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ARTICLE

Genetic Evaluation of Starch Synthesis-Related Genes and Starch Quality Traits in Special Rice Resources

Linyu Tai* Lixia Lei Lan Luo Hang Shen Jiao Chen Ruyu Tang Jing Xiang Zhengwu Zhao

Chongqing Normal University, Chongqing Engineering Research Center of Specialty Crop Resource, Chongqing, 401331, China

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ABSTRACT

The genetic diversity of 36 rice landraces and 43 breeding materials in the upper reaches of the Yangtze River in China was studied by intragenic molecular markers of 26 starch synthesis-related loci. And research on quality traits such as the amylose content (AC), gel consistency (GC) and alkali spreading value (ASV) to analyze genetic differences in quality traits. The results showed that the number of alleles, average gene diversity and polymorphism information content values of landraces were higher than those of breeding materials. The genetic similarity coefficient (GS) of 79 rice materials ranged from 0.392 to 1, with an average of 0.757.There were significant variations in the quality traits of rice landraces and breeding materials, and the high-quality compliance rates were low, only 6.3% of the varieties have an amylose content that reached grade 1. The results of cluster analysis and population structure analysis are generally consistent; that is, the two resource types are closely related and cannot be clustered independently. This study can provide a basis for genetic improvement of rice starch quality. Make full use of the quality genetic diversity of landraces in modern breeding work, further broaden the genetic base of rice and improve rice quality.

1. Introduction

Rice (*Oryza sativa L.*) is the first of the three major grain plants in China, and also one of the most adaptable cultivated crops in the world, supporting more than half of the world's population[1]. With the rapid development of rice breeding technology in China, rice yield has been dramatically increased, while the market demand for rice quality improvement has been increasing [2-3]. As one of the main objectives of rice breeding, high-quality compliance rate of rice in China has shown an apparent upward trend in the past ten years. However, as quality traits tend to be high quality, it is bound to narrow the quality difference between varieties and reduce their genetic diversity[4-6], making the genetic basis gradually narrow. The content of starch in the endosperm is closely related to rice quality. A large number of studies have shown that there are abundant allelic differences between starch synthesis related genes in rice germplasm resources, and these allelic variations are an essential reason for the difference in cooking and eating quality between different rice varieties [7].

Rice landraces are the main components of rice

*Corresponding Author:
Linyu Tai,
Chongqing Normal University, Chongqing Engineering Research Center of Specialty Crop Resource, Chongqing, 401331, China;
Email: longabsent124@163.com

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Table 1. Name, type and serial number of the 79 rice varieties in this study

Serial number	Variety type	Country number	Variety name	Serial number	Variety type	Country number	Variety name
1	Landrace	32-00080	197	41	Breeding material	ZD06675	89-1/527
2	Landrace	32-00081	Shilixiang	42	Breeding material	ZD06676	89-1/CDR22
3	Landrace	32-00082	Sanjiujiu	43	Breeding material	ZD06677	89-1/Gui 99
4	Landrace	32-00083	Sanbaibang	44	Breeding material	ZD06678	184Yi
5	Landrace	32-00084	Fengyou 788	45	Breeding material	ZD06679	355-2
6	Landrace	32-00085	Wujiehuanggu	46	Breeding material	ZD06680	481 Xuan
7	Landrace	32-00086	Kaizhou No. 2	47	Breeding material	ZD06681	486 Yi
8	Landrace	32-00087	Wuminggu	48	Breeding material	ZD06682	CDR22
9	Landrace	32-00088	Niushizhan	49	Breeding material	ZD06683	D62B
10	Landrace	32-00089	Xianxiang B	50	Breeding material	ZD06684	D62B Yi
11	Landrace	32-00090	Banbianzhan	51	Breeding material	ZD06685	G2480B
12	Landrace	32-00091	Baishuigu	52	Breeding material	ZD06686	Wanhui 86
13	Landrace	32-00092	Zaohuangai	53	Breeding material	ZD06687	Wanhui 910
14	Landrace	32-00093	Hongmiaoxiang	54	Breeding material	ZD06688	Lehui 101
15	Landrace	32-00094	Yangcenggu	55	Breeding material	ZD06689	Lehui188
16	Landrace	32-00095	Qimiaoxiang	56	Breeding material	ZD06690	Ranhui
17	Landrace	32-00096	Yijudao	57	Breeding material	ZD06691	Shan B
18	Landrace	32-00097	Zhongzhou No.1	58	Breeding material	ZD06692	Jianghui 151
19	Landrace	32-00098	Beizinuo	59	Breeding material	ZD06693	Yangfu No.6
20	Landrace	32-00099	Honggu	60	Breeding material	ZD06694	Yihui 1577
21	Landrace	32-00100	Kehui 675	61	Breeding material	ZD06695	Yixiang B
22	Landrace	32-00101	Yapengzi	62	Breeding material	ZD06696	Luhui 17
23	Landrace	32-00103	Fuyin No.1	63	Breeding material	ZD06697	Yu 69-1
24	Landrace	32-00104	Fuyin No.2	64	Breeding material	ZD06698	Zheng B
25	Landrace	32-00105	Fuyin No.3	65	Breeding material	ZD06699	Jin 23B
26	Landrace	32-00106	Fuyin No.4	66	Breeding material	ZD06700	Kehui 10
27	Landrace	32-00107	Jinmazhan	67	Breeding material	ZD06701	Kehui 26
28	Landrace	32-00108	Zhandao	68	Breeding material	ZD06702	Kehui 36
29	Landrace	32-00109	Zhandao 69-1	69	Breeding material	ZD06703	Gui 99
30	Landrace	32-00110	Zhandao 69-3	70	Breeding material	ZD06704	Miyang 46
31	Landrace	32-00111	Huangkegu	71	Breeding material	ZD06705	Mianhui 527
32	Landrace	32-00112	Huangbianzhan	72	Breeding material	ZD06706	Fuhui 130
33	Landrace	32-00113	Xinxiangdao	73	Breeding material	ZD06707	Fuhui 838
34	Landrace	32-00114	Aihuanggu	74	Breeding material	ZD06708	Shuhui 162
35	Landrace	32-00115	Nuo 89-1	75	Breeding material	ZD06709	Shuhui 527
36	Landrace	ZD06673	77D	76	Breeding material	ZD06710	Qianhui 15
37	Breeding material	ZD06670	44D(1)	77	Breeding material	ZD06711	Qianhui 718
38	Breeding material	ZD06671	44D(2)	78	Breeding material	ZD06712	Nuo 89-1/725
39	Breeding material	ZD06672	46B	79	Breeding material	ZD06713	Nuuo 89-1/838
40	Breeding material	ZD06674	88/Jinmazhan				

germplasm resources in China, and they are rich in genetic diversity and contain a large number of excellent genes such as stress resistance, high yield, high quality and wide adaptability [8-9]. Therefore, to study the genetic diversity of the resource quality genes of the distinctive landraces of rice, and to fully exploit and utilize the genetic potential of the local germplasm resources of rice is an critical way to improve the quality traits of rice and realize the transformation of germplasm resources to genetic resources [5].

At present, there are many studies on genetic variation and genetic structure of local varieties and selected varieties at the molecular level in China [10-12], but the comparative analysis on genetic diversity of genes related to starch synthesis of different resource types is rarely reported. In this study, genetic variation, population structure and quality characteristics of genes related to starch synthesis were analyzed for 36 local varieties and 43 breeding materials in the upper reaches of Yangtze River in China by using internal molecular markers

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Table 2. The molecular markers of starch synrhesis related genes

Gene	Molecular markers	Sequence of primers(5'~3')	Marker type
GBSS Ⅱ	*GBSSII* M1	F:TTGCTGCGAATTATCTGCG; R:ACCTCCTCCCACTTCTTTGC	STS
Wx	*Wx* M1	F:CACAGCAACAGCTAGACAACCAC; R: CACGACGACGGAGGGGAAC	STS
SBE2	*SBE1* M2	F:GTGGGGAAAACAAGTAAGTCTG; R:AGTTCCATCAGAAGAATCAGGG	STS
	SBE1 M3	F: GGAAATGGGAGTCGCC; R: CGAAGAAACCACGCTCA	STS
SBE3	*SBE3* M1	F:AAGGTTAGCATTGGTTGGTGAG; R:TCTCCTTGAACAGCGACAGC	STS
	SBE3 M2	F:GTGGGGTTCTCAACTTAGC; R:CATCAGCATTGTTAGGCAG	STS
SBE4	*SBE4* M1	F:CACCAATTATATTAGCGTGCTCC; R:CGTGGCTCTTGGCTCTCTTG	STS
	SBE4 M2	F:CCATCACCTCAAATACATCACTC; R:AGACTGGAATGCCCCTTAGG	STS
SS Ⅰ	*SSI* M2	F:CTTCTATCCATTCCTTAATCCCA; R:ATGCTATTGATGTTAAGAGGGC	STS
SS Ⅱ *-1*	*SSII-1* M1	F:CACCCCACCGTTCTACTATGC; R:TCCATAGTTTCATTGAGATTGCTC	STS
	SSII-1 M3	F:AGAGATCAAATCGTGGAAC; R:TGGAGTGAAGTAGTGGAAT	STS
	SSII-1 M4	F:ATCTTTAGACGATTAGCG; R:AAGTCACAAGTAGAAGGG	STS
SS Ⅱ *-2*	*SSII-2* M1	F:AGATTTGAACTCAGGACTTGGTG; R:TCTATGGGCTCTATCCTTACTAGG	STS
	SSII-2 M2	F:CGCTCGTTGCCTAGCTAGC; R:GGCGAGGAAGCGATTGCC	STS
	SSII-2 M3	F:ACAGTATGTTTGCCTCAGCG; R:GTAAATCCACCCAGCCAGTC	STS
SS Ⅱ *-3*	*SSII-3* M1	F:CCAATACCGTAAACTAGCGACTATG; R:TACAGGTAGAATGGCAGTGGTG	STS
SS Ⅲ *-1*	*SSIII-1* M1	F:AAGAAGGGAAGGGAGTCAGC; R:GCCATCTCCATTGCCAGC	SSR
SS Ⅲ *-2*	*SSIII-2* M2	F:GAACTTGTGCCTTAAGCTGACTG; R:GGAATAGTAAGCCGAAGGACTT	STS
ISA	*ISA* M1*	F:ATAGATGCTAATGTGATGTGGC; R:TGGTATAGGCACAACCGTAGA	STS
PUL	*PUL* M3	F:CTGTATGGACTGAGTAGTCGATGG; R:TGAGCCTCATCTGCCAGAGT	STS
	PUL M4	F:TACACCATCCTCACTACCA; R:GCAACATCTAAAACACCAA	STS
	PUL M5	F:ATTGGCATTTGTAAGTTTC; R:CAATCTTGGTTTTATCCTG	STS
AGPlar	*AGPlar* M1	F:CGTTCAGGTTCAGGCAATCA; R:GGAAGGGTGGTGATGTGGAG	STS
AGPiso	*AGPiso* M2	F:CAATCGCTGCCATCGGTTG; R:TTCCACATCGTTAGGTACACG	STS
AGPsma	*AGPsma* M1	F:TCTATTCTCAGCCCTCCAACC; R:GTGTGTTTAGAGGTGCTTTTCG	STS

of genes closely related to rice quality, and clustering analysis were conducted for 79 specific rice species resources based on genetic similarity coefficients between materials.

2. Materials and Methods

2.1 Rice Varieties

A total of 368 resources from 31 districts and counties in the upper reaches of the Yangtze River in China were collected and identified and evaluated according to the "rice germplasm resource description specifications and data standards". Eighty-one specific resources were selected for accurate identification, traditional species cataloguing and enter the national long-term germplasm resource database for preservation and utilization http://www.cgris.net (National unified code: ZD06669--ZD06713; 32-00080-32-00115). The experimental study was carried out using 79 rice resources. The specific numbers and resource names are shown in Table 1.

2.2 Field Trial

The experiment was carried out in the rice test base of Chongqing Normal University from 2017 to 2018. Each material was planted in 3 rows with 12 holes per row. The planting density was 16.7 cm × 26.7 cm. The single seed was inserted, and the spacing between the materials was 33.33 cm. Organic fertilizer and chemical fertilizer were applied in combination, and applying fertilizer with massive base and early topdressing. The application rate of pure nitrogen was 120-150 kg/hm^2 and the ratio of nitrogen, phosphorus and potassium was 6:3:1. After harvesting the middle of the middle ten strains, after drying, the rice seeds are dried in a drying oven at 40 °C for 48 h, and stored in a dry environment for later use; the test analysis of the quality traits of the rice materials is carried out on time.

2.3 DNA Extraction

Fresh leaves were taken back to the laboratory 30 days after sowing, and DNA was extracted and purified by the CTAB method such as Murry and Thompson [13] for PCR analysis. The PCR reaction system was 10 μL, including DNA template of 1 ug, adding 2.5 mmol/L dNTP Mixture 0.2 μL and 2.5 mmol/L 10×PCR Buffer (Mg2+) 1 μL, and 0.2 μL mixture of pre-and post-primers (the concentration of primers were 12.5 umol/L-1), and plus 5 U/L Taq DNA polymerase 0.08 μL. Finally, the volume was set to 10 μL with ddH2O. The procedure of PCR was to pre-denature at 94 ℃ for 5 min, denatured at 94 ℃ for 50 s, annealed at 55 ℃ for 50 s, reached at 72 ℃ for 1 min, 30 cycles, and lastly extended at 72 ℃ for 10 min and stored at 4

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℃ . The annealing temperature in the reaction system depended on the specific primers. The amplified products were detected by 8% polyacrylamide gel.

2.4 Molecular Marker Detection

In this study, 26 molecular markers were selected from 16 starch synthesis related genes in rice. These markers were used to analyze the genetic diversity of 79 rice materials in the upper reaches of the Yangtze River in China. All marking information was provided by Professor Tian Zhixi's [14] Laboratory, and the marker information is listed in table 2.

2.5 Quality Measurement Indicators and Methods

The amylose content (AC) of 79 resources was established with reference to the Ministry of Agriculture standard NY/T 2639-2014; the gel consistency (GC) was determined in accordance with the national standard GB/T 22294-2008; the gelatinization temperature (GT) was established by alkali digestion method, and the gelatinization temperature was expressed by alkali digestion value (ASV). The quality grade of traits was classified according to the national standard GB/T17891-1999.

2.6 Statistical Analysis

Data collation and statistical analysis were completed in Microsoft Excel and IBM SPSS Statistics 22.0 software; genetic diversity analysis was carried out using Power-Marker 3.25[15] software; genetic similarity coefficient was calculated using NTSYS 2.1[16] software, and cluster analysis was carried out according to non-weighted pairing method (UPGMA). Structure 2.3.1 software was used to complete the population genetic structure analysis of 79 tested materials. Referring to Evanno et al.'s [17] method, the preset population subsets K ranged from 1 to 10, each K value was repeated 11 times, the Length of burn-in period was set to 10,000 at each run time, and Marko chain monte Carlo was placed to 100,000, and the optimal subsets were determined according to the maximum likelihood principle.

3. Results

3.1 Diversity Analysis of Starch Synthesis-related Genes

Using 26 molecular markers linked to starch synthesis-related genes, different alleles were detected after amplification with different primers (Table 3). A total of 53 alleles were detected in 36 landraces, with an average of 3.31 alleles per locus. Four alleles were detected in 8 alleles, and the least number of alleles were SBE4, SSI and SSII-1, all of which had only two alleles. The average genetic diversity of local varieties was 0.4339, and the variation range was 0.054~0.6157. The variation range of polymorphic information content (PIC) was 0.0526~0.5374, and the average was 0.3767. Three high polymorphic loci (PIC > 0.5) were detected, 11 moderate polymorphic loci (0.25 < PIC < 0.5) and 2 low polymorphic loci (PIC < 0.25) were detected.

Table 3. The genetic diversity of molecular markers in starch synthesis related genes

	Landrace				Breeding material		
Gene	No.of alleles	Gene disversity	PIC		No.of alleles	Gene disversity	PIC
GBSS Ⅱ	4	0.5340	0.4852		4	0.4532	0.4247
Wx	4	0.3657	0.3302		3	0.2455	0.2247
SBE1	4	0.2948	0.2797		3	0.1320	0.1273
SBE3	4	0.5586	0.4884		3	0.4240	0.3653
SBE4	2	0.3457	0.2859		3	0.4002	0.3491
SS Ⅰ	2	0.4614	0.3550		3	0.4932	0.4223
SS Ⅱ *-1*	2	0.0540	0.0526		2	0.1298	0.1214
SS Ⅱ *-2*	3	0.4799	0.3884		3	0.3916	0.3310
SS Ⅱ *-3*	4	0.6157	0.5374		3	0.5733	0.4842
SS Ⅲ *-1*	4	0.3349	0.3137		3	0.4240	0.3653
SS Ⅲ *-2*	4	0.5293	0.4770		3	0.2791	0.2510
ISA	3	0.4954	0.3972		2	0.3310	0.2762
PUL	3	0.4614	0.3776		3	0.4348	0.3584
AGPlar	3	0.5910	0.5141		3	0.4824	0.4244
AGPsma	4	0.5725	0.5116		5	0.5354	0.5002
AGPiso	3	0.2485	0.2335		2	0.0454	0.0444
Mean	3.31	0.4339	0.3767		3	0.3609	0.3169

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A total of 48 alleles were detected in 43 breeding materials, with an average of 3 alleles per locus. Among them, the highest allele mutation rate was AGPsma locus, with 5 alleles; the lowest number of alleles was detected at AGPiso, ISA and SSII-1 loci, all with 2 alleles. The average genetic diversity was 0.3609, ranging from 0.0454 to 0.5733. The variation range of polymorphic information content (PIC) ranging from 0.0444 to 0.5002, and the average PIC value was 0.3169. There was only one high polymorphic locus, 11 moderate polymorphic loci and 4 low polymorphic loci.

Generally, the richness of allele variation is positively correlated with PIC, that is, the richer the allele variation is, the higher the PIC will be. The more moderate polymorphic loci showed that the genetic differences of 79 tested materials were small, and the genetic basis was relatively narrow. The contrast of the average polymorphic information content between the two types of rice germplasms was 0.07, which indicated that there were some differences in genetic variation level between the two kinds of rice germplasms. Though the genetic diversity and PIC of local cultivars were slightly lower than those of breeding materials at some loci, the average genetic diversity and PIC values were higher than those of the breeding materials.

3.2 Performance of Starch-related Quality Traits

There were significant differences in the performance of the three starch-related quality traits in 79 rice resources (Table 4), and the Max-Min of each quality trait is large. The coefficient of variation is variable that represents a unit amount and can be used to compare the magnitude of variation between different characteristics. The ratio of difference of all three quality traits exceeded 30%, and they were alkali spreading value > gel consistency > amylose content from large to small.

Table 4. Analysis of variation of rice quality traits

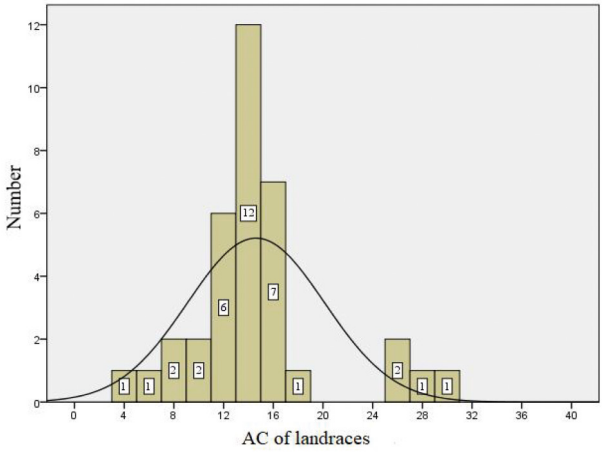
Project	Variation range	Max-Min	Mean	SD	CV/%
Alkali spreading value	1~7	6	3.23	1.85	57.34
Gel consistency /mm	11.2~148.6	137.37	83.08	37.78	45.47
Amylose content /%	3.01~30.72	27.71	15.57	6.08	39.03

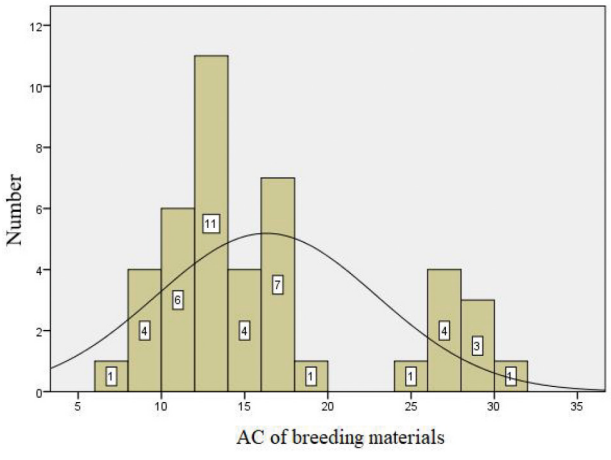
From the high-quality compliance rate of each trait (Table 5), the gel consistency is the best, and the varieties that meet the First-class quality standard are up to 54.4%. The compliance rate of alkali spreading value is low, and the international first-class high-quality rice compliance rate is 8.9%, which is slightly higher than the amylose content. The lowest compliance rate is the amylose content, only 6.3% of the varieties reach the first-class high-quality rice standard, while the proportion of the types that meet the third-grade high-quality rice is only 22.7%.

Table 5. Analysis of excellent rate of rice quality traits

Project	International grade Ⅰ quality rice	International grade Ⅱ quality rice	International grade Ⅲ quality rice	First-class attainment(%)	Secondary attainment (%)	Three-class attainment(%)
Alkali spreading value	6~7	5~6	≥4	8.9	12.7	36.7
Gel consistency /mm	≥70	≥60	≥50	54.4	64.6	77.2
Amylose content /%	17~22	16~23	15~24	6.3	15.2	22.7

3.3 Distribution of Starch-related Quality Traits





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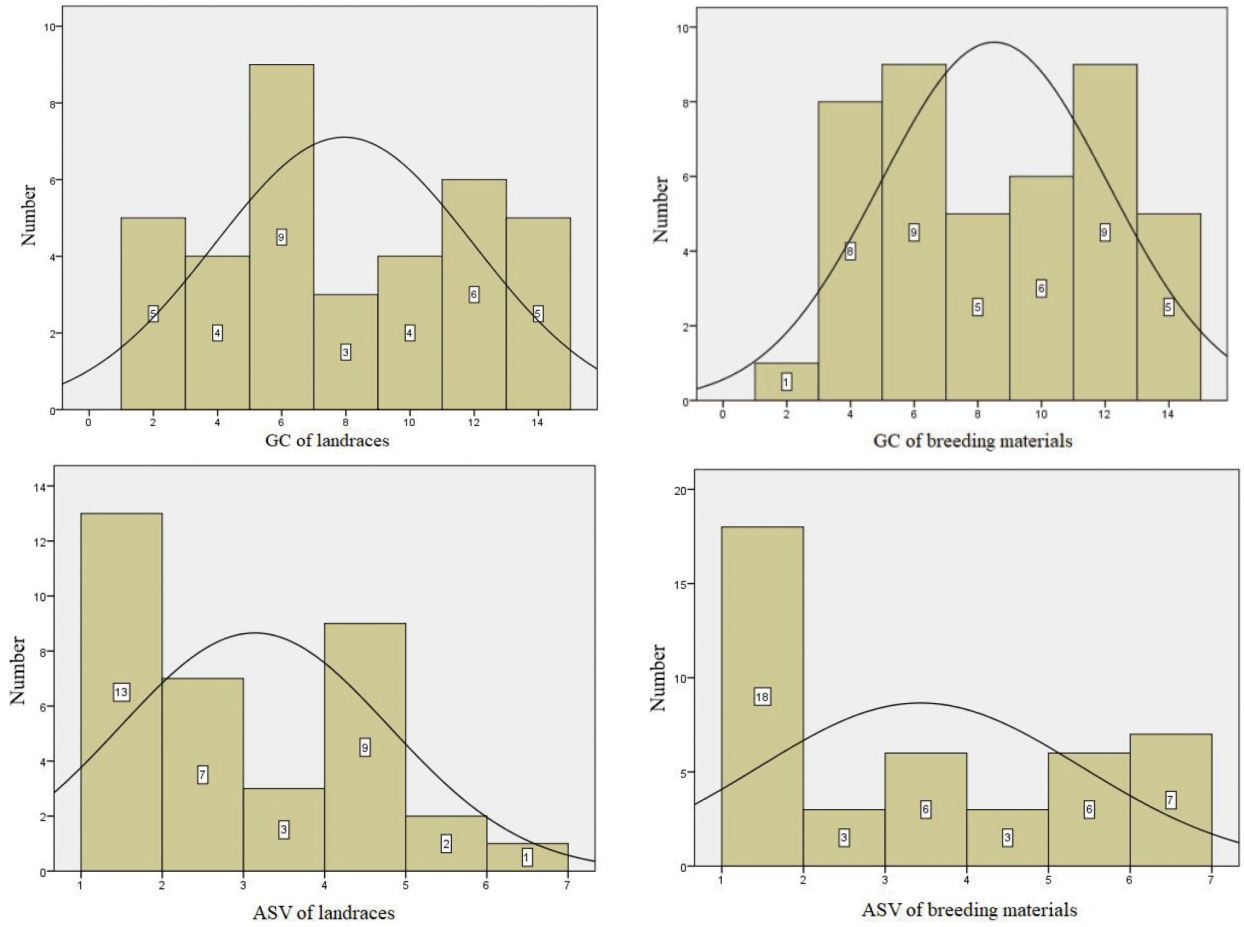

Figure 1. The distribution of several quality traits in different rice varieties

The amylose content (AC), gel consistency (GC) and alkali spreading value (ASV) of 79 rice cultivars in the upper reaches of the Yangtze River in China were determined to determine the quality differences between the tested rice varieties. The results showed that there were significant variations in the quality traits of different types among the 79 certified types (Fig. 1). The AC variation range of 36 rice landraces ranged from 3.0% to 30.0%, of which the AC distribution of 12 varieties was between 14.0% and 16.0%. The AC variation of the breeding materials ranged from 6.0% to 32.0%, a large number of types (including 11 types) distributed between 12.0% and 14.0%. The GC variation of landraces and Breeding materials is between 1.0 and 14.0 cm. The distribution of landraces is scattered, and nine varieties are distributed between 5.0 and 7.0 cm. The breeding materials have nine cultivars in the range of 5.0-7.0 cm and 11.0-13.0 cm, respectively. The rice varieties of the two resource types have similar alkali spreading value (ASV) distributions, with fluctuations ranging from 1 to 7. Rice varieties with ASV values between 1 and 2 are the most.

3.4 Genetic Similarity Coefficient and Cluster Analysis

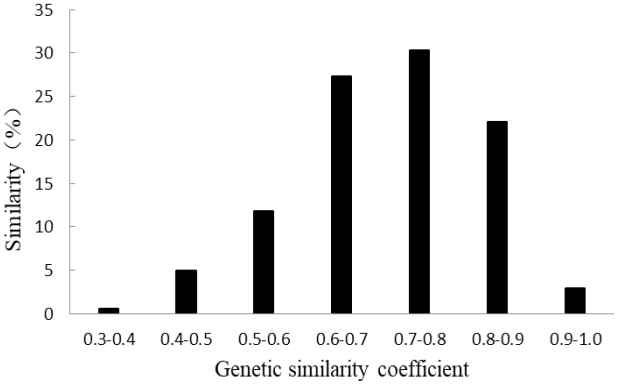

Figure 2. Frequency distribution of similarity coefficients in 79 rice seed resources

Calculating genetic similarity coefficients using NTSYS 2.1, the genetic similarity coefficients of 79 specific rice resources were typically distribution (Fig. 2). Among them, the genetic similarity coefficient was only 0.52 %

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below 0.5, 25.02% was above 0.8, the genetic similarity coefficients of 0.5~0.6 and 0.6~0.7 accounted for 11.85% and 27.30%, respectively, while the genetic similarity coefficient was 0.7~0.8, which was the highest, reaching 30.32%. It indicated that the genes related to starch synthesis showed high genetic similarity among most varieties, and the variation of alleles among most types was small.

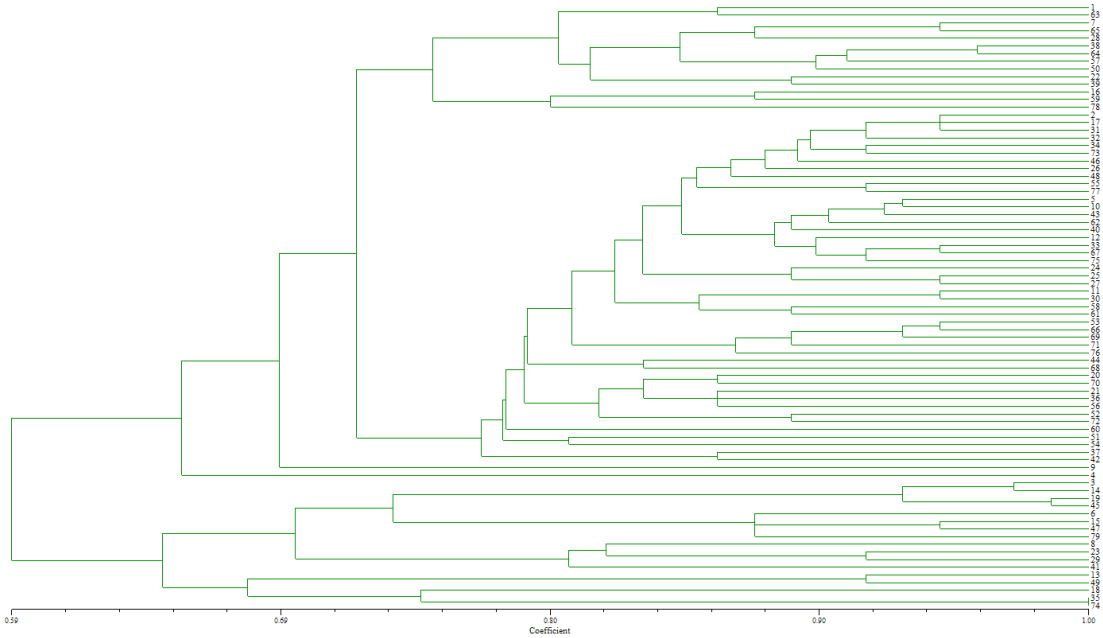

Figure 3. Clustering analysis based on genetic distance

The clustering results of starch synthesis related gene markers showed that the variation of GS of 79 rice resources was 0.324~1, with an average of 0.706 (Fig. 3). Among them, the GS values between Nuo89-1 and Shuhui162 were the largest (up to 1.000), indicating that the two were highly similar in 26 marker sites. If further distinction is needed, some markers need to be added. The GS value between the early yellow dwarf and sticky rice 69-3 was the smallest (0.324), indicating that the genetic relationship was far.

At the GS value of 0. 630, 79 tested materials were divided into two categories (Fig. 3). The first category consists of 11 local varieties and 6 growing materials, and the remaining 62 elements are the second largest category. The second category can be further divided into three sub-categories at the GS value of 0.69. Most of the breeding materials (37 species) are included in the 60 materials of the first sub-category, and the second sub-category and the third sub-category are respectively one local variety (Sanbaibang, Niushizhan). The clustering results of starch synthesis related gene markers showed that local variations and breeding materials existed in different groups at the same time, indicating that there is a specific genetic relationship between the two resource types.

3.5 Population structure of 79 rice varieties

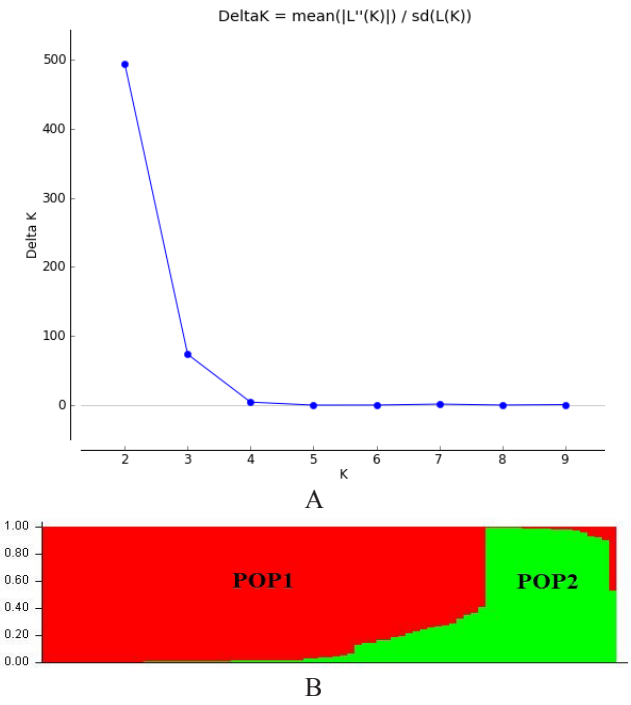

Figure 4. Population genetic structure of 79 rice materials. (A). △K value changes with the number of subgroups; (B). Population structure map based on 26 molecular markers

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Based on the results of molecular marker detection of starch synthesis genes, population structure analysis of 79 specific rice resources was performed using Structure2.3.1 software. As the sample's allelic variation frequency feature type number K (K=1~10) continues to increase, ΔK peaks at K = 2 (Fig. 4A), from which it is concluded that the test material can be divided into two subgroups. The subgroup classification was based on a genetic component value greater than 0.5. The results of the population structure analysis showed that (Fig. 4B), 61 of the 79 rice materials were classified into the first subgroup (POP1), including 25 local varieties and Of the 36 breeding materials, the remaining 11 landraces and 7 breeding materials were classified in the second subgroup (POP2). Landraces and breeding materials are scattered in different sub-groups, and there is a certain genetic exchange between them.

4. Discussion

The success of crop breeding is closely related to the genetic variation of germplasm resources[14] There are many analyses on the genetic diversity of rice population using molecular markers at home and abroad. For example, Deng Hongzhong[18] et al. used 98 pairs of SSR markers to compare the genetic diversity of 202 Chinese rice varieties and selected varieties. Hua Lei[19] et al. used 40 pairs of SSR markers to analyse the genetic diversity of 151 conventional rice varieties in different eras in China. Sun CQ [20] et al. used RFLP markers to evaluate the genetic diversity of ordinary wild rice and cultivated rice in various Asian countries. However, the molecular markers selected for a large number of scientific studies have not been associated with specific phenotypic traits for correlation analysis, and it is difficult to accurately and efficiently play a role in the genetic improvement of essential quality traits of crops [21-22].In this study, 26 intramolecular markers related to rice starch synthesis genes were used to detect allelic variation of rice landraces and breeding materials in the Yangtze River Basin. At the same time, combined with the distribution of three quality traits of amylose content, gel consistency and alkali spreading value, the comparative analysis and evaluation of genetic diversity among different resource types of rice were carried out.

The coefficient of variation of starch-related quality traits and the analysis of international high-quality compliance rates of various characteristics showed that the coefficients of variation of amylose content, gel consistency and alkali spreading value were significant, indicating that these three traits have a full separation range among 79 samples, and the genetic variation is rich. The excellent grade 1 compliance rate of amylose content and gel consistency was lower and less than 10%, indicating that increasing amylose content and gel consistency grade are the key to improving the quality traits of specific rice germplasm resources in the Yangtze River basin. According to the distribution of three quality traits in landraces and breeding materials, the breeding materials show better taste quality, which may be related to the long-term artificial orientation selection of the cultivating materials, and the quality traits of rice varieties are improved and gradually approached to high quality. Among them, the landraces 44D (2) amylose content, gel consistency and alkali spreading value have reached the international level 1 standard, with good starch quality traits, can be applied to the genetic basis of starch synthesis genes and rice breeding.

Many previous studies have shown that rice landraces have higher genetic diversity than modern breeding materials[23-26] The results of this study showed that 26 intramolecular markers of starch synthesis-related genes detected 53 alleles in 36 landraces, with an average genetic diversity of 0.4339 and an average PIC of 0.3767. Although the genetic diversity was slightly lower than the breeding materials at the AGPsma locus, the average genetic diversity and PIC value of the landraces were higher than those of the cultivating materials. In general, the overall genetic diversity of the two resource types is low. According to the results of molecular marker polymorphism detection and genetic similarity analysis, the varieties with a genetic similarity coefficient above 0.7 accounted for 55.34%, the variations below 0.5 were only 5.52%, and the GS range was 0.323~1, with an average of 0.706. It indicates that the genes related to starch synthesis show high genetic similarity among most varieties, and the genetic relationship is relatively close. This may be associated with the geographical location of the test materials in this study, and the number of primers selected is less. These molecular data can be directly used for correlation analysis of starch quality traits, which lays a foundation for excavating the excellent allele loci of rice starch synthesis related genes.

Ao Yan[27] carried out molecular marker-based cluster analysis on 115 rice landraces and 87 breeding materials in Taihu Lake Basin, and found that there were some genetic differences between breeding materials and local varieties. Tang Zhiming[28] et al. carried out cluster analysis based on genetic distance for 50 types of conventional rice in different periods in Guangzhou. It was found that with the increase of years, the genetic distance between variations in each period slowly decreased, and the varieties with distant relatives gradually disappeared. Ma Jing[29] et

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al. UPGMA cluster analysis of 31 selected types in Ningxia showed that the genetic diversity of rice breeding materials in Ningxia has improved in recent years, but the genetic basis is relatively narrow. In this study, the results of cluster analysis and population structure analysis were highly consistent. 36 breeding materials and 43 local varieties existed in different subgroups, and the genetic relationship was relatively close, similar to that in other parts of China.

Among them, the landraces Sanbaibang and Niushizhan have a distant relationship with other types, and the four local varieties of Hongmiaoxiang, Yangcenggu, Zhandao69-1 and Nuo89-1 have good starch-related quality and can be prioritized in the selection of future new breed parent materials. In modern breeding, make full use of the quality genetic diversity of landraces, to broaden the genetic basis of unique varieties of rice breeding in the upper reaches of the Yangtze River in China, and to reduce the loss of beneficial genes in landraces.

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