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Genetic Diversity of Six Duck Populations in India

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ABSTRACT

Background: The variety of indigenous duck germplasm contribute maximum to the poultry industry in India, besides chicken population. In southern part of India, the available duck genetic resource, particularly Arni ducks (comprises of Sanyasi and Keeri ducks) of Tamil Nadu, has its own characteristics with innate potentiality of higher productivity without any input system of management. Genetic characterization and diversity of indigenous duck genetic resources has not been properly studied. In the present study, the genetic diversity of Arni ducks with other indigenous and exotic duck germplasm were analysed with microsatellite markers.

Method: Genomic DNA was isolated from the blood samples of six duck populations. Molecular characterization was carried out with duck specific FAO recommended microsatellite markers. The genotyping of ducks was done based on the size of 4324 PCR amplicons of 23 microsatellite loci, which were subjected to capillary electrophoresis using automatic sequencer.

Result: A total of 222 alleles in six duck populations across 23 microsatellite loci with a mean of 9.65 ± 0.95 alleles were found. Kuttanad duck variety had the highest number of alleles (139) followed by Sanyasi (136), Keeri (129), Muscovy (118), Assam (91) and White Pekin (78) ducks. The mean observed number of alleles was 6.04 ± 0.59, 5.91 ± 0.76, 5.61 ± 3.17, 5.13 ± 0.44, 3.96 ± 0.76 and 3.39 ± 0.40 in Kuttanad, Sanyasi, Keeri, Muscovy, Assam and White Pekin ducks respectively. The overall mean polymorphism information content (PIC) values among the six duck populations was 0.6269. In most of the duck populations, the mean PIC value was more than 0.5 except in Assam (0.4815) and White Pekin (0.3725) ducks. The observed heterozygosity was the highest in Keeri ducks (0.5217) and lowest in White Pekin ducks (0.2766), while, the mean expected heterozygosity was the highest in Sanyasi (0.5628) and lowest in White Pekin (0.4038) ducks. The variations in the observed and expected number of alleles, differences in PIC of various microsatellite loci might be attributed to the genetic variability of the duck populations, number and type of microsatellite primers utilised for analysis and the genetic diversity of the duck breeds under study. Higher F_{ST} value indicates the substantial degree of breed differentiation among the studied duck populations.

Key words: Arni ducks, Sanyasi, Keeri ducks, Genetic diversity, Microsatellite markers.

INTRODUCTION

Among the diversified poultry species, ducks are highly prolific with innate resistance and maximum production potentiality. Small scale duck production makes a significant contribution to household economics and food security. Mainly ducks are being managed by foraging and maintained on free-range system. The distribution and demographic dynamics of duck population revealed that they are mainly concentrated in eastern, north eastern and southern states of the country. The leading states in duck population are West Bengal, Assam, Kerala, Andhra Pradesh, Tamil Nadu, Bihar and Orissa. The indigenous duck varieties are mainly the non-descript ducks with varied phenotypic and genetic makeup, available in large numbers in many states of the country, contributing significantly to the total duck population. These indigenous ducks have innate potential to produce eggs and meat at considerable quantity with lesser input and they are a good dietary source of proteins.

In order to conserve this genetic resource, a study on genetic diversity within and between duck populations using microsatellite markers would provide information for taking priority decisions towards preservation. According to FAO (2004) recommendations, using neutral, highly polymorphic microsatellite markers are currently the method of choice for determination of genetic variation and breed differentiation. This methodology also provides information for establishing preservation priorities for livestock breeds. Microsatellite markers are frequently used in genotype identification, pedigree analysis and estimation of genetic diversity and genetic distance (Kumar *et al*., 2011) and phylogenetic analysis (Seo *et al*., 2015). The characterization of genetic diversity by employing molecular tools is a prerequisite in developing strategies for conservation, utilization of duck genetic resources and to the establishment of a sensible genetic preservation strategy for these populations. Therefore, the present study was undertaken using the microsatellites, which are powerful genetic markers for biodiversity evaluation, to study the genetic diversity of

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Arni ducks (Indigenous duck of Tamil Nadu) with other duck germplasm available in India.

MATERIALS AND METHODS

A total of 190 blood samples were collected from unrelated birds of both sexes of Arni ducks (Sanyasi, Keeri variety) in the study area, Kuttanad ducks from Kerala, Assam ducks, White Pekin and Muscovy ducks for molecular characterisation and genetic diversity analysis of the duck populations. From the blood samples, the genomic DNA was isolated by standard protocol (Sambrook *et al*., 1989). The FAO recommended 23 microsatellite markers with labelled forward primers were used for molecular characterization. These microsatellite primers were standardised for their annealing temperatures. Molecular characterization of various duck varieties was carried out using standard PCR protocol by amplifying all the primers with varying annealing temperatures. The PCR products were checked for its amplification by agarose gel electrophoresis. The PCR amplicons were genotyped by capillary electrophoresis using automatic sequencer. Number of alleles, effective number of alleles, allele frequencies, observed and expected heterozygosities and F-statistics were calculated by using POPGENE version 1.31. (Felsenstein, 1993)

RESULTS AND DISCUSSION

Microsatellite allelic diversity

In the present study, a total of 222 alleles were identified with 23 microsatellite loci among six duck populations and the number of alleles ranged from four (CAUD033) to 25 (CAUD024) per locus. Higher number of total alleles than those observed in the present study was reported by earlier authors (236 alleles in 24 loci for six Chinese duck breeds by Li *et al*., 2006; 281 alleles in 20 loci among six Chinese duck populations by Wu *et al*., 2008). In contrast to this, lesser number of alleles were also reported by several authors (177 alleles in 29 loci for 10 Chinese ducks by Li *et al*., 2010; 50 alleles in 21 primers for Moti Indian native ducks reported by Alyethodi and Kumar, 2010; 37 alleles in 6 loci for three local duck populations of Indonesia by Ismoyowati and Purwantini, 2010; 48 alleles in 9 loci among two Indian duck populations by Kumar *et al*., 2011; 153 alleles in 22 loci for 8 Indonesian duck populations by Hariyona *et al*., 2019). The higher mean value of alleles obtained per locus is indicative of polymorphic nature of the loci and the genetic diversity of the duck populations studied. Higher mean number of alleles at a given locus might be due to automated sequencing followed which is more accurate and sophisticated than the conventional methods followed by most of the workers.

Results revealed that the highest mean observed number of alleles (Table 1) was obtained in Kuttanad ducks (6.04 ± 0.59) and lowest values in White Pekin ducks (3.39 ± 0.40) and Assam ducks (3.96 ± 1.55). Lesser observed number of alleles than the present study was reported in White Pekin (2.22) and Muscovy ducks (2.44) of Iran by Ahmadi *et al*. (2007). Among Indian duck populations, less number of observed alleles was reported in Moti (3.4) and Indian Runner (3.3) by Kumar *et al*. (2011), while higher observed number of alleles (8.0) was reported in Assam ducks by Mukesh *et al*. (2011). They also reported the mean observed number of alleles in West Bengal and Uttarakhand ducks as 4.11 and 5.0 respectively. The reason for lesser mean number of alleles in White Pekin and Muscovy duck breeds in the present study may be due to less number of samples collected for the study (only 15 number of ducks in each breed) as against 50 number of samples collected in other populations.

It was found that the mean effective number of alleles obtained in the present study (Table 1) was highest in Sanyasi (2.86 ± 0.41) and lowest in White Pekin (2.01 ± 0.21) ducks. The mean effective number of alleles in Assam ducks (2.31 ± 0.16) obtained in the present study was lesser than the value obtained (4.21) by Mukesh *et al*. (2011). Higher effective number of alleles than those of the present study was reported (4.80, 3.90 and 3.60) by Li *et al*. (2006), Ying Su *et al*. (2007) and Kumar *et al*. (2011) respectively. The variations in the observed and expected number of alleles might be attributed to the genetic variability of the duck populations, number and type of microsatellite primers utilised for analysis and the difference in the duck breeds under study.

All the 23 microsatellite loci studied were highly polymorphic. This finding is in agreement with the reports of Li *et al*. (2006), Ying Su *et al*. (2007), Gaur *et al*. (2009), Su and Chen (2009), Alyethodi and Kumar (2010), Mukesh *et al*. (2011) and Kumar *et al*. (2011). The effective numbers of alleles is also an index used to reveal the genetic diversity of duck populations and the highly polymorphic loci indicated that these microsatellite loci could be used as effective markers for genetic diversity and phylogenetic relationship analysis among duck breeds. But the Indian duck populations used in the present study have considerably lower mean effective number of alleles, even though the mean number of observed alleles was quite high. The lower value indicates the occurrence of most of the alleles with lesser frequencies and the declining alleleic variation within each duck population might be due to closed nature of the populations.

Polymorphism information content

In general the polymorphism information content (PIC) values are suggestive of high polymorphic nature of the microsatellite loci analysed. The PIC was originally introduced by Botstein *et al*. (1980). It refers to the value of a marker for detecting polymorphism within a population depending upon the number of detectable alleles and distribution of their frequency and has been proved to be a general measure of how informative a marker is (Guo and Elston, 1999), the higher the PIC value the more informative a marker.

The PIC is a good index for genetic diversity evaluation. When PIC is more than 0.5, the locus has high diversity; when PIC is less than 0.25, the locus has low diversity; and the locus has intermediate diversity when PIC is in between 0.25 and 0.5. The overall mean PIC values obtained in the present study among the six duck populations was 0.6269. In most of the duck populations, the mean PIC value (Table 3) was more than 0.5 except in Assam (0.4815) and White Pekin (0.3725) ducks. The PIC value of the microsatellite loci CAUD017 in Muscovy duck was the highest (0.8580), but PIC of the loci CAUD025 in Muscovy, Assam and Kuttanad ducks was zero as it was monomorphic in these populations. Similarly, higher PIC value of more than 0.5 was observed at most of the loci in Chinese ducks (Li *et al*., 2006; Wu *et al*., 2008; Su and Chen, 2009; Seo *et al*., 2016; Hariyona *et al*., 2019), Indonesian ducks (Ismoyovati and Purwantini, 2010) and Indian ducks by Kumar *et al*. (2011) while Alyethodi and Kumar (2010) observed moderate PIC value in Moti ducks (0.45) with the same set of markers. Highest mean PIC value was observed in Kuttanad ducks (0.6264) and lowest value was obtained for White Pekin ducks (0.3725) in this study. The differences in PIC of various microsatellite loci may be due to genetic differences in the population analysed. This indicated that the selected microsatellite loci had high diversity which can reflect the genetic relationship among different populations at molecular level and these loci are highly informative.

Heterozygosity

Genetic diversity can be measured as the amount of actual or potential heterozygosity. Heterozygosity is one of the indices used to assay the genetic variation of each population. The values of heterozygosity indicate the diversity level of the molecular marker. When the value is high, the genetic diversity of the molecular marker is also high. Among the six duck populations studied, the observed heterozygosity (Table 2) was the highest in Keeri ducks (0.5217) and lowest in White Pekin ducks (0.2766). Further the results revealed that the mean expected heterozygosity was the highest in Sanyasi (0.5628) and lowest in White Pekin (0.4038) ducks.

Among different loci analysed, the locus CAUD024 had the highest observed (0.8351) and expected (0.9211) heterozygosity in most of the duck populations (Table 2). Similar heterozygosity value of less than 0.6 was observed by Paulus and Tiederman (2003), Ahmadi *et al*. (2007), Gaur

Table 1: Microsatellite allelic diversity of six duck populations.

Microsatellite Marker	Arni Ducks				Kuttanad Ducks		Assam Ducks		White Pekin Ducks		Muscovy Ducks	
	Sanyasi Ducks		Keeri Ducks									
	n_a	n_e	n_a	n_e	n_a	n_e	n_a	n_e	n_a	n_e	n_a	n_e
CAUD010	9	4.3802	11	2.9054	4	2.6074	4	1.1868	3	2.2924	4	1.6364
CAUD011	10	3.7072	9	4.6972	6	2.1804	4	3.1765	1	1.0000	6	4.5581
CAUD013	5	1.7501	3	2.1383	10	3.2294	6	3.4105	3	1.7422	7	2.7273
CAUD016	5	3.0256	5	2.5167	6	3.5393	4	2.1672	7	3.0000	6	1.9397
CAUD017	3	1.2915	3	1.4378	8	2.1135	2	1.8000	3	2.7108	11	7.1429
CAUD019	10	3.6141	11	2.9653	7	2.6759	5	1.9343	1	1.0000	2	2.0000
CAUD022	4	2.8533	4	3.0885	4	2.9467	3	2.2192	2	1.0689	4	1.5254
CAUD023	6	2.7106	3	1.7175	7	1.9715	4	2.8297	3	1.9481	4	1.5254
CAUD024	19	11.2117	15	5.6609	11	4.3801	4	2.4828	8	5.4878	6	3.2609
CAUD025	4	1.5183	4	2.8046	1	1.0000	1	1.0000	4	1.9824	1	1.0000
CAUD026	4	2.9146	3	1.5765	4	2.5324	3	2.6667	2	1.6423	6	3.3582
CAUD027	3	2.0719	3	2.0237	3	1.8412	4	2.1966	3	1.1450	5	1.9231
CAUD031	3	2.7759	3	1.8344	5	2.5507	5	3.5217	3	2.5424	3	2.4064
CAUD032	6	2.3367	8	2.4100	8	3.4175	5	2.0062	1	1.0000	5	3.6885
CAUD033	3	1.7693	4	1.2947	3	1.6046	2	1.3144	2	1.7241	2	1.2195
CAUD035	5	1.9057	5	1.9634	8	1.7270	4	2.2116	3	1.2262	7	1.6729
CAUD001	4	2.8287	4	2.2778	10	3.4602	4	2.4089	4	2.5281	7	2.9605
CAUD004	8	2.8166	6	2.3839	11	4.1996	7	3.4839	8	3.1250	6	2.5000
APH001	3	1.1112	4	1.3245	2	1.1200	2	1.1803	3	1.9231	4	2.5714
APH007	4	2.7042	4	2.5671	5	2.3481	5	2.8929	3	1.6129	5	4.1284
APH009	4	2.6498	6	3.2089	7	3.3379	7	3.1610	4	2.1429	7	4.2857
APH010	9	2.0255	5	1.4509	3	1.3814	2	1.9059	4	2.1951	5	3.0405
MCW328	5	1.7111	6	1.5663	6	1.7552	4	2.0571	3	1.2262	5	3.3088
Mean	**5.913**	**2.8558**	**5.6087**	**2.4267**	**6.0435**	**2.5183**	**3.957**	**2.3137**	**3.3913**	**2.0116**	**5.1304**	**2.7991**
S. E.	**0.76**	**0.41**	**0.66**	**0.22**	**0.59**	**0.19**	**0.32**	**0.16**	**0.40**	**0.21**	**0.44**	**0.29**

n_a – observed number of alleles , n_e- Effective number of alleles.

Table 2: Observed (H_o) and Expected (H_e) Heterozygosity values for six duck populations.

Microsatellite Marker	Arni Ducks				Kuttanad Ducks		Assam Ducks		White Pekin Ducks		Muscovy Ducks	
	Sanyasi Ducks		Keeri Ducks									
	H_o	H_e	H_o	H_e	H_o	H_e	H_o	H_e	H_o	H_e	H_o	H_e
CAUD010	0.3750	0.7717	0.4792	0.6558	0.6364	0.6165	0.1111	0.1574	0.3571	0.5638	0.2000	0.3889
CAUD011	0.6458	0.7303	0.7500	0.7871	0.1860	0.5414	0.1111	0.6852	0.0000	0.0000	0.3571	0.7806
CAUD013	0.5208	0.4286	0.7708	0.5323	0.5227	0.6903	0.5556	0.7068	0.0714	0.4260	0.6000	0.6333
CAUD016	0.8333	0.6695	0.6667	0.6026	0.5909	0.7175	0.2778	0.5386	0.4667	0.6667	0.2000	0.4844
CAUD017	0.0833	0.2257	0.1042	0.3045	0.2727	0.5269	0.3333	0.4444	0.6000	0.6311	1.0000	0.8600
CAUD019	0.7917	0.7233	0.7708	0.6628	0.4545	0.6263	0.5000	0.4830	0.0000	0.0000	0.4667	0.5000
CAUD022	0.7917	0.6495	0.7292	0.6762	0.8182	0.6606	0.7778	0.5494	0.0667	0.0644	0.1333	0.3444
CAUD023	0.4583	0.6311	0.5417	0.4178	0.5455	0.4928	0.4444	0.6466	0.7333	0.4867	0.2667	0.3444
CAUD024	0.9375	0.9108	1.0000	0.8234	0.6136	0.7717	0.9444	0.5972	0.4000	0.8178	0.9333	0.6933
CAUD025	0.3542	0.3414	0.7292	0.6434	0.0000	0.0000	0.0000	0.0000	0.3333	0.4956	0.0000	0.0000
CAUD026	0.4375	0.6569	0.0625	0.3657	0.5227	0.6051	0.8889	0.6250	0.0000	0.3911	0.4667	0.7022
CAUD027	0.6667	0.5174	0.6875	0.5059	0.5000	0.4569	0.3889	0.5448	0.1333	0.1267	0.2000	0.4800
CAUD031	0.7083	0.6398	0.5833	0.4549	0.5682	0.6080	0.8333	0.7160	0.7333	0.6067	0.7333	0.5844
CAUD032	0.3125	0.5720	0.4375	0.5851	0.3864	0.7074	0.5000	0.5015	0.0000	0.0000	0.5333	0.7289
CAUD033	0.2000	0.4348	0.2292	0.2276	0.2045	0.3768	0.0556	0.2392	0.0667	0.4200	0.0667	0.1800
CAUD035	0.3125	0.4753	0.6250	0.4907	0.2727	0.4210	0.5000	0.5478	0.0667	0.1844	0.3333	0.4022
CAUD001	0.7292	0.6465	0.4375	0.5609	0.8864	0.7110	0.6111	0.5849	0.4667	0.6044	0.2667	0.6622
CAUD004	0.5417	0.6450	0.4792	0.5805	0.6591	0.7619	0.3889	0.7130	0.5333	0.6800	0.5333	0.6000
APH001	0.1042	0.1000	0.2292	0.2450	0.1136	0.1072	0.1667	0.1528	0.0000	0.4800	0.6667	0.6111
APH007	0.3542	0.6302	0.2708	0.6105	0.2727	0.5741	0.3889	0.6543	0.1333	0.3800	0.6667	0.7578
APH009	0.6875	0.6226	0.6667	0.6884	0.4545	0.7004	0.2222	0.6836	0.3333	0.5333	0.8000	0.7667
APH010	0.6458	0.5063	0.3542	0.3108	0.2273	0.2761	0.5556	0.4753	0.6667	0.5444	0.6000	0.6711
MCW328	0.4583	0.4156	0.3958	0.3615	0.4773	0.4303	0.6667	0.5139	0.2000	0.1844	0.8000	0.6978
Mean	**0.5196**	**0.5628**	**0.5217**	**0.5258**	**0.4429**	**0.5383**	**0.4444**	**0.5113**	**0.2766**	**0.4038**	**0.4706**	**0.5597**
S. E.	**0.05**	**0.04**	**0.05**	**0.03**	**0.05**	**0.04**	**0.06**	**0.04**	**0.05**	**0.05**	**0.06**	**0.04**

et al. (2009), Alyethodi and Kumar (2010), Li *et al*. (2010) Kumar *et al*. (2011) and Hariyona *et al*. (2019) in various duck breeds. While mean heterozygosity value of more than 0.6 in Chinese and Indonesian ducks were reported by Li *et al*. (2006), Ying Su *et al*. (2007), Wu *et al*. (2008) and Ismoyovati and Purwantini (2010) in different duck populations.

Generally, a marker to be considered useful for measuring genetic variation in a population should have a heterozygosity value of 0.3 to 0.8. Hence, the markers used in this study are quite suitable for assessing the genetic diversity in duck populations as the range of heterozygosity found in this study fit well within the specified range. This indicates that genetic diversity of each breed is high and there are enough gene resources in duck populations.

Fixation indices

The F-statistics was used in testing the genetic differentiation within and between populations. The overall mean F_{IS}, F_{IT} and F_{ST} values observed in this study were 0.1377, 0.3391 and 0.2336 respectively for all six duck populations (Table 4). The within-breed inbreeding estimate on deficit of heterozygosity measured in the overall populations had a mean of 0.1377 (13.77 per cent). This value indicates moderate deficit of heterozygotes in the overall duck populations. However in a study conducted by Mukesh *et al*. (2011), the overall F_{IS} and F_{IT} values estimated among duck populations of Assam, Uttarakhand and West Bengal were comparatively lower (0.03 and 0.15). Moderately higher F_{IS} and F_{IT} values obtained in this study might be due to increased homozygosity (or deficit of heterozygotes) and less differentiation within the duck breed. On the contrary, higher F_{IS} values than those observed in the present study was reported for Moti (0.44) and lower value (-0.09) for Indian Runner duck populations by Kumar *et al*. (2011) indicating that Moti was more homozygous and inbred than Indian Runner. Further, the higher F_{IS} and F_{IT} values of 0.6477 and 0.6807 were also reported by Wu *et al*. (2008) indicating the inbred Chinese duck populations.

The F_{ST} measures the genetic differentiation among various breeds. The overall F_{ST} value of 0.2336 estimated in the present study (Table 4) indicates that 76.64 per cent of the genetic variability was caused by the differences among individuals within breeds or duck varieties and 23.36 per cent was due to differentiation among duck breeds. However, the F_{ST} value observed in this study is high indicating the substantial degree of breed differentiation

Table 3: Polymorphism Information Content (PIC) and within population inbreeding estimate (F_{IS}) for six duck populations.

Microsatellite Marker	Arni Ducks				Kuttanad Ducks		Assam Ducks		White Pekin Ducks		Muscovy Ducks	
	Sanyasi Ducks		Keeri Ducks									
	PIC	F_{IS}	PIC	F_{IS}	PIC	F_{IS}	PIC	F_{IS}	PIC	F_{IS}	PIC	F_{IS}
CAUD010	0.7698	0.5141	0.6530	0.2694	0.5566	-0.0323	0.1560	0.2941	0.4686	0.3665	0.3873	0.4857
CAUD011	0.7232	0.1156	0.7756	0.0471	0.5121	0.6563	0.6497	0.8378	0.0000	-	0.7676	0.5425
CAUD013	0.4282	-0.2152	0.4366	-0.4480	0.6775	0.2488	0.6949	0.2140	0.3609	0.8323	0.6315	0.0526
CAUD016	0.6605	-0.2447	0.5801	-0.1062	0.7074	0.1764	0.5051	0.4842	0.6659	0.3000	0.4627	0.5872
CAUD017	0.2150	0.6308	0.2797	0.6579	0.5196	0.4824	0.3457	0.2500	0.6048	0.0493	0.8580	-0.1628
CAUD019	0.7188	-0.0945	0.6600	-0.1631	0.6245	0.2742	0.4771	-0.0351	0.0000	-	0.3750	0.0667
CAUD022	0.6425	-0.2188	0.6629	-0.0783	0.6426	-0.2385	0.5259	-0.04157	0.0623	-0.0345	0.3259	0.6129
CAUD023	0.6303	0.2737	0.3665	-0.2966	0.4924	-0.1069	0.6378	0.3126	0.4856	-0.5068	0.3302	0.2258
CAUD024	0.9103	-0.0293	0.8208	-0.2145	0.7710	0.2048	0.5939	-0.5814	0.8131	0.5109	0.6805	-0.3462
CAUD025	0.3014	-0.0375	0.5830	-0.1332	0.0000	-	0.0000	-	0.4470	0.3274	0.0000	-
CAUD026	0.5996	0.3340	0.3365	0.8291	0.5389	0.1362	0.5550	-0.4222	0.3146	1.0000	0.7001	0.3354
CAUD027	0.4212	-0.2886	0.5057	-0.3591	0.4567	-0.0944	0.5418	0.2861	0.1248	-0.0526	0.4701	0.5833
CAUD031	0.5808	-0.1072	0.4261	-0.2824	0.6048	0.0654	0.6972	-0.1638	0.5934	-0.2088	0.5763	-0.2548
CAUD032	0.5512	0.4537	0.5739	0.2522	0.6853	0.4538	0.4566	0.0031	0.0000	-	0.7036	0.2683
CAUD033	0.4260	0.5400	0.2270	-0.0067	0.3640	0.4572	0.2106	0.7677	0.3318	0.8413	0.1638	0.6296
CAUD035	0.4749	0.3425	0.4499	-0.2738	0.4201	0.3521	0.5368	0.0873	0.1772	0.6386	0.3956	0.1713
CAUD001	0.5880	-0.1279	0.4844	0.2201	0.6706	-0.2466	0.5105	-0.0449	0.5369	0.2279	0.6456	0.5973
CAUD004	0.6419	0.1602	0.5796	0.1746	0.7561	0.1349	0.7109	0.4545	0.6765	0.2157	0.5924	0.1111
APH001	0.0983	-0.0412	0.2417	0.0647	0.1014	-0.0602	0.1411	-0.0909	0.4127	1.0000	0.5842	-0.0909
APH007	0.5701	0.4380	0.5406	0.5563	0.5257	0.5250	0.6366	0.4057	0.3470	0.6491	0.7358	0.1202
APH009	0.6162	-0.1042	0.6625	0.0315	0.6929	0.3510	0.6354	0.6749	0.4896	0.3750	0.7460	-0.0435
APH010	0.4773	-0.2756	0.2988	-0.1397	0.2524	0.1768	0.3624	-0.1688	0.4764	-0.2245	0.6500	0.1060
MCW328	0.4005	-0.1029	0.3579	0.0948	0.4154	-0.1092	0.4887	-0.2973	0.1772	-0.0843	0.6825	-0.1465
Mean	**0.5985**	**0.0832**	**0.5001**	**0.0302**	**0.6264**	**0.1655**	**0.4813**	**0.1466**	**0.3725**	**0.2705**	**0.5419**	**0.1935**
S. E.	**0.06**	**0.06**	**0.03**	**0.07**	**0.04**	**0.05**	**0.04**	**0.08**	**0.05**	**0.09**	**0.04**	**0.06**

Table 4: F-statistic analysis.

Locus	F_{IS}	F_{IT}	F_{ST}
CAUD010	0.3156	0.5621	0.6303
CAUD011	0.4183	0.5250	0.1834
CAUD013	0.1100	0.3432	0.2619
CAUD016	0.1750	0.3675	0.2333
CAUD017	0.2002	0.3422	0.1776
CAUD019	0.0039	0.3973	0.3950
CAUD022	-0.1264	0.2715	0.3532
CAUD023	0.0097	0.2701	0.2629
CAUD024	-0.0465	0.1212	0.1603
CAUD025	0.0430	0.3114	0.2804
CAUD026	0.2892	0.4481	0.2236
CAUD027	0.0209	0.3265	0.3121
CAUD031	-0.1524	-0.0713	0.0704
CAUD032	0.2990	0.4813	0.2600
CAUD033	0.5621	0.6685	0.2431
CAUD035	0.1631	0.5100	0.4145
CAUD001	0.0988	0.2110	0.1245
CAUD004	0.2123	0.3319	0.1519
APH001	0.2452	0.4939	0.3295
APH007	0.4215	0.4869	0.1131
APH009	0.2080	0.3250	0.1453
APH010	-0.0954	0.0642	0.1457
MCW328	-0.1516	0.0308	0.1583
Mean	**0.1377**	**0.3391**	**0.2336**

among the studied duck populations. The overall F_{ST} value estimated among duck populations of Assam, Uttarakhand and West Bengal was comparatively low (0.12; Mukesh *et al*., 2011). Higher breed differentiation value obtained in the present study is due to different indigenous duck populations selected from various regions. Lower F_{ST} values of 0.17, 0.094 and 0.184 were also reported in Chinese duck populations by Ying Su *et al*. (2007), Wu *et al*. (2008) and Su and Chen (2009) respectively.

Genetic distance and phylogenetic analysis of ducks

The dendrogram constructed using the neighbour joining procedure of PHYLIP version 3.5 (Fig 1) revealed that the six duck populations were clustered into three groups. The first group included *Keeri*, *Sanyasi*, *Kuttanad* and Assam ducks; the second group included White Pekin ducks; and the third group had Muscovy ducks. Among the Indian duck varieties clustered together in the first group, *Keeri* and *Sanyasi* ducks of Tamil Nadu were found to be closer to each other as indicated by the genetic distance value of 0.11 (11 per cent). However, within this group, 26.33 and 29.87 per cent of differentiation were noticed between Assam and *Sanyasi* and Assam and *Keeri* ducks respectively. Similarly, longer genetic distance of 0.22 and 0.26 was reported between Tamil Nadu ducks and Jharkhand and Khaki Campbell ducks by Gaur *et al.* (2010). Higher genetic

Table 5: Genetic distance among six duck populations.

Duck Variety	*Keeri*	*Sanyasi*	Assam	*Kuttanad*	Muscovy	White Pekin
Keeri	-	-	-	-	-	-
Sanyasi	0.1113	-	-	-	-	-
Assam	0.2987	0.2633	-	-	-	-
Kuttanad	0.1864	0.1240	0.1672	-	-	-
Muscovy	0.8694	0.7358	0.8056	0.6975	-	-
White Pekin	0.7521	0.6534	0.6229	0.5326	0.9270	-

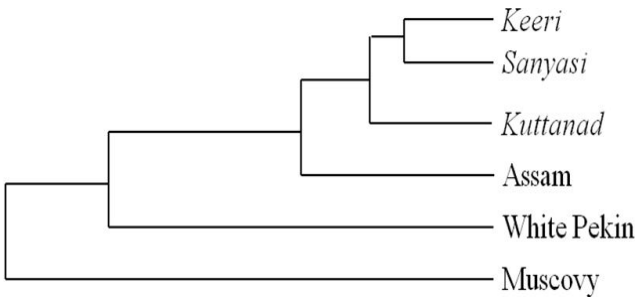

Fig 1: Phylogenetic tree of six duck populations.

distance of 0.64 was observed between Assam and Uttarakhand ducks and lesser genetic distance (0.06) between Assam and West Bengal ducks by Mukesh *et al.* (2011). The result of the cluster was consistent with the breeding history and region / environment of the six populations, since, the genetic distance goes in the ascending order from Tamil Nadu, Kerala, Assam and exotic breeds.

The result of Nei's genetic distance between six duck populations (Table 5) revealed the longest genetic distance of Muscovy and White Pekin ducks with other Indian duck varieties studied. As suggested by the dendrogram, these two duck breeds were separated out from the ducks of Indian origin. Similarly, the longest genetic distance of 0.598 and 0.4558 between Muscovy and Pekin duck of Iran (1) and China (16) respectively. The reason that could be attributed for separation of White Pekin and Muscovy from the rest of the populations was the differences in the form and biological characters that is, the White Pekin ducks are morphologically different (white plumage) and meant for meat; while Muscovy ducks belong to different genera (*Cairina moschata*) with biological distinctiveness such as higher incubation period and broodiness (Seo *et al* (2015, 2016).

CONCLUSION

All the 23 microsatellite markers studied were highly polymorphic indicating that they can be used effectively for genetic diversity and phylogenetic analyses of ducks. Genetic diversity is high among the duck populations studied which could be exploited for improving productivity. Based on the genetic diversity and genetic variation between the Indian duck populations, Arni ducks of Tamil Nadu (comprising of Sanyasi and Keeri), Kuttanad ducks of Kerala and Assam duck varieties may be classified as distinct breeds.

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