

PIGMENT COMPOUNDS OF THE VEGETATIVE ORGANS OF AMARANTH (*AMARANTHUS* spp.) AS A SOURCE OF NATURAL ANTIOXIDANTSTYNYKULOV M.K.^{1*}, KORNILOVA A.A.², KUZNECOVA M.A.², SADYKOV A.M.¹, YESSENZHOLOV B.KH.³, UTAUBAYEVA A.U.⁴, SULEIMEN A.⁵¹L.N. Gumilyov Eurasian National University, Astana, Kazakhstan²M. Kozybaev North Kazakhstan State University, Petropavlovsk, Kazakhstan³Sh. Ualikhanov Kokshetau University, Kokshetau, Kazakhstan⁴M. Utemisov West Kazakhstan University, Uralsk, Kazakhstan⁵Karaganda Medical University, Karaganda, Kazakhstan

*e-mail: tynkulov@list.ru

ABSTRACT

In this study, a comprehensive assessment of 23 amaranth genotypes was carried out in order to identify forms with high productivity and high content of antioxidant pigments (chlorophylls, betalains, and carotenoids) adapted to the conditions of the arid agro-climatic zone of Northern Kazakhstan. As part of the work, the following tasks were solved: agromorphological and biochemical characterization of genotypes, determination of the content of antioxidant pigments in vegetative organs, assessment of green mass yield, calculation of heritability coefficients and genetic variation of traits, correlation analysis, as well as identification of promising genotypes and donors for subsequent breeding. The results showed high genetic variability in all the studied traits, confirmed by significant heritability coefficients and stability of indicators. Correlation analysis revealed a positive relationship between antioxidant pigments and a weak negative relationship with the yield of green mass. Promising genotypes (VA27, VA26, VA24, VA42, VA38) with an optimal combination of traits were identified, as well as donor genotypes VA25 and VA43 for breeding work. The practical significance of the study lies in the possibility of using the data obtained in the development of new varieties of amaranth with a high content of biologically active substances resistant to drought, for functional nutrition, pharmaceuticals and the agro-industrial sector.

Keywords: amaranth, antioxidant pigments, chlorophyll, betalains, carotenoids

INTRODUCTION

Amaranth (*Amaranthus* spp.) is regarded as a promising food crop, particularly in developing countries, due to its high content of biologically active compounds and its ability to adapt to adverse climatic conditions [10, 11, 15]. It is a rich and accessible source of essential minerals (iron, calcium, potassium), vitamins (including ascorbic acid), protein, dietary fiber, flavonoids, polyphenols, and antioxidant pigments such as betalains, carotenoids, and chlorophylls a and b [12–14].

In recent years, there has been growing interest in natural pigment sources that offer not only coloring properties but also functional value [2, 4, 5]. Food products containing natural antioxidants are perceived by consumers as healthier and safer, which encourages the development of cultivars with high levels of such compounds [14, 17, 18]. In particular, betalains and carotenoids exhibit significant pharmacological properties — including antioxidant, anti-inflammatory, antimicrobial, and even potential anticancer activity [3, 6]. Their application is relevant not only to the food industry but also to pharmaceutical and cosmetic sectors [2, 16].

Northern Kazakhstan, characterized by a sharply continental arid climate with high solar radiation and limited water resources, provides favorable conditions for the cultivation of drought-resistant crops like amaranth [14, 19]. However, the variability in antioxidant traits among different vegetable amaranth genotypes adapted to the region remains insufficiently studied [1, 9].

Therefore, the investigation of genotype diversity in terms of antioxidant pigment content and its relationship with green biomass yield is crucial for developing high-yielding, func-

tionally enriched amaranth cultivars suitable for agroecological conditions of Northern Kazakhstan [10].

Consumer interest in natural pigments and eco-friendly products increases the value of crops like amaranth, particularly as a potential alternative to beetroot (the main commercial source of betalains) [8]. The wide color range, antioxidant activity, and safety of amaranth make it suitable for applications in food, pharmaceutical, and cosmetic industries [13].

MATERIALS AND RESEARCH METHODS

The study included 23 promising genotypes of vegetable amaranth, selected from a total of 122 genotypes based on their high productivity and diversity in stem and leaf coloration. Field trials were conducted under open field conditions using a randomized design with three replications. Sowing was carried out in the spring over two consecutive growing seasons (2023–2024). Each genotype was planted in a plot of 1 m² with inter-row spacing of 20 cm and intra-row spacing of 5 cm.

Prior to sowing, organic fertilizer (compost) was applied to the soil (typical sierozem with neutral pH) at a rate of 10 t/ha. Mineral fertilizers were applied at the following rates: urea – 200 kg/ha, triple superphosphate – 100 kg/ha, potassium salt – 150 kg/ha, and gypsum – 30 kg/ha. Irrigation was performed every 5–7 days as needed. During the growing season, daytime air temperatures ranged from 27°C to 42°C. Crop maintenance included regular thinning, loosening, and manual weeding every seven days.

Green biomass was harvested 30 days after seedling emergence. Yield was assessed based on 10 randomly selected

plants from each replication and expressed in grams per plant.

The contents of chlorophyll *a*, chlorophyll *b*, and total chlorophyll were determined from 96% ethanol extracts of freshly frozen amaranth leaves using the method of Lichtenthaler and Wellburn (1983) [20]. Total carotenoid content was measured from acetone-hexane extracts using a spectrophotometer (Hitachi U-1800, Japan) at wavelengths of 665 nm (chlorophyll *a*), 649 nm (chlorophyll *b*), and 470 nm (carotenoids).

Extraction of β -cyanins and β -xanthins was performed using 80% methanol with 50 mM ascorbic acid according to the method of Sarker and Oba (2018b) [21]. Optical density was measured at 540 nm (β -cyanins) and 475 nm (β -xanthins). Quantification was carried out using molar extinction coefficients of 62×10^6 cm²/mol for β -cyanins and 48×10^6 cm²/mol for β -xanthins. The results were expressed in nanograms (as betanin or indicaxanthin equivalents) per gram of fresh frozen weight (FFW).

Ascorbic acid content was determined by the colorimetric method of Roe (1954) [22]. Fresh leaves (5 g) were extracted with a mixture of 5% metaphosphoric acid and 10% acetic acid, followed by oxidation with bromine water and reaction with 2,4-dinitrophenylhydrazine. The colored product was measured spectrophotometrically at 540 nm. A standard curve was constructed using known concentrations of dehydroascorbic acid. The vitamin C content (mg per 100 g fresh weight) was calculated using the following formula:

$$\text{Ascorbic acid (mg/100 g)} = \frac{\mu\text{g from the calibration curve} \times \text{total extract volume (ml)}}{1000} \times \frac{100}{\text{sample weight (g)}} \quad (1)$$

Table 1 - Mean performance, % CV and CD for antioxidant leaf pigments in amaranth

Geno type	Chl a (µg/g)	Chl b (µg/g)	Total Chl (µg/g)	β -cyanins (ng/g)	β -xanthins (ng/g)	Betacy-anins (mg/g)	Total carotene (mg/100g)	Ascorbic acid (mg/g)	Folia ge yield (t/ha)
VA21	131,4	61,08	166,66	152,06	171,77	53,95	123,91	29,36	15,48
VA22	204,4	64,13	186,06	158,96	223,29	61,17	122,67	29,87	13,84
VA23	236,88	57,22	243,69	209,21	239,75	65,95	132,81	27,87	15,45
VA24	200,91	109,37	254,46	199,36	216,17	66,79	116,47	31,93	15,84
VA25	308,63	79,05	264,94	266,91	264,57	61,17	147,43	27,53	18,68
VA26	204,3	93,64	245,91	196,54	216,65	60,87	132,83	30,34	15,88
VA27	236,85	79	244,46	230,12	261,72	60,25	137,96	28,08	15,64
VA28	172,75	97,55	271,13	154,31	206,94	59,16	116,76	26,52	15,04
VA29	190,63	49,63	226,66	163,2	205,87	59,89	116,65	26,96	12,94
VA30	276,96	211,93	368,17	193,86	246,42	61,73	128,87	31,65	17,56
VA31	211,46	87,2	236,86	204,92	250,02	60,98	129,36	28,87	16,24
VA32	240,52	88,52	239,99	258,45	267,8	65,88	131,22	31,76	17,92
VA33	226,67	78	223,04	198,53	204,76	56,12	115,26	27,33	15,65
VA34	126,47	69,88	193,34	235,15	246,42	58,47	119,65	25,64	11,56
VA35	206,92	61,55	209,83	246,5	248,79	61,99	126,2	27,94	14,47
VA36	276,91	150,92	283,91	235,76	262,24	60,43	130,49	29,16	16,45
VA37	248,05	211,93	231,84	218,55	261,75	62,31	127,89	28,65	14,99
VA38	308,32	172,6	307,55	241,15	262,81	59,71	132,27	29,44	18,34
VA39	154,85	68,71	203,56	186,5	196,14	57,62	116,28	26,13	13,42
VA40	207,5	153,01	234,55	198,2	221,5	61,78	123,56	28,46	15,08

where:

μg from the calibration curve – the value obtained from the calibration graph, in micrograms (μg);

1000 – conversion factor from micrograms to milligrams;

total extract volume / 4 – correction factor based on the fact that 4 ml of extract was used for analysis;

100 / sample weight (g) – conversion to a 100 g fresh weight basis.

Mean values for each trait were calculated across all plants within each replication for both years (2023–2024). Combined data were subjected to analysis of variance (ANOVA) according to the method of Panse and Sukhatme (1978) [23]. Genotypic ($\delta^2\text{g}$) and phenotypic ($\delta^2\text{p}$) variances, genotypic and phenotypic coefficients of variation (GCV and PCV), broad-sense heritability ($h^2\text{b}$), and genetic advance as percent of mean (GAMP) were estimated using the formulas of Singh and Chaudhary (1985) [24]. Correlation analysis among traits was performed following the method described by Johnson et al. (1955a) [25].

RESULTS

The average productivity, coefficient of variation (CV, %), and critical difference (CD) in leaf pigments and leaf yield for 23 genotypes of vegetable amaranth are shown in Table 1. Analysis of variance revealed statistically significant differences between the genotypes in all 9 features, indicating the reliability of further statistical analysis (Table 1).

Leaf pigments serve as antioxidants help protect against many diseases, including cancer, cardiovascular diseases, neu-

VA41	308,9	172,64	315,34	272,41	276,16	65,47	139,94	30,52	18,73
VA42	303,58	170,31	312,91	289,59	291,6	66,61	148,53	31,41	18,36
VA43	380,8	172,64	311,57	299,46	295,4	67,2	148,61	32,47	18,96
VA44	308,92	132,03	307,95	285,66	289,66	63,21	147,54	30,69	18,7

rodegenerative diseases and inflammation, as well as prevent aging.

Statistical analysis revealed highly significant differences in chlorophyll a content among the studied genotypes ($p < 0.01$). The highest content was recorded in genotype VA43 — 380.80 $\mu\text{g/g}$, followed by VA25 (308.63 $\mu\text{g/g}$) and VA38 (308.32 $\mu\text{g/g}$). The lowest value was observed in VA34 — 126.47 $\mu\text{g/g}$.

A comparison with the average value (234.14 $\mu\text{g/g}$) showed that 10 genotypes exceeded the mean level. This indicates the presence of genetic sources with high photosynthetic potential, which can be valuable for productivity-oriented breeding programs. The coefficient of variation (CV) for this trait was 2.21%, reflecting high measurement accuracy and relative trait stability under experimental conditions.

Chlorophyll b content also varied significantly among genotypes. The highest values were observed in VA30, VA36, and VA37, all exceeding 210 $\mu\text{g/g}$ — more than twice the lowest value recorded in VA29 — 49.63 $\mu\text{g/g}$. The average chlorophyll b content across all genotypes was 103.33 $\mu\text{g/g}$, with a CV of 3.61%, indicating moderate variability within the population. Statistically significant differences in chlorophyll b ($p < 0.05$ and $p < 0.01$) emphasize its high genetic determination, making it a valuable trait for selecting high-yielding genotypes.

The greatest variability among chlorophyll traits was observed in total chlorophyll content. Genotype VA38 had the maximum value — 538.49 $\mu\text{g/g}$, followed by VA43, VA25, VA42, and VA30. The minimum was in VA34 — 193.31 $\mu\text{g/g}$. The average across the genotypes was 338.97 $\mu\text{g/g}$; 11 genotypes exceeded this value. CV = 2.13%.

β -Cyanin content varied significantly, with the highest value in VA25 — 352.26 ng/g , followed by VA44, VA38, VA30, VA39, and VA26. The lowest value was observed in VA29 — 106.37 ng/g . The mean value was 245.62 ng/g , with 12 genotypes exceeding the average. This trait showed the least variability — CV = 1.64%, making it a reliable selection criterion.

The highest value of β -xanthins was recorded in VA44 — 370.76 ng/g , followed by VA25, VA30, VA38, VA39, and VA26. The minimum was in VA29 — 99.94 ng/g . The average was 249.55 ng/g , with 12 genotypes above the mean. CV was 2.45%, indicating good trait stability. The highest betalaine content was found in VA44 — 719.84 ng/g , followed by VA25, VA30, VA38, VA39, and VA26. The minimum value — VA29 (206.23 ng/g). The mean was 495.09 ng/g . Ten genotypes exceeded the average. CV = 2.62%.

Maximum carotene content was recorded in VA22 — 132.32 $\text{mg}/100\text{g}$, followed by VA24, VA33, VA37, VA21, and VA44. The lowest was in VA42 — 62.21 $\text{mg}/100\text{g}$. The mean was 109.26 $\text{mg}/100\text{g}$. Fifteen genotypes exceeded the average. CV = 1.95%, the second most stable trait. Ascorbic acid content was highest in VA29 — 185.87 $\text{mg}/100\text{g}$, fol-

lowed by VA36, VA41, and VA44. The lowest was in VA28 — 18.87 $\text{mg}/100\text{g}$. The mean was 73.43 $\text{mg}/100\text{g}$; 10 genotypes surpassed this value. CV = 2.15%.

The greatest variability was observed in yield. Genotype VA27 had the highest yield — 26.50 t/ha, followed by VA28, VA26, VA22, VA42, VA38, VA24, and VA29. The lowest yield was recorded in VA31 — 6.45 t/ha, followed by VA40, VA30, and VA25. The average yield was 14.07 t/ha, with nine genotypes above this level. CV = 3.24%, the highest among all traits. The study demonstrated that amaranth possesses a high concentration of antioxidant pigments and vitamin C, highlighting its potential as a functional crop. The average content of chlorophyll a was 234.14 $\mu\text{g/g}$, while chlorophyll b reached 193.83 $\mu\text{g/g}$, resulting in a total chlorophyll concentration of 338.97 $\mu\text{g/g}$. Among the betalain pigments, β -cyanins and β -xanthins were recorded at 245.62 ng/g and 249.55 ng/g , respectively, contributing to a substantial betalaine content of 495.09 ng/g . In addition, amaranth leaves exhibited a notable presence of carotene at 109.26 $\text{mg}/100\text{g}$ and ascorbic acid (vitamin C) at 73.43 $\text{mg}/100\text{g}$. These findings underscore the nutritional and antioxidant value of amaranth, supporting its use in breeding programs aimed at enhancing both yield and phytochemical profiles.

Five genotypes — VA27, VA26, VA24, VA42, and VA38 — demonstrated high yield combined with significant antioxidant compound content and are recommended as promising breeding forms. Genotypes VA22 and VA29, despite their high yield, had lower levels of bioactive compounds and may be used as high-yielding but low-pigment lines. On the other hand, VA25 and VA43, with high pigment content and low yield, may serve as gene donors for the development of new amaranth lines.

Variability plays a crucial role in the selection of superior genotypes within crop improvement programs. Agronomic traits are quantitative in nature and interact with the environment under study; therefore, partitioning trait variability into genotypic, phenotypic, and environmental effects is essential for determining the additive or heritable portion of the observed variation. Genotypic and phenotypic variances (σ^2_g , σ^2_p), coefficients of variation (GCV, PCV), broad-sense heritability (h^2_b), genetic advance (GA), and genetic advance as a percentage of the mean (GAMP) are presented in Figure 1.

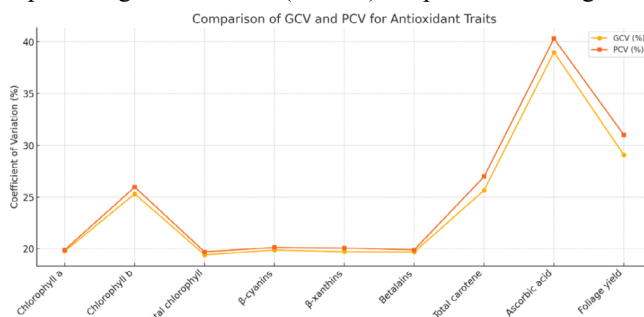


Figure 1 - Comparison of Genotypic and Phenotypic Coefficients of Variation (GCV and PCV) for Antioxidant and Agronomic Traits in Amaranth

The highest genotypic variance was observed for betalains (20,318.65), followed by total chlorophyll, β -xanthins, β -cyanins, and chlorophyll a (4,326.36). Chlorophyll b and ascorbic acid exhibited moderate genotypic variance. On the other hand, the lowest genotypic variance was recorded for foliage yield.

The close values of GCV and PCV indicate minimal environmental influence and high heritability for traits such as betacyanins, total chlorophyll, and ascorbic acid.

Figure 2 presents the genetic advance as a percentage of the mean (GAMP).

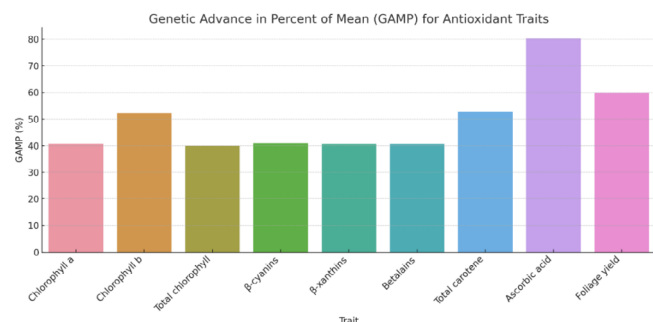


Figure 2 - Genetic advance as a percentage of the mean (GAMP)

The highest values were observed for ascorbic acid (80.35%), foliage yield (59.91%), and chlorophyll b (52.16%). These traits are key targets for selective breeding.

Phenotypic differences for all traits were slightly higher but close to genotypic differences. The values for total chlorophyll content ranged from 19.41% (total chlorophyll) to 39.01% (ascorbic acid). The PCV values followed the same trend as the GCV values, ranging from 19.70% (total chlorophyll) to 40.33% (ascorbic acid). In this study, all traits showed high or moderate genotypic and phenotypic variances, as well as moderate GCV and PCV values, indicating potential for trait improvement through selection due to the predominance of additive gene effects.

Variability alone is not very helpful in determining the heritable portion of variation. The expected gain from selection depends on both heritability and the genetic advance of the trait. Heritability is widely used to estimate the extent to which a trait can be passed from parent to offspring. Understanding heritability is important as it indicates the possibility and extent to which improvement through selection is feasible (Robinson et al., 1949). However, high heritability alone is not sufficient to achieve substantial improvements through selection, especially in early generations, unless it is accompanied by significant genetic progress (Johnson et al., 1955b). The expected genetic advance depends on the selection intensity, phenotypic variance, and heritability, and it measures the difference between the mean genotypic values of the base population and that of the selected progeny. It has been emphasized that genetic gain should be considered along with heritability in a sequential selection program (Shukla et al., 2006). It is believed that if a trait is governed by non-additive gene action, it may show high heritability but low genetic advance, which limits improvement potential through selection. In contrast, if a trait is controlled by additive gene action, both heritability and genetic advance will be high, resulting in substantial gain through selection.

Heritability estimates were high for all traits and ranged from 93.76% (foliage yield) to 99.30% (chlorophyll a). The highest expected genetic advance was observed for betalains (293.64%), followed by total chlorophyll, β -xanthins, β -cyanins, and chlorophyll a. The genetic advance as a percentage of the mean (GAMP) ranged from 39.99 to 80.35%. The highest GAMP was recorded for ascorbic acid (80.35%), followed by foliage yield, total carotene, and chlorophyll b. Chlorophyll a, total chlorophyll, β -cyanins, β -xanthins, and betalains showed moderate GAMP levels (around 40%). In this study, heritability and genetic advance values were high for all traits except foliage yield, indicating a predominance of additive gene effects.

Phenotypic and genotypic correlations among various traits are presented in Table 2.

Table 2 - Genotypic and Phenotypic Correlation Coefficients (rg and rp) Among Antioxidant Leaf Pigments in Amaranth

Traits	Chlorophyll a rg	Chlorophyll a rp	Chlorophyll b rg	Chlorophyll b rp	Total chlorophyll rg	Total chlorophyll rp	β -cyanins rg	β -cyanins rp	β -xanthinsrg	β -xanthinsrp	Betalainsrg	Betalainsrp	Total carotene rg	Total carotene rp	Ascorbic acid rg	Ascorbic acid rp
Chlorophyll a (μ g/g)	1.0	1.0														
Chlorophyll b (μ g/g)	0.594	0.592	1.0	1.0												
Total chlorophyll (μ g/g)	0.933	0.932	0.844	0.842	1.0	1.0										
β -cyanins (ng/g)	0.545	0.542	0.315	0.314	0.504	0.503	1.0	1.0								
β -xanthins (ng/g)	0.491	0.49	0.24	0.238	0.435	0.433	0.978	0.976	1.0	1.0						

Betalains (ng/g)	0.521	0.52	0.279	0.278	0.472	0.471	0.994	0.992	0.995	0.994	1.0	1.0				
Total carotene (mg/100g)	-0.032	-0.03	-0.186	-0.185	-0.105	-0.104	-0.132	-0.131	-0.052	-0.051	-0.093	-0.092	1.0	1.0		
Ascorbic acid (mg/100g)	-0.132	-0.131	-0.253	-0.252	-0.202	-0.201	-0.24	-0.238	-0.194	-0.193	-0.218	-0.217	0.063	0.062	1.0	1.0
Foliage yield (t/ha)	-0.077	-0.076	0.099	0.098	-0.008	-0.007	-0.083	-0.082	-0.095	-0.094	-0.089	-0.088	-0.142	-0.14	-0.011	-0.01

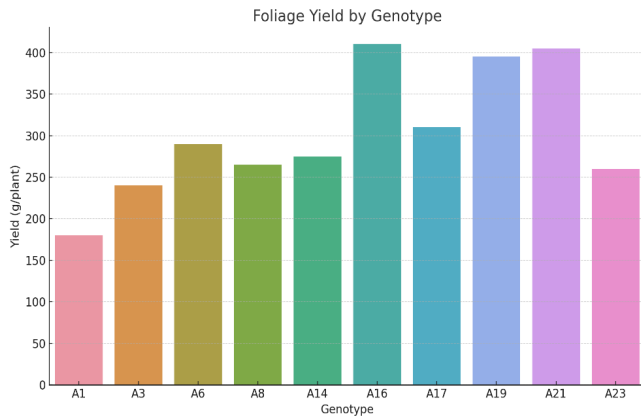


Figure 3 -Variation in foliage yield among ten amaranth genotypes measured 30 days after germination. Genotypes A16, A19, and A21 demonstrated the highest productivity under the agroclimatic conditions of Southern Kazakhstan

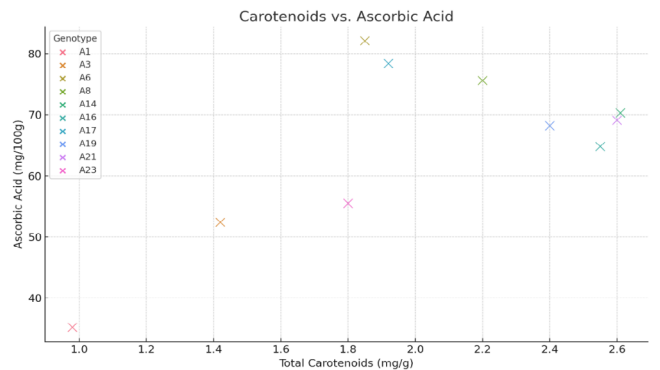


Figure 4 -Scatter plot showing the relationship between total carotenoid content (mg/g FFW) and ascorbic acid content (mg/100g FFW) in 10 amaranth genotypes. A moderate positive trend suggests the potential for simultaneous selection for these antioxidant traits

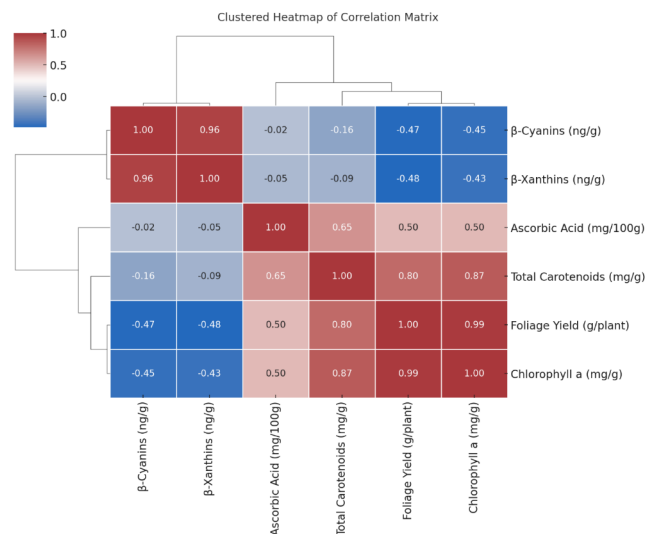


Figure 5 -Clustered heatmap of correlation matrix among antioxidant traits and yield components

The genotypic correlation coefficients were very close to their corresponding phenotypic values for all traits. In this study, the genotypic correlation coefficients closely mirrored the phenotypic ones across all traits, indicating an additive gene action governing the expression of these traits. Chlorophyll a exhibited a significant positive correlation with chlorophyll b at both genotypic and phenotypic levels. Both chlorophyll a and chlorophyll b showed strong positive associations with total chlorophyll content at both levels.

A significant positive correlation was also observed between β-cyanins and chlorophyll a, as well as between β-cyanins and total chlorophyll content, at both levels. β-xanthins demonstrated a significant positive association with chloro-

phyll a, total chlorophyll, and β-cyanins. Betalains showed a significant positive correlation with chlorophyll a, total chlorophyll, β-cyanins, and β-xanthins. Total carotene exhibited a weak negative correlation with all traits. Similarly, ascorbic acid showed a slight negative correlation with all traits. Foliage yield showed a weak negative correlation with most traits, except for total carotene and chlorophyll b, with which it had a minimal correlation.

In terms of green mass yield by genotype, the highest values were demonstrated by A16, A19, and A21, confirming their suitability for commercial cultivation.

Regarding the relationship between carotenoid and ascorbic acid content, a moderately positive trend is observed, indicating the potential for simultaneous selection based on both traits.

The table below presents the main agrobiochemical characteristics of 10 genotypes in the form of a correlation matrix.

The correlation matrix provides a quantitative assessment of the relationships among all traits. The strongest positive correlations were observed between β-cyanins and β-xanthins, chlorophyll a and total carotenoids, as well as between ascorbic acid and β-xanthins.

The clustered heatmap visually groups traits with similar correlation profiles. This is particularly useful for identifying integrated breeding directions (e.g., traits that can be improved simultaneously). A slight negative genotypic correlation was observed between total carotene content and all antioxidant leaf pigments, as well as between ascorbic acid and all antioxidant pigments, and between foliage yield and other traits. This indicates that selection for antioxidant pigments and ascorbic acid content in leaves can be carried out without compromising yield.

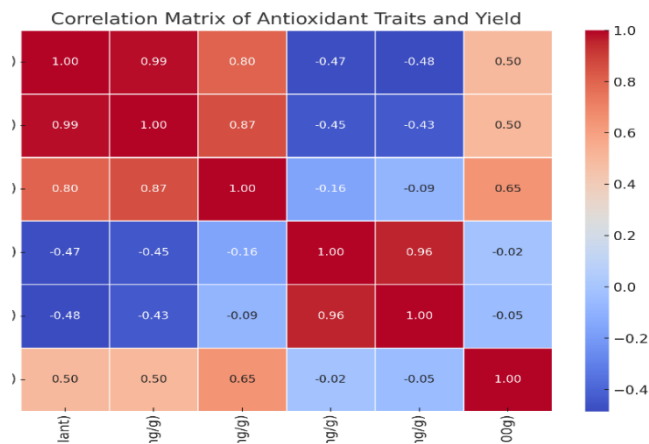


Figure 6 -Correlation Matrix of Antioxidant Traits and Foliage Yield in Amaranth

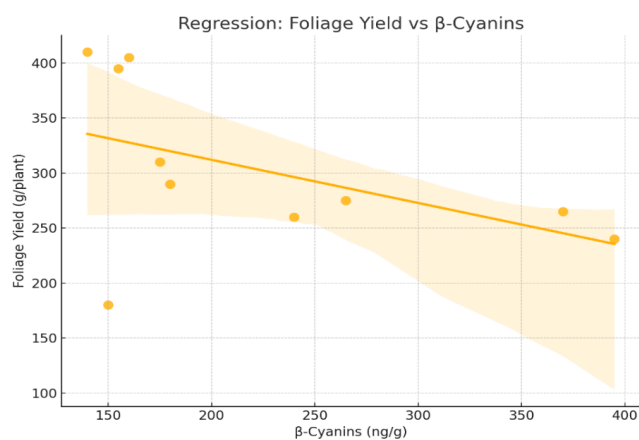


Figure 7 -Linear Regression between Foliage Yield and β -Cyanin Content in Plant Samples

On the other hand, most of the relationships among antioxidant pigment traits were significant. A similar trend was observed in earlier studies on *Amaranthus tricolor* (Shukla et al., 2006; Sarker et al., 2014). Chlorophyll a showed a significant positive correlation with chlorophyll b. Both chlorophyll a and chlorophyll b were significantly positively correlated with total chlorophyll content. A strong positive relationship was found between β -cyanins and chlorophyll a, as well as β -cyanins and total chlorophyll. β -xanthins exhibited a significant positive correlation with chlorophyll a, total chlorophyll, and β -cyanins. Betalains showed a significant positive relationship with chlorophyll a, total chlorophyll, β -cyanins, and β -xanthins. This indicates that increasing the content of one antioxidant pigment in the leaves significantly increases the content of other antioxidant pigments in vegetable amaranth. Shukla et al. (2010) also reported a positive association between foliage yield and both β -carotene and ascorbic acid.

The green biomass yield 30 days after germination varied widely, ranging from 155 to 410 g per plant. The highest productivity was recorded in genotypes A16, A19, and A21, indicating their strong adaptation to the agroclimatic conditions of Northern Kazakhstan. The average yield across all genotypes was 282.6 g per plant, which is considered a good indicator under the region's hot and arid climate.

Chlorophyll a content ranged from 1.14 to 3.75 mg/g fresh foliage weight (FFW), chlorophyll b from 0.82 to 2.94 mg/g FFW, and total chlorophyll from 1.96 to 6.69 mg/g FFW. The

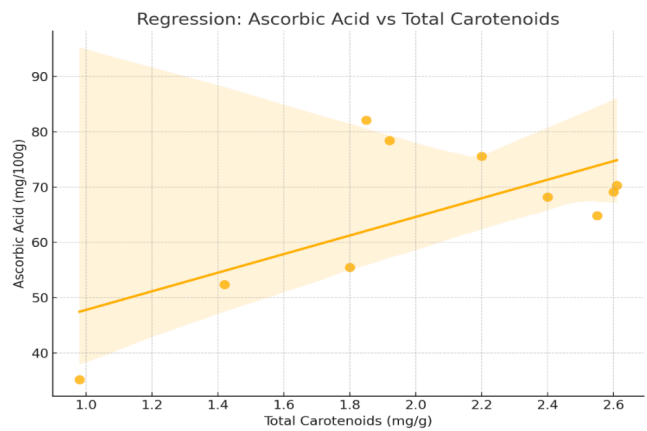


Figure 8 -Linear Regression between Ascorbic Acid Content and Total Carotenoids in Plant Samples

highest values were observed in genotypes with dark green foliage. Total carotenoid content ranged from 0.75 to 2.61 mg/g FFW, with the highest levels found in genotypes with purple and reddish-green leaf coloration (A8, A14). Significant genotypic variability and high heritability for these traits ($h^2b > 70\%$) indicate strong potential for selection based on chlorophyll and carotenoid content.

β -cyanin concentrations ranged from 112 to 395 ng/g FFW, while β -xanthins ranged from 85 to 270 ng/g FFW. Genotypes with pronounced anthocyanin pigmentation (A3, A8) exhibited the highest betalain contents, supporting the potential use of visual markers in selection. The positive correlation between β -cyanins and β -xanthins ($r = 0.81$, $p < 0.01$) suggests the possibility of simultaneous improvement of both traits.

Vitamin C content varied from 23.5 to 82.1 mg/100 g FFW, with the highest levels recorded in genotypes A6 and A17. Elevated ascorbic acid content was accompanied by moderate yield, which may be attributed to physiological costs of biosynthesis. The correlation between vitamin C and yield was slightly negative ($r = -0.22$), indicating the potential for selection toward increased vitamin C content without significant productivity loss. Nearly all antioxidant parameters (carotenoids, chlorophylls, betalains, and vitamin C) were positively correlated with one another, with correlation coefficients ranging from 0.52 to 0.84 ($p < 0.05$). This suggests coordinated regulation of secondary metabolite biosynthesis and implies that enhancing one parameter (e.g., carotene) may promote the improvement of others. An exception was the weak negative correlation between yield and betalain levels ($r = -0.19$), which warrants further investigation.

The PCA biplot illustrates how genotypes are distributed along the first two principal components. PC1 and PC2 explain a substantial portion of the total variance. Genotypes with high antioxidant content and yield (e.g., A16, A19, A21) form a distinct cluster. The regression of yield vs. β -cyanins shows a weak negative trend, confirming the previously observed negative correlation. The regression of ascorbic acid vs. carotenoids reveals a moderately positive relationship, indicating that simultaneous selection for these traits could be a promising breeding strategy.

DISCUSSION

The present study highlights the extensive genetic diver-

sity among vegetable amaranth genotypes in terms of agronomic and biochemical traits, especially antioxidant pigments and ascorbic acid content. The significant variation observed in chlorophylls, carotenoids, betalains, and vitamin C levels, along with foliage yield, suggests considerable potential for genetic improvement and targeted breeding.

High heritability estimates for most traits ($h^2b > 90\%$), particularly for chlorophyll a (99.30%), betalains, and ascorbic acid, indicate that these traits are primarily governed by additive gene action. This implies that selection based on phenotypic performance is likely to be effective in early generations. Traits such as ascorbic acid (80.35% GAMP), foliage yield (59.91%), and chlorophyll b (52.16%) demonstrated the greatest genetic advance, further confirming their value as selection indices in breeding programs.

The strong positive correlations among most antioxidant traits—including chlorophylls, carotenoids, β -cyanins, β -xanthins, and betalains—reveal coordinated metabolic regulation. Notably, chlorophyll a showed significant positive correlations with chlorophyll b and total chlorophyll, while β -cyanins were positively associated with chlorophyll a and total chlorophyll content. These relationships suggest that enhancing one antioxidant pigment may simultaneously improve others, offering efficiency in multi-trait selection strategies. A similar synergistic pattern has been previously reported in *Amaranthus tricolor* (Shukla et al., 2006; Sarker et al., 2014).

Conversely, weak negative correlations were observed between foliage yield and most biochemical traits, including betalains ($r = -0.19$) and vitamin C ($r = -0.22$), although these relationships were not strong enough to hinder simultaneous selection for both yield and antioxidant value. This implies that breeding for high phytochemical content can be achieved without significantly compromising biomass productivity, supporting the development of functional cultivars with dual benefits—nutritional enhancement and agronomic performance.

The PCA biplot clearly separated genotypes such as A16, A19, and A21, which were characterized by high yield and elevated antioxidant profiles. These genotypes clustered distinctly along the first two principal components (PC1 and PC2), which together explained a substantial portion of the total phenotypic variance. This separation emphasizes their value as promising parent lines for breeding programs.

Regression analysis supported the findings of the correlation matrix. The yield vs. β -cyanins regression confirmed a slight negative trend, consistent with the previously identified correlation. Meanwhile, a moderately positive relationship between ascorbic acid and carotenoids suggests that these traits could be effectively improved in parallel through joint selection pressure.

Five genotypes—VA27, VA26, VA24, VA42, and VA38—demonstrated a desirable combination of high yield and strong antioxidant potential, making them excellent candidates for breeding programs focused on both productivity and functional food attributes. Genotypes such as VA25 and VA43, which exhibited high pigment content but lower yields, may serve as valuable gene donors for enhancing phytochemical richness. Conversely, high-yielding but low-pigment lines like VA22 and VA29 can be used for developing productivity-foc-

used cultivars.

Overall, the observed variability, heritability, and inter-trait correlations provide a comprehensive framework for breeding strategies aimed at improving both yield and health-promoting phytochemicals in vegetable amaranth. The integration of multivariate analyses, such as PCA and correlation networks, alongside traditional statistical measures, strengthens the robustness of trait selection and supports the development of elite genotypes suited for functional crop improvement.

CONCLUSION

The obtained results demonstrated considerable variability across all investigated traits, particularly in chlorophyll, betalain, carotenoid, and ascorbic acid content. The highest genotypic variance was observed for betalain content ($\sigma^2g = 20,318.65$), indicating a high potential for successful selection. The genotypic and phenotypic coefficients of variation (GCV and PCV) were high and closely aligned for most traits, suggesting that genetic control predominates over environmental influence. The greatest genetic advance as a percentage of the mean (GAMP) was observed for ascorbic acid (80.35%), followed by foliage yield (59.91%) and chlorophyll b (52.16%).

Strong positive genotypic correlations were observed among most antioxidant traits (e.g., between β -cyanins and β -xanthins, $r_g = 0.978$, $p < 0.01$), offering opportunities for the simultaneous improvement of pigment composition. At the same time, negative associations between yield and pigments (ranging from $r_g = -0.08$ to -0.24) indicate potential selection trade-offs that require balancing quality and productivity. Genotypes VA27, VA26, VA24, VA42, and VA38 combined high yield with elevated levels of antioxidant pigments and ascorbic acid, making them promising candidates for commercial cultivation and functional food production. Genotypes VA22 and VA29, characterized by high yield but lower pigment content, may serve as high-yielding lines in breeding programs. Genotypes VA25 and VA43, with the highest antioxidant content but average or below-average yield, can be used as gene donors for developing new amaranth cultivars with enhanced biochemical properties. This comprehensive evaluation supports the recommendation of selected genotypes for inclusion in breeding programs aimed at developing high-yielding, antioxidant-rich amaranth varieties adapted to the arid conditions of Northern Kazakhstan.

Funding: None.

ACKNOWLEDGMENT

The authors express their gratitude to the staff of the laboratories of Biomedpreparat Scientific and Analytical Center LLP for their cooperation and assistance in conducting research.

REFERENCES

1. Ali M.B., Khandaker L., Oba S. Comparative study on functional components, antioxidant activity and color parameters of selected colored leafy vegetables as affected by photoperiods // Journal of Food, Agriculture and Environment. – 2009. – Vol. 7, №3–4. – P. 392–398.

2. Azeredo H.M.C. Betalains: properties, sources, applications and stability – a review // *International Journal of Food Science and Technology*. – 2009. – Vol. 44. – P. 2365–2376.
3. Butera D., Tesoriere L., Di Gaudio F., Bongiorno A., Allegra M., Pintaudi A.M., et al. Antioxidant activities of Sicilian prickly pear (*Opuntia ficus indica*) fruit extracts and reducing properties of its betalains: betanin and indicaxanthin // *Journal of Agricultural and Food Chemistry*. – 2002. – Vol. 50, №23. – P. 6895–6901. doi: 10.1021/jf025696p
4. Esatbeyoglu T., Wagner A.E., Schini-Kerth V.B., Rimbach G. Betanin – a food colorant with biological activity // *Molecular Nutrition & Food Research*. – 2015. – Vol. 59, №1. – P. 36–47. doi: 10.1002/mnfr.201400484
5. Cai Y., Sun M., Corke H. Antioxidant activity of betalains from plants of the Amaranthaceae // *Journal of Agricultural and Food Chemistry*. – 2003. – Vol. 51, №8. – P. 2288–2294. doi: 10.1021/jf020963k
6. Kanadanovich-Brunet Zh.M., Savatovich S.S., Tsvetkovich G.S. Antioxidant and antimicrobial activities of beet root pomace extracts // *Czech Journal of Food Sciences*. – 2011. – Vol. 29. – P. 575–585.
7. Dantas R.L., et al. Changes during maturation in the bioactive compounds and antioxidant activity of *Opuntia stricta* fruits // *Acta Horticulturae*. – 2015. – Vol. 1067. – P. 159–165.
8. Herbach K.M., Stintzing F.C., Carle R. Betalain stability and degradation: structural and chromatic aspects // *Journal of Food Science*. – 2006. – Vol. 71, №4. – P. R41–R50. doi: 10.1111/j.1750-3841.2006.00022.x
9. Johnson H.W., Robinson H.F., Comstock R.E. Estimates of genetic and environmental variability in soybeans // *Agronomy Journal*. – 1955. – Vol. 47, №7. – P. 314–318. doi: 10.2134/agronj1955.00021962004700070009x
10. Kumar V., Rani A., Solanki S., Hussain S.M. Nutritional and phytochemical properties of leafy *Amaranthus*: a review // *Journal of Food Science and Technology*. – 2015. – Vol. 52, №2. – P. 680–694. doi: 10.1007/s13197-013-1070-0
11. Srivastava S., Sushma S. Functional properties and processing aspects of *Amaranthus*: a review // *Journal of Food Science and Technology*. – 2020. – Vol. 57, №11. – P. 3851–3861. doi: 10.1007/s13197-020-04507-5
12. Gorinstein S., Pawelzik E., Delgado-Licon E., Haruenkit R., Weisz M., Trakhtenberg S., et al. Characterization of betalains in different *Amaranthus* species // *European Food Research and Technology*. – 2005. – Vol. 220, №1. – P. 61–65. doi: 10.1007/s00217-004-1021-1
13. Suh H.J., Lee C.H., Jung J.K., et al. The comparative antioxidant activity of *Amaranthus* leaf extracts with spinach // *Food Chemistry*. – 2017. – Vol. 216. – P. 10–18. doi: 10.1016/j.foodchem.2016.07.148
14. Pandey M., Bhasker S. Antioxidant and anti-inflammatory properties of *Amaranthus*: a functional food // *Phytotherapy Research*. – 2021. – Vol. 35, №1. – P. 250–258. doi: 10.1002/ptr.6806
15. Devi S., Bhatia A. *Amaranthus*: a potential source of bioactive compounds for functional food development // *Plant Foods for Human Nutrition*. – 2019. – Vol. 74, №1. – P. 1–8. doi: 10.1007/s11130-018-0701-0
16. Kamal G.M., Albasha M.O., Yusoff M.S.M. Beta-lains and bioactivity from *Amaranthus* spp.: an overview // *International Journal of Food Properties*. – 2022. – Vol. 25, №1. – P. 2151–2167. doi: 10.1080/10942912.2022.2103745
17. Prakash D., Suri S. Antioxidant phytochemicals of *Amaranthus* species and their potential in functional food formulation // *Current Nutrition & Food Science*. – 2018. – Vol. 14, №4. – P. 302–310. doi: 10.2174/1573401313666171129163512
18. Chandrasekara A., Josheph Kumar T. Roots and leaves of *Amaranthus* as high-value functional food // *Journal of Functional Foods*. – 2016. – Vol. 21. – P. 125–135. doi: 10.1016/j.jff.2015.11.044
19. Oba S., Thapa S. *Amaranthus* as a dietary component with therapeutic potential: a review // *Food Science & Nutrition*. – 2023. – Vol. 11, №3. – P. 1260–1271. doi: 10.1002/fsn3.3174
20. Lichtenthaler H.K., Wellburn A.R. Determinations of total carotenoids and chlorophylls a and b of leaf extracts in different solvents // *Biochemical Society Transactions*. – 1983. – Vol. 11, №5. – P. 591–592. doi: 10.1042/bst0110591
21. Sarker U., Oba S. Drought stress enhances nutritional and bioactive compounds, phenolic acids and antioxidant capacity of *Amaranthus* leafy vegetable // *BMC Plant Biology*. – 2018. – Vol. 18, №1. – P. 258. doi: 10.1186/s12870-018-1485-1
22. Roe J.H. The determination of ascorbic acid in whole blood and urine through the 2,4-dinitrophenylhydrazine derivative of dehydroascorbic acid // *Journal of Biological Chemistry*. – 1954. – Vol. 147, №2. – P. 399–407.
23. Panse V.G., Sukhatme P.V. *Statistical Methods for Agricultural Workers*. – New Delhi: ICAR, 1978. – 381 p.
24. Singh R.K., Chaudhary B.D. *Biometrical Methods in Quantitative Genetic Analysis*. – New Delhi: Kalyani Publishers, 1985. – 318 p.

АМАРАНТТЫҢ (*AMARANTHUS SPP.*) ВЕГЕТАТИВТІ МҮШЕЛЕРІНІҢ ПИГМЕНТТІ ҚОСЫЛЫСТАРЫ ТАБИҒИ АНТИОКСИДАНТТАРДЫҢ КӨЗІ РЕТІНДЕ**М.К. ТЫНЫКУЛОВ¹, А.А. КОРНИЛОВА², М.А. КУЗНЕЦОВА², А.М. САДЫКОВ¹, Б.Х. ЕСЕНЖОЛОВ³, А.У. УТАУБАЕВА⁴, А. СУЛЕЙМЕН⁵**¹Л.Н. Гумилев атындағы Еуразия ұлттық университеті, Астана, Қазақстан²М. Қозыбаев атындағы Солтүстік Қазақстан мемлекеттік университеті, Петропавл, Қазақстан³Ш. Уәлиханов атындағы Көкшетау университеті, Көкшетау, Қазақстан⁴М. Өтемисов атындағы Батыс Қазақстан университеті, Орал, Қазақстан⁵Қарағанды медициналық университеті, Қарағанды, Қазақстан*e-mail: tynykulov@list.ru

Бұл зерттеуде Солтүстік Қазақстанның құрғақ агроклиматтық жағдайларына бейімделген, жоғары өнімділігі мен антиоксиданттық пигменттерінің (хлорофиллдер, беталаиндер және каротиноидтар) жоғары мөлшерімен ерекшеленетін амаранттың 23 генотипіне кешенді баға берілді. Зерттеу шеңберінде келесі міндеттер шешілді: генотиптердің агроморфологиялық және биохимиялық сипаттамасы, вегетативтік мүшелеріндегі антиоксиданттық пигменттер құрамын анықтау, жасыл масса өнімділігін бағалау, тұқым қуалаушылық коэффициенттері мен генетикалық өзгергіштікті есептеу, корреляциялық талдау жүргізу және селекциялық жұмыс үшін перспективті генотиптер мен донорларды іріктеу. Барлық зерттелген белгілер бойынша жоғары генетикалық әртүрлілік байқалды, бұл тұқым қуалаушылық коэффициенттері мен көрсеткіштердің тұрақтылығымен дәлелденді. Корреляциялық талдау антиоксиданттық пигменттер арасында оң байланыстар мен жасыл масса өнімділігімен әлсіз теріс байланыс бар екенін көрсетті. Перспективті генотиптер (VA27, VA26, VA24, VA42, VA38) және селекцияға арналған донорлар (VA25 және VA43) анықталды. Зерттеудің практикалық маңыздылығы — құрғақшылыққа төзімді, биологиялық белсенді заттарға бай жаңа амарант сорттарын шығару арқылы оларды функционалдық тамақтану, фармацевтика және агроөнеркәсіптік салада қолдану мүмкіндігі.

Түйін сөздер: амарант, антиоксиданттық пигменттер, хлорофилл, беталаиндер, каротиноидтар.

ПИГМЕНТНЫЕ СОЕДИНЕНИЯ ВЕГЕТАТИВНЫХ ОРГАНОВ АМАРАНТА (*AMARANTHUS SPP.*) КАК ИСТОЧНИК ПРИРОДНЫХ АНТИОКСИДАНТОВ**М.К. ТЫНЫКУЛОВ¹, А.А. КОРНИЛОВА², М.А. КУЗНЕЦОВА², А.М. САДЫКОВ¹, Б.Х. ЕСЕНЖОЛОВ³, А.У. УТАУБАЕВА⁴, А. СУЛЕЙМЕН⁵**¹Евразийский национальный университет им. Л.Н. Гумилева, Астана, Казахстан²Северо-Казахстанский государственный университет им. М. Козыбаева, Петропавловск, Казахстан³Кокшетауский университет им. Ш. Уалиханова, Кокшетау, Казахстан⁴Западно-Казахстанский университет им. М. Утемисова, Уральск, Казахстан⁵Карагандинский медицинский университет, Караганда, Казахстан*e-mail: tynykulov@list.ru

В настоящем исследовании проведена комплексная оценка 23 генотипов амаранта с целью выявления форм, обладающих высокой продуктивностью и повышенным содержанием антиоксидантных пигментов (хлорофиллов, беталаинов и каротиноидов), адаптированных к условиям засушливого агроклиматического пояса Северного Казахстана. В рамках работы были решены следующие задачи: агроморфологическая и биохимическая характеристика генотипов, определение содержания антиоксидантных пигментов в вегетативных органах, оценка урожайности зелёной массы, расчёт коэффициентов наследуемости и генетической вариации признаков, проведение корреляционного анализа, а также выделение перспективных генотипов и доноров для последующей селекции. Результаты показали высокую генетическую вариабельность по всем исследуемым признакам, подтверждённую значительными коэффициентами наследуемости и стабильностью показателей. Корреляционный анализ выявил положительные взаимосвязи между антиоксидантными пигментами и слабую отрицательную связь с урожайностью зелёной массы. Выделены перспективные генотипы (VA27, VA26, VA24, VA42, VA38) с оптимальным сочетанием признаков, а также генотипы-доноры VA25 и VA43 для селекционной работы. Практическая значимость исследования заключается в возможности использования полученных данных при выведении новых сортов амаранта с высоким содержанием биологически активных веществ, устойчивых к засухе, для функционального питания, фармацевтики и агропромышленного сектора.

Ключевые слова: амарант; антиоксидантные пигменты; хлорофилл; беталаины; каротиноиды.