Genetic Differentiation between Sheep and Goats Based on Microsatellite DNA

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ABSTRACT : The 7 sheep microsatellite markersOarFCB48. OarAE101, MAF33. OarFCB11, MAF70. OarFCB304 and OarFCB128, which were located on chromosomes 2, 4, 6, 9, 17 and 19, were selected to PCR in Hu sheep. Tong sheep and their closely related species, the goat. They were studied with the amplifying result of 7 microsatellite sites of Hu Sheep, Tong Sheep and goats, the data of allele number and range of allele's size of amplifying were analyzed with ANOVA. The results showed that there were no significant differences (p < 0.05) in microsatellite DNA sites among 3 populations. Concerning the conservation of microsatellites in closely related species, selecting microsatellite sites located on the chromosome where the Robertsonian fusion was caused between sheep and goat, may be used in research into genetic differentiation and evolutionary relationships between sheep and goats. (Asian-Aust. J. Anim. Sci. 2004. Vol 17, No. 5 : 583-587)

Key Words : Sheep, Goat, Microsatellite DNA, Genetic Differentiation

INTRODUCTION

The karyotype of sheep and goats is 2n=54 and 60. respectively. They belong to the genera Ovis and Capra of the familyCaprinae. Archaeological and morphological research indicates that the sheep and the goat originated from the same ancestor: Rupicaprids. goat-antelopes in the Pleistocene era. Cellular genetic research showed that the sheep and the goat were evolved from a common ancestor: Rupicaprids and the karyotype of the goat is similar to the ancestral form (Li et al., 2000). It can be seen that the two species have a close relationship, but it is still necessary to study their genetic differentiation using on modern molecular technology. There are some reports about genetic differentiation between the sheep and the goat (Upholt et al., 1977; Li et al., 2000), but there are no reports on genetic differentiation of the two species based on microsatellite DNA. In the last 10 years, research on polymorphic markermicrosatellite DNA markers has been greatly advanced because of new techniques, especially PCR. The usefulness of microsatellites for the analysis of genetic relationships among closely related populations has been documented by numerous studies (Buchanan et al., 1994; Bancroft et al., 1995: Arranz et al., 1998; Joseph et al., 1999).The first genetic linkage map of the sheep genome was published in 1995 (Crawford et al., 1995), the second genetic linkage map of the sheep genome was published by de Gortari in 1998 and the first genetic linkage map of the goat genome was published by Vaiman in 1996 (Wu. 1999). This paper is

concerned with Hu sheep, Tong sheep and their closely related speciesthe goat. We discuss the probability of studying the genetic differentiation between sheep and goats based on 7 sheep microsatellites, so as to provide a basis for the data bank of sheep (goat) microsatellites, and also put forward theoretical grounds for genetic differentiation among closely related species using microsatellite DNA.

MATERIALS AND METHODS

Materials and sampling

The Hu and Tong sheep studied were from Lianshi Town of Huzhou city in Zhejiang province and Baishui country in Shannxi province of China, respectively. The sample size was 63 and 65 respectively. Blood sampling was performed by the "Random sampling in typical colonies of central area" method and we tried to avoid sampling two (or more) individuals that had traceable genetic relationships. Some external morphological characteristics were also investigated (Sun et al., 2002). At the same time, 49 Yangtse River Delta White Goats were sampled by the same method as the contrast population from the suburb of Yangzhou city in Jiangsu province of China.

Microsatellites, PCR conditions and fragment analysis

The genomic DNA was separated according to procedures described by Sun (2002). The 7 sheep microsatellites studied and their characteristics are shown in Table 1. Each 20 μ l reaction contained: 0.4 μ l dNTP (10 m mol/l). 2 μ l 10×buffer. 25 m mol/l MgCl₂(shown in Table 1). 1 μ l GT and CA primer (8 μ mol/ μ l) (2 μ l for OarFCB11), 0.2 μ l of Taq polymerase (5 U/ μ l) and 2 μ l template DNA (50 ng/ μ l). then super purified water was added. After an initial denaturation at 94°C for 5 min. the

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Received November 14, 2002: Accepted February 19, 2004

Table 1. Microsatellite primer sequences, chromosome assignment and part of PCR conditions of the microsatellites used in this study

Site Chromosome		Drimer vention tes	Anneal	$MgCl_2$
	assignment	Timer sequences	temp. (°C)	amount (µl)
OarFCB 11	2	(CAstrand): GGCCTGAACTCACAAGTTGATATATCTATCAC	63	1.8
		(GT strand): GCAAGCAGGTTCTTTACCACTAGCACC		
OarFCB 128	2	(CA strand): CAGCTGAGCAACTAAGACATACATGGCG	60	1.0
		(GT strand): ATTAAAGCATCTTCTCTTTATTTCCTCGC		
OarFCB 304	19	(CA strand): CCCTAGGAGCTTTCAATAAAGAATCGG	61	1.5
		(GT strand): CGCTGCTGTCAACTGGGTCAGGG		
OarFCB 48	17	(CA strand): GAGTTATGTACAAGGATGACAAGAGGCAC	53	1.6
		(GT strand): GACTCTAGAGGATCGCAAAGAACCAG		
MAF 70	4	(CA strand): GCAGGACTCTACGGGGGCCTTTGC	63.5	1.0
		(GT strand): CACGGAGTCACAAAGAGTCAGACC		
MAF 33	9	(CA strand): GATCATCTGAGTGTGAGTATATACAG	58	1.5
		(GT strand): GACTTTGTTTCAATCTATTCCAATTTC		
OarAE 101	6	(CA strand): TAAGAAATATATTTGAAAAAACTGTATCTCCC	57	1.0
		(GT strand): TCCTTATAGATGCACTCAAGCTAGG		

Table 2 (i). Estimates of gene frequencies of the microsatellite DNA sites

	OarF0	CB 48		OarAE 101				MAF 33			
Allele	Hu	Tong	Goat	Allele	Hu	Tong	Goat	Allele	Hu	Tong	Goat
127	0.0000	0.0122	0.0000	75	0.0000	0.0000	0.0526	110	0.0000	0.0000	0.1200
141	0.0125	0.0122	0.0000	77	0.0000	0.0000	0.0263	112	0.0000	0.0000	0.4800
145	0.0125	0.0122	0.0000	79	0.0000	0.0000	0.0263	114	0.0000	0.0000	0.0800
147	0.0250	0.0366	0.0000	85	0.0000	0.0172	0.0000	116	0.0250	0.0000	0.1600
149	0.1125	0.0976	0.0000	87	0.0000	0.0000	0.0263	120	0.0125	0.0125	0.0000
151	0.0750	0.0732	0.0000	93	0.0588	0.0517	0.0263	122	0.0125	0,0000	0.0600
153	0.0375	0.0488	0.0227	95	0.1176	0.0690	0.0263	124	0.0250	0.0750	0.0600
155	0.0375	0.0976	0.0681	97	0.2059	0.1724	0.0263	126	0.1500	0.1500	0.0400
157	0.0500	0.0610	0.0909	99	0.1765	0.1034	0.0000	128	0.0500	0.0875	0.0000
159	0.0375	0.0366	0.1136	101	0.0588	0.0690	0.0000	130	0.0125	0.0250	0.0000
161	0.0250	0.0366	0.0227	103	0.1176	0.0862	0.0789	132	0.0500	0.0500	0.0000
163	0.0500	0.0244	0.0909	105	0.1029	0.0862	0.0526	134	0.0250	0.0500	0.0000
165	0.0125	0.1098	0.0227	107	0.1176	0.1207	0.1052	136	0.1750	0.0750	0.0000
167	0.0875	0.0122	0.0455	109	0.0000	0.0517	0.1052	138	0.1250	0.1625	0.0000
169	0.1375	0.0732	0.0455	111	0.0294	0.0517	0.0526	140	0.0750	0.1125	0.0000
171	0.0500	0.0610	0.1136	113	0.0147	0.0690	0.0789	142	0.0750	0.0500	0.0000
173	0.1000	0.0854	0.0000	115	0.0000	0.0344	0.0263	144	0.0000	0.0125	0.0000
175	0.0125	0.0732	0.1136	117	0.0000	0.0000	0.0526	146	0.0375	0.0000	0.0000
177	0.0375	0.0122	0.0227	119	0.0000	0.0172	0.0789	148	0.0000	0.0250	0.0000
179	0.0125	0.0244	0.0909	121	0.0000	0.0000	0.0263	150	0.0500	0.0500	0.0000
181	0.0250	0.0122	0.0227	123	0.0000	0.0000	0.0526	152	0.0500	0.0375	0.0000
185	0.0000	0.0000	0.0227	125	0.0000	0.0000	0.0263	154	0.0125	0.0250	0.0000
187	0.0125	0.0000	0.0681	127	0.0000	0.0000	0.0263	156	0.0125	0.0000	0.0000
189	0.0125	0.0000	0.0227	131	0.0000	0.0000	0.0263	158	0.0125	0.0000	0.0000
197	0.0250	0.0000	0.0000					160	0.0125	0.0000	0,0000

PCR was performed with 30 cycles: denaturation at 94°C for 1 min. anneal at temperatures in Table 1 for 1 min. extension at 72°C for 1 min; the final cycle was followed by an extension at 72°C for 5 min. Amplified fragments were analyzed on 8% denaturation-polyacrylamide gel and detected with EB. Fragment sizes were calculated by Kdak Digital Science ID Image Analysis Software according to pBR322/Msp I Marker.

content (PIC) (Bostein et al., 1980) and effective allele number (Ne) (Kimura et al., 1974) were calculated using the SAS package (Sun, 2002). The Nei's genetic distances were calculated using PAPP (Guo et al., 1996). The results of 7 sheep microsatellite primers amplifying in goat and sheep were analyzed with ANOVA procedure using the SPSS package.

RESULTS

Statistical analysis

Allele frequencies were computed by the gene counting method. Heterozygosity (H), polymorphism information

From Table 2-7, we saw that the PIC and H of each microsatellite site is more than 0.7. the genetic information

· **삭제됨:** BASED ON MICROSATELLITE DNA

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Fable 2 (ii). Estimates o	f gene frequencies o:	f the microsatellite DNA sites
	E and the destruction of the second sec	

Allele Hu Tong Goat Allele Hu Tong Goat 120 0.4405 0.0351 0.0000 133 0.0119 0.0000 126 0.0000 0.0000 0.0000 0.0000 0.0172 91 0.0000 0.0250 0.133 126 0.0000 0.0714 0.0000 137 0.0595 0.0625 0.133 0.0000 0.0000 0.0000 0.0517 97 0.0000 0.0000 0.0250 0.1333 128 0.0400 0.0833 0.0000 143 0.0952 0.0121 132 0.0000 0.0517 97 0.0313 0.1060 0.0000 134 0.0541 0.0357 0.0476 147 0.0238 0.0124 138 0.0000 0.0517 101 0.0313 0.0500 0.0576 0.0000 1207 105 0.0000 0.0576 0.0000 0.0576 0.0000 0.0576 0.0000 0.0576 0.0000 0.0576 0.0000 0.		OarF	CB 11	-	-	MA	F 70			OarF	TB 304			OarFC	TB 128	
120 0.0405 0.0551 0.0000 133 0.0119 0.0469 0.0000 122 91 0.0000 0.0000 0.0667 124 0.0135 0.0238 0.0000 135 0.019 0.0469 0.0000 0.0000 0.0172 93 0.0000 0.0250 0.1333 126 0.0000 0.0144 0.0000 137 0.0595 0.0625 134 0.0000 0.0317 97 0.0000 0.0230 0.1333 128 0.0000 0.0033 0.0000 144 0.0952 0.0122 138 0.0000 0.00517 97 0.0000 0.0333 0.0000 1040 10400 10400 10400 10400 1040 1055 0.0000 0.0571 101 0.0313 0.1400 0.0157 104 0.0000 0.0571 0.0000 0.0333 1042 0.0000 0.0571 0.0333 0.0313 0.1400 0.0333 1142 0.0000 0.0000 0.0313 0.1400	Allele	Hu	Tong	Goat	Allele	Hu	Tong	Goat	Allele	Hu	Tong	Goat	Allele	Hu	Tong	Goat
124 0.0135 0.0238 0.0000 135 0.0119 0.0625 0.0625 0.0000 0.0000 0.03450 95 0.0000 0.0250 0.1333 126 0.0000 0.03714 0.0000 137 0.0555 0.0625 0.0001 0.0000 0.03450 95 0.0000 0.0300 0.0313 0.0000 0.0313 0.0000 0.0313 0.0000 0.0313 0.0000 0.0313 0.0000 0.0313 0.0000 0.0313 0.0000 0.0313 0.0000 0.0300 0.0300 0.0000 0.0313 0.0000 0.0313 0.0300 0.0313 0.0300 0.0313 0.0300 0.0313 0.0300 0.0313 0.0300 0.0313 0.0300 0.0331	120	0.0405	0.0351	0.0000	133	0.0119	0.0000	0.0000	126	0.0000	0.0000	0.0172	91	0.0000	0.0000	0.0667
126 0.0000 0.0714 0.0000 137 0.0595 0.0625 130 0.0000 0.0000 0.0315 95 0.0000 0.0250 0.1333 123 0.0000 0.0833 0.0000 141 0.0952 0.0625 0.133 0.0000 0.0000 0.0517 99 0.0313 0.1000 0.0000 0.0517 99 0.0313 0.0000 0.0000 0.0517 99 0.0313 0.0000 0.0000 0.0517 101 0.0313 0.0500 0.0000 0.0500 0.0000 0.0517 101 0.0313 0.0500 0.0000 0.0517 1010 0.0313 0.0500 0.0000 0.0517 103 0.0000 0.0517 1038 0.055 0.0161 144 0.0000 0.0000 0.0517 10.038 0.0750 0.0333 142 0.0270 0.0357 0.0238 153 0.0055 0.116 144 0.0000 0.0017 111 0.0166 0.0166 0.0000 0.0171 111 0.0166 0.0166 0.0000 0.0172 111 0.0166 <td< td=""><td>124</td><td>0.0135</td><td>0.0238</td><td>0.0000</td><td>135</td><td>0.0119</td><td>0.0469</td><td>0.0000</td><td>128</td><td>0.0000</td><td>0.0000</td><td>0.0172</td><td>93</td><td>0.0000</td><td>0.0250</td><td>0.1333</td></td<>	124	0.0135	0.0238	0.0000	135	0.0119	0.0469	0.0000	128	0.0000	0.0000	0.0172	93	0.0000	0.0250	0.1333
128 0.0405 0.1548 0.0000 139 0.1667 0.0231 0.1042 132 0.0000 0.0000 0.0517 97 0.0000 0.0250 0.1633 130 0.0270 0.0333 0.0000 141 0.0952 0.0250 0.0251 134 0.0000 0.0000 0.0517 97 0.0000 0.0313 0.1000 0.0000 134 0.0541 0.0714 0.0000 145 0.0119 0.0255 0.168 0.0000 0.0000 0.0517 101 0.0313 0.0300 0.0000 136 0.0457 0.0714 0.0000 145 0.0156 0.0000 0.0000 0.0517 107 0.0303 0.0000 0.0517 107 0.0300 0.0000 0.0517 107 0.0300 0.0000 0.0517 107 0.0335 0.1657 0.0238 116 0.0000 0.0517 107 0.0335 0.1657 0.0238 0.0300 0.0517 107 0.0300 0.0517 107 0.0300 0.0000 0.0517 107 0.0300 0.0000 0.0	126	0.0000	0.0714	0.0000	137	0.0595	0.0625	0.0625	130	0.0000	0.0000	0.3450	95	0.0000	0.0000	0.0667
130 0.00270 0.0833 0.00000 141 0.0925 0.0925 0.0225 0.0225 0.0000 0.0000 0.0517 99 0.0313 0.0000 0.0300 134 0.0541 0.0374 0.0000 145 0.0114 0.0015 0.0125 0.0125 0.0125 0.0000 0.0000 0.0517 101 0.0313 0.0300 0.0000 0.0000 0.0517 103 0.0000 0.0000 0.0000 0.0517 103 0.0000 0.0000 0.0517 103 0.0000 0.0000 0.0517 109 0.0313 0.1000 0.0000 0.0001 0.0017 109 0.0313 0.1000 0.0000 0.0001 144 0.0000 1055 0.0000 110 0.0313 0.1000 0.0000 0.0017 110 0.0133 0.1000 0.0000 0.0017 111 0.0133 0.1000 0.0000 0.0017 111 0.0133 0.1000 0.0000 0.0017 111 0.0133 0.1000 0.0000 0.0001 0.0117 113 0.00333 0.0000 0.0000 0.00	128	0.0405	01548	0.0000	139	0.1667	0.2031	0.1042	132	0.0000	0.0000	0.0517	97	0.0000	0.0250	0.1333
132 0.0000 0.0833 0.0000 143 0.0922 0.0312 0.0000 0.0017 10 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0017 110 0.0033 0.0000 0.0000 0.0017 111 0.0133 0.0000 0.0000 0.0017 111 0.0133 0.0000 0.0000 0.0017 111 0.0133 0.0000 0.0000 0.0151 111 0.0033 0.0000 0.0000 0.0151 111 0.0000 0.0000 <	130	0.0270	0.0833	0.0000]4]	0.0952	0.0625	0.0208	134	0.0000	0.0000	0.0517	99	0.0313	0.1000	0.1000
134 0.0541 0.0714 0.0000 145 0.0126 0.1042 138 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0127 105 0.0000 0.0750 0.0000 0.0333 0.1750 0.0000 0.0000 0.0156 0.0000 0.0000 0.0517 107 0.0333 0.1000 0.0333 142 0.0270 0.0357 0.0238 153 0.0955 0.0719 0.1575 146 0.0000 0.0001 0.0117 113 0.0156 0.0000 144 0.0551 0.0052 0.0000 157 0.158 0.0938 0.0208 150 0.0000 0.0171 113 0.0338 0.0350 0.0360 0.0000 0.0172 113 0.0338 0.0360 0.0000 0.0172 119 0.0000 0.0000 0.057 0.0300 0.0075 0.0000 0.0075 0.0000	132	0.0000	0.0833	0.0000	143	0.0952	0.0312	0.0625	136	0.0000	0.0000	0.0517	101	0.0313	0.0500	0.0000
136 0.0541 0.0377 0.0476 147 0.0286 0.0156 0.0000 0.0000 0.0207 105 0.0000 0.0750 0.0000 138 0.0577 0.0238 151 0.0000 0.0156 0.0000 0.0000 0.0517 107 0.0338 0.0750 0.0000 142 0.0270 0.0357 0.0238 153 0.0595 0.1719 0.1875 146 0.0000 0.0517 109 0.0313 0.0000 0.0314 0.0334 0.0308 0.0760 0.0000 144 0.0557 0.0235 0.0252 0.0001 155 0.0452 0.0000 0.0000 0.0315 111 0.0166 0.0000 0.0171 113 0.0383 0.0300 0.0300 0.0000 0.0315 0.0400 0.0000 0.0000 117 0.1313 0.1300 0.0000 0.0000 0.0000 117 0.0313 0.0000 0.0000 117 0.0313 0.0400 0.0000 117 0.0314 0.0000 0.015 0.0000 0.0000 0.015 0.0000 0.0172	134	0.0541	0.0714	0.0000	145	0.0119	0.0625	0.1042	138	0.0000	0.0000	0.0862	103	0.0000	0.0500	0.0000
138 0.0405 0.0714 0.0000 149 0.0476 0.0833 142 0.0000 0.0000 0.0517 107 0.0333 0.1300 0.0333 142 0.0270 0.0357 0.0238 153 0.0955 0.1719 0.1816 144 0.0000 0.0000 0.0517 109 0.0313 0.1300 0.0304 144 0.0541 0.0952 0.0000 155 0.0952 0.0781 0.6525 148 0.0000 0.0000 0.0172 113 0.0333 0.1300 0.0300 146 0.0946 0.0557 0.0100 157 0.1548 0.0208 152 0.0000 0.0000 0.0557 113 0.0333 0.466 0.0405 150 0.4055 0.0000 157 0.1548 0.0136 0.0416 152 0.0000 0.0000 0.0597 0.172 119 0.0000 0.0200 0.0272 0.1250 0.0750 0.0000 151 0.0405 0.0517 163 0.0476 0.0119 0.0000 0.0208 160 0.0166	136	0.0541	0.0357	0.0476]47	0.0238	0.0156	0.0208	140	0.0156	0.0000	0.1207	105	0.0000	0.0750	0.0000
140 0.0811 0.0057 0.0238 151 0.0000 0.0166 144 0.0000 0.0000 0.0517 109 0.0313 0.1000 0.0303 142 0.0270 0.0357 0.0238 153 0.0595 0.1719 0.1875 146 0.0000 0.0000 0.0517 111 0.01563 0.0000 0.0000 144 0.0445 0.0595 0.0000 155 0.0595 0.1719 0.1875 148 0.0156 0.0000 0.0172 113 0.0333 0.0300 0.0000 148 0.0405 0.0535 0.2143 159 0.0533 0.0469 0.1042 152 0.0000 0.0517 117 0.0133 0.1000 0.0200 150 0.0435 0.0194 161 0.0238 0.0156 0.0000 0.0761 0.0000 121 0.1250 0.0750 0.0000 152 0.0135 0.0119 0.0476 163 0.0166 0.0761 0.0000 123 0.0303 0.0000 0.0303 154 0.0500 0.0208	138	0.0405	0.0714	0.0000	149	0.0476	0.0312	0.0833	142	0.0000	0.0000	0.0517	107	0.0938	0.0750	0.0000
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144 0.0541 0.0925 0.0000 155 0.0952 0.0751 0.0000 0.0172 113 0.0938 0.0750 0.0000 146 0.0946 0.0595 0.0000 157 0.1548 0.0938 0.0208 150 0.0000 0.0000 0.0345 115 0.0338 0.0500 0.0000 150 0.0405 0.0537 0.1143 159 0.0469 0.0142 152 0.0000 0.0000 0.0595 117 0.0313 0.0000 0.0000 117 0.0313 0.0000 0.0000 10750 0.0000 0.0000 117 0.0313 0.0000 0.0000 10750 0.0000 0.0000 117 0.0133 0.0000 0.0000 10750 0.0000 113 0.0000 0.0000 1154 0.0000 1075 0.0000 112 0.1250 0.0750 0.0000 154 0.0405 0.0475 0.0475 0.0000 0.0000 0.0208 162 0.0938 0.0317 125 0.0313 0.0200 0.0000 164 0.1406 0.0761 0	142	0.0270	0.0357	0.0238	153	0.0595	0 1719	0.1875	146	0.0000	0.0000	0.0517	111	0.01563	0.0000	0.0000
146 0.0946 0.0595 0.0000 157 0.1548 0.0938 0.0208 150 0.0000 0.0000 0.0345 115 0.0338 0.0300 0.0300 148 0.0405 0.0257 0.2143 159 0.0833 0.0469 0.1042 152 0.0000 0.0001 0.0591 117 0.0313 0.1000 0.0600 150 0.0405 0.0238 0.1994 161 0.0238 0.0476 0.0156 0.0416 158 0.0000 0.0761 0.0000 121 0.1250 0.0750 0.0000 154 0.0541 0.0119 0.0476 165 0.0116 0.0156 0.0416 158 0.0000 123 0.0338 0.0000 0.0000 123 0.0338 0.0000 0.0000 155 0.0000 123 0.0338 0.0000 0.0000 123 0.0333 0.0300 0.0000 0.0312 0.0000 162 0.0938 0.0326 0.0517 129 0.0313 0.0250 0.0000 0.0313 0.0250 0.0000 0.0313 0.0250	144	0.0541	0.0952	0.0000	155	0.0952	0.0781	0.0625	148	0.0156	0.0000	0.0172	113	0.0938	0.0750	0.0000
148 0.0405 0.0357 0.2143 159 0.0833 0.0469 0.1042 152 0.0000 0.0000 0.069 117 0.0313 0.1000 0.0000 150 0.0405 0.0238 0.1904 161 0.0238 0.0156 0.0416 156 0.0166 0.0000 0.0761 0.0000 121 0.1250 0.0750 0.0000 154 0.0541 0.019 0.0476 165 0.0156 0.0156 0.0751 0.0000 121 0.1250 0.0750 0.0000 156 0.0541 0.0000 0.0238 167 0.0000 0.0208 162 0.0938 0.0326 0.0517 125 0.0313 0.0000 0.0000 158 0.0405 0.0537 0.0000 169 0.0000 0.0312 0.0000 164 0.1406 0.0761 0.0600 127 0.0313 0.0250 0.0001 160 0.0238 0.0000 0.0000 169 0.0000 0.0312 0.0000 164 0.0455 0.0261 0.0000 131 0.0250	146	0.0946	0.0595	0.0000	157	0.1548	0.0938	0.0208	150	0.0000	0.0000	0.0345	115	0.0938	0.0500	0.0000
150 0.0405 0.0238 0.1904 161 0.0238 0.0156 0.0166 0.0000 0.0172 119 0.0000 0.0750 0.0000 152 0.0135 0.0119 0.0046 163 0.0146 158 0.0000 0.0761 0.0000 121 0.1230 0.0750 0.0000 154 0.0541 0.0119 0.0000 0.0208 160 0.0156 0.0517 0.0000 123 0.0600 0.0000 158 0.0455 0.0357 0.0000 169 0.0000 0.0312 0.0000 164 0.1466 0.0761 0.0600 127 0.0313 0.0500 0.0000 160 0.0270 0.0238 0.0000 0.0312 0.0000 164 0.1466 0.0761 0.0600 127 0.0313 0.0250 0.0300 162 0.0405 0.0000 0.0000 169 0.0288 0.0326 0.0000 131 0.0625 0.0000 131 0.0250 0.0000 164 0.0946 0.0000 0.0714 172 0.156	148	0.0405	0.0357	0.2143	159	0.0833	0.0469	0.1042	152	0.0000	0.0000	0.069	117	0.0313	0.1000	0.0000
152 0.0135 0.0119 0.0952 163 0.0476 0.0156 0.0416 158 0.0000 0.0761 0.0000 121 0.1250 0.0750 0.0000 154 0.0541 0.0109 0.0208 167 0.0000 0.0156 0.0516 0.0517 125 0.0313 0.0500 0.0000 0.0000 158 0.0455 0.0557 0.0000 169 0.0000 0.0512 0.0000 164 0.1466 0.0571 129 0.0313 0.0500 0.0000 0.0333 160 0.0270 0.0238 0.0000 0.0015 0.0000 166 0.1719 0.0978 0.0517 129 0.0313 0.0250 0.0333 162 0.4055 0.0000 0.0000 166 0.1719 0.0978 0.0517 129 0.0313 0.0250 0.0000 164 0.0466 0.0000 0.1714 172 0.0156 0.0425 0.0000 133 0.0313 0.0250 0.0000 0.0313 0.0250 0.0000 0.0326 0.0000 133 0.0	150	0.0405	0.0238	0 1904	161	0.0238	0.0156	0.0416	156	0.0156	0.0000	0.0172	119	0.0000	0.0750	0.0000
154 0.0541 0.0119 0.0476 165 0.0119 0.0000 0.0208 160 0.0156 0.0761 0.0000 123 0.0338 0.0000 0.0000 158 0.0405 0.0357 0.0000 169 0.0000 0.0312 0.0000 164 0.0761 0.0600 127 0.0313 0.0300 0.0000 158 0.0270 0.0238 0.0000 0.0312 0.0000 164 0.1406 0.0517 129 0.0313 0.0300 0.0000 160 0.0270 0.0238 0.0000 0.0000 166 0.1719 0.0313 0.0250 0.0331 0.0250 0.0331 0.0250 0.0000 0.0000 0.0000 0.0667 164 0.0946 0.0000 0.0714 172 0.0156 0.0455 0.0000 133 0.0313 0.0250 0.0000 168 0.0000 0.0000 0.0714 172 0.0156 0.0435 0.0000 133 0.0200 0.0000 0.0000 0.0238 174 0.0000 0.0238 139 0.0	152	0.0135	0.0119	0.0952	163	0.0476	0.0156	0.0416	158	0.0000	0.0761	0.0000	121	0.1250	0.0750	0.0000
156 0.0541 0.0000 0.0238 167 0.0000 0.0156 0.0288 162 0.0938 0.0326 0.0517 125 0.0313 0.0500 0.0000 158 0.0405 0.0357 0.0000 0.0512 0.0000 164 0.1406 0.0761 0.0690 127 0.0313 0.0000 0.0333 160 0.0270 0.0238 0.0000 0.1512 0.0179 0.0978 0.0517 129 0.0313 0.0260 0.0333 162 0.0946 0.0000 0.1900 166 0.1719 0.0978 0.0001 131 0.0250 0.0000 164 0.0946 0.0000 0.0714 172 0.0156 0.0469 0.0325 0.0000 133 0.0250 0.0000 168 0.0500 0.0714 174 174 0.0000 0.0325 0.0000 133 0.0250 0.0000 0.0250 0.0000 0.0333 170 0.135 0.0000 0.	154	0.0541	0.0119	0.0476	165	0.0119	0.0000	0.0208	160	0.0156	0.0761	0.0000	123	0.0938	0.0000	0.0000
158 0.0405 0.0357 0.0000 169 0.0000 0.0312 0.0000 164 0.1406 0.0761 0.0690 127 0.0313 0.0000 0.0000 160 0.0270 0.0238 0.0000 0.0000 166 0.1719 0.0978 0.0517 129 0.0313 0.0200 0.0000 162 0.0405 0.0000 0.0000 166 0.0719 0.0928 0.0000 131 0.0625 0.0000 0.0667 164 0.0946 0.0000 0.0714 170 0.0455 0.0000 133 0.0250 0.0000 0.0667 166 0.0541 0.0000 0.0714 172 0.0156 0.0435 0.0000 133 0.0250 0.0000 0.0667 168 0.0000 0.0000 0.0714 174 0.0000 0.0325 0.0000 137 0.0000 0.0000 0.0333 0.0250 0.0000 0.0333 0.0250 0.0000 0.0333 0.0250 0.0000 0.0333 137 0.0000 0.0000 0.0000 0.0000	156	0.0541	0.0000	0.0238	167	0.0000	0.0156	0.0208	162	0.0938	0.0326	0.0517	125	0.0313	0.0500	0.0000
160 0.0270 0.0238 0.0000 166 0.1719 0.0978 0.0517 129 0.0313 0.0250 0.0333 162 0.0405 0.0000 0.0000 168 0.0938 0.0326 0.0000 131 0.0625 0.0000 0.0667 164 0.0946 0.0000 0.1190 170 0.0469 0.0326 0.0000 133 0.0313 0.0250 0.0000 166 0.0541 0.0000 0.0714 172 0.0156 0.0435 0.0000 133 0.0250 0.0000 0.0250 0.0260 0.0300 0.0000 0.0250 0.0260 0.0333 170 0.1135 0.0000 0.0714 174 0.0000 0.0326 0.0000 137 0.0000 0.0250 0.0261 170 0.1135 0.0000 0.0714 174 0.0000 0.0345 141 0.0313 0.0000 0.0300 0.0000 0.0333 174 0.0000 0.0000 0.0238 178 0.0469 0.0109 0.0345 141 0.0313 0.0000	158	0.0405	0.0357	0.0000	169	0.0000	0.0312	0.0000	164	0.1406	0.0761	0.0690	127	0.0313	0.0000	0.0000
162 0.0405 0.0000 0.0000 168 0.0938 0.0326 0.0000 131 0.0625 0.0000 0.0667 164 0.0946 0.0000 0.1190 170 0.0469 0.0326 0.0000 133 0.0250 0.0000 0.0000 166 0.0541 0.0000 0.0000 0.0714 172 0.0156 0.0435 0.0000 133 0.0250 0.0000 0.0300 0.0000 0.0000 0.0000 0.0000 0.0000 0.0325 0.0000 133 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0000 0.0325 0.0000 133 0.0000 </td <td>160</td> <td>0.0270</td> <td>0.0238</td> <td>0.0000</td> <td></td> <td></td> <td></td> <td></td> <td>166</td> <td>0.1719</td> <td>0.0978</td> <td>0.0517</td> <td>129</td> <td>0.0313</td> <td>0.0250</td> <td>0.0333</td>	160	0.0270	0.0238	0.0000					166	0.1719	0.0978	0.0517	129	0.0313	0.0250	0.0333
164 0.0946 0.0000 0.1190 170 0.0469 0.0326 0.0000 133 0.0313 0.0250 0.0000 166 0.0541 0.0000 0.0714 172 0.0156 0.0435 0.0000 133 0.0313 0.0250 0.0000 168 0.0000 0.0000 0.0714 174 0.0000 0.0345 139 0.0000 0.0250 0.0667 170 0.135 0.0000 0.0476 176 0.0625 0.0217 0.0345 139 0.0000 0.0300 0.0000 0.0333 0.0000 0.0300 0.0250 0.0667 174 0.0000 0.0000 0.0238 178 0.0469 0.0109 0.0345 141 0.0313 0.0000 0.0300 174 0.0000 0.0000 0.0238 178 0.0469 0.0172 143 0.0000 0.0300 0.0300 0.0300 0.0300 0.0300 0.0300 0.0300 0.0300 0.0300 0.0300	162	0.0405	0.0000	0.0000					168	0.0938	0.0326	0.0000	131	0.0625	0.0000	0.0667
166 0.0541 0.0000 0.0714 172 0.0156 0.0435 0.0000 135 0.0000 0.0000 0.0000 0.0000 0.0000 0.0250 0.0000 0.0250 0.0000 0.0250 0.0000 0.0250 0.0667 170 0.0135 0.0000 0.0476 176 0.0625 0.0217 0.0345 139 0.0000 0.0000 0.0333 174 0.0000 0.0000 0.0238 178 0.0465 0.0172 143 0.0000 0.0000 0.0333 174 0.0000 0.0000 0.0238 178 0.0465 0.0172 143 0.0000 0.0000 0.0333 182 0.0781 0.0870 0.0172 153 0.0000 <	164	0.0946	0.0000	0.1190					170	0.0469	0.0326	0.0000	133	0.0313	0.0250	0.0000
168 0.0000 0.0000 0.0714 174 0.0000 0.0326 0.0000 137 0.0000 0.0250 0.0467 170 0.0135 0.0000 0.0476 176 0.0625 0.0217 0.0345 139 0.0000 0.0300 0.0333 174 0.0000 0.0000 0.0238 178 0.0469 0.0109 0.0345 141 0.0300 0.0000 0.0300 180 0.0625 0.0476 0.0469 0.0172 143 0.0000 0.0300<	166	0.0541	0.0000	0.0714					172	0.0156	0.0435	0.0000	135	0.0000	0.0000	0.1000
170 0.0135 0.0000 0.0476 176 0.0625 0.0217 0.0345 139 0.0000 0.0000 0.0333 174 0.0000 0.0000 0.0238 178 0.0469 0.0109 0.0345 141 0.0313 0.0000 0.0000 0.0333 180 0.0625 0.0476 0.0875 0.0172 143 0.0000 0.0000 0.0333 182 0.0781 0.0870 0.0172 153 0.0000 0.0000 0.0667 184 0.0156 0.0761 0.0000 159 0.0000 0.0000 0.0667 184 0.0156 0.0000 0.0000 0.0000 0.0000 0.0000 0.0667 186 0.0156 0.0000 0.0000 159 0.0000 0.0000 0.0667 190 0.0156 0.0543 0.0000 0.0000 0.0667 190 0.0156 0.0543 0.0000 0.0000 0.0667 190 0.0156 0.0543 0.0000 0.0667 192 0.0469 0.0453 0.0000 <td>168</td> <td>0.0000</td> <td>0.0000</td> <td>0.0714</td> <td></td> <td></td> <td></td> <td></td> <td>174</td> <td>0.0000</td> <td>0.0326</td> <td>0.0000</td> <td>137</td> <td>0.0000</td> <td>0.0250</td> <td>0.0667</td>	168	0.0000	0.0000	0.0714					174	0.0000	0.0326	0.0000	137	0.0000	0.0250	0.0667
174 0.0000 0.0238 178 0.0469 0.0109 0.0345 141 0.0313 0.0000 0.0333 180 0.0625 0.0435 0.0172 143 0.0000 0.0333 182 0.0781 0.0870 0.0172 153 0.0000 0.0300 0.0333 184 0.0156 0.0781 0.0000 0.0000 0.0667 184 0.0156 0.0000 0.0000 0.0000 0.0000 0.0667 186 0.0156 0.0581 0.0000 0.0000 0.0000 0.0000 0.0667 186 0.0156 0.0253 0.0000 0.0000 0.0000 0.0000 0.0000 188 0.0156 0.0543 0.0000 0.0000 0.0000 0.0000 190 0.0156 0.0526 0.0000 0.0000 192 0.0469 0.0000 0.0000 194 0.0156 0.0266 0.0000 0.0000 194 0.0000 198 0.0000 0.0000 198 0.0000 0.0109 0.00000 0.0000 </td <td>170</td> <td>0.0135</td> <td>0.0000</td> <td>0.0476</td> <td></td> <td></td> <td></td> <td></td> <td>176</td> <td>0.0625</td> <td>0.0217</td> <td>0.0345</td> <td>139</td> <td>0.0000</td> <td>0.0000</td> <td>0.0333</td>	170	0.0135	0.0000	0.0476					176	0.0625	0.0217	0.0345	139	0.0000	0.0000	0.0333
180 0.0625 0.0435 0.0172 143 0.0000 0.0000 0.0333 182 0.0781 0.0870 0.0172 153 0.0000 0.0600 0.0667 184 0.0156 0.0056 0.0000 159 0.0000 0.0667 186 0.0156 0.0000 0.0000 0.0667 188 0.0156 0.0000 0.0000 0.0667 190 0.0156 0.0543 0.0000 0.0600 192 0.0469 0.0435 0.0000 0.0000 194 0.0156 0.0526 0.0000 0.0000 192 0.0469 0.0435 0.0000 0.0000 194 0.0156 0.0266 0.0000 0.0000 196 0.0000 0.0109 0.0000 0.0000	174	0.0000	0.0000	0.0238					178	0.0469	0.0109	0.0345	141	0.0313	0.0000	0.0000
182 0.0781 0.0870 0.0172 153 0.0000 0.0667 184 0.0156 0.0761 0.0000 159 0.0000 0.0667 186 0.0156 0.0000 0.0000 0.0000 0.0667 188 0.0156 0.0000 0.0000 0.0000 198 0.0156 0.0543 0.0000 190 0.0156 0.0543 0.0000 192 0.0469 0.0435 0.0000 194 0.0156 0.0326 0.0000 196 0.0000 0.0109 0.0000 198 0.0156 0.0526 0.0000									180	0.0625	0.0435	0.0173	143	0.0000	0.0000	0.0333
184 0.0156 0.0761 0.0000 159 0.0000 0.0000 0.0000 186 0.0156 0.0000 0.0000 188 0.0156 0.0000 188 0.0156 0.057 0.0000 190 0.0000 190 0.0156 0.0533 0.0000 192 0.0469 0.0435 0.0000 194 0.0156 0.0326 0.0000 196 0.0000 196 0.0000 198 0.0000 0.0000 198 0.0000 0.0000									182	0.0781	0.0870	0.0172	153	0.0000	0.0000	0.0667
186 0.0156 0.0000 0.0000 188 0.0156 0.1087 0.0000 190 0.0156 0.0543 0.0000 192 0.0469 0.0435 0.0000 194 0.0156 0.0326 0.0000 196 0.0000 0.0109 0.0000 198 0.0000 0.0109 0.0000									184	0.0156	0.0761	0.0000	159	0.0000	0.0000	0.0667
188 0.0156 0.1087 0.0000 190 0.0156 0.0543 0.0000 192 0.0469 0.0435 0.0000 194 0.0156 0.0326 0.0000 196 0.0000 0.0109 0.0000 198 0.0000 0.0109 0.0000									186	0.0156	0.0000	0.0000				
190 0.0543 0.0000 192 0.0469 0.0435 0.0000 194 0.0156 0.0256 0.0000 196 0.0000 0.0109 0.0000 198 0.0000 0.0109 0.0000									188	0.0156	0.1087	0.0000				
192 0.0469 0.0435 0.0000 194 0.0156 0.0326 0.0000 196 0.0000 0.0109 0.0000 198 0.0000 0.0109 0.0000									190	0.0156	0.0543	0.0000				
194 0.0156 0.0326 0.0000 196 0.0000 0.0109 0.0000 198 0.0000 0.0109 0.0000									192	0.0469	0.0435	0.0000				
196 - 0.0000 - 0.0000 198 - 0.0000 - 0.0000									194	0.0156	0.0326	0.0000				
198 0.0000 0.0109 0.0000									196	0.0000	0.0109	0.0000				
									198	0.0000	0.0109	0.0000				

Table 3. Polymor	phism information co	ontent (PIC) and Hete	rozygosity (H) of	each microsatellite :	site	
Site		PIC			Н	
	Hu sheep	Tong sheep	Goat	Hu sheep	Tong sheep	Goat
OarFCB 48	0.9240	0.9252	0.9138	0.9284	0.9296	0.9195
OarAE 101	0.8520	0.8987	0.9343	0.8664	0.9061	0.9378
MAF 33	0.8985	0.8986	0.6869	0.9056	0.9059	0.7144
OarFCB 11	0.9409	0.9198	0.8636	0.9438	0.9246	0.8753

0.8992

0.9374

0.9068

0.9000

0.9101

0.9120

0.8856

0.9291

0.9242

was abundant: and we also found that more than 7 alleles could be detected at each sheep microsatellite site in 3 populations. According to the protocol for the estimation of the global animal breeds distance. Barker put torward the selection standard for microsatellite DNA: only if there are more than 4 can the microsatellite site be used (Barker et al. 1994). At the same time, we compared the genetic distances obtained from 7 sheep microsatellites and found that the distance between goat and sheep was longer than the distance between Hu sheep and Tong sheep, thus, OarFCB48, OarAE101, MAF33, OarFCB11, MAF70, OarFCB304 and OarFCB128 could be used in the research of the genetic differentiation between sheep and goats. And

0.8926

0.9036

0.9055

MAF 70

OarFCB 304

OarFCB 128

we also could see from the tables the difference in gene amount and gene frequencies in the same microsatellite site in the different breeds (species), which could show the difference in heredity among them., This also indicated that the 7 microsatellite sites could be used to distinguish the breeds and species. The common alleles were the most antique and conservative, the other alleles were the result of insertions, deletions and mutations.

0.9008

0.9331

0.9288

0.9063

0.9406

0.9133

Table 8-9 also shows the conservation of each microsatellite among 3 breeds (species): there were no significant differences in allele number of amplifying (p>0.05) and allele size of amplifying (p>0.05) in each microsatellite site.

Table 4. Effective number of alleles of each microsatellite DNA $-\frac{T}{2}$ site

Site	Hu sheep	Tong sheep	Goat
OarFCB 48	13.9738	14.2043	12.4183
OarAE 101	7,4839	10.6474	16.0637
MAF 33	10,5960	10.6312	3.5014
OarFCB 11	17.7772	13.2683	8.0218
MAF 70	10.0796	9.4382	10.6676
OarFCB 304	11.1285	14.9520	16.8237
OarFCB 128	11.3673	14.0351	11.5385

Table	5.	Mean	polyi	norphis	m i	nform	nation	content	(mean P	IC)
mean	he	terozyg	gosity	(mean	H)	and	mean	effective	number	i o
alleles	(π	iean N	e) of r	nierosat	ellit	te DN	Asite	s in 3 pop	oulations	

	Hu sheep	Tong sheep	Goat
mean PIC	0.9024	0.9116	0.8821
mean H	0.9095	0.9184	0.8906
mean Ne	11.7723	12.4538	11.6104

DISCUSSION

Sun (2002) also used 20 biochemical markers to detect the Tong sheep. Hu sheep and goats, and concluded that the microsatellite sites provided more accurate values and more abundant information than protein markers, for example, the PIC. H and Ne of each protein marker was smaller than those of microsatellite sites. The 20 biochemical markers indicated that the genetic distance between Hu sheep and Tong sheep, Hu sheep and goats, Tong sheep and goats was 0.0268, 0.2411, 0.2476, respectively, which is also smaller than those of the 3 populations based on microsatellite sites. but the dendrogram of the 3 populations based on the microsatellites is similar to that of the 3 populations based on biochemical markers. Thus, the microsatellites might be a better indicator than protein polymorphisms of evolutionaryrelationships among populations or between closely related species.

Nei thought that the time polymorphism alleles existed in a population were longer than the time the breed existed,

able 6. The standard genetic distance among 3 populations	
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	Hu sheep	Tong sheep	Goat
Hu sheep	0.0000	0.2321	1.2313
Tong	0.2321	0.0000	1.0921
Goat	1.2313	1.0921	0.0000

so that gene diversity (heterzygosity) in one locus in one breed is relative with the gene diversity of its closely related species (Nei et al., 1983). For the conservation of microsatellite profile sequences, microsatellites could be used in many species, the microsatellite primer of one breed might be used in its relative species, and for example, the sheep microsatellite could amplify in goats, which provided the possibility of expediting the obtaining of microsatellite primers and speeding up the comparative genome map. Because of the slow research progress of goat microsatellites, the first genetic linkage map of the goat published by Vaiman (Wu et al., 1999) only reported 10 goat microsatellite to study the genetic diversity of goats.

In general, during the evolution of mammalian chromosomes, the difference in chromosomes of closely related species was caused by the Robertsonian translocation (fusion or fission) near the centromere. Comparing the G band type between *Capra* and *Ovis* could yield significant homology. Some M chromosomes of SM chromosomes of *Ovis*: M1, M2. M3 and M4 were caused from the Robertsonian fusion of *Capra* 1/5. 3/10, 4/9 and 11/17 chromosomes. Thus, it was believed that the sheep and the goat originated from the same ancestor, the goat karyotype was similar to the ancestral, the telocentric chromosome of goat caused the Robertsonian fusion, the number of goat chromosomes was reduced from 2n=60 to 2n=54 and differentiated into the sheep karyotype (Chang, 1995).

Concerning the conservation of microsatellites and the similar origin of closely related species, selecting microsatellite sites located on the chromosome where the

Table 7. The comparison of 7 microsatellite primers amplifying in Goat. Hu sheep and Tong sheep

Site	Hu sh	eep	Tong s	heep	Goa	Goat	
	Allele numbers	Size	Allele numbers	Size	Allele numbers	Size	
OarFCB 48	23	141-197	21	127-181	17	153-189	
OarAE 101	10	93-113	14	85-119	21	75-131	
MAF 33	20	116-160	16	120-154	7	110-126	
OarFCB 11	22	120-170	19	120-160	13	136-174	
MAF 70	16	133-165	17	135-169	16	137-167	
OarFCB 304	20	140-194	20	158-198	22	126-18	
OarFCB 128	17	99-141	17	93-137	14	91-159	

Table 8. The comparison of microsatellite allele number and range of allele's size in Goat, Hu sheep and Tong sheep Allele number of amplifying

	Sum of squares	df	Mean square	F	P value
Between groups	25.524	2	12.762	0.745	0.489
Within groups	308.286	18	17.127		
Total	333.810	20			

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Table 9. The comparison of microsatellite allele number and range of allele's size in goat, Hu sheep and Tong sheep Allele size of amplifying

	Sum of squares	df	Mean square	F	P value
Between groups	34,667	2	17.333	0.096	0.909
Within groups	3244,571	18	180.254		
Total	3279.238	20			



Figure 1. Electrophoresis photograph of some microsatellite sites

Robertsonian fusion occurred between sheep and goat, may be used in the study of genetic differentiation and evolutionary relationships between sheep and goats.

ACKNOWLEDGEMENTS

This work was supported by the International Cooperation Item of the National Natural Science Foundation of China (30310103007).

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