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BREEDING CERTAIN TRAITS OF THE SNAKE CUCUMBER (CUCUMIS MELO VAR. FLEXUOSUS) THROUGH HONEYCOMB SELECTION

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Abstract

The study was conducted as a field experiment over four seasons at the Abu Ghraib Research Station during fall of 2022 and 2023 and spring 2023 and 2024. It involved three cycles of the honeycomb selection method and three snake cucumber genotypes. The plants from the second selection cycle significantly outperformed the others in all traits. The results showed that the second genotype (V2) significantly outperformed the others in all traits providing the highest values for leaf area (222.24 dm^2), sex ratio (14.14%), and main branches plant^{-1} (4.71 branches plant^{-1}) compared to the lowest for the V3 genotype at 139.44 dm^2, 8.84%, and 3.00 branches plant^{-1}, respectively. The interaction between the study's two factors had a significant effect, with the second genotype plants in the second selection cycle achieving the highest values for all traits, compared to the lowest for the original plants of the third genotype. The narrow-sense heritability values for the two selection criteria in the second selection cycle were 40.67% and 39.14% for branch numbers and sex ratio, respectively, confirming the high yield efficiency of these two criteria. Selection led to increased homogeneity of the plants for several traits, as indicated by an increase in the mean and a decrease in the standard deviation and coefficient of variation as the selection cycles progressed.

Keywords: Honeycomb selection, Snake cucumber, Heritability.

تحسين بعض صفات خيار القثاء بالانتخاب بخلية النحل

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الخلاصة

أجريت الدراسة كتجربة حقلية على مدى أربعة مواسم في محطة أبحاث أبو غريب، خلال موسم الخريف 2022، 2023، وربيع 2023 و2024. شملت الدراسة ثلاث دورات انتخابية باستخدام طريقة الانتخاب بخلية النحل، باستخدام ثلاثة تراكيب وراثية محلية من الخيار القثاء. تفوقت نباتات دورة الانتخاب الثانية معنوياً على نباتات الدورات الأخرى في جميع الصفات. أظهرت النتائج تفوق التركيب الوراثي الثاني معنوياً على التركيبين الوراثيين الآخرين في جميع الصفات، حيث اعطى أعلى القيم للمساحة الورقية والنسبة الجنسية وعدد الأفرع الرئيسية بالنبات وبلغ 222.24 دسم^2 و14.14% و4.71 فرع نبات$^{-1}$ على التوالي، مقارنة بأقل القيم لنباتات التركيب الوراثي الثالث وبلغت 139.44 دسم^2 و8.84% و3.00 فرع نبات$^{-1}$ التوالي. وكان للتداخل بين عاملي الدراسة تأثيراً معنوياً حيث حققت نباتات التركيب الوراثي الثاني في دورة الانتخاب الثانية أعلى القيم لجميع الصفات مقارنة بأقل القيم التي سجلتها نباتات التركيب الوراثي الثالث الأصلية لنفس الصفات، كما بلغت نسبة التوريث بالمعنى الضيق لمعياري الانتخاب في دورة الانتخاب الثانية 40.67% و39.14% لعدد الأفرع والنسبة الجنسية على التوالي، مما يؤكد كفاءة هذين المعيارين في الانتخاب للحصول على إنتاجية عالية، وقد أدى الانتخاب إلى زيادة تجانس النباتات المنتخبة لعدة صفات كما يتضح من زيادة المتوسط الحسابي وانخفاض الانحراف المعياري ومعامل التباين مع تقدم دورات الانتخاب.

كلمات مفتاحية: الانتخاب بخلية النحل، خيار القثاء، معدل التوريث.

Introduction

The snake cucumber (*Cucumis melo var. flexuosus*) belongs to the Cucurbitaceae family and is an important summer vegetable crop grown for its fruits, which are consumed either fresh or pickled (15). It has a chromosome number of 2n = 24 and is a monoecious, cross-pollinated crop, carrying both male and female flowers on the same plant. This increases the rate of cross-pollination, which is a major factor contributing to the genetic deterioration of cultivars and strains (1). Many locally grown cultivars have excellent qualitative traits and are characterized by high genetic stability and adaptation to the local environment. However, due to the nature of flowering and pollination, these genotypes are susceptible to genetic mixing and degradation over generations, necessitating their inclusion in programs to maintain their purity and improve certain quantitative and qualitative traits. Selection is one of the most effective methods for improving cross-pollinated plants.

Generally, selection relies on the extent of genetic variation within a population, increasing the genetic frequency of the selected trait (5). Whether natural or artificial, selection is a process that isolates desirable plants within a heterogeneous population by increasing the genetic frequency of those traits (11). Environmental factors are among the greatest challenges in the success or failure of selection programs. Therefore, it is essential to minimize their impacts by increasing planting distances, which allows each plant to become an independent population. This approach is primarily available through the honeycomb selection method (10) which is particularly effective in improving both quantitative and qualitative traits of most crops, as it conducts selection in a competition-free environment, reducing environmental variations, and increasing genetic variation, particularly the additive gene effect (8).

As a result, the overall mean of the selected trait increases, and the coefficient of variation decreases across selection cycles, indicating an increase in genetic improvement and population homogeneity This method allows for the selection of high and early yields by relying on vegetative growth criteria that correlate positively with yield. Choosing the correct selection criterion is crucial for increasing the efficiency of the honeycomb selection program. Typically, qualitative traits that are positively associated with yield and controlled by a limited number of genes are used as effective selection criteria (18).

Given the lack of breeding and improvement programs for this crop in the local environment, and the widespread genetic mixing and deterioration in growth and yield traits from existing cross-pollination applications, this study aimed to:

1. Improve certain quantitative and qualitative traits of local genotypes of snake cucumber using the honeycomb selection method.
2. Identify the best selection criterion for growth, and high and early yield.
3. Determine the best genotypes and their suitability for cultivation under the conditions in the central Iraq region.

Materials and Methods

The study was conducted at the Abu Ghraib Research Station, Horticulture Department of the Ministry of Agriculture, with the aim of improving some local snake cucumber (*Cucumis melo var. flexuosus*) cultivars using the honeycomb selection method. The experiment involved two factors:

1. Selection - comprising three cycles:
    - C0: Representing seeds of the original genotypes before any selection was made.
    - C1: Representing seeds from the first selection cycle obtained from self-pollinated plants selected from the C0 seeds.
    - C2: Representing seeds from the second selection cycle, obtained from self-pollinated plants selected from the first selection cycle (C1).
2. Three local genotypes of snake cucumber (V1, V2, and V3) obtained from different regions. (As they are local varieties, they have not been approved and named).

The field experiment was conducted over four planting seasons:

- First season (Fall 2022): Seeds of 11 genotypes were planted after selection from a field survey of the northern, central, and southern regions of Iraq to assess their phenotypic traits. They were sown on August 1st, and all necessary agricultural practices were applied. Vegetative growth traits and yield were recorded for all lines. Based on morphological traits, three genetically diverse genotypes were selected for purification through honeycomb selection. The honeycomb design was used only for the first and second selection cycles, while the first season was used for screening the studied genotypes.
- Spring season 2023 (First selection cycle, C1): Seeds from the three selected genotypes were planted in special dishes to produce seedlings on February 25th. After the seedlings reached the two true leaves stage, they were transplanted into the permanent field according to the honeycomb selection program's design (17). (The studied characteristics were maintained in the materials and methods).

The seedlings were planted on raised beds that were 2 meters apart, with plants placed on one side only. The distance between plants was 1.7 meters, following the equation: $a = d\sqrt{2/3}$, where a represents the distance between plants, and d the distance between rows (9). The seedlings of the three genotypes were planted with 50 replications for each cultivar, where each hexagonal cell, consisting of seven plants with one at the center, representing one replication (7).

A random sample of 25 plants, on which no selection or exclusion criteria was applied, was taken to determine the average traits of the cultivar before selection (C0). The mean ($\overline{X}$), standard deviation (σρ), and coefficient of variation (C.V%) for the two selection criteria (number of main branches per plant and sex ratio) were calculated for each genotype. After the plants reached the flowering stage, the necessary data were recorded for each genotype. Plants meeting both selection criteria were selected, and their flowers covered with perforated paper bags one day before blooming. The following day, they were self-pollinated, and the bags were reapplied. The bags were removed after the fruit set was confirmed. Upon reaching full maturity, the fruits from each genotype were harvested separately, and the seeds extracted and mixed for each genotype to represent the seeds of the first selection cycle (C1).

- Third season (Fall 2023 - Second selection cycle, C2): The seeds from the first selection cycle of the three genotypes were directly sown in the permanent field soil on August 1st, following the same design and with the same number of plants per genotype. The outstanding plants that met both selection criteria were selected, bagged, and self-pollinated as in the first selection cycle. After full fruit maturity, the fruits were harvested, and the seeds extracted for each genotype separately to represent the seeds of the second selection cycle (C2).
- Fourth season (Spring 2024 - Evaluation of field performance for original and selected plants): A field experiment was conducted to evaluate the field performance of the three genotypes across the selection cycles. The seeds from the three cycles (C0, C1, and C2) of the three genotypes were sown on February 25th, in polystyrene trays. After the seedlings reached the two true leaves stage, they were transplanted into the permanent field based on the spacing mentioned in (12).

The study was conducted as a factorial experiment using a randomized complete block design (RCBD) with three replications. Data were collected and analyzed using GenStat statistical software, and the means compared using the least significant difference (LSD) test at a 0.05 probability level (4).

Upon completion of plant growth, a random sample of five plants was taken from each experimental unit to determine the average vegetative growth and yield traits, as follows:

1. Number of days from transplanting in the permanent field to the appearance of the first female flower on the main stem: This trait indicates the earliness of yield (16).
2. Number of main branches per plant (branches plant^{-1}).
3. Number of nodes on the main stem before opening of the first female flower (nodes plant^{-1}).
4. Leaf area (dcm^2 plant^{-1}): the area of a single leaf multiplied by the total number of leaves, measured using the Digimizer software, as described by (19).
5. Dry weight of the vegetative mass (g plant^{-1}).
6. Sex ratio of the plant 60 days after transplanting (6).
7. Plant yield (kg plant^{-1}).
8. Heritability in the narrow sense based on the parent-offspring regression method (20) and using the following equation:

$$\mathrm{h^2n.s}\ \% = \frac{\overline{X}_o - \overline{X}_p}{\overline{X}_s - \overline{X}_p} \text{ X } 100$$

with $\mathrm{h^2n.s}$ % representing the narrow-sense heritability percentage, $\overline{X}_0$ the mean trait value of the offspring, $\overline{X}_p$ the mean trait value in the original population, and $\overline{X}_s$ the mean trait value of the selected parents.

This equation helps in estimating the percentage of heritability in the narrow sense between the original genotypes, the selected parents, and the resulting offspring for the studied traits, as well as by knowing the averages of those traits during the different selection cycles.

Results and Discussion

The results in Table 1 indicate significant differences between the selected cycles. The C2 cycle outperformed the others, producing plants with the highest values across all growth and yield traits compared to the original C0 plants except for the number of days to the appearance of the first female flower and number of nodes before it bloomed. Here, the lower value is used as it is an indicator of early flowering and fruit setting. The selection process led to an increase in the mean values of the studied traits and a reduction in σρ resulting in in a gradual decrease in the coefficient of variation (CV %).

As the selection cycles progressed high CV values were recorded for some quantitative traits, such as plant yield, due to the presence of a large number of genes controlling them, as well as environmental factors. This indicates higher homogeneity of the selected genotypes after two selection cycles, reflecting the genetic improvements achieved through selection, which increased the genetic frequency of desirable traits in the selected individuals compared to those that were not selected

(18). Continued selection ultimately leads to increased homogeneity among plants which is one of the three essential traits, namely distinctness, uniformity, and stability (DUS) in improved cultivars (21). These findings are consistent with those reported by (3).

The superior performance of the selected plants can be attributed to the effectiveness of the selection criteria used, which are positively correlated with plant yield and quality. Moreover, the selection process increased the genetic frequency of genes responsible for the number of branches and the sex ratios in the selected plants, leading to an increase in female flowers, thereby enhancing the likelihood of fruit set and ultimately increasing yield. These results align with the findings of (14).

Table 1: The effect of selection cycles on growth and yield traits in snake cucumbers.

Cycle		Days to first female flower	Number of nodes before flowering	Leaf area (dm^2)	Vegetative dry weight (g)	Sex ratio (%)	Main branches per plant	Plant yield (kg)
C$_0$	$\overline{X}$	55.98	9.14	139.63	321.60	9.31	2.86	1.52
	$\sigma\rho$	4.02	1.25	31.44	74.09	1.69	0.53	0.84
	%C. V	7.18	13.68	22.52	23.04	18.15	18.53	55.26
C$_1$	$\overline{X}$	51.74	8.14	171.82	396.52	11.97	3.80	1.95
	$\sigma\rho$	3.61	1.02	29.38	72.82	1.60	0.50	0.78
	%C. V	6.98	12.53	17.10	18.36	13.36	13.16	40.0
C$_2$	$\overline{X}$	49.46	6.63	213.56	492.38	13.54	4.57	2.56
	$\sigma\rho$	3.37	0.68	28.22	69.39	1.24	0.49	0.60
	%C. V	6.81	10.25	13.21	14.09	9.15	10.72	23.44
LSD 5%		0.714**	0.462**	0.492**	1.804**	0.289**	0.059**	0.050**

As shown in Table 2, there were significant differences among the genotypes under study. The V2 genotype significantly outperformed the others, achieving the highest values across all growth and yield traits, while the lowest values were observed in the V3 genotype for the same traits. These variations in genotype performance may be attributed to their different genetic makeup, as the genes of each genotype expressed themselves differently under similar environmental conditions (13).

The superiority of the V2 genotype could be attributed to its high productivity and common usage in the central region. It has high adaptability to the conditions in the production area as well as exceptional genetic and environmental stability, being one of the well-known and adapted genotypes in the region. In contrast, the V1 and V3 genotypes were likely less adaptable and genetically stable as they were developed under environmental conditions (northern and southern Iraq) that are entirely different from those of the central region.

Table 2: The effect of genotypes on growth and yield traits in snake cucumbers.

Genotype	Days to first female flower	Number of nodes before flowering	Leaf area (dm^2)	Vegetative dry weight (g)	Sex ratio (%)	Main branches per plant	Plant yield (kg)
V1	52.25	7.92	163.34	376.65	11.83	3.52	1.67
V2	47.42	6.58	222.24	512.77	14.14	4.71	3.36
V3	57.51	9.41	139.44	321.08	8.84	3.00	1.01
LSD 5%	0.714**	0.462**	0.492**	1.804**	0.289**	0.059**	0.050**

The results presented in Table 3 highlight the significant role of the interaction between the study factors in creating highly important differences among the treatments. The second genotype plants after two selection cycles (C2V2) outperformed all other treatments in all growth and yield traits, compared to the lowest values observed in the third genotype which did not undergo selection (C0V3). This superiority can be attributed to the synergistic effect of the combined study factors. In other words, the genetically superior, stable, and well-adapted genotype, when subjected to two consecutive selection cycles, produced a significant increase in vegetative growth indicators, which positively influenced yield indicators. This is due to the positive correlation between vigorous vegetative growth and the expected yield quantity. These findings agree with those of (2).

Table 3: The effect of interactions between selection cycles and cultivars on growth, yield traits and heritability in snake cucumbers.

Cycle	Geno type	Para meter	Days to first female flower	Number of nodes before flowering	Leaf area (dm^2)	Vegetative dry weight (g)	Sex ratio (%)	Main branches per plant	Plant yield (kg)
CO	V$_1$	$\bar{X}$	55.24	8.97	131.12	301.74	9.19	2.63	1.26
		$\sigma\rho$	1.77	0.58	9.36	25.12	0.71	0.12	0.11
		%C. V	3.20	6.47	7.14	8.32	7.72	4.56	8.73
	V$_2$	$\bar{X}$	51.83	7.99	179.09	414.11	11.27	3.56	2.63
		$\sigma\rho$	1.87	0.44	11.19	26.27	0.64	0.19	0.17
		%C. V	3.61	5.50	6.24	6.34	5.67	5.33	6.46
	V$_3$	$\bar{X}$	60.86	10.46	108.69	248.94	7.48	2.40	0.68
		$\sigma\rho$	2.00	0.46	9.44	22.55	0.49	0.17	0.06
		%C. V	3.29	4.39	8.68	9.06	6.51	7.08	8.82
C1	V$_1$	$\bar{X}$	51.97	8.11	159.40	367.86	12.58	3.62	1.61
		$\sigma\rho$	1.34	0.38	8.77	24.76	0.69	0.10	0.11
		%C. V	2.58	4.68	5.50	6.73	5.48	2.76	6.83
	V$_2$	$\bar{X}$	46.69	6.72	222.10	512.55	14.68	4.79	3.30
		$\sigma\rho$	1.09	0.31	10.74	25.22	0.48	0.19	0.16
		%C. V	2.33	4.61	4.83	4.92	3.26	3.97	4.85
	V$_3$	$\bar{X}$	56.57	9.59	133.96	309.15	8.66	3.01	0.94
		$\sigma\rho$	1.35	0.33	8.54	20.10	0.42	0.15	0.04
		%C. V	2.38	3.44	6.37	6.50	4.80	4.98	4.25
C2	V$_1$	$\bar{X}$	49.54	6.68	199.48	460.35	13.73	4.31	2.12
		$\sigma\rho$	1.05	0.26	7.89	24.50	0.49	0.07	0.10
		%C. V	2.11	3.89	3.96	5.32	3.57	1.62	4.72
	V$_2$	$\bar{X}$	43.72	5.03	265.52	611.64	16.49	5. 80	4.14
		$\sigma\rho$	0.87	0.18	9.66	23.19	0.43	0.17	0.13
		%C. V	1.99	3.57	3.63	3.79	2.58	2.93	3.14
	V$_3$	$\bar{X}$	55.11	8.19	175.67	405.15	10.40	3.61	1.41
		$\sigma\rho$	1.11	0.19	7.86	19.94	0.40	0.13	0.03
		%C. V	2.02	2.32	4.47	4.92	3.86	3.60	2.13
	LSD		1.237*	Ns	0.851**	3.125**	0.500**	0.103**	0.086**

Table 4 shows the variations in heritability percentages depending on the traits studied i.e., number of main branches per plant and sex ratio (two criteria for selecting high yield) across the two selection cycles for the three genotypes. The percentages for main branches per plant and sex ratio were relatively high with values of 42.75% and 41.49% for the V2 genotype, 40.08% and 38.85% for V1, and 39.19% and 37.08% for V3, respectively. An increase in narrow-sense heritability for a particular trait indicates a higher genetic variance for that trait or an increase in the additive genetic effect. This implies an ease in transferring this trait to the offspring and its stability across generations due to the influence of the additive genetic effect (6**)**. Additionally, these results suggest the efficiency of the selection criteria used in the improvement program, as higher heritability for a trait means a greater expected genetic improvement from the selection program. This confirms the possibility of using the number of main branches per plant and sex ratio as effective selection criteria for high yield due to their high heritability (3).

The heritability percentage for plant yield was relatively low compared to that for yield components at 26.67% for the V1 genotype, 28.64% for V2, and 23.8% for V3. This suggests that selecting for yield components may be more efficient than selecting for yield itself. This may be because plant yield is a quantitative trait controlled by a large number of genes, along with low genetic variance for yield and high environmental variance, making it difficult to use yield itself as a criterion for selecting for high yield The heritability values in the narrow sense for number of nodes to flowering and the number of female flowers are relatively high. This because these two traits are considered qualitative traits with high heritability, which encourages their selection as effective criteria to achieve high yields

These findings are consistent with (22), who noted that the heritability of quantitative traits, such as the number of fruits, fruit weight, and total yield, is low because these traits are influenced more by non-additive rather than additive genes, and the returns for selection of these traits is minimal. They recommended not using them as criteria for selecting high yield.

Table 4: Narrow-sense heritability rates (h^2ns%) for growth, yield, and heritability in snake cucumbers.

Genotype	Days to first female flower	Number of nodes before flowering	Leaf area (dm^2)	Vegetative dry weight (g)	Sex ratio (%)	Main branches per plant	Plant yield (kg)
V1	34.18	36.19	29.31	30.14	38.85	40.08	26.67
V2	37.08	38.24	31.43	33.17	41.49	42.75	28.64
V3	32.26	35.48	27.58	28.19	37.08	39.19	23.8
Mean	34.51	36.64	29.44	30.5	39.14	40.67	26.37

Conclusions

The honeycomb selection program effectively improved certain quantitative and qualitative traits of snake cucumber (*Cucumis melo var. flexuosus*) and enhanced the genetic homogeneity of the selected individuals. Based on the findings, the number of main branches per plant and the sex ratio can be adopted as selection criteria for high yield. Also, there is potential to consider other criteria based on the high narrow-sense heritability values. The V2 genotype outperformed the others in all growth and yield traits, making it a promising line for inclusion in breeding and improvement programs due to its strong adaptability and genetic stability under the conditions existing in the central region.

Supplementary Materials:

No Supplementary Materials.

Author Contributions:

Author M. M. M. Alabdaly: writing the methodology, preparation of the original draft; Fouad N. Mohammed Al-Khazarji: Review the writing and editing. Both authors have read and approved the published version of the manuscript.

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All data availability statement.

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The authors declare no conflict of interest.

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