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Genetic diversity and relationships among loose-curd cauliflower and related varieties as revealed by microsatellite markers

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ABSTRACT

Loose-curd cauliflower is widely cultivated in China for its excellent quality, but most of the genetic studies have used the compact-curd variety that is more popular in Europe and America. In the present study, we analyzed the genetic diversity of 57 *Brassica oleracea* genotypes worldwide, including 33 loose-curd cauliflower accessions, by using 14 simple sequence repeat primer pairs. The overall genetic similarity for all the cauliflower genotypes was 0.74 but that of the loose-curd cauliflower accessions was 0.83. The genetic similarity values were used to conduct a two-dimensional principal coordinate analysis and a cluster analysis that generally classified loose-curd cauliflower and compact-curd cauliflower into separate groups. Cauliflower accessions that displayed early maturity or had close geographic origins tended to be clustered together. On the basis of these results, 49 cauliflower genotypes were classified into three relatively independent types (loose-curd cauliflower from China, compact-curd cauliflower from China, and compact-curd cauliflower from Europe and America). This study suggested that commercial cauliflower cultivars have a very narrow genetic diversity, and that it is important to enhance gene introgression among different types to improve their genetic diversity.

1. Introduction

Cauliflower (*Brassica oleracea* var. *botrytis*) is an important vegetable crop that is cultivated worldwide. It is characterized mainly by its specific edible "curd", which displays rich polymorphism in morphology and chemical makeup. In addition to the most common white and compact curded cauliflower, several other forms that possess varying antioxidant profiles, such as pigmented curd cauliflower and Romanesco cauliflower, are still grown in some regions (Branca, 2008; Branca et al., 2002; Lo Scalzo et al., 2008). Several recent studies (Gu et al., 2012; Zhao et al., 2012, 2013) have noted the popularity of another special form called loose-curd cauliflower in China, especially in Zhejiang and Fujian Provinces. Although post-harvest storage and transport of the loose-curd variety is difficult, it is well-known for its quality, which is mainly due to its tender and crisp floret stems and high level of soluble sugar content (Gu et al., 2012). Therefore, the loose-curd cauliflower is a high-profit vegetable crop in China.

The loose-curd variety was first cultivated in Fujian Province and later introduced to other areas in China. Because of the economic importance of this variety, a number of genetic improvement programs have been conducted, mainly to enlarge curd size and improve curd shape. Other goals of the improvement programs were to enhance the green color of the floret stems, reduce plant size so that crops may be planted more densely, and increase resistance to biotic and abiotic stress. The cultivars that resulted from such intense selection represent a narrow portion of the cauliflower gene pool and therefore have low genetic diversity. Accordingly, assessing genetic diversity among loose-curd cauliflower cultivars can help breeders understand the need to enhance genetic variation.

Microsatellites, also known as simple sequence repeats (SSRs), are short tandem nucleotide repeats that are widely known for their high level of frequency of length polymorphisms. Since they are stable, highly informative, easy to detect, and relatively inexpensive to use in research, SSR markers are routinely used for purposes such as assessing genetic diversity and tagging genes (Formisano et al., 2012; Powell et al., 1996). Recently, there has been a great increase in publicly available SSR markers for *B. oleracea* species (Burgess et al., 2006; Iniguez-Luy et al., 2008; Louarn et al., 2007; Lowe et al., 2004; Tonguc and Griffiths, 2004). Tonguc and Griffiths (2004) used 13 SSR markers from *B. oleracea* to demonstrate that cauliflower cultivars had the lowest genetic diversity among cultivars of three botanical varieties of *B. oleracea*. In another cluster analysis that included five varietal groups, all the cauliflower accessions formed a completely separate cluster, indicating their independent origin

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Table 1
Brassica oleracea accessions evaluated for SSR polymorphisms in this study.

Code	Accessions	Varietal group	Curd description	Origin	Maturity[a]	Type
1	Bisheng	Cauliflower	Loose curd	Fujian, China	100 d	Open pollinated cultivar
2	C12011	Cauliflower	Loose curd	Zhejiang, China	50 d	F_1 hybrid
3	Cangnan	Cauliflower	Loose curd	Zhejiang, China	100 d	Landrace
4	Changsheng	Cauliflower	Loose curd	Shaihai, China	90 d	F_1 hybrid
5	Dadi	Cauliflower	Loose curd	Fujian, China	55 d	F_1 hybrid
6	Fengtian	Cauliflower	Loose curd	Fujian, China	80 d	F_1 hybrid
7	Fujian	Cauliflower	Loose curd	Fujian, China	80 d	Open pollinated cultivar
8	Haochi	Cauliflower	Loose curd	Zhejiang, China	80 d	Open pollinated cultivar
9	Huawang	Cauliflower	Loose curd	Jiangxi, China	90 d	Open pollinated cultivar
10	IB01	Cauliflower	Loose curd	Netherlands	90 d	Inbred line
11	IB31	Cauliflower	Loose curd	France	110 d	Inbred line
12	Jialijia	Cauliflower	Loose curd	Fujian, China	80 d	F_1 hybrid
13	Jimei80	Cauliflower	Loose curd	Fujian, China	80 d	F_1 hybrid
14	Jinsong66	Cauliflower	Loose curd	Tianjin, China	70 d	F_1 hybrid
15	Lvnong	Cauliflower	Loose curd	Jiangsu, China	75 d	F_1 hybrid
16	Nongle55	Cauliflower	Loose curd	Fujian, China	60 d	F_1 hybrid
17	Nongmei45	Cauliflower	Loose curd	Fujian, China	45 d	F_1 hybrid
18	Qingnong65	Cauliflower	Loose curd	Fujian, China	75 d	F_1 hybrid
19	Qingsong55	Cauliflower	Loose curd	Zhejiang, China	55 d	F_1 hybrid
20	Qingsong65	Cauliflower	Loose curd	Fujian, China	80 d	F_1 hybrid
21	Qingxiu	Cauliflower	Loose curd	Gansu, China	70 d	F_1 hybrid
22	Shuangfeng	Cauliflower	Loose curd	Hunan, China	110 d	Open pollinated cultivar
23	Songhua 1	Cauliflower	Loose curd	Zhejiang, China	80 d	F_1 hybrid
24	Taisong65	Cauliflower	Loose curd	Zhejiang, China	75 d	F_1 hybrid
25	Tongan	Cauliflower	Loose curd	Fujian, China	75 d	Landrace
26	Xiahua	Cauliflower	Loose curd	Fujian, China	70 d	F_1 hybrid
27	Xingnong	Cauliflower	Loose curd	Fujian, China	80 d	F_1 hybrid
28	Xingui	Cauliflower	Loose curd	Shandong, China	80 d	F_1 hybrid
29	Xinhong 1	Cauliflower	Loose curd	Zhejiang, China	75 d	F_1 hybrid
30	Yulong	Cauliflower	Loose curd	Yunnan, China	80 d	Open pollinated cultivar
31	Yunsong	Cauliflower	Loose curd	Shandong, China	90 d	F_1 hybrid
32	Zhe017	Cauliflower	Loose curd	Zhejiang, China	70 d	F_1 hybrid
33	Zhe091	Cauliflower	Loose curd	Zhejiang, China	85 d	F_1 hybrid
34	Jinpin60	Cauliflower	Compact curd	Tianjin, China	60 d	F_1 hybrid
35	Tezao50	Cauliflower	Compact curd	Zhejiang, China	50 d	F_1 hybrid
36	Dongya 3	Cauliflower	Compact curd	Liaoning, China	50 d	F_1 hybrid
37	Chongming cauliflower	Cauliflower	Compact curd	Shanghai, China	200 d	Open pollinated cultivar
38	Yinxue	Cauliflower	Compact curd	Shanghai, China	70 d	Open pollinated cultivar
39	Xuesheng	Cauliflower	Compact curd	Zhejiang, China	180 d	F_1 hybrid
40	Chenggong 1	Cauliflower	Compact curd	Zhejiang, China	100 d	F_1 hybrid
41	Chenggong 2	Cauliflower	Compact curd	Zhejiang, China	120 d	F_1 hybrid
42	Zhonghua 1	Cauliflower	Compact curd	Beijin, China	60 d	F_1 hybrid
43	CJ115	Cauliflower	Compact curd	Japan	60 d	F_1 hybrid
44	Deyou100	Cauliflower	Compact curd	Italy	100 d	F_1 hybrid
45	Bai039	Cauliflower	Compact curd	America	90 d	F_1 hybrid
46	Xuepan	Cauliflower	Compact curd	America	100 d	F_1 hybrid
47	Xueba	Cauliflower	Compact curd	Netherlands	85 d	F_1 hybrid
48	Nanjixue	Cauliflower	Compact curd	Netherlands	90 d	F_1 hybrid
49	Xuejie70	Cauliflower	Compact curd	France	100 d	F_1 hybrid
50	Sailuoma	Cauliflower	Pyramidal curd	Zhejiang, China	90 d	Open pollinated cultivar
51	Zijing	Cauliflower	Purple curd	Japan	100 d	Open pollinated cultivar
52	Hailv	Broccoli	–	Zhejiang, China	70 d	F_1 hybrid
53	Youxiu	Broccoli	–	Japan	70 d	F_1 hybrid
54	Lvxiong90	Broccoli	–	Japan	90 d	F_1 hybrid
55	Xique	Broccoli	–	Japan	95 d	F_1 hybrid
56	G055	Cabbage	–	Zhejiang, China	80 d	F_1 hybrid
57	Qianglv60	Cabbage	–	Japan	60 d	F_1 hybrid

[a] Maturity was expressed by the days lasted from transplant to harvest when the accessions were cultivated in autumn in Zhejiang, China. Maturity less than or equal to 60 d was defined as early maturing.

and limited genetic variation relative to that of other *B. oleracea* varieties (Louarn et al., 2007). A more recent study revealed that kai-lan was more closely related to cauliflower than to the other varietal groups of *B. oleracea*, such as broccoli, cabbage, and kale (Izzah et al., 2013).

All the cauliflower accessions sampled in previous studies were compact curded forms obtained mainly from seed suppliers in Europe, Japan, and America. Therefore, a more comprehensive characterization of the genetic variation in cauliflower is required in order to explicate the relationship between loose-curd and compact-curd cauliflowers and understand the genetic diversity among loose-curd cauliflower cultivars. Here, we applied publicly available SSR markers to 33 loose-curd and 16 compact-curd cauliflower cultivars or lines and compared these with several related varieties of *B. oleracea*, in order to estimate the genetic diversity within loose-curd cauliflower and characterize the genetic relationships within the clade.

2. Materials and methods

2.1. Plant materials

A total of 57 *B. oleracea* accessions were used in this study, including 51 cauliflower varieties (33 loose-curd, 16 compact-curd,