SNPs in AKIRIN2, TTN, EDG1 and MYBPC1 genes are associated with growth-related traits in Chinese Qinchuan cattle

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Short report

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Abstract

Growth performances are the main breeding objectives in Chinese beef cattle. The objective of this study was to confirm genetic effects of the *c.*188G > A* SNP in *AKIRIN2*, the *g.231054C > T* SNP in *TTN*, the *g.1471620G > T* SNP in *EDG1*, and the *g.70014208A > G* SNP in *MYBPC1* gene on growth-related traits in Chinese Qinchuan cattle, as well as to compare the frequencies of the well-characterized alleles of these SNPs among six Chinese cattle populations, three Japanese cattle populations, two European cattle populations and one Korean cattle breed. In this study, a total of 655 cattle samples were genotyped using MassARRAY and PCR-RFLP. Association analysis indicated that the four SNPs have effects one to six indexes of growth-related traits including body length, wither height, hip height, hip width, rump length, chest depth and chest circumference in Chinese Qinchuan cattle ($P < 0.05$ to $P < 0.001$). The well-characterized *A* (*c.*188G > A*), *T* (*g.231054C > T*) and *T* (*g.1471620G > T*) alleles in Japanese Black cattle were significantly higher than Chinese cattle breeds, on the contrary, the *G* allele (*g.70014208A > G*) was markedly higher in Chinese cattle breeds than other cattle breeds. These results suggest that the four SNPs might be useful as a molecular marker for growth-related traits in Chinese Qinchuan cattle.

Background

Growth-related traits are complex and controlled by multiple genes [1, 2], which have an important effect on the economics of beef production. Thus, a better knowledge of the molecular architecture of growth-related traits is important as it may generate new opportunities for more effective marker-assisted breeding, leading to economic benefits to the beef production industry.

To date, numerous candidate genes and molecular markers associated with economic traits have been identified, and some of them have already been used in cattle breeding [3–5]. Among them, the *akirin 2* (*AKIRIN2*) gene is located within genomic regions of quantitative trait loci (QTLs) for marbling score and longissimus muscle area in Japanese Black (JB) cattle [6], as well as marbling score in Angus cattle [7]; the *titin* (*TTN*) gene was found in the genomic regions within QTLs for marbling score, longissimus muscle area and subcutaneous fat thickness in JB [6], as well as subcutaneous fat thickness in a Brahman × Hereford sire developed half-sib family [8]; the *endothelial differentiation sphingolipid G-protein-coupled receptor 1* (*EDG1*) gene is located within genomic regions of QTLs for marbling score and body weight in JB [6], subcutaneous fat thickness in Angus [9], as well as marbling score in a Belgian Blue × MARC III developed half-sib family and a Piedmontese × Angus sire developed half-sib family [10], and the *myosin binding protein C, slow type* (*MYBPC1*) is included in the genomics regions of QTLs for rib thickness and subcutaneous fat thickness in JB [6], marbling score in Angus [7], hip height, rump length, rump width and chest depth in a cattle population including 1554 AI bulls distributed in 14 half-sib families (nine in Holstein, three in Normande and two in Montbéliarde breeds [11], as well as intramuscular fat in a Brangus heifers population [12]. Thus, these genes could be considered as important candidate genes for growth-related traits in beef cattle. Furthermore, the *c.*188G > A* single nucleotide polymorphism (SNP) in *AKIRIN2*, the *g.231054C > T* SNP in *TTN*, the *g.1471620G > T* SNP in *EDG1*, and the *g.70014208A > G* SNP in the *MYBPC1* genes showed associations of these SNPs with growth-related traits in JB [13–18] and Korean native (KN) [19] cattle. However, the relationships of these SNPs with growth-related traits in Chinese cattle breed have not been investigated.

Chinese indigenous cattle breeds can be divided into three groups based on their geographic distribution, morphological characteristics and sex chromosome polymorphisms: the northern type in North China (NC), the central type in the middle (CC) and lower areas of the Yellow River and the southern type in South China (SC) [20]. The Qinchuan (QC) cattle which is a typical breed of CC, are well-known to be good beef cattle in China, because of distinctive qualities including good adaptability and fine beef flavor among others. However, Qinchuan cattle exhibit a number of limitations compared to imported commercial beef cattle breeds, such as slow growth rate and underdeveloped hind hips. Accordingly, it is necessary to select important functional genes and molecular marker to increase the economic traits in Chinese native cattle.

Therefore, the objectives of this study reported herein were to: (1) perform association analyses between these SNPs in the *AKIRIN2*, *TTN*, *EDG1* and *MYBPC1* genes and growth-related traits in QC, (2) investigate the genetic diversity of these SNPs in three Chinese typical cattle groups (including QC, Mongolia cattle (MG), Luxi (LX), Wuling (WL) and Longlin (LL) cattle breeds), and (3) compare the frequencies of the well-characterized alleles of these SNPs in Chinese cattle breeds to those of JB, Japanese Brown (JBR), Japanese Short Horn (JSH), Holstein (HOL), Brown Swiss (BS) [21–23], and KN breeds [19].

Methods

Ethics Standards and Animals

The 41 MG population in Inner Mongolia Autonomous Region of China (MGC) and 24 LX cattle were sampled from Chifeng Shengquan Ecological Animal Husbandry Co., Ltd. Ten milliliters of blood was collected from each cattle for DNA extraction.

DNA and Phenotypic Data

Genomic DNA of 41 MGC and 24 LX were extracted from blood samples with a TIANamp Blood DNA kit (TIANGEN Biotech, Beijing, China). The quality and quantity of the extracted DNA were evaluated using a Nanodrop® spectrophotometer (Thermo Fisher Scientific, Waltham, MA, USA) and by agarose gel electrophoresis. The DNA samples and phenotypes of 350 QC adult females were provided from the National Beef Cattle Improvement Center, Northwest A & F University [24, 25]. The measurement of growth-related traits (body length, withers height, hip height, hip width, rump length, chest depth and chest circumference) were described in the previous reports [24, 25]. The DNA samples of 50 MG population in Mongolia (MGM), 50 WL and 50 LL cattle were provided from the Inner Mongolia Key Laboratory of Biomanufacture, Inner Mongolia Agriculture University. The geographical distribution of cattle breeds in this study are shown in Figure 1, as well as the information is shown in Table 1.

Genotyping

For 350 QC cattle population, the *c.*188G>A* SNP in *AKIRIN2*, the *g.231054C>T* SNP in *TTN*, the *g.1471620G>T* SNP in *EDG1*, and the *g.70014208A>G* SNP in the *MYBPC1* genes were genotyped with the MassARRAY® SNP genotyping system (Agena Bioscience, San Diego, CA, USA). PCR and extension primers were designed from sequences containing each target mutation and ~ 100 upstream and downstream bases with Assay Design Suite (<http://agenabio.com/assay-design-suite-20-software>) using the default settings. The genotype of each

allele was analyzed using the Sequenom MassARRAY iPLEX platform [26]. The resulting data was analyzed using the MassARRAY Typer 4.0 Analyzer software (Agena Bioscience).

For 41 MGC, 50 MGM, 50 WL, 50 LL and 24 LX cattle populations, the *c.*188G>A* SNP in *AKIRIN2*, the *g.231054C>T* SNP in *TTN*, the *g.1471620G>T* SNP in *EDG1*, and the *g.70014208A>G* SNP in the *MYBPC1* genes were genotyped using PCR-restriction fragment length polymorphism (RFLP) method as described previously [13, 14, 16, 17]. The PCR primers and restriction enzymes used for PCR-RFLP are shown in Table S1.

Data Analyses

Genotypic and allelic frequencies and Hardy-Weinberg equilibria were calculated for QC, MGC, MGG, WL, LL and LX cattle populations. Population genetic indices, observed heterozygosity (H_o), expected heterozygosity (H_e), effective allele numbers (n_e) and polymorphism information content (PIC) were calculated by Nei's methods [27]. The allelic frequencies of each SNP among cattle breeds or groups were compared by a χ^2 test. The relationship between different genotypes and growth-related traits of QC cattle was analyzed in SPSS 24.0 (SPSS, Inc., Chicago, IL, USA). The statistical linear model for this analysis was the same as previous reports [25]:

$$Y_{ijk} = \mu + G_i + A_j + A_k + E_{ijk}$$

Where, Y_{ijk} = trait value per individual, μ = overall population mean per trait, G_i = fixed effect associated with genotype, A_j = fixed effect of age, A_k was the fixed effect due to the age of dam and E_{ijk} = standard error. The Bonferroni correction was used to adjust P values [25].

Results

Genetic Diversity of Four SNPs in Six Cattle Populations

For the *c.*188G>A* SNP in *AKIRIN2*, the *g.231054C>T* SNP in *TTN*, the *g.1471620G>T* SNP in *EDG1*, and the *g.70014208A>G* SNP in *MYBPC1* genes, the frequencies of the two alleles and the three genotypes of each SNP in the MGC, MGG, QC, LX, WL and LL cattle populations are listed in Table 2, as are the genetic indices (H_o , H_e , n_e and PIC). No significant departures at the 5% level were detected by any test for the four SNPs, except for the *c.*188G>A* SNP of the *AKIRIN2* gene in MGC (Table 2). In this study, the LL populations showed the lowest value of PIC in each of four SNPs than the other cattle populations.

Associations Between Four SNPs and Growth-related Traits in Qinchuan Cattle

The effects of the *c.*188G>A* SNP in *AKIRIN2*, the *g.231054C>T* SNP in *TTN*, the *g.1471620G>T* SNP in *EDG1*, and the *g.70014208A>G* SNP in *MYBPC1* genes on growth-related traits were analyzed in 350 QC population (Table 3). For the *c.*188G>A* SNP in *AKIRIN2*, the individuals with *AA* and *GA* genotypes had significantly longer body length than individuals with *GG* genotype ($P < 0.05$); the individuals with *AA* and *GA* had significantly greater hip width than the individuals with *GG* ($P < 0.05$, $P < 0.001$, respectively); and the SNP genotype also had statistically significant effect on chest depth ($P < 0.05$) and chest circumference ($P < 0.05$). For the *g.231054C>T* SNP in *TTN*, there were only six cattle with the *TT* genotype. Therefore, their associations and effects could not be reliably estimated and they were excluded from the analysis. The individuals with *CT*

genotype had significantly greater wither height ($P < 0.05$), chest depth ($P < 0.05$) and chest circumference ($P < 0.05$) compared with *CC* genotype. For the *g.1471620G>T* SNP in *EDG1*, there were significant difference between *GG* and *GT* genotype of hip width ($P < 0.05$), whereas no association was detected in the other growth-related traits. For the *g.70014208A>G* SNP in *MYBPC1*, the only six cattle with *AA* genotype were excluded from the analysis. Thus, the *GG* homozygotes exhibited significantly greater body length ($P < 0.001$), wither height ($P < 0.05$), hip height ($P < 0.05$), hip width ($P < 0.01$), rump length ($P < 0.01$) and chest depth ($P < 0.05$) compared to *GA* heterozygotes (Table 3).

Statistical Comparations of Allele Frequencies among Six Cattle Groups

To further analyze the distribution of the well-characterized allele of each SNP in the cattle populations based on the different genetic background, the MGC, MGG, QC, LX, WL and LL of this study as well as the JB, JBR, JSH, HOL, BS and KN [19, 21-23] were divided into six groups which including North China (MGC and MGG) [20], Central China (QC and LX) [20], South China (WL and LL) [20], Japanese cattle group (JB, JBR and JSH), European cattle group (HOL and BS), and Korean cattle (KN) (Figure 2; Table 4). At the *c.*188G>A* locus in *AKIRIN2*, the well-characterized *A* allele frequency in the JG and KN was obviously higher than the other cattle groups (Fig. 2a; Table 4). At the *g.231054C>T* locus in *TTN*, the well-characterized *T* allele frequency in the JG was significantly higher than the other cattle groups ($P < 0.001$, Fig. 2b; Table 4). At the *g.1471620G>T* locus in *EDG1*, the well-characterized *T* allele frequency in the JG was significantly higher than the other cattle groups ($P < 0.001$, Fig. 2c; Table 4). On the contrary, the distribution of the well-characterized *G* allele frequency of the *g.70014208A>G* SNP in *MYBPC1* was significantly higher in NC, CC and SC than the JG and EG ($P < 0.001$, Fig. 2d; Table 4). In addition, the well-characterized *T* allele of the *g.231054C>T* SNP were almost absent in SC and EG (Fig. 2b), the same to the well-characterized *T* allele of the *g.1471620G>T* SNP in SC and EG (Fig. 2c), as well as the well-characterized *G* allele frequency of the *g.70014208A>G* SNP in EG (Fig. 2d). The χ^2 tests for different distribution of allele frequencies of the *c.*188G>A* SNP in *AKIRIN2*, the *g.231054C>T* SNP in *TTN*, the *g.1471620G>T* SNP in *EDG1* and the *g.70014208A>G* SNP in *MYBPC1* genes between any two cattle groups are listed in Tables S2-5.

Discussion

The *AKIRIN2*, *TTN*, *EDG1* and *MYBPC1* genes were first found in relationship with economic traits in cattle, because they showed different expression levels in the *musculus longissimus* muscle between JB and HOL [28]. These genes are located within genomic regions of QTLs for carcass and growth-related traits in JB, Angus and crossbred populations [6–12]. Since then, the *c.*188G>A* SNP, the *g.231054C>T* SNP, the *g.1471620G>T* SNP and the *g.70014208A>G* SNP were identified in the *AKIRIN2*, *TTN*, *EDG1* and *MYBPC1* genes of JB [13, 14, 16, 17], respectively. The *c.*188G>A* SNP of *AKIRIN2* showed associations with marbling score in JB [13] as well as marbling score and longissimus muscle area in KN [19]. The *g.231054C>T* SNP of *TTN* showed associations with marbling score and longissimus muscle area in JB [14, 15]. The *g.1471620G>T* SNP of *EDG1* showed association with marbling score in JB [16], simultaneously one other SNP in *EDG1* had effects on marbling score, subcutaneous fat thickness, carcass weight, longissimus muscle area and rib thickness [29, 30]. The *g.70014208A>G* SNP of *MYBPC1* had effects on marbling score and longissimus muscle area [17, 18]. Furthermore, the results of the present study confirmed the significant genetic effects of

the *c.*188G > A* SNP in *AKIRIN2*, the *g.231054C > T* SNP in *TTN*, the *g.1471620G > T* SNP in *EDG1*, and the *g.70014208A > G* SNP in *MYBPC1* genes on growth-related traits in Chinese QC cattle population.

Around the second century A.D., cattle migrated from North Asian continent via the Korean peninsula to Japan [31]. In Japanese cattle, both genetic [32] and morphological [33] studies have illustrated that native Japanese cattle are *Bos taurus* and are representatives of the “Turano-Mongolian” type [34]. The Korean native cattle (Hanwoo), is a cattle breed that is native to the Korean peninsula and the Japanese islands, which is considered to belong to the *Bos taurus* [35–37]. Actually, the JB has been performed for a strong selection for high marbling over the last 50 years, but not in other breeds such as JBR and JSH [38], as well as all Korean indigenous cattle breeds are just used for beef meat production [39] and consumer demands are driving efforts to increase meat production and produce higher quality meat in Korea [40]. Unlike them, the growth performance was regarded as the major breeding goal in most breeds of Chinese indigenous cattle. In this study, the frequencies of the well-characterized *A* allele (in the *c.*188G > A* SNP of *AKIRIN2*) in the JB and KN, *T* allele (in the *g.231054C > T* SNP of *TTN*) in JB and *T* allele (in the *g.1471620G > T* SNP of *EDG1*) in JB were significantly higher than Chinese cattle breeds (Tables S2-4), maybe due to the breeding aim for high marbling in JB and KN. Interestingly and conversely, the well-characterized *G* allele (in the *g.70014208A > G* SNP of *MYBPC1*) was markedly higher in Chinese cattle breeds than JB (Table S5). Moreover, the *G* allele (in the *g.70014208A > G* SNP of *MYBPC1*) was almost null in JBR and JSH, on the contrary, the *G* allele was almost full in WL and LL breeds of CC (Tables S2-5), likely due to genetic background. In addition, the PIC of each SNPs in HOL and BS of EG was related low or null, maybe due to the breeding aim for dairy-related traits in HOL and BS. We noted that such as Angus and Shorthorn breeds should be tested to better understand the distribution of the well-characterized allele in European beef cattle in the future study.

Recently, China cattle beef industry is growing rapidly to meet the meat demand of large population. Although a lot of researches have been performed to identify marker for breeding, it still needs more useful molecular markers for more effectively breeding [5]. In this study, there were significant associations between the *c.*188G > A* SNP of *AKIRIN2* and body length, hip width, chest depth and chest circumference, between the *g.231054C > T* SNP of *TTN* and wither height, chest depth and chest circumference, between the *g.1471620G > T* SNP of *EDG1* and hip width, between the *g.70014208A > G* SNP of *MYBPC1* and body length, wither height, hip height, hip width, rump length and chest depth in Chinese QC cattle.

Conclusions

In conclusion, the results of this study suggest that the *c.*188G > A* SNP of *AKIRIN2*, the *g.231054C > T* SNP of *TTN*, the *g.1471620G > T* SNP of *EDG1*, and the *g.70014208A > G* SNP of *MYBPC1* genes might be useful as a specific DNA marker for growth-related traits in Chinese QC cattle.

Additional Files

Additional file 1: Table S1. PCR primers and restriction enzymes used for PCR-RFLP method. (DOCX 16kb)

Additional file 2: Table S2. Statistical significance for differences in the allele frequency of *c.*188G > A* SNP among 12 cattle populations. (DOCX 17kb)

Additional file 3: Table S3. Statistical significance for differences in the allele frequency of *g.231054C>T* SNP among 11 cattle populations. (DOCX 17kb)

Additional file 4: Table S4. Statistical significance for differences in the allele frequency of *g.1471620G>T* SNP among 11 cattle populations. (DOCX 17kb)

Additional file 5: Table S5. Statistical significance for differences in the allele frequency of *g.70014208A>G* SNP among 11 cattle populations. (DOCX 17kb)

Abbreviations

SNP: Single nucleotide polymorphism; *AKIRIN2*: akirin 2 gene; *TTN*: titin gene; *EDG1*: the endothelial differentiation sphingolipid G-protein-coupled receptor 1 gene; *MYBPC1*: the myosin binding protein C, slow type gene; PCR: Polymerase Chain Reaction; RFLP: Restriction Fragment Length Polymorphism; QTL: quantitative trait locus

Declarations

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Availability of data and materials

All data analyzed during this study are included in this article and its supplementary files.

Authors' contributions

BT, LZ, GL, HZ and TY designed the experiments. YL, GC, YZ, CB, SW, KW, LC and SS carried out the experiments and calculated data. YL, GC, LZ and BT wrote the manuscript and edited. All authors read and approved the final manuscript.

Ethics approval and consent to participate

The animal care and experiments were conducted according to the Administration of Affairs Concerning Experimental Animals (Ministry of Science and Technology, 2004) China. The protocol was approved on

01/03/2018 by the Institutional Animal Care and Use Ethics Committee of Inner Mongolia University, with the permit number for conducting animal experiments of (IMU-2018-01).

Consent for publication

Not applicable.

Competing interests

The authors declare no conflict of interest.

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Tables

Table 1 Information of six cattle populations selected for genotyping

Breed	Abbreviation	Number	Type
Mongolia cattle population (Inner Mongolia Autonomous Region of China)	MGC	41	North China (NC)
Mongolia cattle population (Mongolia)	MGG	50	North China (NC)
Qinchuan cattle	QC	350	Central China (CC)
Luxi cattle	UM	24	Central China (CC)
Wuling cattle	WL	50	South China (SC)
Longlin cattle	LL	50	South China (SC)

Table 2 Genotypic frequencies, allelic frequencies and diversity parameters of four SNPs in six cattle populations

SNP	Breed (Type ^a)	Number	Genotypic frequency			Allelic frequency		Diversity parameter				
								<i>Ho</i>	<i>He</i>	<i>n_e</i>	<i>PIC^b</i>	χ^2 (HWE ^c)
<i>c.*188G>A (AKIRIN2)</i>			<i>AA</i>	<i>AG</i>	<i>GG</i>	<i>A</i>	<i>G</i>					
	MGC (NC)	41	0.073	0.146	0.780	0.146	0.854	0.750	0.250	1.333	0.218	7.037
	MGG (NC)	50	0.140	0.460	0.400	0.370	0.630	0.534	0.466	1.873	0.358	0.009
	QC (CC)	350	0.051	0.343	0.606	0.223	0.777	0.654	0.346	1.530	0.286	0.036
	LX (CC)	24	0.000	0.167	0.780	0.083	0.917	0.847	0.153	1.180	0.141	0.198
	WL (SC)	50	0.020	0.220	0.760	0.130	0.870	0.774	0.226	1.292	0.201	0.038
	LL (SC)	50	0.000	0.000	1.000	0.000	1.000	1.000	0.000	1.000	0.000	-
<i>g.231054C>T(TTN)</i>			<i>TT</i>	<i>TC</i>	<i>CC</i>	<i>T</i>	<i>C</i>					
	MGC (NC)	41	0.000	0.122	0.878	0.061	0.939	0.885	0.115	1.129	0.185	0.173
	MGG (NC)	50	0.020	0.180	0.800	0.110	0.890	0.804	0.196	1.243	0.282	0.326
	QC (CC)	350	0.017	0.249	0.734	0.141	0.859	0.757	0.243	1.321	0.213	0.194
	LX (CC)	24	0.000	0.125	0.878	0.063	0.938	0.883	0.117	1.133	0.240	0.107
	WL (SC)	50	0.000	0.060	0.940	0.030	0.970	0.942	0.058	1.062	0.090	0.048
	LL (SC)	50	0.000	0.000	1.000	0.000	1.000	1.000	0.000	1.000	0.000	-
<i>g.42041062G>T(EDG1)</i>			<i>TT</i>	<i>GT</i>	<i>GG</i>	<i>T</i>	<i>G</i>					
	MGC (NC)	41	0.000	0.146	0.854	0.073	0.927	0.864	0.136	1.157	0.126	0.256
	MGG (NC)	50	0.000	0.180	0.820	0.090	0.910	0.836	0.164	1.196	0.150	0.489
	QC (CC)	350	0.034	0.237	0.729	0.153	0.847	0.741	0.259	1.349	0.226	2.489
	LX (CC)	24	0.000	0.042	0.854	0.021	0.979	0.959	0.041	1.043	0.040	0.011
	WL (SC)	50	0.000	0.060	0.940	0.030	0.970	0.942	0.058	1.062	0.057	0.048
	LL (SC)	50	0.000	0.020	0.980	0.010	0.990	0.980	0.020	1.020	0.020	0.005
<i>g.70014208A>G(MYBPC1)</i>			<i>GG</i>	<i>GA</i>	<i>AA</i>	<i>G</i>	<i>A</i>					
	MGC (NC)	41	0.829	0.146	0.024	0.902	0.098	0.824	0.176	1.214	0.161	1.170
	MGG (NC)	50	0.820	0.180	0.000	0.910	0.090	0.836	0.164	1.196	0.150	0.489
	QC (CC)	350	0.831	0.151	0.017	0.907	0.093	0.832	0.168	1.203	0.154	3.581
	LX (CC)	24	0.917	0.083	0.024	0.958	0.042	0.920	0.080	1.087	0.077	0.045
	WL (SC)	50	0.940	0.060	0.000	0.970	0.030	0.942	0.058	1.062	0.057	0.048
	LL (SC)	50	0.980	0.020	0.000	0.990	0.010	0.980	0.020	1.020	0.020	0.005

^aNC, North China; CC, Central China; SC, South China^bThe classification was conducted according to the PIC values (PIC value < 0.25, low polymorphism; 0.25 < PIC value < 0.5, moderate polymorphism; and PIC value > 0.5, high polymorphism)^cNo Hardy-Weinberg departure was detected from the obtained genotype frequencies

QC, Qinchuan; LX, Luxi; MGC, Mongolia cattle population (Inner Mongolia Autonomous Region of China); MGG, Mongolia cattle population (Mongolia); WL, Wuling; LL, Longlin; Ho, observed heterozygosity; He, expected heterozygosity; n_e , effective allele numbers; PIC, polymorphism information content; HWE, Hardy-Weinberg equilibrium

Table 3 Association of four SNPs with growth-related traits in Qinchuan cattle

SNP	Genotype (No.)	Body Length (cm)	Wither Height (cm)	Hip Height (cm)	Hip Width (cm)	Rump Length (cm)	Chest Depth (cm)	Chest Circumference (cm)
<i>c.*188G>A (AKIRIN2)</i>	GG (212)	133.24 ± 0.90 ^a	120.53 ± 0.61	123.33 ± 0.55	38.24 ± 0.41 ^{ae}	41.68 ± 0.32	58.02 ± 0.43 ^a	161.33 ± 1.22 ^a
	GA (120)	134.60 ± 1.18 ^b	121.67 ± 0.84	123.89 ± 0.72	38.42 ± 0.55 ^f	41.98 ± 0.40	59.30 ± 0.66	164.04 ± 1.68 ^b
	AA (18)	141.03 ± 3.89 ^b	124.19 ± 2.25	126.64 ± 2.06	41.36 ± 1.77 ^b	43.53 ± 1.17	62.69 ± 2.38 ^b	169.06 ± 4.50
<i>g.231054C>T(TTN)</i>	CC (257)	133.78 ± 0.82	120.41 ± 0.55 ^a	123.22 ± 0.48	38.17 ± 0.38	41.75 ± 0.27	58.19 ± 0.42 ^a	161.21 ± 1.10 ^a
	CT (87)	134.28 ± 1.49	122.62 ± 1.06 ^b	124.71 ± 0.91	39.03 ± 0.67	42.05 ± 0.57	59.90 ± 0.80 ^b	165.95 ± 2.11 ^b
<i>g.42041062G>T(EDG1)</i>	GG (255)	134.77 ± 0.82	121.28 ± 0.54	123.89 ± 0.47	38.87 ± 0.35 ^a	41.98 ± 0.28	58.83 ± 0.42	163.52 ± 1.09
	GT (83)	131.37 ± 1.49	120.13 ± 1.12	122.73 ± 1.02	37.02 ± 0.77 ^b	41.42 ± 0.55	58.23 ± 0.85	159.25 ± 2.24
	TT (12)	138.96 ± 3.67	124.33 ± 2.27	126.08 ± 1.98	39.83 ± 1.80	42.75 ± 1.44	59.13 ± 1.78	167.83 ± 4.87
<i>g.70014208A>G(MYBPC1)</i>	GG (291)	135.19 ± 0.80 ^e	121.71 ± 0.53 ^a	124.11 ± 0.47 ^a	38.89 ± 0.35 ^c	42.19 ± 0.27 ^c	59.04 ± 0.41 ^a	163.93 ± 1.07
	GA (53)	128.17 ± 1.41 ^f	118.25 ± 1.17 ^b	121.57 ± 1.03 ^b	35.97 ± 0.82 ^d	40.22 ± 0.54 ^d	56.85 ± 0.91 ^b	156.16 ± 2.31

Values are shown as the means ± standard error; Values with different superscripts within the same column differ significantly at $P < 0.05$ (^{a, b}), $P < 0.01$ (^{c, d}) and $P < 0.001$ (^{e, f}) after Bonferroni correction

Table 4 Statistical significance for differences in the allele frequencies of the four SNPs among the six groups of cattle populations

SNP/Group	Group ^a				
	Central China	South China	Japanese cattle group	European cattle group	Korean cattle ^b
<i>c.*188G>A</i> SNP (<i>AKIRIN2</i>) ^c					
North China	n.s. ^d	***	**	n.s.	***
Central China		***	***	n.s.	***
South China			***	***	***
Japanese cattle group				***	n.s.
European cattle group					***
<i>g.231054C>T</i> SNP (<i>TTN</i>) ^c					
North China	n.s.	**	***	***	-
Central China		***	***	***	-
South China			***	**	-
Japanese cattle group				***	-
<i>g.1471620G>T</i> SNP (<i>EDG1</i>) ^c					
North China	*	**	***	***	-
Central China		***	***	***	-
South China			***	*	-
Japanese cattle group				***	-
<i>g.70014208A>G</i> SNP (<i>MYBPC1</i>) ^c					
North China	n.s.	**	***	***	-
Central China		***	***	***	-
South China			***	***	-
Japanese cattle group				***	-

^aNorth China: MGC and MGG; Central China: QC and LX; South China: WL and LL; Japanese cattle group: JB, JBR and JSH; European cattle group: HOL and BS, and Korean cattle;

^bThe data of the *c.*188G>A* SNP (*AKIRIN2*) in Korean native cattle was referred from [119];

^cThe data of the *g.1471620G>T* SNP (*EDG1*), *c.*188G>A* SNP (*AKIRIN2*), *g.231054C>T* SNP (*TTN*), and *g.70014208A>G* SNP (*MYBPC1*) in Japanese and European cattle breeds were referred from [21-23];

^dn.s., Non-significant. *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$

Figures

Figure 1

Geographic map indicating the origins of the cattle populations. Cattle populations of North China type in green; Cattle breeds of Central China type in yellow; Cattle breeds of South China type in blue; Japanese cattle breeds in purple; Korean native cattle in red; European cattle breeds in brown. MGM, Mongolia cattle population (Inner Mongolia Autonomous Region of China); MGC, Mongolia cattle population (Mongolia); QC, Qinchuan; LX, Luxi; WL, Wuling; LL, Longlin; JB, Japanese Black; JBR, Japanese Brown; JSH, Japanese Short Horn; KN, Korean native cattle; HOL, Holstein; BS, Brown Swiss. Note: The designations employed and the presentation of the material on this map do not imply the expression of any opinion whatsoever on the part of Research Square concerning the legal status of any country, territory, city or area or of its authorities, or concerning the delimitation of its frontiers or boundaries. This map has been provided by the authors.

Figure 2

Distribution of allele frequency in the six cattle groups. (a-d) The allele frequency of the c.*188G>A SNP in AKIRIN2, the g.231054C>T SNP in TTN, the g.1471620G>T SNP in EDG1, and the g.70014208A>G SNP in MYBPC1 genes, respectively. NC: North China; CC: Central China; SC: South China; JG: Japanese cattle group [21-23]; EC: European cattle group [21-23]; KN: Korean cattle [19].

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