# Karyotype Analysis of Two Kinds of Red Petioles Leaf Beet

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**Abstract.** Leaf beet is one of the important leaf vegetables and has important ornamental value. Red petioles leaf beet includes two kinds, green leaves and red leaves. They are new types of leaf beet and are popular with consumers. In this research, we try to obtain cytological parameters on these two leaf beet. Seven chromosomal parameters were measured and calculated: chromosome length, relative length, index of relative length, type of relative length, arm ratio, centromere index, and centromere type. The results showed that the maximum chromosome length of green leaves kind of red petioles leaf beet was measured 2.54 μm and max arm ratio was determined 1.75, relative length ranged between 8.19%-12.93%. The maximum centromeric index was measured in 47.43%, Karyotype asymmetry index was 57.16%, and the karyotype formula was 2n=18=16m+2sm (2SAT). The maximum chromosome length of red leaves kind of red petioles leaf beet was measured 2.92 μm and max arm ratio was determined 1.92, relative length ranged between 9.00%-12.86%. The maximum centromeric index was measured in 46.07%, Karyotype asymmetry index was 60.10%, and the karyotype formula was 2n=18=14m(2SAT)+4sm. The karyotype characteristics of two kinds of leaf beet were all type 1A. The findings revealed the karyotypic characteristics of leaf beet from the cytogenetic aspects, and inferred the revolution of red leaves kind of red petioles leaf beet is higher than green leaves kind.

## 1. Introduction

Leaf beet (Beta vulgaris var. cicla) belongs to Chenopodiaceae, and it is the variety of beet. Leaf beet is origin from Mediterranean coast, so it prefer warm and humid climate[1]. And it is the common leaf vegetable consumed in summer, for containing abundant nutrients. Besides, leaf beet is a common ornamental plant. Leaf beet has good high yield, disease resistance and adaptability. It is widely planted in China because of its good comprehensive characters. Varieties of leaf beet exist in the world, and they present significant difference in appearance. The difference in plant phenotype is mainly influenced by genetic material and environment, in which genetic material is the main factor. Karyotype analysis is a basic method to study chromosomes, it is a basic work in cytogenetics research. It has been reported that sugar beet has diploid, triploid and tetraploid[2]. However, the chromosomes of different types of plants, even different cultivars vary widely. In this experiment, the karyotype analysis was carried out on two kinds of red petioles leaf beet (green leaves and red leaves) to reveal theirs chromosome composition and diversity, and to provide the basis for determining the genetic composition of red petioles leaf beet.

## 2. Materials and Methods

### 2.1 Plant Materials.

There are two kinds of red petioles leaf beet was used as experimental material, including the representative Beta vulgaris cv. Jindiyongfeng with green leaves and Beta vulgaris cv. Hajinlong with red leaves (Table1).

Table 1. Experimental materials

| No | Cultivar | Kind |
|---|---|---|
| G | Jindiyongfeng | Green leaves |
| R | Hajinlong | Red leaves |

### 2.2 Chromosome Preparation.

The seeds were soaked for 2 h, then cultured in dark in petri dishes with moist filter paper at 25 °C incubator to the root length of 1-1.5 cm and cut root tips of about 1 cm. Pretreated in 0.002 mol·L-1 8-hydroxyquinoline at 4 °C for 9h, and fixed in Carnoy's solution (acetic acid: absolute ethanol, 1:3, v/v) at 4 °C for 24 h, subsequently, the root tips were macerated in 1 mol·L-1 hydrochloric acid at 60 °C for 12 min, stained with Carbol Fuchsin, and observed under microscope[3].

### 2.3 Karyotype Analysis.

Chromosome counts were performed on 30 well-spread metaphase chromosomes from five different root tips. Karyotype analysis referred to the standard of Li et al.[4]. Following parameters were calculated: chromosome relative length, arm ratio, type of chromosomes, index of chromosomes relative length and centromere index. karyotypic formula referred to the standard of Levan et al.[5], and the asymmetry coefficient of karyotypes was calculated by the method of Arano[6], the karyotypes were calculated according to Stebbins' standard[7].

## 3. Results

### 3.1 Chromosome Number of Green Leaf Beet and Red Leaf Beet.

Metaphase chromosomes and karyotype of two kinds of red petioles leaf beet root tips were shown in Fig. 1, detailed karyotype parameters of chromosome were listed in Table 2. The chromosome number of the two kinds of red petioles leaf beet both were 2n=18.

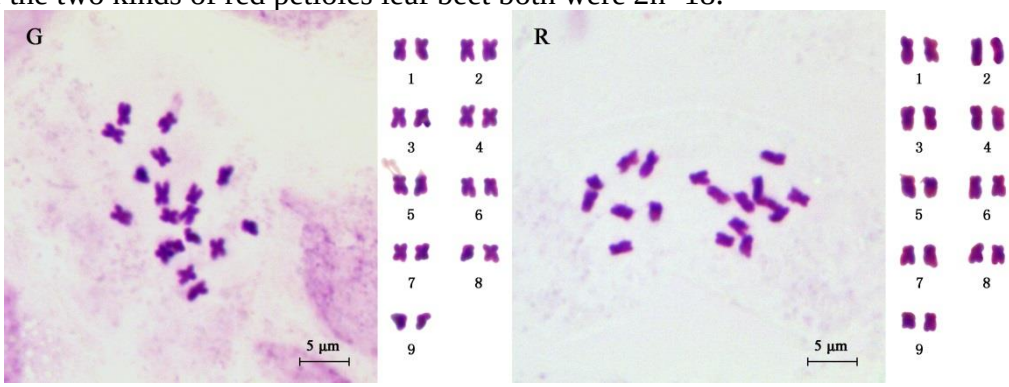


Fig. 1. Metaphase chromosomes and karyotype of two kinds of red petioles leaf beet root tips
Note: The number 1-9 represent chromosome No.

### 3.2 Karyotype Analysis.

The chromosome relative length of green leaves kind of red petioles leaf beat ranged from 8.19% to 12.93% and red leaf beat ranged from 9.00% to 12.86%, and chromosome length ratio (longest chromosome / shortest chromosome) of green and red leaves kinds of red petioles leaf beet were 1.58 and 1.43, respectively. The chromosome types included long chromosomes (L), medium long chromosomes2 (M2), medium short chromosomes1 (M1) and short chromosome (S), the relative length constitution of green leaves kind was 4M2+4M1+S, and red leaves kind was 5M2+4M1. The centromeric index of green leaves kind ranged from 36.33% to 47.43%, and arm ratio ranked from 1.11 to 1.75. The centromeric index of red leaves kind ranged from 34.19% to 46.07%, and arm ratio ranked from 1.17 to 1.92. There were one pair (the fifth chromosome) of submetacentric chromosomes (sm) and other eight pairs of metacentric chromosomes (m) in green leaves kind of leaf beat. And two pairs (the second and seventh chromosome) of submetacentric chromosomes (sm) and seven pairs (number One, three, four, five, six, eight, nine, chromosome) of metacentric chromosomes (m) in red leaves kind of leaf beat. Moreover, the green and red leaves kinds of red petioles leaf beet all had one pairs of satellites, and the two satellites were both observed at the five pair of chromosomes. The karyotype formula of green leaves kind of leaf beet was 2n=18=16m+2sm (2SAT), and red leaves kind was 2n=18=14m (2SAT)+4sm. Karyotype asymmetry index of green leaves kind was 57.16%, and red leaves kind was 60.10%. The karyotype characteristics of two kinds of red petioles leaf beet fell into type 1A according to Stebbins's classification criteria. The chromosome idiogram of green leaf beet and red leaf beet were shown in Fig. 2.

Table 2. Karyotype parameters of chromosome of two kinds of red petioles leaf beet

| No. | Chromosome No. | Relative length / % | | | Index of relative length | Type of relative length | Arm ratio | Centromere index / % | Centromere type |
|---|---|---|---|---|---|---|---|---|---|
| | | Short arm | Long arm | Total length | | | | | |
| G | 1 | 5.64 | 7.29 | 12.93 | 1.16 | M2 | 1.29 | 43.59 | m |
| | 2 | 5.14 | 7.36 | 12.51 | 1.13 | M2 | 1.43 | 41.12 | m |
| | 3 | 5.34 | 6.58 | 11.92 | 1.07 | M2 | 1.23 | 44.78 | m |
| | 4 | 5.38 | 6.38 | 11.77 | 1.06 | M2 | 1.19 | 45.74 | m |
| | 5* | 3.97 | 6.95 | 10.92 | 0.98 | M1 | 1.75 | 36.33 | sm |
| | 6 | 4.13 | 6.61 | 10.74 | 0.97 | M1 | 1.60 | 38.45 | m |
| | 7 | 5.06 | 5.61 | 10.68 | 0.96 | M1 | 1.11 | 47.43 | m |
| | 8 | 4.49 | 5.86 | 10.35 | 0.93 | M1 | 1.30 | 43.39 | m |
| | 9 | 3.69 | 4.50 | 8.19 | 0.74 | S | 1.22 | 45.11 | m |
| R | 1 | 5.18 | 7.67 | 12.86 | 1.16 | M2 | 1.48 | 40.31 | m |
| | 2 | 4.71 | 8.14 | 12.85 | 1.16 | M2 | 1.73 | 36.64 | sm |
| | 3 | 5.40 | 6.32 | 11.72 | 1.06 | M2 | 1.17 | 46.07 | m |
| | 4 | 4.64 | 6.79 | 11.43 | 1.03 | M2 | 1.46 | 40.59 | m |
| | 5* | 4.45 | 6.41 | 10.86 | 0.98 | M2 | 1.44 | 40.96 | m |
| | 6 | 4.17 | 6.51 | 10.68 | 0.96 | M1 | 1.56 | 39.05 | m |
| | 7 | 3.56 | 6.85 | 10.41 | 0.94 | M1 | 1.92 | 34.19 | sm |
| | 8 | 4.22 | 5.97 | 10.19 | 0.92 | M1 | 1.41 | 41.43 | m |
| | 9 | 3.57 | 5.42 | 9.00 | 0.81 | M1 | 1.52 | 39.71 | m |

Note: * means the chromosomes with satellites, and the length of satellites is not included in the chromosome length.

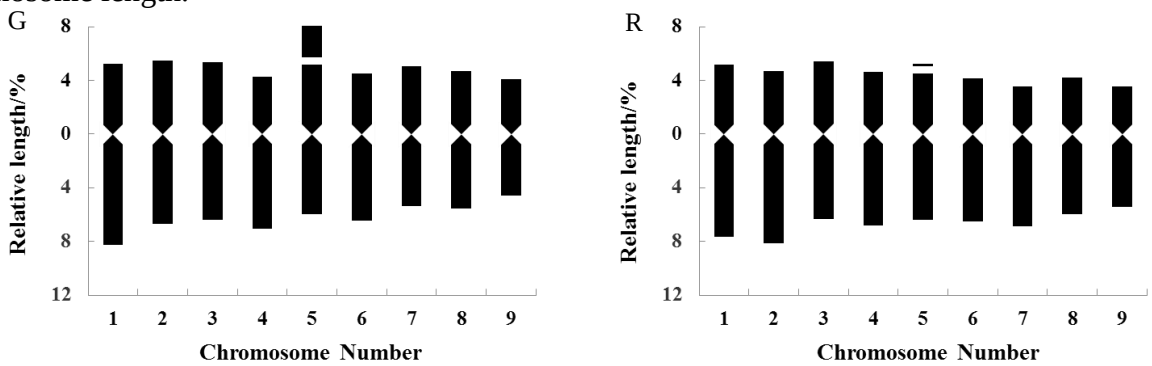


Fig. 2. Chromosome idiogram of green leaf beet and red leaf beet

## 4. Summary

The results of karyotype analysis of green leaves kind of red petioles leaf beet have consistent with red leaves kind at some aspects. All of them are diplont with 18 chromosomes, and they have a pair of satellites. Besides, the number of chromosomes with satellites is the fifth. The karyotype characteristics of two kinds of leaf beet all fell into type 1A. However, diversities were also existence between two kinds of red petioles leaf beet. The chromosomes composition is different between green and red leaves kinds. Green leaves kind of red petioles leaf beet have one pair of chromosomes is submetacentric chromosomes, while red leaves kind have two. And the results revealed that the karyotype asymmetry index of red leaves kind of leaf beet (60.10%) is higher than green leaves kind (57.16%). The basic evolutionary trend of plant karyotypes is from symmetry to asymmetry. Thus, primitive plants have symmetrical karyotypes. And the more asymmetric the plant karyotype is, the higher its degree of evolution [8]. That is implied the evolution of red leaves kind of red petioles leaf beet is higher than green leaves kind.

As reported by Chen, the sugar beet had three kinds of ploidy (diploid, triploid and tetraploid) [2]. However, the leaf beet only had one kind of ploidy (diploid). An increase in chromosome ploidy may induce the increased of plant size and nutrition content. The reason why the ploidy of leaf beet is single was inferred that human have no higher demand for its yield and nutrition content. Our research provides a reference for the genetic evolution of red petioles leaf beet.

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