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Comparative karyotype analysis and chomosomal characterization of *Lycopersicon esculentum* Mill.

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Abstract

Lycopersicon esculentum Mill. a member of the Solanaceae family was examined following orcein staining. The analysis revealed that, interphase nuclei and prophase chromosomes represented a simple chromocentric and gradient type in BARI Tomato-11 and -15, respectively. They were identified as having been 2n = 24 chromosomes. The chromosomal length ranged from 0.87–1.56 μm in BARI Tomato-11 and from 0.78–1.32 μm in BARI Tomato-15. The total chromosome length of 2n were 28.29 μm for BARI Tomato-11 and 25.14 μm for BARI Tomato-15. The centromeric formula was determined to be 18m+6sm in BARI Tomato-11 and 24m in BARI Tomato -15. No gradual decrease in chromosome length was observed in either variety. However, the range between the highest to lowest chromosome length was greater in BARI Tomato-11 compared to BARI Tomato-15. The presence of exclusively metacentric chromosomes and the absence of gradual decrease of chromosome length in BARI Tomato-15 suggest its primitive karyotypic nature . In contrast, the combination of metacentric and submetacentric chromosomes in BARI Tomato-11 indicates a relatively ~~more~~ advanced chromosomal architecture . These karyotypic characteristics clearly distinguish BARI Tomato-11 and BARI Tomato-15 at the cytogenetic level.

Key words: Karyotype, Characterization , Solanaceae.

INTRODUCTION

Lycopersicon esculentum Mill. (Tomato) is one of the most important vegetable plants in the world. Its use as food dates back to Mexico, but it originated in western South America. Nowadays, a variety of them are grown, sometimes in greenhouses in colder locations. Tomatoes have been developed to increase fruit quality, productivity, and tolerance to biotic and abiotic stresses due to their significance as food. In addition to being utilized as food, tomatoes have also been employed extensively in research. Other model plants (such as rice and Arabidopsis) lack certain intriguing characteristics that the tomato plant possesses, such as succulent fruit, a sympodial stalk, and compound leaves. There are also thirteen designated wild species, which are significant for breeding, for providing desirable features, and for research on evolution. Tomatoes are a significant research material because knowledge from these studies can be easily applied to other plants. These characteristics make the tomato a model plant for the Solanaceae family, and more especially for plants with fleshy fruits.

Tomatoes are eaten in a various ways, such as raw or as an ingredient in a wide range of foods, sauces, salads, and beverages. Despite being a fruit in the botanical sense, they are regarded as a vegetable. They are consumed raw or cooked, added to salads, and used as a flavoring in dishes with meat or fish.

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Lycopene, which is abundant in the fruit, may have positive health effects. One of the strongest natural antioxidants is lycopene. According to certain research, lycopene, which is primarily contained in cooked tomatoes, may help prevent prostate cancer. It has also been demonstrated that lycopene enhances the skin's defense against damaging UV radiation. Eating tomatoes has been linked to a lower incidence of head and neck malignancies, breast cancer, and may provide significant protection against neurological illnesses. The tomato is now grown and eaten around the world. In many countries, it comes in second place to potatoes (Tiwari & Chowdhury, 1986). Unripe green tomatoes can also be pickled, made into salsa, or breaded and fried. Tomato juice can be purchased as a beverage and added to mixed drinks. Because they are acidic, tomatoes are particularly simple to can at home—either whole, in bits, or as tomato paste or sauce. Additionally, the fruit is preserved by drying, frequently under the sun, and is then sold in jars or bags that contain oil. Mediterranean cuisine, particularly that of Italy and the Middle East, uses a lot of tomatoes. They are frequently used in pasta sauces and are an essential component of pizza.

Approximately 7,500 tomato varieties are cultivated for diverse reasons. Shape and size are the primary factors used to categorize different tomato cultivars into multiple groups. Such as- Slicing or globe, Ox heart, Plum, Pear, Cherry, Grape, Campari, Early and cool-summer tomatoes.

Since tomatoes are mostly an inbreeding species, genetic variety tends to decline. There is very little genetic diversity in the cultivated tomato species. Genetic diversity has been regarded as a key component and prerequisite for hybridization programs in crop enhancement programs. There is a great scope for genetic improvement of tomato since a number of specific traits are available in the wild *Lycopersicon* species.

Bangladesh Agriculture Research Institute (BARI) has implemented several breeding initiatives to generate variability to transfer desirable traits and produce an appropriate tomato variety. Following this, BARI published several variants. Only morphological characteristics distinguish these variations. Regarding these variations, no genetic data is known. Genetic data has been regarded as a crucial component and prerequisite for plant improvement initiatives. (Chaudhuri *et al.*, 1976). Therefore, for accurate identification and characterization, comprehensive genetic information about the tomato varieties now in production is required.

Cultivated tomatoes are known to be diploid, with 2n=24 chromosomes (Rick, 1960). A few cases also reported having 2n=25 chromosomes (Chaney & Reeves 1973; Gill, 1978). There are 2n=25 trisomic specimens. Trisomic individuals often produce half as many sterile gametes as other types because of an uneven chromosomal distribution during anaphase, which reduces yield. As such, a screening is required before the variety is distributed to the farmers. Furthermore, as the BARI variety represents our country's wealth, it need to be protected by a patent in genomics to stop trade theft.

A specimen's karyotype is a permanent characteristic that frequently helps to distinguish between distinct taxa. The issue, though, comes when distinct taxa have the same number of chromosomes and centromeric location. Other karyotypic parameters may be helpful in this regard in such a scenario.

A further criterion for differentiating across species is the staining characteristics of prophase chromosomes and interphase nuclei. The study's findings demonstrated that staining characteristics might be used to identify between different taxa, including variants of numerous plant species. DAPI, CMA, and Orcein differential staining is typically used to do this (Warasy & Alam 2009; Shahla & Alam 2011; Jahan *et al*., 2012). In this investigation, an effort was made to comparative karyotype analysis and characterize BARI tomato-11 and -15. The aim was to –

1. find out the characteristic features of interphase nuclei and prophase chromosomes.
2. determine the diploid chromosome number of these specimen after the staining with orcein.
3. prepare the karyotype and ideogram.
4. karyotype analysis and characterization of these specimens.

MATERIALS AND METHODS

For the karyotype analysis and characterization, two vegetable plants viz. BARI Tomato-11 and -15 were used as plant materials.

We gathered healthy roots. For these specimens, the best time to gather roots was at 1:00 pm to achieve the greatest number of proliferating cells. The extracted roots were first treated for one hour at room temperature (28–30°C) with a 0.002M solution of 8-hydroxyquinoline. This was followed by a 15-minute fixation in 45% acetic acid at 4°C. After that, these were hydrolyzed for 10–12 seconds at 60°C in a solution of 1 N HCl and 45% acetic acid (2:1). Subsequently, the hydrolyzed roots were placed in a clean slide after being soaked in filter paper. Using a fine blade, the meristematic area was sliced. The substance received a 1% aceto-orcein drop added to it. The material was covered with a spotless cover glass. Next, a toothpick was used to lightly tap the materials, and then thumbs were used to squish them. Ultimately, the slides were examined using an Olympus-DP72 microscope (Japan).

RESULTS AND DISCUSSION

Orcein-stained interphase nuclei and prophase chromosomes: These staining characteristics offer karyomorphological characteristics that aid in the characterization of various germplasm. After the cytogenetical investigation, the stained interphase nuclei of both the samples (BARI Tomato-11 and -15) showed some common features (Figs. 1, 2). Such as – a minute darkly stained heterochromatic region was found, it was not easy to distinguish the nucleus from the cytoplasm, no nucleolus was present. According to

Tanaka (1971) it is simple chromocentric type (Figs. 1, 2 Table 1). The prophase chromosomes of these samples show gradual staining from one end to another with orcein (Figs. 3, 4, Table 1). A prominent nucleolus was found in both samples. The nature of the prophase chromosome's staining properties may be considered as gradient type (Tanaka 1971).

Tanaka (1971) discovered that different species have distinct heterochromatin staining characteristics in their prophase chromosomes and interphase nuclei. It was he who first suggested these standards for karyomorphological characteristics. Tanaka (1971) categorized prophase chromosomes and interphase nuclei into five groups according to the staining quality in each instance. These standards were then used by many scientists to characterize various plant materials (Begum & Alam 2004).

Table 1. Types of interphase nuclei and prophase chromosomes

Samples	Type of interphase nuclei	Type of prophase chromosomes
BARI Tomato-11	Simple chromocenter type	Gradient type
BARI Tomato-15	Simple chromocenter type	Gradient type

2n chromosome number: In the present investigation, both the samples of *Lycopersicon esculentum* Mill. viz. BARI Tomato-11 and BARI Tomato-15 were found to possess 2n=24 chromosomes (Figs. 5, 6 Table 2). Alam *et al*., 2012 reported the same chromosome number for *Lycopersicon esculentum*. So, the present findings supported the earlier report.

Total chromosomal length: The total length of 2n chromosome complement was recorded as 28.29 μm for BARI Tomato-11 and 25.14 μm for BARI Tomato-15. The total length of the 2n chromosome compliments of BARI Tomato-11 was larger than BARI Tomato-15. Therefore, the present results indicate the diversification of chromatin length among the specimens studied in this research.

Average and Range of individual chromosomal length: Between the two samples of *Lycopersicon esculentum* Mill. the range of chromosomal length was 0.87–1.56 μm in BARI Tomato -11 and 0.78–1.32 μm in BARI Tomato-15. There is no gradual decrease of chromosome length observed in any sample but the distance between highest to lowest chromosome length was larger in BARI Tomato-11 than BARI Tomato-15 (Figs. 7, 8, 9, 10, Table 2).

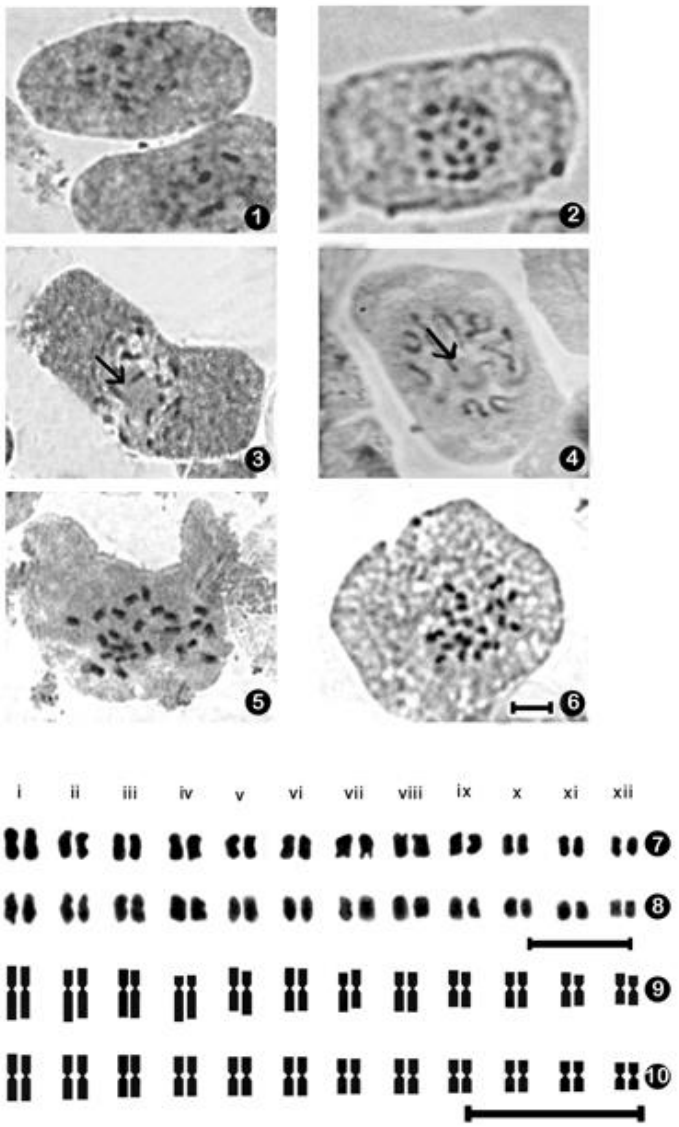
Centromeric feature: Range of centromeric index was 25.19-50.00 for BARI Tomato-11 and 37.62-50.00 for BARI Tomato-15. In case of the centromeric formula, all metacentric chromosomes were found in BARI Tomato-15 representing a strictly symmetric karyotype. Whereas BARI Tomato-11 was found to possess 18 metacentric and 6 sub-metacentric chromosomes representing relatively asymmetric karyotype (Figs. 7, 8, 9, 10, Table 2). Stebbins (1971) mentioned that the symmetric karyotype indicates a primitive character and the asymmetric karyotype indicate an advanced character. From that point

of view, among the two samples BARI Tomato-11 was comparatively advanced than that of BARI Tomato-15.

Table 2. Comparative karyotype analysis in two samples of *Lycopersicon esculentum* Mill.

Cytogenetical Parameters	BARI Tomato-11	BARI Tomato -15
2n	24	24
Total length of 2n (μm)	28.29	25.14
Range of individual chromosome length (μm)	0.87–1.56	0.78–1.32
Difference between largest and smallest chromosome	0.69	0.54
Range of relative length	0.03-0.06	0.03-0.05
Difference of relative length (DRL)	0.03	0.02
Centromeric index range (CI)	25.19-50.00	37.62-50.00
Centromeric formula	18m+6sm	24m

m = metacentric, sm = sub-metacentric chromosome.



Figs. 1-10. Orcein-stained mitotic interphase, prophase, metaphase, karyotype and Idiogram of two samples of *Lycopersicon esculentum* Mill. 1. Interphase nuclei of BARI Tomato-11, 2. Interphase nuclei of BARI Tomato-15, 3. Prophase chromosome of BARI Tomato-11, 4. Prophase chromosome of BARI Tomato-15, 5. Mitotic metaphase of BARI Tomato-11, 6. Mitotic metaphase of BARI Tomato-15, 7. Karyotype of BARI Tomato -11, 8. Karyotype sof BARI Tomato -15, 9. Idiogram of Tomato-11, 10. Idiogram of BARI Tomato-15. Bar=5µ. In figure arrow (→) indicates nucleolus.

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