



## GROWTH AND GENETIC CHARACTERISTICS OF CUCUMBER (*CUCUMIS SATIVUS* L.) CULTIVAR MERCY F<sub>1</sub> HYBRID AND MUTANT POPULATIONS

F. FATHURRAHMAN

Department of Agrotechnology, Islamic University of Riau, Pekanbaru, Indonesia  
 Corresponding author's email: fathur@agr.uir.ac.id

### SUMMARY

Phenotypic and genotypic diversity is essential and needs more enhancement in different ways to achieve higher productivity and better quality vegetables. The presented research aimed to study the phenotypic characteristics and determine the genetic diversity of F<sub>1</sub> populations compared with the colchicine mutant variants in the Mercy cultivar of cucumber (*Cucumis sativus* L.), held from March to May 2019 at the experimental field of the Department of Agrotechnology, Islamic University of Riau, Pekanbaru, Indonesia. A total number of 144 studied plants comprised 72 from each of the two plant groups. The data recorded on various parameters attained statistical analysis using paired T-test at a significant  $P < 0.05$ . Comparisons between the cucumber plants of both groups included karyotypes and chromosome shapes, idiograms, and sequencing. The results revealed that, on average, the taller plants came from F<sub>1</sub> populations (93.49 cm), followed by mutants of the same cultivar (67.83 cm). The F<sub>1</sub> hybrids showed early flowering (29.00 days) compared with colchicine-treated mutants (33.31 days). However, the number of fruits and fruit weight were higher in the mutant variants (9.39 fruits and 1055.39 g), followed by F<sub>1</sub> populations. The karyotypes of cucumber cultivar Mercy F<sub>1</sub> hybrids and mutants have different chromosomes, especially with the arm size. An idiogram also exhibited differences in chromosome length between the variants of both plant groups, while the primers of *trnL-F* and *trnL-R* target sequences were the same with the DNA sequence length. Using mutant cucumber seeds demonstrates a change in phenotypic character to increase fruit production. However, polyploidy did not occur, with genetic alterations measured by the length of chromosome arms. As a result, a future study with increased concentrations of colchicine is imperative to obtain significant chromosomal mutations.

**Keywords:** Cucumber (*Cucumis sativus* L.), F<sub>1</sub> hybrids, colchicine mutants, growth traits, phenotypic characteristics, genetic diversity, chromosomes

**Key findings:** The number of fruits and fruit weight were higher in the mutant populations, while the Brix values and plant height were better in F<sub>1</sub> hybrids compared with the mutants in cucumber (*Cucumis sativus* L.). The varied karyotypes between F<sub>1</sub> and mutant populations with short-arm (p) and long-arm (q) size indicate significant differences in the number of genes. The variation in chromosome arm length phenotypically causes changes in the morphological characters. Therefore, it is necessary to enhance the colchicine concentration and immersion time to obtain polyploidy.

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## INTRODUCTION

Agricultural growth and production have fluctuated yearly, with unstable production that can lead to Indonesia's food security decline. Some horticultural crops' production has decreased in Indonesia, including cucumber (*Cucumis sativus* L.). Noticeably, the decreasing cucumber production brings fear of reducing the fulfillment of Indonesian people's nutrition. In addition, a decrease in production can also lead to a significant reduction in the number of cultivars, eventually degrading the germplasm resources of cucumber (Pratami *et al.*, 2020; Bazargaliyeva *et al.*, 2023).

In Indonesia, the total cucumber production was 521.53 t in 2011. In 2017, it decreased to 424.92 t, with an 18.2% yearly reduction. During the last four years (2018–2021), cucumber production has increased (471.94 t), but it has not been able to match 2011. The area of West Java is the largest cucumber producer in Indonesia, producing 148.272 t in 2021. East Java follows after, yielding 53.570 t of cucumbers (Mahmudan, 2021). The decreasing cucumber production is due to a reducing cultivation area and using less superior varieties. Meanwhile, in Riau province, cucumber production was only 16,808 t in 2021 and is still far behind others. In Riau province, Indonesia, the cucumber cultivation area has decreased over time due to land conversion for urbanization, including housing and other physical needs.

Furthermore, in 2017 the cucumber cultivated area reached 1,729 ha, increasing to 1,804 ha in 2018, but decreasing again to 1,511 ha in 2019 (BPS, 2023). The reduction in area cultivated with cucumber also causes a productivity decline. It makes it necessary to vary the genotypes of cucumbers suitable to the agro-climate of an area to increase production. The conventional breeding constraints of cucumbers delay obtaining new varieties with high-quality phenotypic characteristics. To date, seldom use genetic engineering methods on cucumbers, preferring the use of breeding techniques instead. Moreover, there is an approach with mutagens, with colchicine as the most frequently used for mutations known to be more effective in chromosomal polyploidy. Zheng *et al.* (2019) found that the colchicine treatment caused the size of the stomata to be larger in tetraploid cucumbers than in control diploids, with more chloroplasts per guard cell. This phenomenon

is following the previous characteristic study of stomata at different ploidy levels. Findings of Diminda *et al.* (2021) showed an increase in the size of the red grape callus, which also grew faster than the control. Inducing polyploid can be artificially, through colchicine, oryzalin, trifluralin, amiprofos-methyl, and scopolamine (Gallone *et al.*, 2014; Madani *et al.*, 2015; Podwyszynska *et al.*, 2015).

Colchicine has a significant role in plant breeding, namely, in reduced autopolyploid, it has a direct contribution to plant improvement (Zhang *et al.*, 2015). However, the genetic background of cucumber genotypes gradually becomes narrower due to the genetic erosion of the plant's genetic resources and the hybridization of the different cultivars (Zhang *et al.*, 2016). Polyploidy developed with colchicine is a current technique used for increasing genetic variance and as a plant breeding tool (Maghfirah *et al.*, 2018).

The cucumber cultivar Mercy F<sub>1</sub> and Mercy-Mutant seeds provide useful genetic information for further development. A comparison of growth and production traits appears in phenotypic and genotypic differences. The PCR technique aims to amplify millions of DNA segments in just a few hours (Handoyo and Rudiretna, 2000). However, identifying molecular characteristics has become easier and faster when DNA barcode sequence data is available (Will and Rubinof, 2004; Roslim and Herman, 2017). DNA barcodes are often available in databases, such as, GenBank, that are easily accessed. The chloroplast marker (cpDNA) is also widely used as a molecular marker, with a non-coding part of the cpDNA genome as the *trnL-F* intergenic spacer. These areas are more varied than coding and can aid in recognizing evolutionary relationships at lower taxa levels. Roslim (2019) also used the *trnL-trnF* intergenic spacer primer on *Syzygium* sp., which was most effective for molecular identification.

Therefore, the recent study aimed to determine the phenotypic and genotypic characteristics of the two forms of Mercy cucumber cultivar, namely, Mercy F<sub>1</sub> hybrids and Mercy-Mutant populations obtained through colchicine treatment. The hypothesis was acquiring a genetic diversity presence, having the expected results as the basis for identifying genotypic characters that vary between the two different population types of cucumber cultivar Mercy.

## MATERIALS AND METHODS

The presented study ran from March to May 2019 in the experimental field. The processes implemented during this research included land preparation, soil processing, and treatment with liquid organic fertilizer applied at 15 ml L<sup>-1</sup> during planting once a week for the first two months. The oil palm empty fruit bunches (OPEFB) compost application also had a rate of 500 g per plant one week before planting. The design used was a factorial, completely randomized design with one factor. The subplot size was 100 cm x 100 cm, with the seeds placed inside the planting hole, at 2 cm deep with a spacing of 50 cm x 50 cm. In addition, the crop maintenance processes employed at the planting field included replanting, irrigation, weeding, heaping, and controlling pests and diseases using a modified method from Loleh *et al.* (2018).

### Phenotype analysis

Biological samples subjected to this research included 72 cucumber plant populations derived from each of the F<sub>1</sub> hybrids of Mercy seeds and mutant specimens treated with 1500 ppm of colchicine, making a total of 144 plants. The number of samples observed was 50% of the population. The data recording on plant characteristics included plant height (cm), flowering (day), number of fruits, and Brix content.

### Statistical analysis

The observed data subjected to the statistical analysis used the T-test at the significant level of  $P < 0.05$ .

### Genotype analysis

Analysis of seed samples of the cucumber cultivar Mercy F<sub>1</sub> hybrids and Mercy-Mutant populations ensued at the Genetics and Breeding Laboratory using three specimens of each plant in the chromosome analysis. The analysis included the number, shape, and length of the chromosomes.

### Chromosome preparation of F<sub>1</sub> cucumber and results of mutant induction

The samples of cucumber sprout roots were inserted into the flacon bottle and fixed with 45% glacial acetic acid (AAG) at 4 °C for 24 h. Rinsing with distilled water three times

followed, then macerating the well-rinsed samples with 1N hydrochloric acid solution (HCl) at 60 °C for 11 min. Afterward, rinsing again with distilled water three times, samples got soaked with 1% aceto orcein for 3 h using a modified method by Nathewet *et al.* (2009). Observing chromosomes three times proceeded with a light microscope with a size of 100x10 magnifications on the Olympus BX-41 microscope. Selecting the best image from Prometaphase was then measured and cut using Image Raster 3. Furthermore, processing data size employed the application from Microsoft Excel 2007 to produce an ideograph. Character karyotyping followed the description of Levan *et al.* (1964) on the short sleeve of chromosome (p), the long arm of chromosome (q), centromere index ( $CI = p/[p+q]$ ), arm length ratio of the chromosomes on the short arm chromosomes, and the shape of the chromosomes.

The materials used in this research are the young leaves from a four-week-old cucumber. The Cetyl Trimethyl Ammonium Bromide (CTAB) method aided the DNA extraction. Analysis of DNA samples consisted of 20 biological specimens for each of the Mercy F<sub>1</sub> hybrids and Mercy-Mutant populations (M1), taking from each 20 small leaf samples (0.5 cm<sup>2</sup>), then combining all the samples, DNA extraction proceeded. The process included fresh leaves crushed with a mortar pistil, sterilized with liquid nitrogen, and added with extract buffers. DNA isolation using the CTAB method (Saghai-Marooof *et al.*, 1984) with a slight modification by centrifuging at 4000 rpm for 10 min after incubation. The total DNA solution obtained gained storage at -20 °C, while the solution Working DNA at 4 °C. Total DNA electrophoresis continued on 1.2% agarose gel in 1X TBE buffer, at 190 volts for 20 min. The electrophoresis results in gel form received staining with 5 µg/µl ethidium bromide. DNA band detection used a UV lamp, then photographed with a digital camera.

The total PCR reaction was 10 µl with the PCR component, including 1 µl of total DNA, 1X PCR buffer, 0.1 mM dNTPs, 0.4 µM primer, and two units of Taq DNA Polymerase. The run PCR included pre-PCR at 94 °C for 5 min, followed by 35 cycles with three stages, i.e., denaturation at 94 °C for 45 s, annealing at a temperature of 50.60 °C for 1 min, and elongation at 72 °C for 1 min 30 s. After that, post PCR with a temperature of 72 °C for 10 min ensued. PCR product electrophoresis progressed on 1.5% agarose gel in 1X TBE buffer, at 80 volts for 60 min. Then, gel

staining continued with 5 µg/µl ethidium bromide. Detecting DNA bands used a UV lamp and photographed with a digital camera.

For intra-specific genetic variations, the primers used in amplifying the *trnL-F* sequence were the universal primers, i.e., forward primer (F- 5'-GGTTCAAGTC CCTCT ATCCC-'3) and reverse primer *trnL-R* (R-5'-ATTTGAAGTGGTGACACGAG-'3). This research sent the PCR product samples to 1<sup>st</sup> Base in Malaysia for sequencing analysis.

## RESULTS AND DISCUSSION

### Plant height, flowering, the number of fruits, Brix content, and fruit weight

Results revealed significant differences occurred among the cucumber plant populations for plant height. On average, the height of the Mercy F<sub>1</sub> populations (93.49 cm) was taller than Mercy-Mutant populations (67.83 cm), showing a 37.82% difference (Table 1). It follows the research conducted by Wienda and Pharmawati (2019), who reported that treatment of colchicine (0.01%) and 24-h soaking resulted in slower growth and production of fewer stomata in Garden balsam (*Impatiens balsamina* L.).

According to the analysis, notable differences showed between cucumber Mercy

F<sub>1</sub>s and Mercy-Mutant populations for days to flowering. The reported early flowering in F<sub>1</sub> was 29.00 days compared with Mercy-Mutant populations (33.31 days), showing a delay of 4.31 days (Table 1). It contradicts past findings of Poehlman and Sleper (1995), who reported the size of polyploid plant cells always increases but with a decreasing number of cells. The increase in cell size reduces the plant's metabolism efficiency, affecting its growth with a delay in flowering time. In addition, Sofia (2007) reported that colchicine concentration in cucumber enables slowing down the flowering. Adib *et al.* (2017) reported flowering age of 16–18 days in Ridged gourd (*Luffa acutangula* L.) plants by treating them with colchicine compared with the control (12–13 days).

On average, the cucumber Mercy F<sub>1</sub> and Mercy-Mutant populations also indicated substantial differences in the number of cucumber fruits. In cucumber mutants, the average fruit number was 9.39 compared with the F<sub>1</sub> hybrids (6.22). According to Achmad *et al.* (2013), applying 15 ppm colchicine concentration produced more fruits than the control sample in chili plants. Their research further stated that treated chili plant sprouts enhanced their growth and production, visible in the number of fruits and the wet pod's weight.

**Table 1.** Analysis of T-Test based on mean squares between the cucumber cultivar Mercy F<sub>1</sub> hybrids and colchicine-mutated (M1) populations.

Source	F <sub>1</sub>	Mutant	T-Stat (F <sub>1</sub> )	P (T< = t) two-tail (F <sub>1</sub> )
Plant height (cm)	93.49±1.53 b	67.83±0.78 a	32.71	5.01-7
Flowering (day)	29.00±0.32 a	33.31±0.46 b	-24.7	2.02 -6
Fruit number	6.22±0.30 b	9.39±0.36 a	-9.26	0.0002
Brix	4.44±0.14 a	3.72±0.11 b	8.01	0.0004

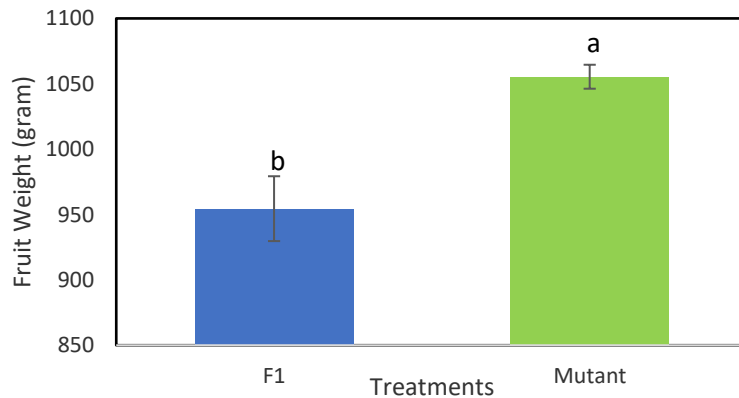
Note: Mean±Standard error (S.E.) followed by a different letter of treatment is significantly tested by T-test at  $P < 0.05$ .

Furthermore, the analysis showed significant differences between the cucumber Mercy F<sub>1</sub>s and Mercy-Mutant populations for fruit weight (Figure 1). The cucumber mutants produced a higher fruit weight per plant (1055.39 g) than the F<sub>1</sub> populations (954.51 g). The cucumber mutant populations showed an increment of 10.56% in fruit weight compared with F<sub>1</sub> hybrid progenies. According to Fathurrahman (2016), the colchicine (0.1%) application with 18 h of soaking on black soybeans produced the highest seed weight. Also, past research has shown colchicine effects in tissue culture, reporting its use increased the production of secondary

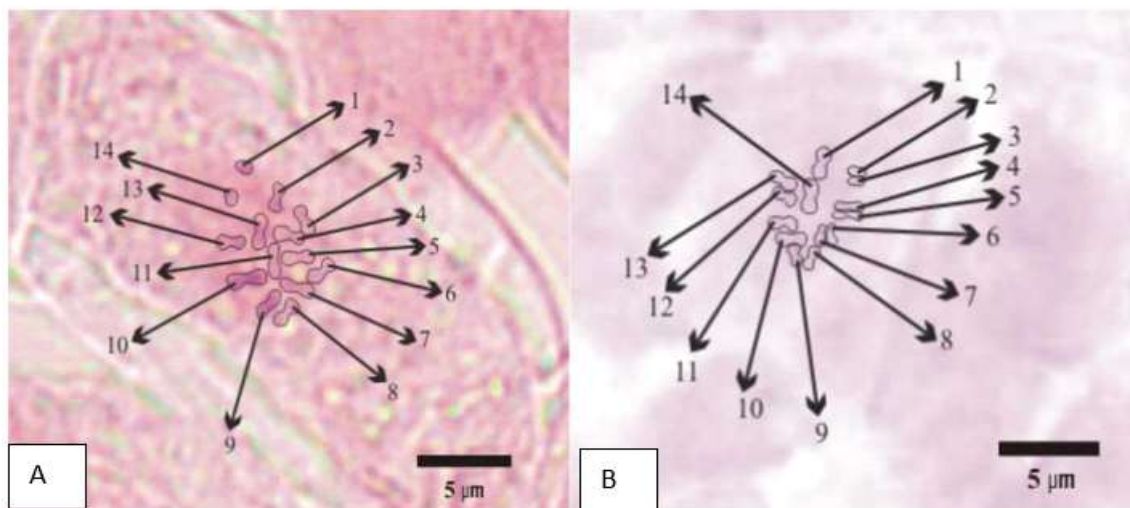
compounds in Cape gooseberry (*Physalis peruviana* L.) in vitro, limiting its concentration to 0.6% to obtain more secondary compounds (Comlekcioglu and Ozden, 2020).

### Number, shape, and length of chromosomes

In the cucumber (*Cucumis sativus*) cultivar Mercy F<sub>1</sub> hybrids and Mercy-Mutants, determining and counting the number of chromosomes employed the prometaphase phase of mitotic division (Figure 2). The two different types of populations (Mercy F<sub>1</sub> hybrids and mutants) showed the same number of



**Figure 1.** Fruit weight of cucumber cultivar Mercy  $F_1$  and mutant populations. Mean  $\pm$  standard error (S.E.) followed by a different letter of treatment is significantly tested by using T-test at  $P < 0.05$ .



**Figure 2.** Comparison of the number of chromosomes at pro-metaphase in cucumber cultivar Mercy (A) and Mercy-Mutant (B) populations.

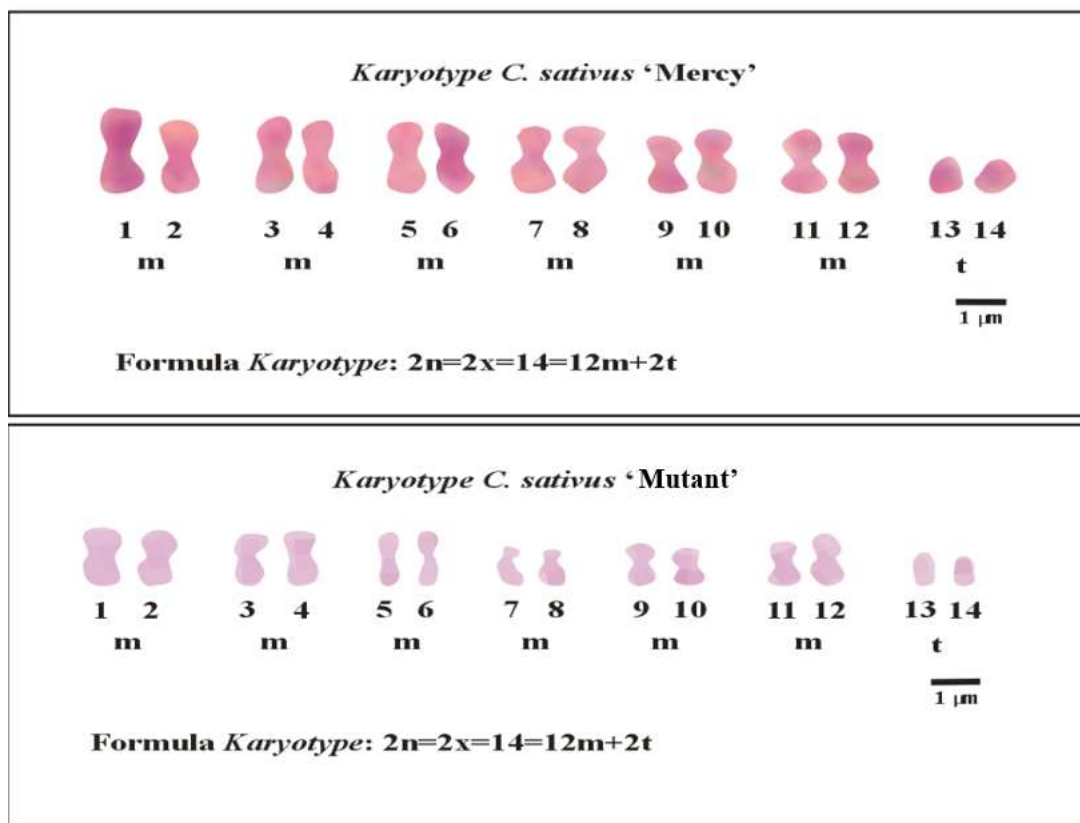
chromosomes ( $2n = 14$ ). During this phase, a jerky movement caused the condensed chromosomes to spread out the cytoplasm. It also facilitated chromosome counting without observing the overlapping ones. Plant populations of the same species always have the same basic number of chromosomes. It is because the number of chromosomes is the character that denotes an organism in the same species besides the morphological parameters of the flowers as a means of reproduction (Mayrose and Lysak, 2020).

Based on the presented results, a difference in the number of chromosomes of the Mercy  $F_1$  hybrid and Mercy-Mutant populations did not occur; thus, they have the same genotype. However, the processes of cross-hybridization and treatment with colchicine could have influenced this. The difference between Mercy  $F_1$  and Mercy-Mutant populations can affect the course of mitosis, pattern, and composition of genes spread across the plant's chromosomes. Furthermore, the variations in mitotic periods within the

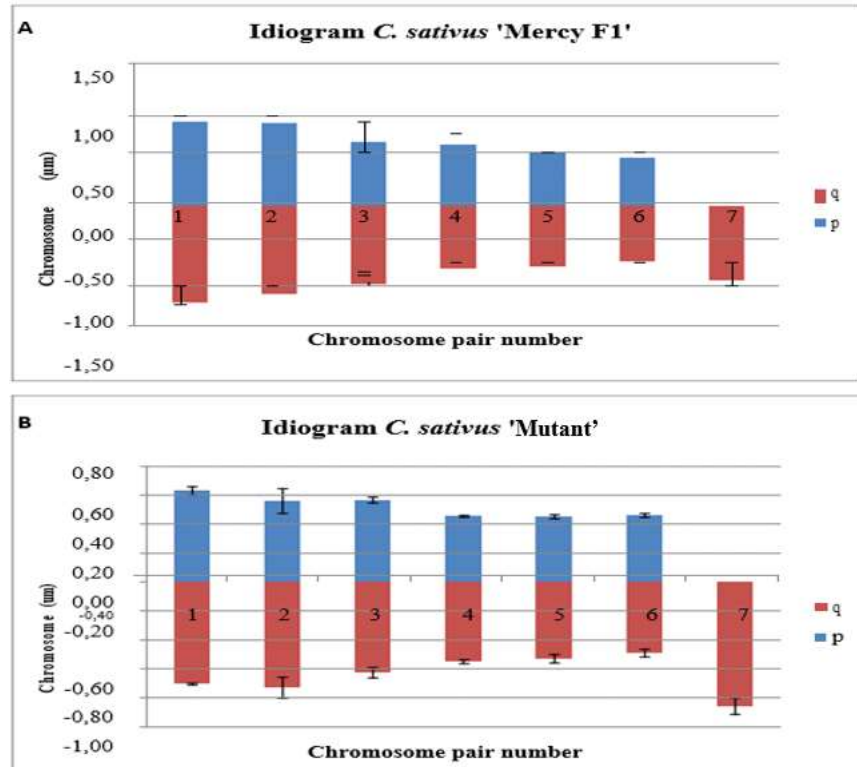
same species were also visible through phenotypic differences. It shows an expression of the variations in chromosomal structure and gene sequences found in the two species. The study results further support such findings, with the molecular analysis showing differences in the length and number of DNA fragments and the results of pCR amplification between the two cucumber samples in this research.

Karyotype measurements of both populations of cucumber (*Cucumis sativus* L.) cultivars showed different chromosomal shapes. Based on the results, both populations have the same number of chromosomes ( $2n = 14$ ), as presented in Figure 3. The karyotype measurements indicated 12 chromosomes have a metacentric form (chromosome pair numbers one to six) and two telocentric chromosome pair number seven in both populations. It indicated that the basis

for classifying cucumber populations was the number of chromosomes, without any difference as expressed by the karyotype formula. The observed differences were only in the chromosome shape between the cucumber cultivar Mercy  $F_1$  hybrids and Mercy-Mutants. It is according to the interpretation depicting the shape and differences in the absolute size of the arms ( $p+q$ ) in both populations. Cultivar Mercy  $F_1$  hybrids have absolute arm size that was relatively long compared with mutant populations. Analysis of the idiogram of Mercy  $F_1$  hybrid populations showed a longer pair of chromosomes (Figure 4A) than Mercy-Mutant populations (Figure 4B). The idiogram results showed differences in the relative length of the chromosomes in Mercy  $F_1$  and Mercy-Mutant populations, both on the short arm ( $p$ ) and long arm ( $q$ ).



**Figure 3.** Comparison of karyotype formulas and chromosome shapes in cucumber cultivar Mercy  $F_1$  (A) and Mercy-Mutant (B) populations.  $n$  = unit of the chromosome,  $x$  = unit of the genome,  $m$  = metacentric,  $t$  = telocentric.



**Figure 4.** Comparison of idiograms in cucumber cultivar Mercy F<sub>1</sub> (A) and Mercy-Mutant (B) populations.

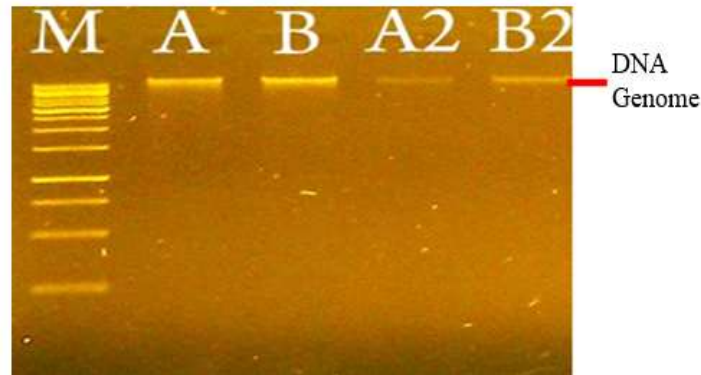
#### TRNL-F and TRNL-R spacer PCR analysis

Using the *trnL-F* and *trnL-R* intergenic spacer as molecular markers helped identify the chloroplast marker (cpDNA). It was part of the cpDNA genome that is non-coding, with several diversities compared with the coding area. Therefore, it was suitable in uncovering the evolutionary relationships at a lower taxonomic level and the kinship of populations within a family. Several studies on non-coding areas of chloroplasts showed higher variation and frequent mutations in Ophioglossaceae (Small *et al.*, 2005). In cucumber Mercy F<sub>1</sub> hybrids and Mercy-Mutant populations, a molecular-based study by optimizing DNA isolation and amplifying the *trnL-F* and *trnL-R* intergenic spacer transpired. The obtained DNA bands were clean, showing very few contaminants, and can serve the subsequent process (Figure 5).

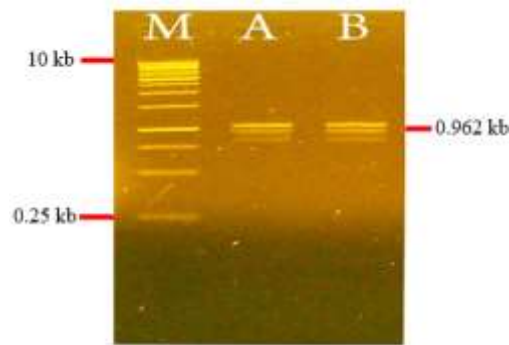
The amplification of the target sequence *trnL-F* and *trnL-R* against both populations of cucumber cultivar Mercy has three bands each, with the same length of DNA sequences. Meanwhile, the longest sequence for band number one was ±0.962 kb (Figure 6), and the two bands were shorter in size.

#### Comparison of DNA sequencing in Mercy F<sub>1</sub> hybrids and mutant populations

The results of the PCR amplification of the purified samples of the DNA of cucumber cultivar Mercy F<sub>1</sub> hybrids and Mercy-Mutant populations underwent sequencing services from a company that provided said services (1<sup>st</sup> Base). Figure 7 shows that the nucleotide sequencing of the PCR amplification results using the primers *trnL-F* and *trnL-R* in the two samples showed no difference in the number of produced nucleotides. Furthermore, the sequencing of both populations yielded 962 bp with no difference in nucleotide yields.



**Figure 5.** Profile of total DNA on 1% agarose gel. M is a 1 kb DNA ladder for Thermo Scientific, A, and A2: total DNA of cucumber Mercy-Mutant, B and B2: total DNA of cucumber Mercy  $F_1$  hybrids.



**Figure 6.** PCR amplification of *trnL-F-trnL-R* intergenic spacer. M: is a 1 kb DNA ladder (Thermo Scientific). A: Biological sample of Mercy-Mutant, B: Biological sample of cucumber Mercy  $F_1$ .

**A.** Cucumber cultivar Mercy  $F_1$  hybrids (962 bp).

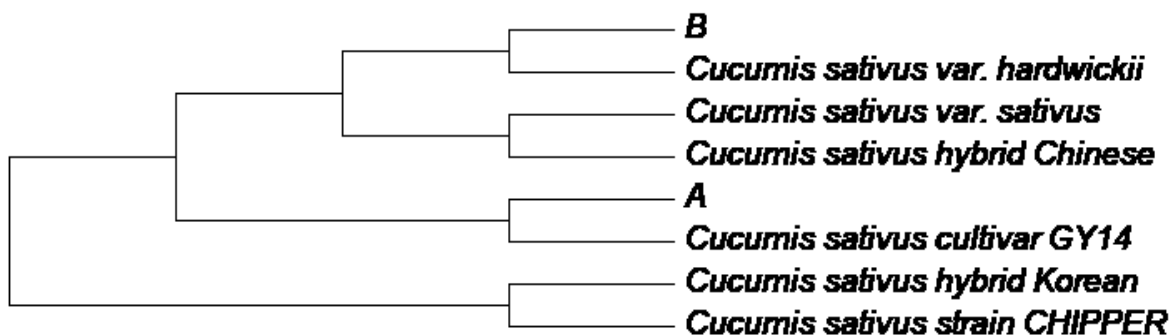
```
TGGAAACCTACTAAGTGATAACTTTCAAATTCAGAGAAACCCTGGAATTAATAATGGGCAATCCTGAGCCAAATCCTTTTTCCGAAAAAAA
AAAAGGGGTAGGTGCAGAGACTCAATGGAAGCTGTTCTAACAAATGGAGTTGACGACGTTGCGTTAGTAAAGGAATGAATCCTTCCATC
GAACTCCAGAAACGATGAAAGATAAATTTATTACGTACTGAAATACTATATTGATTAATGACAACTCGAATCTCTATTTTTATATTTATA
TATTTTTAGATTTATATATATTTTTTTATTTATATAGATATATAAAATTATATATAAAAAATGAACGAATTGTTATGAATCGATTCCAAGACCT
CCAAGTTGAAAAAAGAATCGAATATTCATTGATCAAATCATTTACTCCATCATAACATAGTCTGATAGATCTTTTGAAGAAGTGAATTAATCAG
ATCGAATAGAATAAAGATAGAGTCCCATTCTACATGTCAATACCGACAAAAATGAAATTTATAGTAAGAGGAAAAATCCGTCGACTTTTTAAA
ATCGTGAGGGTTCAAGTCCCTCTATCCCAAACCCGAAAAAGGGCCCGTTGGCCTCTTTAATTATTTATCCTTTTCATTAGCAATTCACAA
TTTGTTATCTTTCTCATTCAATTCGACTCTTTCAAAAACGTATTTGAGCGGAAATTTGATTTCTTATCACAAGGCTTGTTGATATATTCTATA
TGATACACGTACAAACGAACATCCTTGCGCAAGTAATCTGTGAAATTTGAATGATTAACAATACTATCTACTGACTGAAACTTCGAAAGT
CTTATCCAAGCCCTGAAATTTCTGTATCTTCAAAAAGAAGACTTTGGAATACCTTTTTTTCTTATTTACAATTGACATAGACCAAAGTCATC
TATTAATAAAGGATAATGTGTCGGAAATGGCCGGGATAGCTCAGT
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**B.** Cucumber cultivar Mercy-Mutants (962 bp).

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TGGAAACCTACTAAGTGATAACTTTCAAATTCAGAGAAACCCTGGAATTAATAATGGGCAATCCTGAGCCAAATCCTTTTTCCGAAAAAAA
AAAAGGGGTAGGTGCAGAGACTCAATGGAAGCTGTTCTAACAAATGGAGTTGACGACGTTGCGTTAGTAAAGGAATGAATCCTTCCATC
GAACTCCAGAAACGATGAAAGATAAATTTATTACGTACTGAAATACTATATTGATTAATGACAACTCGAATCTCTATTTTTATATTTATA
TATTTTTAGATTTATATATATTTTTTTATTTATATAGATATATAAAATTATATATAAAAAATGAACGAATTGTTATGAATCGATTCCAAGACCT
CCAAGTTGAAAAAAGAATCGAATATTCATTGATCAAATCATTTACTCCATCATAACATAGTCTGATAGATCTTTTGAAGAAGTGAATTAATCAG
ATCGAATAGAATAAAGATAGAGTCCCATTCTACATGTCAATACCGACAAAAATGAAATTTATAGTAAGAGGAAAAATCCGTCGACTTTTTAAA
ATCGTGAGGGTTCAAGTCCCTCTATCCCAAACCCGAAAAAGGGCCCGTTGGCCTCTTTAATTATTTATCCTTTTCATTAGCAATTCACAA
TTTGTTATCTTTCTCATTCAATTCGACTCTTTCAAAAACGTATTTGAGCGGAAATTTGATTTCTTATCACAAGGCTTGTTGATATATTCTATA
TGATACACGTACAAACGAACATCCTTGCGCAAGTAATCTGTGAAATTTGAATGATTAACAATACTATCTACTGACTGAAACTTCGAAAGT
CTTATCCAAGCCCTGAAATTTCTGTATCTTCAAAAAGAAGACTTTGGAATACCTTTTTTTCTTATTTACAATTGACATAGACCAAAGTCATC
TATTAATAAAGGATAATGTGTCGGAAATGGCCGGGATAGCTCAGT
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**Figure 7.** The *trnL-F* and *trnL-R* intergenic spacer sequences on cucumber cultivar Mercy.





**Figure 8.** Dendrogram of six populations of cucumber cultivar Mercy. Blume-based Kimura-2 genetic distance (1980). A: Cucumber Mercy-Mutant populations, B: Cucumber Mercy  $F_1$  hybrid populations.

### Clustering of DNA

Accordingly, the Neighbor-Joining Tree-Consensus Tree-Kimura-2-parameter model-bootstrap 1000× replication obtained the phylogeny construction. Generally, the genetic relationship of *Cucumis sativus* forms two main groups. The first group consisted of Korean *Cucumis sativus* hybrid, *Cucumis sativus* var. 'sativus,' and *Cucumis sativus* var. 'hardwickii' clustered together into one genetic similarity (Figure 8). The second group consisted of populations of *Cucumis sativus* hybrid 'Chinese,' *Cucumis sativus* strain Chipper, and *Cucumis sativus* cultivar GY14, also grouped to show genetic similarities.

### CONCLUSIONS

The cucumber growth traits showed that the number of fruit and fruit weight were also higher in the Mercy-Mutant populations. However, the plant height and the Brix values were better in the  $F_1$  hybrids than in the Mercy-Mutant populations. Using mutant cucumber seeds demonstrates a change in phenotypic character to increase fruit production. However, polyploidy does not occur, with genetic alterations measured by the length of chromosome arms.

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