Evaluation of Coat Color Inheritance and Production Performance for Cossbreed from Chinese Indigenous Chenghua Pig Crossbred with Berkshire

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Research

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Abstract

Background: Crossbreeding programs have been used extensively to improve the native pig's overall production performance while maintaining superior meat quality. Chenghua (CH) pig is a traditional Chinese indigenous breed with superior meat quality characteristic but poor growth and carcass traits. In recent years, we implemented Berkshire × Chenghua (BC) crossbreeding scheme and have bred the new crossbreed BC pig through selection for four generations. The present objective was to determine the black coat inheritance and evaluate production performance for crossbreed BC F4 pigs in comparison with those of control purebred CH pigs.

Results: Coat color of crossbreed BC pigs exhibits a "dominant black" hereditary pattern. Twelve mutation sites for MC1R gene were identified between "uniform black type" and "domino black spotting type" pigs and all piglets derived from boars or sows genotyped *ED1ED1* homozygous for MC1R gene showed uniform black coat phenotype. The crossbreed BC F4 gilts displayed a relatively good reproductive performance, showing a higher litter (total no. born: 12.06 and no. born alive: 11.14) and tear size (13.14), heavier at farrowing litter (11.92 kg) and at weaning litter (65.87 kg) than purebred CH gilts, but they reached puberty later than CH gilts (178.44 vs.125.45). The crossbreed BC F4 pigs exhibited improved growth and carcass characteristics with a higher average daily live weight gain (535.28 g vs. 447.11 g), lower feed-to-gain ratio (3.06 vs. 4.03), and higher carcass lean meat rate (50.76% vs. 42.58%) than purebred CH pigs. Importantly, similar to those of purebred CH pigs, the crossbreed BC F4 pigs produced super meat-quality characteristics, showing ideal pH and meat-color values, high intramuscular fat content (3.24%) and water-holding capacity (drip loss: 1.68%), and acceptable muscle-fibre parameters. C18:1, C16:0, C18:0, and C18:2 were the main fatty acids in *M. longissimus lumborum* in the two breeds, and a very high polyunsaturated/saturated fatty acid ratio of ~0.39 was observed in the BC F4 pigs.

Conclusion: These results indicate that crossbreed BC F4 pigs exhibit a uniform black coat pattern and acceptable total production performance, and it can be extensively used in commercial pig production to provide high-quality niche products.

Background

The Chenghua (CH) pig is a traditional black breed native to South-Western China in the Sichuan province and it is characterized by superior meat quality traits and good adaptability for extensive and management [1]. However, owing to the undesirable traits such as slow growth rate and low lean meat percentage [2], the production system of purebred CH pigs has been almost displaced and CH pigs have been included in the National Program for Farm Animal Resources since 2014.

Crossbreeding programs have been used extensively to improve the native pig's overall production performance while maintaining superior meat quality for F1 hybrid pigs from Duroc × Dahe, Duroc or Landrace × Celta, Duroc × Korean Native Black Pig, and Duroc × Berkshire × Yanan [3–6]. However attribution to the break-up of epistatic complexes since F2 generation [7, 8], improving and stabilizing the obtained heterosis basing on breed additive and dominance effects is a considerable challenge for a new breed formation arisen from the cross of two or more existing breeds.

In recent years, so as to utilize the genetic resource of CH pig to improve its overall production performance and produce superior pork, we have implemented the crossbreeding scheme of Berkshire (BS) × CH (BC) and bred the new crossbreed BC pig through selection for four generations. Nowadays, the core breeding group of BC pigs contains 30 unrelated boar strains and more than 1000 sows, and the production system of BC pigs can supply approximate 50,000 black fattening pigs per year to meet the market demand for high quality pork. However, there is no scientific data evaluating the production performance as well as coat color variation for the new crossbreed BC pigs. Therefore, the present objective was to determine the black coat inheritance and evaluate production performance for crossbreed BC F4 pigs in comparison with those of control purebred CH pigs.

Materials And Methods

Breeding group structure and management

All pigs were kept on the Qionglai Jialin Ecological Farm, Chengdu city, China. The core breeding group of purebred CH pigs included 8 unrelated boar strains and 320 sows. The BC crossbreed base population derived from the progeny of 10 Berkshire (BS) unrelated boar strains and 218 CH sows with above synthetic selection index. Then the more advanced generations were bred by the method of population subgeneration breeding, and the cross breeding population included 30 unrelated boar strains and about 600 sows in each generation. Mating was done in a way that reduced inbreeding. Animals with a relationship coefficient of above 5% were not mated to each other. All matings were through artificial service. The selection of boars and gilts was mainly based on the paternal and matrilineal selection index, respectively; meanwhile, the selection also was in combination with pedigree and phenotypic characteristics. The management and feeding conditions of all pigs at different stages of production were largely designed to the conditions that are experienced in modern breeding areas. The diets met the National Research Council (NRC) (2012) recommendations for the different production phases [9].

Observation of coat color variation and collection of reproductive performance data

Coat color was observed for cross piglets per litter and their parents, and "the uniform black" or "domino black spotting" phenotypes were recorded for more than 16,000 pigs. The reproductive performance for 927 crossbreed BC F4 gilts and 320 purebred CH gilts were recorded and collected from January 2019 to July 2020, respectively. Number of teat was recorded for gilts, meanwhile the puberty of gilts was defined as the first observed estrus followed by a second estrus approximately 21 or 42 d later. The total number of pigs born and number of pigs born alive per litter were recorded, meanwhile piglets per litter were weighed within 12 h of birth and at 28 d of age for litter weight at birth and litter weight at weaning, respectively.

Identification of MC1R SNPs

Hairs with follicles were collected after disinfection with 75% alcohol at the shoulder and then were washed twice with PBS and stored in a refrigerator at -20°C. Genomic DNA was extracted according to the instructions using a DNA extraction kit (Magen, Guangzhou, China). Two pairs of primers were designed according to MC1R reference sequence (GenBank accession number FJ6655467.1) to amplify the complete MC1R DNA sequence (Table 1). The PCR reaction system was 25μL, containing 22μL 2 × TsingKE Master Mix (TsingKE, Beijing, China), 1μL upstream primer, 1μL downstream primer, and 1μL DNA. Thermocycling conditions began with a denaturing at 98 °C for 2 min, followed by 34 cycles of denaturing at 98 °C for 10 s, annealing at the Tm (Table 1) for 10 s, and extension at 72 °C for 10 s, and finally extension step at 72 °C for 5 min, Store at 4 °C. The amplification process was carried out using the Genemate Series PCR machine (Analytik Jena, Germany).

5 μl PCR products were used for 1.5% agarose gel electrophoresis to determine whether the MC1R gene was amplified. BigDye® Terminator V3.1 was used for sequencing purification. 3730 sequencer was used for sequencing and 3730XL was used for data collection. The sequencing sequence and peak graph were obtained by Chromas. The obtained sequence is spliced by CExpress to obtain a complete sequence. The sequence was aligned by Blast in NCBI.

Table 1
Primer sequences and amplification conditions for MC1R gene

Gene	Primer	Primer sequence	Primer binding region	Product size (bp)	Annealing temperature (°C)
MC1R	M-F1	GCTGAGCACAGGCGAGGTT	5'UTR	884	61
	M-R1	GGAAGCAGAGGCTGGACACC	Exon1		
	M-F2	CATCGCCAAGAACCGCAACC	Exon1	903	61
	M-R2	GGTCCAGCGTCCATACCTTCA	3'UTR		

Measurement of fattening and slaughtering performance

In total, 80 pigs (20 castrated males and 20 females for BC F4 or CH pigs, respectively) were randomly selected at about 60 days of age (with weight at about 15 ~ 20 kg). These pigs in each breed were born to a total of 10 litters that were produced by five sires and 10 sows. All pigs were housed in individual pens (2 m^2) located in the same room and were fed twice a day with the same diet, and pigs had ad libitum access to diet and water (nipple drinkers). To gain expected market slaughter weight and age, the fattening experiment lasted for 180 days for BC pigs and 240 days for CH pigs after 7 days of the adaptation period, respectively. The experimental diets met the National Research Council (NRC) (2012) recommendations for the two different growth phases [9]. In the fattening period, the data of initial live weight, final live weight and feed consumption were recorded to determine daily live weight gain and feed-to-gain ratio.

At the end of the fattening period, 20 pigs (10 castrated males and 10 females) per breed were slaughtered to determine carcass-composition characteristics according to the described methods [3, 6]. The measured carcass traits included carcass length, dressing percentage, back fat thickness, loin muscle area, skin thickness, number of ribs, and dissection ratio of bone, muscle, subcutaneous fat and skin. The *M. longissimus lumborum* of the left side of the carcass at the last third to fourth rib was sampled and used to measure meat quality according to the described methods [3, 6]. The measured meat quality traits included pH values, color parameters, water-holding capacity, and muscle-fibre parameters. The muscular fatty acid (FA) composition was analyzed using a gas chromatography (Agilent 6820, Agilent Technologies, Palo Alto, CA, USA) and capillary column (HP-Innowax, Agilent, 30 m long, 0.32 mm internal diameter, 0.25 mm film thickness) according to the described method [10].

Statistical analyses

Statistical testing was implemented by IBM SPSS Statistics 22. The data are quantified as the mean ± SEM for one group. The differences of groups were calculated using independent T test. $P < 0.05$ is considered to be significant, *$P < 0.05$, **$P < 0.01$, and ***$P < 0.001$.

Results

Specific black coat were selected for the crossbreed BC pigs basing on $E^{D1}E^{D1}$ homozygous genotype of MC1R gene

To discover the hereditary pattern of black coat for the crossbreed BC pigs, we firstly observed the phenotypic changes of coat color in cross generation. As a result, all F1 crosses (3,140), which derived from 10 BS boars (domino black spotting type) and 320 CH sows (black type), were uniform black; however F2 cross pigs (5,906), which derived from 30 F1 black boars and 588 F1 black sows, were black or domino in the proportion of about 3:1. Interestingly, when 26 F2 black boars were used in the production system, F3 cross pigs (2, 038 out of 2,041) derived from 9 F2 black boars (called homozygote) and 182 F2 black sows were uniform black, but F3 cross pigs (3,698) derived from other 17 F2 black boars (called heterozygote) were black or domino in the proportion of about 5:1 (Table 2). The results indicated that the black coat of crossbreed BC pigs might be controlled by a dominant single gene and it could inherit in accordance with Mendel's law of segregation.

Table 2
Observation of coat color variation in crossbreed BC pigs

Progeny	Boar		Sow		Piglets		
	Breed	Coat color	Breed	Coat color	Coat color		Ratio of black: domino
F1	BS (10)	Black	CH (320)	Black	Black (3140)	Domino (0)	-
F2	F1 (30)	Black	F1 (588)	Black	Black (4434)	Domino (1472)	3:1
F3	F2 (9)	Black	F2 (182)	Black	Black (2038)	Domino (3)	-
F3	F2 (17)	Black	F2 (330)	Black	Black (3088)	Domino (610)	5:1

BC, Berkshire × Chenghua; BS, Berkshire; CH, Chenghua. F1, F2, and F3 means the BC cross pigs from first, second, and third generation, respectively. "-"means that the ratio cannot be calculated or very large because coat color pattern of (nearly) all pigs is black.

Basing on the important regulatory role of melanocortin 1 receptor gene (MC1R) on body melanin deposition [11], we considered MC1R gene as a potential candidate gene for the black coat of crossbreed BC pigs, and cloned and sequenced the complete DNA of MC1R gene for these samples from BS, CH, F1 crosses, F2 black boars (called homozygote or heterozygote), and F2 domino black spotting cross pigs, respectively. As a result, we obtained a 1552 bp DNA sequence of MC1R (GenBank accession number AY960624) and screened out 12 mutation sites in complete DNA sequence of MC1R gene from these samples (Table 3). According to the definitely established alleles at the MC1R locus [11], we found that the CH pigs and F2 black boars (called homozygote) showed the typical $E^{D1}E^{D1}$ homozygous genotype while BS and F2 crosses with domino black spotting showed other opposite $E^{P}E^{P}$ homozygous genotype; meanwhile F1 crosses and F2 black boars (called heterozygote) displayed the same $E^{D1}E^{P}$ heterozygous genotype (Table 3). The result indicates that the E^{D1} allele is associated with a black coat phenotype and inherits in a dominant pattern in the crossbreed BC pigs. According to the above results, we selected these black boars and sows genotyped with $E^{D1}E^{D1}$ since F3 generation to reproduce offspring. As expected, all BC F4 cross pigs showed uniform black in whole production system.

Table 3
Mutation sites of MC1R gene in CH, BS, and crossbreed BC pigs

Breed	Coat color pattern	Genotype locus	Allele/ Genotype	Mutation locus											
				5'UTR					CDS						
				215	220	242	371	414	490	505	722	744	802	809	1338
CH, F2	Black	$E^{D1}E^{D1}$	Allele	A	G	C	G	C	A	-	A	C	C	G	A
			Genotype	AA	GG	CC	GG	CC	AA	-	AA	CC	CC	GG	AA
F1, F2	Black	$E^{D1}E^{P}$	Allele	A&G	A&G	C&T	A&G	C&T	A&G	-CC	A&G	C&T	C&T	A&G	A&G
			Genotype	AG	AG	CT	AG	CT	AG	-CC	AG	CT	CT	AG	AG
BS, F2	Domino black spotting	$E^{P}E^{P}$	Allele	G	A	T	A	T	G	-CC	G	T	T	A	G
			Genotype	GG	AA	TT	AA	TT	GG	-CC	GG	TT	TT	AA	GG

5'UTR, 5'-untranslated regions, BC, Berkshire × Chenghua; BS, Berkshire; CH, Chenghua; CDS, coding sequence. F1 and F2 mean the BC cross pigs from first and second generation, respectively.

Reproductive performance of crossbreed BC gilts compared with purebred CH gilts

Table 4 summarized the reproductive performance of crossbreed BC F4 gilts compared with purebred CH gilts. The mean number of teat was higher for BC gilts than for CH gilts (*P*< 0.001, 13.14 vs. 12.14). The mean age at puberty of BC gilts was 178.44 d although it was older than CH gilts (*P*< 0.001, 125.45 d). Total number of pigs born (12.06 pigs) and number of pigs born alive (11.14 pigs) per litter were higher (*P*< 0.001) for BC sows than for CH sows (10.31 pigs and 9.82 pigs, respectively). Breed effects were significant for litter birth weight and litter weaning weight. At birth and weaning at 28 d age, litters from BC sows (11.92 kg and 65.87 kg, respectively) were heavier (*P*< 0.001) than those from CH sows (8.33 kg and 49.4 kg, respectively).

Table 4
Reproductive performance of crossbreed BC F4 gilts compared with purebred CH gilts

Traits	Breeds						SEM	*P*
	BC (N = 927)			CH (N = 322)				
	Mean	SD	CV (%)	Mean	SD	CV (%)		
Teat number	13.14	1.21	9.21	12.14	0.87	7.17	0.03	***
Puberty age (day)	178.44	28.38	15.90	125.45	21.39	17.05	2.60	***
Total no. born	12.06	1.73	14.34	10.31	3.79	36.76	0.07	***
No. born alive	11.14	1.74	15.62	9.82	2.30	23.42	0.03	***
Litter birth wt (kg)	11.92	2.34	19.63	8.33	2.37	28.45	0.25	***
Litter weaning wt (kg)	65.87	12.79	19.42	49.40	13.50	27.33	0.64	***

BC, Berkshire × Chenghua; CH, Chenghua. ***, *P* < 0.001.

Growth and carcass traits of crossbreed BC pigs compared with purebred CH pigs

As expected, throughout the fattening period, the crossbreed BC F4 pigs grew faster than that of purebred CH pigs (*P*< 0.001), with a higher average daily live weight gain (535.28 g vs. 447.11 g); meanwhile the feed consumption was more efficient for BC crosses than for CH pigs (*P*< 0.001, feed-to-gain ratio: 3.06 vs. 4.03) (Table 5).

As shown in Table 5, the crossbreed BC F4 pigs exhibited an improved carcass composition compared with those of purebred CH pigs. The slaughter weight was heavier for BC pigs at 236.7 d age than for CH pigs at 302 d age (*P*< 0.001, 112.56 kg vs. 105 kg). The carcasses of BC pigs were longer than those of CH pigs (*P*< 0.001, 83.35 cm vs. 74.85 cm), meanwhile they had more rib than those of CH pigs (*P*< 0.001, 14.5 vs. 13.1). Importantly, the carcasses of BC pigs were more muscular than that of CH pigs (*P*< 0.001), with a higher carcass lean meat rate (50.76% vs. 42.58%), bigger loin muscle area (32.61 cm^2 vs. 24.15 cm^2), higher ham rate (29.98% vs. 25.03%), thinner back fat (26.44 mm vs. 35.99 mm), and lower carcass fat rate (23.65% vs. 32.46%). Similarly to CH pigs, BC pigs had a thick skin (5.77 mm) and high carcass skin rate (14.93%).

Table 5
Growth and carcass traits of crossbreed BC F4 pigs compared with purebred CH pigs

Traits	Breeds						SEM	*P*
	BC (N = 40 and 20)			CH (N = 40 and 20)				
	Mean	SD	CV (%)	Mean	SD	CV (%)		
Daily live weight gain (g/day)	535.28	20.02	3.74	447.11	42.72	9.55	10.44	***
Feed-to-gain ratio	3.06	0.18	5.88	4.03	0.36	8.93	0.09	***
Slaughter age (day)	236.70	4.07	1.72	302.05	4.82	1.60	1.41	***
Slaughter weight (kg)	112.56	2.64	2.35	105.00	7.18	6.84	1.71	***
Carcass length (cm)	83.35	1.80	2.16	74.85	3.38	4.52	0.86	***
Dressing percentage (%)	73.27	2.39	3.26	74.21	1.14	1.54	0.59	n.s.
Back fat thickness (mm)	26.44	3.56	13.46	35.99	3.52	9.78	1.12	**
Loin muscle area (cm^2)	32.61	6.24	19.14	24.15	3.99	16.52	1.66	***
Skin thickness (mm)	5.77	0.87	15.08	6.53	1.15	17.61	0.41	n.s.
Number of ribs	14.50	0.53	3.66	13.10	0.45	3.44	0.18	***
Ham (%)	29.98	1.64	5.47	25.03	1.39	5.55	0.48	***
Carcass lean (%)	50.76	2.95	5.81	42.58	2.39	5.61	0.85	***
Carcass fat (%)	23.65	2.54	10.74	32.46	2.49	7.67	0.92	***
Carcass skin (%)	14.93	1.02	6.83	14.66	1.68	11.46	0.54	n.s.
Carcass bone (%)	10.74	0.85	7.91	10.29	0.87	8.45	0.27	n.s.

BC, Berkshire × Chenghua; CH, Chenghua. **, $P < 0.01$; ***, $P < 0.001$. n.s., not significant ($P > 0.05$).

Meat quality and muscle fatty acid composition of crossbreed BC pigs compared with purebred CH pigs

Similarly to CH pigs, crossbreed BC F4 pigs displayed excellent meat quality traits (Table 6). The meat from BC pigs showed ideal pH value (pH_1: 6.32 and pH_2: 5.9) and meat-color parameter (L_{45min}: 41.68 and L_{24h}: 44.41); meanwhile, the meat from BC pigs had strong water-holding capacity, with less water content (72.64%), very low drip loss (1.68%) and cooking loss (34.06%). Noteworthily, the BC meat contained more intramuscular fat (IMF) content than CH pigs ($P < 0.01$, 3.24% vs. 2.64%). In addition, the BC pigs displayed ideal muscle-fibre parameters, with small myofibre area (2641.75 $\mu m^{2)}$, low shear force (6.16 kg) and firmness (26.59 kg/s).

Table 6
Meat quality traits of crossbreed BC F4 pigs compared with purebred CH pigs

Traits	Breeds						SEM	*P*
	BC (N = 20)			CH (N = 20)				
	Mean	SD	CV (%)	Mean	SD	CV (%)		
$pH_{45\ min}$	6.32	0.25	3.96	6.49	0.11	1.69	0.09	n.s.
$pH_{24\ h}$	5.90	0.25	4.24	5.95	0.23	3.87	0.11	n.s.
L (luminosity)$_{45\ min}$	41.68	2.06	4.94	41.41	2.58	6.23	0.74	n.s.
L (luminosity)$_{24\ h}$	44.41	3.32	7.48	42.35	3.77	8.90	1.12	n.s.
Crude protein content(%)	22.87	2.05	8.96	23.48	0.96	4.09	0.51	n.s.
Intramuscular fat content (%)	3.24	1.36	41.98	2.64	1.95	73.86	0.53	***
Water content (%)	72.64	2.53	3.48	72.30	1.21	1.67	0.63	n.s.
Drip loss (%)	1.68	0.27	16.07	1.55	0.75	48.39	0.25	n.s.
Cooking loss (%)	34.06	2.32	6.81	28.87	2.42	8.38	0.75	***
Shear force (kg)	6.16	2.93	47.56	9.37	3.09	32.98	0.97	**
Firmness (kg/s)	26.59	13.50	50.77	41.54	13.03	31.37	4.25	**
Myofibre area (μm^2)	2641.75	711.55	26.93	2723.72	533.87	19.60	281.30	n.s.

BC, Berkshire × Chenghua; CH, Chenghua. **, $P < 0.01$; ***, $P < 0.001$. n.s., not significant ($P > 0.05$).

More than 16 fatty acids (FAs) were identified in the *longissimus dorsi* from both crossbreed BC F4 pigs and purebred CH pigs, and the most prevalent FAs in all pigs were C18:1, C16:0, C18:0 and C18:2, accounting for more than 85% of all FAs (Table 7). The predominant saturated fatty acid (SFA) were C16:0 and C18:0 in all pigs, and total concentrations of SFA accounted for 34.14% in BC pigs and 49% in CH pigs, respectively. The predominant monounsaturated fatty acid (MFA) in all pigs was C18:1 (52.48% for BC and 41.41% for CH). C18:2 was the main polyunsaturated fatty acid (PUFA) in all pigs, and the total concentrations of PUFA were significantly affected by breed, with BC pigs exhibiting a higher PUFA content than did CH pigs ($P < 0.05$, 14.03% vs. 9.59%), which leading to a PUFA:SFA ratio of 0.39 for BC crosses and 0.2 for CH pigs.

Table 7
Fatty acid composition of M. longissimus lumborum from crossbreed BC F4 pigs compared with purebred CH pigs (% total fatty acids)

Traits	Breeds						SEM	*P*
	BC (N = 20)			CH (N = 20)				
	Mean	SD	CV (%)	Mean	SD	CV (%)		
C10:0	0.10	0.01	12.31	0.15	0.07	43.05	0.03	n.s.
C12:0	0.08	0.01	7.61	0.1	0.02	22.74	0.01	n.s.
C14:0	1.59	0.14	8.91	1.48	0.22	15.17	0.11	n.s.
C16:0	14.93	0.67	4.47	28.22	3.6	12.77	1.64	***
C16:1	4.01	0.22	5.43	3.93	1.02	26.06	0.47	n.s.
C17:0	0.15	0.02	14.91	0.21	0.04	16.87	0.02	**
C18:0	16.08	0.51	3.15	12.53	1.76	14.03	0.81	***
C18:1n9c	45.13	3.72	8.23	36.26	8.48	23.39	3.95	*
C18:2n6c	11.95	2.19	18.29	8.21	2.47	30.07	1.23	**
C18:3n3	0.27	0.04	15.32	0.25	0.04	15.69	0.02	n.s.
C20:0	0.25	0.04	14.28	0.29	0.07	23.71	0.03	n.s.
C20:2	0.39	0.07	18.14	0.61	0.24	39.76	0.11	n.s.
C20:3n3	0.44	0.08	19.01	0.27	0.09	34.25	0.01	n.s.
C21:0	0.81	0.05	6.47	4.05	6.59	162.72	1.11	n.s.
C22:1n9	3.33	0.69	20.61	0.03	0.01	24.77	0.16	***
C23:0	0.05	0.02	36.47	1.60	0.66	41.58	0.30	***
SFA	34.14	1.22	3.56	49.00	6.69	13.66	3.05	***
PUFA	14.03	11.39	81.19	9.59	2.84	29.67	1.41	*
MUFA	52.48	3.14	5.99	41.41	8.3	20.05	3.84	**
MUFA:SFA	1.54	0.14	8.81	0.85	0.23	27.55	0.11	***
PUFA:SFA	0.39	0.07	16.79	0.20	0.05	26.13	0.03	***

BC, Berkshire × Chenghua; CH, Chenghua. *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$. n.s., not significant ($P > 0.05$). MUFA, monounsaturated fatty acid; PUFA, polyunsaturated fatty acid; SFA, saturated fatty acid; MUFA:SFA, the ratio of MUFA to SFA; PUFA:SFA, the ratio of PUFA to SFA.

Discussion

Black coat variation of pig being associated with MC1R gene

Coat color is an important characteristic of various pig breeds and color variations may be useful in identifying the components of some specific crossbreeding schemes as well as contributing to the image associated with high quality regional products [12]. Here, the observation dealing with crossbreeding experiments between the Chinese indigenous CH pigs (uniform black) and imported BS pigs (domino black spotting) exhibits a “dominant black” coat color hereditary pattern. Similar result was reported that the allelism between “uniform black type” and “domino black spotting type” may also be inferred from Large Black × Berkshire cross pigs [13].

Our observed segregation results prompted the important discovery that the coat color variation of crossbreed BC pigs is determined by the single MC1R gene although more than eight color loci have been discovered [14]. Twelve mutations were screened for MC1R gene in BC crosses, which represent two typical E^{D1} and E^{P} alleles inferred according to the previous report [9]. Our result indicates that the E^{D1} allele associated with a black coat phenotype inherits in a dominant pattern in the crossbreed BC pigs. Consequently, we have selected these the black boars and gilts genotyped with homozygous $E^{D1}E^{D1}$ since F3 progeny, and largely succeeded in the BC breed standard of black coat.

Crossbreeding improving sow reproductive performance

Level of sow productivity is one of the most importance production traits affecting the efficiency of a swine enterprise [15]. Crossbreeding programs have been used extensively to improve reproduction by exploiting breed additive effects, breed maternal effects, and heterosis. Young (1995) reported that Chinese indigenous breed Meishan, Fengjing, and Minzhu pigs can be used to produce crossbreed gilts that have a higher level of reproductive performance than Duroc crossbreed gilts [16]. Here, we found that the crossbreed BC F4 gilts have a higher litter and tear size, and meanwhile the BC gilts also are heavier at farrowing and at weaning than purebred CH gilts. Noteworthy, a mean of 12.1 pigs for total litter size and 11.4 pigs for alive litter size of BC sows has an advantage in litter size during the breeding process. Similar result also was reported that the cross sows from the Chinese native Meishan, Fengjing and Minzhu pigs show a total number of pigs born (12.0 to 11.0 pigs) and number of pigs born alive (11.3 to 10.7 pigs) per litter [14].

A favorable mean age at puberty of 178 d for crossbreed BC gilts was found although BC gilts reached puberty later than CH gilts. Similar result was reported that a mean age at puberty of 118 and 217 d for purebred Meishan and its crossbred gilts [17]. However, purebred Duroc averaged 234 d at puberty, compared with 210, 205, and 201 d for Hampshire, Pietrain, and Spot pigs [18].

Crossbreeding improving growth performance and carcass composition

Previous reports revealed that the growth performance of hybrid pigs from Duroc × Dahe, Celta × Landrace, Celta × Duroc, and Duroc × Yanan was largely improved compared with that of native pig breeds [3, 4, 6]. Here, two important growth traits including weight gain and feed efficiency are largely improved in crossbreed BC F4 pigs, indicating that the BC pigs reach a competitive slaughter age (about 200 d) at above 100 kg slaughter weight.

Meanwhile, the crossbreed BC pigs exhibit an improved carcass characteristic such as moderate lean meat ratio (~ 50%) and backfat thickness (~ 2.6 cm). The present result is similar to those of previous studies [3, 4, 6], which reported that the carcass characteristics of hybrid pigs were largely improved compared with those of the native pig breeds and a mean of 51 ~ 55% for lean meat ratio was found in the crosses from Duroc × Dahe, Celta × Landrace, Celta × Duroc, and Duroc × Yanan. According to the market demand of black pork in China, we suggest that it is perfectly suitable for black breed to reach a mean of 53 ~ 55% for lean meat ratio (about 3 ~ 5% increase). So, to achieve an ideal lean meat ratio, we will select back fat further down to 20 mm for alive back fat thickness at 180 d age in the subsequent breeding process of BC pigs.

Breed affecting meat quality characteristics

Meat quality is an important factor affecting how pork can be utilized. When choosing the best crossbreeding strategy, it is important to recognize pig breeds determine meat quality traits [19]. In this study, the crossbreed BC and purebred CH pigs produce excellent meat-quality characteristics, which showing normal and high pH values referred to the recommended normal levels (pH_1 > 6.1 and pH_2: 5.5 ~ 6.0) [20], normal and low meat color parameters according to NPPC (2000) standards (Minolta L-value levels of 37–49) [21], lower drip loss than those for foreign breeds with above 3% [22], and smaller muscle-fibre areas than did foreign hybrid pigs with above 5,000 μm^2 [19].

As the most important parameter of meat quality, The IMF content is related to the organoleptic characteristics of pig meat and influences meat and meat-product quality [23]. An IMF content of 2 ~ 3% is suggested to be optimal for food quality [24, 25]. Interestingly, the crossbreed BC pigs in the present study exhibited relatively high IMF content (3.24%). Meanwhile, a higher PUFA:SFA ratio of IMF leads to better digestion rates and an improved digestibility of SFAs with emulsifying agents [26, 27] and the recommended PUFA:SFA ratio is more than 0.4 [28]. Here, a similar PUFA:SFA ratio of ~ 0.39 also was found in the BC pigs. The BC pig meat with high IMF content and PUFA:SFA ratio can meet the demand for high-quality niche pork products.

The super meat quality properties for the crossbreed BC pigs might be attributed to the breed attributes of their parent. Because previous studies found that the Berkshire sire pigs are superior for loin meat and eating [22], and these characteristics are, consequently, thought to attribute to its higher overall likeability score [29, 30] and improved acceptability compared with European commercial pork breeds [31]. Meanwhile, the present result and previous study indicate that the CH pigs also is characterized by superior meat quality traits [1].

Conclusions

The coat color of Berkshire × Chenghua (BC) cross pigs exhibits a “dominant black” phenotypic hereditary pattern and the new crossbreed BC F4 pigs exhibit a uniform black coat pattern by selecting their sire and maternal pigs with $E^{D1}E^{D1}$ homozygous genotype for MC1R gene. Meanwhile, the crossbreed BC F4 pigs have an outstanding overall production performance, which showing that BC pigs have a relatively good maternal reproductive performance, market-competitive improved growth and carcass characteristics, and super meat-quality characteristics. These results indicate that the new crossbreed BC black pigs can be extensively used in commercial pig production to provide high-quality niche products.

Abbreviations

BC

Berkshire × Chenghua; BK:Berkshire; CH:Chenghua; FA:fatty acid; IMF:intramuscular fat; MC1R:melanocortin 1 receptor; MFA:monounsaturated fatty acid; NRC:National Research Council; PUFA:polyunsaturated fatty acid; SFA:saturated fatty acid.

Declarations

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Availability of data and materials

All analysis results data generated during this study are included in this published article. Requests for the raw data should be made to the corresponding authors.

Authors' contributions

YJ, XW, ZG, LZ, GT, ML and XL led the experiments and designed the analytical strategy, YJ, XW, ZG, QZ, ZZ, ZX and YS performed the experiments, YJ, XW, ZG and GT analyzed the data, YJ and XW wrote the manuscript. All authors contributed to the production of the final manuscript. All authors read and approved the final manuscript.

Ethics approval and consent to participate

All animal experimental procedures were approved by the Care and Use Committee of Sichuan Agricultural University (permit number: 2018216014).

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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