

## DETERMINATION OF PLOIDY LEVELS IN SUGAR BEET PLANTS:

### III. CHROMOSOME NUMBER OF TWENTY SUGAR BEET BREEDING MATERIALS.

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#### ABSTRACT

During the last three decades of sugar beet breeding program has been started in Egypt by several Egyptian investigators and breeders, in the Agricultural Research Station, Alexandria, Sugar Crops Research Institute, Agricultural Research Center, Egypt. The data were very encouragement; breeding materials were introduced for this program from different countries. Twenty sugar beet breeding materials were chosen and cytologically examined to determine chromosome number to employ these breeding materials in hybridization and improvement the prospective genotypes. This study also includes some yield component characters (root weight and sucrose percentage) and 100 seeds weight for all studied breeding materials to make all knowledge of them available for sugar beet breeding program. The results indicated that there are thirteen sugar beet genotypes have diploid chromosome numbers  $2n = 2x = 18$  (C31/6, C12T, C39, C92, C4612, C790-68H26, Sp-220, Sp-270, Sp-3915, Mo140, Gif. 7, Gif. 8 and Gif. 9), while seven of them have tetraploid chromosome number  $2n = 4x = 36$  (El-Kasr, D2-R18, D2-R66, D2-R811, C11-R48, C11-R540 and C11T). In root weight character the highest value was found in diploid genotype (C92), 1.530 Kg, while the lowest value in tetraploid genotype (C11T), 0.700 Kg. In sucrose percentage character highest value was found in diploid genotype (Gif. 9), 19.6 %, while the lowest values was found in diploid genotype (C4612), 17.4 %. In 100 seeds weight character highest value was found in tetraploid genotype (D2-R811) with mean value of 2.55gm., while, lowest one was found in diploid genotype (C92), with a mean value of 1.43 gm.

#### INTRODUCTION

Sugar beet (*Beta vulgaris* L.), which belongs to the family Chenopodiaceae, is one of the most important arable crops. Sugar beet is a biennial plant species. Approximately 30 percent of World Sugar production are provided by Sugar beet (USDA, Nov. 2008). In Egypt, sugar beet has recently been introduced for sugar production to reduce the gap between the national consumption and the total sugar production. The production of sugar beet in Egypt accounts for about 25% of the total sugar production. Sugar beet as a second sugar crops in Egypt needs a good and safe source of seeds to insure the raw materials for five or six beet sugar factories. For this reason sugar beet breeding program has been started in Egypt by several breeders and investigators (Younan, 1984; El-Manhaly *et al.*, 1987; Saleh, 1993; Ghura, 1995 and Ghonema, 2005).

The main goal of sugar beet breeding programs is to develop sugar beet varieties with high root yield and high sugar content; better extraction yield (juice purity); higher seed germination percentages; lower tendency to

“bolt” and higher resistance to leaf diseases. For that, Egyptian Sugar Beet Breeding Program is concentrated on collecting different sugar beet breeding materials from different countries to achieve this goal. The Egyptian sugar beet breeding materials were exposed to different evaluation and testing experiment to select the good genotypes which have good characteristics to join to the Egyptian sugar beet breeding program.

Since the discovery of polyploidy role in sugar beet breeding, many breeders produced sugar beet tetraploid genotypes in breeding work and there are two types of sugar beet commercial varieties were found anisoploid in the first and later since 1970,s triploid hybrids was found. Determination of ploidy level is a common practice in sugar beet breeding and seed production (Sliwiska and Steen, 1995). Wang and Wang (1998) used deferent methods for ploidy determination in sugar beet (*Beta vulgaris* L.). These methods included measurements of leaf stomatal size (longer and diameter), stomatal density, chloroplast number in stomatal guard cells, pollen size and somatic chromosome counting. All these methods were conducted among a diploid population, and its equivalent triploid and tetraploid population. The results indicated that somatic chromosome counting was the most reliable and efficient method, however, good cytological technique is required. Saleh (2008) established good method for sugar beet chromosome counting in both root tips and young leave cells.

In the present work twenty sugar beet breeding materials, were cytologically examined in both root tips and young leave cells to determine the chromosome number of them and yield component characters (root weight and sucrose percentage) and 100 seeds weight were studied as an attempt for introducing a full knowledge about these breeding materials available to whom work in Egyptian sugar beet breeding program, and can be used later in hybridization to produce triploid hybrids.

## **MATERIALS AND METHODS**

### **1. Sugar beet materials:**

Twenty sugar beet breeding materials used in this study were obtained from Sugar Crops Research Institute, Agriculture Research Center, Egypt. However, 11 genotypes of them were joined recently to the Egyptian sugar beet breeding program. Table (1) presents the twenty sugar beet breeding materials (all of these genotypes were multigerm).

### **2. Methods:**

#### **2.1. Field experiment**

The field experimental part of study was carried out at Faculty of Agriculture Farm, University of Alexandria. The materials were sown in October 2007 and harvested in May 2008. Root weight and sucrose percentages were taken after harvest.

#### **2.2. Green house and laboratory experiments**

Twenty sugar beet studied material seeds were sown at January 2008 in both (Petri dishes and green house), to collect root tips or young leaves for chromosome examination. Root tips and leaf samples were taken for chromosome analysis and prepared as the method described by (Saleh, 2008).

**Table (1): Twenty sugar beet breeding materials and its origin**

No.	Genotypes	Origin	Employment at breeding program
1	C31/6	U.S.A.	New
2	C11T	U.S.A.	New
3	C12T	U.S.A.	New
4	C39	U.S.A.	New
5	C92	U.S.A.	New
6	C4612	U.S.A.	New
7	C790-68H26	U.S.A.	New
8	Sp-220	U.S.A.	New
9	Sp-270	U.S.A.	New
10	Sp-3915	U.S.A.	New
11	Mo140	Turkey	New
12	Gif. 7	Turkey	Old
13	Gif. 8	Turkey	Old
14	Gif. 9	Turkey	Old
15	El-Kasr	U.S.A.	Old
16	D2-R18	U.S.A.	Old
17	D2-R66	U.S.A.	Old
18	D2-R811	U.S.A.	Old
19	C11-R48	U.S.A.	Old
20	C11-R540	U.S.A.	Old

### **2.3. Preparation of investigated materials**

Root tips and leaf samples taken for cytogenetic investigation were collected and transferred to 8-hydroxyquinoline in concentration of 0.002% solution as a pretreatment for 2.30 hours and the samples were fixed by Carnoy's solution (consists of 3 parts of absolute alcohol: one part of glacial acetic acid) for 24 hours at least, and then they were transferred to 70% ethyl alcohol and kept in a refrigerator until usage. Hydrolysis of the studying materials was done by 1N HCL at 60 °C for ten minutes for root tips and two minutes for young leaves. Staining was done by lacto-propionic orcein for 15 minutes at least.

### **2.4. Yield component characters**

Yield component characters were measured after harvest time on the following characters:

- Root weight (Kg.).
- Sucrose percentage.

### **2.5. 100 seeds weight**

Average of 100 seeds weight of the twenty studied genotypes was measured at twenty seed samples for each genotype to detect this trait.

### **2.6. Statistical analysis**

The experimental design was performed in Randomized Complete Block Design (RCPD) with three replicates, and the data were analyzed according to (Snedecor and Cochran, 1990).

## RESULTS AND DISCUSSION

### 1. Cytological examination:

The twenty sugar beet breeding materials were cytologically examined and the data classified the twenty studied materials into thirteen genotypes were diploid and the other seven genotypes were tetraploid. Table (2), illustrates the twenty studied breeding materials and its ploidy levels.

**Table (2): Ploidy levels of the twenty sugar beet of the examined materials**

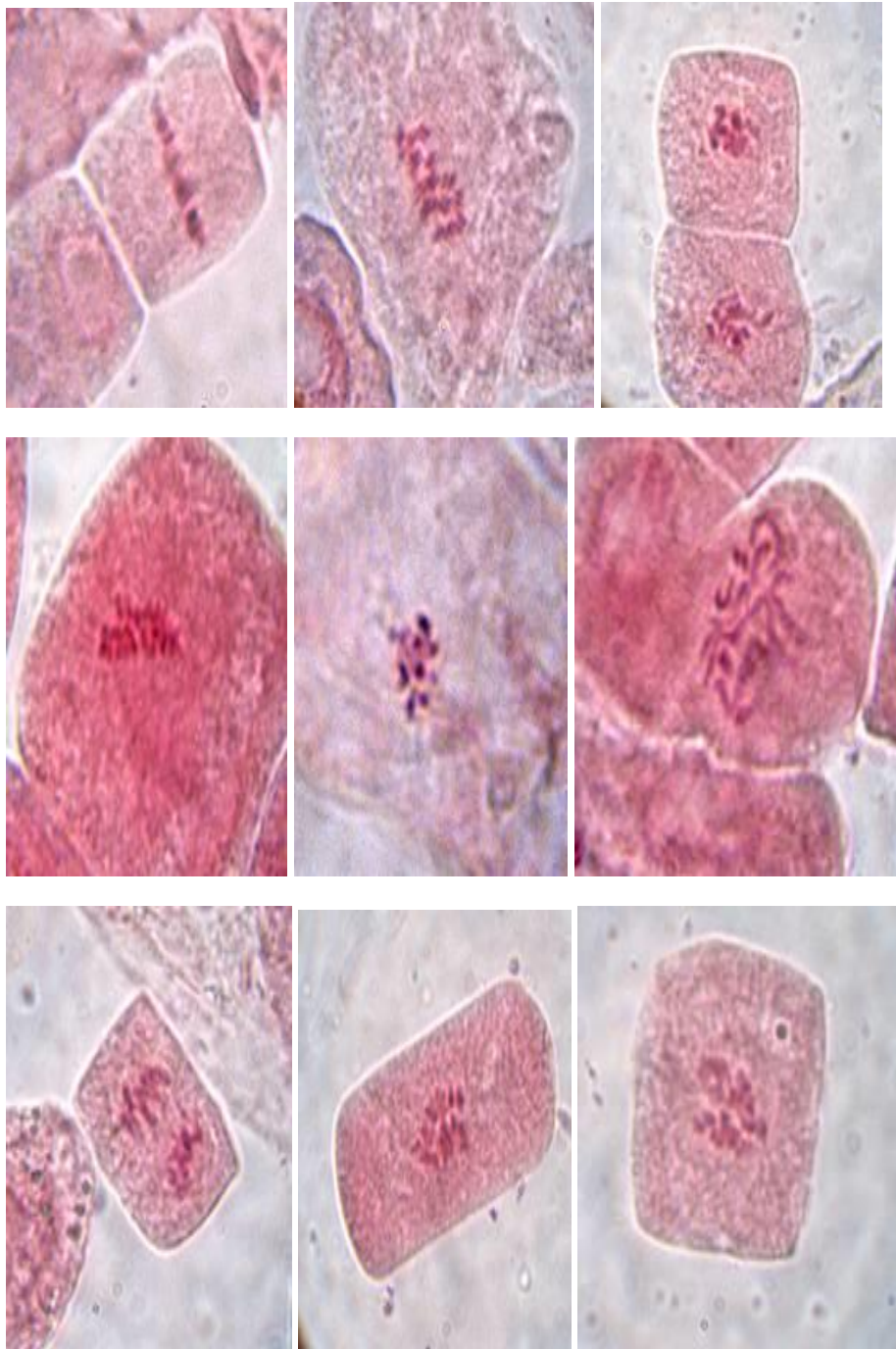
No.	Genotypes	Ploidy levels	No.	Genotypes	Ploidy levels
1	C31/6	Diploid	11	Mo140	Diploid
2	C11T	Tetraploid	12	Gif. 7	Diploid
3	C12T	Diploid	13	Gif. 8	Diploid
4	C39	Diploid	14	Gif. 9	Diploid
5	C92	Diploid	15	El-Kasr	Tetraploid
6	C4612	Diploid	16	D2-R18	Tetraploid
7	C790-68H26	Diploid	17	D2-R66	Tetraploid
8	Sp-220	Diploid	18	D2-R811	Tetraploid
9	Sp-270	Diploid	19	C11-R48	Tetraploid
10	Sp-3915	Diploid	20	C11-R540	Tetraploid

#### A. Diploid breeding materials:

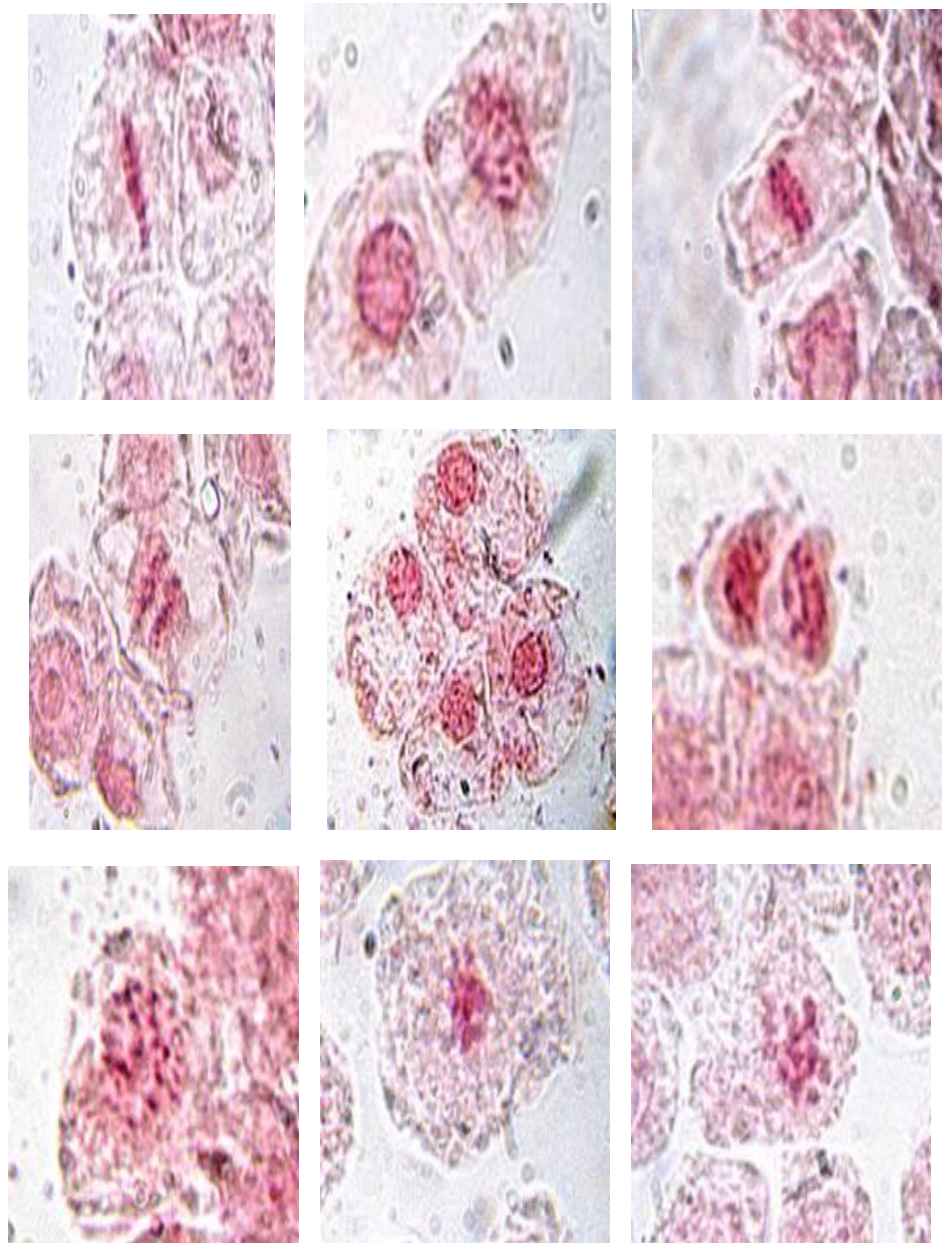
The examined data showed that there were thirteen sugar beet genotypes had diploid chromosome number ( $2n = 2x = 18$ ), ten of these genotypes were employed recently in Egyptian Sugar Beet Breeding Program (C31/6, C12T, C39, C92, C4612, C790-68H26, Sp-220, Sp-270, Sp-3915 and Mo140), while three of the thirteen sugar beet genotypes were previously employed ones (Gif. 7, Gif. 8 and Gif. 9). Figure (1) illustrate root tip sugar beet diploid cells at different stages of mitotic division, while (Figure 2), shows young leave diploid cells.

#### B. Tetraploid breeding materials:

Data illustrated that there were seven sugar beet genotypes had tetraploid chromosome number ( $2n = 4x = 36$ ), six of them were previously employed genotypes (El-Kasr, D2-R18, D2-R66, D2-R811, C11-R48 and C11-R540), while (C11T), was new employed genotype. Tetraploid sugar beet cells were illustrated in Figure (3) and (4) for root tips and young leave sugar beet cells at different stages of mitotic division respectively. Saleh, *et al.* (2008), examined the chromosome number of twenty imported sugar beet varieties they found that 1- four sugar beet varieties were diploid (Gloria, Rhist, Armure and Despreze poly), 2- seven varieties were triploid (Francesca, Amile, Sprint, Puma, Toro, Ymer and Hilma), 3- four varieties were tetraploid (Oscar poly, Sultan, Toro poly and Farida) and 4- five varieties were anisoploid (Gloria poly, Baraca, Athos poly, Top and Ras poly).

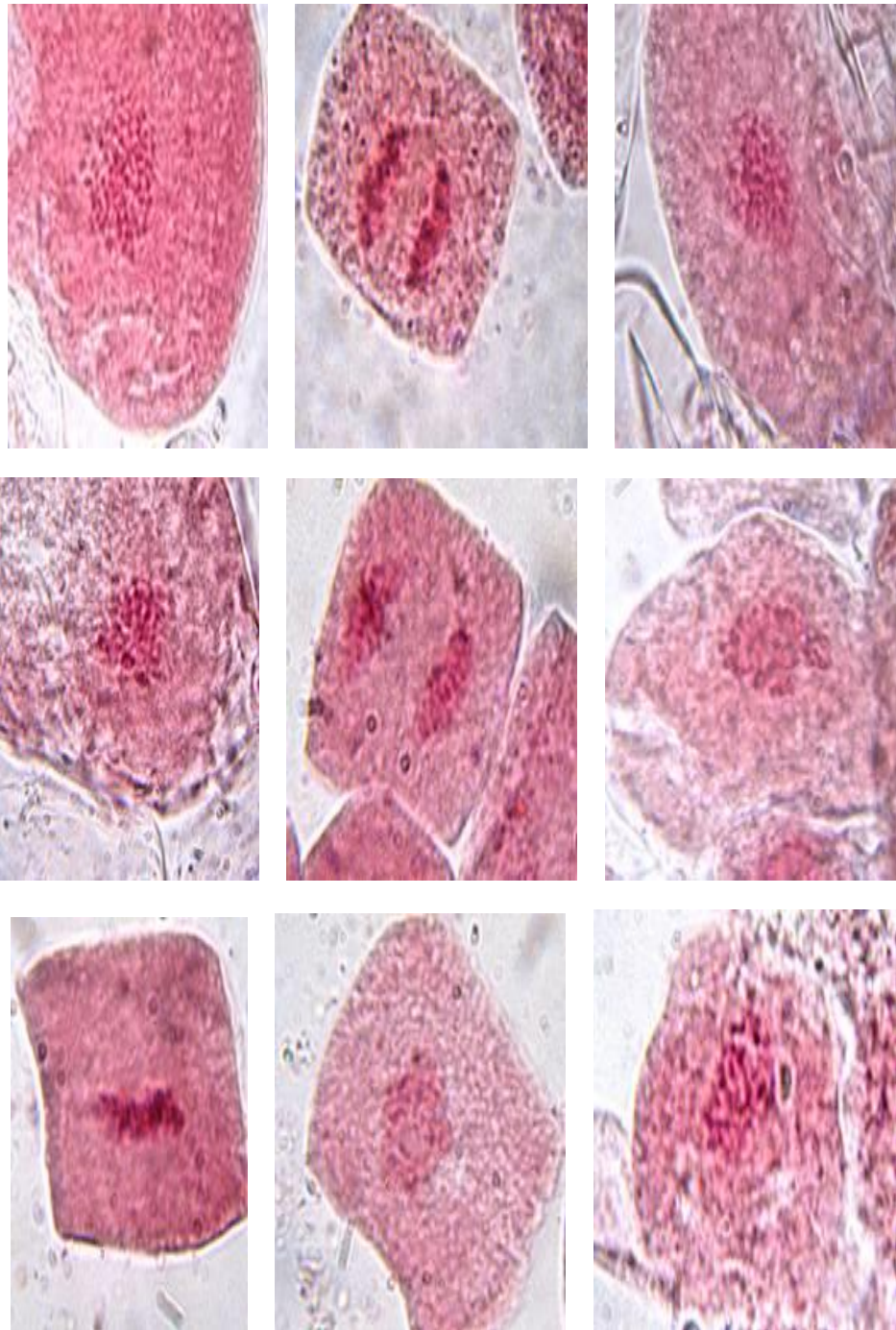


**Figure (1): Diploid sugar beet root tip cells at different stages of mitotic division.**

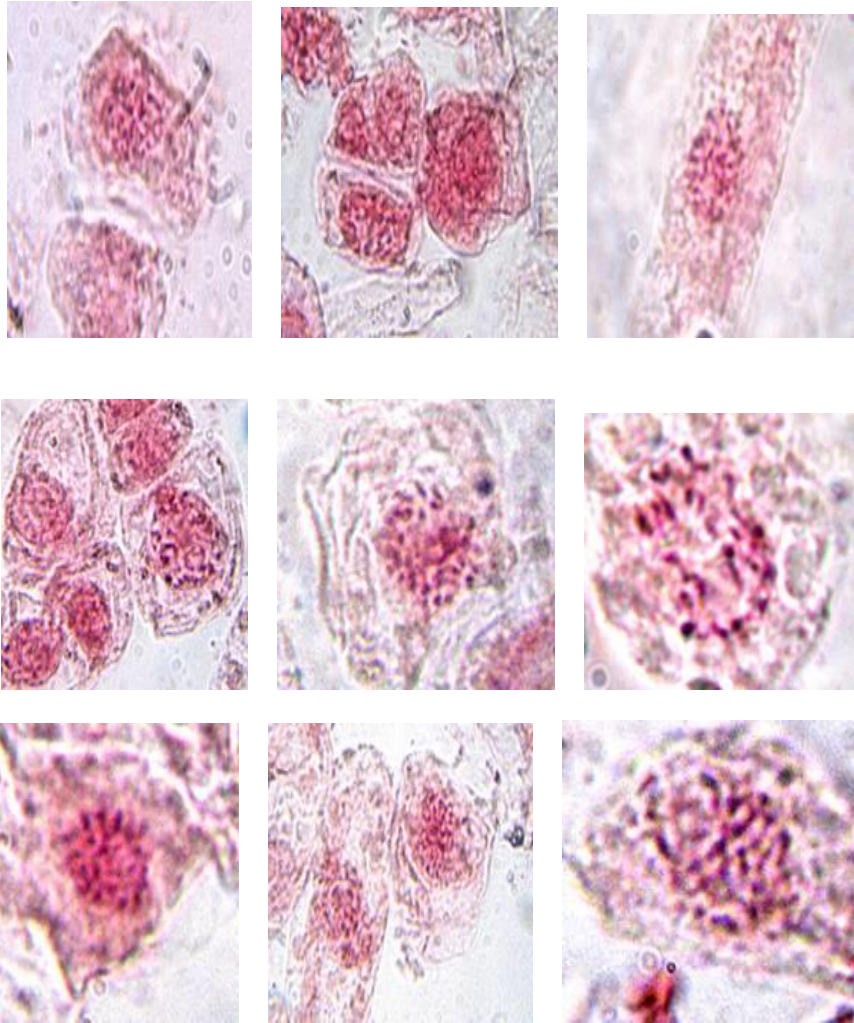


**Figure (2): Diploid sugar beet young leaf cells at different stages of mitotic division.**





**Figure (3): Tetraploid sugar beet root tip cells at different stages of mitotic division.**



**Figure (4): Tetraploid sugar beet young leaf cells at different stages of mitotic division.**

## **2. Yield component characters:**

It is well-known that tetraploid beets physiologically grow somewhat slower. About the time of the longest day they have one or two leaves less than the diploid. Their cells are larger and they have larger stomata. At harvesting most of the tetraploids have wider shoulders than the diploids and this may be the smaller of leaves inserted on the apex and wider leaf stalks (Kloen and Speckmann, 1956). Root weight and sucrose percentage were studied in twenty sugar beet breeding materials to examine the effect of ploidy levels in yield component characters. Table, (3), shows means of root weight and sucrose percentage. Data showed that there were significant



differences between twenty sugar beet varieties in these characters. In root weight character highest value was found in diploid genotype (C92) with root weight mean 1.530 Kg, while lowest value in this character was found in tetraploid genotype (C11T) with root weight mean 0.700 Kg. Sucrose percentage character displayed high value. It was found in diploid genotype (Gif. 9) with sucrose percentage value of 19.6 %, while the lowest one was found in diploid genotype (C4612) with sucrose percentage value of 17.4 %. The obtained data are in agreement to some extent with Poschne Potyi (1982), who concluded that diploids tended to have more leaves but not greater foliage area than the polyploids. Foliage area and root yield were positively related, and there was a close negative correlation between foliage area and sugar content of the roots.

**Table (3): Means of the root weight, sucrose percentage and 100 seeds weight of the twenty sugar beet**

No.	Genotypes	Means		
		Root weight	Sucrose percentage	100 Seeds weight
1	C31/6	0.830 cd	18.4 abcde	1.54 jk
2	C11T	0.700 d	19.3 abc	1.62 ij
3	C12T	0.850 cd	17.9 bcde	1.54 jk
4	C39	0.770 d	17.5 e	1.6 ij
5	C92	1.530 a	18.2 abcde	1.43 k
6	C4612	1.100 bcd	17.4 e	1.69 i
7	C790-68H26	1.310 ab	17.9 bcde	1.44 k
8	Sp-220	0.860 cd	18.5 abcde	1.71 hi
9	Sp-270	0.720 d	18.9 abcde	1.87g
10	Sp-3915	0.850 cd	18.0 bcde	1.85 gh
11	Mo140	0.890 cd	17.7 de	1.61 ij
12	Gif. 7	0.810 d	17.8 cde	2.32 bcd
13	Gif. 8	1.210 abc	18.7 abcde	2.09 f
14	Gif. 9	1.040 bcd	19.6 a	2.33 bc
15	El-Kasr	0.970 bcd	18.4 abcde	2.42 b
16	D2-R18	0.920 cd	18.5 abcde	2.24 cde
17	D2-R66	0.790 d	18.7 abcde	2.13 ef
18	D2-R811	1.070 bcd	19.2 abcd	2.55 a
19	C11-R48	1.030 bcd	19.4 ab	2.18 def
20	C11-R540	1.030 bcd	19.2 abcd	2.39 bc
L.S.D. 0.05		0.3274	1.2774	0.1363

### 3. 100 seeds weight:

100 seeds weight character was examined to detect seed size and weight for the twenty studied genotypes which are influenced on seedling vigor in field and either effect on seed number/Kg. Table (3), shows means of 100 seeds weight character and the highest value for this character was found in tetraploid genotype (D2-R811) with a mean value of 2.55gm, while lowest value was found in diploid genotype (C92) with a mean value of 1.43 gm.

**REFERENCES:**

- El-Manhaly, M.A.; N.Z Younan and M.A. Farage (1987). Sugar beet flowering and seed production in Egypt. Com. In Sci. Dev. Res., 19:45-61.
- Ghonema, M.A.A. (2005). Genetical and cytological studies on bolting in sugar beet *Beta vulgaris* L. plant. Ph. D. Thesis, Faculty of Agriculture (Saba Basha), University of Alexandria Egypt.
- Ghura, Nabwya S.A. (1995). Studies on sugar beet. Evaluation of sugar beet monogerm lines and estimation of general and specific combining ability. Ph.D. Thesis, Faculty of Agriculture University of Alexandria Egypt.
- Kloen, D. and G. J. Speckmann (1956). The creation of tetraploid beets IV. morphological and physiological characteristics of C<sub>2</sub> beets. Euphytica (5): 308-322.
- Poschne Potyi, E. (1982). Study of foliage and yield components in sugar beet hybrids. Pl. Breed. Abs. (1982) 52(12): 10662.
- Saleh, M.S. (1993). Genetical studies on sugar beet. M.Sc. Thesis, Faculty of Agriculture University of Alexandria Egypt.
- Saleh, M.S (2008). Determination of ploidy levels in sugar beet plants I. methods of chromosome counting in sugar beet plants. International Conference IS. Meeting the Challenges of Sugar Crops & Integrated Industries in Developing Countries, Al Arish, Egypt, pp 192-199.
- Saleh, M. S; M. Z. Attalah and M.A. Ghonema (2008). Determination of ploidy levels in sugar beet plants. II. Chromosome Number of Twenty Sugar Beet Varieties Based on Root Tips and Young Leave Cells. International Conference IS. Meeting the Challenges of Sugar Crops & Integrated Industries in Developing Countries, Al Arish, Egypt, pp 200-204.
- Sliwinska E. and P. Steen (1995). Flow cytometric estimation of ploidy in anisoploid sugar-beet populations. Journal-of-Applied-Genetics. 36(2): 111-118.
- Snedecor, C.W. and W.G. Cochran (1990). Statistical methods 7<sup>th</sup> ed. The Iowa State Univ. Press. Ames Iowa. USA. P. 593.
- Wang, S. and S.K. Wang (1998). Evaluation of various methods for ploidy determination in *Beta vulgaris* L. Journal-of-Genetics-and-Breeding. 52: 83-87.
- Younan, Z.N. (1984). Genetical studies on sugar beet. Ph. D. Thesis, Faculty of Agriculture University of Alexandria Egypt.

### تحديد درجة تعدد المجموعة الكروموسومية في نباتات بنجر السكر

٣- العدد الكروموسومي لعشرون مادة من مواد تربية بنجر السكر

مجدى سعد صالح و محمد عبد المنعم غنيمه

قسم التربية والوراثة - معهد بحوث المحاصيل السكرية

أجرى هذا البحث في محطة البحوث الزراعية بالصباحية بالإسكندرية وفي مزرعة كلية الزراعة بالإسكندرية في موسم ٢٠٠٧-٢٠٠٨ والغرض الأساسى من هذا البحث هو دراسة العدد الكروموسومى لعشرون مادة من مواد تربية بنجر السكر وكذلك تم دراسة بعض الصفات المحصولية (متوسط وزن الجذر و نسبة السكر) للعشرون مادة تحت الدراسة بالإضافة إلى وزن ١٠٠ بذرة وذلك بهدف إعطاء صورة متكاملة وواضحة عن مواد التربية هذه لامكانية استخدامها فى برامج التربية ولتكون هذه البيانات متاحة أمام كل الباحثين بغرض الاستفادة منها.

وقد أظهرت النتائج وجود ثلاثة عشر تركيب وراثى تحتوى العدد الثنائى من

الكروموسومات وهم

(C31/6, C12T, C39, C92, C4612, C790-68H26, Sp-220, Sp-270, Sp-3915, Mo140, Gifera 7, Gifera 8 and Gifera)

أما العدد الرباعى فقد وجد فى سبعة تراكيب وراثية هي:

(C11T) و (C11-R540 و C11-R48 و D2-R811 و D2-R66 و D2-R18 و El-Kasr).

عند دراسة بعض الصفات المحصولية مثل صفة وزن الجذر و صفة نسبة السكر وجدت

أختلافات معنوية بين العشرون صنفا تحت الدراسة فى الصفات التى تمت دراستها وقد أظهر التركيب الوراثى (C92) الثنائى المجموعة الكروموسومية أعلى القيم فى صفة متوسط وزن الجذر ١,٥٣٠ كجم، بينما أظهر التركيب الوراثى (C11T) الرباعى المجموعة الكروموسومية أقل القيم فى هذه الصفة ٠,٧٠٠ كجم وفى صفة نسبة السكر أظهر التركيب الوراثى الثنائى المجموعة الكروموسومية (Gifera 9) أعلى القيم ١٩,٦ % بينما أظهر التركيب الوراثى (C4612) الثنائى المجموعة الكروموسومية أيضا أقل القيم فى هذه الصفة ١٧,٤ % وفى صفة وزن مئة بذرة فقد أظهر التركيب الوراثى رباعى المجموعة الكروموسومية (D2-R811) أعلى القيم ٢,٥٥ جم بينما أعطى التركيب الوراثى الثنائى المجموعة الكروموسومية (C92) أقل القيم فى هذه الصفة ١,٤٣ جم.