Effect of the oestrogen receptor (*ESR*) gene on reproductive traits of Large White, White Meaty and Landrace pigs

R. Omelka¹, M. Bauerová¹, J. Mlynek², B. Buchová³, D. Peškovičová³, J. Bulla³

ABSTRACT: We investigated the effect of oestrogen receptor (*ESR*) gene on total number of born (TNB), number of born alive (NBA) and number of weaned (NW) piglets in Large White (LW), White Meaty (WM) and Landrace (L) sows from six Slovak breeding farms. Detection of *ESR* (*Pvu II*) genotypes was performed by the PCR-RFLP method. The frequencies of favourable *B* allele in LW, WM and L were 0.33, 0.25 and 0.08, respectively. We found a highly significant effect of *ESR* locus on NW ($P \le 0.01$) in LW, however, we observed a negative effect of *BB* genotype on the trait. In WM, a positive association of *B* allele with TNB, NBA and NW was found but the differences were not confirmed statistically. A highly significant effect of *ESR* locus on TNB, NBA and NW ($P \le 0.01$) was identified only in L breed. We found an increase of +0.62 ± 0.18 (TNB), +0.65 ± 0.18 (NBA) and +0.51 ± 0.16 (NW) pigs per copy of *B* allele.

Keywords: oestrogen receptor gene; reproductive traits; litter size; pig

Reproduction plays a key role in the successful production of farm animals. Selection for increased litter size is rather difficult because of low heritability of this trait. So the identification of genes or genetic markers associated with reproductive traits in pigs could have a great economic impact on pork production. Such revealed genetic polymorphisms could be employed in marker-assisted selection programs to improve reproductive efficiency.

The oestrogen receptor (*ESR*) gene has been identified as a major gene for litter size in Meishan and Large White breeds. Rothschild et al. (1996) described a polymorphism (substitution of adenosine and thymidine for guanosines) between the third and fourth exons of the gene which produces two common alleles, *A* and *B*. The preferred *B* allele was associated with the first parity increase of 1.15 pigs

per litter for each copy of the allele in Meishan crosses. In later parities, the effect of B allele was about 0.5 pigs per litter. The B allele also segregates in Large White breed and the effect varies from 0.31 to 0.42 pigs per litter in the first and later parities (Short et al., 1997).

The aim of our work was to determine the frequencies of *ESR* genotypes and alleles and to estimate their associations with some reproductive traits in Large White, White Meaty and Landrace pigs from Slovakia.

MATERIAL AND METHOD

We analysed 155 sows of Large White (LW), 134 sows of White Meaty (WM) and 132 sows of

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¹Department of Botany and Genetics, Constantine the Philosopher University, Nitra, Slovak Republic

²Slovak University of Agriculture, Nitra, Slovak Republic

³Research Institute of Animal Production, Nitra, Slovak Republic

Landrace (L) breeds from six Slovak breeding farms. The groups of pigs consisted of nucleus herds and involved 37, 23 and 14 lines of LW, WM, and L, respectively. The samples were collected at random from the herds.

Genomic DNA was isolated from the hair roots by silica matrix according to Bauerová et al. (1999). Genetic polymorphism at *ESR* (*Pvu II*) was genotyped by the PCR-RFLP method described by Short et al. (1997).

Allele and genotype frequencies were calculated and Hardy-Weinberg equilibrium was tested by comparing expected and observed genotype frequencies using a chi-square goodness of fit test. A total of 1 664 litters from 421 sows (1st–8th litters) were included in litter size analyses in which the following traits were recorded: total number of born (TNB), number of born alive (NBA) and number of weaned (NW) piglets. General statistics for all tested traits are given in Table 1.

The associations between *ESR* genotypes and TNB, NBA, NW were evaluated for each breed separately by a linear model as follows (SAS[®] v. 8.2; 2002):

$$y_{ijklm} = GS_i + GB_j + PAR_k + hys_l + anim_m + b_1 \times vop_{ijklm} + b_2 \times vop_{ijklm}^2 + e_{ijklm}$$

where: y = trait value

GS = ESR genotype of sow

GB = ESR genotype of mated boar

PAR = parity effect

hys = random herd-year-season effect

anim = random effect of animal

 $b_1 \times vop_{ijklm} + b_2 \times vop_{ijklm}^2$ = linear and quadratic regression on age at farrowing

e = random error

The variance components for random effects were estimated by MIXED procedure and REML method. Differences of Least-Squares Means (LSM) were tested by Scheffe multiple range test. Linear model used in our study was close to the statistical model used in routine genetic evaluation of litter size traits in pigs in Slovakia. Additional effects of genotype (according to ESR gene) were included in the model as genotype of sow (GS) and genotype of boar (GB). According to analysis of variance the effects of genotype of sow and genotype of boar were statistically significant only for L breed. Other effects included in the model were not statistically significant for the given data set. As we wanted to keep the model as close as possible to a routine

Table 1. General statistics for all tested traits

Trait	Mean	S _D	Min	Max				
Large White (<i>n</i> = 155; NL = 640)								
TNB	11.93	1.55	6.00	18.00				
NBA	10.95	0.99	4.00	17.00				
NW	9.19	0.76	4.00	15.00				
White Meaty (<i>n</i> = 134; NL = 489)								
TNB	11.68	1.34	4.00	18.00				
NBA	11.12	1.34	4.00	16.00				
NW	10.02	0.86	4.00	15.00				
Landrace (<i>n</i> = 132; NL = 535)								
TNB	10.46	0.48	5.00	15.00				
NBA	10.10	0.48	5.00	14.00				
NW	9.58	0.33	5.00	14.00				

TNB – total number of born; NBA – number of born alive; NW – number of weaned piglets; S_D – standard deviation; Min – minimum value; Max – maximum value; NL – number of litters

genetic evaluation model all effects were kept in the model. The proportion of variability explained by random animal effect ranged from 10 to 20% of total variability for different breeds in our study. The proportion of residual variability (the part of variability which could not be explained by other effects in the model) was close to 80% of total variability. These values correspond to usually published values for the litter size traits in pigs.

RESULTS AND DISCUSSION

According to the general statistics (Table 1) WM breed showed the highest mean values of NBA and NW. TNB was found to be the best in LW.

The frequencies of *ESR* genotypes and alleles in tested pigs are given in Table 2. The locus was in Hardy-Weinberg equilibrium in all tested herds except herd IV (absence of *BB* genotype). The frequency of *B* allele, which is supposed to be positively associated with litter size (Rothschild et al., 1996; Short et al., 1997), was 0.33, 0.25 and 0.08 in LW, WM and L, respectively. Our results in LW could be compared with data published by Short et

Table 2 The frequencies of *ESR* genotypes and alleles in Large White (LW), White Meaty (WM) and Landrace (L) sows

Breed	77 1	Number ofsows	ESR genotypes (%)			2	ESR alleles	
	Herd		AA	AB	BB	χ^2	A	В
LW	I	76	46.05	47.37	6.58	1.49	0.697	0.303
	II	79	39.24	49.37	11.39	0.49	0.639	0.361
	Total	155	42.58	48.39	9.03	0.82	0.668	0.332
WM	III	65	46.15	49.23	4.62	3.61	0.708	0.292
	IV	69	59.42	40.58	0	6.48*	0.797	0.203
	Total	134	52.99	44.78	2.23	4.25	0.754	0.246
L	V	47	89.36	10.64	0	0.31	0.947	0.053
	VI	85	81.18	18.82	0	1.08	0.906	0.094
	Total	132	84.09	15.91	0	0.74	0.920	0.080

 $[\]chi^2$ – chi-square test; * $P \le 0.05$

Table 3. The effect of ESR genotypes (least-squares means ± standard error) on total number of born (TNB), number of born alive (NBA) and number of weaned (NW) piglets in Large White (LW), White Meaty (WM) and Landrace (L)

Breed	ESR genotypes	Number _ of litters	TNB		NBA		NW	
			LSM ± S _E	P	LSM ± S _E	P	LSM ± S _E	\overline{P}
LW	AA	264	11.64 ± 0.54		10.85 ± 0.42		9.17 ± 0.33	
	AB	308	12.04 ± 0.54		11.03 ± 0.41		9.19 ± 0.33	
	BB	68	11.88 ± 0.62		10.89 ± 0.47		8.35 ± 0.37	
	AB- AA		$+0.40 \pm 0.23$	0.09	$+0.18 \pm 0.17$	0.29	$+0.02 \pm 0.13$	0.87
	BB-AA		$+0.24 \pm 0.40$	0.55	$+0.04 \pm 0.29$	0.90	$-0.82 \pm 0.23^{**}$	< 0.01
	BB-AB		-0.16 ± 0.39	0.67	-0.14 ± 0.28	0.62	$-0.84 \pm 0.22^{**}$	< 0.01
WM	AA	259	10.48 ± 1.07		9.29 ± 1.05		8.40 ± 0.74	
	AB	219	10.61 ± 1.03		9.29 ± 1.01		8.58 ± 0.72	
	BB	11	10.73 ± 1.18		9.42 ± 1.03		8.63 ± 0.83	
	AB- AA		$+0.13 \pm 0.33$	0.70	-0.01 ± 0.30	0.99	$+0.18 \pm 0.22$	0.42
	BB-AA		$+0.25 \pm 0.38$	0.49	$+0.13 \pm 0.42$	0.86	$+0.23 \pm 0.32$	0.52
	BB-AB		$+0.12 \pm 0.26$	0.61	$+0.13 \pm 0.40$	0.82	$+0.05 \pm 0.21$	0.92
L	AA	447	10.82 ± 0.58		10.37 ± 0.56		9.45 ± 0.51	
	AB	88	11.43 ± 0.59		11.02 ± 0.57		9.96 ± 0.52	
	AB- AA		+0.62 ± 0.18**	< 0.01	+0.65 ± 0.18**	< 0.01	+0.51 ± 0.16**	< 0.01

 $^{**}P \le 0.01$

al. (1997), who found a much higher frequency of *B* allele ranging from 0.64 to 0.74. Data acquired in Landrace based breeds showed a very low frequency of *B* allele (0.06 in Polish Landrace; Kmiec et al., 2002) or no *Pvu II* polymorphism (Drögemüller et al., 2001). In the Czech Republic, the *B* allele frequency varies from 0.27 to 0.40 in LW (Vrtková and Dvořák, 2001; Matoušek et al., 2003) and from 0.02 to 0.13 in L (Vrtková and Dvořák, 2001). No other study of the *ESR* gene breed has been published in WM and therefore we have no opportunity to confirm the results.

The effects of ESR genotypes on reproduction traits (least-squares means with standard errors) are presented in Table 3. There was a highly significant effect of ESR locus on NW ($P \le 0.01$) in LW, however, in contrast with other studies (Short et al., 1997) we observed a negative effect of BB genotype on the trait. We suppose this could be due to higher dependence of NW on environmental factors. The AB genotype showed a tendency to improve other tested traits but the results were not statistically significant. Short et al. (1997) reported a significant increase ($P \le 0.01$) in TNB and NBA by 0.42 (0.31) and 0.39 (0.31) pigs per litter in the first parity (later parities), respectively. In the Czech Republic, an increase by 0.14–1.75 (TNB) and 0.13-1.96 (NBA) was found in LW (Putnová et al., 2001; Matoušek et al., 2003).

A highly significant effect of ESR locus on TNB, NBA and NW ($P \le 0.01$) was identified in L breed. An increase by $+0.62 \pm 0.18$ (TNB), $+0.65 \pm 0.18$ (NBA) and $+0.51 \pm 0.16$ (NW) pigs per copy of B allele was found. The BB genotype was not evaluated because of its absence. It is also possible that the differences could be a consequence of dominant effect. Our results can be compared with Kmiec et al. (2002), who found a small and statistically insignificant effect of B allele in Polish Landrace breed.

This study gives primary information about associations between *ESR* genotypes and reproductive traits in WM breed. We could observe a positive association of *B* allele with TNB, NBA and NW but the differences were not confirmed statistically.

In our study, the magnitude of the *ESR* effect on reproductive traits varied in individual breeds. It is apparent that the background genetics of each different line plays an important role in the manner and magnitude of genetic control of these traits. The number of analysed pigs was also limited. It would be necessary to examine a larger sample pop-

ulation for a more conclusive evaluation of the *ESR* effect on litter size in tested populations.

The mechanism through which the polymorphism in the ESR gene affects litter size is not known yet. Results of Van Rens et al. (2000) suggest that a difference in litter size between the ESR genotypes is probably caused by a difference in foetal survival. Embryos of BB gilts had a significantly longer placenta than embryos of AA gilts. A higher chance for placental insufficiency in AA gilts could be a cause of higher foetal mortality as compared with BB gilts.

The use of genetic markers associated with reproductive traits can lead to increased rates of genetic response and bring a higher economic profit to pig industry. However, the results of our study showed the possibility of *ESR* utilization in marker-assisted selection to increase litter size only in Landrace pigs.

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Corresponding Author

RNDr. Radoslav Omelka, PhD., Department of Botany and Genetics, Constantine the Philosopher University, Trieda A. Hlinku 1, 949 74 Nitra, Slovak Republic Tel. +421 137 414 182, e-mail: romelka@pobox.sk