

Short Communication

Correlation between an oestrogen receptor gene and reproductive traits in purebred and crossbred pig populations

**F.E. Li., C.Y. Deng, S.W. Jiang, M.G. Lei, B. Zuo, R. Zheng,
J.L. Li, D.Q. Xu and Y.Z. Xiong[#]**

Key Laboratory of Pig Genetics and Breeding, Ministry of Agriculture, China
College of Animal Science, Huazhong Agricultural University, Wuhan 430070, P. R. China

Abstract

The relationship between an oestrogen receptor (ESR) gene and reproductive traits in 11 Large White (LW), 19 Landrace (L), 22 Meishan (MS), 22 Meishan × Large White (MS × LW) F₁, 26 Large White × Meishan (LW × MS) F₁, 16 Landrace × Large White (L × LW) F₁ and nine Large White × Landrace (LW × L) F₁ female populations was studied. Reproductive characteristics and some performance data were analyzed for associations with ESR genotype, parity and breed. The ESR genotype was associated with length of oviduct (LO). Uterine weight (UW), LO and the weight of two ovaries (OW) of purebred Meishan and LW × MS females were higher than those of purebred Large White and MS × LW females. In Large White and Meishan crosses, crossbreds with Meishan dams had higher UW, LO, uterine horn length (LUH) and OW than pigs with Large White dams. Significant effects of paternal breed of sire were observed for LO and OW. In Large White and Landrace combinations, differences in values between purebred and crossbred animals were found for UW and LO. However, maternal breed effects were not detected for UW, LO, LUH, OW. A significant effect of paternal breed of sire was observed for LO. Females with Landrace sires had a larger LO than females with Large White sires.

Keywords: Pig, ESR locus, female reproductive tract components, breed effects

[#] Corresponding author. E-mail: lifener@mail.hzau.edu.cn

Improvement in reproduction traits of livestock species is limited when using traditional methods of genetic selection because of the low heritability and sex-limited nature of these traits. Marker-assisted selection programs could be included in genetic selection and be very effective in genetic improvement, especially for improvements in reproductive traits. Using the candidate gene approach, the oestrogen receptor (ESR) gene (Rothschild *et al.*, 1994), the follicle stimulating hormone-β subunit (FSH-β) gene (Li *et al.*, 1998) and other genes have been shown to be associated with reproductive efficiency in pigs. The ESR gene was the first candidate or major gene studied, and in recent years there are many reports about the ESR locus and its relationship with reproductive traits and some performance traits (Rothschild *et al.*, 1994; Southwood *et al.*, 1995; Short *et al.*, 1997; Southwood *et al.*, 1999; Li *et al.*, 2004). However, little attention has been focused on the relationship between the ESR gene and components of reproductive traits. The objective of the current study was to evaluate the effect of ESR genotype and breed on several of these previously less intensively investigated reproductive traits.

Seven pig populations, *viz.* Large White (LW × LW) (n = 11), Landrace (L × L) (n = 19), Chinese Meishan (MS × MS) (n = 22) and some of their crosses: the Large White × Landrace (LW × L) (n = 9), Landrace × Large White (L × LW) (n = 16), Large White × Meishan (LW × MS) (n = 26) and Meishan × Large White (MS × LW) (n = 22) F₁ gilts were created in 2001 from purebred Large White, Landrace and Chinese Meishan seed stock. All animals were raised on the farm owned by the Huazhong Agricultural University, and received the same diet. Experimental animals were slaughtered at about 180 days of age. Reproductive tract characteristics, including length of uterine horn (LUH), uterine weight (UW), weight of two ovaries (OW) and length of oviduct (LO) were recorded. LUH were measured according to the method of Lin (1992). Internal fat percentage (IFP), average back-fat thickness over three points (ABFT), loin eye area (LEA) and lean meat percentage (LMP) were measured according to the methods of Xiong & Deng

(1999). A blood sample was collected from each animal and DNA was extracted from peripheral lymphocytes (Xiong, 1999). The ESR genotype was determined according to the PCR procedure of Short *et al.* (1997) and Li *et al.* (2000). Allelic and genotypic frequencies were calculated within each population.

All the data obtained were analyzed using the General Linear Model (GLM) procedure of the SAS package (SAS, 1999). The Pair-wise t-test was used to test the differences between ESR genotypes. Both additive and dominance effects were estimated using the REG procedure of SAS, where the additive effect was estimated as -1, 0, 1 for AA, AB, BB, respectively, and the dominance effect represented as 1, -1, 1 for AA, AB, BB, respectively (Liu, 1998).

Allelic and genotypic frequencies for all animals included in the study are shown in Table 1. Large White pigs in the present study were found to have a B allele frequency of 0.545. In comparison, the frequency of the B allele of other Large White pig lines ranged from 0.41 to 0.57 (Short *et al.*, 1997). The ESR B allele frequency for Meishan was 0.591, in agreement with results obtained by Li *et al.* (2000). The ESR B allele frequency for Landrace animals was 0 in the present study, similar to the observation by Drogemuller *et al.* (2001), while Kmiec *et al.* (2002) found the frequency of the B allele in the Landrace to be 0.058 and Chen (1999), 0.159. This demonstrates that the ESR B allele has different frequencies in different pig populations.

Table 1 The allele (A and B) and genotype frequencies of the oestrogen receptor gene (ESR) in the pig populations included in the present study

Populations	No	Allele frequencies		Genotype frequencies		
		A	B	AA	AB	BB
L × L	19	1.000	0.000	1.000	0.000	0.000
LW × LW	11	0.455	0.545	0.272	0.364	0.364
MS × MS	22	0.409	0.591	0.091	0.636	0.273
MS × LW	22	0.250	0.750	0.136	0.227	0.637
LW × MS	26	0.615	0.385	0.462	0.308	0.230
L × LW	16	0.625	0.375	0.250	0.750	0.000
LW × L	9	0.556	0.444	0.111	0.889	0.000

L - Landrace; LW - Large White; MS - Meishan

Previous studies have shown the ESR B allele was associated with an increased number of piglets born and piglets born alive per litter (Rothschild *et al.*, 1996; Short *et al.*, 1997). In the present study the ESR genotype was associated with LO ($P < 0.05$) (Table 2) but not with OW, LUH or UW. Our previous research results on Large White × Meishan F_2 offspring showed that the UW of AB gilts was lower than that of AA gilts ($P < 0.05$), with an additive effect of -18.058 g (Li *et al.*, 2004). Isler *et al.* (1999) showed that the ESR genotypes were not associated with UW ($P > 0.05$). Comparing our results with those of Isler *et al.* (1999), it can be concluded that ESR effects can differ between populations. The pigs used by Isler *et al.* (1999) were Yorkshire (Y × Y), Large White (LW × LW) and crossbreds (LW × Y, Y × LW) at approximately 75 days of gestation, while the pigs used in the present study were non-pregnant gilts from Large White, Landrace, Chinese Meishan breeds and their two-way cross populations. No evidence was detected of an association of the ESR B allele with ABFT, LEA, and LMP. Favourable pleiotropic effects were detected for back fat thickness (BF) ($P < 0.05$) with the additive effect of -0.11 mm per copy of the B allele (Short *et al.*, 1997). Earlier research of Rothschild *et al.* (1996) suggested that the effect of the favourable B ESR allele might be antagonistic to BF.

For the LW × MS cross, breed effects were found for several of the traits studied (Table 3). Purebred Meishan and LW × MS females had higher UW, LUH, OW than purebred Large White and MS × LW females. Purebred Large White and Meishan pigs differed significantly ($P < 0.05$) in all traits tested except

Table 2 Least squares means (LSM) and standard errors (s.e.) for selected reproductive traits and some performance traits with respect to oestrogen receptor (ESR) genotypes

ESR genotypes	LSM ± s.e.			Additive effect, a	Dominant effect, d	Dominance D
	AA	AB	BB			
LO/cm	19.50 ^a ± 0.631	19.77 ^a ± 0.590	21.54 ^b ± 0.854	1.018	-0.748	-0.735
LUH/cm	101.34 ± 4.482	98.31 ± 4.190	102.15 ± 6.064	0.403	-3.441	-8.538
OW/g	10.43 ± 0.615	11.97 ± 0.657	10.43 ± 0.889	0.000	1.544	-
UW/kg	0.44 ± 0.018	0.45 ± 0.017	0.45 ± 0.024	0.005	-0.007	-1.400
IFP/%	3.30 ^a ± 0.137	3.21 ^a ± 0.112	2.78 ^b ± 0.156	-0.261	0.176	-0.661
ABFT/cm	2.06 ± 0.066	2.00 ± 0.054	2.07 ± 0.074	0.004	-0.069	-17.25
LEA/cm ²	37.08 ± 0.770	36.83 ± 0.628	38.00 ± 0.874	0.463	-0.709	-1.531
LMP/%	63.15 ± 0.452	63.38 ± 0.368	64.00 ± 0.512	0.424	-0.193	-0.455

Note: additive effect (a) = (BB-AA)/2; dominance effect (d) = AB-(AA+BB)/2; dominance degree (D) = d/a

Means in the same row with different superscripts differ significantly at P < 0.05

LO - length of oviduct; LUH - length of uterine horn; OW - weight of two ovaries; UW - uterine weight;

IFP - internal fat percentage; ABFT - average back-fat thickness; LEA - loin eye area; LMP - lean meat percentage

Table 3 Least squares means and standard errors for reproductive traits influenced by heterosis, maternal or paternal effects in Large White (LW) and Meishan (MS) cross combinations

Population	No	UW/kg	LO/cm	OW/g	LUH/cm	IFP/%	ABFT/cm	LEA/cm ²	LMP/%
LW x LW	11	0.207 ^a ± 0.042	18.35 ^{ac} ± 1.07	7.00 ^a ± 1.23	47.22 ^a ± 10.03	2.05 ^a ± 0.22	1.603 ^a ± 0.105	43.22 ^a ± 1.23	71.46 ^a ± 0.72
MS x MS	22	0.592 ^b ± 0.031	20.91 ^b ± 0.80	10.47 ^b ± 1.45	129.8 ^b ± 7.47	5.71 ^b ± 0.16	2.784 ^b ± 0.078	17.17 ^b ± 0.92	44.99 ^b ± 0.54
MS x LW	22	0.298 ^{ac} ± 0.031	15.79 ^a ± 0.80	7.08 ^a ± 0.93	68.42 ^{ad} ± 5.74	4.07 ^c ± 0.16	2.346 ^c ± 0.078	28.65 ^c ± 0.91	56.13 ^c ± 0.54
LW x MS	26	0.619 ^b ± 0.027	20.97 ^b ± 0.71	14.93 ^b ± 0.81	132.10 ^b ± 6.62	4.23 ^c ± 0.15	2.585 ^c ± 0.069	31.89 ^c ± 0.81	58.24 ^c ± 0.48
H		-0.133	-0.148	0.063	-0.261	0.07	0.124	0.003	-0.018
Pure vs. crosses ¹		0.835	0.162	0.062	0.725	0.499	0.331	0.098	0.209
Maternal effect P-value ²		< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001
Paternal effect P-value ³		0.734	< 0.0001	0.005	0.509	< 0.0001	0.011	< 0.0001	< 0.0001

¹For the linear contrast for purebred animals vs. crossbred animals, where H₀ = no differences in the indicated trait between purebred and crossbred animals (Isler *et al.*, 2002)

²For the linear contrast for maternal effects, where H₀ = no differences in the indicated trait between animals with a different breed of dam (Isler *et al.*, 2002)

³For the linear contrast for paternal effects, where H₀ = no differences in the indicated trait between animals with a different breed of sire (Isler *et al.*, 2002)

LO - length of oviduct; LUH - length of uterine horn; OW - weight of two ovaries; UW - uterine weight; IFP - internal fat percentage; ABFT - average back-fat thickness; LEA - loin eye area; LMP - lean meat percentage

for LO (Table 3). No significant difference in values between purebred and crossbred animals was detected for all the test traits ($P > 0.05$). Maternal breed effects were also found for all tested traits. Animals with Meishan dams differ ($P < 0.01$) from those with Large White dams for all tested traits. Significant ($P < 0.01$) effects of paternal breed of sire were observed for LO, OW, IFP, LEA and LMP.

Table 4 Least squares means and standard errors for reproductive traits influenced by heterosis, maternal or paternal effects in Landrace (L) and Large White (LW) cross combinations

Population	No	UW/kg	LO/cm	OW/g	LUH/cm	IFP/%	ABFT/cm	LEA/cm ²	LMP/%
L x L	18	0.232 ± 0.040	20.31 ± 1.02	7.01 ± 1.18	46.70 ± 9.53	2.02 ± 0.21	1.618 ± 0.010	43.21 ^a ± 1.17	70.49 ^a ± 0.69
LW x LW	11	0.207 ± 0.042	18.35 ± 1.07	7.00 ± 1.23	47.22 ± 10.03	2.05 ± 0.22	1.603 ± 0.105	43.22 ^a ± 1.23	71.46 ± 0.72
L x LW	16	0.225 ± 0.037	21.08 ± 0.95	6.63 ± 1.14	65.53 ± 8.93	2.03 ± 0.20	1.842 ^a ± 0.093	49.54 ^b ± 1.1	69.80 ^a ± 0.64
LW x L	9	0.242 ± 0.049	21.55 ± 1.26	6.55 ± 1.54	59.75 ± 11.80	1.58 ± 0.26	1.523 ^b ± 0.124	47.43 ^b ± 1.45	73.46 ^b ± 0.85
H		-0.064	-0.103	-0.334	0.059	-0.114	0.045	0.122	0.009
Pure vs. crosses ¹		<.0001	0.027	0.143	0.317	0.595	0.130	0.003	0.519
Maternal effect P-value ²		0.141	0.474	0.411	0.330	0.104	0.065	0.347	0.016
Paternal effect P-value ³		0.476	0.012	0.170	0.482	0.044	0.019	0.084	0.002

¹For the linear contrast for purebred animals vs. crossbred animals, where H_0 = no differences in the indicated trait between purebred and crossbred animals (Isler *et al.*, 2002)

²For the linear contrast for maternal effects, where H_0 = no differences in the indicated trait between animals with a different breed of dam (Isler *et al.*, 2002)

³For the linear contrast for paternal effects, where H_0 = no differences in the indicated trait between animals with a different breed of sire (Isler *et al.*, 2002)

LO - length of oviduct; LUH - length of uterine horn; OW - weight of two ovaries; UW - uterine weight; IFP - internal fat percentage; ABFT - average back-fat thickness; LEA - loin eye area; LMP - lean meat percentage

As for the Large White and Landrace crosses, significant differences in values between purebred and crossbred animals were found for UW ($P < 0.01$), LO ($P < 0.05$) and LEA ($P < 0.01$) (Table 4). Maternal breed effects were detected only for LMP ($P < 0.05$). A significant effect of paternal breed of sire was observed for LO, IFP and LMP ($P < 0.05$).

The sample size in the present study was limited due to practical problems of collecting the samples. Further research should be conducted on larger numbers of pigs. The results presented here will allow researchers to understand some aspects of ESR genetic effects and breed effects on reproduction and production traits and how the ESR gene can be incorporated as a selection tool in pig genetic improvement programs.

Acknowledgements

This research was supported by the Key Project of the Chinese Ministry of Education (104132), and Science and Technology Innovation Foundation of Huazhong Agricultural University and the National High Science and Technology Foundation of China (2002AA211041).

References

- Chen, K.F., 1999. Effect of ESR and FSH- β haplotypes on litter size in pigs. Master thesis, Jiangxi Agricultural University. (in Chinese).
- Drogemuller, B., Hamann, H. & Distl, O., 2001. Candidate gene markers for litter size in different German pig lines. *J. Anim. Sci.* 79, 2565-2570.
- Isler, B.J., Irvin, K.M., Neal, S.M., Moeller, S.J., Davis, M.E. & Meeker, D.L., 1999. Examination of the relationship between the estrogen receptor gene and reproductive tract components in swine. Special circular-Ohio Agricultural Research and Development Center. 171, 54-59.
- Isler, B.J., Irvin, K.M., Neal, S.M., Moeller, S.J. & Davis, M.E., 2002. Examination of the relationship between the estrogen receptor gene and reproductive traits in swine. *J. Anim. Sci.* 80, 2334-2339.
- Kmiec, M., Dvorak, J. & Vrtkova, I., 2002. Study on a relation between estrogen receptor (ESR) gene polymorphism and some pig reproduction performance characters in Poland Landrace breed. *Czech. J. Anim. Sci.* 47 (5), 189-193.
- Li, F.E., Xiong, Z., Deng, C.Y., Zheng, Y.R. & Qu, Y.C., 2000. Study on the PCR-RFLP at Estrogen Receptor (ESR) locus in different pig breeds. *Journal of Huazhong Agricultural University (P. R. China)*, 19 (1), 37-39. (in Chinese).
- Li, F.E., Lei, M.G., Zheng, R., Zuo, B., Jiang, S.W., Deng, C.Y. & Xiong, Y.Z. 2004. The effects of estrogen receptor locus on reproduction tracts components and performance traits in large white \times Meishan F2 offspring. *Asian-Aust. J. Anim. Sci.* 17, 1223-1226.
- Li, N., Zhao, Y., Xiao, L., Zhang, F.J., Chen, Y.Z., Dai, R.J., Zhang, J.S., Shen, S.Q., Chen, Y.F. & Wu, C.X., 1998. Candidate gene analysis for identification of genetic loci controlling litter size in swine. In: *Proc. 6th World Congr. Genet. Appl. Livest. Prod. Armidale, Australia*, 183-186.
- Lin, H., 1992. *Porcine Dissection Map*. 1st ed. Agricultural Publishers of P. R. China, Beijing. p. 158. (in Chinese).
- Liu, B.H., 1998. *Statistical Genomics: Linkage, Mapping, and QTL Analysis*. CRC Press, LLC 404-409.
- Rothschild, M.F., Jacobson, C., Vaske, D.A., Tuggle, C.K., Short, T.H., Sasaki, S., Eckardt, G.R. & McLaren, D.G., 1994. A major gene for litter size in pigs. In: *Proc. 5th World Congr. Genet. Appl. Livest. Prod. Guelph, Canada*. 21, 225-228.
- Rothschild, M., Jacobson, C., Vaske, D., Tuggle, C., Wang, L., Short, T., Eckardt, G., Sasaki, S., Vincent, A., McLaren, D., Southwood, O., Van der Steen, H., Mileham, A. & Plastow, G., 1996. The estrogen receptor locus is associated with a major gene influencing litter size in pigs. *Proc. Natl. Acad. Sci. USA*. 93, 201-205.
- SAS, 1999. *Statistical Analysis Systems user's guide: Version 8.0*, SAS Institute Inc., Cary, North Carolina, USA.
- Short, T.H., Rothschild, M.F., Southwood, O.I., McLaren, D.G., De Vries, A., Van der Steen, H., Eckardt, G.R., Tuggle, C.K., Helm, J., Vaske, D.A., Mileham, A.J. & Plastow, G.S., 1997. Effect of the estrogen receptor locus on reproduction and production traits in four commercial pig lines. *J. Anim. Sci.* 75, 3138-3142.
- Southwood, O.I., Short, T.H., Plastow, G.S. & Rothschild, M.F., 1999. A genetic marker for litter size in Landrace-based pig lines. In: *Proc. 50th Eur. Assoc. Anim. Prod., Zurich, Switzerland*. 5:1.
- Southwood, O.I., Van der Steen, H.A.M., Mileham, A.J. & Cuthbert-Heavens, D., 1995. Evaluation of the oestrogen receptor (ESR) gene in Meishan synthetic and Large White pigs. *Eur. Assoc. Anim. Prod., Prague*, Sept. 4-7. (Abstr.).
- Xiong, Y.Z., 1999. *Pig Biochemistry and Molecular Genetics: Experimental Introduction*. 1st Ed. Agricultural Publishers of P. R. China, Beijing. pp. 39-41. (in Chinese).
- Xiong, Y.Z. & Deng, C.Y., 1999. *Principle and Method of Swine Tests*. 1st Ed. Agricultural Publishers of P. R. China, Beijing. (in Chinese).