Plant Breeding

UDC 633.36/.37:631.527 https://doi.org/10.31867/2523-4544/0252

SOURCE MATERIAL FOR RED LENTIL (*LENS CULINARIS* MEDIK.) BREEDING

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Topicality. *As an important food crop, red lentils account for 70 % of gross consumption, and the demand for these lentils among consumers in Ukraine is constantly increasing, so it is important to develop and promote varieties of this species into production.* ***Purpose.*** *To create and evaluate a new source material based on a set of economically valuable traits for the red lentil breeding.* ***Methods.*** *The research was conducted at the State Enterprise Experimental Farm "Krasnohradske" in the 2020–2022. The main method used for lentil breeding is intraspecific hybridization followed by individual selection in nurseries. Records and observations were carried out according to generally accepted methods.* ***Results.*** *Plant height of the most productive hybrids ranged from 47.0 to 56.5 cm. The coefficient of variation for this indicator was in the range of 6.4–10.8 %. The number of lateral branches in the studied samples was 5-18 pcs, the coefficient of variation ranged from 15.2 to 26.2 %. The highest number of beans per plant was in the following combinations: natural hybrid with CN 111395 – 281 pcs, and CN 111396 × (Stanka 2 × K. 1212) – 261 pcs on average, the coefficient of variation of this trait was within 32–45 %. Among the studied samples of red lentils, the number of full seeds per plant reached 520 pcs. The best combinations in terms of this indicator were Stanka 2 × K. 1212 with an average of 283 seeds and CN 111396 × (Stanka 2 × K. 1212) with an average of 275 seeds. The coefficient of variation for this indicator was quite high (36–52 %), depending on the combination. The grain weight per plant was 6.3–8.6 g. The coefficient of variation of this trait was 46–51 %, which indicates a significant potential for the selection of highly productive lines. The high yielding combinations by grain weight per plant were hybrid combinations Stanka 2 × K. 1212; CN 111396 × (Stanka 2 × K. 1212).* ***Conclusions.*** *It was found that extreme growing conditions reduced both average and maximum values for all traits. Significant variation in the number of beans and seeds per plant as well as in the seed weight per plant was recorded. In the near future, the developed hybrid combinations are planned to be included in the breeding programme for new high-yielding red lentil varieties.*

Key words: *lentil, variety, hybridization, cotyledons, yield, traits, hybrid combinations*

Introduction. Lentil (*Lens culinaris* Medic.) is a grain legume that belongs to the vetch tribe. It is an important food for a large part of humanity and ranks fifth among the major grain legumes in terms of cultivation area. In 2021, the area under lentils was 5.58 million hectares, with gross production of 5.61 million tonnes, according to FAO [1]. Lentil seeds contain high levels of protein (22–30 %) and carbohydrates (50–60 %) and are rich in B vitamins and other biologically active substances [2, 3]. It should also be noted that lentils, like all legumes, contribute to the nitrogen accumulation in the soil, improving its fertility and structural properties. The moderately arid climate of the Steppe zone of Ukraine is quite favourable for the lentil growth and development, and provides an opportunity to obtain high-quality seeds [4, 5].

The red lentil is a marketing name for lentils that have orange cotyledons and are consumed mainly after the seed coat has been removed.

This significantly reduces the cooking time to 5–10 minutes compared to 40–50 minutes for unprocessed lentils, and increases their nutritional value. Red lentils account for 70 per cent of the total lentil production [2, 4].

In general, the requirements for red lentil breeding are almost the same as for green lentil breeding. Plants must be well developed, resistant to lodging and major diseases, especially Ascochyta leaf spot and Fusarium wilt, and cha-

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racterised by high productivity and plasticity. In addition, the seeds should have an orange colour of the cotyledons, and the seed coat should be easily separated from the cotyledons without damage. This feature of the seeds depends on many factors, such as plant growth and development conditions, damage by pests or diseases, and genetic characteristics. The more convex shape of the seeds makes it easier to separate the seed coat from the cotyledons. According to a number of studies, seeds with a grey seed coat are the most suitable for processing into groats [6]. Another key indicator is the yield of finished products, which should be at least 85 %. It should be noted that the larger seed size, the less by-product in the form of seed coat. However, increasing the seed size causes a decrease in the yield of undamaged cotyledons, which are the most valuable for sale, and a significant increase in the share of by-product in the form of broken cotyledons [7]. The optimal weight of 1000 seeds is 30–44 g.

The colour of the lentil cotyledons is controlled by a rather complex interaction of genes called dominant epistasis. It means that there are several genes that control the colour of the cotyledons: Y (yellow) in the dominant state controls the yellow colour of cotyledons; B (brown) controls the brown colour of cotyledons. If both genes are in the dominant state, the cotyledons are orange in colour (*Y– B –*). If they are in the recessive state of yybb, the pigment does not develop and the cotyledons are light green in colour [8, 9]. Therefore, the cotyledons will be orange in colour with the following combinations of genes: *YY BB, Yy BB, Yy Bb, YY Bb.* However, in order for a variety to be sustainable, the combination of genes that control cotyledon colour should only be *YY BB.*

For a long time, no attention was paid to the development of red lentil varieties in Ukraine. The first red lentil variety to be listed in the State Register of Plant Varieties Suitable For Dissemination in Ukraine in 2019 was Darynka, bred at the State Enterprise Institute of Grain Crops of the National Academy of Agrarian Sciences of Ukraine. The ever-growing demand for red lentils both on the world market and in our country makes it expedient to intensify breeding work to develop new varieties of red lentils and introduce them into production [10, 11]. The purpose of the study is to develop and evaluate a new source material for the complex of economically valuable traits for breeding lentils with orange cotyledons.

Materials and Methods. The main method of lentil breeding is intraspecific hybridisation followed by individual selection in nurseries. Lentil breeding, hybrid nurseries and nurseries for the study of lentil source material were placed in a stationary crop rotation after the winter wheat and black fallow. Breeding and hybrid nurseries were sown with a manual planter at a given plant density of 20 plants per linear metre with a row spacing of 30 cm. The sown area of the plots was 1.5 m^2.

In nurseries, phenological observations were carried out, the duration of the interphase and general growing season of lentil plants was analysed. The resistance of lentil plants to lodging, shedding, drought and disease was determined in accordance with existing recommendations and methods. For the analysis of economically valuable traits, 50 plants were selected from breeding and hybrid nurseries in the stage of full grain ripeness. The obtained research results were subjected to statistical processing using analysis of variance and correlation analyses according to the method of B. O. Dospekhov [12].

The research was carried out in Krasnohrad district of Kharkiv region on the basis of the State Enterprise Research Farm "Krasnohradske" of the State Enterprise Institute of Grain Crops of NAAS in 2020–2022. The soils are deep chernozems with a humus horizon of 70–80 cm. The humus content is 4.7–4.9 %, the pH in the topsoil is 6.9–7.0, the total nitrogen content is 0.28–0.30 %, and the phosphorus and potassium content is 0.15–0.16 % and 2.1–2.2 %, respectively. The mechanical composition of the soils is heavy loamy, silt-coarse silty, relatively homogeneous throughout the territory.

The bulk density of the undisturbed topsoil averages 1.23 g/cm^3, with a slight increase with depth. Groundwater is deep (12–15 m) and does not significantly affect plant life processes [13]. The average annual precipitation is 530 mm, of which about 300 mm falls from April to September. The highest average monthly temperatures are observed in July (20.8 ºC), the lowest in January (-7.0 ºC). It should be noted that over the past 10 years, the average annual temperature in the region has increased by more than 1 ºC and is 8.9 ºC (according to the Kras-

nohrad Meteorological Station, which is located on the farm's territory). Over the years of research, the average annual temperature was 10.7 ºC in 2020, 9.2 ºC in 2021, and 9.6 º C in 2022.

In 2020, weather conditions were quite favourable for lentil growth and development. The soil was physically ripe in the third decade of March, and by the end of the first decade of April it had warmed up enough to begin sowing. The lentils were sown on 10 April and seedlings were obtained on 24 April. Despite the low precipitation, the soil had enough moisture accumulated during the autumn-winter period to produce seedlings. Conditions in May and early June were rather cool and wet, with late frosts (down to -9 °C on the soil on 10 May). However, these conditions did not affect the lentil plants and their development. There was sufficient soil moisture content; in particular, there was 82 mm of rainfall during the third part of May (Table 1), which was almost twice higher than the average monthly rate. Flowering began 48 days after the emergence of seedlings on 9 June.

Table 1. Weather conditions during the growing season of lentils (2020–2022)

Month	Ten days	Average temperature, °C				Precipitation, mm			
		*	2020	2021	2022	*	2020	2021	2022
April	I	7.6	7.6	6.6	10.1	11.3	0.0	13.4	3.7
	II	10.6	7.8	8.7	7.9	6.2	7.7	15.9	52.9
	III	12.2	10.6	8.7	12	12.8	11.1	20.8	12.7
May	I	13.9	14.4	13.2	12.2	13.6	23.0	16.4	5.2
	II	15.3	13.2	14.4	14.7	16.2	23.3	30.0	1.5
	III	18.5	13.0	18.2	15.9	20.2	82.0	33.0	26.6
June	I	19.3	19.1	15.2	21.1	13.7	21.9	45.0	37.2
	II	20.2	23.9	21.2	21.6	19.7	8.1	34.1	4.3
	III	20.2	23.2	25.2	20.8	34.7	9.8	31.6	85.5
July	I	21.0	24.8	23.2	24	30.7	0.1	7.2	0.8
	II	22.8	21.1	26.7	19.3	26.8	29.3	3.7	29.8
	III	23.4	22.9	23.1	20.8	14.1	2.1	10.3	16.3
∑≥5°C/mm		1405.0	1531.0	1504.0	1457.0	274	218.4	261.4	276.5

*Note: * Long-term average indicator*

The weather was very hot and dry during the flowering period. The average air temperature in the second and third ten days of June was 23.9 °C and 23.2 °C, respectively, which is more than 3 °C higher than the long-term average. During this period, rainfall was less than 20 mm. Such weather conditions reduced the flowering period, however, considering the significant moisture reserves in the soil, the plants survived this period successfully, which resulted in a good harvest.

Weather conditions in 2021 were favourable for lentils. A wet and rather cool April allowed us to prepare the soil and sow only on 2 May, almost 10 days later than the optimal time. Seedlings were obtained 12 days after sowing, on 14 May. In the first ten days of June, the temperature was 3.8 °C below the long-term average, with a high amount of precipitation (281 % of the norm). In the second and third ten-day periods, temperatures reached long-term norms, but precipitation fell out 178 % of the norm. Flowering began on 20 June. Such weather conditions contributed to prolonged flowering, the formation of a large number of pods and seeds, and the lentil harvest was the highest this year, in spite of the widespread occurrence of diseases and plant lodging.

In 2022, the meteorological conditions of the early spring, pre-sowing period for legumes were unfavourable in terms of temperature. In April, the weather was cool, temperature contrasts were observed throughout the month, and there was a significant amount of precipitation (69 mm). In the second and third ten days of April, frosts were observed on the soil surface. Therefore, experiments were laid out on 6 May this year. In May, temperatures were cool com-

pared to long-term averages, with an average temperature of 14.2 °C, which is 1.5 °C below the norm. Such weather conditions in May contributed to the growth and development of early grain legumes, however, during the flowering and ripening period of lentils, the weather conditions were not favourable. June was moderately warm, with high precipitation, and flowering began on 22 June. A significant amount of precipitation in the third ten-day period (85.5 mm) contributed to intensive plant growth, caused lodging, and significantly extended the flowering stage. Precipitation in July had a negative impact on seed quality and increased yield loss due to pod cracking. The crop reached maturity on 30 July.

Involvement of the maximum diversity of genetic material in the breeding process is crucial for the development of highly productive lentil varieties with high adaptive capacity. Another important stage in the development of lentil breeding material is the study of morphological characteristics of the involved varieties and elements of their productivity.

The source material was lentil varieties bred by genetic centres: the State Enterprise Institute of Grain Crops of NAAS (the Laboratory of Spring Cereals and Grain Legumes breeding), and the Plant Production Institute named after V.Ya. Yuriev of NAAS (Ukraine), the Plant Gene Resources of Canada http://pgrc.agr.gc.ca (Canada), the Türkiye Tohum Gen Bankası (Turkey), the Germplasm Resources Information Network (USA). In order to obtain hybrid material with desirable traits and properties, 15–20 crosses were conducted annually between both cultivars with orange cotyledons and cultivars with yellow cotyledons, using method of multiple stepwise individual selections. The selection of pairs for crossing took into account the elements of productivity, the length of the growing season, drought tolerance, resistance to lodging and disease. According to the results of the study of the available gene pool, we identified varieties with desirable traits to be involved in the breeding process. The best samples include the following: CDC Maxim, CDC Imperial, CDC Redberry (Canada), Stanka 2, Tuija (Bulgaria), K. 1212 (Iran), Fırat 87, Altıntoprak (Turkey).

In the lentil breeding process, these varieties were used as donors of the following traits: productivity, developed habit, seed shape, and cold resistance.

The resulting hybrid material was improved by a set of economically valuable traits in hybrid and breeding nurseries. Individual selection from hybrid populations was based on the analysis of variability and inheritance of quantitative and qualitative traits.

Every year, 500–600 lentil samples were tested in the breeding nursery. The main areas of breeding and genetic improvement of red lentil source material were to increase seed yield and quality (protein content). An important indicator for red lentils is the separability of the seed coat from the cotyledons. For red lentil varieties, the grey colour of the seed coat is more desirable, due to the fact that such a seed coat is easier to separate from the cotyledons. Considerable attention was also paid to seed size uniformity, resistance to pod cracking, absence of cotyledon colour splitting, tolerance and resistance to diseases and pests, plant height, and lodging resistance. Every year, 800–900 plants of red lentil with breeding valuable traits were selected for further work. Essential approbation and morphological parameters of lentils are plant height, number of lateral branches, lower pod insertion height, number of pods per plant, number of pods per pedicel, number of seeds per plant, number of seeds per pod, 1000 seed weight. These indicators were used to study hybrid material of different generations and compare them with parental forms.

Results and Discussion. Morphological traits are determined not only by the characteristics of varieties, productivity and adaptive properties of the genotype, but also by a number of technological indicators that characterise the suitability of red lentils for production. Therefore, the issues of studying the morphological characteristics of plants and breeding improvement of their performance are relevant.

Plant height is both the most accessible indicator for study and quite informative at the same time. This trait is highly correlated with the duration of the growing season. It also correlates with the number of lateral branches and lodging resistance [13]. Lentils are a short-growing plant with an average height of 35–40 cm, which makes harvesting quite difficult. Therefore, high plants are desirable, but at the same time, the tendency of the plants to lodge

also increases. Therefore, lentil plants should have a spreading habit and a semi-spreading compact bush shape.

The highest plant height was in 2022, slightly lower in 2020 and 2021. In the hybrid combinations studied, the average plant height varied from 47 to 56.5 cm (Table 2). The maximum height (56.5 ± 1.0 cm) was in the hybrid combination Stanka 2 × K. 1212, the minimum (47.0 ± 1.0 cm) – in the combination Stanka 2 × Tuija. The coefficient of variation of this indicator varied in the range of 6.4–9.8 %. The average number of lateral branches in the selected hybrid combinations ranged from 10 in the combination Stanka 2 × K. 1212 to 13 in the combination CN 111396 × Firat. The trait is highly dependent on the conditions of plant growth and development.

Table 2. Parameters of variation in plant height and number of lateral branches of the best hybrid combinations of red lentils (2020–2022)

Name of hybrid combination	Plant height, cm			Number of lateral branches, pcs		
	$X \pm S_x^*$	Lim (min–max)	V, %**	$X \pm S_x^*$	Lim (min–max)	V, %**
CN 111396 × (Stanka 2 × K. 1212)	52.7 ± 1.2	46–64	9.4	11 ± 0.8	6–15	26.1
Stanka 2 × K. 1212	56.5 ± 1.0	41–65	10.6	10 ± 0.6	5–17	23.0
Natural hybrid CN 111395	49.7 ± 0.9	42–55	8.2	12 ± 0.7	8–15	15.2
CN 111396 × Fırat	55.6 ± 1.2	44–67	10.8	13 ± 0.8	5–18	26.2
Stanka 2 × Tuija	47.0 ± 1.0	44–48	6.4	12 ± 0.6	9–18	18.5
CN 111396 × Altıntoprak	54.4 ± 1.4	45–63	10.1	13 ± 0.7	9–17	21.3

*Note * Standard square deviation, ** Coefficient of variation*

Lentil yield is a complex integrated indicator that is formed by the elements of the plant structure: the number of pods and seeds per plant, the seed weight per plant, the 1000 seed weight [14]. Red lentils are characterised by a large number of pods per plant, but not all of them produce a full grain. The fullest pods are formed on the lower part of the plant at the beginning of flowering. The pods that form on the upper tiers have smaller grains, and often under adverse weather conditions, their seeds are underdeveloped and small. A significant number of pods of the upper part of the crop may have no seeds at all, because they are formed at the end of the growing season and under unfavourable conditions the seeds do not have time to develop. The largest number of pods per plant was in the combinations – natural hybrid with CN 111395 281 ± 22 pods and CN111396 × (Stanka 2 × K. 1212) with an average of 261 ± 17 pods, the coefficient of variation of this indicator varied within 32–45 % (Table 3).

Table 3. Parameters of variation in number of pods and seeds per plant of the best hybrid combinations of red lentils, (2020–2022)

Name of hybrid combination	Number of pods per plant, pcs			Number of seeds per plant, pcs		
	$X \pm S_x^*$	Lim (min-max)	V, %**	$X \pm S_x^*$	Lim (min-max)	V, %**
CN 111396 × (Stanka 2 × K. 1212)	261 ± 17	87–402	38	275 ± 15	71–308	41
Stanka 2 × K. 1212	256 ± 23	98–421	39	283 ± 27	95–567	42
Natural hybrid CN 111395	281 ± 32	76–417	45	268 ± 33	68–514	49
CN 111396 × Fırat	212 ± 21	83–430	42	183 ± 20	58–427	52
Stanka 2 × Tuija	258 ± 22	151–419	27	262 ± 24	82–464	36
CN 111396 × Altıntoprak	246 ± 19	92–385	32	281 ± 39	75–449	46

*Note * Standard square deviation, ** Coefficient of variation*

An important prerequisite for high yields is a high number of seeds per plant. Among the red lentil samples, the number of full seeds per plant reached 520. However, the average number of seeds per plant is a more important characteristic of productivity. Tested hybrid combi-

nations have high breeding value because their average number of seeds per plant was very high. This indicator ranged from 183 ± 20 in the hybrid combination CN 111396 × Firat to 283 ± 27 in the combination Stanka 2 × K. 1212.

Depending on the hybrid combination, the coefficient of variation of this indicator is quite high – 36–52 %.

The seed weight per plant in the samples averaged 6.3–8.6 g. This indicator was very variable; the coefficient of variation was 46–51 % (Table 4).

The highest productive hybrid combinations in terms of seed weight per plant were Stanka 2 × K. 1212 – (8.6 ± 0.8 g), CN 111396 × (Stanka 2 × K. 1212) – 8.3 ± 0.6 g, Stanka 2 × Tuija – 8.1 ± 0.9 g. Plant productivity largely depends on seed size. The 1000 seed weight is a relatively stable genetically determined trait, with a coefficient of variation of 10–13.7 % in the experiment. The highest weight of 1000 seeds among the hybrid combinations was observed in the following: Stanka 2 × Tuija and the natural hybrid CN 111395 – 45 and 44 g, respectively.

In 2021, the yield was the highest for all samples, while 2022 was the lowest for most samples. The selected best hybrid combinations significantly exceeded the standard in terms of productivity, in particular, the combinations CN 111396 × (Stanka 2 × K. 1212), Stanka 2 × K. 1212, P. g. CN 111395 were equal to it or within the statistical error (Table 5).

Table 4. Parameters of variation in weight of seeds per plant and 1000 seed weight of the best hybrid combinations of red lentils, (2020–2022)

Name of hybrid combination	Seed weight per plant, g			1000 seed weight, g		
	$X \pm S_x^*$	Lim (min-max)	V, %**	$X \pm S_x^*$	Lim (min–max)	V, %**
CN 111396 × (Stanka 2 × K. 1212)	8.3 ± 0.6	2.1–12.6	46	38.3 ± 0.9	33.4–35.2	11.9
Stanka 2 × K. 1212	8.6 ± 0.8	2.3–13.6	46	39.3 ± 1.2	31.4–49.1	13.7
Natural hybrid CN 111395	6.7 ± 1.0	1.6–12.6	51	44.0 ± 1.0	34.8–50.3	11.7
CN 111396 × Fırat	6.3 ± 0.7	2.3–11.5	52	35.0 ± 1.0	29.6–38.1	12.3
Stanka 2 × Tuija	8.1 ± 0.9	2.5–11.2	47	45.0 ± 1.1	37.5–48.2	10.0
CN 111396 × Altıntoprak	7.1 ± 0.8	1.7–10.9	46	40.0 ± 0.9	36.4–44.3	11.1

In general, red lentils are 5–10 % higher in productivity than green lentils, depending on weather conditions and varietal characteristics.

Table 5. Productivity of the most promising hybrid combinations of red lentils (the data of breeding nursery for 2020–2022)

Name of hybrid combination	Seed weight per 1 m^2, g			Average
	2020	2021	2022	
CN 111396 × (Stanka 2 × K. 1212)	269	335	252	285
Stanka 2 × K. 1212	276	365	275	305
Natural hybrid CN 111395	225	342	265	277
CN 111396 × Fırat	241	256	183	227
Stanka 2 × Tuija	271	363	228	287
CN 111396 × Altıntoprak	253	284	216	251
Standard	228	256	236	239
LSD_{05}	18	21	20	17

Conclusions. The research and the results obtained have shown that the yield of the developed hybrid combinations of red lentils is equal to the standard, and in some cases significantly exceeds it.

Analysing the yield structure, we found that most of the developed hybrid combinations have the following parameters: plants are medium in height (47–54 cm), with up to 10–12 lateral branches, highly productive in terms of both the number of pods (212–281) and seeds per plant (183–283), and with a 1000 seed weight

ranging from 35 to 45 g.

It was found that both average and maximum indicators for all traits decreased under extreme growing conditions. Significant variation was observed in the traits "number of pods and seeds per plant" and "seed weight per plant".

The developed hybrid combinations are planned to be included in the breeding programme for new high-yielding varieties of red lentil in the near future.

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УДК 633.36/.37:631.527

Кулініч О. О. Вихідний матеріал для селекції червоної сочевиці (Lens culinaris* Medik.*).
Зернові культури. 2023. 7 (1). 5–12.

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Актуальність. Сочевиця є важливою харчовою культурою. Червона сочевиця зокрема становить 70 % її валового споживання, також попит на сочевицю цього типу постійно зростає серед споживачів в Україні, тому важливим є створення і поширення у виробництві сортів цього типу. **Мета роботи.** Створення і оцінка нового вихідного матеріалу за комплексом господарсько-цінних ознак для селекції червоної сочевиці. **Методи.** Дослідження проводилися на ДП ДГ «Красноградське» Державної установи Інститут зернових культур протягом 2020–2022 рр. Основним методом селекції сочевиці є внутрішньовидова гібридизація з наступним індивідуальним добором у розсадниках. Обліки і спостереження проводили за загальноприйнятими методиками. **Результати.** Висота рослин найбільш урожайних гібридів змінювалася від 47,0 до 56,5 см. Коефіцієнт варіації цього показника знаходився в межах 6,4–10,8 %. Кількість бокових гілок у зразках, що досліджувалися була 5–18 шт., коефіцієнт варіації – від 15,2 до 26,2 %. Найбільше бобів на одну рослину мали комбінації: природний гібрид з CN 111395 – 281 шт., та CN 111396 × (Станка 2 × К. 1212) в середньому 261 шт., коефіцієнт варіації цієї ознаки коливався в межах 32–45 %. Серед досліджуваних зразків сочевиці

червоної кількість повноцінного насіння з однієї рослини сягала 520 шт. Найкращими за цим показником виявилися комбінації Станка 2 × К. 1212 із середньою кількістю 283 насінини та CN 111396 × (Станка 2 × К. 1212) із середньою кількістю 275 насінин. Коефіцієнт варіації цього показника був досить високий – 36–52 % залежно від комбінації. Маса зерна з рослини становила 6,3–8,6 г. Ця ознака є найбільш мінливою V = 46–51 %, що вказує на значний потенціал виділення високопродуктивних ліній. Найбільш урожайними за масою зерна з рослини були гібридні комбінації Станка 2 × К. 1212; CN 111396 × (Станка 2 × К. 1212). **Висновки.** Встановлено, що за екстремальних умов вирощування зменшувалися як середні, так і максимальні показники за всіма ознаками. Значне варіювання спостерігається за ознаками "кількість бобів" та "насіння з рослини" та ознакою "маса насіння з рослини". Створені гібридні комбінації заплановано найближчим часом включати до програми з селекції нових високопродуктивних сортів червоної сочевиці.

Ключові слова: *сочевиця, сорт, гібридизація, сім'ядолі, урожайність, ознаки, гібридні комбінації*