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## **ORIGINAL PAPER**

# The effect of mineral fertilizer doses on amaranth grain quality in the wet climate of Western Ukraine

Mariia Tyrus<sup>1</sup>, Volodymyr Lykhochvor<sup>1</sup>, Petro Hnativ<sup>2</sup>, Wieslaw Szulc<sup>3</sup>, Beata Rutkowska<sup>3</sup>, Oleg Andrushko<sup>4</sup>, Ivanna Rozko<sup>5</sup>, Olga Lytvyn<sup>1</sup>, Ivan Dudar<sup>6</sup>, Serhii Pavkovych<sup>6</sup>

 <sup>1</sup> Department of Technologies in Crop Production
<sup>2</sup> Institute of Agriculture of Carpathian Region National Academy of Agrarian Science, Lviv, Ukraine
<sup>3</sup> Division of Agricultural and Environmental Chemistry Warsaw University of Life Sciences, Warsaw, Poland
<sup>4</sup> Department of Genetics, Plant Breeding and Plant Protection
<sup>5</sup> Department of Horticulture and Vegetable Growing
<sup>6</sup> Department of Animal Husbandry and Fodder Production Lviv National Environmental University, Lviv, Ukraine

#### Abstract

Amaranth (Amaranthus spp.) is a promising alternative crop, which is gaining more popularity and a more widespread distribution throughout the world. The aim of this study has been to find out how the doses of mineral fertilizers affect the yield and quality of amaranth grain in the wet and warm climate of the Western Forest Steppe of Ukraine. The research was conducted from 2020 to 2022, on an experimental field of the Lviv National Environmental University (LNEU - Dublyany). The soil of the experimental field is Gravic Luvic Phaeozem. The experiment comprised seven different fertilizer treatments:  $N_0P_0K_0$ ,  $N_{40}P_{20}K_{40}$ ,  $N_{80}P_{40}K_{80}$ ,  $N_{120}P_{40}K_{80}$ ,  $N_{160}P_{60}K_{120}$ ,  $N_{200}P_{80}K_{120}$ ,  $N_{200}P_{80}K_{120}$ ,  $N_{200}P_{80}K_{120}$ ,  $N_{200}P_{80}K_{120}$ . The tested plant was amaranth of the Kharkivskyi 1 variety. Asystematic increase in fertilizer doses, particularly nitrogen, led to a gradual rise in amaranth grain yield. In the control without fertilizers, the yield was 2.31 Mg ha<sup>-1</sup>. With an increase in fertilizer application, it rose by 2.57 Mg ha<sup>-1</sup>, reaching 4.88 Mg ha<sup>-1</sup> at the highest fertilizer dose  $N_{200}P_{s0}K_{160}$ . The grain bulk density was higher (790 g l<sup>-1</sup>) in the variants without fertilizers, and the lowest when  $N_{200}P_{s0}K_{160}$  and  $N_{200}P_{s0}K_{180}$  were applied, where it was 755 g l<sup>-1</sup> and 750 g l<sup>-1</sup>. The protein content in amaranth grains increased from 16.2% to 18.9%, reaching its peak under the influence of an increase in mineral fertilizer doses from  $N_{40}P_{20}K_{40}$  to  $N_{200}P_{s0}K_{160}$ . The starch content decreased from 67.4% on the plot without fertilizers to 64.8% when applying the highest doses of fertilizers, i.e.  $N_{200}P_{80}K_{120}$  and  $N_{200}P_{80}K_{160}$ . Statistical models of correlations showed that the doses of mineral fertilizers, especially nitrogen, have a strong influence on the indicators of yield and grain quality of amaranth in the warm and humid climate of western Ukraine. High doses of fertilizers significantly increase yield, but negatively affect the grain bulk density and starch content, while significantly increasing the proportion of protein.

Keywords: amaranth, fertilization, yield, quality

Wiesław Szulc, Division of Agricultural and Environmental Chemistry, Warszaw, Nowoursynowska 166, 02-787 Warszawa, Poland, e-mail: wieslaw\_szulc@sggw.edu.pl

## INTRODUCTION

Agricultural intensification is based on the widespread use of a small number of high-yielding, profitable crops. This makes the field of crop production vulnerable and narrows the variety of crops (Piecyk et al. 2009, Weerasekara, Waisundara 2020). It also causes agronomic problems, deteriorates the environment, and impoverishes the menu in human nutrition. These facts encourage the involvement of new alternative plant resources in agricultural production.

Amaranth (*Amaranthus* spp.) is a promising alternative crop (Kozak et al. 2011), which is gaining more and more popularity and broader distribution (Weerasekara, Waisundara 2020, Tyrus et al. 2023). It is believed that growers are rediscovering amaranth today. It is known in the food, pharmaceutical, and cosmetic industries. Amaranth is used for the prevention and treatment of certain diseases, such as coronary heart disease, hypertension, diabetes, etc. (Repo-Carrasco et al. 2009). The cultivation of amaranth is a promising option for the pharmaceutical industry and for the production of many functional food products. Amaranth grain has unique nutritional properties and various uses (Skwaryło-Bednarz 2012, Lykhochvor et al. 2022). It has excellent nutritional value and ensures high digestibility of starch and proteins in products (Valentiuk 2019). It is also noted that amaranth as a source of high-quality protein, nutritionally balanced, can be a solution to famine in developing countries (Grobelnik et al. 2010).

Amaranth possesses a valuable property: the unique protein content in the grain (15-18%), which exceeds that of other grain crops (Venskutonis, Kraujalis 2013). It has a higher protein content (by 4.4%) and fats (by 5.8%) than products made from wheat flour (Malkar et al. 2009). Amaranth is distinguished by its high-quality proteins, which are balanced in terms of amino acid composition. The grain contains an elevated amount of vitamins and mineral compounds (Adegbola et al. 2020).

Amaranth contains high-quality proteins with a balanced amino acid content. The grain also has an increased amount of vitamins and mineral salts (Adegbola et al. 2020). These aspects of amaranth grain quality have not been comprehensively considered in research for a long time (Repo--Carrasco et al. 2009, Goptsiy 2018).

Amaranth oil contains more than 70% mono- and polyunsaturated fatty acids. These are linoleic (Omega-6), oleic (Omega-9), linolenic (Omega-3), arachidonic, palmitoleic and other acids. Amaranth seed oil contains 9% phospholipids (dominated by phosphatidylcholine), squalene (more than 8%), approximately 2% vitamin E, as well as phytosterols (more than 2%), carotenoids (provitamin A), vitamin D, bile acids, various macro- and trace elements (potassium, iron, phosphorus, calcium, magnesium, copper, and others) (Akin-Idowu et al. 2017, Karamać et al. 2019). Amaranth production has been positively evaluated as a source of dietary fiber and some bioactive compounds. The exceptional nutritional value and unique chemical composition of amaranth grain make it a crop of the future.

Amaranth seeds are a potentially rich source of polyphenols and glutenfree protein compounds, particularly when grown on fertilized soil (Vedmedeva et al. 2021). It contains macronutrients (lipids, proteins, carbohydrates and dietary fibers) and other important components, in particular squalene, tocopherols, phenols, phytates and vitamins. These aspects of amaranth have not been comprehensively reviewed in research for a long time (Rastogi, Shukla 2013, Goptsiy 2018).

Amaranth oil contains more than 70% of mono- and polyunsaturated fatty acids: linoleic (Omega-6), oleic (Omega-9), linolenic (Omega-3), arachidonic, palmitoleic acids, etc., more than 9% of phospholipids (dominated by phosphatidylcholine), squalene (over 8%), about 2% of vitamin E, phytosterols (over 2%), carotenoids (precursors of vitamin A), vitamin D, bile acids, various macro- and microelements (potassium, iron, phosphorus, calcium, magnesium, copper, etc.) – Akin-Idowu et al. (2017), Karamać et al. (2019).

Amaranth grain products have various medicinal properties, as they are a source of antioxidants (Gimplinger et al. 2007, Akin-Idowu et al. 2017).

Amaranth is very adaptable to growing conditions; it is a good precursor in crop rotation, has an ameliorative effect on the soil, and increases its fertility (Adegbola et al. 2020).

Amaranth surpasses maize in terms of nutrient requirements (Aderibigbe et al. 2020). Fertilizer doses and sowing rates have a significant effect on the oil content and fatty acid content of amaranth grains (Ardali 2014).

Amaranth culture is gaining more and more popularity among producers, industry and consumers of products owing to the unique nutritional quality of the grain, especially considering the properties of its proteins, fats and starch (Skwaryło-Bednarz 2012). Amaranth is used both as a leafy vegetable and as a grain crop all over the world. Amaranth is a potential source of antioxidants and thus of great interest to nutritionists. Amaranth products are vital for the prevention and treatment of diseases caused by reactive oxygen species (Adegbola et al. 2020).

Some scientists (Alegbejo 2015) determine the difference in the chemical composition of plants that were grown in different climatic zones. Saturated fatty acids predominate in grains in southern latitudes. In northern regions, mono- and polyunsaturated fatty acids prevail. Studies have shown (Kaźmierczak et al. 2011) that amaranth grown in the Left Bank Forest Steppe of Ukraine accumulates more unsaturated fatty acids than saturated ones, making it a more valuable raw material for oil production and a source of bioactive substances.

Other data suggest that the composition of fatty acids in the oil obtained from amaranth grain is primarily influenced by varietal characteristics. Other factors are the weather during the growing season in the years of research and the doses of nitrogen fertilizers (Valentiuk 2019). Despite a large number of experiments with amaranth, several questions regarding its cultivation, nutritional properties, and potential applications remain unanswered, highlighting the need for additional research (Mlakar et al. 2009).

Amaranth is highly adaptable to growing conditions. As a crop in crop rotation, it serves as a good precursor and ameliorant, contributing to increased soil fertility (Adegbola et al. 2020). However, the high yield of amaranth entails its great need for mineral nutrients. Amaranth is even more demanding of nutrients than maize, which also belongs to crops of tropical origin (Aderibigbe et al. 2020). Mineral fertilizers affect both the level of productivity and quality indicators of amaranth grain. A particularly high content of fat in grain was obtained at the highest level of NPK fertilization. Increased levels of NPK had a significant positive effect on the amount of  $\alpha$ -tocopherol. Fertilizer application dose and sowing rate affected the oil content and fatty acid content (Ardali 2014).

Our goal was to investigate how different doses of mineral fertilizers affect the yield and quality of amaranth grain in the wet and warm climate of the Western Forest Steppe of Ukraine. The soil type in this region is darkgrey, low-humus, light loam, specifically classified as a Greyic Luvic Phaeozem.

## MATERIALS AND METHODS

We conducted the research from 2020 to 2022, on an experimental field of the Lviv National University of Nature Management (LNUP – Dublyany). The university is located in the Western Forest Steppe of Ukraine (N 49°54'14"; E 24°05'10"). The soil of the experimental field is a Grayic Luvic Phaeozem (WRB 2015). The soil of the control plot of the experiment had the following agrochemical parameters before applying fertilizers: the humus content is average in the arable layer of the soil, at 2.3%, and decreases with depth (DSTU 4289:2004). The soil is classified as poorly supplied with mobile nitrogen as its content in the soil according to the Kornfield method is 80-84 mg kg<sup>-1</sup> (DSTU 7863:2015. 2018). The soil has an average supply of mobile phosphorus compounds – 108-114 mg kg<sup>-1</sup> of soil, and potassium – 98-108 mg kg<sup>-1</sup> of soil (according to the Chirykov method (DSTU ISO 10390:2001. 2008).

Hydrothermal conditions differed relative to long-term averages. Warmer temperatures and increased rainfall were observed during 2020-2022. In 2019, the average temperature during the growing season was 16.1°C, which was 1.3°C higher than the long-term average. This figure was 15.3°C in 2020, which was 0.5°C higher, and 14.8°C in 2021, which was in line with the long-term average. During the growing season, precipitation exceeded the long-term average by 53 mm in 2019, 129 mm in 2020, and 73 mm in 2021.

The experiment was set up in a completely randomized design. The area of the experimental plot was 30 m<sup>2</sup>. Seven fertilizer combinations were tested in three replications:  $N_0P_0K_0$ ,  $N_{40}P_{20}K_{40}$ ,  $N_{80}P_{40}K_{80}$ ,  $N_{120}P_{40}K_{80}$ ,  $N_{160}P_{60}K_{120}$ ,  $N_{200}P_{80}K_{120}$ ,  $N_{200}P_{80}K_{160}$ . Mineral fertilizers were applied for the crops of the rotation system in the form of ammonia nitrate (34.4%), double superphosphate (42%), potassium chloride (60%). The preceding crop for amaranth was winter wheat. We started tilling the field after harvesting the preceding crop, and we ploughed the field in October. In autumn of each year, phosphorus and potash fertilizers were applied under ploughing. We prevented moisture loss by applying heavy harrows in early spring. Sowing was carried out in rows spaced 45 cm apart to a depth of 1 cm on April 17th of each year. A Horsch Pronto 4 DS seeder was used. Inter-row tillage for weed control was performed. Amaranth harvesting took place in the phase of full ripeness of the grain. Amaranth was threshed after drying.

We conducted field studies of amaranth productivity, which included the determination of grain bulk density (ISO 7971-3:2019) and the measurement of the mass of 1000 seeds (g) (Methodology for...2016). Additionally, we analyzed the grain dry matter composition, focusing on indicators such as crude protein content determined by the Kjeldahl method or protein collection per unit area (Tkachyk 2016). Starch content was analyzed with the polarimetric method according to Ewers (1908).

The results were statistically analyzed using MS Excel and analysis of variance (ANOVA) in the Statistica software.

## **RESULTS AND DISCUSSION**

The fertilization regime implemented in the experiment had a significant impact on the yield of amaranth. It was  $2.31 \text{ t} \text{ ha}^{-1}$  in the control and increased by  $2.57 \text{ t} \text{ ha}^{-1}$  to the level of  $4.88 \text{ t} \text{ ha}^{-1}$  at the highest dose of fertilizers. The increase was more than two times (Table 1).

Amaranth is a plant that responds well to mineral fertilization. Nitrogen fertilization increased the content of chlorophyll, carotenoids, protein, activity of antioxidant enzymes, and amaranth biomass. Previous authors (Karami et al. 2020) found that the highest yields in water deficit conditions were obtained with the combined application of low doses of nitrogen fertilizers (80 kg N ha<sup>-1</sup>) in combination with zeolite. Under these conditions, the highest plant yields of over 9 Mg ha<sup>-1</sup> were achieved, which was more than 30% higher compared to the no-fertilizer treatment. In our previous studies (Tyrus, Lykhochvor 2022), it was shown that amaranth yield increased in the entire range of applied doses from 0 to 200 kg N ha<sup>-1</sup>, from 2.31 Mg ha<sup>-1</sup> to 4.88 Mg ha<sup>-1</sup>. Nitrogen was the element that most affected the yield of this plant.

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The research conducted showed that the bulk grain density of amaranth Kharkivskyi 1 variety was the highest (790 g l<sup>-1</sup>) in the control without fertilizers (Table 1). The bulk density of grain decreased with the dose of fertilization to 784 g l<sup>-1</sup> on  $N_{40}P_{20}K_{40}$  object, and it decreased to 750 g l<sup>-1</sup> with a further increase in the amount of applied fertilizers to  $N_{200}P_{80}K_{160}$ . Bulk density depends most on the shape and size of the grain. The more seeds fit in one hectoliter volume, the higher the bulk density will be. A larger grain size usually reduces the bulk density, as the porosity of one hectoliter of grain increases (Venskutonis, Kraujalis 2013). Mass of 1000 seeds varied under different doses of mineral fertilizers (Table 1). The fertilizer dose  $N_{160-200}P_{60-80}K_{120-160}$  had the greatest effect on increasing the mass of 1000 grains – up to 0.89-0.91 g.

Table 1

| Fertilization          | Grain yield<br>(Mg ha <sup>.1</sup> ) | Bulk density<br>(g l <sup>·1</sup> ) | Mass of 1000<br>seeds (g) | Protein<br>(% of DM) | Starch<br>(% of DM) |
|------------------------|---------------------------------------|--------------------------------------|---------------------------|----------------------|---------------------|
| $N_0 P_{0K} 0$         | 2.311 <i>a</i>                        | 790.1a                               | 0.861a                    | 16.21a               | 67.42a              |
| $N_{40}P_{20}K_{40}$   | 2.980b                                | 784.5a                               | 0.872a                    | 16.83a               | 67.01 <i>a</i>      |
| $N_{80}P_{40}K_{80}$   | 3.542c                                | 775.2a                               | 0.885a                    | 17.45ab              | 66.21 <i>a</i>      |
| $N_{120}P_{40}K_{80}$  | 3.951cd                               | 772.1a                               | 0.881 <i>a</i>            | 18.01ab              | 65.53a              |
| $N_{160}P_{60}K_{120}$ | 4.503d                                | 760.5a                               | 0.892a                    | 18.51b               | 65.35a              |
| $N_{200}P_{80}K_{120}$ | 4.842d                                | 755.2a                               | 0.903a                    | 18.93b               | 64.82a              |
| $N_{200}P_{80}K_{160}$ | 4.883d                                | 750.1a                               | 0.911 <i>a</i>            | 18.90b               | 64.83a              |

Grain yield, bulk density, mass of thousand seeds, protein and starch content in amaranth depending on fertilization, averages for 2019-2021

Research conducted in Ukraine showed that there is a significant effect of fertilization on the physical parameters of amaranth grain quality, depending on the fertilizer doses (Goptsiy 2018). Bulk grain density was lower (702 g and 699 g) in variants with  $N_{30}P_{60}K_{60}$  and  $N_{60}P_{60}K_{60}$  fertilizer doses. A further increase in the nitrogen dose increases the grain bulk density. The authors attribute it to an increase in the share of small grain, which occurs as a result of its additional formation on side shoots under the influence of higher mineral nutrition. The species *Amaranthus hypochondriacus*, which is characterized by a larger grain than *Amaranthus hybridus*, achieved lower bulk density (Goptsiy 2018).

We found that the protein content in amaranth seeds ranged from 16.2 to 18.9%. It was the lowest in the control without fertilizers – 16.2% (Table 1). The protein content reached the level of 16.8% in the treatment with the of  $N_{40}P_{20}K_{40}$  fertilization, and increased to 17.4% in response to the  $N_{80}P_{40}K_{80}$  fertilization. It exceeded the control variant by 1.2%. With a further

Values marked with the same letter within each variant of the experiment do not differ significantly from each other according to the Tukey test (P<0.05).

increase in the dose of fertilizers, the protein content increased even more, and was 18.9% against the background of  $N_{200}P_{80}K_{120}$ . The increase in the content of protein was 2.7% compared to the variant without fertilizers. These results are also confirmed by Karami et al. (2020), who also achieved a reduction in grain bulk density by increasing doses of nitrogen fertilizers. These authors also pointed out that the bulk density of grain was influenced by soil moisture. Researchers have found that fertilization significantly influences the protein content in amaranth seeds. The lowest content was observed in the species Amaranthus hypochondriacus on unfertilized soil, measuring 16.0% in studies conducted in the Kharkiv region. The highest protein content (18.8%) was obtained with the application of the  $N_{30}P_{60}K_{60}$ fertilizer dose. However, in the object with the application of  $N_{60}P_{60}K_{60}$ the protein content decreased to 18.3%. Furthermore, when the dose of fertilizers was increased to  $N_{_{90}}P_{_{60}}K_{_{60}}$  and  $N_{_{120}}P_{_{60}}K_{_{60}}$ , the protein content decreased to 17.8% and 16.6%, respectively. The authors of the research explain this decrease as a result of the proportion of different quality seeds due to their formation on side shoots and their inferiority (Goptsiy 2018). The amaranth grains contain approximately 61.3-76.5% carbohydrates (mostly starch), 13.1-21.5% protein, 5.6-10.9% fat, 2.7-5% fiber, and 2.5-4.4% of ash (Skwaryło-Bednarz 2012). The protein content in amaranth grains averaged 16.2% in the control, 18.9% in the  $N_{200}P_{80}K_{160}$  fertilizer doses according to the results of our research.

In the study by Kozak et al. (2011), the protein content in amaranth seeds was 163 g kg<sup>-1</sup>. An increase in the nitrogen dose from 60 to 120 kg ha<sup>-1</sup> increased seed yield by 33%, reduced the fat content in grain, but increased the collection of protein fats with the harvest (Nasirpour-Tabrizi et al. 2020).

Gimplinger et al. (2007) investigated the grain quality of different genotypes of amaranth in the Pannonian region in Eastern Austria. The grain composition was within the following limits: crude protein 15.2-18.6%, crude fat 5.4-8.6%, crude fiber 3.5-4.2%, ash 2.7-3.2%, carbohydrates 66.7-72.7%. It changed according to the genotype. The density of amaranth crops did not affect either yield or grain quality.

Energy carbohydrates in amaranth grain are represented mainly by starch. The content of starch varied in the range of 64.8-67.4% (Table 1). The starch content was the highest in the object without fertilizers (67.4%). When applying  $N_{200}P_{80}K_{120}$  and  $N_{200}P_{80}K_{160}$ , the amount of starch in grain decreased to 64.8%. In other studies, it has also been found that mineral fertilization reduces the starch content in amaranth grains (Channab et al. 2023).

Thus, according to our research, the highest indicators of productivity of amaranth of the Kharkivskyi 1 variety were obtained at the doses of fertilization  $N_{200}P_{80}K_{160}$  and  $N_{200}P_{80}K_{120}$ : yield in the experiment – 4.88 and 4.84 t/ha, respectively, oil content – 8.0 and 7.9%, respectively, and the same protein content – 18.9%.

We calculated correlations between amaranth productivity indicators and nitrogen application doses (Table 2).

Table 2

| Indicator                            | Yield<br>(Mg ha <sup>-1</sup> ) | Protein<br>(%) | Starch<br>(%) | Bulk density<br>(g l <sup>-1</sup> ) |
|--------------------------------------|---------------------------------|----------------|---------------|--------------------------------------|
| Nitrogen dose (kg ha <sup>.1</sup> ) | 0.99                            | 0.99           | -0.98         | -0.98                                |
| Yield (Mg ha <sup>·1</sup> )         | Х                               | 0.99           | -0.98         | -0.98                                |
| Protein (%)                          |                                 | X              | -0.99         | -0.98                                |
| Starch (%)                           |                                 |                | Х             | 0.96                                 |

Correlation coefficients of yield indicators, grain quality and fertilizer rates

The correlation analysis showed that both the yield and protein content in amaranth grain significantly increase with the increase in nitrogen dose. However, the starch content and grain bulk density decrease with the increase in nitrogen dose. Simultaneously, a positive correlation was demonstrated between the protein content in the grain and the yield of amaranth. For starch content, this relationship was negative.

Based on polynomial regression analysis, it has been demonstrated that there is a direct relationship between nitrogen dosage and the grain yield as well as the protein content in amaranth seeds (Figure 1).

The 3D model of the slope of the regression plane visually shows that in the case of a proportional increase in the dose of nitrogen fertilizers, the proportion of starch in the amaranth grain decreases both for a small and for a large harvest.

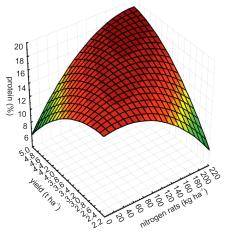


Fig. 1. The polynomial regression of the content of protein in the grain in accordance with the dose of nitrogen fertilization and yield of amaranth

Figure 2 shows that in the case of increase in the dose of nitrogen fertilizers, the content of starch in the amaranth grain decreases both for a small and for a large yield of grain.

A similar relationship was obtained for the bulk density of the grain (Figure 3). However, the slope of the plane indicates that increasing doses of

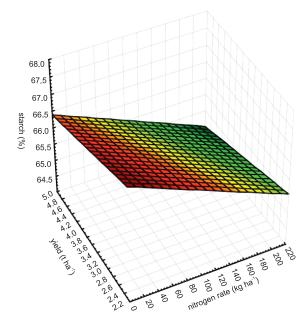


Fig. 2. The polynomial regression of the content of starch in the grain in accordance with the dose of nitrogen fertilization and yield of amaranth

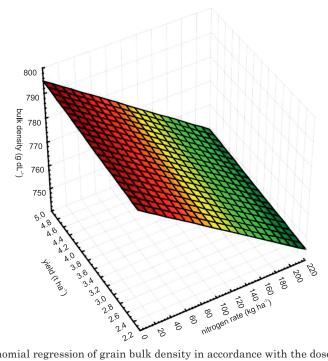


Fig. 3. The polynomial regression of grain bulk density in accordance with the dose of nitrogen fertilization and yield of amaranth

nitrogen have a stronger impact on reducing the value of this parameter than on the starch content in amaranth grain.

## CONCLUSIONS

Mineral fertilization, especially nitrogen fertilization, significantly affects the yield and quality characteristics of amaranth seeds. The yield of amaranth grown in the humid and warm climate of the Western Forest Steppe of Ukraine increased with the rising fertilizer doses, reaching 4.88 Mg ha<sup>-1</sup> at the highest dose ( $N_{200}P_{80}K_{160}$ ), which was twice as high as in the control variant. At the same time, high fertilizer doses negatively impacted the bulk density of the grain and starch content while increasing the protein content in amaranth seeds. In the absence of fertilization, amaranth seeds were the smallest, as evidenced by the bulk density of 790 g l<sup>-1</sup>. However, fertilization with high doses ( $N_{200}P_{80}K_{120}$  and  $N_{200}P_{80}K_{160}$ ) resulted in an increase in seed size and a decrease in bulk density to 755 g l<sup>-1</sup> and 750 g l<sup>-1</sup>. Under the influence of the highest fertilizer doses, the starch content in amaranth seeds by 3.4% compared to the control, while the protein content increased by 2.7%.

The results of the study prove that in order to obtain the highest productivity indicators of amaranth: grain yield within 4.88 t/ha, protein content – 18.9%, oil content – 8.0%, in conditions of sufficient moisture in the western forest-steppe of Ukraine, it is advisable to recommend growing the Kharkivskyi 1 variety for fertilizer standards  $N_{200}P_{s0}K_{160}$ .

## Author contributions

M.T., V.L., P.H. – conceptualization, M.T., V.L., P.H., O.A., I.R., O.L. – formal analysis, M.T., V.L., P.H., S.P. – methodology, M.T., V.L., O.L., I.D., I.R., P.H. – investigation, M.T., V.L., P.H., W.S., B.R. – visualization, M.T., V.L., P.H., W.S., B.R. – writing, original draft preparation, M.T., V.L., P.H., W.S., B.R. – writing, review and editing. All authors have read and agreed to the published version of the manuscript.

### **Conflicts of interest**

The authors declare no conflict of interest. The authors ensure that they have neither professional nor financial connections related to the manuscript sent to the Editorial Board.

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We dedicate the manuscript to our colleague Taras Bahaya, who died in the war in Ukraine.

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