BOAR SNP VARIABILITY IN GENETIC RESOURCE PŘEŠTICE BLACK-PIED PIG

Falková L., Vrtková I., Kratochvílová L.

Mendel University in Brno, Laboratory of Agrogenomics, Czech Republic

Abstract

The aim of study was to genotype Přeštice Black-Pied boars in the SNP markers and to evaluate the genetic variability. The genetic variability of genetic resource Přeštice Black-Pied (Pc) was analysed by genotyping 92 breeding boars. The research of molecular variability is useful for optimal conduct of their breeding and knowledge of relations between molecular markers and traits. In this study was chosen markers with important association with production and economical traits. Seven single nucleotide polymorphisms were used for analysis: *CRC*, *ESR2*, *FUT1*, *MUC4*, *Mx1*, *MC4R*, *TEAD3*. We observed genetic variability in these markers. Some alleles was due to breeding almost lost and occurred only in heterozygous genotypes.

Key Words: Pig, genetic variability, Přeštice Black-Pied , SNP

The Přeštice Black-Pied (Pc) was declared as a genetic resource in 1992. The genetic resources are very important branch of research, because these populations are not influenced by selection as much as the commercial hybrid lines. The study of genetic polymorphisms is useful for preservation and conservation. The Přeštice Black-Pied pigs are characterised by high fertility and excellent maternal properties, on the other hand they carry higher back-fat thickness and lower ratio of muscles.

The *CRC* gene effected manifestation of malignant hyperthermia syndrome (MHS) with important impact on meat quality (e.g. FUJII et al., 1991). The pigs of CRC^n/CRC^n genotype achieve lower total number of born piglets, number piglets born alive, number of weaned piglets (WITTMANN et al., 1992; DVOŘÁK, 1994).

The *FUT1* gene has been determined as a candidate gene for *Escherichia coli* F18 receptor locus (MEIJERINK et al., 1997). Fimbriated *E. coli* adheres to brush border membranes in the small intestine and brings about oedema disease and post-weaning diarrhoea in pigs (BERTSCHINGER et al., 1990). The M307 polymorphism of the *FUT1* gene influences susceptibility to adhesion of *E. coli* F18 to intestinal mucosa and an outbreak of illness (MEIJERINK et al., 2000). In spite of the noted expression of the *FUT1* gene in the murine uterus, no evident effect of this gene on fertility has been confirmed so far (SIDHU & KIMBER, 1999; DOMINO et al., 2001).

In the *ESR* gene, the *PvuII* polymorphism (*ESR2*) was described by ROTHSCHILD et al. (1991). Associations between the *ESR2* polymorphism and litter size in pigs have been observed many times, but the outcomes are ambiguous (SHORT et al., 1997).

The *MUC4* gene are associated with diarrhoea piglets and post-weaning pigs caused by enterotoxic *E. coli* with F4 fibres. The ETEC F4 resistence shows autosomal recessive inheritance (SELLWOOD, 1979).

Gene *Mx1* is connected with resistance again influensa virus. Deletion of 11 bp influences replication of virus (NAKAJIMA et al., 2007) and lost of ability to inhibit dispersion of virus (VRTKOVÁ et al., 2006).

Widely studied in pigs has also been the effect of *MC4R* polymorphism on feed intake and carcass fatness traits. When studying the relationship between melanocortin receptor polymorphism and carcass fatness, feed intake and weight gain traits in pigs, CHEN et al. (2004), KIM et al. (2006) and MEIDTNER et al. (2006) found that the *MC4R* gene can be used as a genetic marker for these traits in animal selection. VAN DEN MAAGDENBERG et al. (2007) demonstrated that mutation in the melanocortin receptor gene affects the weight gain and carcass fatness in pigs, but not quality of their meat.

The *TEAD3* gene is considered as a candidate gene of QTL for accumulation of androstenone in fat. It was found one SNP in *TEAD3*-related sequences with the right profile to explain the androstenone QTL (ROBIC et al., 2012). The T allele was connected with a lower androstenone level in LW.

Material and Methods

The genetic variability was analysed by genotyping 92 Pc breeding boars. The extraction of genomic DNA from blood cell was carried out by Genomic DNA Mini Kit (Blood/ Cultured Cell) (Geneaid) according to the instructions. The PCR-RFLP methods according to Laboratory of Agrogenomics standard were used.

Results

We detected dominant homozygous, only two heterozygous and no boars of recessive homozygous genotypes for *CRC* gene (tab. 1). The frequency of the allele CRC^n (0,011) was markedly lower than the frequency of the allele CRC^N (0,989).

In *ESR2* locus, the higher frequency of the $ESR2^C$ allele was detected (0,958) than $ESR2^D$ (0,042) and genotype $ESR2^D/ESR2^D$ missed too.

A higher frequency of the allele $FUT1^G$ (0,860) compared with the allele $FUT1^A$ (0,140) was determined. Higher frequency of homozygous $FUT1^G$ was found.

We detected lower frequency of the allele $MUC4^B$ (0,242) than $MUC4^A$ (0,758) and no genotype $MUC4^B/MUC4^B$ was observed.

Markedly higher presence of the allele $Mx1^O$ (0,909) was recorded in comparison with $Mx1^P$ (0,091) and only 0,011 genotype $Mx1^P/Mx1^P$ frequency was detected.

Similar allelic frequencies were found out in the *MC4R* gene – $MC4R^A$ (0,517) and $MC4R^B$ (0,483). Heterozygous $MC4R^A/MC4R^B$ individuals dominated.

The allelic frequency of $TEAD3^C$ (0,624) was higher than $TEAD3^T$ (0,376) and number of homozygous $TEAD3^T$ / $TEAD3^T$ was very low.

Discussion

A higher incidence of CRC^n alleles in Pc found ČEPICA et al. (1982) (0,348) and HORÁK et al. (2004) (0,083) and they also confirmed absence CRC^n/CRC^n genotype. The reduction of the recessive allele must have been caused by efforts of exclusion of heterozygous and CRC^n/CRC^n genotypes from reproduction in pedigree breedings. We can found differences in the *CRC* gene variability in Czech Landrace (CL) and Czech Large White (CLW) breed in the Czech Republic, where the recessive allele frequency 0,01 in Czech Landrace was detected (BEČKOVÁ et al., 2002) and 0,025 – 0,045 in LW (MATOUŠEK et al., 2003).

A higher frequency of the allele $ESR2^A$ than the allele $ESR2^B$ similarly as our $ESR2^C$ $ESR2^D$ found HORÁK et al. (2005). VRTKOVÁ & DVOŘÁK (2001) found the frequency of the allele $ESR2^B$ to be 0,35 in CLW sows and 0,10 in CL sows. Thus in the Pc sows, the variability in the *ESR2* polymorphism is more similar to the CL breed than the CLW breed.

The similar frequency of alleles $FUT1^A$ (0,216) and $FUT1^G$ (0,784) found HORÁK et al. (2005). On the basis of past research on other breeds, higher incidence of a recessive allele $FUT1^A$ was expected in Pc as well. For example, KLUKOWSKA et al. (1999) noted that in the Polish local breed, Zlotnicka Spotted, the frequency of this allele was 0,63. The lower frequency in Pc may have originated in the absence of the natural selection against susceptible animals in the past.

The $MUC4^B/MUC4^B$ was not detected in Pc sows by MATOUŠEK et al. (2011). The allelic frequency of $MUC4^A$ in commercial breeds was 0,52 (VRTKOVÁ et al., 2007).

MATOUŠEK et al. (2011) found in Pc sows missing genotype $Mx1^P/Mx1^P$ and allelic frequencies were similar to our – $Mx1^O$ 0,907, $Mx1^P$ 0,093. In boars of commercial breeds (CLW, CL, D) recorded similar allelic frequencies VRTKOVÁ et al. (2007) – for allele $Mx1^O$ 0,85, and in Pc breed 0,85.

The highest frequency of heterozygous $MC4R^A/MC4R^B$ described MATOUŠEK et al. (2011) too. They found the similar allelic frequencies $MC4R^A$ (0,547) a $MC4R^B$ (0,453) too. The higher occurrence of $MC4R^B$ allele was recorded in Pietrain (Pn) (0,924) (PIORKOWSKA et al., 2010).

Degree of admixture of commercial breeds and Pc breed based on variability of microsatellite markers described VRTKOVÁ (2014). Author focused on evaluation of three breeds - CL, CLW a Pn which participated in improving Pc breed. During clustering of breeds, Pn was the last one separated from Pc. According to microsatellite markers is Pc close to Pn.

VRTKOVÁ et al. (2007) analysed three SNP markers identically with us (*FUT1*, *MUC4* and *Mx1*) in Pn breed. Frequencies of genotypes in given markers for Pc vs. Pn are following: $FUT1^A/FUT1^A$ 0.023/0.088, $FUT1^A/FUT1^G$ 0.233/0.415, $FUT1^G/FUT1^G$ 0.744/0.497, $MUC4^A/MUC4^A$ 0.517/0.766, $MUC4^A/MUC4^B$ 0.483/0.222, $MUC4^B/MUC4^B$ 0.000/0.012, $Mx1^O/Mx1^O$ 0.921/0.439, $Mx1^O/Mx1^P$ 0.068/0.421, $Mx1^P/Mx1^P$ 0.011/0.140.

The same frequency of $TEAD3^T$ allele (0,37) described ROBIC et al. (2012) in Landrace and slightly higher in Duroc (0,52).

Table 1. Allelic and genotypes frequencies of six SNP markers in Pc

MARKER	allele	allele frequency	genotype	genotype frequency
CRC	*N*	0.989	*NN*	0.978
	n	0.011	*Nn*	0.022
ESR2	*C*	0.958	*CC*	0.917
	D	0.042	*CD*	0.083
FUT1	*A*	0.140	*AA*	0.023
	G	0.860	*AG*	0.233
			GG	0.744
MUC4	*A*	0.758	*AA*	0.517
	B	0.242	*AB*	0.483
Mx1	*O*	0.909	*OO*	0.921
	P	0.091	*OP*	0.068
			PP	0.011
MC4R	*A*	0.517	*AA*	0.213
	B	0.483	*AB*	0.607
			BB	0.180
TEAD3	*C*	0.624	*CC*	0.337
	T	0.376	*CT*	0.573
			TT	0.090

Conclusion

In this study we detected considerable molecular genetic variability in genes connected with economical traits in Přeštice Black Pied breed. The results confirmed an importance of preservation and conservation genetic resources. The obtained allelic frequencies of markers *CRC, FUT1*, *Mx1* and *TEAD3* was similar to commercial breeds (CLW, CL, Pn).

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Corresponding Address:
Mgr. Lenka Falková
Mendel University in Brno,
Zemědělská 1, 613 00 Brno
Czech Republic
E-mail: lenka.falkova@mendelu.cz

The research was supported by project MZe ČR QJ1210253.