have not been any studies in rabbits or other mammals to detect major genes affecting the number of implanted embryos and their subsequent survival.

In a divergent selection experiment for uterine capacity in rabbits, a large difference between lines in uterine capacity (1.25 rabbits) and in the number of implanted embryos (1.65 embryos) was found after the first generation of selection (Argente *et al.* 1997). These large divergences could be due to the segregation of major genes affecting uterine capacity or related traits. Segregation analysis (*e.g.*, Hill and Knott 1990) can be used to investigate the presence of major genes.

The objective of this study was to investigate whether major genes are controlling uterine capacity and other components of litter size: ovulation rate, implanted embryos, prenatal survival, embryo survival, and fetal survival.

MATERIALS AND METHODS

Animals: Rabbits came from a divergent selection experiment on uterine capacity in unilaterally ovariectomized does (ULO does). Uterine capacity was estimated as litter size in the remaining overcrowded horn of ULO does. Both lines

[1]*Corresponding author:* Universidad Miguel Hernández, Departamento de Tecnología Agroalimentaria, División de Producción Animal, Carretera de Beniel Km 3,2, 03312 Orihuela, Spain.
E-mail: mj.argente@umh.es

TABLE 1

Number of records per line

	Base generation	ULO+	ULO−
UC	196	1453	1347
OR	61	349	325
IE	61	349	325
ES	61	349	325
FS	57	327	300
PS	57	327	300

UC, uterine capacity; OR, ovulation rate; IE, number of implanted embryos; ES, embryo survival; FS, fetal survival; PS, prenatal survival; ULO+, high line of uterine capacity; ULO−, low line of uterine capacity.

were derived from a synthetic breed, described by Argente *et al.* (1996). Each divergent line had ~40 females and 12 males per generation, each female had up to four parities, and data from 10 generations of selection were analyzed. Selection was performed on estimated breeding values for uterine capacity, by using a BLUP procedure and a repeatability animal model with year-season and parity fixed effects up to four parities. Data came from 929 does. Table 1 shows the number of records used in the experiment.

Surgical techniques: *Unilateral ovariectomy technique:* The left ovary was removed before puberty in ULO does via midventral incision. The does were anesthetized using a ketamine (50 mg/ml):promethazine (25 mg/ml) mixture injected intramuscularly; 5 min later this injection was followed by an intravenous dose of the same solution in the marginal ear vein. The anesthetized does were lying down on a surgical table, and the surgical table was inclined by 30°. The ovary was grasped with a hemostat, a ligature was placed around the oviduct and blood vessels, and the ovary was removed.

Laparoscopy technique: The does were anesthetized and placed on the surgical table using the same protocol as for the unilateral ovariectomy technique. A Verres needle was introduced laterally beneath the last rib to inflate the abdominal cavity with CO_2 gas. A skin incision was made on the midline, 1 cm below the sternum. A trocar-cannula was inserted through this incision and after the trocar was replaced by endoscopy connected with a cold light fountain of 250 W. A second trocar-cannula was introduced into the right side of the abdominal cavity, through a lateral incision at 3 cm from the sternum. A palpation probe, which allowed manipulating the right ovary and uterine horn of ULO does, was introduced through this second cannula.

Traits: All the traits were measured in unilateral ovariectomized does. The analyzed traits were uterine capacity (UC), estimated as the litter size measured up to four parities; ovulation rate (OR), estimated as the number of corpora lutea at day 12 of the second gestation; number of implanted embryos (IE), estimated as the number of implantation sites at day 12 of the second gestation; prenatal survival (PS) (UC/OR); embryo survival (ES) (IE/OR); and fetal survival (FS) (UC/IE). The numbers of corpora lutea and implanted sites were counted by means of the laparoscopy as described before.

Statistical analysis: *Response to selection:* Phenotypic differences between lines from the divergence selection experiment were calculated for OR, IE, ES, FS, and PS using the GLM procedure of SAS Institute (1996). The statistical model included the fixed effects of generation (with 11 levels), lactation (with 3 levels: nulliparous does, multiparous lactating, and multiparous nonlactating does during pregnancy), and line (with three levels: base generation, high line, and low line of uterine capacity). The random effect of a permanent environment was included in the basic model for UC, and the MIXED procedure of SAS Institute (1996) was used for analysis of phenotypic differences between divergent lines for this trait.

Simple tests for heterogeneity of variance across families: If a major gene of large effect is segregating in the population, we would expect heterogeneity of variance across full-sib and half-sib families and a relationship between the family mean and family variance. Residual values were estimated with a model including the fixed effects of year-season (with 30 levels) and lactation (with 3 levels). Bartlett's and Fain's tests were performed on these estimated residuals. Bartlett's test (Sokal and Rohlf 1995) was used to test for heterogeneity of within-half-sib-family variance. In addition, the regression of family variances on family means (Fain 1978) was performed. The regression model was $V = b_0 + b_1\mu + b_2\mu^2$, where V is the within-family variance and μ is the family mean.

Complex segregation analysis: Janss *et al.* (1995) have presented a Bayesian approach for segregation analysis in livestock. For the segregation analysis, the mixed model assumed to describe phenotypic observation for each trait **y** was

$$\mathbf{y} = \mathbf{Xb} + \mathbf{Zu} + \mathbf{ZWm} + \mathbf{e},$$

where **b**, in a frequentist context, is considered as a vector of fixed nongenetic effects (all effects are random in a Bayesian context; see, for example, Blasco 2001), and **X** is an incidence matrix relating effects of year-season and lactation to observations in **y**. **Z** is an incidence matrix relating the genetic effects to observations in **y**. Genetic effects are separated in polygenic effects in **u** and single-gene effects in **Wm**. Vector **e** contains the errors. Polygenic effects were modeled to be additive. Single-gene effects are expressed using $\mathbf{W} = \{\mathbf{w}_i\}$, a three-column matrix with 0/1 variables to indicate the genotypes of each individual, and the vector $\mathbf{m}' = (-a, d, a)$ that contains the genotypic values, where a and d are referred to as the additive and dominance effects at the single locus. The single major locus was assumed to be autosomal and diallelic (A_1, A_2) with Mendelian transmission probabilities. A common permanent environmental effect was included in the model for UC. Statistical inference was based on a Bayesian approach computing marginal posterior densities of the unknown parameters by the Markov chain Monte Carlo (MCMC) method known as Gibbs sampling. The Bayesian (MCMC) approach for segregation analysis allows the estimation of marginal posterior distributions for nongenetic effects, genotypic values $\mathbf{m}' = (-a, d, a)$, recessive allele frequency q, polygenic variance (V_{polyg}), residual variance (V_e), and permanent environmental variance for UC (V_{per}). On the basis of the allele effects (a and d) and the allele frequencies (p and q), the additive (V_{ga}), dominant (V_{gd}), and total major gene variance (V_g) were calculated as $V_g = V_{ga} + V_{gd} = 2pq[a + d(q - p)]^2 + (2pqd)^2$ (Falconer and Mackay 1996). Also, the proportion of total variance due to polygenic effects ($R_{polyg} = V_{polyg}/V_{TOTAL}$), the additive effect of a major gene ($R_{ga} = V_{ga}/V_{TOTAL}$), and the total effect of a major gene ($R_g = V_g/V_{TOTAL}$) were estimated and their marginal posterior distributions were computed. Uniform prior distributions were assumed in the range ($-\infty$; $+\infty$) for nongenetic effects and genotypic values, in the range (0; $+\infty$) for the variance components and in the range [0; 1] for the allele frequencies. The probability of the genotypic configuration in the pedigree was defined as

$$P(\mathbf{W}') = \prod_{\text{founders } i} P(\mathbf{W}_i) \prod_{\text{nonfounders } j} P(\mathbf{W}_j|\mathbf{W}_{\text{mother } j}, \mathbf{W}_{\text{father } j}).$$

Alleles were assumed to be randomly sampled from the par-

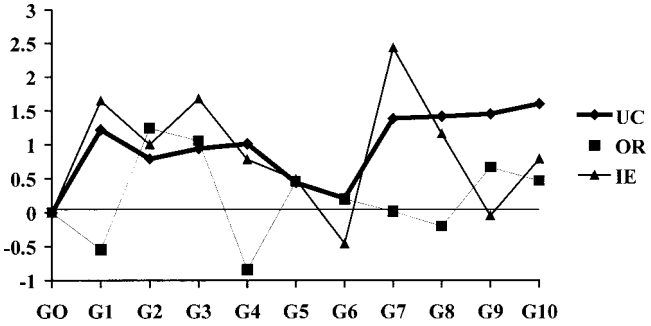

FIGURE 1.—Phenotypic differences among lines for uterine capacity (UC), number of implanted embryos (IE), and ovulation rate (OR).

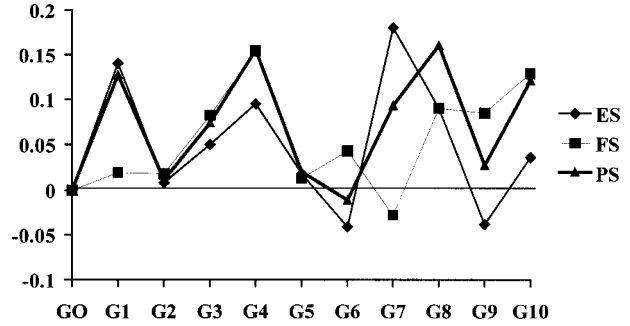

FIGURE 2.—Phenotypic differences among lines for embryo survival (ES), fetal survival (FS), and prenatal survival (PS).

ents' genotypes according to Mendelian rules. For founders, genotypes are assumed to be randomly sampled from the available genotypes given the frequency of the alleles in the base population (q and $1 - q$) under the assumption of Hardy-Weinberg equilibrium. The basis theory and methodology are explained in more detail in JANSS *et al.* (1995) and SORENSEN (1996). For each analysis two chains were run. The length of each chain was set to 500,000 iterations. Exploratory analyses suggested a burn-in period of 300,000 iterations, higher than the minimum required according to the method of RAFTERY and LEWIS (1992). Samples were saved every 25 iterations thereafter, so that the total number of saved samples per chain was 8000. Convergence was tested using the criterion of GELMAN and RUBIN (1992). For each variance, a scale parameter ("shrink" factor, $\sqrt{R}$), which involves variance between and within chains, is computed. The shrink factor can be interpreted as the factor by which the scale of the marginal posterior distribution of each variable would be reduced if the chain were run to infinity. It should be close to 1 to convey convergence. Monte Carlo standard errors were also calculated. The posterior mean, standard deviation, and highest posterior density region at 95% ($HPD_{95\%}$) were calculated from the sampled values of the marginal posterior distributions. The sampled values of each marginal posterior distribution were sorted. $HPD_{95\%}$ was constructed by finding the shortest interval between two values including 95% of the sample. The MaGGic statistical package was used for all complex segregation analyses (JANSS *et al.* 1995).

RESULTS

Figures 1 and 2 show the evolution of the differences between lines estimated by least-square means of the divergent selection experiment for all traits. In the first generation of the selection experiment, a large difference between lines was found for UC (1.25 young rabbits) and IE (1.65 number of implanted embryos). This large difference in UC seems to be associated more with differences in IE than in OR (Figure 1). The evolution of the differences in IE between lines shows a similar pattern to ES. The difference between lines in PS seems to be related to differences in ES. FS does not show any clear pattern (Figure 2).

Tests for heterogeneity of within-family variances (Bartlett's test) were significant for UC ($P < 0.001$) and IE ($P < 0.05$). This is consistent with the presence of a major gene segregating for these traits. OR, ES, FS, and PS did not show any presence of a major gene using Bartlett's test. Linear (b_1) and quadratic (b_2) regressions of family variance on family means (Fain's test) were significantly different from zero only for UC, IE, and ES. Both the phenotypic divergence results and these tests are consistent with the segregation of a major gene for UC and number of IE.

Tables 2 and 3 present the mean, standard deviation, and highest posterior density region at 95% of the marginal posterior distributions for effects of the major gene (a and d), frequency q of the unfavorable allele, and the variance ratios for all traits analyzed. In the same tables, Monte Carlo standard errors (MCSE) and results of Gibbs sampler convergence tests for all traits are also displayed. Monte Carlo standard errors were low for all traits and variables. The number of iterations to be discarded according to the procedure of RAFTERY and LEWIS (1992) ranged from 50 to 1925, the latter corresponding to the frequency of the unfavorable allele (q) for UC, so that the burn-in period used was much higher than the minimum recommended. The shrinking factors for the application of the procedure of GELMAN and RUBIN (1992) were near unity. From this and the above results it was concluded that convergence was achieved; therefore we combined the samples from the two chains to estimate features of the marginal posterior distributions of each variable.

Figures 3 and 4 show the marginal posterior distributions of the variance ratios for all traits. The means of the marginal posterior distributions of the polygenic variance ratio (R_{polyg}) were similar for OR and IE, and lower estimates were obtained for UC, ES, FS, and PS (Tables 2 and 3). The proportion of the total variance due to the total major gene variance (R_g) would be related to the presence of a major gene segregating in this population. The means of the marginal posterior distribution of R_g were the highest for IE and ES (around double the means for OR, FS, and PS) and the lowest for UC (Tables 2 and 3). These results seem to suggest the presence of a major gene with a large additive effect on IE and ES; a moderate effect on OR, FS, and PS; and a lower effect on UC. The ratio of the additive

TABLE 2

Features of marginal posterior distributions of the major gene effects and variance ratios

Traits	*M*	SD	$HPD_{95\%}$	MCSE	B-in	$\sqrt{R}$
UC						
a	1.04	0.49	0.01, 1.79	0.02	800	1.04
d	1.04	0.79	−0.97, 2.16	0.03	1225	1.04
q	0.31	0.15	0.01, 0.56	0.006	1925	1.31
R_{polyg}	0.08	0.02	0.03, 0.13	0.001	1200	1.03
R_{ga}	0.04	0.03	0.00, 0.10	0.001	150	1.02
R_g	0.08	0.05	0.01, 0.17	0.002	625	1.03
OR						
a	1.73	1.41	0.00, 3.16	0.04	750	1.10
d	−1.55	1.52	−3.51, 2.28	0.07	1350	1.31
q	0.68	0.18	0.37, 1.00	0.007	750	1.15
R_{polyg}	0.22	0.09	0.05, 0.39	0.003	1500	1.02
R_{ga}	0.10	0.10	0.00, 0.28	0.003	225	1.02
R_g	0.23	0.13	0.00, 0.45	0.006	700	1.01
IE						
a	3.14	0.32	2.50, 3.76	0.002	100	1.00
d	3.48	0.48	2.56, 4.43	0.004	300	1.00
q	0.30	0.07	0.18, 0.44	0.001	75	1.00
R_{polyg}	0.19	0.06	0.08, 0.32	0.002	250	1.00
R_{ga}	0.15	0.09	0.01, 0.32	0.001	75	1.00
R_g	0.39	0.10	0.20, 0.57	0.001	75	1.00

Mean (*M*), standard deviation (SD), and highest posterior density region at 95% ($HPD_{95\%}$) of the marginal posterior distributions. UC, uterine capacity; OR, ovulation rate; IE, number of implanted embryos; *a*, additive effect; *d*, dominant effect; *q*, frequency of unfavorable allele; R_{polyg}, polygenic variance ratio (V_{polyg}/V_{TOTAL}); R_{ga}, additive major gene variance ratio (V_{ga}/V_{TOTAL}); R_g, total major gene variance ratio ($V_{ga} + V_{gd}/V_{TOTAL}$); MCSE, Monte Carlo standard error; B-in, burn-in of Raftery and Lewis test; $\sqrt{R}$, scale factor of the Gelman and Rubin test.

TABLE 3

Features of marginal posterior distributions of the major gene effects and variance ratios

Traits	*M*	SD	$HPD_{95\%}$	MCSE	B-in	$\sqrt{R}$
ES						
a	0.20	0.02	0.17, 0.24	0.001	150	1.00
d	0.28	0.03	0.23, 0.33	0.001	200	1.00
q	0.32	0.07	0.19, 0.45	0.001	50	1.00
R_{polyg}	0.05	0.03	0.01, 0.11	0.002	800	1.02
R_{ga}	0.13	0.08	0.00, 0.28	0.002	50	1.02
R_g	0.47	0.10	0.29, 0.66	0.001	50	1.01
FS						
a	0.17	0.05	0.08, 0.27	0.002	375	1.04
d	0.21	0.06	0.09, 0.34	0.002	300	1.02
q	0.31	0.11	0.11, 0.52	0.004	600	1.03
R_{polyg}	0.08	0.06	0.01, 0.19	0.002	1500	1.05
R_{ga}	0.09	0.09	0.00, 0.25	0.002	75	1.05
R_g	0.27	0.10	0.07, 0.46	0.002	250	1.01
PS						
a	0.16	0.06	0.04, 0.28	0.002	700	1.04
d	0.26	0.07	0.11, 0.41	0.003	1750	1.11
q	0.20	0.10	0.02, 0.40	0.004	400	1.03
R_{polyg}	0.07	0.04	0.01, 0.14	0.002	1650	1.02
R_{ga}	0.03	0.03	0.00, 0.10	0.001	50	1.02
R_g	0.18	0.11	0.01, 0.38	0.004	400	1.01

Mean (*M*), standard deviation (SD), and highest posterior density region at 95% ($HPD_{95\%}$) of the marginal posterior distributions. ES, embryo survival; FS, fetal survival; PS, prenatal survival; *a*, additive effect; *d*, dominant effect; *q*, frequency of unfavorable allele; R_{polyg}, polygenic variance ratio (V_{polyg}/V_{TOTAL}); R_{ga}, additive major gene variance ratio (V_{ga}/V_{TOTAL}); R_g, total major gene variance ratio ($V_{ga} + V_{gd}/V_{TOTAL}$); MCSE, Monte Carlo standard error; B-in, burn-in of Raftery and Lewis test; $\sqrt{R}$, scale factor of the Gelman and Rubin test.

major variance to the total additive genetic variance (additive major gene and polygenic variance) was higher in IE, ES, and FS (0.44, 0.72, and 0.53, respectively) than in UC, OR, and PS (0.33, 0.31, and 0.30, respectively).

Figure 5 shows the estimates of the marginal posterior distributions of major gene additive effects for all traits. All of them suggest the presence of a major gene. The mean of the marginal posterior distribution of the additive effect (*a*) was higher in IE than in OR and UC, and the highest posterior density region at 95% ($HPD_{95\%}$) for IE did not include zero (Table 2). An advantage of the Bayesian approach trough MCMC procedures is the possibility of easy construction of all kinds of confidence intervals (Bayesians prefer to call them "credibility intervals"). We can find intervals of the type [*k*, +∞) having 95% of the probability area of the marginal posterior distribution. With these intervals we know that the probability of the trait of being $<k$ is 5%. The intervals [0.28, +∞) and [0.27, +∞) contained 95% of the area of the marginal posterior distributions for OR and UC, respectively. Thus, the probability of the additive effect of the gene for OR being lower than 0.28 and that for UC being lower than 0.27 is <5%. The means of the marginal posterior distribution of the additive effect (*a*) for ES, FS, and PS were similar (Table 3). Zero was not included in either the $HPD_{95\%}$ for ES, FS, and PS (Table 3) or the intervals at 95% of the marginal posterior density [0.17, +∞), [0.10, +∞), and [0.05, +∞) for ES, FS, and PS, respectively. The largest additive effects for the major gene were found in IE and ES (1.12 and 1.06 phenotypic standard deviations respectively). The additive effects were lower for OR, FS, PS, and UC (0.75, 0.86, 0.85, and 0.43 phenotypic standard deviations, respectively). The differences between the two homozygous genotypes are 2, 3, and 6 rabbits for UC, OR, and IE and 0.40, 0.34, and 0.32 for ES, FS, and PS, respectively. These estimates are larger than the phenotypic differences found between lines (Figures 1 and 2). The estimates of the dominant effects were similar to the estimates of the additive effects for IE, ES, FS, and PS (Tables 2 and 3); zero was not included in the $HPD_{95\%}$; and the values of *k* for the intervals [*k*, +∞)

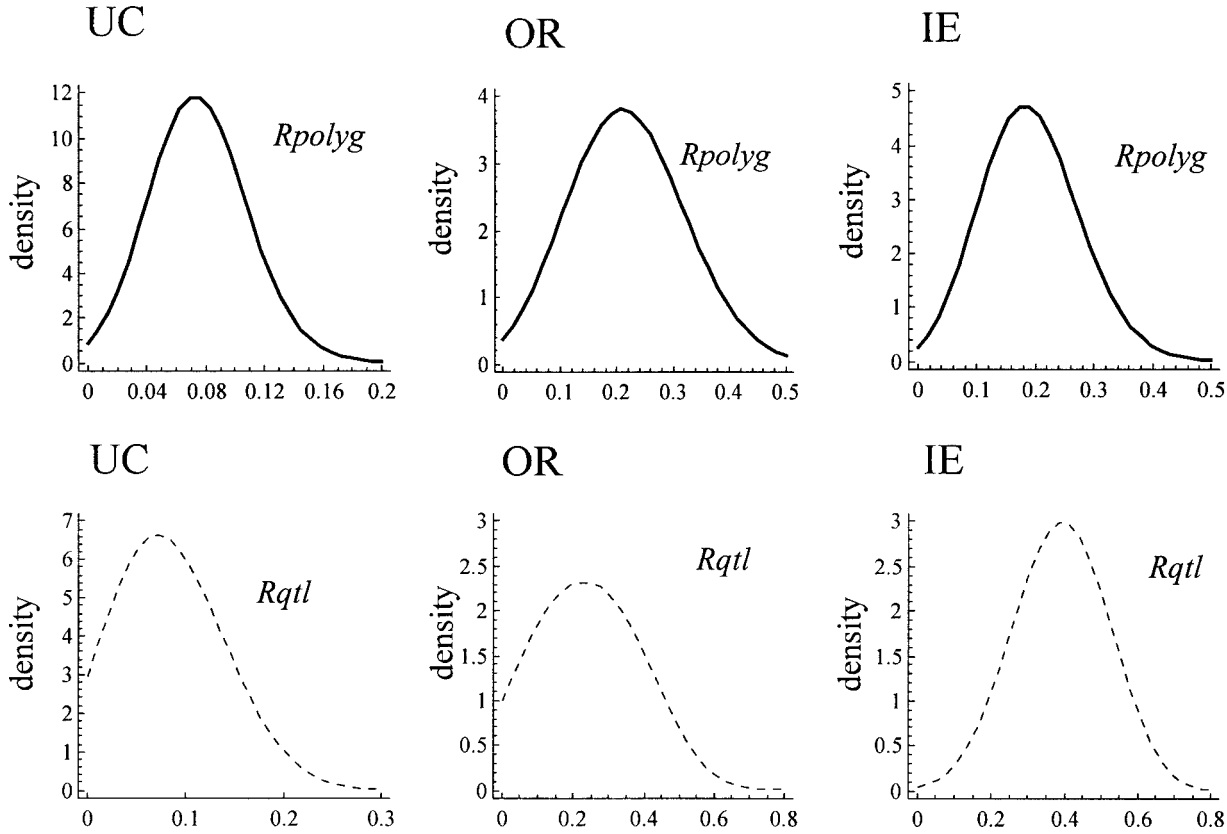

FIGURE 3.—Marginal posterior distributions for polygenic variance ratio (—, $R_{polyg} = V_{polyg}/V_{TOTAL}$) and total major gene variance ratio (- - -, $R_g = V_g/V_{TOTAL}$; with V_g = total major gene variance) for uterine capacity (UC), ovulation rate (OR), and number of implanted embryos (IE).

were 2.69, 0.23, 0.11, 0.12, respectively. Hence, the gene action was found to be dominant for these traits. Tables 2 and 3 also show that the gene was segregating with a frequency of the unfavorable allele near 0.3 for most traits.

Evidence was found for segregation of major genes affecting uterine capacity and other components of litter size: ovulation rate, implanted embryos, embryo survival, and fetal survival. However, the major genes with the larger additive effects and the larger contribution to total variance are connected with embryo survival and implantation.

DISCUSSION

Identification of major genes or QTL affecting reproductive traits in livestock could have a considerable impact on genetic improvement, for example, by increasing the accuracy of selection. Moreover, the presence and identification of major genes related to implantation, embryo survival, or fetal survival can orientate research not only in other livestock species but also in human medicine. However, there have been only a few reports of detected major genes or QTL for reproductive traits in pigs and mice, none in rabbits, and no QTL have been found for components of reproduction, such as the number of IE, ES, and FS. Rabbit is the only domestic animal in which ovulation rate, number of implantation sites, and litter size can be recorded in the same animal by laparoscopy. In sheep, two major genes affecting reproductive performance have been identified, one affecting ovulation rate (GALLOWAY *et al.* 2000) and the second affecting both ovulation rate and litter size (WILSON *et al.* 2001).

The estimates of the polygenic variance ratio obtained in ULO does for UC, OR, number of IE, ES, FS, and PS were in the range of those obtained in intact pigs, rabbits, and mice (see review in BLASCO *et al.* 1993).

Selection on litter size shows an annual improvement between 0.02 and 0.20 newborn per year (ROTHSCHILD and BIDANEL 1998, in pigs; BLASCO 1996, in rabbits; NIELSEN 1994, in mice). ARGENTE *et al.* (2000) found a high genetic correlation (0.92) between litter size and UC, so the genetic trend found for litter size is expected to be the same for UC. However, a larger difference between lines for UC was found in the first generation (1.25 rabbits). Divergent selection for a major gene of moderate effect ($a < \frac{1}{2}\sigma_p$) and not fixed ($p = 0.69$) on UC, as was found in this population, could explain a large amount of response obtained in the first generation of selection.

Selection on UC seems to be associated with large differences between lines in IE (1.65 rabbits) and ES (0.14). These large differences could be explained be-

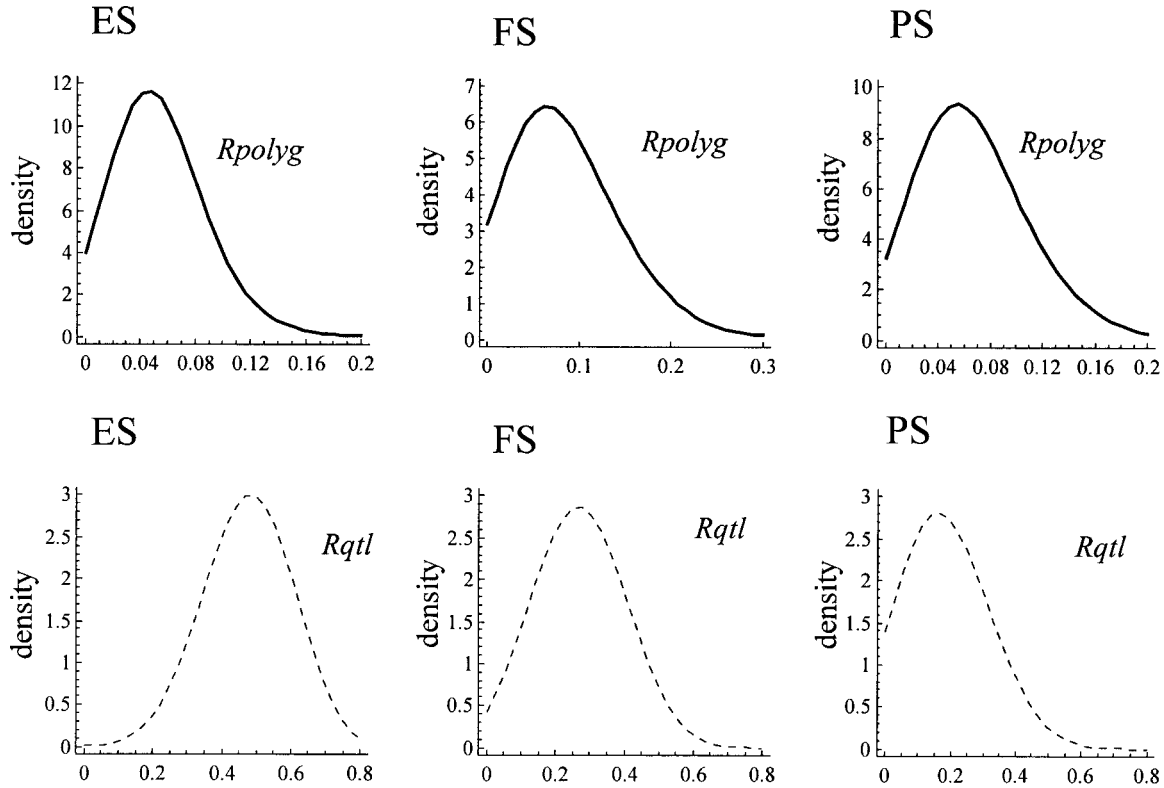

FIGURE 4.—Marginal posterior distributions for polygenic variance ratio (—, $R_{polyg} = V_{polyg}/V_{TOTAL}$) and total major gene variance ratio (- - -, $R_g = V_g/V_{TOTAL}$; with V_g = total major gene variance) for embryo survival (ES), fetal survival (FS), and prenatal survival (PS).

cause UC has a high and moderate correlation with IE (0.71) and ES (0.59), respectively (ARGENTE *et al.* 1997), and also because there are major genes of large effect on IE and ES ($a > 1\ \sigma_p$), which are completely dominant and not fixed ($p \simeq 0.7$). For IE, nearly all of the response was obtained by the second generation of selection, and little response was observed subsequently (Figure 1). Using simulation studies, VILLANUEVA *et al.* (1999) found a maximum response to selection in the second generation and a subsequent decrease in later generations, because a simulated recessive allele with a large additive and dominant effect was fixed. These authors used BLUP and genotype information on the major gene for selection decisions. In our study, the response did not level off, possibly because the favorable and unfavorable alleles were not fixed in the high and low lines, respectively.

In our divergent lines, we found evidence for the segregation of major genes for UC and OR. In a line selected for 21 generations for high litter size in mice, CLUTTER *et al.* (1994) found a difference of 2.97 ova and 2.27 fetuses at day 17 of gestation (close to birth) with the control line. MASSER *et al.* (1999) localized QTL affecting ovulation rates and number of fetuses in this line. RATHJE *et al.* (1993) reported a difference of 6.7 ova and 3.3 fetuses at day 50 of gestation in pigs between a line selected for 10 generations using an index of ovulation rate and embryo survival and a control line. These authors found a QTL affecting ovulation rate and litter size (CASSADY *et al.* 2001). In another line of pigs selected on ovulation rate, ROHRER *et al.* (1999) identified QTL affecting ovulation rate and uterine capacity. The difference between the two breeds of Meishan and Yorkshire pigs was estimated to be 6.4 corpora lutea and 5 piglets (WHITE *et al.* 1993). In the F_2 population generated from crossing these breeds, QTL for ovulation rate and litter size were detected (WILKIE *et al.* 1999). ROTHSCHILD *et al.* (1996) have shown that a specific allele of the estrogen receptor (*ER*) locus is associated with increased litter size, but the *ER* marker was not associated with variation in either OR or UC (ROHRER *et al.* 1999). To our knowledge, this is the first study reporting evidence for major genes of large effect on other important reproductive traits such as IE and ES.

Methods to detect the segregation of genes of large effects on quantitative traits from phenotypic data only are notoriously difficult and usually not robust to violations of assumptions such as normality of residuals and homogeneity of variances (LYNCH and WALSH 1998). Therefore, results from variance analysis should be treated with caution, and conclusive evidence for the segregation of QTL of large effects should be obtained from collecting genotypic (marker) data. Nevertheless, in summary, all our results suggest that there may be

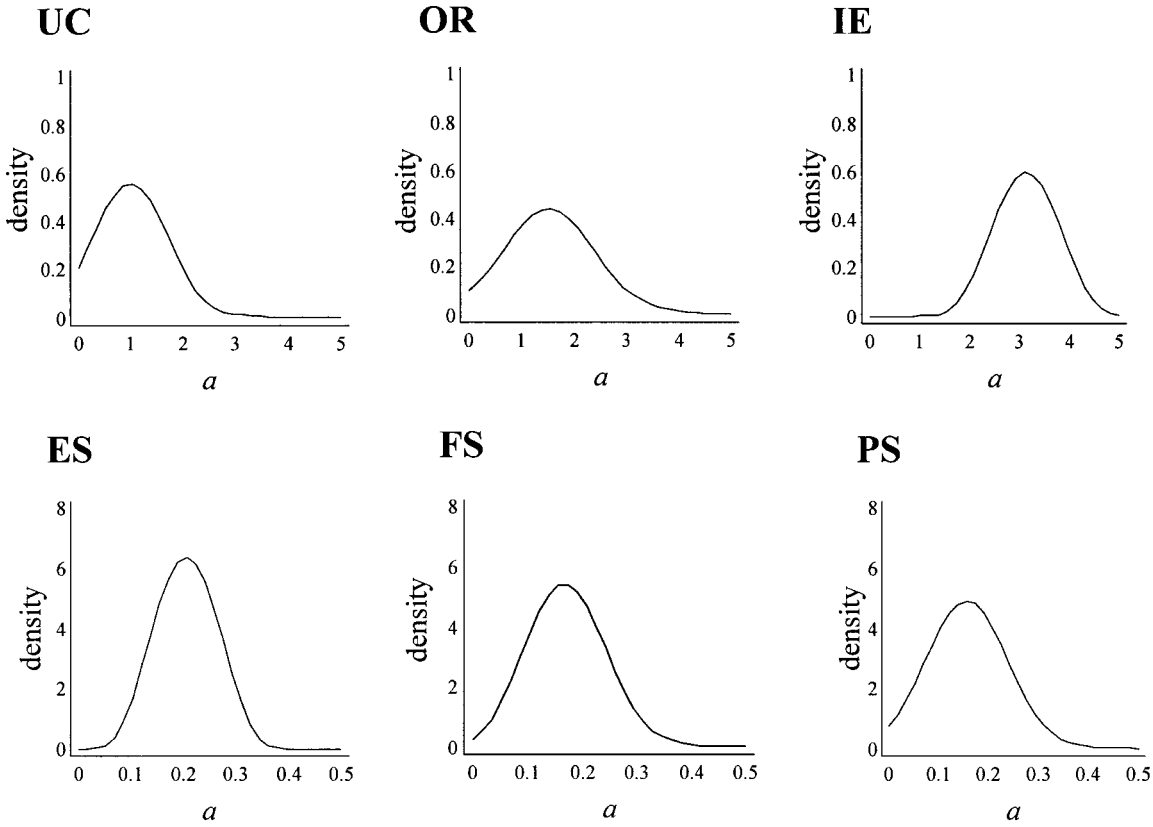

FIGURE 5.—Marginal posterior distributions for major gene additive effects: uterine capacity (UC), ovulation rate (OR), number of implanted embryos (IE), embryo survival (ES), fetal survival (FS), and prenatal survival (PS).

genes of large effect segregating in this population on IE and ES ($a > 1\sigma_p$), and major genes of relatively moderate effect affecting OR ($\frac{1}{2}\sigma_p \leq a \leq 1\sigma_p$), FS ($\frac{1}{2}\sigma_p \leq a \leq 1\sigma_p$), PS ($\frac{1}{2}\sigma_p \leq a \leq 1\sigma_p$), and UC ($a < \frac{1}{2}\sigma_p$). Further study is needed to confirm these results and to map this gene.

Thanks go to Pau Navarro for her assistance with the statistical analyses. We gratefully acknowledge the Ministerio de Educación y Ciencia (Spain) for granting M. J. Argente a "ayuda del Subprogramas de Estancias de Investigadores Españoles en Centros de Investigacion Extranjeros," which allowed her stay at Edinburgh. This study was supported by a CICYT (AGF98-0382-C02-01) and British Council Acciones Integradas(1999/2000/8504) grant. C.S.H. acknowledges support from the BBSRC.

LITERATURE CITED

ARGENTE, M. J., M. A. SANTACREU, A. CLIMENT and A. BLASCO, 1996 Selection for uterine efficiency in rabbits. 6th World Rabbit Congr. **2:** 241–244.

ARGENTE, M. J., M. A. SANTACREU, A. CLIMENT, G. BOLET and A. BLASCO, 1997 Divergent selection for uterine capacity in rabbits. J. Anim. Sci. **75:** 2350–2354.

ARGENTE, M. J., M. A. SANTACREU, A. CLIMENT and A. BLASCO, 2000 Genetic correlations between litter size and uterine capacity. 7th World Rabbit Congr. **A:** 333–338.

BENNET, G. L., and K. A. LEYMASTER, 1989 Integration of ovulation rate, potential embryonic viability and uterine capacity into a model of litter size in swine. J. Anim. Sci. **67:** 1230–1241.

BLASCO, A., 1996 Genetics of litter size and does fertility in the rabbit. 6th World Rabbit Congr. **2:** 219–227.

BLASCO, A., 2001 The Bayesian controversy in animal breeding. J. Anim. Sci. **79:** 2023–2046.

BLASCO, A., J. P. BIDANEL, G. BOLET, C. S. HALEY and M. A. SANTACREU, 1993 The genetics of prenatal survival of pigs and rabbits: a review. Livestock Prod. Sci. **37:** 1–21.

BLASCO, A., M. J. ARGENTE, C. S. HALEY and M. A. SANTACREU, 1994 Relationships between components of litter size in unilaterally ovariectomized and intact rabbit does. J. Anim. Sci. **72:** 3066–3072.

CASSADY, J. P., R. K. JOHNSON, D. POMP, G. A. ROHRER, L. D. VAN VLECK *et al.*, 2001 Identification of quantitative trait loci affecting reproduction in pigs. J. Anim. Sci. **79:** 623–633.

CHRISTENSON, R. K., K. A. LEYMASTER and L. D. YOUNG, 1987 Justification of unilateral hysterectomy-ovariectomy as a model to evaluate uterine capacity in swine. J. Anim. Sci. **67:** 738–744.

CLUTTER, A. C., M. K. NIELSEN and R. K. JOHNSON, 1990 Alternative methods of selection for litter size in mice. I. Characterization of base population and development of methods. J. Anim. Sci. **68:** 3536–3542.

CLUTTER, A. C., Y. L. KIRBY and M. K. NIELSEN, 1994 Uterine capacity and ovulation rate in mice selected 21 generations on alternative criteria to increase litter size. J. Anim. Sci. **72:** 577–583.

FAIN, P. R., 1978 Characteristics of simple sibship variance tests for the detection of major loci and application to height, weight and spatial performance. Ann. Hum. Genet. **42:** 109–120.

FALCONER, D. S., and T. F. C. MACKAY, 1996 *Introduction to Quantitative Genetics*, Ed. 4. Longman, Harlow, London.

GALLOWAY, S. M., K. P. MCNATTY, L. M. CAMBRIDGE, M. P. LAITINEN, J. L. JUENGEL *et al.*, 2000 Mutations in an oocyte-derived growth factor gene (BMP15) cause increased ovulation rate and infertility in a dosage-sensitive manner. Nat. Genet. **25:** 279–283.

GELMAN, A., and D. B. RUBIN, 1992 Inference for iterative simulation using multiple sequences. Stat. Sci. **7:** 457–472.

HILL, W. H., and S. KNOTT, 1990 Identification of major genes with large effects, pp. 447–494 in *Advances in Statistical Methods for*

Genetic Improvement of Livestock, edited by D. Gianola and K. Hammond. Springer-Verlag, Berlin.

Janss, L. L. G., R. Thompson and J. A. M. Van Arendonk, 1995 Application of Gibbs sampling for inference in a mixed major gene-polygenic inheritance model in animal population. Theor. Appl. Genet. **91:** 1137–1147.

Janss, L. L. G., A. M. Johan, J. A. M. Van Arendonk and E. W. P. Brascamp, 1997 Segregation analysis for presence of major genes affecting growth, backfat, and litter size in Dutch Meishan crossbreds. J. Anim. Sci. **75:** 2864–2876.

Lynch, M., and B. Walsh, 1998 *Genetics and Analysis of Quantitative Traits*. Sinauer Associates, Sunderland, MA.

Messer, L., M. Mousel, M. Nielsen and D. Pomp, 1999 Identification of quantitative trait loci (QTL) affecting components of litter size in unilaterally ovariectomized mice. Plant & Animal Genome VII Conference, San Diego, Vol. 1, p. 430.

Nielsen, M. K., 1994 Selection experiments for reproductive rate in mice. Proceedings of the 5th World Congress on Genetics Applied to Livestock Production, Guelph, ON, Canada, Vol. 19, pp. 219–225.

Raftery, A. E., and S. Lewis, 1992 How many iterations in the Gibbs sampler?, pp. 763–773 in *Bayesian Statistics 4*, edited by J. O. Berger, J. M. Bernardo, A. P. Dawid and A. F. M. Smith. Oxford University Press, Oxford.

Rathje, T. A., L. T. Gama and R. K. Johnson, 1993 Statistical evidence for the existence of genes with large effects on ovulation rate and number of fetuses in pig selected for ten generations using an index of embryo survival and ovulation rate. J. Anim. Sci. **71** (Suppl. 1): 94.

Rohrer, G. A., J. J. Ford, T. H. Wise, J. L. Vallet and R. K. Christenson, 1999 Identification of quantitative trait loci affecting females reproductive traits in a multigeneration Meishan-White composite swine population. J. Anim. Sci. **77:** 1385–1391.

Rothschild, M. F., and J. P. Bidanel, 1998 Biology and genetics of reproduction, pp. 313–343 in *The Genetics of the Pig*, edited by M. F. Rothschild and A. Ruvinsky. C.A.B International, Cambridge, UK.

Rothschild, M. F., C. Jacobson, D. Vaske, C. Tuggle, L. Wang *et al.*, 1996 The estrogen receptor locus is associated with a major gene influencing litter size in pigs. Proc. Natl. Acad. Sci. USA **93:** 201–205.

Santacreu, M. A., M. P. Viudes and A. Blasco, 1990 Evaluation par coelioscopie des corps jaunes et des embryons. Influence sur la taille de portée chez la lapine. Reprod. Nutr. Dev. **30:** 583–588.

SAS Institute, 1996 *SAS/STAT User's Guide (Release 6.12)*. SAS Institute, Cary, NC.

Sokal, R. R., and F. J. Rohlf, 1995 *Biometry*, Ed. 2. W. H. Freeman, New York.

Sorensen, D., 1996 *Gibbs Sampling in Quantitative Genetics*. Internal Report No. 82, Danish Institute of Animal Science, Foulum, Denmark.

Villanueva, B., R. Pong-Wong, B. Grundy and J. A. Woolliams, 1999 Potential benefit from using an identified major gene in BLUP evaluation with truncation and optimal selection. Genet. Sel. Evol. **31:** 115–133.

White, B. R., D. G. Mclaren, P. J. Dziuk and M. B. Wheeler, 1993 Age at puberty, ovulation rate, uterine length, prenatal survival and litter size in chinese Meishan and Yorkshire females. Theriogenology **40:** 85–97.

Wilkie, P. J., A. A. Paszed, C. W. Beattie, L. J. Alexander, M. B. Wheeler *et al.*, 1999 A genomic scan of porcine reproductive traits reveals possible quantitative trait loci (QTLs) for number of corpora lutea. Mamm. Genome **10:** 573–578.

Wilson, T., W.-X. Yang, J. L. Juengel, I. K. Ross, J. M. Lumsden, *et al.*, 2001 Highly prolific Booroola sheep have a mutation in the intracellular kinase domain of bone morphogenetic protein in IB receptor (ALK-6) that is expressed in both oocytes and granulosa. Biol. Reprod. **64:** 1225–1235.

Communicating editor: J. A. M. van Arendonk