

Genetic characterization of Andaman Desi pig, an indigenous pig germplasm of Andaman and Nicobar group of islands, India by microsatellite markers

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Abstract

Aim: The present study was carried out to characterize Andaman Desi pig genetically using 23 FAO recommended microsatellite markers.

Materials and Methods: 25 blood samples were collected from genetically unrelated Andaman Desi pigs and DNA was isolated by standard procedure of phenol/chloroform. The genomic DNA was amplified by polymerase chain reaction (PCR) at 23 microsatellite loci and the PCR products were resolved by denaturing urea polyacrylamide gel electrophoresis and alleles were visualized after silver nitrate staining. The data were analyzed for allele size range, number of alleles, allelic frequencies, heterozygosity and polymorphism information content (PIC) for each locus.

Results: The allele size range varied from 86–116 bp at locus SW936 to 280–296 at locus IGFI. The total number of alleles ranged between 5 (S0228, SW122, SW951, SW24 and S0178) and 12 (S0355). The effective number of alleles ranged from 3.14 (SW24) to 8.1 (S0355). The mean observed and expected heterozygosities were 0.69 ± 0.01 and 0.77 ± 0.01 respectively. The mean PIC for all the 23 studied loci was 0.74 ± 0.01 .

Conclusions: The Andaman Desi pig was characterized at genetic level and it was found that the genetic diversity of this indigenous breed was high.

Keywords: Andaman Desi pig, genetic characterization, microsatellite, polymorphic information content

Introduction

The Andaman and Nicobar Islands are a group of 572 big and small islands & islets in the south eastern part of Bay of Bengal. The temperature of these Islands varies from 27°C to 30°C. The annual rainfall of these islands is about 3100 mm and enjoys a tropical hot humid climate [1]. Geographically these islands are distinguished into two groups, i.e. the Andaman group and Nicobar group, separated by 10° N channel. The total land area of all these islands is about 8249 sq. km of which about 86 percent is covered by tropical rain forest.

Local indigenous breeds of domestic animals are very much adapted to the local environmental conditions, so conservation of these breeds is very important [2-4]. Pigs constitute 27.26 % of the total livestock of Andaman and Nicobar Islands and mostly reared by the tribes and native people of Andaman and Nicobar Islands [1]. There are four different genetic groups of pigs available in these Islands. They are Andaman wild pig, Nicobari pig, Andaman Desi pig and pure and cross breeds of Large White Yorkshire.

The Andaman Desi pig (Figure-1) is mostly found in Andaman group of islands. Its body color varies from rusty grey to brown and black. The hair on the neck and back portion are thick and long and those on the flank and sides are relatively thinner and shorter.



Figure-1. Andaman Desi pig

At present, this indigenous pig breed needs immediate conservation effort. No information is available regarding genetic structure of this indigenous pig breed. For development of cost effective conservation approach, genetic characterization is very much

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Table-1. Observed and effective number of alleles, observed and expected heterozygosity and polymorphic information content in 23 FAO-recommended microsatellite loci in Andaman Desi pig

Locus	Allele size range (bp)	Observed no. of alleles	Effective no. of alleles	Observed heterozygosity	Expected heterozygosity	Polymorphism information content
CGA	266–302	10	5.78	0.68	0.74	0.73
S0005	216–255	8	4.56	0.71	0.71	0.65
S0090	241–253	6	4.32	0.68	0.79	0.79
S0215	127–155	7	5.56	0.69	0.76	0.72
S0218	164–184	6	4.52	0.68	0.83	0.8
S0226	185–217	7	5.9	0.67	0.71	0.67
S0228	231–255	5	4.26	0.74	0.8	0.79
S0386	156–172	6	4.35	0.65	0.76	0.74
SW72	100–122	8	4.77	0.68	0.79	0.72
SW122	112–138	5	5.54	0.76	0.83	0.77
SW632	151–163	6	4.62	0.61	0.72	0.69
SW857	145–157	7	5.72	0.63	0.71	0.7
SW911	156–178	8	5.59	0.65	0.75	0.73
SW936	86–116	9	5.52	0.66	0.7	0.71
SW951	115–125	5	4.15	0.62	0.76	0.69
S0355	247–263	12	8.1	0.77	0.87	0.83
SW24	90–110	5	3.14	0.74	0.79	0.78
S0225	172–180	7	5.63	0.68	0.75	0.77
S0227	243–239	8	5.52	0.67	0.73	0.72
S0178	110–124	5	4.66	0.67	0.76	0.74
S0068	218–248	6	5.53	0.68	0.78	0.76
IGFI	280–296	8	4.78	0.69	0.78	0.73
S0026	91–109	8	4.64	0.76	0.81	0.79
Mean±SEM		7.04±0.37	5.09±0.20	0.69±0.01	0.77±0.01	0.74±0.01

essential [5]. Monitoring the genetic variability of the actual population by use of DNA molecular markers is essential also to address the correct breeding policy and its fitness in the future [6]. The microsatellite markers have evolved as useful polymorphic markers and have been used in genetic characterization of various pig breeds across the globe [7-16].

The objective of the present study was to characterize Andaman Desi pig by 23 FAO recommended microsatellite markers.

Materials and Methods

Ethical approval: The present experiment comply with all relevant institutional and national animal welfare guidelines and policies. Blood samples from pigs were collected aseptically following standard national welfare guidelines.

Biological sample collection and DNA isolation: Twenty five blood samples of Andaman Desi pigs were collected from different villages of Andaman and Nicobar islands. DNA was isolated by standard procedure of phenol/chloroform. The DNA samples were stored at -20°C and/or at 4°C.

Polymerase chain reaction: 23 FAO recommended microsatellite primers were used for amplification of the genomic DNA by polymerase chain reaction (PCR). PCR reactions were carried out as described by Kaul et al. [17]. Briefly, each 25 µl reaction consisted of DNA (approximately 100 ng), primers (60 ng each), dNTPs (40 mm of each), 10X buffer (10 mm Tris, 50 mm KCl, 0.1% gelatin, pH 8.4) (2.5 µl), MgCl₂ (1.5 mm) and Taq DNA polymerase (0.75 units). The thermocyclic conditions were initial denaturation at 94°C for 2 min followed by 30 cycles of denaturation at 94°C for 45 s, annealing at optimum temperature (FAO

1998) for 45 s and extension at 72°C for 45 s with a final extension at 72°C for 10 min. The amplified fragments were analyzed on 8% denaturing urea-polyacrylamide gel and detected by silver staining.

Statistical analysis: The observed and expected heterozygosity, observed and effective number of alleles were calculated using popgene computer package [18]. The polymorphism information content (PIC) was calculated as described by Botstein et al. [19].

Results

The allele size range, observed and effective number of alleles, observed and expected heterozygosity and polymorphic information content (PIC) at 23 loci in Andaman Desi pig is presented in Table-1. The allele size range varied from 86–116 bp at locus SW936 to 280–296 at locus IGFI. The total number of alleles ranged between 5 (S0228, SW122, SW951, SW24 and S0178) and 12 (S0355). The effective number of alleles ranged from 3.14 (SW24) to 8.1 (S0355). The mean observed and expected number of alleles for all 23 loci in Andaman Desi pigs were 7.04±0.37 and 5.09±0.20 respectively. The observed heterozygosities of Andaman Desi pig were lower than the expected values at 22 studied loci whether in S0005 the value was same. The mean observed and expected heterozygosities were 0.69±0.01 and 0.77±0.01 respectively. The mean PIC for all the 23 studied loci was 0.74±0.01.

Discussion

Domestic pig is one of the most widespread domestic animal species in the world and complex factors such as men-deriving forces and natural selection have influenced the actual diversity and

population structure of the species [20]. Andaman Desi pig (Figure-1) is one of the indigenous pig breeds of Andaman and Nicobar islands which is mainly found in Baratang and Mayabander area of Andaman. Its body color varies from rusty grey to brown and black. Both male and female have a long tail. The approximate adult body weight ranges from 75-80 Kg for male and 60-70 Kg for female. Age at first farrowing is about 300 days with litter size of about 7-8. They maintain good health with low plane of nutrition [1]. Microsatellite markers have been used widely for the genetic characterization of animal breeds including pig [21-24]. The use of microsatellites has revealed high levels of genetic diversity amongst the huge pool of genetic resources of pigs around the world [23]. In the present study 23 FAO recommended microsatellite markers were used to characterize Andaman Desi pig genetically.

The microsatellite analysis of 23 loci of Andaman Desi pig revealed that the mean observed number of alleles and mean effective number of alleles were 7.04 ± 0.37 and 5.09 ± 0.20 respectively. The mean value of effective number of alleles for Andaman Desi pig was found higher than South-African pig breeds Mozambique (8.45), Kolbroek (6.18) and Kune-Kune (5.97) but was lower than Duroc (3.98) [25]. The mean effective number of alleles of the Indian pig breeds Desi, Gahuri and Ankamali were 5.00, 5.33 and 5.34 respectively [5] which were comparable with our present study. The relatively higher number of alleles in the India populations over European breeds indicates that the effects of isolation and selection of these populations has been mild.

Mean observed and expected heterozygosities of 23 microsatellite loci of Andaman Desi pig were 0.69 ± 0.01 and 0.77 ± 0.01 respectively which were higher when compared with the Large White Yorkshire and other European pig breeds [7-9]. Higher observed and expected heterozygosities were also reported in South-African pig breeds Mozambique, Kolbroek and Kune-Kune [25]. The mean of observed heterozygosity in our study is higher than that reported in Cinta Senese pig breed of Italy [6].

The genetic diversity in Andaman Desi pig is higher than the European pig breeds. Genetic diversity in other Indian pig breeds like Desi, Gahuri and Ankamali was also found high [5]. The observed heterozygosities of Desi, Gahuri and Ankamali were 0.71 ± 0.14 , 0.68 ± 0.12 and 0.74 ± 0.09 respectively and the expected heterozygosities were 0.80 ± 0.06 , 0.79 ± 0.07 and 0.83 ± 0.03 respectively [5]. High genetic diversity was also reported in Chinese pig populations [12]. Large White Yorkshire and other European pig breeds are maintained in small populations whereas Indian and Chinese breeds have large effective populations; that may be the reason behind high heterozygosity values of Indian and Chinese breeds [5].

Mean PIC of all the 23 microsatellite loci of Andaman Desi pig was 0.74 ± 0.01 which was found

higher than Large White Yorkshire but comparable with other Indian pig breeds like Desi, Gahuri and Ankamali [5]. Polymorphism information content (PIC) values of Taiwan black pigs and other indigenous pig breeds of Taiwan were reported to be above 0.50 [26]. PIC values of all the microsatellite loci in the present study were above 0.5 which indicates that the microsatellite loci were suitable for detection of genetic diversity in Andaman Desi pig.

Mean observed and expected heterozygosities of 23 microsatellite loci of Andaman Desi pig were found high indicating high genetic diversity of this pig breed. From the microsatellite data it was also found that this pig breed is distinguishable from other pig breeds. As the pig breed is under the threat of extinction due to extensive cross breeding, serious effort must be initiated to conserve this breed in its breeding tract. As the current experiment confirms this breed as a distinguishable breed, it will assist in formulation of effective conservation strategy and breeding policy for this important indigenous pig breed.

Conclusion

Genetic characterization of Andaman Desi pig was carried out by 23 FAO recommended microsatellite markers. It was found that the genetic diversity of this pig breed was very high compared to Large White Yorkshire and other European pig breeds. High genetic diversity of this pig breed was found consistent with other Indian pig breeds like North Indian Desi, Gahuri and Ankamali. It may be concluded that Andaman Desi pig is genetically distinguishable from other Indian and exotic pig breeds. This genetic characterization of the pig breed will be helpful in their conservation effort.

Authors' contributions

AKDe, SJ and MSK were involved in the design of the experiment. The experiment was done by AKDe, JS and MR. AK revised the final draft of manuscript. All authors read and approved the final manuscript.

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Competing interests

The authors declare that they have no competing interests.

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