

## SHORT COMMUNICATION

# Identification of Local Melon (*Cucumis melo* L. var. *Bartek*) Based on Chromosomal Characters

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**Bartek is one of local melon varieties mainly cultivated in Pemalang, Central Java. Bartek has three variations of fruits; Long-Green, Ellipsis-Green, and Yellow. Chromosome characterization of the Bartek was investigated to determine the genetic variation. The main purpose of this research was to determine the genetic characters of Bartek including chromosome number, mitosis, cell cycle, and karyotype. Squash method was used for chromosome preparation. The results showed that all of Bartek observed in this study have similar diploid ( $2n$ ) chromosome number = 24. According to the total number of chromosome, Bartek is closer to melon than cucumber. The mitotic analysis exhibited that the Bartek has similar karyotype formula,  $2n = 2x = 24m$ . Based on the  $R$  value of the three kinds of Bartek ( $R < 0.27$ ), it indicated that three kinds of Bartek were considered to be originated from similar species and one of melon varieties (*Cucumis melo* L. var. *Bartek*).**

Key words: *Cucumis* sp., Bartek, chromosome, karyotype

## INTRODUCTION

Melon (*Cucumis melo* L.) is a climbing herbaceous annual fruit vegetable belonging to the family Cucurbitaceae consisting of 90 genera and 750 species. It is a cross-pollinated diploid ( $2n = 2x = 24$ ) species. Melon was originally monoecious, as are many modern cucurbit plants, but gynoeceous and andromonoecious cultivars were subsequently bred. Male and female flowers were typically borne at different nodes, with the female flowers at higher nodes than the male. Fruits are round to ellips or narrowly cylindrical, with small tubercles and spines of trichome origin on the rind. *Cucumis* is a genus of about 25 species, mostly tropical African in distribution. Some are confined to tropical Asia. (Robinson & Walters 1999).

Melon is commonly planted in several regions in Indonesia. In Java and Sumatra Islands, melon planted in lowland area. It is usually used for table fruit, fresh consumed fruit, and cocktail. However, the improvement of melon quality is slow, especially for local variety of melon, because it has small and un-similar fruit shape. Most of farmers choose hybrid melon for this economic reason (Trehane *et al.* 1995). Bartek is one of local melon varieties which is mainly cultivated in Pemalang, Central Java. Bartek is usually planted in lowland farm on the dry season with 21-27 °C temperature and it needs 2 months to be harvested. Bartek is a unique local melon, since it has three variations of fruit in one cultivar; Long-Green, Ellipse-Green, and Yellow. Hence, chromosome studies of

Bartek, are absolutely needed due to their implications for melon breeding program. In addition, the melon industry is today critically dependant genetically on superior cultivars to improve fruit quality, disease resistance, and ability to tolerate in environmental stresses. The objective of this research was to examine the mitosis period, chromosome number, chromosome size, and the karyotypes of Bartek. The findings of the research are expected to enrich valuable information concerning the genetic identity of the Bartek to develop the quality of local melon through breeding program.

Chromosome characterization can be used to determine the genetic differences among plant cultivars. Stace (1979) stated that closer relation among the cultivars will show a higher similarity in their chromosome number. The chromosome number inside the nucleus was generally stable in each species (Kimball 1983). Chromosome characterization usually done by study the mitotic cell division, because the morphological characters of chromosome in this cell division is stable and can be seen clearly (Min *et al.* 1984). The whole components of chromosome inside the cell is called *karyotype*. *Karyotype* is an important tool to determine the identity of organisms, such as identify the taxonomical unit, biological diversity, evolution process analisis, and to determine the genetic abnormality of organisms (Jones & Luchsinger 1979).

## MATERIALS AND METHODS

**Plant Materials.** Bartek used in this study were: long-green Bartek, ellipsis-green Bartek, and yellow Bartek. Seeds

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of Bartek collected from Pemalang, Central Java were sown and germinated in Petri-dishes at Genetics Laboratory, Faculty of Biology, Gadjah Mada University, Yogyakarta. Those samples were collected in February 2007 on the dry season, when Bartek usually grown.

**Chromosome Preparation.** A chromosome preparation procedure of the Bartek followed those described in Jahier and Tanguy (1996). To analyze the chromosome characters of Bartek, the measurement of chromosome size was made from the chromosome observed at prometaphase. Chromosome preparation was conducted from 08.00 to 11.00 a.m. (WIB) with 15 minutes interval. Fresh root tips from germinated seeds of the Bartek were fixed in 45% acetic acid at 4 °C for 15 minutes. Fixed root tips were then macerated in 1 N hydrochloric acid for about 11 minutes at 55 °C. The root tips were stained in 1% aceto-orcin for about 24 hours before they squashed. The slides were then photographed using Olympus C-35-AD-4 and Fuji ASA 200 film.

**Chromosome Analysis.** The measurement of chromosome size was made on the chromosomes observed at prometaphase using Adobe Photoshop CS2 for Windows program. Centromere position of chromosome was classified by centromeric index calculated by short arm/total length accorded to Levan *et al.* (1964): metacentric chromosome with centromeric index of 37.50-50.00; submetacentric chromosome with centromeric index of 25.00-37.49; subtelocentric chromosome with centromeric index of 12.5-14.99; and telocentric chromosome with centromeric index of 0-12.49. Data of chromosome size and centromere position of chromosomes were then arranged to construct karyogram using Adobe Photoshop CS2 for Windows program, and ideogram using CorelDRAW Graphic Suite X3.

## RESULTS

**Fruits and Seeds Morphology of Bartek.** Bartek has three variations of fruit; Long-Green, Ellips-Green, and Yellow (Figure 1). However, there are no specific morphological differences among the plants producing those kinds of fruit. Bartek is generally used as a mix of a cocktail. The stems of Bartek are usually herbaceous, climbing, and branched. It can reach 138-215 cm in length with 0.7-1 cm diameters. The leaf of Bartek is about 13-18 cm long with 14-18 cm wide. The fruits of Bartek have many variations in size, but commonly, it is about 10-12 cm in length. The morphological characters differentiating the three kinds of Bartek's fruit are the color and shape of the fruits. The color of the flesh fruits of Bartek usually change from white or pale-yellow to orange when it starts to mature.

**Chromosomal Characters and Analysis.** To observe the chromosomal characters of Bartek, fresh root tips from germinated seeds were cut and prepared in squash method. The results exhibited that mitosis period of Bartek mainly occurred from 08.00-11.00 a.m. The finding of mitosis period of Bartek investigated in this study is useful to obtain prometaphase stages used to examine chromosome characters. Prometaphase stage of Yellow

Bartek was found at 08.50 a.m. (WIB), whereas prometaphase stage of Long-green and Ellips-green Bartek were found at 09.00 a.m. Three kinds of Bartek investigated in this study had similar chromosome numbers,  $2n = 24$  (Figure 2).

The longest chromosome of Yellow, Ellips-Green, and Long-Green Bartek was 1.845, 1.844, and 1.887  $\mu\text{m}$ , respectively, whereas the shortest chromosome total length of Yellow, Ellips-Green, and Long-Green Bartek was 1.167, 1.118, and 1.114  $\mu\text{m}$ , respectively. Short arm chromosome of Long-Green Bartek was shorter than the others. While the long arm chromosome of Ellips-Green Bartek was longer than the others. The shortest short arm chromosome of Yellow, Ellips-Green, and Long-Green Bartek was 0.529, 0.512, and 0.511  $\mu\text{m}$ , respectively, while the longest short arm chromosome of Yellow, Ellips-Green, and Long-Green Bartek was 0.859, 0.887, and 0.884  $\mu\text{m}$ , respectively. The shortest of long arm chromosome of Yellow, Ellips-Green, and Long-Green Bartek was 0.617, 0.605, and 0.603  $\mu\text{m}$ , respectively whereas the longest long arm of Yellow, Ellips-Green, and Long-Green Bartek was 0.923, 1.062, and 1.004  $\mu\text{m}$ , respectively (Figure 3).

## DISCUSSION

In this study, the chromosomal characterization of Bartek was observed to determine the genetic characters. The finding of mitosis period of Bartek investigated in this study is useful to obtain prometaphase stages used to examine chromosomal characters. Three kinds of Bartek belonging to Cucurbitaceae investigated in this study had similar chromosome numbers, i.e.  $2n = 24$  (Figure 2). This count was not similar from the previous studies on cucumber reported by Robinson and Walters (1999), who recorded that the diploid cucumber chromosome number was 14. However, Robinson and Walters (1999) also explain that the most common haploid numbers of Cucurbitaceae's chromosome were 11 and 12. Therefore, it could be concluded that Bartek was Cucurbitaceae family member and one of melon varieties. Based on the research of PI 371795 melon and American muskmelon reported by Winarsih (2007) and Dewei *et al.* (2005) the chromosome number of Bartek is more related to Melon ( $2n=2x=24$ ). In previous studies, the *karyotype* formula of several species in *Cucurbitaceae* family had been investigated. In this case, melon used as the comparison of Bartek because their similarity in chromosome number ( $2n=2x=24$ ). The *karyotype* formula of several cultivars of *Cucurbitaceae* showed variation. *Karyotype* similar to Bartek belongs to PI 371795 melon ( $2n=2x=24$ ) (Winarsih 2007). While, the *karyotype* different to Bartek belongs to *American muskmelon* (*Cucumis melo* var. *reticulatus*) with a chromosome formula of  $2n=2x=24=20m+2sm+2sm$  (SAT), and Hetian melon (*Cucumis melo* spp. *melo* Pang) with  $2n=2x=24=18m+4m+2sm$  (Dewei *et al.* 2005). Those *karyotype* formula showed that *American muskmelon* and *Hetian melon* have two types of chromosome, metacentric and submetacentric chromosome, which then determined to be symmetry and asymmetry. Singh (1999) stated that

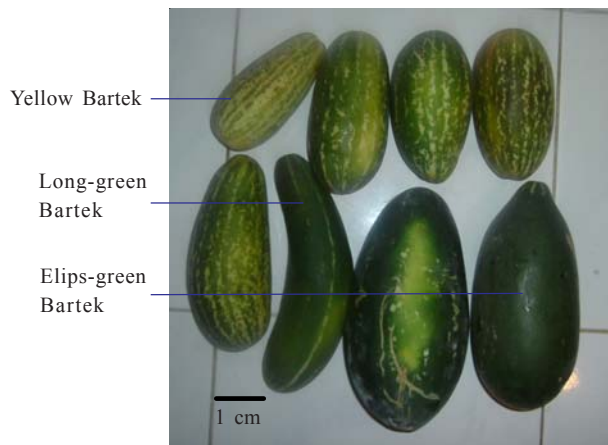
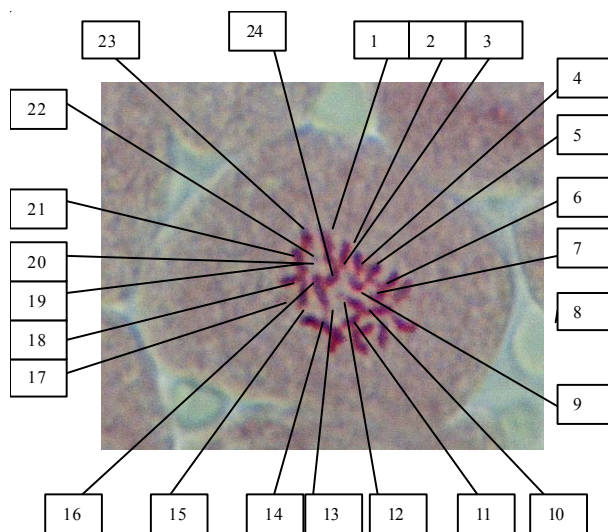


Figure 1. The shape and color of Bartek's fruits.

Figure 2. Total number of diploid chromosomes ( $2n$ ) = 24 in Bartek.

symmetric *karyotype* was more primitive than the asymmetric *karyotype*. Therefore, it could be known that plant varieties with asymmetric chromosome in its *karyotype* have better step of evolution than plant varieties with symmetric chromosome in all of its part. It also could be revealed that Bartek has ancient evolution step because it doesn't have asymmetric chromosome in its *karyotype*.

Moreover, the chromosome number of Bartek is also similar to white Bartek (Ariwarsi 2008) and Timun Suri (Wicaksono 2008). All chromosomes of Bartek also appeared to have the centromere in the median region and were thus classified as metacentric. according to Levan *et al.* (1964) displaying similar karyotype formula  $2n = 2x = 24 = 24m$  (Table 1). In addition, autotetraploid chromosome numbers were appeared in Ellips-Green Bartek (2.22%). It was small number compare to diploid chromosome (97.78%). Autotetraploid chromosome indicates natural mutation because Bartek has not been developed using polyploidization mutation agent. The centromeric index of Yellow, Ellips-Green, and Long-Green Bartek was about 43.283-48.065; 42.404-47.531; and 41.649-47.460, respectively. The finding of metacentric chromosomes in Bartek revealed that Bartek investigated in this study have symmetry *karyotypes* indicating that Bartek have not been widely developed through breeding program. The phylogenetic research reported by Renner *et al.* (2007) showed that the relation between cucumber and melon is not too close. Based on congruent nuclear and chloroplast

Table 1. Comparison of chromosome absolute length, *karyotype* formula, and R value among Bartek and Melon cultivars

Type	Absolute length of chromosome ( $\mu\text{m}$ )	<i>Karyotype</i> formula	R value
Long-green Bartek	1.114-1.887	$2n = 2x = 24m$	1.694
Ellips-green Bartek	1.118-1.844	$2n = 2x = 24m$	1.649
Yellow Bartek	1.167-1.845	$2n = 2x = 24m$	1.581

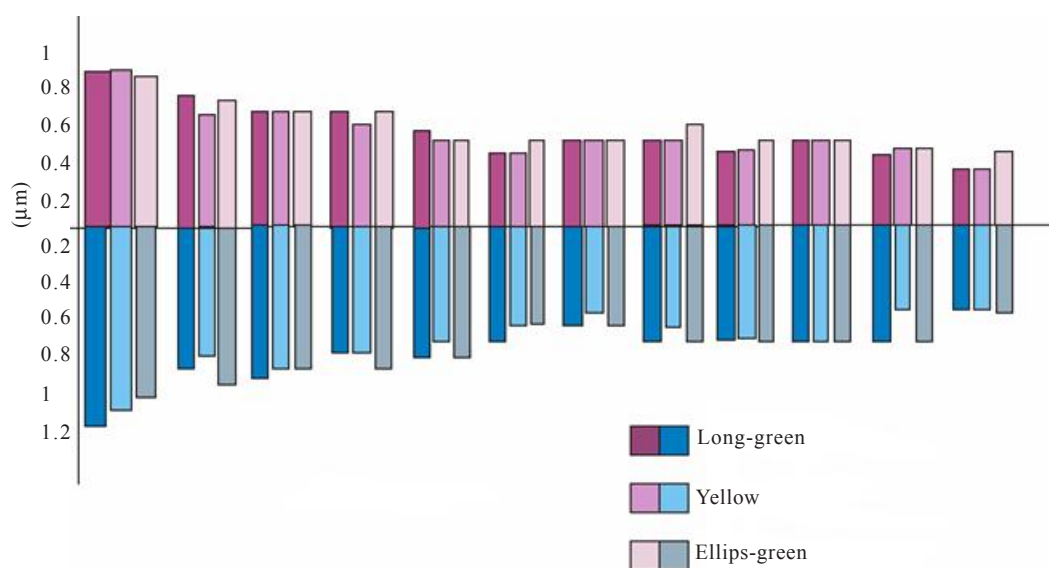


Figure 3. Ideogram showing comparison chromosome size among yellow, ellips-green, and long-green Bartek.



phylogenies, they concluded that *Cucumis* comprises an old Australian/Asian component that was therefore unsuspected. *Cucumis sativus* evolved within Australian/Asian clade is phylogenetically far more distant from *Cucumis melo* than being implied by the current morphological classification. Hence, since Bartek has similar chromosome numbers with melon, it could be concluded that Bartek and melon are the same species.

The result also exhibited that the proportion of the longest chromosome with the shortest chromosome (R) of of Yellow, Ellips-Green, and Long-Green Bartek was 1.581, 1.649, and 1.694, respectively (Table 1). This data revealed that chromosome size of three types of Bartek had similar variation compare to the others. The close genetic relationship among three kinds of Bartek gave valuable information concerning to breeding program. According to several researches, R value that same as 0.27 or lower can be used to strengthen the species placement. For example between *Ranunculus ternatus* (R=1.70) and *R. polii* (R=1.96) (Liang *et al.* 1991), *Arthrolaxis cupressoides* (R=2.05) and *A. selangoides* (R=2.32) (Lin-Chu 1992), *Larix sibirica* (R=1.85) and *L. olgensis* (R=2.06) (Lin-Chu 1993), *Cupressus duclauxiana* (R=1.54) and *C. lusitanica* (R=1.79) (Lin-Chu & Yu-Xi 1996), *Amentoraxis argotaenia* (R=2.71) and *A. yunnanensis* (R=2.59) (Qi-Xing *et al.* 2000). Furthermore, on the basis of the difference of R value in the three kinds of Bartek (R < 0.27) indicated that three kinds of Bartek had close genetic relationship.

Further chromosome studies on other *Cucumis* species from Indonesia will be needed to identify more precisely genetic identity of cucumber species of this country. This is due to plant breeders have always been appreciating enhancement of the existing gene pool, and chromosome studies would be helpful in the enrichment of the existing *Cucumis* germplasm resources of the country to develop *Cucumis* industry.

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#### REFERENCES

- Ariwarsi T. 2008. Siklus Sel dan Jumlah Kromosom Timun Bartek Bule (*Cucumis* spp.) [Thesis]. Yogyakarta: Gadjah Mada Univ.
- Dewei M, Zhenhuai G, Chenghe Z, Suozhu G, Ming W. 2005. Chromosome number and karyotype of melons (*Cucumis melo* L.). *Acta Hort* 17:61-65.
- Jahier J, Tanguy AM. 1996. *Observation of Chromosomes: Basic Techniques (Mitosis) of Plants Cytogenetics*. New Hampshire. Science Publishers Inc.
- Jones SB, Luchsinger AE. 1979. *Plant Systematics*. McGraw-Hill Book Company. USA.
- Kimball JW. 1983. *Biologi. Edisi Kelima*. Jakarta: Erlangga.
- Levan A, Fredga K, Sanberg AA. 1964. Nomenclature for centromeric position on chromosome. *Hereditas* 52:201-202. <http://dx.doi.org/10.1111/j.1601-5223.1964.tb01953.x>
- Liang L, Ling-Ling X, Di-Qing Y. 1991. Studies on karyotypes of 5 species in *Ranunculus* from Jiangxi. *Acta Phytotax Sin* 29:178-181.
- Lin-Chu L. 1992. The karyotype of arthrotaxiz (taxodiaceae) and its systematic position. *Acta Phytotax Sin* 30:331.
- Lin-Chu L. 1993. Studies on the karyotype and systematic position of larix Mill. (Pinaceae). *Acta Phytotax Sin* 31:407-408.
- Lin-Chu L, Yu-Xi F. 1996. The karyotype and the cytogeography of cupressus (Cupressaceae). *Acta Phytotax Sin* 34:119-121.
- Min HG, Ma HT, Liang GH. 1984. *Studies of Genetics*. New York: The MacMillan Company.
- Qi-Xing Z, Zhi-Jian G, Zhong-Shu Y. 2000. Karyomorphology and relationships of Amentotaxus Pilg. *Acta Phytotax Sin* 38:525-532.
- Renner SS, Schaefer H, Kocyan A. 2007. Phylogenetics of Cucumis (Cucurbitaceae): Cucumber (*C. sativus*) belongs in an Asian/Australian clade far from Melon (*C. melo*). *BMC Evolutionary Biology* 7:58. <http://dx.doi.org/10.1186/1471-2148-7-58>
- Robinson RW, Walters DS. 1999. *Cucurbits*. Cambridge: CAB International.
- Singh G. 1999. *Plant Systematics*. Science Publishers, Inc. USA.
- Stace CA. 1979. *Plant Taxonomy and Biosystematics*. Second Edition. London. Edward Arnold.
- Trehane P, Brickell CD, Baum BR, Hettterscheid WLA, Leslie AC, McNeill J, Spongberg SA, Vrugtman F. 1995. *International code of nomenclature for cultivated plants-1995*. Wimborne. Quaterjack Publ.
- Wicaksono CE. 2008. Perbandingan Karyotype Timun Suri (*Cucumis* spp.) Lokal Jawa, Kalimantan dan Sulawesi [Thesis]. Yogyakarta: Gadjah Mada Univ.
- Winarsih. 2007. *Karakterisasi Kromosom Melon (Cucumis melo L.) PI 371795* [Thesis]. Yogyakarta: Gadjah Mada Univ.