

# Ancient DNA Reveals a Migration of the Ancient Di-Qiang Populations Into Xinjiang as Early as the Early Bronze Age

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**ABSTRACT** Xinjiang is at the crossroads between East and West Eurasia, and it harbors a relatively complex genetic history. In order to better understand the population movements and interactions in this region, mitochondrial and Y chromosome analyses on 40 ancient human remains from the Tianshanbeilu site in eastern Xinjiang were performed. Twenty-nine samples were successfully assigned to specific mtDNA haplogroups, including the west Eurasian maternal lineages of U and W and the east Eurasian maternal lineages of A, C, D,

F, G, Z, M7, and M10. In the male samples, two Y chromosome haplogroups, C\* and N1 (xN1a, N1c), were successfully assigned. Our mitochondrial and Y-chromosomal DNA analyses combined with the archaeological studies revealed that the Di-qiang populations from the Hexi Corridor had migrated to eastern Xinjiang and admixed with the Eurasian steppe populations in the early Bronze Age. *Am J Phys Anthropol* 157:71–80, 2015. © 2014 Wiley Periodicals, Inc.

Xinjiang is a geographic region in the northwest part of China, and it connects East and West Eurasia. In this region, even before the opening of the Silk Road, population migrations, interactions, and trade between regional tribes took place continuously (Lin, 2001). Archaeological and anthropological research has shown that several populations of West Eurasian ancestry immigrated into Xinjiang in the beginning of the Bronze Age (Han, 1991, 1998; Lin, 2003). In recent years, ancient DNA studies of Xinjiang have suggested that the East and West Eurasian maternal lineages were admixed in this region (Gao et al., 2008; Cui et al., 2009, 2010; Li et al., 2010a).

The eastern part of Xinjiang is situated between the Hexi Corridor of China and the Eurasian steppe zone, where large scale population movements have occurred since the Bronze Age. The Hexi Corridor is considered to be the area where the Di-qiang populations originated. The corridor has a long history of human habitation and primitive agriculture. The Qijia and Siba cultures, both of which can be traced back to the more ancient Yangshao Culture in the Yellow River valley, were two major Di-qiang cultures that flourished in the Hexi Corridor during the Neolithic to the Bronze Age (Yan, 1978; Li, 1993, 2009). According to ethnological studies, the Di-qiang populations contributed to the development of the current Han and Tibeto-Burman (TB) speaking populations (Yang and Ding, 2003). As the eastern part of Xinjiang is located in the transition zone of the steppe and ancient oriental cultures, this region has played a substantial role in research on the transmissions of early agriculture and the technologies of ceramics and bronze metallurgy (Chen and Hiebert, 1995; Mei, 2000; Lin,

2001, 2003; Svyatko et al., 2013). Questions on the relationship between ceramic, bronze, and agricultural technologies, and population migrations and interactions are thus paramount to archaeologists and geneticists. The Tianshanbeilu site (also called the Linya site) is located in the Hami city of eastern Xinjiang, in northwestern China (Fig. 1). According to archaeological research and C<sup>14</sup> data, the date of the Tianshanbeilu site is 1900–1300 BC (Ding, 1996; Li, 2002, 2009). A large number of bronze vessels and pottery were excavated from the site, and the archaeological culture of this site was named the Tianshanbeilu Culture. The characteristics of the bronze vessels and the burial styles are consistent with

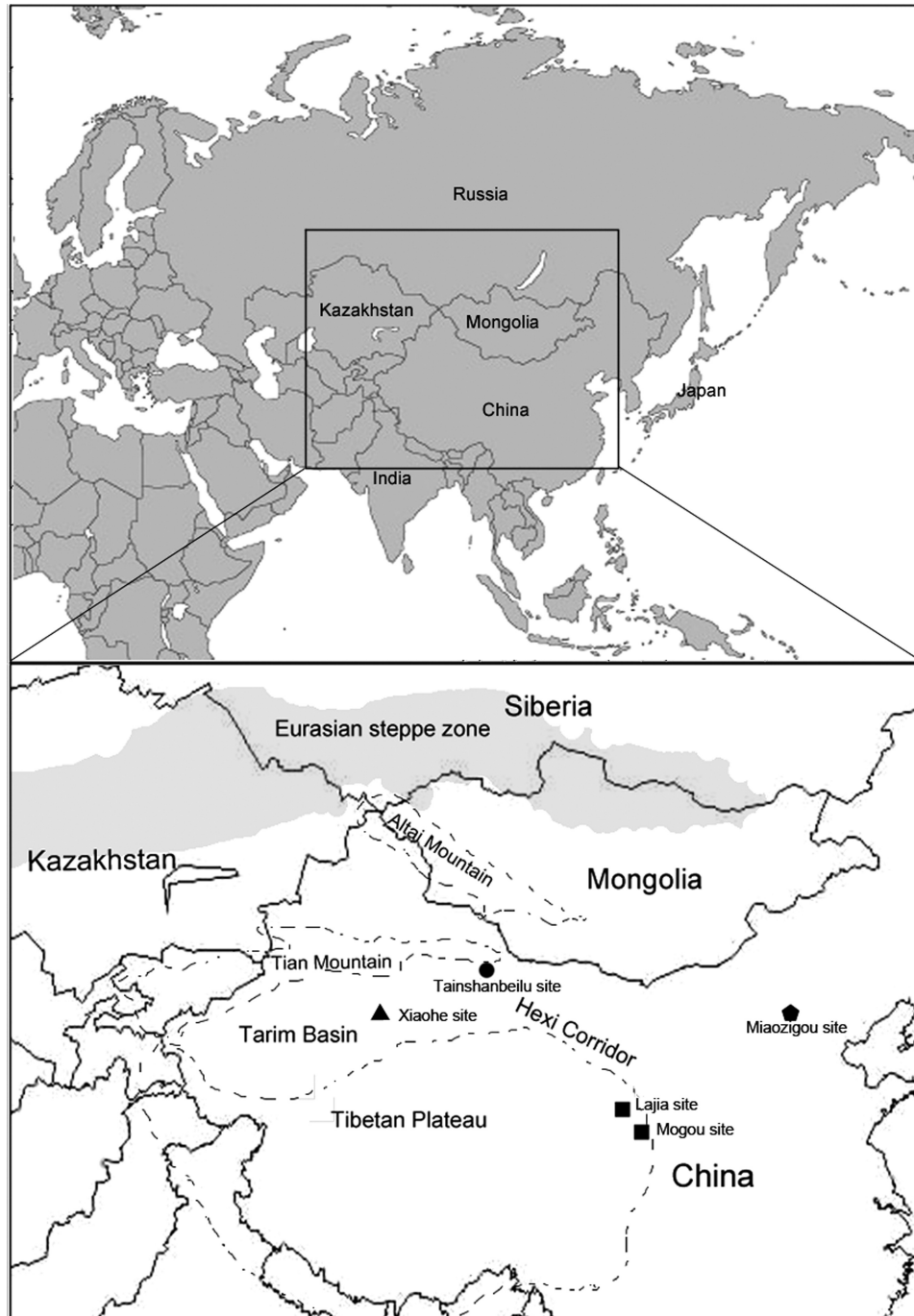
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**Fig. 1.** Geographic location of the Tianshanbeilu population. The *black circle* represents the site sampled for ancient DNA in this study. The *black squares* represent the sites of the ancient Di-qiang populations: the Lajia and Mogou. The *black diamond* represents the Miaoziqou site of the ancient Yangshao Culture. The *black triangle* represents the Xiaohe site in the Tarim Basin. The *gray area* signifies the location of the Eurasian Steppe.

those of the Eurasian steppe cultures, such as the Okunev Culture in South Siberia (Li, 2002; Lin, 2003; Li, 2009). Of note, the abundant amount of pottery discovered at this Bronze Age site is unusual to see in Xinjiang. For example, no pottery was found in the Bronze Age Xiaohe site of the Tarim Basin (Han, 2007), which is about 360 km away from the Tianshanbeilu site. The pottery here can be classified into two groups. One group, which has never been found elsewhere in Xinjiang, has a relatively

high similarity to the pottery of the Siba or pre-Siba Cultures (Late Neolithic to the early Bronze Age). In the Late Neolithic, the Machang type of the Yangshao Culture was transmitted westward into the Hexi Corridor, where it first formed the pre-Siba Culture, which then developed into the Siba Culture. The Siba Culture was one of the primary Bronze cultures found west of the Hexi Corridor (Li, 1993). The other group of pottery is very similar to that of the Eurasian steppe cultures (Li, 2002, 2009).

Thus, the Tianshanbeilu Culture is evidence of cultural integration between the Eurasian steppe cultures and the Di-qiang cultures (Li, 2002; Lin, 2003; Li, 2009). Two hypotheses were proposed to explain the formation of the Tianshanbeilu Culture (Li, 2009). These hypotheses are: (1) The Eurasian steppe populations in Siberia moved south into the eastern part of Xinjiang and settled there. Through the diffusion of pre-Siba and Siba culture to the Eurasian steppe populations through cultural exchanges or pottery trade, the Eurasian steppe populations mastered the ceramic technology of the Di-qiang populations living in the Hexi Corridor. In this hypothesis, these two populations did not admix. (2) Both the Eurasian steppe populations and the ancient Di-qiang populations of the Hexi Corridor immigrated into the eastern part of Xinjiang and admixed there. The Di-qiang populations brought their ceramic technology to this region in the Early Bronze Age. The human remains excavated from this site offer an opportunity to study the prehistoric demographic events using genetic tools. The molecular genetic data provide invaluable data to test these two hypotheses.

In this study, we aim to study the maternal and paternal lineages of the Tianshanbeilu population. As there is a clear geographical distribution of the mtDNA and Y-DNA haplogroups, the data may provide genetic evidence for the origins of this prehistoric population. This study will provide further insight for understanding the patterns of population interaction between the East Eurasians and the Eurasian steppe populations in the eastern region of Xinjiang, China.

## MATERIALS AND METHODS

### Sample collection

The Tianshanbeilu site is situated in northwest China where the climate is cold and dry. Organic remains are well-preserved at this site. Samples for DNA research were collected from teeth without caries and cracks of 40 human remains excavated at the Tianshanbeilu site in Xinjiang.

### Ancient DNA extraction

Teeth were soaked in 5% sodium hypochlorite solution for 10 min, then washed with distilled water and ethanol. Each side of teeth were exposed to ultraviolet (UV) light (254 nm) for 30 min. A freezer mill was used (6750 Freezer Mill, Spex SamplePrep, United States) to pulverize the teeth in liquid nitrogen, and the subsequent powder was stored at  $-20^{\circ}\text{C}$ . Tooth powder (0.2 g) and a blank control were incubated in a 4 mL solution containing 0.5% SDS, 0.5 M EDTA, and 2 mg/mL proteinase K at  $50^{\circ}\text{C}$  for 24 h in a shaker (220 rpm/min). DNA was carefully extracted using the QIAamp DNA Mini Kit (Qiagen, Germany), according to the manufacturer's protocol.

### Mitochondrial DNA amplification, sequencing, and APLP typing

Amplification of mtDNA was performed on two subregions (a and b) of the first hypervariable segment of the control region (HVSI) using the primers listed in Table 1. PCR amplification was carried out in 25  $\mu\text{L}$  of a reaction mixture containing 2 mM  $\text{MgCl}_2$ , 67 mM Tris-HCl (pH 8.8), 50 mM KCl, 0.5  $\mu\text{M}$  of each primer, 500 mM each of dNTPs, 1.3 mg/mL BSA, 1 Unit of Taq polymerase (Promega), and 2  $\mu\text{L}$  extract. PCR conditions were  $94^{\circ}\text{C}$  for 10 min, followed by 32 cycles with  $94^{\circ}\text{C}$

for 50 s, 45 s at  $54^{\circ}\text{C}$ , 1 min at  $72^{\circ}\text{C}$ , extension for 10 min at  $72^{\circ}\text{C}$ , and storage at  $4^{\circ}\text{C}$ . Negative controls were included in each DNA extraction and PCR run. PCR products were checked on a 2% agarose gel and purified with a QIAquick Gel Extraction Kit (Qiagen, Germany). Amplification products were sequenced using the ABI 310 Terminator Sequencing kit (Applied Biosystems, United States) according to the manufacturer's instructions. The products were analyzed on the ABI PRISM 3100 automatic sequencer (Applied Biosystems, United States).

Eleven sets of primers (Table 1) were used to type the key SNPs of the mtDNA coding regions. Haplogroups M/N and F were identified by using the amplified product-length polymorphism (APLP) method (Shinoda et al., 2006). Haplogroups A, C, D, G, Z, M7, M10, U, and W were examined by direct sequencing. The amplifying conditions and cycling parameters were the same as for HVSI. For each sample, the characteristic site at position 10,400 for macrohaplogroup M/N was screened first. Then, on the basis of the HVSI motifs and the mtDNA phylogenetic tree, other characteristic sites of the coding region were used to further define the haplogroup.

### Y chromosome SNP and STR analysis

Sex identification was performed using amelogenin fragment polymorphism (Liu et al., 2004). The male samples were analyzed further. Y-chromosomal single nucleotide polymorphisms (Y-SNPs) were typed, as they are diagnostic for major branches in the Y chromosome haplogroup tree (Underhill et al., 2000, 2001; Karafet et al., 2008). These Y-SNPs include C-M216, C1-M8, C2-M38, C3-M217, F-M89, K-M9, P-M45, NO-M214, N-M231, N1a-M128, N1b-P43, and N1c-TAT, and they were identified by direct sequencing. The primers used in SNP markers are shown in Table 1. Y chromosome STR analysis of the ancient samples was performed on 17 loci (DYS19, DYS385, DYS389I, DYS389II, DYS390, DYS391, DYS392, DYS393, DYS437, DYS447, DYS438, DYS439, DYS448, DYS456, DYS458, DYS635, and Y GATA H4) using the AGCU® mini STR Kit (AGCU ScienTech, China).

### Cloning of PCR Products

To test the authenticity of the results, four samples (3, 8, 14, and 18) were randomly chosen to be cloned. The mtDNA HVSI products were cloned using the pGEM-T Easy Vector System I (Promega, United States) according to the manufacturer's instructions. Eight to ten clones from two independent amplifications were chosen for DNA sequencing, using vector M13 primers.

### DNA analysis

Sequence alignments were performed using the Clustal X 2.0 software (Larkin et al., 2007). Comparison of DNA sequence homology was performed with BLAST from the National Centre for Biotechnology Information (<http://blast.ncbi.nlm.nih.gov>). Principal component analysis (PCA) and Fisher's Exact test was computed using SPSS 16.0 software (SPSS, United States).

### Precautions against contamination

Standard procedures were followed to prevent exogenous DNA contamination. Pre- and post-PCR experiments were conducted in separate buildings. Every operator wore facemasks, gloves, and lab coats. Pipettes

TABLE 1. PCR primers used for mtDNA and Y-DNA analysis in this study

Primer	Sequence	Length (bp)	Sites	Haplogroup
L16017	5'-TTCTCTGTTCTTTCATGGGGA-3'	235	HVSI	
H16251	5'-GGAGTTGCAGTTGATGTGTGA-3'			
L16201	5'-CAAGCAAGTACAGCAATCAAC-3'	209	HVSI	
H16409	5'-AGGATGGTGGTCAAGGGA-3'			
10400T	5'-TAATTATACAAAAAGGATTAGACTGTGCT-3'	149(M) 142(N)	10400T/C	M/N
10400C	5'-TACAAAAAGGATTAGACAGAACC-3'			
10400R	5'-GAAGTGAGATGGTAAATGCTAG-3'			
L648	5'-CCCACATCACCCATAAAACA-3'	136	663G	A
H745	5'-GCTTGATGCTTGTCCTTT-3'			
L8215	5'-ACAGTTTCATGCCCATCGTC-3'	121	8251A	W
H8297	5'-ATGCTAAGTTAGCTTTACAG-3'			
L4687	5'-CTATCCTCTTCAACAATATACTCT-3'	179	4715G	C
H4866	5'-ATGTGAGAAGAAGCAGGC-3'			
L 5159	5'-GCACCACGACCCTACTACTA-3'	166	5178A	D
H 5287	5'-GGGATGATGAGGCTATTGT-3'			
3970T	5'-TAAAATGTATTTCGGCTATGAAGATAA-3'	70(F)/66	3970T	F
3970C	5'-GTGTATTTCGGCTATGAAGATAG-3'			
3970R	5'-AGTCTCAGGCTTCAACATCG-3'			
L 4701	5'-CTATCCTCTTCAACAATATACTCT-3'	179	4833G	G
H 4848	5'-ATGTGAGAAGAAGCAGGC-3'			
L48	5'-CATTTGGTATTTTCGTCGGG-3'	158	152C	Z
H205	5'-CGCTTTGGTAAGTATGTTTCGC-3'			
L9787	5'-CTCAACATTTTGTAGCCAC-3'	161	9824C	M7
L9947	5'-CCACATCTACAAAATGCCAG-3'			
L10600	5'-CTACTCTCATAACCCTCAAC-3'	163	10646A	M10
H10762	5'-CATTGGAGTAGGTTTAGG-3'			
L12269	5'-TAACAACATGGCTTTCTCAACT-3'	132	12308G	U
H12357	5'-GAAGTCAGGGTTAGGGTGGT-3'			
XBamel-1	5'-CCTGGGCTCTGTAAAGAATAG-3'	103/109	AMG gene	Sex
XBamel-2	5'-CAGAGCTTAAACTGGGAAGCTG-3'			
	5'-TCACTTTTATATCCTCAACCA-3'	113	M216	C
	5'-AATCTGAATTCTGACACTGC-3'			
	5'-GCAAGTTTAGTGCCTCAGTATC-3'	117	M8	C1
	5'-TAAAGACCCCAAGCAAGAC-3'			
	5'-TGGCAATGGTATGTAGGC-3'	128	M38	C2
	5'-GCTGGCACATCTGTCATAA-3'			
	5'-ACTTGTGAAGGAGAATGAAA-3'	101	M217	C3
	5'-GCATTTGATAAAGCTGCTGTG-3'			
	5'-CCACAGAAGGATGCTGCTCA-3'	123	M89	F
	5'-CACACTTTGGGTCCAGGATCAC-3'			
	5'-GGACCCTGAAATACAGAAC-3'	117	M9	K
	5'-AAGCGCTACCTTACTTACAT-3'			
	5'-GGGTGTGGACTTTACGAAC-3'	125	M45	P
	5'-AAATCCTACTATCTCCTGGC-3'			
	5'-ACTGGAAAGAAAAGAATGCTG-3'	109	M214	NO
	5'-ATGGAAATGCCACTTCACTC-3'			
	5'-CCTGGAAAATGTGGGCTC-3'	133	M231	N
	5'-TTCTTTGACGATCTTTCCCC-3'			
	5'-GAACTGCCTCTTATAAAATCAT-3'	106	M128	N1a
	5'-ATCTACCTCTTCAAAGTGT-3'			
	5'-GCTACTTGGGAGGCTGAGG-3'	125	P43	N1b
	5'-ACGGAGTCTCGCTCTGTG-3'			
	5'-GGACTCTGAGTGTAGACTGTG-3'	122	TAT	N1c
	5'-GAGAAGGTGCCGTAAAAGTG-3'			

with aerosol-resistant tips were used. Negative extraction and PCR controls were conducted. DNA contamination removal solution (DNA-OFF™, Takara, Japan) and UV irradiation at 254 nm were used to treat equipment and benches. The mtDNA of the researchers were typed and compared with the results. The extraction and amplification of Y-SNP and Y-STR were only conducted by female researchers. To assess the authenticity of the results, at least two extractions and two amplifications were independently done at different times with different reagents for each sample. With the application of the authentication criteria, the possibility of contamination was minimized.

## RESULTS

### Authenticity of results

Strict precautions and controls were employed to prevent modern DNA contamination (see "Materials and Methods" section). The results from the clones were the same as that of direct sequencing (Table 2 and Supporting Information Fig. S1). Two samples re-tested in Research Center for Chinese Frontier Archaeology of our university for independent confirmation by another operator yielded consistent results (Table 2, Supporting Information Tables S1 and S2). The sequences of all researchers who came into contact with these samples



TABLE 2. HVSI polymorphic sites and haplogroups of mtDNA

No.	Mutations in HVSI (16,000+)	Coding region SNPs	Haplogroup	Sex <sup>a</sup>	Authenticity
1	223-292-311	10400C, 8251A	W	♂	Retested
2	223-295	10400T, 9824C	M7		
3	256-270	10400C, 12308G	U		Cloned
4	185-223-260	10400T, 152C	Z		
5	126-223-298-314-327	10400T, 4715 G	C		
6	093-169-183C-189-304	10400C, 3970T	F		
7	189-223-242-311	10400T, 5178A	D		
8	223-362	10400T, 5178A	D		Cloned
9	129-223-362	10400T, 5178A	D		
10	079-262-321-356	10400C, 12308G	U		
11	223-290-319-362	10400C, 663G	A		
12	127-223-362	10400T, 5178A	D		
13	051-129C-168-183C-189-362	10400C, 12308G	U		
14	223-290-293C-319-329	10400C, 663G	A		Cloned
15	185-223-260-298-380	10400T, 152C	Z		
16	129-193-223-311-357	10400T, 5178A	D		
17	148-223-239-290-319-362-376	10400C, 663G	A		
18	223-227-278-362	10400T, 4833 G	G		Cloned
19	223-245-362	10400T, 5178A	D		
20	223-265-362-378	10400T, 5178A	D		
21	239-256-270-321-377	10400C, 12308G	U		
22	223-278-362	10400T, 4833 G	G		
23	223-256-319	10400T, 5178A	D		
24	223-362	10400T, 5178A	D		Retested
25	185-223-260-362	10400T, 152C	Z		
26	189-223-311-362	10400T, 5178A	D		
27	223-362	10400T, 5178A	D		
28	129-150-223-298-327	10400T, 4715G	C		
29	129-223-311	10400T, 10646A	M10		
R1 <sup>b</sup>	126-294-296-304				
R2	126-174-223-311-362				
R3	189-261-278-311-362				

<sup>a</sup> – indicates that the sex of the sample was not identified.

<sup>b</sup> R represents researcher.

were typed, and the results showed that none of them were matches for the samples (Table 2).

**mtDNA haplogroup identification and distribution**

The 29 mtDNA sequences were successfully amplified from positions 16,035–16,409. The sequence data was submitted to the GenBank, and the corresponding accession numbers were granted to KM035805–KM035833. Twenty-seven mtDNA haplotypes were obtained, as represented in Table 2. Ten haplogroups were found in the samples from the Tianshanbeilu population and of these, two (U and W) are generally considered West Eurasian lineages and are commonly found in the West. The other eight haplogroups are A, C, D, F, G, Z, M7, and M10, which are found in East Eurasian lineages. The haplogroup distribution in the Tianshanbeilu population is A (10.3%), C (6.9%), D (37.9%), F(3.4%), G (6.9%), Z (10.3%), M7 (3.4%), M10 (3.4%), U (13.8%), and W (3.4%), which indicates the population is composed of a mix of East Eurasian (A, C, D, F, G, Z, M7, and M10, 82.8%) and West Eurasian (U and W, 17.2%) lineages.

**Y chromosome SNP and STR analysis**

Thirteen of the twenty-nine ancient samples were identified as male when the sexually dimorphic amelogenin fragment was typed. We used Y chromosome SNP primers and of the male samples, seven failed to amplify. Five male individuals were assigned to N1-M231 (Table 3). Because ancient samples have such short

amplicons, it is not possible to type the diagnostic site P43 (sub-haplogroup N1b) on these N1 samples, so samples that yielded negative M128 and TAT mutations were defined as N1 (xN1a, N1c). One sample was determined to belong to Y haplogroup C\*, after screening M216, M8, M38, and M217. The six male samples successfully assigned to haplogroups were further analyzed at 17 Y chromosome STR loci. Of the six individuals, only three of them were successfully screened at three or more of the 17 loci in two independent extractions. Consensus data are reported in Table 4.

**Comparison with the extant and ancient populations of Eurasia**

In order to identify the genetic relationship of the Tianshanbeilu population to other Eurasian Neolithic and Bronze Age ancient and present day populations, mtDNA haplogroup distributions were compared using PCA (Supporting Information Table S3). The PCA plot of the first two components (51.58% of the total variance, Fig. 2) shows that present day populations largely segregate into three main clusters: Central Asians, North Asians, and East Asians. The North Asians cluster in the upper left corner, while East Asians, including the Han and TB populations, cluster in the lower left corner of the plot. The middle and lower parts of the PCA plot contain the Central Asians. The Tianshanbeilu population occupies a position between the North and East Asians.

TABLE 3. Y-SNP and Y haplogroup data for six male samples

No.	Y-SNP <sup>a</sup>										Haplogroup		
	M216 C->T	M8 G->T	M38 T->G	M89 C->T	M217 A->C	M9 C->G	M45 G->A	M214 T->C	M231 G->A	M128 -2bp		P43 G->A	TAT T->C
1	C	-	-	T	-	G	G	C	A	+2bP	×	T	NI(xN1a, N1c)
17	C	-	-	T	-	G	G	C	A	+2bP	×	T	NI(xN1a, N1c)
19	C	-	-	T	-	G	G	C	A	+2bP	×	T	NI(xN1a, N1c)
25	C	-	-	T	-	G	G	C	A	+2bP	×	T	NI(xN1a, N1c)
28	T	G	T	-	-	-	-	-	-	-	-	-	C*
29	C	-	-	T	-	G	G	C	A	+2bP	×	T	NI(xN1a, N1c)

<sup>a</sup> - represents a SNP site that was not tested and × represents sites that failed to be detected.

DISCUSSION

Maternal origin of the Tianshanbeilu population

The Tianshanbeilu site is adjacent to the Eurasian steppe zone, and the Hexi Corridor is in an important area of cultural contact between East and West Eurasia. It is, thus, important to understand the origin and migration history of populations in this region. The archaeological research suggests that characteristics of both the Eurasian steppe cultures and the Di-qiang cultures can be observed in the Tianshanbeilu Culture. Two hypotheses were put forward to explain this phenomenon (Li, 2009). In both hypotheses, Eurasian steppe populations of Siberia migrated south into the eastern part of Xinjiang, where the Di-qiang Cultures moved to as well in the early Bronze Age. In the first hypothesis, these two populations did not admix, and the emerging Siba and pre-Siba Cultures in this region were a result of cultural diffusion from the Di-qiang Cultures to the Eurasian steppe populations. In this case, the genetic composition of the Tianshanbeilu population should include only lineages that are found in early Bronze Age Siberia. In the second hypothesis, these two populations did admix, and the emerging cultures in this region results from population admixture. In this case, both prehistoric Siberian and East Asian lineages should be found in the Tianshanbeilu population.

A morphological study of the Tianshanbeilu samples suggests they are a mixture of East Asian and European populations (Wei et al., 2012). The mtDNA haplogroups of the Tianshanbeilu individuals also suggest there was an admixture of East and West Eurasian lineages. The East Eurasian matrilineages, such as A, C, D, F, G, Z, M7, and M10, have been detected in the Tianshanbeilu population. Haplogroups C and D (10.3% and 37.9%) are the most common lineages found in extant Central, North, and East Asians (Quintana-Murci et al., 2004; Tanaka et al., 2004; Wen et al., 2004a; Derenko et al., 2007). These two haplogroups were prevalent in the Neolithic or Bronze Age populations in Siberia and the Hexi Corridor (Table 5). Haplogroups A and F are widely distributed in present day North and East Asians, with relatively high frequencies in the Han and TB populations (Tanaka et al., 2004; Wen et al., 2004a, 2004b; Derenko et al., 2007). Haplogroups G and Z are found throughout extant Central and East Eurasia at low levels (Quintana-Murci et al., 2004; Tanaka et al., 2004; Wen et al., 2004a). These four haplogroups are also found in prehistoric Siberians and Di-qiang populations (Table 5). This suggests that Siberians and Di-qiang populations are two candidate source populations for the Tianshanbeilu population. M7, a common haplogroup in the Han and TB populations, is rarely found in other parts of Eurasia (Quintana-Murci et al., 2004; Tanaka et al., 2004; Wen et al., 2004a, 2004b; Derenko et al., 2007). The Neolithic Qijia and Yangshao populations from the Yellow River valley near the Hexi Corridor also possess the M7 lineage (Table 5). Though no mtDNA data from the Siba and pre-Siba populations are available, the fact that haplogroup M7 is found in the Tianshanbeilu population, the Qijia population, and the Yangshao population indicates the close genetic relationship between the Tianshanbeilu population and the Di-qiang populations. Haplogroup M10 has very a low frequency in Central Asia and South Siberia, is mainly distributed in East Asia, has its highest frequency in the TB populations, and has the largest diversity in China

TABLE 4. 17 loci of Y STR data from three male samples

No.	DYS 391	DYS 389I	DYS 439	DYS 389II	DYS 438	DYS 456	DYS 458	DYS 437	DYS 635	DYS 448	H4	DYS 447	DYS 19	DYS 392	DYS 393	DYS 390	DYS 385
1	10	–	13	–	–	16	16	–	–	–	13	22	14	–	12	–	–
19	11	14	8, 13	26	–	18	16	13	22	19	13	22	18	13	13	24	11,13,2
28	10	13	10	–	–	16	16	–	–	–	12	28	–	–	13	–	–

– indicates that the loci of the sample was not identified.

(Tanaka et al., 2004). Haplogroup M10 can be found in the two Neolithic Di-qiang populations from the Hexi Corridor, the Lajia and Mogou populations (Table 5). This haplogroup is also found in the Yangshao population. Therefore, the distribution of M7 and M10 in extant and ancient populations suggests that the Di-qiang populations contributed to the formation of the Tianshanbeilu population. In addition, among the twenty two East Eurasian haplotypes identified in the twenty nine Tianshanbeilu samples, fifteen haplotypes were a perfect or near perfect match to sequences from the Han or TB populations in NCBI Genbank, and ten haplotypes had the same sequence as the ancient Di-qiang populations (Table 6). These results suggest that part of the Tianshanbeilu population derives from the Di-qiang populations.

The West Eurasian lineages identified in the Tianshanbeilu population are W and U. U is one of the oldest European haplogroups (Torroni et al., 1996), and it is also widely distributed in southern Siberia and Central Asia in the early period of the Neolithic (Table 5). Haplogroup W appears in Europe, Siberia, and Southwest Asia (Torroni et al., 1996; Metspalu et al., 2004; Derenko et al., 2007). To explore the origins of these West Eurasian lineages, the U and W haplotypes found in the Tianshanbeilu site were compared with the Eurasian haplotypes found at other Neolithic and Bronze Age sites. We detected four samples from the Tianshanbeilu site with the U haplotype. The haplotype 16,256–16,270 was also found in three Neolithic Southern Siberians (Mooder et al., 2006) and could be linked to the haplogroup U5a. No matching sequences for the haplotype 16,239–16,256–16,270–16,321–16,377 have been found. This haplotype could potentially also be classified as haplogroup U5a. Besides Siberia, U5a is also present in Europe and Central Asia in the Neolithic and Bronze Age (Lalueza-Fox et al., 2004; Brandt et al., 2013). The haplotype 16,079–16,262–16,321–16,356 can be assigned to haplogroup U4 according to the mutation found at position 16,356. The haplogroup U4 was found in ancient European and Southern Siberian populations (Keyser et al., 2009; Brandt et al., 2013). The haplotype 16,051–16,129C–16,168–16,183C–16,189–16,362 has a two-step neighbor, detected in the Neolithic Southern Siberians (Keyser et al., 2009), and this haplotype could be classified as the haplogroup U2e, which is found in prehistoric Europeans and Southern Siberians (Keyser et al., 2009; Brandt et al., 2013). The haplotype 16,223–16,292–16,311 was assigned to the haplogroup W. A one-step neighbor of this haplotype was found in the Bronze/Iron Kazakh samples (Lalueza-Fox et al., 2004). These results combined with the archaeological studies (Lin, 2003; Li, 2009) indicate that the Southern Siberians may be a major source population for the West Eurasian lineages of the Tianshanbeilu population.

The analysis of the mtDNA haplogroups and haplotypes of the Tianshanbeilu population reveals that the

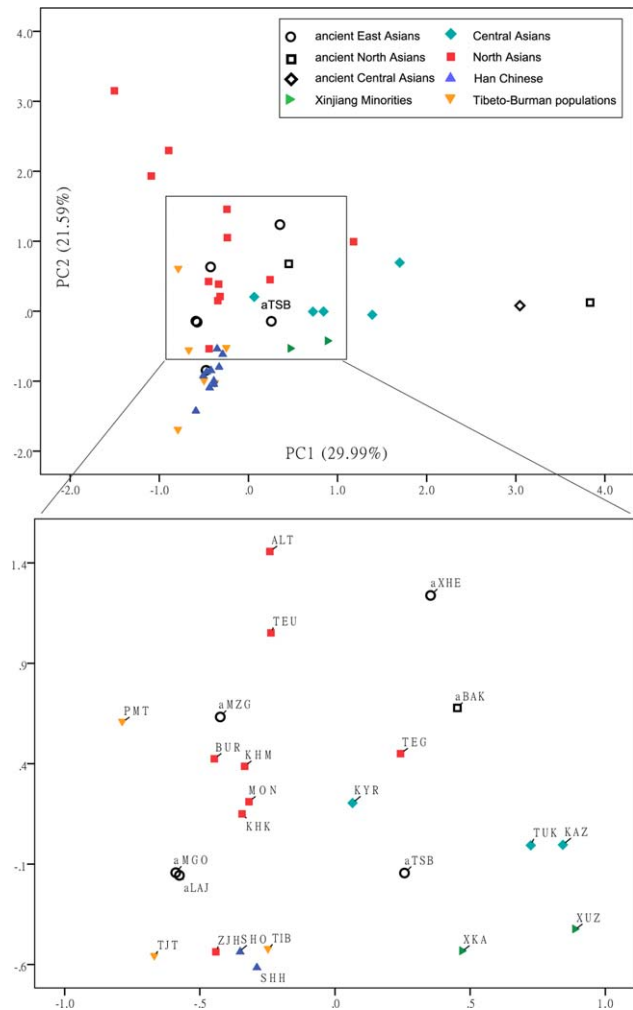


Fig. 2. PCA of mtDNA haplogroup frequencies of the Tianshanbeilu population and other Eurasian populations. The first two dimensions account for 51.58% of the total variance. The labels of ancient and extant populations were listed in Supporting Information Table S3. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

composition of this ancient population was a mixture of the Di-qiang and Eurasian steppe populations. On the PCA plot, the Tianshanbeilu population occupied a position between the North and East Asians, which may also indicate mixed origins for this population. Using Fisher's Exact test, we show that the Tianshanbeilu population is significantly different from the extant and ancient North Asians ( $P < 0.05$ , Table 7). However, there seems to be no significant differences between the Tianshanbeilu and ancient Di-qiang populations ( $P > 0.05$ , Table 7). These results suggest that the second

TABLE 5. *mt-DNA haplogroup distributions in regions near the Tianshanbeilu site from the Bronze Age or earlier*

Region	Number	Age	Haplogroup
Kazakhstan <sup>a</sup>	13	Bronze Age 14th–10th BC	H, HV, T, U, W
Mongolia, Altai <sup>b</sup>	3	Bronze Age	D
Gorny, Altai <sup>c</sup>	4	Neolithic and Bronze Age	West Eurasian lineages
South Siberia <sup>d</sup>	11	Bronze Age 19th–9th BC	U, UK, H, T, Z
Lake Baikal, South Siberia <sup>e</sup>	27	Neolithic	A, C, D, G, F, U
Northeastern Siberia <sup>f</sup>	1	Neolithic	C
Lajia, Hexi Corridor <sup>g</sup>	14	Qijia, Late Neolithic 3,800 BP	A, B, C, D, M10
Mogou, Hexi Corridor <sup>h</sup>	36	Qijia, Late Neolithic 4,000 BP	A, B, C, D, F, G, Z, M7, M8, M10, N9
Miaozigou, Inner Mongolia <sup>i</sup>	6	Yangshao, Neolithic	A, C, D, M9, M10
Xiaohu, Xinjiang <sup>j</sup>	33	Andronovo, Bronze Age	C, M*, R*, H, K
Tianshanbeilu, Xinjiang <sup>j</sup>	29	Tianshanbeilu, Bronze Age	A, C, D, F, G, Z, M7, M10, U, W

<sup>a</sup> Lalueza-Fox et al., 2004.

<sup>b</sup> Gonzalez-Ruiz et al., 2012.

<sup>c</sup> Chikisheva et al., 2007.

<sup>d</sup> Keyser et al., 2009.

<sup>e</sup> Mooder et al., 2006.

<sup>f</sup> Ricaut et al., 2005.

<sup>g</sup> Gao et al., 2007.

<sup>h</sup> Our unpublished data.

<sup>i</sup> Li et al., 2010.

<sup>j</sup> This study.

TABLE 6. *Shared sequences in Han and TB populations and ancient Di-qiang populations*

Haplotype	Number of shared sequences in Genbank	Number of shared sequences with Di-qiang populations
223-295	Chinese Han (12)	Qijia populations (1) <sup>a</sup>
185-223-260	Chinese Han (6)	TB populations (7)
223-362	Chinese Han (11)	TB populations (7)
129-223-362	Chinese Han (14)	Qijia populations (3)
223-290-319-362	Chinese Han (16)	Qijia populations (1)
127-223-362	Chinese Han (9) <sup>a</sup>	Qijia populations (1) <sup>a</sup>
223-290-293C-319-329	Chinese Han (1) <sup>a</sup>	TB populations (7) <sup>a</sup>
185-223-260-298-380	Chinese Han (11)	Qijia populations (1) <sup>a</sup>
223-227-278-362	Chinese Han (1)	TB populations (9)
223-278-362	Chinese Han (5)	Qijia populations (1) <sup>a</sup>
223-256-319	Chinese Han (1) <sup>a</sup>	TB populations (3)
185-223-260-362	Chinese Han (1) <sup>a</sup>	TB populations (23) <sup>a</sup>
189-223-311-362	Chinese Han (1)	TB populations (23) <sup>a</sup>
129-150-223-298-327	Chinese Han (3)	Qijia populations (1) <sup>a</sup>
129-223-311	Chinese Han (2)	TB populations (3)
		TB populations (1)
		Qijia populations (2) <sup>a</sup>
		Qijia populations (3) <sup>a</sup>

<sup>a</sup> Indicates one-step neighbors of the haplotype.

hypothesis of admixture between the Di-qiang and Eurasian steppe populations is more likely than the first hypothesis of no admixture and only cultural diffusion. According to archaeological records, the Di-qiang populations engaged in cereal agriculture since the Neolithic. Millet was one of the cereals that the Di-qiang populations planted (Li, 1993). Stable isotope dietary analysis of the Tianshanbeilu populations revealed that the diet of this Bronze Age population was a mix of C3 plants (53.7%–72.3%, mainly wheat) and C4 plants (27.7%–46.3%, mainly millet) (Zhang et al., 2010). The report by Svyatko et al. (2013) indicates that the diet of Neolithic to Middle Bronze Age populations in South Siberia were primarily based on C3 plants, with C4 plants only becoming an important component of the diet in the Late Bronze Age (Svyatko et al., 2013). This leads us to infer that the Di-qiang populations might have moved to the eastern part of Xinjiang and transmitted millet to this region. Archaeological studies also show that after experiencing a period of development, the Siba Culture declined and did not continue in the Hexi Corridor. This might be due to population migration (Li et al., 2010b).

Above all, the archaeological, anthropological, and genetic evidence suggest that the ancient Di-qiang populations of the pre-Siba and Siba cultures that lived in the Hexi Corridor had immigrated into the eastern part of Xinjiang in the early Bronze Age. These results support the latter hypothesis that the presence of pre-Siba and Siba cultural elements in the Tianshanbeilu Culture was a product of demic diffusion.

### Paternal origin of the Tianshanbeilu population

Six of the thirteen male samples of the Tianshanbeilu site were successfully assigned to haplogroup C\* and N1 (xN1a, N1c), each with a frequency of 16.7% and 83.4%, respectively. C\* is prevalent in extant Siberians and in regions north of East Asia (Pakendorf et al., 2006; Xue et al., 2006). N1 (xN1a, N1c) is also common in Siberians and widely distributed in the northern areas of East Asia (Xue et al., 2006). Haplotype N1 (xN1a, N1c) was detected in the Neolithic samples of the West Liao River Valley, Northeast China (Cui et al., 2013). It is noteworthy that in samples from the Yangshao population from



TABLE 7. The results of Fisher's Exact tests between the Tianshanbeilu population and other Eurasian populations

Label <sup>a</sup>	P-value <sup>b</sup>	Label	P-value
UZB	0.000	GDH	0.000
TUK	0.000	JSH	0.001
TAJ	0.000	SHH	0.015
KAZ	0.145	SCH	0.000
KYR	0.042	YNH	0.000
MON	N/A	ZJH	0.000
YAT	0.000	BAT	0.000
ALT	0.000	LHT	0.000
EVE	0.000	TIB	0.000
BUR	N/A	TJT	0.000
TUV	N/A	YIT	0.000
TUB	0.000	XKA	0.340
SHO	0.000	XUZ	0.304
KHM	0.030	aKAZ	0.000
TEG	0.023	aBAK	0.002
KHK	0.000	aKUR	0.004
TEU	0.008	aMGO	0.312
LNH	0.002	aLAJ	0.167
SDH	0.001	aXHE	0.002
SXH	0.008	aDDZ	0.458
FJH	0.000	aMZG	0.145

<sup>a</sup>The labels of the populations are consistent with those listed in Supporting Information Table S3.

<sup>b</sup>N/A represents the P-value cannot be computed.

the Yellow River valley, the identified haplogroups of Y-DNA were all classified as N1 (xN1a, N1c). Although we lack Y-DNA data for the Neolithic and Bronze Age in the Hexi Corridor, N1 (xN1a, N1c) was found in both the Tianshanbeilu and the Yangshao samples in high frequency, which is consistent with the mtDNA results in suggesting that the Di-qiang population of the Hexi Corridor might have migrated to the eastern part of Xinjiang.

## CONCLUSIONS

The present study reveals that the maternal lineages of the Tianshanbeilu population contained matrilineal lines from both East and West Eurasia. The phylogeographic analysis of the mtDNA haplogroups, combined with the archaeological data, suggests that the Di-qiang populations living in the Hexi Corridor migrated to eastern Xinjiang and mixed with the Eurasian steppe populations that settled there in the early Bronze Age; these conclusions are further supported by the results from Y-DNA.

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## LITERATURE CITED

Brandt G, Haak W, Adler CJ, Roth C, Szécsényi-Nagy A, Karimnia S, Möller-Rieker S, Meller H, Ganslmeier R, Friederich S, Dresely V, Nicklisch N, Pickrell JK, Sirocko F, Reich D, Cooper A, Alt KW; Genographic Consortium. 2013. Ancient DNA reveals key stages in the formation of central

European mitochondrial genetic diversity. *Science* 342:257–261.

Chen KT, Hiebert FT. 1995. The late prehistory of Xinjiang in relation to its neighbors. *J World Prehist* 9:243–300.

Chikisheva T, Gubina M, Kulikov I, Karafet T, Voevoda M, Romaschenko A. 2007. A paleogenetic study of the prehistoric populations of the Altai. *Archaeol Ethnol Anthropol Eurasia* 32:130–142.

Comas D, Calafell F, Mateu E, Pérez-Lezaun A, Bosch E, Martínez-Arias R, Clarimon J, Faccioli F, Fiori G, Luiselli D, Pettener D, Bertranpetit J. 1998. Trading genes along the silk road: mtDNA sequences and the origin of central Asian-populations. *Am J Hum Genet* 63:1824–1838.

Comas D, Plaza S, Wells RS, Yuldaseva N, Lao O, Calafell F, Bertranpetit J. 2004. Admixture, migrations, and dispersals in Central Asia: evidence from maternal DNA lineages. *Eur J Hum Genet* 12:495–504.

Cui YQ, Gao SZ, Xie CZ, Zhang QC, Wang HJ, Zhu H, Zhou H. 2009. Analysis of the matrilineal genetic structure of population in the early Iron Age from Tarim Basin., Xinjiang, China. *Chinese Sci Bull* 54:3916–3923.

Cui YQ, Li CX, Gao SZ, Xie CZ, Zhou H. 2010. Early Eurasian migration traces in the tarim basin revealed by mtDNA polymorphisms. *Am J Phys Anthropol* 142:558–564.

Cui YQ, Li HJ, Ning C, Zhang Y, Chen L, Zhao X, Hagelberg E, Zhou H. 2013. Y Chromosome analysis of prehistoric human populations in the West Liao River Valley, Northeast China. *BMC Evol Biol* 13:216.

Derenko MV, Grzybowski T, Malyarchuk BA, Dambueva IK, Denisova GA, Czarny J, Dorzhu CM, Kakkapov VT, Miščicka-Sliwka D, Woźniak M, Zakharov IA. 2003. Diversity of mitochondrial DNA lineages in South Siberia. *Ann Hum Genet* 67:391–411.

Derenko M, Malyarchuk B, Grzybowski T, Denisova G, Dambueva I, Perkova M, Dorzhu C, Luzina F, Lee HK, Vanecsek T, Villems R, Zakharov I. 2007. Phylogeographic analysis of mitochondrial DNA in northern Asian Populations. *Am J Hum Genet* 81:1025–1041.

Ding XL. 1996. The report of the carbon dating (33). *Archaeology* 7:66–70.

Gao SZ, Yang YD, Xu Y, Zhang QC, Zhu H, Zhou H. 2007. Tracing the genetic history of the Chinese people: mitochondrial DNA analysis of a neolithic population from the Lajia Site. *Am J Phys Anthropol* 133:1128–1136.

Gao SZ, Cui YQ, Yang YD, Duan RH, Abuduresule I, Mair VH, Zhu H, Zhou H. 2008. Mitochondrial DNA analysis of human remains from the Yuansha site in Xinjiang, China. *Sci China Ser C* 51:205–213.

Gokcumen O, Dulik MC, Pai AA, Zhadanov SI, Rubinstein S, Osipova LP, Andreenkov OV, Tabikhanova LE, Gubina MA, Labuda D, Schurr TG. 2008. Genetic variation in the enigmatic Altaian Kazakhs of south-central Russia: insights into Turkic population history. *Am J Phys Anthropol* 136:278–293.

Gonzalez-Ruiz M, Santos C, Jordana X, Simon M, Lalueza-Fox C, Gigli E, Aluja MP, Malgosa A. 2012. Tracing the origin of the East-West population admixture in the Altai region (Central Asia). *Plos One* 7:e48904.

Han JY. 2007. Cultures in Xinjiang from the bronze age to the early iron age. Beijing: Cultural Relics Press. p 40–90.

Han KX. 1991. Anthropological research of ancient populations in Xinjiang and physical characteristics of Uigur. *West Region Res* 2:1–13.

Han KX. 1998. The physical anthropology of the ancient populations of the Tarim Basin and surrounding areas. In: Mair VH, editor. The bronze age and early iron age peoples of eastern central Asia. Washington, DC: Institute for the Study of Man, Inc. in collaboration with the University of Pennsylvania Museum Publications. p 558–570.

Karafet TM, Mendez FL, Meilerman MB, Underhill PA, Zegura SL, Hammer MF. 2008. New binary polymorphisms reshape and increase resolution of the human Y chromosomal haplogroup tree. *Genome Res* 18:830–838.

Keyser C, Bouakaze C, Crubezy E, Nikolaev VG, Montagnon D, Reis T, Ludes B. 2009. Ancient DNA provides new insights

- into the history of south Siberian Kurgan people. *Hum Genet* 126:395–410.
- Kivisild T, Tolk HV, Parik J, Wang YM, Papiha SS, Bandelt HJ, Villems R. 2002. The emerging limbs and twigs of the East Asian mtDNA tree. *Mol Biol Evol* 19:1737–1751.
- Lalueza-Fox C, Sampietro ML, Gilbert MTP, Castri L, Facchini F, Pettener D, Bertranpetit J. 2004. Unravelling migrations in the steppe: mitochondrial DNA sequences from ancient Central Asians. *Proc R Soc Lond B* 271:941–947.
- Larkin MA, Blackshields G, Brown NP, Chenna R, McGettigan PA, McWilliam H, Valentin F, Wallace IM, Wilm A, Lopez R, Thompson JD, Gibson TJ, Higgins DG. 2007. Clustal W and Clustal X version 2.0. *Bioinformatics* 23:2947–2948.
- Li SC. 1993. The study of the Siba Culture. In: Qi S, editor. *Archaeological culture (three)*. Beijing: Cultural Relics Press. p 80–121.
- Li SC. 2002. Interaction between Northwest China and Central Asia during the second millennium BC: an archaeological perspective. In: Boyle K, Renfrew C, Levine M, editors. *Ancient interactions: east and west in Eurasian Steppe*. Cambridge, England: McDonald Institute Monographs, University of Cambridge. p 171–180.
- Li SC. 2009. The archaeological analysis of the Tianshanbeilu site. In: Yu W, editor. *School of Archaeology and Museology PU*. Beijing: Cultural Relics Press. p 193–202.
- Li CX, Li HJ, Cui YQ, Xie CZ, Cai DW, Li WY, Mair VH, Xu Z, Zhang QC, Abuduresule I, Jin L, Zhu H, Zhou H. 2010a. Evidence that a West-East admixed population lived in the Tarim Basin as early as the early Bronze age. *BMC Biol* 8:15.
- Li SC, Shui T, Wang H. 2010b. Report on prehistoric archaeological survey in Hexi Corridor. *Acta Archaeol Sin* 2:229–264.
- Li HJ, Zhao X, Zhao YB, Li CX, Si DY, Zhou H, Cui YQ. 2011. Genetic characteristics and migration history of a bronze culture population in the West Liao-River valley revealed by ancient DNA. *J Hum Genet* 56:815–822.
- Lin Y. 2001. The transport routes in Xinjiang before the building of the Silk Road. *Steppe Cultural Relics* 1:57–64.
- Lin MC. 2003. Origin and migration of Tocharian. *West Region Res* 3:9–23.
- Liu SB, Cui YQ, Xie CZ, Zhu H, Zhou H. 2004. Sex identification of archaeological human remains based on amplification of AMG gene via fluorescent-labeled primers. *J Jilin Univ: Sci Ed* 1:139–142.
- Mei JJ. 2000. Copper and bronze metallurgy in late prehistoric Xinjiang: its cultural context and relationship with neighbouring regions. Oxford: British Archaeological Reports Ltd.
- Metspalu M, Kivisild T, Metspalu E, Parik J, Hudjashov G, Kaldma K, Serk P, Karmin M, Behar DM, Gilbert MT, Endicott P, Mastana S, Papiha SS, Skorecki K, Torroni A, Villems R. 2004. Most of the extant mtDNA boundaries in South and Southwest Asia were likely shaped during the initial settlement of Eurasia by anatomically modern humans. *BMC Genet* 5:26.
- Mooder KP, Schurr TG, Bamforth FJ, Bazahiski VI, Savel'ev NA. 2006. Population affinities of Neolithic Siberians: a snapshot from prehistoric Lake Baikal. *Am J Phys Anthropol* 129:349–361.
- Pakendorf B, Novgorodov IN, Osakovskij VL, Danilova AP, Protod'jakonov AP, Stoneking M. 2006. Investigating the effects of prehistoric migrations in Siberia: genetic variation and the origins of Yakuts. *Hum Genet* 120:334–353.
- Qian YP, Chu ZT, Dai Q, Wei CD, Chu JY, Tajima A, Horai S. 2001. Mitochondrial DNA polymorphisms in Yunnan nationalities in China. *J Hum Genet* 46:211–220.
- Quintana-Murci L, Chaix R, Wells RS, Behar DM, Sayar H, Scozzari R, Rengo C, Al-Zahery N, Semino O, Santachiara-Benerecetti AS, Coppa A, Ayub Q, Mohyuddin A, Tyler-Smith C, Qasim Mehdi S, Torroni A, McElreavey K. 2004. Where west meets east: the complex mtDNA landscape of the southwest and Central Asian corridor. *Am J Hum Genet* 74:827–845.
- Ricaud FX, Fedoseeva A, Keyser-Tracqui C, Crubezy E, Ludes B. 2005. Ancient DNA analysis of human neolithic remains found in northeastern Siberia. *Am J Phys Anthropol* 126:458–462.
- Shinoda K, Adachi N, Guillen S, Shimada I. 2006. Mitochondrial DNA analysis of ancient Peruvian highlanders. *Am J Phys Anthropol* 131:98–107.
- Starikovskaya EB, Sukernik RI, Derbeneva OA, Volodko NV, Ruiz-Pesini E, Torroni A, Brown MD, Lott MT, Hosseini SH, Huoponen K, Wallace DC. 2005. Mitochondrial DNA diversity in indigenous populations of the southern extent of Siberia, and the origins of Native American haplogroups. *Ann Hum Genet* 69:67–89.
- Svyatko SV, Schulting RJ, Mallory J, Murphy EM, Reimner PJ, Khartanovich VI, Chistov YK, Sablin MV. 2013. Stable isotope dietary analysis of prehistoric populations from the Minusinsk Basin, Southern Siberia, Russia: a new chronological framework for the introduction of millet to the eastern Eurasian steppe. *J Archaeol Sci* 40:3936–3945.
- Tanaka M, Cabrera VM, Gonzalez AM, Larruga JM, Takeyasu T, Fuku N, Guo LJ, Hirose R, Fujita Y, Kurata M, Shinoda K, Umetsu K, Yamada Y, Oshida Y, Sato Y, Hattori N, Mizuno Y, Arai Y, Hirose N, Ohta S, Ogawa O, Tanaka Y, Kawamori R, Shimoto-Nagai M, Maruyama W, Shimokata H, Suzuki R, Shimodaira H. 2004. Mitochondrial genome variation in Eastern Asia and the peopling of Japan. *Genome Res* 14:1832–1850.
- Torroni A, Huoponen K, Francalacci P, Petrozzi M, Morelli L, Scozzari R, Obinu D, Savontaus ML, Wallace DC. 1996. Classification of European mtDNAs from an analysis of three European populations. *Genetics* 144:1835–1850.
- Underhill PA, Shen PD, Lin AA, Jin L, Passarino G, Yang WH, Kauffman E, Bonne-Tamir B, Bertranpetit J, Francalacci P, Ibrahim M, Jenkins T, Kidd JR, Mehdi SQ, Seielstad MT, Wells RS, Piazza A, Davis RW, Feldman MW, Cavalli-Sforza LL, Oefner PJ. 2000. Y chromosome sequence variation and the history of human populations. *Nat Genet* 26:358–361.
- Underhill PA, Passarino G, Lin AA, Shen P, Lahr MM, Foley RA, Oefner PJ, and Cavalli-Sforza LL. 2001. The phylogeography of Y chromosome binary haplotypes and the origins of modern human populations. *Ann Hum Genet* 65:43–62.
- Wei D, Zhao YS, Chang XE, Zhu H. 2012. Craniometric variation of ancient skulls from the Hami Tianshan North Road Cemetery. *Acta Anthropol Sin* 31:395–406.
- Wen B, Li H, Lu DR, Song XF, Zhang F, He YG, Li F, Gao Y, Mao XY, Zhang L, Qian J, Tan J, Jin J, Huang W, Deka R, Su B, Chakraborty R, Jin L. 2004a. Genetic evidence supports demic diffusion of Han culture. *Nature* 431:302–305.
- Wen B, Xie XH, Gao S, Li H, Shi H, Song XF, Qian TZ, Xiao CJ, Jin JZ, Su B, Lu D, Chakraborty R, Jin L. 2004b. Analyses of genetic structure of Tibeto-Burman populations reveals sex-biased admixture in southern Tibeto-Burmans. *Am J Hum Genet* 74:856–865.
- Xue Y, Zerjal T, Bao W, Zhu S, Shu Q, Xu J, Du R, Fu S, Li P, Hurler ME, Yang H, Tyler-Smith C. 2006. Male demography in East Asia: a north–south contrast in human population expansion times. *Genetics* 172:2431–2439.
- Yan WM. 1978. The origin of the ancient painted pottery found in Gansu. *Cultural Relics* 10:62–76.
- Yang SM, Ding H. 2003. *The ethnography of China*. Beijing: Central University for Nationalities Press.
- Yao YG, Zhang YP. 2002. Phylogeographic analysis of mtDNA variation in four ethnic populations from Yunnan Province: new data and a reappraisal. *J Hum Genet* 47:311–318.
- Yao YG, Kong QP, Bandelt HJ, Kivisild T, Zhang YP. 2002a. Phylogeographic differentiation of mitochondrial DNA in Han Chinese. *Am J Hum Genet* 70:635–651.
- Yao YG, Nie L, Harpending H, Fu YX, Yuan ZG, Zhang YP. 2002b. Genetic relationship of Chinese ethnic populations revealed by mtDNA sequence diversity. *Am J Phys Anthropol* 118:63–76.
- Yao YG, Kong QP, Man XY, Bandelt HJ, Zhang YP. 2003. Reconstructing the evolutionary history of China: a caveat about inferences drawn from ancient DNA. *Mol Biol Evol* 20:214–219.
- Yao YG, Kong QP, Wang CY, Zhu CL, Zhang YP. 2004. Different matrilineal contributions to genetic structure of ethnic groups in the Silk Road region in China. *Mol Biol Evol* 21:2265–2280.
- Zhang QC, Chang XE, Liu GR. 2010. Stable isotopic analysis of human bones unearthed from Tianshan Beilu Cemetery in Hami, Xinjiang. *The Western Regions Studies* 2:38–43.