

# Cortisol-binding globulin and meat quality in five European lines of pigs<sup>1</sup>

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**ABSTRACT:** The gene (*Cbg*) encoding cortisol-binding globulin (CBG) has been proposed as a candidate gene to explain genetic variation in cortisol secretion and carcass composition in pigs. The objective of this study was to evaluate the association between CBG and pork quality in 5 European breeding lines, Piétrain, Large White (LW), and Landrace purebred lines, a Duroc synthetic line, and a Meishan (MS) × LW advanced intercross. Cortisol-binding globulin maximum binding capacity (CBG-Bmax) was twice as high ( $P < 0.05$ ) in MS × LW pigs compared with the other lines. There was no ( $P \geq 0.364$ ) association between CBG-Bmax and carcass quality traits in Piétrain gilts, but CBG-Bmax was associated with increased loin yields in LW ( $P = 0.010$ ) and Landrace ( $P = 0.103$ ) gilts, decreased ham

yields ( $P = 0.082$ ) in Duroc gilts, and increased fat depth ( $P = 0.064$ ) and leaf fat ( $P = 0.001$ ) in MS × LW gilts. There was no association between CBG-Bmax and pork quality traits in Piétrain ( $P \geq 0.269$ ) and Duroc ( $P \geq 0.114$ ) gilts. Conversely, CBG-Bmax was associated with lighter (higher  $L^*$  values;  $P < 0.05$ ) pork in Landrace gilts, as well as lower ( $P \leq 0.055$ ) ultimate pH in the LM and semimembranosus, and a tendency for lower ( $P = 0.095$ )  $L^*$  values of pork from LW gilts. Within MS × LW pigs, CBG-Bmax was associated with increased drip loss ( $P = 0.001$ ) and decreased i.m. fat in the semimembranosus ( $P = 0.005$ ). Because drip loss is an economically important pork quality trait, results of this study could be used in the selection of improved water-holding capacity of pork from synthetic lines involving the MS breed.

**Key words:** cortisol-binding globulin, drip loss, gilt, meat quality, pork

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## INTRODUCTION

Pigs are exposed to a number of stressors before slaughter, and hormones released in response to stress may influence ultimate meat quality. Catecholamines and glucocorticoids can lead to aberrant meat quality by triggering rapid glycogenolysis and excessive lactate production (Tarrant, 1993; Foury et al., 2005). Large differences in hypothalamic-pituitary-adrenal axis activity have been shown within breeds (Ruis et al., 2000;

Geverink et al., 2002), as well as among breeds (Mormède et al., 1984; Désautés et al., 1997).

Meishan (MS) and Large White (LW) purebreds have been shown to differ largely in neuroendocrine traits, behavior, growth performance, body composition, and meat quality (Bidanel et al., 1990; Hay and Mormède, 1998; Désautés et al., 1999). Experimental F<sub>2</sub> crosses between these breeds have been used in several QTL studies. On several chromosomes, QTL for growth and fatness have been demonstrated (Bidanel et al., 2001; Milan et al., 2002). For fatness traits, a QTL was found on chromosome 7 in the swine leukocyte antigens (SLA) region (Milan et al., 2002). A nearby QTL showed association with basal and poststress cortisol levels (Désautés et al., 2002), as well as linkage to loin weight, and suggestive association with the percentage of ham and loin and estimated carcass lean content (Milan et al., 2002). The gene (*Cbg*) encoding cortisol-binding globulin (CBG) maps at the peak of this QTL. Cortisol-binding globulin is a circulating glycoprotein that binds most of the glucocorticoids with high affinity, and there

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is evidence that CBG influences both cortisol concentrations and carcass composition (Ousova et al., 2004).

The objective of the current study was to evaluate the association between CBG and pork quality in 5 European genetic lines. In one of these lines, a MS × LW advanced intercross, a microsatellite marker at the peak of the SLA region (Sw1856) was genotyped and included in the association study.

## MATERIALS AND METHODS

### *Subjects*

The study was done in 22 identical successive replicates. Gilts from each of 5 divergent genetic lines ( $n = 100$  per genetic line), including Piétrain, LW, and Landrace purebreds (Pig Improvement Co., Oxford, UK), a Duroc synthetic line, and a MS × LW advanced intercross population, were reared under the same environment and production regimen on a farm in France. Piglets were weaned at 21 d of age and kept in pens with concrete fully slatted floors, with a stocking density of 1.1 m<sup>2</sup>/pig. Pigs were fed commercial pelleted starter diet until weaning and, after weaning, liquid grower and finisher diets (Maisadour-Landal, Mont de Marsan, France). Gilts were weighed regularly, and ADG from 70 to 140 d of age was calculated.

### *Urine Collection and Cortisol Analysis*

Urine samples were collected for individual gilts between 0800 and 1000 under basal conditions at the farm for determination of cortisol concentrations. Samples were frozen immediately after addition of 10% EDTA (1 mL/40 mL of urine). Cortisol was assayed using a solid-phase extraction procedure, using an HPLC with UV absorbance detection, and expressed as a function of creatinine excretion, to correct for the variable dilution related to water intake (Hay and Mormède, 1997).

### *Lairage and Slaughter*

In each replicate, an equal number of gilts from each of 5 genetic lines was selected for slaughter. In general, there were 5 gilts per genetic line, resulting in 25 pigs that were slaughtered in 1 replicate, 7 replicates that contained 4 gilts per genetic line, and 1 replicate that contained 2 gilts per genetic line. Gilts were slaughtered at an average age and live weight of 188.8 d and 109.7 kg, respectively. Gilts that were selected for slaughter did not originate from the same litter. Gilts were fasted 8 h before a 10-h journey to a research abbatoir in Monells, Spain (IRTA Monells). After an overnight lairage where gilts had ad libitum access to water, gilts from different genetic lines were slaughtered in an alternating order according to normal commercial practices after weighing and CO<sub>2</sub> stunning.

### *Blood Sampling*

Three blood samples per gilt were collected at exsanguination for serum cortisol, CBG analysis, and DNA-

extraction. Blood samples for analysis of cortisol were collected in plain tubes, allowed to coagulate for 20 min, centrifuged at 3,000 × *g* at room temperature, and serum was frozen at -20°C until RIA for total cortisol (Désautés et al., 1997). Blood samples for analysis of CBG maximum binding capacity (CBG-**Bmax**) were collected in heparinized tubes, centrifuged at 3,000 × *g* at room temperature, and plasma samples were frozen at -20°C until analysis. Additionally, whole blood samples were collected in heparinized tubes and frozen at -20°C until DNA-extraction and genotyping.

### *Carcass Composition and Pork Quality Measurements*

Measurements of fat depth and muscle depth at 45 min postmortem were made using the Fat-O-Meat'er (SFK Technology, Denmark) inserted 60 mm from the midline between the 10th and 11th ribs. Fat and LM depths were used to calculate carcass lean content using the equation of Gispert and Diestre (1994). At 24 h postmortem, the right side of each carcass was sectioned between the 10th and 11th ribs, a digital image of the LM was collected, and LM area (cm<sup>2</sup>) was determined using the computer program of Pomar et al. (2001). Each left carcass side was subsequently fabricated into primal cuts and dissected following the method of Walstra and Merkus (1996).

The left side of each carcass was used to assess meat quality. Muscle pH was measured using a Crison portable meter equipped with a xerolyt electrode (Crison, Barcelona, Spain) in the LM at the level of the caudal edge of the last rib, and in the semimembranosus (**SM**) in the middle of the muscle in the exposed visible part, at 24 h postmortem. Drip loss from the LM was determined according to the method of Honikel (1998). Objective color measurements were collected 24 h postmortem on the exposed cut surface of the LM at the last rib, using a spectrophotometer Minolta C2002 (Minolta, Japan) in the CIE L\*a\*b\* space (CIE, 1976), using an illuminant D65 and 10° observer. Intramuscular fat content was measured in the LM and SM by near-infrared transmittance apparatus (Infratec 1265, Foss-Tecator, Höganäs, Sweden) according to the procedure of Gispert et al. (1997).

### *CBG-Binding Assay*

Each plasma sample was incubated for 30 min on a shaking platform at room temperature with an equal volume of dextran-coated charcoal [5% charcoal and 0.5% dextran (wt/wt)] to remove endogenous steroids. Charcoal was removed from the plasma by centrifugation at 9,000 × *g* for 15 min. The binding capacity of CBG in the stripped plasma was determined at 4°C using a modification of the solid-phase assay method described by Pugeat et al. (1984). Cortisol-binding globulin was absorbed from plasma onto a solid phase matrix of Con A-Sepharose (Amersham Biosciences, Upp-

**Table 1.** Least squares means for cortisol concentrations and cortisol-binding globulin maximum binding capacity (CBG-Bmax) in 5 divergent genetic lines

Trait	No.	Line					SEM
		MS × LW <sup>1</sup>	Piétrain	LW	Landrace	Duroc	
Urinary cortisol, ng/mg of creatinine	494	24.99 <sup>z</sup>	17.04 <sup>y</sup>	11.05 <sup>x</sup>	12.64 <sup>x</sup>	19.90 <sup>y</sup>	1.66
Total plasma cortisol, ng/mL	500	37.69 <sup>z</sup>	24.30 <sup>y</sup>	21.96 <sup>x</sup>	17.41 <sup>w</sup>	21.30 <sup>wx</sup>	2.61
Free plasma cortisol, ng/mL	500	5.81 <sup>z</sup>	4.18 <sup>y</sup>	4.05 <sup>xy</sup>	2.80 <sup>x</sup>	3.86 <sup>xy</sup>	0.68
CBG-Bmax, nM	473	60.36 <sup>z</sup>	33.02 <sup>y</sup>	27.01 <sup>x</sup>	27.69 <sup>x</sup>	26.82 <sup>y</sup>	1.94

<sup>w-z</sup>Within a row, least squares means that do not have common superscript letters differ,  $P < 0.05$ .

<sup>1</sup>MS × LW = Meishan × Large White intercross.

sala, Sweden) by incubating 100  $\mu$ L plasma with 250  $\mu$ L of a 50% (vol/vol) slurry of Con A-Sepharose in each of 8 scintillation vials. Incubation took place on a shaking platform and lasted 30 min. The gel sediment in each vial was washed once with 2 mL of 0.05 M Tris-buffer (pH 7.4). After centrifugation for 15 min at 3,000  $\times g$ , the aqueous phase was decanted to remove nonadsorbed proteins. Then, 200  $\mu$ L [<sup>3</sup>H]cortisol ( $35 \times 10^3$  counts per minute) and 200  $\mu$ L of different concentrations of unlabeled cortisol (0, 0.3125, 0.625, 1.25, 2.5, 4.0, 5.0, and 500 ng/vial) were added, followed by 2 mL of Tris-buffer. The mixture was incubated for 45 min on a shaking platform. After centrifugation for 15 min at 3,000  $\times g$ , the aqueous phase was decanted, 3.6 mL of scintillation fluid was added, and the radioactivity was determined. The binding capacity of CBG for cortisol was calculated by nonlinear regression, using “bound” as the quantity of cortisol specifically bound to the glycoproteins adsorbed to the gel, whereas “free” was the concentration of cortisol in the aqueous phase.

### DNA Isolation and Genotyping

For all MS × LW pigs, genomic DNA was isolated from whole blood using the Wizard Genomic DNA Purification Kit (Promega, Madison, WI). Genotyping of microsatellite Sw1856 in the SLA-region of SSC 7 was performed at the Laboratoire de Génétique Cellulaire (INRA, Castanet-Tolosan, France). Microsatellite locus was amplified from 20 ng of pig genomic DNA. The reaction was performed in 10  $\mu$ L with 0.2 U of Taq DNA Polymerase (Invitrogen, Cergy Pointoise, France), 200  $\mu$ M of dNTP (Invitrogen), 0.2  $\mu$ M of each primer, and 1.5 mM of MgCl<sub>2</sub>. Polymerase chain reactions were carried out in a PCR GeneAmp 9700 Dual 384 well (Applied Biosystems, Foster City, CA) under the following cycling conditions: initial denaturation for 5 min at 94°C, followed by 35 cycles of PCR amplification (each cycle consisted of 30 s at 94°C, 30 s at 55°C, and 45 s at 72°C), and a final extension for 30 min at 72°C. A 2- $\mu$ L aliquot of PCR product was mixed with 7.85  $\mu$ L of deionized formamide and 0.15  $\mu$ L of ABI Genescan 400 HD Rox size standard (Applied Biosystems). A Genesis RSP Low volume 200/8 robot and Gemini software (Tecan, Mannedorf, Switzerland) were used for all pipetting. Amplified fragments were denatured at 94°C

for 5 min before being resolved on Pop6 capillary on the 3700 ABI sequencer (Applied Biosystems). Fragment sizes and peak intensities were analyzed to identify microsatellite alleles with ABI Genotyper software (Applied Biosystems), and size range of the alleles was between 171 and 196 bp.

### Statistical Analyses

The experimental unit for all measurements was the individual pig. Free cortisol concentrations in plasma were calculated using the equation of Sodergard et al. (1982), and urinary cortisol and plasma cortisol data were normalised by log-transformation. Partial Pearson correlations, corrected for replicate, were calculated between urinary cortisol, plasma cortisol, and CBG-Bmax.

The effect of CBG-Bmax on meat quality variables in all 5 breeds was studied with the mixed model procedure of SAS (SAS Inst., Inc., Cary, NC). Genetic line, CBG-Bmax, and their interaction were included in the model as fixed effects; repetition was the lone random effect. Quadratic regression relationships, different for the 5 lines, between meat quality variables and CBG-Bmax were assumed. Because the quadratic terms of CBG-Bmax were not significant at an  $\alpha$  level of 5%, we removed them from the models, reporting the results for linear relationships only. The linear regression lines of the meat quality variables on CBG-Bmax were allowed to be different for the 5 genetic lines.

In addition to the previously described analysis for MS × LW gilts, the effect of CBG-Bmax on meat quality variables was analyzed using the Mixed procedure of SAS, which included the Sw1856-alleles as covariates with a fixed effect. One variable was included for each allele, with values of 0, 1, and 2 corresponding to the pig having 0, 1, or 2 copies of the allele in question. The CBG-Bmax was assumed to have a fixed effect and repetition a random effect.

## RESULTS AND DISCUSSION

### Cortisol Concentrations and CBG-Binding Capacity in 5 Genetic Lines

Both urinary and plasma cortisol concentrations were greatest ( $P < 0.05$ ) in MS × LW pigs. Urinary

**Table 2.** Pearson correlations (corrected for replicate) between cortisol concentrations and cortisol-binding globulin maximum binding capacity (CBG-Bmax) in 5 divergent genetic lines

Trait	Line									
	MS × LW <sup>1</sup>		Piétrain		LW		Landrace		Duroc	
	r	No.	r	No.	r	No.	r	No.	r	No.
Urinary cortisol to plasma cortisol	0.03	97	0.07	98	0.02	100	0.07	99	0.00	98
CBG-Bmax to urinary cortisol	0.02	95	0.17	89	-0.07	97	0.15	93	-0.02	93
CBG-Bmax to plasma cortisol	0.34***	96	0.16	91	0.21*	98	0.19	93	0.18	95

<sup>1</sup>MS × LW = Meishan × Large White intercross.

\* $P < 0.05$ ; \*\*\* $P < 0.001$ .

cortisol concentrations were least ( $P < 0.05$ ) in Landrace and LW gilts, whereas plasma total cortisol was least ( $P < 0.05$ ) in Landrace pigs but did not differ ( $P > 0.05$ ) from Duroc pigs (Table 1). Plasma free cortisol concentrations were least ( $P < 0.05$ ) in Landrace pigs, but they did not differ from those of LW or Duroc pigs ( $P > 0.05$ ). This finding agrees with other studies showing that purebred MS pigs, as well as MS × LW F<sub>1</sub> pigs, have greater plasma and urinary cortisol concentrations than purebred LW pigs (Hay and Mormède, 1998; Désautés et al., 1999). Similarly, CBG-Bmax was roughly twice as high ( $P < 0.05$ ) in MS × LW gilts compared with the other lines. Furthermore, Piétrain gilts had greater ( $P < 0.05$ ) CBG-Bmax values than Duroc, LW, and Landrace gilts. These results agree with those of Ousova et al. (2004), who showed that CBG-Bmax in purebred Meishan pigs was 3 times greater than in LW pigs. Other breed differences were demonstrated by Marple et al. (1974), in which Poland China pigs had greater CBG-Bmax values than Berkshire and Hampshire pigs.

Plasma cortisol concentrations were correlated with CBG-Bmax in MS × LW ( $P < 0.001$ ) and LW ( $P < 0.05$ ) gilts (Table 2). Conversely, CBG-Bmax was not significantly correlated with urinary cortisol, nor were plasma and urinary cortisol concentrations correlated, regardless of genetic line. It also has been shown in humans that plasma, but not urine, cortisol concentrations were significantly influenced by variation in CBG-Bmax (Bright and Darmaun, 1995; Dhillon et al., 2002). Thus,

according to Pol et al. (2002) and Hay et al. (2000), urine cortisol excretion seems to be more suitable for the detection of variations in cortisol production because concentrations are not influenced by variation in CBG-Bmax, rapid changes in hormone secretion (urine accumulates over several hours), or sample handling (urine collection is a noninvasive method).

### *Carcass Composition and Pork Quality in Relation to CBG-Bmax*

Published results on carcass composition in similar genotype gilts (Plastow et al., 2005) showed that the Piétrain line produced the leanest, heaviest-muscled carcasses, whereas carcasses from the MS × LW line had the greatest fat depth and lowest lean content, with the other lines being intermediate (Plastow et al., 2005). As we demonstrated in this study, MS × LW gilts also had the greatest ( $P < 0.05$ ) percentage of leaf fat and the least ( $P < 0.05$ ) ham yields (Table 3). Loin yield was greater ( $P < 0.05$ ) in Duroc gilts. With regard to pork quality, Plastow et al. (2005) reported that pork from Landrace pigs had the highest L\* values, indicating a lighter pork color. Drip losses were less in the Duroc than Landrace lines (Plastow et al., 2005). The i.m. fat content was greatest ( $P < 0.05$ ) in the SM of Duroc and MS × LW gilts (Table 3), which is similar to results for i.m. fat content of the LM (Plastow et al., 2005).

The 5 breeds differed in the effect of CBG-Bmax on carcass composition and pork quality (Table 4). With

**Table 3.** Least squares means for pork carcass and pork quality characteristics of 5 divergent genetic lines

Trait	No.	Line					SEM
		MS × LW <sup>1</sup>	Piétrain	LW	Landrace	Duroc	
ADG, g/d (70 to 140 d of age)	500	765.46 <sup>w</sup>	758.70 <sup>x</sup>	796.30 <sup>z</sup>	790.72 <sup>z</sup>	783.32 <sup>yz</sup>	13.487
Ham yield, %	480	22.94 <sup>w</sup>	27.03 <sup>z</sup>	24.67 <sup>x</sup>	25.03 <sup>y</sup>	24.83 <sup>xy</sup>	0.097
Loin yield, %	480	16.70 <sup>x</sup>	16.84 <sup>x</sup>	16.90 <sup>xy</sup>	17.10 <sup>y</sup>	17.36 <sup>z</sup>	0.126
Leaf fat, %	500	10.85 <sup>z</sup>	5.65 <sup>x</sup>	5.56 <sup>x</sup>	7.20 <sup>y</sup>	6.93 <sup>y</sup>	0.238
Semimembranosus pH	500	5.51 <sup>y</sup>	5.55 <sup>z</sup>	5.54 <sup>z</sup>	5.49 <sup>x</sup>	5.54 <sup>z</sup>	0.015
Semimembranosus i.m. fat, %	495	2.15 <sup>z</sup>	1.49 <sup>y</sup>	1.19 <sup>x</sup>	1.53 <sup>y</sup>	2.17 <sup>z</sup>	0.080

<sup>w-z</sup>Within a row, least squares means that do not have common superscript letters differ,  $P < 0.05$ .

<sup>1</sup>MS × LW = Meishan × Large White intercross.

**Table 4.** Regression coefficients ( $\pm$ SE) for carcass composition and pork quality variables on cortisol-binding globulin maximum binding capacity (10 nM increase) in 5 divergent genetic lines

Trait	No.	Line									
		MS $\times$ LW <sup>1</sup>		Piétrain		LW		Landrace		Duroc	
		Regression coefficient	P-value	Regression coefficient	P-value	Regression coefficient	P-value	Regression coefficient	P-value	Regression coefficient	P-value
ADG, g/d (70 to 140 d of age)	473	-6.04 $\pm$ 3.09	0.051	-5.87 $\pm$ 6.46	0.364	-8.59 $\pm$ 7.67	0.264	-10.27 $\pm$ 7.26	0.158	-1.47 $\pm$ 6.82	0.830
Ham yield, %	456	-0.012 $\pm$ 0.033	0.709	0.027 $\pm$ 0.068	0.689	0.053 $\pm$ 0.082	0.523	-0.100 $\pm$ 0.076	0.187	-0.125 $\pm$ 0.072	0.082
Loin yield, %	456	-0.009 $\pm$ 0.034	0.779	0.049 $\pm$ 0.070	0.483	0.223 $\pm$ 0.086	0.010	0.129 $\pm$ 0.079	0.103	-0.080 $\pm$ 0.075	0.284
Fat depth, mm	473	-0.232 $\pm$ 0.125	0.064	-0.087 $\pm$ 0.259	0.739	-0.237 $\pm$ 0.306	0.440	-0.198 $\pm$ 0.291	0.499	-0.080 $\pm$ 0.275	0.771
Leaf fat, %	473	0.257 $\pm$ 0.077	0.001	0.056 $\pm$ 0.160	0.730	0.082 $\pm$ 0.190	0.667	0.048 $\pm$ 0.181	0.790	0.167 $\pm$ 0.170	0.326
LM area, cm <sup>2</sup>	473	-0.106 $\pm$ 0.199	0.596	-0.009 $\pm$ 0.416	0.983	-0.519 $\pm$ 0.492	0.292	-0.497 $\pm$ 0.468	0.289	-0.016 $\pm$ 0.440	0.971
Estimated lean content, <sup>2</sup> %	473	-0.187 $\pm$ 0.115	0.105	0.071 $\pm$ 0.239	0.765	-0.304 $\pm$ 0.281	0.280	-0.201 $\pm$ 0.268	0.454	0.035 $\pm$ 0.252	0.888
Semimembranosus pH	473	-0.001 $\pm$ 0.003	0.685	0.003 $\pm$ 0.006	0.596	-0.014 $\pm$ 0.007	0.055	-0.009 $\pm$ 0.007	0.217	-0.003 $\pm$ 0.006	0.665
LM pH	473	-0.000 $\pm$ 0.004	0.954	-0.006 $\pm$ 0.007	0.396	-0.022 $\pm$ 0.009	0.015	-0.002 $\pm$ 0.008	0.797	-0.012 $\pm$ 0.008	0.114
Drip loss, %	434	-0.140 $\pm$ 0.043	0.001	-0.012 $\pm$ 0.087	0.890	0.186 $\pm$ 0.101	0.067	0.161 $\pm$ 0.103	0.117	-0.039 $\pm$ 0.089	0.660
Lightness (L*) value <sup>3</sup>	473	-0.012 $\pm$ 0.099	0.907	-0.230 $\pm$ 0.207	0.269	0.410 $\pm$ 0.245	0.095	0.470 $\pm$ 0.233	0.044	0.284 $\pm$ 0.219	0.196
Semimembranosus i.m. fat, %	469	-0.066 $\pm$ 0.024	0.005	0.018 $\pm$ 0.049	0.710	0.008 $\pm$ 0.057	0.894	-0.007 $\pm$ 0.055	0.892	0.021 $\pm$ 0.051	0.683
LM i.m. fat, %	471	-0.000 $\pm$ 0.021	0.979	0.005 $\pm$ 0.043	0.895	0.045 $\pm$ 0.051	0.382	0.016 $\pm$ 0.049	0.738	0.014 $\pm$ 0.046	0.768

<sup>1</sup>MS  $\times$  LW = Meishan  $\times$  Large White intercross.

<sup>2</sup>Estimated lean content = 61.56 - (0.878  $\times$  fat depth, mm) + (0.157  $\times$  LM depth, mm), Gispert and Diestre (1994).

<sup>3</sup>L\* is a measure of darkness to lightness (a large L\* value indicates a lighter color).

**Table 5.** Regression coefficients ( $\pm$ SE) for carcass composition and pork quality variables on cortisol-binding globulin maximum binding capacity (10 nM increase) in Meishan  $\times$  Large White gilts, when Sw1856-alleles were included in the analysis as covariates

Trait	No.	Regression coefficient	P-value	Main effect P-value for Sw1856-alleles
ADG, g/d (70 to 140 d of age)	96	-2.96 $\pm$ 3.45	0.392	0.345
Ham yield, %	92	0.028 $\pm$ 0.032	0.382	0.052
Loin yield, %	92	-0.025 $\pm$ 0.035	0.484	0.003
Fat depth, mm	96	0.299 $\pm$ 0.202	0.143	0.672
Leaf fat, %	96	0.238 $\pm$ 0.134	0.079	0.670
LM area, cm <sup>2</sup>	96	-0.019 $\pm$ 0.179	0.916	0.006
Estimated lean content, <sup>1</sup> %	96	-0.269 $\pm$ 0.182	0.144	0.554
Semimembranosus pH	96	-0.005 $\pm$ 0.004	0.214	0.627
LM pH	96	-0.005 $\pm$ 0.004	0.243	0.498
Drip loss, %	84	0.156 $\pm$ 0.055	0.006	0.271
Lightness <sup>2</sup> (L*) value	96	0.089 $\pm$ 0.091	0.332	0.093
Semimembranosus i.m., fat, %	92	-0.064 $\pm$ 0.040	0.114	0.176
LM i.m. fat, %	94	-0.011 $\pm$ 0.035	0.763	0.022

<sup>1</sup>Estimated lean content = 61.56 - (0.878  $\times$  fat depth, mm) + (0.157  $\times$  LM depth, mm); Gispert and Diestre (1994).

<sup>2</sup>L\* is a measure of darkness to lightness (a larger L\* value indicates a lighter color).

the exception of a positive relationship between CBG-Bmax and carcass loin yield ( $P = 0.010$ ) in LW gilts and ham yield ( $P = 0.082$ ) in Duroc gilts, CBG-Bmax was not associated with carcass cutability characteristics of Piétrain ( $P \geq 0.364$ ), Duroc ( $P \geq 0.284$ ), Landrace ( $P \geq 0.103$ ), and LW ( $P \geq 0.264$ ) gilts. With regard to carcass composition, a positive effect of CBG-Bmax on leaf fat percent ( $P = 0.001$ ) and a trend for greater ( $P = 0.064$ ) fat depth were found in MS  $\times$  LW gilts, but only leaf fat percent tended ( $P = 0.079$ ) to be positively associated with CBG-Bmax when the Sw1856-alleles were included as covariates in the analysis (Table 5). Conversely, there was no ( $P \geq 0.596$ ) association between CBG-Bmax and LM area or ham and loin yields in MS  $\times$  LW gilts (Table 4), or when the Sw1856 alleles were included as covariates ( $P \geq 0.143$ ; Table 5). Ousova et al. (2004) found positive correlations between CBG-Bmax and fat, and negative correlations with muscle content in 39 male MS  $\times$  LW F<sub>2</sub> pigs, and they suggested that the CBG-gene may be a regulator of fat accumulation and muscle content.

Pork quality of Piétrain ( $P \geq 0.269$ ) or Duroc ( $P \geq 0.114$ ) gilts was not associated with CBG-Bmax (Table 4); however, in Landrace gilts, a 10-nM increase in CBG-Bmax resulted in a 0.47 increase ( $P = 0.044$ ) in L\* values, whereas, in LW gilts, a 10-nM increase in CBG-Bmax was associated with a 0.022 decrease ( $P = 0.015$ ) in LM pH and increases ( $P \leq 0.095$ ) in both drip loss percent and L\* values. In MS  $\times$  LW gilts, CBG-Bmax was associated with increased ( $P = 0.001$ ) drip loss and decreased ( $P = 0.005$ ) i.m. fat content in the SM. Furthermore, when Sw1856-alleles were included as covariates in the statistical analysis, a 10-nM increase in CBG-Bmax was associated with a 0.156% increase ( $P = 0.006$ ) in drip loss (Table 5). As water-holding capacity of pork determines juiciness, this is

an important aspect of palatability that affects overall acceptability of meat. Unexplained variation in water-holding capacity is a consistent problem in the industry and a major source of consumer dissatisfaction (Forrest et al., 2000). Drip development during storage of meat is principally caused by shrinking myofibrils because of changes in pH and temperature postmortem (Offer and Knight, 1988). Rapid pH decline while muscle temperature is still high causes denaturation of many proteins, including those involved in binding water. Changes in pH and temperature postmortem are influenced by preslaughter muscle activity and/or elevated preslaughter muscle temperature (Klont and Lambooij, 1995a,b). How CBG-Bmax could influence these processes is unclear, but this may be related to the metabolic role of cortisol on muscle protein catabolism (Devenport et al., 1989).

Pork quality depends on multiple factors, including fat content, appearance, water-holding capacity, pH, color, and temperature. Ousova et al. (2004) reported that the Cbg-gene may be the causal gene of a QTL on chromosome 7 associated with plasma cortisol concentrations and carcass composition. These authors found a highly significant genetic linkage between CBG-binding capacity and chromosome 7 markers flanking the cortisol-associated QTL in MS  $\times$  LW F<sub>2</sub> intercross pigs. Furthermore, CBG levels were positively correlated with fat and negatively correlated with muscle content in a subset of male pigs, leading Ousova et al. (2004) to suggest that CBG might be a good predictor of carcass composition. In the current study, no major effects of CBG on fat or muscle content were found in the MS  $\times$  LW intercross, nor any of the other 4 genetic lines in the study. Nonetheless, an effect of CBG-Bmax on drip loss was demonstrated in MS  $\times$  LW gilts, which had CBG-Bmax values twice as high as the other lines,

perhaps explaining why no effects were found in the other genetic lines in this study. Because CBG has no apparent circadian rhythm (Barnett et al., 1981), shows a high intraindividual correlation when measured at different times (Nyberg et al., 1988), hardly varies between days, and is not influenced by temperature or humidity (Aberle et al., 1976), it could be a valuable predictor for the level of drip loss in synthetic lines involving the Meishan breed.

## IMPLICATIONS

The cortisol-binding globulin gene is suggested to be an important candidate gene to explain genetic variation in cortisol secretion and carcass composition in pigs. Results of this experiment indicated little to no effect of cortisol-binding globulin maximum binding capacity on either carcass composition or pork quality traits in Piétrain, Duroc, Landrace, or Large White gilts; however, a prominent effect of cortisol-binding globulin maximum binding capacity was found in Meishan × Large White gilts, with greater levels being associated with increased drip loss. Furthermore, results of this study suggest that sequencing the gene encoding cortisol-binding globulin for polymorphisms is warranted to more accurately explain the relationship between cortisol-binding globulin binding capacity and drip loss. Because drip loss is an economically important pork quality trait, results of this study could be used in the selection of improved water-holding capacity of pork from synthetic lines involving the Meishan breed.

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