

RESEARCH ARTICLE

Genetic diversity and population structure of 10 Chinese indigenous egg-type duck breeds assessed by microsatellite polymorphism

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Abstract

The genetic structure and diversity of 10 Chinese indigenous egg-type duck breeds were investigated using 29 microsatellite markers. The total number of animals examined were 569, on average 57 animals per breed were selected. The microsatellite marker set analysed provided 177 alleles (mean 6.1 alleles per locus, ranging from 3 to 10). All populations showed high levels of heterozygosity with the lowest estimate of 0.539 for the Jinding ducks, and the highest 0.609 observed for Jingjiang partridge ducks. The global heterozygote deficit across all populations (F_{IT}) amounted to -0.363 . About 10% of the total genetic variability originated from differences among breeds, with all loci contributing significantly. An unrooted consensus tree was constructed using the NeighborNet tree based on the Reynold's genetic distance. The *structure* software was used to assess genetic clustering of these egg-type duck breeds. Clustering analysis provided an accurate representation of the current genetic relations among the breeds. An integrated analysis was undertaken to obtain information on the population dynamics in Chinese indigenous egg-type duck breeds, and to better determine the conservation priorities.

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Introduction

Genetic diversity can be observed within and between breeds or populations. However, there is a trend that high producing breeds or strains are replacing indigenous, locally adapted breeds, which subsequently decline in numbers and sometimes become extinct. This loss of genetic diversity within and among breeds is a negative trend, not only from the perspective of culture, but also with regard to utility. Traits, genotypes and alleles with possible economic interest are at risk of being lost. Further, breeds are exposed to a great loss of alleles and haplotypes as a consequence of small effective population size or, equivalently, high rates of inbreeding (Falconer and Mackay 1996). Continued loss of within-population genetic diversity also diminishes the possibility of genetic improvement of breeds in future (Eding 2001).

With its long history of animal husbandry and diversified geographical conditions, China has a wide variety of

indigenous poultry resources. There are 27 native duck breeds recorded in China (China Agriculture Press 2004), mainly distributed along the Yangtze River and in southern regions of China. Many of these local duck varieties have valuable genetic features. Liancheng white ducks in Liancheng, Fujian province, for instance, are used not only for egg production, but also as an important source of traditional Chinese medicine. However, the population sizes of some indigenous duck breeds have been rapidly decreasing. According to a report from Ministry of Agriculture, Wending black ducks, Zhongshan partridge ducks, Jianchang ducks and Sichuan partridge ducks are also facing extinction (China Agriculture Press 2004). The decrease in population sizes of indigenous ducks is mainly attributed to the introduction of exotic duck breeds and the limited conservation measures for local breeds.

Conservation efforts should be as efficient as possible, securing a maximum amount of genetic diversity in a given limited resources. The question to be answered is, which breeds we need to conserve? Decisions on which breeds to

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conserve can be based on a number of different considerations (Ruane 1999). However, the quantitative assessment of genetic diversity within and between populations is an important tool for decision making in genetic conservation plans (Weigend *et al.* 1995). In the process of developing strategies to conserve genetic diversity in domestic ducks, it is important to assess the genetic uniqueness of a given population, which may be deduced from genetic distances (Hillel *et al.* 2003). According to FAO (2004) recommendations, determination of genetic distances using neutral, highly polymorphic microsatellite markers are currently the method of choice for investigating genetic relationships and breed differentiation. This methodology also provides information for establishing preservation priorities for livestock breeds (Barker 1999).

Ducks are appreciated for their meat and eggs. There are 10 egg-type duck breeds among 27 indigenous duck breeds in China and are defined according to their utilization patterns. Egg-type ducks are mainly used for their eggs. Research on duck genetics and breeding has been developed recently (Cheng *et al.* 2003). The aim of the current study was to assess the genetic structure and diversity in 10 Chinese indigenous egg-type duck breeds with 29 microsatellite markers, and to determine their genetic relationships by different methods. The results may be useful to understand genetic differentiation of these important local breeds in China also in developing more efficient conversation strategies.

Materials and methods

Experimental populations

A total of 569 individuals originating from 10 Chinese indigenous egg-type duck breeds (*Anas platyrhynchos*) were analysed in the present study. Information about breeds, main original area of their distribution in China and number of individuals sampled are given in table 1 and figure 1. All breeds were kept at their own conservation farm or conservation zone. Individuals from each breed were sampled with the proportion of male : female equaling 1:4, according to Barker's (1994) guidelines for sample requirements of genetic diversity evaluation.

DNA isolation

From each individual, 0.4 mL of whole blood was collected from the ulnar vein with heparin as anticoagulant. Then, 4 mL of DNA lysate solution (2 M urea, 100 mM Tris-HCl (pH 8.0), 1% SDS, 100 mM EDTA) was added, and the mixture was stored at 4°C. DNA was isolated by using a phenol/chloroform based method (Sambrook and Russell 2001).

Genotyping

Microsatellites were chosen such as (i) to be well spaced across the genome, (ii) to give good typing performance on an automatic sequencer with multiplexing, and (iii) to be

Table 1. Description of the 10 indigenous Chinese egg-type duck breeds.

Breed (abbreviation)	Longitude and latitude	Main original area	Number of animals studied
Jingjiang partridge duck JJP	29°54'N; 112°42'E	Jiangling, Jianli and Mianyang countries, Hubei province	59
Enshi partridge duck ESP	30°18'N; 108°56'E	Lichuan county, Hubei province	59
Weishan partridge duck WSP	35°06'N; 117°12'E	Nanyang, Dushan, Zhaoyang and Weishan lakes in Shandong province	56
Jinding duck JD	24°18'N; 117°48'E	Longhai city, Fujian province	56
Liancheng white duck LCW	25°42'N; 116°42'E	Liancheng county, Fujian province	58
Putian black duck PTB	25°24'N; 119°08'E	Putian county, Fujian province	58
Shan partridge duck SP	25°06'N; 117°01'E	Longyanhu town, Fujian province	60
Sansui duck SS	25°6'N; 104°48'E	Sansui county, Guizhou province	56
Youxian partridge duck YXP	26°72'N; 113°27'E	Youxian county, Hunan province	57
Shaoxing duck SX	30°12'N; 120°12'E	Shaoxing, Xiaoshan and Zhuji counties, Zhejiang province	50

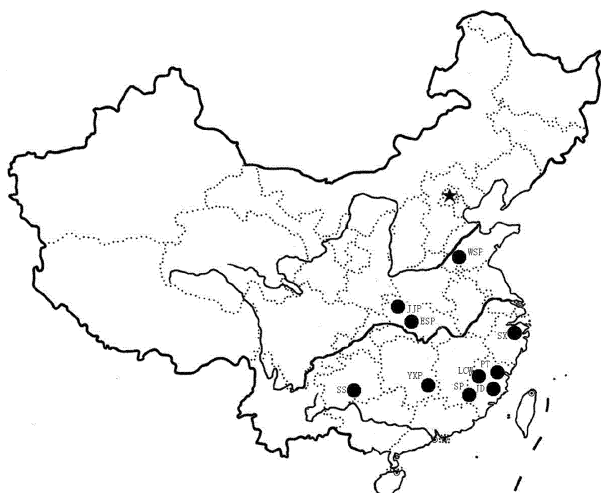


Figure 1. The geographic location of each of the 10 egg-type duck breeds in China.

polymorphic in the duck populations. Thus, 29 primers that produced clear and reproducible bands were selected from 35 primers (from GenBank) (table 2) (Maak *et al.* 2000; Paulus and Tiedemann 2003; Denk *et al.* 2004). For the entire sample, amplifications and analyses were performed in the same laboratory, the PCR products were labelled with the fluorescent dyes and genotyped using a capillary sequencer ABI PRISM 3100 Genetic Analyzer (Applied Biosystems, Foster City, USA).

Statistical analysis

Genetic diversity: Total number of alleles, allele frequencies, average number of alleles per locus, observed (H_o) and expected heterozygosity (H_e) for each locus across populations and for each population across the loci, were estimated with microsatellite-toolkit for Excel (Park 2001). Polymorphism information content (PIC) for each locus was obtained according to Botstein *et al.* (1980):

Table 2. The observed number of alleles (N_a), effective number of alleles (N_e), observed heterozygosity (H_o), expected heterozygosity (H_e), PIC values, F -statistics, for each of the 29 microsatellite markers in 10 Chinese egg-type duck breeds.

Locus	N_a	N_e	H_o	H_e	PIC	$F_{IT} = F$	$F_{ST} = \theta$	$F_{IS} = f$
APH01	4	2.07	0.949	0.517	0.401	-0.837	0.003***	-0.843
APH07	7	1.17	0.120	0.144	0.142	0.175	0.047***	0.135
APH09	7	5.62	0.936	0.823	0.798	-0.132	0.052***	-0.195
APH10	6	3.65	0.989	0.726	0.682	-0.345	0.134***	-0.552
APH11	3	2.23	0.954	0.551	0.448	-0.715	0.104***	-0.915
APH14	3	2.47	0.931	0.596	0.523	-0.554	0.059***	-0.652
APL2	7	5.15	0.979	0.807	0.779	-0.210	0.030***	-0.247
APL11	6	4.84	0.948	0.794	0.765	-0.188	0.058***	-0.261
APL12	8	3.85	0.993	0.741	0.704	-0.335	0.037***	-0.386
APL23	9	6.05	0.909	0.835	0.815	-0.08	0.075***	-0.167
APL26	5	3.89	0.983	0.743	0.701	-0.32	0.029***	-0.359
APL36	8	3.03	0.995	0.671	0.612	-0.481	0.021***	-0.512
APL77	5	2.65	0.989	0.624	0.549	-0.562	0.161***	-0.861
APL78	5	2.51	0.998	0.601	0.519	-0.66	0.004**	-0.667
APL79	7	1.33	0.240	0.248	0.240	0.041	0.079***	-0.041
APL80	10	5.75	0.982	0.827	0.803	-0.182	0.049***	-0.244
APL81	5	2.71	0.998	0.632	0.563	-0.567	0.089***	-0.719
APL82	3	2.32	0.935	0.569	0.473	-0.635	0.062***	-0.743
APL83	6	3.38	0.948	0.705	0.654	-0.308	0.283***	-0.825
CMO11	10	1.78	0.180	0.438	0.423	0.606***	0.399***	0.344***
CMO12	10	7.46	0.943	0.867	0.852	-0.078	0.094***	-0.19
SMO4	6	2.10	0.699	0.524	0.440	-0.331	0.023***	-0.362
SMO6	8	5.58	0.981	0.821	0.796	-0.16	0.29***	-0.634
SMO7	4	2.43	0.998	0.589	0.501	-0.681	0.098***	-0.863
SMO8	8	4.27	0.998	0.767	0.733	-0.295	0.059***	-0.375
SMO9	4	1.89	0.649	0.473	0.39	-0.366	0.048***	-0.436
SMO11	5	2.12	0.996	0.529	0.417	-0.885	0.002**	-0.888
SMO12	4	2.10	0.819	0.523	0.424	-0.561	0.035***	-0.617
SMO13	4	3.39	0.963	0.706	0.652	-0.322	0.317***	-0.936
Mean	6.1	3.37	0.862	0.634	0.579	-0.363	0.095***	-0.506
S.d.	2.1	1.61	0.251	0.173	0.183	0.050	0.019	0.053

** $P < 0.01$; *** $P < 0.001$.

$$\text{PIC} = 1 - \sum_{i=1}^n p_i^2 - 2 \sum_{i=1}^{n-1} \sum_{j=i+1}^n p_i^2 p_j^2,$$

where n , the number of alleles; p_i , frequency of the allele i ; p_j , frequency of the allele j .

Genetic differentiation: Population differentiation was estimated by Wright's (1978) fixation indices F_{IT} , F_{ST} and F_{IS} in the form of F , θ , and f , respectively, for each locus across populations according to the variance-based method of Weir and Cockerham (1984) using FSTAT software version 2.9.3 (Goudet 2002). Allelic richness was also computed using FSTAT. The significance of the F -statistics was determined by permutation tests with the sequential Bonferroni procedure applied over loci (Hochberg 1988). The extent of inbreeding was further studied with Genepop software (Raymond and Rousset 1995) by estimating the F_{IS} values and their significance level within each of the populations.

Pair-wise F_{ST} values were computed for all combinations of 10 populations using GENEPOP program. Gene flow among populations, defined as the number of reproductively successful migrants per generation (Nm), was estimated based on the island model of population structure (Slatkin and Barton 1989). The estimate was based on the relationship $F_{ST} = 1/(4Nm + 1)$, where N is the effective population size, m is the migration rate, and F_{ST} is calculated as mean over loci.

Clustering of breeds: The program *structure* (Pritchard et al. 2000) which implements a model-based clustering method for inferring population structure using multilocus genotypes was utilized. This program uses a Monte Carlo Markov chain (MCMC) algorithm to assess the presence of a structure underlying the genetic information provided by the genetic markers. We ran the program *structure* 100 times with 50,000 iterations, after a burn-in period of 20,000 iterations, for each number of genetic clusters (K) chosen a priori. Pairwise similarities (G) between runs were computed using *clumpp* (Jakobsson and Rosenberg 2007). Solutions with over 95% similarity were considered as identical. The most frequent solution for each K was taken as the most probable clustering and visualized using *distrupt* software (Rosenberg 2007).

The matrix of Reynold's unweighted distances D_R (Reynolds et al. 1983) was computed using POPULATION (Olivier Langella; <http://www.pge.cnrs.fr/bioinfo/populations/>). Regarding the D_R distance, a NeighbourNet tree was drawn using SPLITSTREE 4.8 (Hudson and Bryant 2006).

Result

Genetic diversity within and among 10 egg-type duck breeds

A total of 177 alleles were observed in 10 Chinese indigenous egg-type duck breeds. All microsatellite loci typed were polymorphic, the average PIC value was 0.634 (table 2).

The number of alleles per locus ranged from three (*APH11*, *APH14* and *APL82*) to 10 (*APL80*, *CMO11* and *CMO12*), and the average number of alleles observed was 6.1. The observed and expected heterozygosities for each marker are also given. Across the 10 egg-type duck breeds, locus *APH07* had the lowest expected heterozygosity (H_e) estimate (0.144), and locus *CMO12* showed the highest (0.867).

Genetic differentiation was examined by fixation indices F_{IT} , F_{ST} , F_{IS} for each locus across all populations (table 2). The fixation coefficients of subpopulations within the total population, measured as F_{ST} value, for the 29 loci varied from 0.002 (*SMO11*) to 0.399 (*CMO11*), with a mean of 0.095 ($P < 0.001$). All loci contributed significantly to this differentiation. The global deficit of heterozygotes across populations (F_{IT}) amounted to -0.363 . Mean F_{IS} was found to be -0.506 within populations, at which two loci showed deficit of heterozygotes, while 27 markers, to some extent, showed excess of heterozygotes, with a negative F_{IS} value calculated (table 2).

Average number of alleles per locus ranged from 3.57 in Jinding duck breed to 4.40 in Jingjiang partridge duck breed. All 10 Chinese egg-type duck breeds showed a relatively large expected heterozygosity with a mean of 0.578. The lowest estimate (0.539) was obtained for Jinding breed, while the highest (0.609) was found in Jingjiang partridge breed (table 3).

A breakdown of inbreeding estimates (F_{IS}) of populations and their statistical significance over loci are given in table 3. All duck breeds showed significant excess of heterozygous genotypes with respect to the expected value ($P < 0.001$).

Genetic distances and clustering of breeds

Estimates of gene flow (Nm) and Reynold's genetic distance (D_R) between each population pair are given in table 4. Reynold's distance values varied between 0.045 (Putian black-Youxian partridge) and 0.174 (Jingjiang partridge-Liancheng white). The Nm value ranged from 1.313 (between Jingjiang partridge-Liancheng white duck) to 5.406 (between Putian black-Youxian partridge duck). Most Nm values between pairs of breeds were above 2.0.

The results of the *structure* clustering are displayed in figure 2. At $K = 2$, two main groups were formed. Jingjiang partridge, Enshi partridge, Jinding, Sansui, and Shan partridge clustered together, while Putian black, Youxian partridge, Weishan partridge, Shaoxing, and Liancheng white clustered together. At $K = 3$, the most frequent solution showed Jingjiang partridge duck and Enshi partridge duck split from others to form a separate cluster. At $K = 4$, Liancheng white ducks made up their own separate cluster. At $K = 5$, Shaoxing ducks formed a separate cluster. From $K = 6$ to 9, Shan partridge duck formed a separate cluster first, and then did Weishan partridge duck, and then Sansui duck separated from the remaining. Jingjiang partridge

Table 3. Allelic richness (AR), mean number of alleles per locus, mean estimates of expected (H_e) and observed (H_o) heterozygosity and F_{IS} estimates per population.

Breed	AR	No. of mean	H_o	H_e	F_{IS}
		alleles per locus (Mean \pm s.d.)			
JJP	4.31	4.40 \pm 2.13	0.880 \pm 0.008	0.609 \pm 0.030	-0.451***
ESP	3.97	4.10 \pm 2.40	0.865 \pm 0.008	0.586 \pm 0.039	-0.481***
WSP	4.05	4.23 \pm 2.08	0.889 \pm 0.008	0.592 \pm 0.027	-0.509***
JD	3.45	3.57 \pm 1.59	0.855 \pm 0.009	0.539 \pm 0.034	-0.595***
LCW	3.77	3.90 \pm 1.67	0.858 \pm 0.008	0.563 \pm 0.027	-0.530***
PTB	3.86	4.00 \pm 2.02	0.863 \pm 0.008	0.563 \pm 0.033	-0.539***
SP	4.00	4.13 \pm 1.85	0.851 \pm 0.008	0.584 \pm 0.037	-0.463***
SS	3.99	4.10 \pm 2.06	0.886 \pm 0.008	0.588 \pm 0.031	-0.514***
YXP	4.05	4.20 \pm 1.99	0.863 \pm 0.008	0.585 \pm 0.035	-0.482***
SX	3.87	3.93 \pm 2.20	0.857 \pm 0.009	0.569 \pm 0.038	-0.515***

*** $P < 0.001$ **Table 4.** Reynold's genetic distances and the gene flow, Nm between breeds. Numbers in bold are highest and lowest values of D_R and Nm . The data in upper and lower diagonal are gene flow, Nm and Reynold's genetic distances between breeds, respectively.

Breed	JJP	ESP	WSP	JD	LCW	PTB	SP	SS	YXP	SX
JJP	–	2.338	2.620	2.050	1.313	1.429	2.017	1.970	1.771	1.887
ESP	0.102	–	2.537	2.270	1.424	1.563	2.044	2.371	1.992	1.869
WSP	0.091	0.094	–	4.633	2.766	3.493	3.526	3.057	4.075	2.447
JD	0.115	0.104	0.052	–	2.050	2.459	4.295	3.917	3.161	2.473
LCW	0.174	0.161	0.087	0.115	–	2.459	1.846	2.766	2.245	1.554
PTB	0.161	0.149	0.069	0.097	0.097	–	2.191	2.438	5.406	2.401
SP	0.117	0.115	0.068	0.057	0.127	0.108	–	3.782	2.597	2.485
SS	0.120	0.100	0.079	0.062	0.087	0.098	0.064	–	2.453	1.982
YXP	0.132	0.119	0.060	0.076	0.105	0.045	0.092	0.098	–	4.700
SX	0.124	0.126	0.098	0.097	0.150	0.099	0.095	0.119	0.052	–

duck and Enshi partridge duck did not separate until $K = 10$. Youxian partridge duck always showed a mixed population.

The NeighbourNet tree derived from the Reynold's genetic distance is given in figure 3. The clustering results are in accordance with the results obtained from *structure* in general. Weishan partridge duck clustered together with Jingjiang partridge duck and Enshi partridge duck.

Discussion

The average expected heterozygosity within populations exceeded the value reported in the 24 Chinese native duck breeds (Li *et al.* 2006), but was lower than the values estimated for six endangered local duck populations in China (Su *et al.* 2007). The number of alleles observed in these 10 Chinese native egg-type populations (6.1) were greater than that observed in eight Chinese native concern duck breeds using 28 microsatellite markers (Tang *et al.* 2007), but lower than that observed in five Fujian native duck breeds in China using 32 microsatellite markers (Xiao *et al.* 2009).

On average, the genetic differentiation index, F_{ST} , among breeds was 0.095 (table 2). About 10% of the total genetic

variation corresponds to differences between breeds and the remaining 90% was the result of variation among individuals within breeds. All loci contributed to this differentiation significantly. This level of differentiation value is very similar to the values reported among 78 Chinese indigenous chicken breeds ($F_{ST} = 0.106$; Qu *et al.* 2006), but higher than that reported, in 95 red-winged blackbirds using 10 microsatellites ($F_{ST} = 0.009$; Williams *et al.* 2004), African cattle breeds ($F_{ST} = 0.060$; Ibeagha-Awemu and Erhardt 2005), and human populations ($F_{ST} = 0.054$; Rosenberg *et al.* 2002).

The overall F_{IS} value (-0.506), estimated at the marker level (table 2), was lower than zero. Twenty-seven loci contributed to this result. One microsatellite *CMO11* showed significant deficit of heterozygotes, a possible explanation for this observation might be genetic drift or that this locus is linked to loci affecting morphological, productive or adaptive traits of selective interest and have undergone selection (Ibeagha-Awemu and Erhardt 2005). All breeds showed negative F_{IS} values. The avoidance of mating among closely related animals might be one reason why significant excess of heterozygotes was found in these populations.

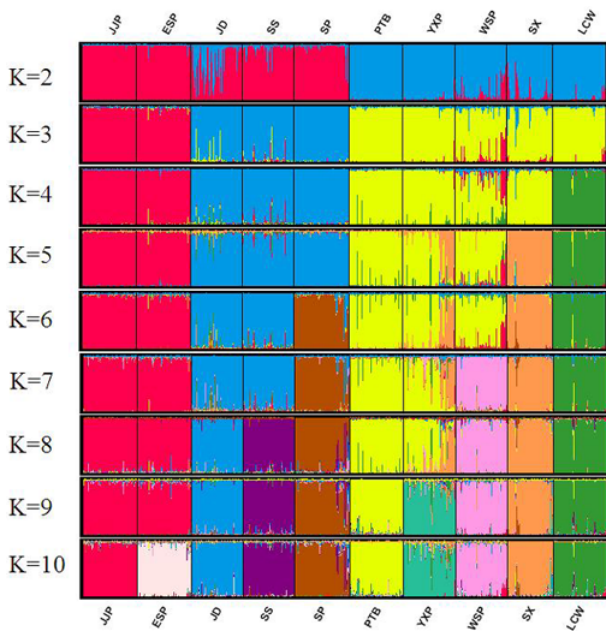


Figure 2. Clustering diagrams of 10 egg-type duck breeds obtained from $K = 2$ to $K = 10$ using Q matrices of runs with best similarities.

Clustering results were somewhat consistent with the geographical locations. Jingjiang partridge and Enshi partridge duck clustered together in the NeighborNet tree, indicating a close genetic relationship. These two breeds are distributed in Hubei province, thus raising the possibility of interbreeding. During the *structure* runs, they could not be distinguished until the number of clusters, K , equalled the number of breeds. Thus, these two populations can be considered as genetically similar (Rosenberg *et al.* 2001). Two high production breeds

Jinding and Shan partridge, from Fujian province also clustered together, interbreeding may occur in these two breeds. This may explain the relative high gene flow ($Nm = 4.295$) and low Reynold's genetic distance ($D_R = 0.057$), between Jinding and Shan partridge duck.

In general, the model-based clustering analysis of the breeds concurred with the relationships determined by genetic distance. Similar results were also obtained from genetic structure analysis and phylogenetic relationships in other studies (Liu *et al.* 2003; Ibeagha-Awemu and Erhardt 2005). Rosenberg *et al.* (2001) using model-based clustering method and 27 microsatellite markers achieved a 98% success rate of correctly assigning individuals from 20 distinct chicken breeds to their correct populations. Therefore, cluster analysis can resolve effectively the genetic similarity of a group of highly diverged breeds and has great potential to help identifying individuals with different or similar multi-locus genotypes (Ibeagha-Awemu and Erhardt 2005). In our study, the *structure* program clustered the analysed populations well, and suggested that the Youxian partridge duck breed is a mixed population. Such information can not be obtained from the methods based on genetic distance.

Youxian partridge egg-type duck breed appeared as a mixed breed (figure 2). The gene flow between Youxian partridge and other duck breeds range from 1.771 to 5.406, with seven of them over 2.0. This, however, may be due to lack of management during breed development. As Liancheng white ducks are not only used for egg production, but are also as an important source of traditional Chinese medicine, selection measures may have been applied in this breed. This may be the reason why Liancheng white duck split from the other breeds at the early K value in *structure* and resulted in the different clustering result between the *structure* and NeighborNet tree based on Reynold's genetic distance.

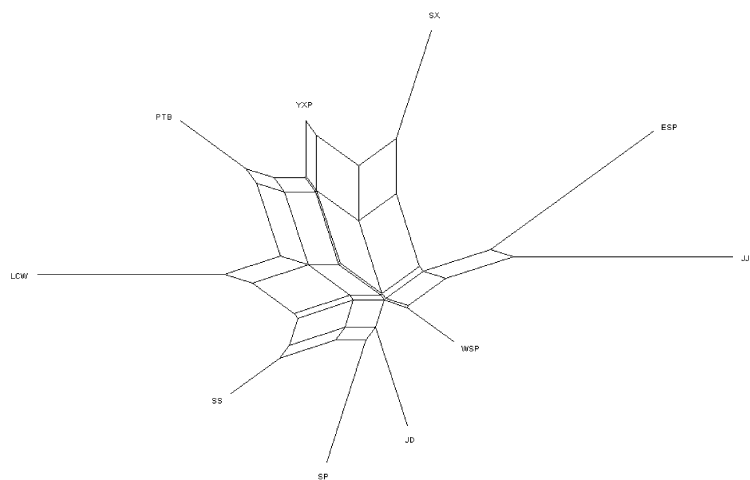


Figure 3. NeighbourNet tree for the 10 egg-type duck breeds using the Reynold's genetic distance

Based on the various genetic diversity measures used in this study, a high genetic diversity was observed in the Chinese indigenous egg-type duck breeds. The considerably rich genetic diversity of egg-type duck breeds in China can be attributed to its complicated local geographical conditions where different farming practices and agro-ecosystems exist. The diversity may also be significantly associated with its rich culture diversity that promotes miscellaneous needs and applications of duck breeds. The genetic diversity information, evaluated by integrating within and between population analyses may allow conservation priorities to be better established. For example, Jinding duck has a lower genetic diversity and a higher inbreeding, so a better conservation should be made for this breed to avoid inbreeding depression and genetic drift. However, it should be noted that Chinese native chicken breeds also have more morphological diversity. If setting conservation priorities based exclusively on the diversity of molecular markers might lead to the loss of locally adapted populations (Mckay *et al.* 2001), so additional phenotypic performance and population history should be considered jointly to provide more reliable guidelines in choosing populations for practical and conservation purposes in the future.

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References

- Barker J. S. F. 1994 A global protocol for determining genetic distance among domestic livestock breeds. In Proceedings of 5th world congress on genetic application of livestock production. Guelph, Canada. **21**, 501–508.
- Barker J. S. F. 1999 Conservation of livestock breeds diversity. *Anim. Genet. Res. Inf.* **25**, 33–43.
- Botstein D., White R. L., Skolnick M. and Davis R. W. 1980 Construction of a genetic linkage map in man using restriction fragment length polymorphisms. *Am. J. Hum. Genet.* **32**, 314–331.
- Cheng Y. S., Rouvier R., Hu Y. H., Tai J. J. L. and Tai C. 2003 Breeding and genetics of waterfowl. *World's Poult. Sci. J.* **59**, 511–521.
- China Agriculture Press 2004 Country report for the preparation of SoW-AnGR Report on domestic animal genetic resources in China. China Agriculture Press, Beijing, P. R. China.
- Denk G. A., Gautschi B., Carter K. and Kempnaers B. 2004 Seven polymorphic microsatellite loci for paternity assessment in the mallard (*Anas platyrhynchos*). *Mol. Ecol.* **4**, 506–508.
- Eding H. 2001 Conservation of genetic diversity: assessing genetic variation using marker estimated kinships. Ph.D. thesis, Animal Breeding and Genetics Group, Department of Animal Sciences, Wageningen University, Wageningen, The Netherlands.
- Falconer D. S. and Mackay T. F. C. 1996 *Introduction to quantitative genetics*. Longman House, Harlow, UK.
- FAO 2004 Guidelines for development of national management of farm animal genetic resources plans. <http://dad.fao.org/en/refer/library/guidelin/marker.pdf>.
- Goudet J. 2002 *FSTAT* version 2.9.3.2. Department of Ecology and Evolution, University of Lausanne, Lausanne, Switzerland.
- Hillel J., Groenen A. M. M., Tixier-Boichard A. B., Korol L., David V. M. and Kirzhner T. 2003 Biodiversity of 52 chicken populations assessed by microsatellite typing of DNA pools. *Genet. Sel. Evol.* **35**, 533–557.
- Hochberg Y. 1988 A sharper Bonferroni procedure for multiple tests of significance. *Biometrika* **75**, 800–802.
- Hudson D. H. and Bryant D. 2006 Application of phylogenetic networks in evolutionary studies. *Mol. Biol. Evol.* **23**, 254–267.
- Ibeagha-Awemu E. M. and Erhardt G. 2005 Genetic structure and differentiation of 12 African *Bos indicus* and *Bos taurus cattle* breeds, inferred from protein and microsatellite polymorphisms. *J. Anim. Breed. Genet.* **122**, 12–20.
- Jakobsson M. and Rosenberg N. A. 2007 CLUMPP: a cluster matching and permutation program for dealing with label switching and multimodality in analysis of population structure. *Bioinformatics* **23**, 1801–1806.
- Li H. F., Li B. C., Chen K. W., Yang N., Ma Y. H., Tang Q. P. and Tu Y. J. 2006 Study on molecular genetic diversity of native duck breeds in China. *Acta Vet. Zootech. Sin.* **37**, 1107–1113.
- Liu K., Goodman M., Muse S., Smith J. S., Buckler E. and Doebley J. 2003 Genetic structure and diversity among maize inbred lines as inferred from DNA microsatellites. *Genetics* **165**, 2117–2128.
- Maak S., Neumann K. and von Lengerken G. 2000 First seven microsatellites developed for the Peking duck (*Anas platyrhynchos*). *Anim. Genet.* **31**, 233.
- Mckay J. K., Bishop J. G., Lin J., Richards J. H. and Sala A. 2001 Local adaptation across a climatic gradient despite small effective population size in the rare sapphire rockcross. *Proc. R. Soc. Lond. B* **268**, 1715–1721.
- Park S. D. E. 2001 *The Excel microsatellite toolkit*, version 3.1. Animal Genomics Laboratory, University College Dublin, Ireland. (<http://animalgenomics.ucd.ie/sdepark/ms-toolkit/>).
- Paulus B. K. and Tiedemann R. 2003 Ten polymorphic autosomal microsatellite loci for the Eider duck *Somateria mollissima* and their cross-species applicability among waterfowl species (Anatidae). *Mol. Ecol.* **3**, 250–252.
- Pritchard J. K., Stephens M. and Donnelly P. 2000 Inference of population structure using multilocus genotype data. *Genetics* **155**, 945–959.
- Qu L. J., Li X. Y., Xu G. F., Chen K. W., Yang H. J., Zhang L. C. *et al.* 2006 Evaluation of genetic diversity in Chinese indigenous chicken breeds using microsatellite markers. *Sci. Chin. C Life Sci.* **49**, 332–341.
- Raymond M. and Rousset F. 1995 GENEPOP (version 1.2): population genetics software for exact test and ecumenicism. *J. Hered.* **86**, 248–249.
- Reynolds J., Weir B. S. and Cockerham C. C. 1983 Estimation of the coancestry coefficient: basis for a short-term genetic distance. *Genetics* **105**, 767–779.
- Rosenberg N. A. 2007 *Distrupt: a program for the graphical display of population structure*. (<http://rosenberglab.bioinformatics.med.umich.edu/distrupt.html>).
- Rosenberg N. A., Burke T., Elo K., Feldman M. W., Freidlin P. J., Groenen M. A. M. *et al.* 2001 Empirical evaluation of genetic clustering methods using multilocus genotypes from 20 chicken breeds. *Genetics* **159**, 699–713.
- Rosenberg N. A., Pritchard J. K., Weber J. L., Cann H. M., Kidd K. K., Zhivotovsky L. A. and Feldman M. W. 2002 Genetic structure of human populations. *Science* **298**, 2981–2985.
- Ruane J. 1999 A critical review of the value of genetic distances studies in conservation of animal genetic resources. *J. Anim. Breed. Genet.* **116**, 317–323.
- Sambrook J. and Russell D. W. 2001 *Molecular cloning: a laboratory manual*, 3rd edition. Cold Spring Harbor Laboratory, New York, USA.

- Slatkin M. and Barton N. H. 1989 A comparison of three indirect methods of estimating average levels of gene flow. *Evolution* **43**, 1349–1368.
- Su Y., Long R. J., Chen G. H., Wu X. S., Xie K. Z. and Wan J. H. 2007 Genetic analysis of six endangered local duck populations in China based on microsatellite markers. *J. Genet. Genomics* **34**, 1010–1018.
- Tang Q. P., Li H. F., Tu Y. J. and Chen K. W. 2007 Analysis of genetic diversity of the domestic concern duck breeds in China. *Journal of Northwest A & F University (Nat Sci Ed)*. **35**, 47–52.
- Weigend S., Vef E., Wesch G., Meckenstock E., Seibold R. and El-lendorff F. 1995 Conception for conserving genetic resources in poultry in Germany. *Arch. Geflügelkunde* **59**, 327–334.
- Weir B. S. and Cockerham C. C. 1984 Estimation of F-statistics for the analysis of population structure. *Evolution* **38**, 1358–1370.
- Williams L. C., Homan H. J., Johnston J. J. and Linz G. M. 2004 Microsatellite variation in red-winged blackbirds (*Agelaius phoeniceus*). *Biochem. Genet.* **42**, 35–41.
- Wright S. 1978 *Evolution and the genetics of populations*, vol. 4. *Variability within and among natural populations*. University of Chicago Press, Chicago, USA.
- Xiao T. F., Ke L. Y., Zhang L. and Jiang X. B. 2009 Genetic diversity of duck breeds: a study with microsatellite markers. *Chin. J. Appl. Ecol.* **20**, 190–196.

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