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基于SSR标记的中亚生态区核桃（*Juglans regia* L.）遗传多样性与种群结构分析

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摘 要：【目的】从分子水平探究中国新疆与吉尔吉斯斯坦和塔吉克斯坦2个中亚国家共6个区域核桃种质资源间的遗传多样性及亲缘关系。**【方法】**采用SSR分子标记方法对采自6个区域的137份核桃种质资源进行遗传多样性和居群遗传结构分析。**【结果】**利用14对引物共检测到130个等位变异位点，变异范围为6~13，平均每对SSR引物可检测到9.285 7个等位位点；14个SSR位点的占比范围为0.625 7~0.865 1，平均值0.763 4；平均Shannon's多样性指数（*I*）为0.459 8；种群总基因多样性（*Ht*）平均值为3.248 1；种群内基因多样性（*Hs*）平均值为2.108 9；种群间的遗传分化指数（*Gst*）范围为0.057 8~0.406 8，平均值为0.259 5；基因流（*Nm*）平均值为3.017 4；各个等位基因频率整体分布不均匀。UPGMA聚类结果显示，137份供试材料在遗传相似性系数为0.75时分成3个类群。其中，第一类群共42份，包括中国新疆阿克苏地区16份，中国新疆和田地区21份，吉尔吉斯斯坦5份；第二类群共44份，包括中国新疆伊犁地区12份、吉尔吉斯斯坦32份；第三类群共51份，包括中国新疆喀什地区19份、塔吉克斯坦13份和中国新疆伊犁地区19份。聚类结果在一定程度上表明了居群间亲缘关系的距离，总体分布与地理分布有关，不同区域之间存在一定的基因交换。聚类结果与居群间遗传距离和遗传一致度结果一致。**【结论】**SSR分子标记结果表明，中亚生态区不同地理分布的核桃种质资源遗传多样性存在差异，为揭示中亚生态区核桃种群结构及传播途径提供了依据。

关键词：核桃；种质资源；SSR；遗传多样性；遗传结构

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Analysis of genetic diversity and population structure of walnut (*Juglans regia* L.) in Central Asia ecological region based on SSR markers

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Abstract:【Objective】In the distribution areas of walnut（*Juglans regia* L.）in Central Asia, researchers in different countries have studied the genetic diversity of walnut in their own countries. However, there have been few studies on the germplasm of walnut in several regions, especially the related studies between Xinjiang and many other countries in Central Asia. In this study, genetic diversity and population genetic structure of 137 walnut germplasm resources collected from four regions of Ili, Kashgar, Hotan and Aksu and six regions of Kyrgyzstan and Tajikistan were analyzed by SSR (Simple Sequence Repeat) molecular markers. The genetic diversity and genetic relationships of walnut germplasm in six regions of Xinjiang and Central Asia were explored at the molecular level, in order to provide material references for further revealing the origin of walnut in the Central Asian ecology-region.【Methods】137

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accessions of walnut germplasm resources were collected a in six areas with individual random sampling, including 19 of Kashgar region, 15 of Aksu area, Hotan region in 21, 31 of Ili region, 37 of Kyrgyzstan, 13 of Tajikistan. The fresh walnut leaves collected were wiped clean and immediately put into the self-sealed bag containing silica gel for preservation. The DNA of each material was extracted by Plant Genomic DNA Extraction Kit (Tiangen Company). After testing the quality and concentration of DNA,30 accessions of randomly selected walnut germplasms were used to construct the six group's genomic DNA mixed gene pool, using published 64 SSR primers .The PCR products were detected by 1% agarose gel electrophoresis, and the bright and clear primers were screened and amplified. The 14 pairs of primers screened were used for PCR amplification of all the test materials. PCR products were separated with 6% non-denatured polyacrylamide gel electrophoresis. The bands were detected by silver staining. The amplification of each primer in each individual was counted. Genetic diversity indices including the number of alleles (*Na*), the number of effective alleles (*Ne*), Shannon's information index (*I*) and allele frequency were calculated by Popgene32 software, and the genetic structure indices were calculated. The results included genetic differentiation coefficient (*Gst*), gene flow between populations (*Nm*), Nei's standard genetic distance (*GD*) and genetic consistency (*GI)*. Clustering analysis of 137 walnut germplasm was carried out using NTSYS- PC2.10 program and the map was made.【Results】A total of 130 alleles were detected by 14 pairs of SSR primers, with the range of variation ranging from 6 to 13. An average of 9.285 7 alleles could be detected by each pair of SSR primers. The percentage of 14 SSR loci ranged from 0.625 7 to 0.865 1, with an average of 0.763 4. The average Shannon's diversity index (*I*) was 0.459 8. The mean value of total genetic diversity (*Ht*) was 3.248 1. The mean value of gene diversity (*Hs*) within the population was 2.108 9; The genetic differentiation index (*Gst*) between populations ranged from 0.057 8 to 0.406 8, with an average of 0.259 5. The mean value of gene flow (*Nm*) was 3.017 4; All allele frequencies were not uniformly distributed. UPGMA clustering results showed that 137 samples were divided into three groups when the genetic similarity coefficient was 0.75. The first group consisted of 42 samples, including 16 samples from Aksu, 21 samples from Hotan and 5 samples from Kyrgyzstan. The second group contained 44 accessions, including 12 samples from Ili and 32 samples from Kyrgyzstan. The third group contained 51 samples, including 19 samples from Kashgar, 13 samples from Tajikistan and 19 samples from Ili. To a certain extent, the clustering results indicated the distance of the genetic relationship among the populations, and the overall distribution were related to the geographical distribution, and there was a certain gene exchange among the different regions. The analysis results of genetic distance and genetic consistency were consistent with the tree cluster diagram constructed by UPGMA method based on genetic distance frequency.【Conclusion】14 pairs of SSR primers with clear bands and strong polymorphism in walnut were screened , The genetic diversity of the six regions detected by SSR markers was high, among walnut germplasm resources from different geographical distribution in the Central Asian ecological region, and the level of genetic differentiation among populations was high. The gene exchange among walnut communities was high, which might be caused by the long-distance dispersal of walnut seeds by human or birds The walnut germplasm in Ili region would be likely the origin of walnut in Central Asia..

Key words: Walnut（*Juglans regia* L.）; Germplasm resources; SSR; Genetic diversity; Genetic structure

核桃（*Juglans regia* L.）是胡桃科胡桃属多年生木本油料植物，风味独特，营养价值高，具有健胃、补血、润肺、养神等功效[1-3]，栽培广泛，经济、生态、社会效益显著[4-5]。核桃属植物全世界约有23种，主要分布在北半球从中亚到地中海盆地的温带和亚热带地区，包括其原始祖先在内都存在芽鳞性状，证明其是

适应山区温带气候进化而来的[6-7]。由于中国新疆与中亚吉尔吉斯斯坦、塔吉克斯坦等国家都有大面积的核桃栽培以及野生核桃林分布，中亚地区被认为是核桃种质资源的多样性中心，但也存在一些争议[8]。我国核桃种植历史已有3000多年，栽培的核桃是在汉代从新疆一带传入陕西，然后再传至西北和华北各地，并特别注明古代西域在今新疆一带[9-10]。林培钧等[11]也认为，我国普遍栽培的核桃原产于欧洲东南部、亚洲西部和我国新疆，并指出新疆野核桃与栽培核桃属于同一种，前者是后者的直接祖先，这进一步证明亚洲西部实际为现代伊犁地区。据实地考察，中亚国家核桃种质资源较为丰富，但对包括中国新疆在内的中亚生态区核桃种质资源遗传多样性方面的研究并不多。

随着分子生物学的发展，分子标记技术在果树遗传多样性研究方面得到广泛应用。Ebrahimi等[12]利用10个SSR引物对3大洲25个国家/地区的189份核桃种质资源进行分析，确定了欧洲和北非的种质资源与希腊和近东的种质资源是分开的。Bernard等[13]利用13对SSR引物对来自于世界各地的253份核桃种质进行分析，确定了东欧和亚洲的资源与西欧和美洲的资源2个主要类群。Pollegioni等[14]利用14对SSR引物对来自8个国家39份核桃种质资源进行了分析。相关研究多数都涉及中亚生态区的种质资源，足以见其重要性。因此，需要在这一区域更大范围内结合国内外更加丰富的材料，利用分子标记技术研究核桃种质资源的遗传变异与种群结构，为探究中亚生态区核桃种质资源起源分化、遗传变异与传播途径提供依据。

1 材料和方法

1.1 供试材料

试验材料选自中国新疆与吉尔吉斯斯坦、塔吉克斯坦2个中亚国家6个相对集中的核桃分布区，根据各区域核桃分布情况进行单株采样，采样所涉及的核桃树都为20年生以上进入结果期的大树。采集到137份核桃种质资源，包括中国新疆喀什地区19份，中国新疆阿克苏地区15份，中国新疆和田地区21份，中国新疆伊犁地区31份，吉尔吉斯斯坦37份，塔吉克斯坦13份。其中中国新疆和田与中国新疆阿克苏部分样品采集于阿克苏地区实验林场。将所采集的新鲜核桃叶片擦拭干净后放入含硅胶的自封袋中干燥保存。采集样品的基本信息见表1。

1.2 试验方法

1.2.1 DNA提取 取出自封袋中0.2 g干质量叶片材料在液氮中迅速研磨成粉末，使用植物基因组DNA提取试剂盒（天根公司）提取试材的DNA，经1%（*w*，后同）琼脂糖凝胶电泳检测合格后，用Nanodrop微量紫外分光光度计（德国耶拿）检测DNA的质量及浓度，用ddH_2O将质量浓度稀释至50 μg·mL^{-1}，于−20 ℃冰箱保存备用。

1.2.2 PCR扩增及引物的筛选 选取已公开发表的64对核桃SSR引物，引物由生工生物工程（上海）合成。在总样本中随机挑选的30个供试材料的基因组DNA构建混合基因池，根据实验室已建立的SSR-PCR初始反应条件，在温度范围内对各引物进行梯度扩增，筛选得到不同引物最适退火温度。PCR反应体系为25 μL：50 ng模板DNA，上下游引物各1 μL，10 μL PCR预混Mix（*Taq* DNA聚合酶、dNTPs、$MgCl_2$、KCl反应缓冲液、其他稳定剂和增强剂），7 μL ddH_2O共20 μL的PCR反应体系。PCR反应程序为：94 ℃预变性5 min；94 ℃变性30 s，50~64 ℃复性40 s，72 ℃延伸40 s，共35个循环；最后72 ℃延伸8 min。1%琼脂糖凝胶电泳检测PCR产物，筛选出有明亮、清晰条带的引物。

1.2.3 聚丙烯酰胺凝胶电泳及数据分析 将筛选引物对所有供试材料进行PCR扩增，在6%非变性聚丙烯酰胺凝胶电泳下对PCR产物进行分离，银染检测条带并观察、拍照。对条带进行统计，在不同的位点上将有条带记录为1，无条带记录为0。统计每条引物在每个个体的扩增情况。利用Popgene32软件计算遗传多样性指标，包括等位基因数（*Na*）、有效等位基因数（*Ne*）、Shannon's信息指数（*I*）和等位基因频率，及反映居群遗传结构的指标，包括居群的遗传分化系数（*Gst*）、居群间的基因流（*Nm*）、*Nei's*标准遗传距离（*GD*）和遗传一致度（*GI*）。利用NTSYS-pc2.10程序进行137份核桃种质的聚类分析并作图。

2 结果与分析

2.1 PCR扩增及引物的筛选

通过设置温度梯度，得到各SSR引物最适退火温度。并从64对SSR引物中筛选出14对条带清晰、杂带少且多态性高的SSR引物，引物具体信息见表

表 1　137 份供试核桃种质资源

Table 1　137 germplasm resources of walnut for SSR analysis

国家 Country	省份 Province	地区 Region	编号 Code	经度 Longitude	纬度 Latitude	海拔 Altitude/m	立地条件等 Site conditions, etc
中国 China	新疆 Xinjiang	喀什 Kashgar	K1	77°08′01"	37°32′52"	1 663.07	浅山区,壤砂土,树势中等 Shallow mountainous area, sandy loam, medium tree vigor
			K2	77°08′20"	37°33′09"	1 659.53	浅山区,壤砂土,树势强 Shallow mountainous area, sandy loam, strong tree vigor
			K3	77°08′20"	37°33′09"	1 659.54	浅山区,壤砂土,树势中等 Shallow mountainous area, sandy loam, medium tree vigor
			K4	77°07′35"	37°32′30"	1 660.90	浅山区,壤砂土,树势中等 Shallow mountainous area, sandy loam, medium tree vigor
			K5	77°24′39"	37°57′05"	1 311.60	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			K6	77°25′30"	37°58′40"	1 313.70	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			K7	77°16′29"	37°57′20"	978.11	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			K8	78°37′13"	37°26′28"	1 394.62	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			K9	78°37′17"	37°26′27"	1 383.11	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			K10	78°37′14"	37°26′35"	1 373.09	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			K11	78°35′00"	37°23′52"	1 468.23	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			K12	78°35′01"	37°23′48"	1 470.55	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			K13	78°30′29"	37°12′03"	1 827.40	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			K14	77°19′50"	37°51′29"	1 391.14	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			K15	77°19′51"	37°51′34"	1 386.82	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			K16	77°20′06"	37°51′04"	1 381.64	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			K17	77°20′20"	37°51′44"	1 376.95	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			K18	77°19′51"	37°51'33"	1 386.12	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			K19	77°19′52"	37°51'34"	1 387.18	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
		阿克苏 Aksu	A1	80°31′55"	41°15′30"	1 103.42	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			A2	80°32′15"	41°15′32"	1 103.55	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			A3	80°32′04"	41°15′35"	1 103.32	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			A4	80°32′02"	41°15′29"	1 103.42	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			A5	80°32′02"	41°15′30"	1 103.42	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			A6	80°32′04"	41°15′32"	1 104.12	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			A7	80°14′12″	41°18′16″	1 212.00	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			A8	80°14′12″	41°18′16″	1 212.00	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			A9	80°20′33″	41°13′32″	1 156.60	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			A10	80°20′33″	41°13′32″	1 156.60	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			A11	80°20′33″	41°13′32″	1 156.60	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			A12	80°20′33″	41°13′32″	1 156.60	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			A13	80°20′33″	41°13′32″	1 156.60	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			A14	80°20′33″	41°13′32″	1 156.60	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			A15	80°20′33″	41°13′32″	1 156.60	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			A16	80°20′33″	41°13′32″	1 156.60	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor

续表

Continued Table

国家 Country	省份 Province	地区 Region	编号 Code	经度 Longitude	纬度 Latitude	海拔 Altitude/m	立地条件等 Site conditions, etc
		和田 Hotan	H1	79°43′27"	37°03′47"	1 225.17	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			H2	77°51′04"	37°19′15"	2 102.04	浅山区,壤砂土,树势中等 Shallow mountainous area, sandy loam, medium tree vigor
			H3	77°50′32"	37°19′39"	1 753.25	浅山区,壤砂土,树势中等 Shallow mountainous area, sandy loam, medium tree vigor
			H4	79°40′47"	37°17′55"	1 339.58	平原区,壤砂土,树势强Plain area, sandy loam, strong tree vigor
			H5	79°40′47"	37°17′55"	1 339.58	平原区,壤砂土,树势强Plain area, sandy loam, strong tree vigor
			H6	77°49′56"	37°15′28"	2 120.56	浅山区,壤砂土,树势中等 Shallow mountainous area, sandy loam, medium tree vigor
			H7	79°41′09"	37°18′09"	1 328.49	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			H8	79°41′21"	37°18′10"	1 325.80	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			H9	79°40′56"	37°18′56"	1 322.18	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			H10	79°48′32"	37°06′27"	1 384.97	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			H11	79°48′25"	37°06′31"	1 378.09	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			H12	79°43′05"	37°01′49"	1 457.28	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			H13	79°48′24"	37°03′54"	1 426.13	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			H14	78°27′00"	37°11′09"	1 912.18	浅山区,壤砂土,树势中等 Shallow mountainous area, sandy loam, medium tree vigor
			H15	78°27′04"	37°11′11"	1 918.38	浅山区,壤砂土,树势中等 Shallow mountainous area, sandy loam, medium tree vigor
			H16	78°27′04"	37°11′11"	1 918.38	浅山区,壤砂土,树势中等 Shallow mountainous area, sandy loam, medium tree vigor
			H17	79°68′93″	37°30′28″	1 322.00	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			H18	79°68′60″	37°30′26″	1 328.60	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			H19	79°68′60″	37°30′26″	1 328.60	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			H20	79°68′60″	37°30′26″	1 328.60	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			H21	79°68′60″	37°30′26″	1 328.60	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
		伊犁 Ili	Y1	82°16′10"	43°20′35"	1 557.34	山区阴坡中部,壤土,树势中等 The middle part of the shady slope in mountainous area, loamy soil, medium tree vigor
			Y2	82°16′26"	43°20′33"	1 597.58	山区阳坡上部,壤土,树势强 The upper part of the sunny slope in the mountainous area, loamy soil, strong tree vigor
			Y3	82°16′02"	43°20′34"	1 635.33	山区阴坡上部,壤土,树势中等 The upper part of the shady slope in the mountainous area, loamy soil, medium tree vigor
			Y4	82°16′20"	43°20′49"	1 435.21	山区阴坡下部,壤土,树势中等 The lower part of the shady slope in the mountainous area, loamy soil, medium tree vigor
			Y5	82°16′20"	43°21′02"	1 223.40	山区阴坡下部,壤土,树势强 The lower part of the shady slope in the mountainous area, loamy soil, strong tree vigor
			Y6	82°16′20"	43°21′02"	1 222.80	山区阴坡下部,壤土,树势强 The lower part of the shady slope in the mountainous area, loamy soil, strong tree vigor
			Y7	82°16′20"	43°21′02"	1 220.40	山区阴坡下部,壤土,树势强 The lower part of the shady slope in the mountainous area, loamy soil, strong tree vigor
			Y8	82°16′20"	43°20′59"	1 233.83	山区阴坡下部,壤土,树势中等 The lower part of the shady slope in the mountainous area, loamy soil, medium tree vigor

续表

Continued Table

国家 Country	省份 Province	地区 Region	编号 Code	经度 Longitude	纬度 Latitude	海拔 Altitude/m	立地条件等 Site conditions, etc
			Y9	82°16′17"	43°20′54"	1 284.76	山区阴坡下部,壤土,树势中等 The lower part of the shady slope in the mountainous area, loamy soil, medium tree vigor
			Y10	82°16′15"	43°20′52"	1 275.82	山区阴坡下部,壤土,树势中等 The lower part of the shady slope in the mountainous area, loamy soil, medium tree vigor
			Y11	82°16′15"	43°20′50"	1 277.23	山区阴坡下部,壤土,树势中等 The lower part of the shady slope in the mountainous area, loamy soil, medium tree vigor
			Y12	82°16′16"	43°20′38"	1 366.89	山区阴坡下部,壤土,树势中等 The lower part of the shady slope in the mountainous area, loamy soil, medium tree vigor
			Y13	82°16′19"	43°20′35"	1 404.03	山区阴坡中部,壤土,树势中等 The middle part of the shady slope in mountainous area, loamy soil, medium tree vigor
			Y14	82°16′19"	43°20′33"	1 427.60	山区阴坡中部,壤土,树势中等 The middle part of the shady slope in mountainous area, loamy soil, medium tree vigor
			Y15	82°16′20"	43°20′33"	1 415.45	山区阴坡中部,壤土,树势中等 The middle part of the shady slope in mountainous area, loamy soil, medium tree vigor
			Y16	82°16′23"	43°20′32"	1 459.09	山区阴坡中部,壤土,树势中等 The middle part of the shady slope in mountainous area, loamy soil, medium tree vigor
			Y17	82°16′23"	43°20′30"	1 473.30	山区阴坡中部,壤土,树势中等 The middle part of the shady slope in mountainous area, loamy soil, medium tree vigor
			Y18	82°16′24"	43°20′30"	1 483.66	山区阳坡中部,壤土,树势强 The middle part of the sunny slope in the mountainous area, loamy soil, strong tree vigor
			Y19	82°16′26"	43°20′29"	1 503.55	山区阳坡中部,壤土,树势强 The middle part of the sunny slope in the mountainous area, loamy soil, strong tree vigor
			Y20	82°16′19"	43°20′32"	1 408.34	山区阳坡中部,壤土,树势强 The middle part of the sunny slope in the mountainous area, loamy soil, strong tree vigor
			Y21	82°16′11"	43°20′36"	1 346.52	山区阳坡中部,壤土,树势强 The middle part of the sunny slope in the mountainous area, loamy soil, strong tree vigor
			Y22	82°16′06"	43°20′34"	1 406.11	山区阳坡中部,壤土,树势中等 The middle part of the sunny slope in the mountainous area, loamy soil, medium tree vigor
			Y23	82°16′03"	43°20′33"	1 441.26	山区阴坡中部,壤土,树势中等 The middle part of the shady slope in mountainous area, loamy soil, medium tree vigor
			Y24	82°16′12"	43°20′36"	1 408.91	山区阳坡中部,壤土,树势强 The middle part of the sunny slope in the mountainous area, loamy soil, strong tree vigor
			Y25	82°16′13"	43°20′39"	1 364.74	山区阳坡中部,壤土,树势强 The middle part of the sunny slope in the mountainous area, loamy soil, strong tree vigor
			Y26	82°16′31"	43°20′32"	1 525.83	山区阳坡上部,壤土,树势中等 The upper part of the sunny slope in the mountainous area, loamy soil, medium tree vigor
			Y27	82°16′33"	43°20′29"	1 448.74	山区阴坡下部,壤土,树势中等 The lower part of the shady slope in the mountainous area, loamy soil, medium tree vigor
			Y28	82°16′35"	43°20′26"	1 462.54	山区阴坡下部,壤土,树势强 The lower part of the shady slope in the mountainous area, loamy soil, strong tree vigor

续表

Continued Table

国家 Country	省份 Province	地区 Region	编号 Code	经度 Longitude	纬度 Latitude	海拔 Altitude/m	立地条件等 Site conditions, etc
			Y29	82°16′36"	43°20′26"	1 479.10	山区阴坡中部,壤土,树势强 The middle part of the shady slope in mountainous area, loamy soil, strong tree vigor
			Y30	82°16′38"	43°20′35"	1 495.96	山区阴坡下部,壤土,树势中等 The lower part of the shady slope in the mountainous area, loamy soil, medium tree vigor
			Y31	82°16′22"	43°20′57"	1 308.99	山区阴坡下部,壤土,树势中等 The lower part of the shady slope in the mountainous area, loamy soil, medium tree vigor
吉尔吉斯斯坦 Kyrgyz-stan	奥什州 Osh State	奥什 Osh	J1	72°44′45"	40°30′50"	980.11	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
	贾拉拉巴德州 Jalalabad	贾拉拉巴德 Jalalabad	J2	73°11′10"	40°41′20"	953.93	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
		杰尔格塔尔 Jargotar	J3	73°19′12"	41°10′54"	1 335.22	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor
			J4	73°20′48"	41°12′46"	1 404.06	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor
			J5	73°20′32"	41°12′37"	1 378.77	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor
			J6	73°19′14"	41°10′56"	1 347.42	浅山区,壤土,树势强 Shallow mountainous area , loamy soil, strong tree vigor
			J7	73°19′15"	41°10′57"	1 353.68	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor
	奥什州 Osh State	奥什 Osh	J8	73°38′43"	40°50′40"	1 319.69	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor
			J9	73°38′19"	40°50′44"	1 321.03	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor
			J10	73°38′20"	40°50′43"	1 326.46	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor,
			J11	73°36′19"	40°49′05"	1 260.06	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor
			J12	72°45′03"	40°31′03"	955.12	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
	贾拉拉巴德州 Jalalabad	贾拉拉巴德 Jalalabad	J13	72°58′15"	40°56′36"	771.01	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			J14	71°56′59"	41°50′38"	1 451.44	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor
		纳奈 Nanai	J15	71°57′55"	41°51′23"	1 833.76	山区阴坡上部,壤土,树势中等 The upper part of the shady slope in the mountainous area, loamy soil, medium tree vigor
			J16	71°56′56"	41°51′12"	1 585.08	山区阴坡下部,壤土,树势中等 The lower part of the shady slope in the mountainous area, loamy soil, medium tree vigor
			J17	71°56′30"	41°51′23"	1 516.60	山区阴坡下部,壤土,树势中等 The lower part of the shady slope in the mountainous area, loamy soil, medium tree vigor
			J18	71°56′31"	41°51′21"	1 506.21	山区阳坡下部,壤土,树势中等 The lower part of the sunny slope in the mountainous area, loamy soil, medium tree vigor
			J19	71°57′22"	41°50′11"	1 397.98	山区阳坡下部,壤土,树势中等 The lower part of the sunny slope in the mountainous area, loamy soil, medium tree vigor
			J20	71°57′24"	41°50′10"	1 391.90	山区阳坡下部,壤土,树势中等 The lower part of the sunny slope in the mountainous area, loamy soil, medium tree vigor
			J21	71°57′25"	71°45′15"	1 355.01	山区阳坡下部,壤土,树势中等 The lower part of the sunny slope in the mountainous area, loamy soil, medium tree vigor
		阿拉布卡 Allabka	J22	71°45′15"	41°29′20"	1 295.21	浅山区,壤土,树势中等 Shallow mountainous area, loamy soil, medium tree vigor
		贾拉拉巴德 Jalalabad	J23	72°57′20"	41°17′35"	1 462.17	山区阴坡下部,壤土,树势强 The lower part of the shady slope in the mountainous area, loamy soil, strong tree vigor
			J24	72°57′16"	41°17′32"	1 469.81	山区阴坡下部,壤土,树势强 The lower part of the shady slope in the mountainous area, loamy soil, strong tree vigor

续表

Continued Table

国家 Country	省份 Province	地区 Region	编号 Code	经度 Longitude	纬度 Latitude	海拔 Altitude/m	立地条件等 Site conditions, etc
		巴扎尔库尔干 Bazar Korgan	J25	72°57′12"	41°17′35"	1 470.67	山区阴坡下部,壤土,树势中等 The lower part of the shady slope in the mountainous area, loamy soil, medium tree vigor
			J26	72°57′15"	41°17′33"	1 452.55	山区阴坡下部,壤土,树势中等 The lower part of the shady slope in the mountainous area, loamy soil, medium tree vigor
			J27	72°58′01"	41°17′56"	1 506.90	山区阴坡中部,壤土,树势强 The middle part of the shady slope in mountainous area, loamy soil, strong tree vigor
			J28	72°58′01"	41°17′54"	1 513.10	山区阴坡中部,壤土,树势中等 The middle part of the shady slope in mountainous area, loamy soil, medium tree vigor
			J29	72°57′59"	41°17′58"	1 495.31	山区阴坡中部,壤土,树势强 The middle part of the shady slope in mountainous area, loamy soil, strong tree vigor
			J30	72°58′00"	41°17′58"	1 504.98	山区阴坡中部,壤土,树势中等 The middle part of the shady slope in mountainous area, loamy soil, medium tree vigor
			J31	72°54′49"	41°20′18"	1 542.80	山区阴坡中部,壤土,树势中等 The middle part of the shady slope in mountainous area, loamy soil, medium tree vigor
			J32	72°54′50"	41°20′20"	1 524.03	山区阴坡中部,壤土,树势强 The middle part of the shady slope in mountainous area, loamy soil, strong tree vigor
			J33	72°58′25"	41°19′35"	1 338.72	浅山区,壤土,树势中等 Shallow mountainous area, loamy soil, medium tree vigor
			J34	72°58′25"	41°19′37"	1 334.62	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor
			J35	72°58′27"	41°19′36"	1 330.22	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor
			J36	73°04′45"	41°22′18"	1 349.00	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor
			J37	73°04′50"	41°22′18"	1 364.12	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor
塔吉克斯坦 Tajikistan	哈特隆州 Hartron	库尔干秋别 Korgan autun farewell	T1	68°43′42"	37°46′41"	495.30	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			T2	68°38′42"	37°51′54"	517.12	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
	列宁纳巴德州 Leninabadezhou	苦盏 Bitter cup	T3	69°41′21"	40°12′25"	446.12	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			T4	69°37′56"	40°10′14"	391.22	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
			T5	69°26′48"	40°07′48"	408.33	平原区,壤砂土,树势中等 Plain area, sandy loam, medium tree vigor
			T6	69°01′21"	39°54′01"	908.12	平原区,壤砂土,树势强 Plain area, sandy loam, strong tree vigor
	塔吉克斯坦直辖州 Tajikistan	杜尚别 Dushanbe	T7	68°48′33"	38°45′38"	1 017.13	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor
			T8	69°00′47"	38°30′44"	1 232.12	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor
			T9	69°22′33"	38°35′27"	1 464.63	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor
			T10	69°22′24"	38°35′27"	1 387.58	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor
			T11	68°48′37"	38°55′09"	1 484.28	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor
			T12	68°45′48"	38°58′00"	1 709.27	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor
			T13	69°54′37"	38°49′59"	1 623.12	浅山区,壤土,树势强 Shallow mountainous area, loamy soil, strong tree vigor

2。14对SSR引物对137份核桃种质资源样品进行扩增，均表现出较高的遗传多样性。图1为引物WJR014在137份核桃种质资源样品的扩增结果。

2.2 核桃种质资源的遗传多样性分析

6个区域的核桃种质资源遗传多样性结果见表3，14对SSR引物共检测出130个基因位点，等位基因数（*Na*）的平均值为9.285 7。位点WJR007与WC7329等位基因数最少，均为6。位点WJR115、JR6160与JH86514等位基因数最多，均为13。有效等位基因数（*Ne*）的平均值为7.098 1。位点WC7329有效等位基因数最少，为4.047 1。位点JH86514有效等位基因数最多，为9.580 1。多态性位点占比范围为0.625 7~0.865 1，平均值为0.763 4。平均Shannon's多样性指数（*I*）为0.459 8。种群总基因多样性（*Ht*）平均值为3.248 1；种群内基因多样性（*Hs*）平均值为2.108 9；种群间的遗传分化指数（*Gst*）范围为0.057 8~0.406 8，平均值为0.259 5；基因流（*Nm*）平均值为3.017 4。

由表4可知，各个等位基因频率整体分布不均匀。其中，WJR007的B、C、D等位基因频率较高，均大于0.200 0。除JM61666主要为C等位基因频率高外，JC5411、WJR115、WJR041主要为B等位基因频率高，WJR265、WC7329、JR6160、WJR065、WJR014主要为D等位基因频率高，WJR031、JH89978、JR4616、JH86514主要为I等位基因频率高，其余等位基因频率相对较低。

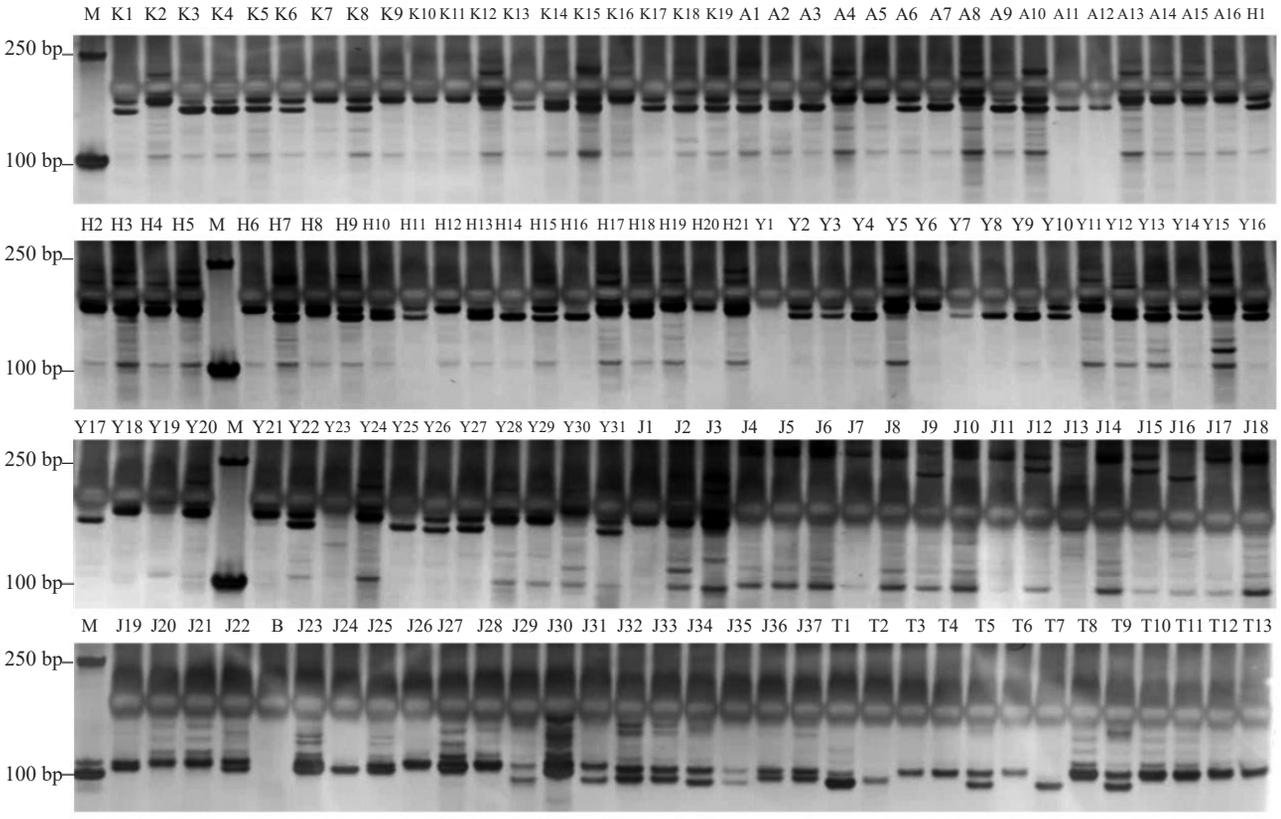
2.3 核桃种质资源聚类分析

根据14对SSR引物扩增数据统计结果，利用NTSYS软件，采用UPGMA法聚类得到核桃聚类树状图（图2），在相似系数为0.75时可划分为3个类

表2 筛选出的14对引物序列及适宜的退火温度

Table 2 Sequences and proper annealing temperatures of 14 selected primer pairs

引物名称 Primer name	引物序列（5'-3'） Primer Sequence（5' to 3'）	引物长度 Primer length/bp	重复基序 Repeat motif	退火温度 Annealing temperature/℃
JC5411-F	AAGCTGTTTGTGCCAAAAGC	300-315	$(GAT)_7$	55
JC5411-R	TTCTAGCGAGAATTCCGGCC			
WJR007-F	AATGCATGACATGGTGGTCA	228	(AT)n	58
WJR007-R	GTATGAAACAATCATTTTCACTCAA			
WJR265-F	TGGCTATTGCAAAATCAGGTC	268	(AAT)n	56
WJR265-R	CAAAAGCATGTAGGTCGGGT			
JC7329-F	TGCAGCGCATCAGTGAGTTA	330-363	$(TGA)_8$	55
JC7329-R	ACGCTCGAGTGTAGTAGCAAG			
WJR115-F	CTCACCCTTGTAGAGCGAGG	102	(GA)n	57
WJR115-R	TGCAAACTCAGTGCTAAAATCAA			
JR6160-F	ACTTCAGGTTCCCAACGCAA	180-217	$(GA)_{10}$	58
JR6160-R	TAGAGGGAAGGTCTCCGGTG			
WJR065-F	CAGCCATTCGAGTTAAGACTTG	102	(GATGA)n	56
WJR065-R	TTCGATTAAGAGCTCGTTTGG			
WJR014-F	ACATGATGAGTAGGGCAGGG	236	(TA)n	57
WJR014-R	GCTTATTGATCAAGATTGGACG			
WJR031-F	AGCTTCCCCCATTCTCCTAA	225	(TA)n	58
WJR031-R	GGACCTCCACAACCAAAAGA			
JH89978-F	ACCTTCCCTGCTCCTCTCTT	190-201	$(GGT)_6$	55
JH89978-R	GAGCCTTGTGGAAGCAAACG			
WJR041-F	ATCTGCCTTCCCATAAGGCT	161	$(ATAC)_8$	60
WJR041-R	TTCTCCCATTGTCGGATCTC			
JR4616-F	AGCCCTTTTGCATCGGCTAT	160-180	$(AGAC)_5$	55
JR4616-R	AGCTGACCGATCGATCAACA			
JH86514-F	CGTTACGTCGGGAGGATGAG	132-162	(TTAGGG)6	55
JH86514-R	ATTGAGCGACGAAAAGGGGT			
JM61666-F	AACTGTTGCCGGAGCTTTCT	266-280	$(GA)_{11}$	55
JM61666-R	TGGGATAACACCACATGCAGT			



M. Marker; B. 空白。编号同表 1。

M. Marker; B. Blank. The codes are the same as in Table 1.

图 1　引物 WJR014 的扩增结果

Fig. 1　DNA fragments amplified by primer WJR014 in *J. regia*

表 3　采自 6 个区域的核桃遗传多样性

Table 3　Genetic diversity of *J. regia* in six regions

引物名称 primer name	等位基因数 Detected alleles number，Na	有效等位基因数 Effective number of alleles，*Ne*	多态性位点占比 Percent of polymorphic bands/%	多样性指数 Shannon's-index，*I*	种群总基因多样性 Total genetic diversity of the population，*Ht*	种群内基因多样性 Genetic diversity within the population，*Hs*	种群间的遗传分化指数 The index of genetic differentiation between populations，*Gst*	基因流 Gene flow，*Nm*
JC5411	7	4.379 6	0.625 7	0.278 1	1.184 8	1.075 6	0.057 8	14.789 9
WJR007	6	5.063 7	0.844 0	0.550 9	2.250 4	1.241 2	0.406 8	0.924 1
WJR265	9	6.843 0	0.760 3	0.475 2	2.766 6	2.261 3	0.172 4	3.640 8
WC7329	6	4.047 1	0.674 5	0.322 2	1.314 8	0.683 4	0.274 2	15.263 5
WJR115	13	9.512 2	0.731 7	0.437 4	3.813 4	3.067 9	0.157 7	6.515 3
JR6160	13	9.492 2	0.730 2	0.421 8	3.731 8	2.671 5	0.233 3	3.018 7
WJR065	8	6.467 7	0.808 5	0.512 4	2.604 5	1.747 6	0.297 1	2.071 1
WJR014	7	5.215 4	0.745 1	0.459 7	2.044 3	1.162 2	0.338 7	1.146 6
WJR031	9	7.301 1	0.811 2	0.516 2	3.039 0	1.799 0	0.335 2	2.899 6
JH89978	10	7.740 0	0.774 0	0.469 8	3.196 1	2.058 4	0.286 5	8.190 3
WJR041	8	6.325 3	0.790 7	0.514 5	2.751 0	2.134 4	0.230 5	2.286 6
JR4616	11	9.515 6	0.865 1	0.585 5	4.314 6	2.625 5	0.399 4	0.979 8
JH86514	13	9.580 1	0.736 9	0.407 1	3.498 9	2.268 1	0.224 7	7.717 6
JM61666	10	7.890 6	0.789 1	0.486 4	3.248 1	2.108 9	0.282 2	3.017 4
平均值 Mean	9.2857	7.098 1	0.763 4	0.459 8	2.839 9	1.921 8	0.259 5	5.175 8

表4 采自6个区域的核桃等位基因频率

Table 4 Overall allele frequency of *J. regia* in six regions

等位基因 Allele	位点 Locus													
	JC5411	WJR007	WJR265	WC7329	WJR115	JR6160	WJR065	WJR014	WJR031	JH89978	WJR041	JR4616	JH86514	JM61666
A	0.122 1	0.029 4	0.087 0	0.029 6	0.006 3	0.107 1	0.153 0	0.171 3	0.109 6	0.073 0	0.124 2	0.139 2	0.133 3	0.115 8
B	0.494 2	0.226 5	0.065 2	0.197 6	0.211 6	0.128 4	0.233 0	0.217 2	0.164 1	0.107 0	0.264 1	0.082 6	0.019 7	0.147 2
C	0.004 8	0.259 4	0.145 2	0.221 2	0.176 8	0.143 8	0.097 0	0.054 2	0.196 4	0.185 1	0.138 5	0.151 6	0.127 8	0.210 4
D	0.111 6	0.263 5	0.273 8	0.544 6	0.041 7	0.202 5	0.152 1	0.242 4	0.222 7	0.007 9	0.241 6	0.115 9	0.063 2	0.163 6
E	0.198 7	0.172 8	0.128 1	0.003 4	0.058 1	0.187 0	0.239 8	0.179 7	0.147 3	0.225 8	0.108 0	0.053 5	0.119 9	0.161 0
F	0.063 7	0.048 4	0.114 9	0.003 4	0.064 4	0.078 9	0.089 7	0.057 9	0.019 6	0.137 2	0.075 9	0.118 8	0.203 9	0.014 4
G	0.004 8		0.156 4		0.151 5	0.025 2	0.007 8	0.077 3	0.051 1	0.022 2	0.036 3	0.072 0	0.018 4	0.053 3
H			0.014 7		0.046 2	0.018 4	0.027 5		0.008 9	0.002 2	0.011 4	0.079 5	0.018 6	0.096 2
I			0.032 1		0.193 3	0.111 0			0.238 2	0.608 7		0.463 0	0.319 3	0.022 9
J					0.101 0	0.021 1				0.055 6		0.049 5	0.169 6	0.029 7
K					0.014 5	0.016 5						0.014 1	0.001 3	
I					0.056 6	0.016 5							0.005 3	
M					0.006 4	0.016 5							0.008 2	

群。第一类群共42份种质资源，包括中国新疆阿克苏16份，中国新疆和田21份，吉尔吉斯斯坦5份；第二类群共44份种质资源，包括中国新疆伊犁12份，吉尔吉斯斯坦32份；第三类群共51份，包括中国新疆喀什19份，塔吉克斯坦13份和中国新疆伊犁19份。聚类结果在一定程度上表明了各居群间亲缘关系的远近，整体分布与地理分布有一定的关联，各区域间也存在一定的基因交流。

2.4 不同居群间遗传距离和遗传一致度

为了进一步分析核桃种质资源不同居群间的遗传关系，计算了6个居群间的遗传距离（*GD*）和遗传一致度（*GI*），由表5可知，居群遗传一致度在0.740 5~0.959 5之间，遗传距离在0.041 3~0.300 5之间,说明群体间的相似程度较高，遗传距离小。其中，中国新疆阿克苏居群与中国新疆和田居群的遗传相似性最高，为0.959 5，遗传距离最小，为0.041 3；中国新疆和田居群与塔吉克斯坦居群的遗传距离最大，为0.300 5，遗传相似性最低，为0.740 5。遗传距离、遗传一致度的分析结果与UPGMA法构建的树状聚类图相互印证、吻合一致。

3 讨 论

物种具备一定的遗传多样性，遗传多样性水平越高，该物种或种群对新环境的适应能力就越强[15]。陆婷等[16]利用SSR分子标记技术对云南省迪庆藏族自治州3个群体共161份深纹核桃种质资源进行遗传多样性分析，揭示资源的遗传多样性水平及遗传结构特征。肖良俊等[17]利用SSR分子标记技术对滇西北13个群体共332份深纹核桃种质进行遗传多样性分析，揭示长期的自然选择及人为影响导致了滇西北核桃种质资源遗传多样性水平及特有的分布格局。张浩等[18]利用SRAP分子标记技术对山东鹊山省级自然保护区不同山体部位核桃居群进行遗传多样性分析，认为人为干扰、环境筛选和种内竞争是影响核桃居群更新的主要原因。

在中亚各核桃分布区，不同国家对本国的核桃遗传多样性研究均有所涉及。然而对中亚生态区更大范围内核桃种质资源间的研究不多。笔者在本研究中利用筛选出的14对SSR引物在137份核桃种质资源中共检测出130个基因位点，等位基因数（*Na*）平均值为9.285 7，有效等位基因数（*Ne*）平均值为7.098 1。多态性位点占比范围为0.625 7~0.865 1，平均值0.763 4；平均Shannon's多样性指数（*I*）为0.459 8。种群总基因多样性（*Ht*）平均值为3.248 1；种群内基因多样性（*Hs*）平均值为2.108 9。试验中所有SSR位点在样品中大部分表现出较好的多态性，各指标之间体现了较高的一致性，这说明中亚生态区核桃种质资源具有较高的遗传多样性。

群体的遗传结构受遗传漂变和基因流等很多因素的影响，基因流值与遗传分化系数等都是评价群体遗传结构的重要参数和指标[19]。本试验中137份核桃种质资源种群间的遗传分化指数（*Gst*）范围为0.057 8~0.406 8，平均值为0.259 5，说明6个分布区之间的遗传分化程度较高。基因流是影响群体内部

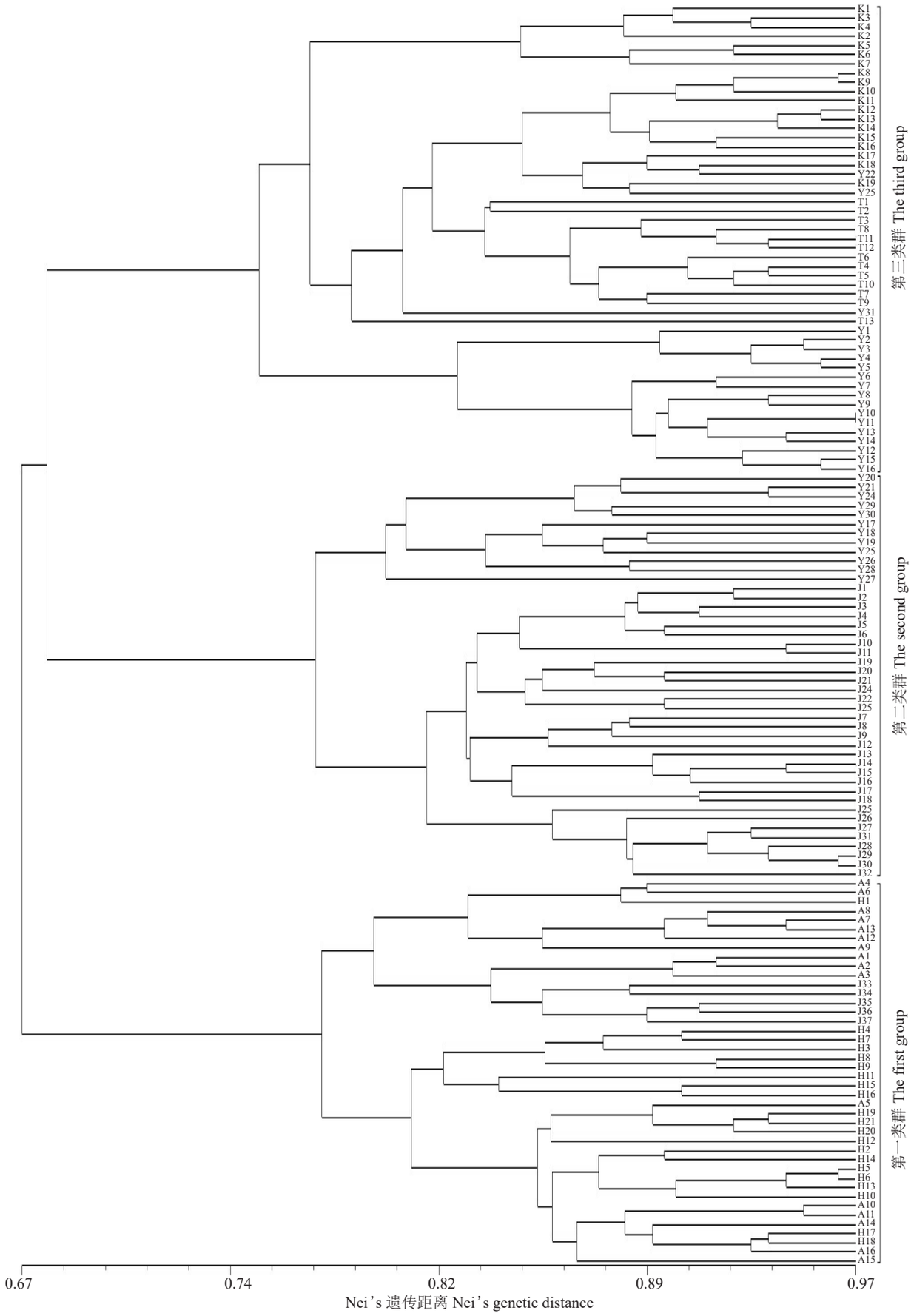


图 2　采自 6 个区域的核桃聚类分析

Fig. 2　Clustering analysis of *J. regia* in six regions based on SSR data

表5 中亚地区137份核桃种质遗传一致度和遗传距离分析

Table 5 The genetic identity and genetic distance of 137 walnut in Central Asia

地区 Region	中国新疆喀什 Kashgar, Xinjiang, China	中国新疆阿克苏 Aksu, Xinjiang, China	中国新疆和田 Hotan, Xinjiang, China	中国新疆伊犁 Ili, Xinjiang, China	吉尔吉斯斯坦 Kyrgyzstan	塔吉克斯坦 Tajikistan
中国新疆喀什 Kashgar, Xinjiang, China		0.821 3	0.791 5	0.941 9	0.835 7	0.931 6
中国新疆阿克苏 Aksu, Xinjiang, China	0.196 9		0.959 5	0.880 5	0.867 9	0.766 5
中国新疆和田 Hotan, Xinjiang, China	0.233 8	0.041 3		0.856 8	0.844 9	0.740 5
中国新疆伊犁 Ili, Xinjiang, China	0.059 9	0.127 3	0.154 6		0.931 6	0.884 2
吉尔吉斯斯坦 Kyrgyzstan	0.179 5	0.141 7	0.168 5	0.070 9		0.802 9
塔吉克斯坦 Tajikistan	0.070 8	0.265 9	0.300 5	0.123 1	0.219 5	

注：右上为遗传一致度，左下为遗传距离。

Note: The upper right represent genetic identity and the lower left is genetic distance.

和群体间遗传变异程度的重要因素[20]。核桃分布区之间的基因流（*Nm*）平均值为5.175 8，表明分布区间的基因交流程度较大，因此推断核桃虽为风媒花，花粉可远距离传播，但本研究采样地新疆与中亚周边国家有山脉等地理屏障，核桃群体间的基因交流更可能是通过古代丝绸之路和人类活动等方式携带种子远距离传播。

遗传相似系数是判断品种间亲缘关系及遗传基础的标准之一[21]。笔者通过基于Nei's遗传距离的UPGMA聚类，显示137份供试材料在遗传相似性系数为0.75时分成3个类群，其中，聚类第一类群显示吉尔吉斯斯坦与中国新疆阿克苏以及和田地区亲缘关系近，第二类群显示吉尔吉斯斯坦与中国新疆伊犁地区亲缘关系近，第三类群显示喀什与塔吉克斯坦以及中国新疆伊犁三地区亲缘关系近，中国新疆伊犁地区核桃种质混合分布于二、三类群中，存在地区间的基因交流。聚类结果与居群间遗传距离和遗传一致度结果一致。

中亚生态区核桃的起源尚无定论。从核桃原始分布区来看，从欧洲东南部通过西亚、中亚、喜马拉雅分布到东亚，主要是为适应山区温带气候而发展起来的[22]。据《西京杂记》、《本草纲目》等史料记载，中国核桃多引自于西域，而西域多指今新疆及周边地区[10]。新疆伊犁地区巩留县凯特明山海拔1280~1700 m的深峡谷，冬季有逆温层的保护免于寒流侵袭，气候温暖湿润，形成了"植物避难所"的地貌特征和气候要素，野核桃群落范围大且生长的野核桃与普通核桃在形态及品质上极其相似，可能是普通核桃的直系祖先[23-24]。据实地考察，吉尔吉斯斯坦南部也保留有较大面积的天然核桃林，一些区域可见到百年以上大树，分布海拔、气候、土壤条件等与中国新疆伊犁野核桃林十分相似。本研究结果也表明来自中国新疆伊犁的种质资源与吉尔吉斯斯坦种质资源亲缘关系较近，初步分析这两个区域的核桃种质资源为中亚生态区核桃起源的可能性更大。

4 结 论

利用筛选出的14对SSR引物，对中国新疆与吉尔吉斯斯坦、塔吉克斯坦2个中亚国家共6个核桃种质资源分布区的137份资源进行分子水平遗传多样性分析。结果表明，不同地理分布的核桃种质资源遗传多样性存在差异，为揭示中亚地区核桃种群结构及传播途径提供了依据。

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