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MACRONUTRIENT CONTENT AND RATIOS IN QUINOA SEEDS (*Chenopodium quinoa* WILLD.) DEPENDING ON SULPHUR FERTILISATION

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ABSTRACT

Background. The present study concerns the species quinoa (*Chenopodium quinoa* Willd). The aim of the study was to evaluate the effect of fertilizer sulfur on the formation of mineral composition and quantitative proportions of major nutrients in quinoa seeds.

Material and methods. Titicaca variety of quinoa was used for the study. A one-factor field experiment was established in four replications. The factor examined was the variation in sulphur fertilisation. This component was applied pre-sowing in spring in the form of Wigor S fertilizer in the amount of: 0 (control), 20, 40 and 60 kg $S \cdot ha^{-1}$.

Results. The study showed that in the years with significantly lower precipitation the contents of nitrogen, potassium, phosphorus, magnesium and calcium in seeds were generally lower. Their highest contents were found in the year with high precipitation. Sulphur fertilisation in respective years of vegetation generally significantly increased the nitrogen and calcium contents in quinoa seeds, to a lesser extent it was also true for magnesium. The content of phosphorus under these conditions increased in the quinoa seeds only in one year of the study, while the content of potassium under affected by sulphur application generally decreased. There was no significant effect of sulphur fertilisation on the values of ionic ratios ($K^+:Ca^{2+}$, $K^+:Mg^{2+}$, $(K^++Na^+):(Ca^{2+}+Mg^{2+})$), however, differences were observed in respective years of the study.

Conclusion. A correlation was found between the mineral composition of quinoa seeds and weather conditions during the vegetation period. Under conditions of high moisture of the soil high doses of sulphur increased the content of most of the elements studied in quinoa seeds. In drier years the effect of sulphur on macronutrient content was less.

Key words: *Chenopodium quinoa*, seeds, mineral composition, sulphur fertilisation

INTRODUCTION

Sulphur is an essential component in the life processes of plants as it has specific physiological functions in their metabolism and cannot be replaced by any other nutrient. Since the end of the last century in Poland, as a result of eco-friendly activities, as well as changes in the assortment of mineral fertilisers and reduction in the use of manure, deficiencies of this element in agrosystems have been observed (Grzebisz and Przygocka-Cyna, 2000; Motowicka-Terelak and Terelak, 2000). Disturbance of sulphur balance in Polish soils (Szulc, 2008) resulted in a growing interest in the problem of the influence of this element on the

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quantity and quality of crop yields. One of the important criteria for assessing yield quality is nutrient content and nutrient ratios (Filipek-Mazur and Tabak, 2016; Jakubus and Bakinowska, 2020). Many studies deal with C:N:P ratios (Chen *et al*., 2016; Zeng *et al*., 2017), much less with K^+:Mg^{2+}, K^+:Na^+, Ca^{2+}:P, Ca^{2+}:Mg^{2+}, K^+:(Ca^{2+}+Mg^{2+}) or (K^++Na^+):(Ca^{2+}+Mg^{2+}) ratios. According to Jakubus and Bakinowska (2020), their balanced ratios determine the nutritional value of crops for consumption and fodder purposes. It is also assumed that quantitative ratios of macronutrients can be a better indicator of their deficiency than their contents (Ostrowska and Porębska, 2017).

Relatively many sulphur fertilising effect studies have concerned oilseed rape, which is classified as a sulphur-loving plant (Wielebski *et al*., 2000, Lošák and Richter, 2003, Malhi *et al*., 2007), much fewer studies covered cereals and *Fabaceae* plants (Barczak, 2010), while only a few – the plants grown in a small area, however, showing many advantages which make them attractive raw materials for some industries. Such species include quinoa (*Chenopodium quinoa* Willd.) (Nowak *et al*., 2016). It is an extremely valuable plant native to South America, cultivated as early as the time of the Incas. It belongs to the pseudo-cereals and the family *Chenopodiaceae*. The seeds do not contain gluten and can, therefore, be consumed by people with celiac disease (Bhargava *et al*., 2006), while offering a high nutritional value due to a relatively high amount of valuable protein, fat, carbohydrates and ash, found in the right proportions (Meyhuay, 2014). Quinoa seeds contain significant amounts of minerals, especially phosphorus, potassium and magnesium, however, also calcium, sodium, iron (50% more than in cereals), copper, manganese and zinc (Ando *et al*., 2007; Jacobsen, 2015; Vilcacundo and Hernández-Ledesma, 2017). However, the greatest advantage of the species is its high protein quality (Cauda *et al*., 2013), with a high proportion of essential amino acids – lysine and tryptophan (Ruas *et al*., 1999). Quinoa is also a valuable source of vitamins: B (riboflavin and folic acid, thiamine), E (tocopherols) and vitamin C, and stands out due to its high amount of polyunsaturated fatty acids (omega-3 and omega-6) (Cauda *et al*., 2013). The above numerous qualities call for values research on the adaptation of the species for cultivation in Polish soils. This requires research on the fertilizer needs of quinoa, also in relation to sulphur, especially since, as indicated by the study by Gęsiński (2012), the seeds of this species show a low amount of methionine, an essential amino acid containing that element.

The aim of this study was to evaluate the influence of fertiliser sulphur on the mineral composition and quantitative proportions of the main nutrients in quinoa seeds.

MATERIAL AND METHODS

A single-factor field experiment was conducted in 2014–2016 at the Experimental Station for Variety Evaluation in Chrząstów (53°11′ N; 17°35′ E), the Nakło district of the Kujawsko-Pomorskie province. The experiment was set up in a split-plot system.

Research plots were located on soil of class IVa, silty loamy sand, on medium clay. The content of available forms in soil (in mg·kg^{-1}) for phosphorus was 68.17 (low abundance), for potassium – 157.7 (high) and for magnesium – 36.18 (medium). The soil had a slightly acid reaction (pH_{KCl} 6.1). Total dose of nitrogen in amount of 120 kg N·ha^{-1} was divided into two equal portions and applied in the form of ammonium nitrate at pre-sowing and post-sowing periods. Post-sowing nitrogen fertilizers were sown at 11 pairs of leaf stage. Phosphorus and potassium fertilizers were applied pre-sowing in a form of triple superphosphate (42 kg P·ha^{-1}) and 60% potassium salt (120 kg K·ha^{-1}). In the field experiment, no manure was applied and the soil was characterised by low sulphate sulphur content. The variation in sulphur fertilisation was studied. The sulphur was applied pre-sowing in spring in a form of fertilizer Wigor S in the amount of: 0 (control), 20, 40 and 60 kg S·ha^{-1} in four replications. Wigor S is a granulated fertilizer containing 90% of elementary sulphur. Bentonite in the fertiliser, due to its ability to swell in contact with water, causes fragmentation of sulphur, which in the soil with a participation of microorganisms is oxidised to the sulphate form ($S\text{-}SO_4^{2-}$), assimilable by plants. In this study, quinoa of the Titicaca variety was grown. Seeds were sown at 9 kg·ha^{-1} in the first decade of May at a row spacing of 40 cm and a sowing depth of 1–2 cm. The plants were harvested at the stage of full seed maturity after the drying of inflorescences.

The macronutrients were determined using the following methods:

– total nitrogen and phosphorus – with flow spectrophotometry,
– potassium and sodium – with flame emission spectrometry,
– calcium and magnesium – with atomic absorption spectrometry.

Based on the results, the quantitative ratios were calculated the most common in literature: K^+:Ca^{2+}, K^+:Mg^{2+}, and (K^++Na^+):(Ca^{2+}+Mg^{2+}), taking into account the gram equivalents of the elements studied. The results were processed using analysis of variance in a randomized block design using ANALWAR-5FR software. Calculations were performed for a significance level of $P < 0.05$.

RESULTS AND DISCUSSION

During the growing seasons of quinoa in 2014-2016, a large variation in weather conditions was observed (Table 1). The years 2014 and 2015 were demonstrated dry conditions with rainfall totals from May to October slightly exceeding 200 mm, while 2016 was a wet year, with rainfall totals during the growing season (464.9 mm) significantly higher than the value multi-year (330 mm) and twice as high as in the first two years of the study. These differences were particularly pronounced in June, July and August, months in which weather conditions determine the size and mineral composition of the quinoa yield. The average air temperature in all growing months, except August, was the lowest in 2015, as compared with the other years of the study.

It was shown that among the macronutrients studied, quinoa seeds recorded the highest content of nitrogen (average 17.24 $g \cdot kg^{-1}$) (Table 2) and potassium (17.14 $g \cdot kg^{-1}$) (Table 3). The average contents of phosphorus, magnesium and calcium were lower and, as expressed in $g \cdot kg^{-1}$, they were, amounted respectively: 3.84 (Table 4), 2.32) (Table 5) and 1.25 (Table 6). The lowest content was found for sodium (average 0.005 $g \cdot kg^{-1}$) (Table 7).

Table 1. Monthly total rainfall and mean daily, monthly temperature in the years of the study – during the growing period of *Chenopodium quinoa*

Month	Year							
	2014		2015		2016		Multi-year	
	rainfall mm	temp. ºC	rainfall mm	temp. ºC	rainfall mm	temp. ºC	rainfall mm	temp. ºC
May	59.7	12.8	22.2	11.9	56.1	14.9	47.0	14.1
June	31.2	15.6	40.4	15.2	85.4	18.0	78.1	17.0
July	46.3	21.3	57.4	18.5	121.8	18.9	71.4	19.2
August	58.7	17.4	18.2	21.5	71.3	17.4	57.1	18.6
September	23.6	14.5	53.3	14.1	4.7	15.6	46.0	13.8
October	28.9	9.7	20.9	7.0	125.6	7.1	31.0	8.8
∑ / ºC	248.4	15.2	212.4	14.7	464.9	15.3	330.0	15.3

Table 2. Content of total nitrogen (g·kg^{-1}) in *Chenopodium quinoa* seeds

Wigor kg S·ha^{-1}	Year			
	2014	2015	2016	2014–2016
0	12.83	16.70	18.80	16.11
20	16.43	17.70	19.03	17.72
40	16.23	17.77	19.50	17.83
60	14.97	16.33	20.63	17.31
Mean	15.12	17.13	19.49	17.24
$LSD_{0.05}$	1.99	0.85	0.92	1.28

The weather factors, which determine the yield and its chemical composition, influenced the accumulation of macronutrients and their ionic ratios. In the case of nitrogen, potassium, phosphorus, magnesium and calcium (Table 2–6), regardless of the sulphur dose, lower contents were obtained in quinoa seeds from 2014 and 2015 than 2016, which recorded significantly lower rainfall during the growing season. The macronutrient content in quinoa seeds appears to be shaped largely by soil moisture conditions, especially during flowering and seed-filling periods. The highest contents of those nutrients were found in 2016, which may have been contributed to by temperatures much higher than the multi-year averages in May, June and September, as well as relatively high rainfall throughout the growing season.

Many authors (Klikocka, 2004; Paszkiewicz-Jasińska, 2005; Barczak, 2010) draw attention to a greater importance of the weather factor in shaping the mineral composition of seeds than sulphur fertilisation. This is confirmed by the analysis of nitrogen content in quinoa seeds (Table 2). In the present studies much more differentiated contents of this element were reported in respective years than as a result of sulphur application. Nevertheless, the application of sulphur significantly shaped the nitrogen content in each year of the study. In 2014 and 2015, already a dose of 20 kg S·ha^{-1} significantly increased the nitrogen content, as compared with the control, by 28.1% and 6.0%, respectively. In 2016, a significant difference was found only under the influence of 60 kg S·ha^{-1} (by 9.7%). It can be considered that the effectiveness of sulphur in shaping nitrogen content under drought conditions, which occurred in 2014 and 2015, was higher than in the third year of the study, showing high humidity (precipitation about twice as high as in the first two years). Hydrothermal conditions probably influenced the activity of sulphur bacteria (*Thiobacillus*), affecting the intensity of microbial transformation of sulphur and also determining its availability to plants.

Table 3. Content of potassium (g·kg^{-1}) in *Chenopodium quinoa* seeds

Wigor S kg S·ha^{-1}	Year			
	2014	2015	2016	2014–2016
0	16.33	18.40	18.00	17.58
20	16.27	15.87	18.27	16.80
40	16.90	17.13	17.77	17.27
60	14.70	15.97	20.07	16.91
Mean	16.05	16.84	18.53	17.14
$LSD_{0.05}$	1.35	1.68	1.91	ni

ni – non significant

Based on the literature (Barczak, 2010; Kozera *et al*., 2017; Jakubus and Bakinowska, 2020), it should be assumed that the application of sulphur, as a consequence of its effect on the concentration of ions in the soil solution and on the degree of saturation of the soil sorption complex with them, changed the conditions of plant nutrition and differentiated the accumulation of respective elements in quinoa seeds. Noteworthy is the high content of potassium compared to the literature data (Grzebisz, 2004), is noteworthy, which is probably due to the high abundance of this component in the soil and the relatively high dose of potassium fertilizer.

Many authors have noted a positive effect of sulphur application on nitrogen content, and thus on protein biosynthesis in crop seeds (Inal *et al*., 2003;

Podleśna, 2009; Barczak, 2010). Kopcewicz and Lewak (2005) explain this dependence by the presence of sulphur in ferrodoxin and nitrogenase, the enzymes playing a key role in these processes. Their activity is determined by the spatial arrangement formed by these enzymes thanks to sulphur bridges, formed by joining disulfide groups of sulphur amino acids, such as methionine and cysteine.

Studies in respective years showed a significant but differentiated effect of sulphur application on potassium content in quinoa seeds (Table 3). In 2014, seeds of plants fertilised with 60 kg S·ha^{-1} showed a significant reduction in the content of that nutrient, as compared with the seeds from the objects fertilised with lower doses of sulphur. In 2015, the highest potassium content was found in seeds of the plants not fertilised with sulphur, and in 2016 – fertilised with the highest dose (60 kg S·ha^{-1}).

Only in two years (2014 and 2016) a significant effect of sulphur fertilisation on phosphorus content in quinoa seeds was found (Table 4). In 2014, each of the doses resulted in a significant increase in that content, as compared with the control; the differences (%) were, respectively, 39.2, 44.4 and 38.4. The other years of the study showed no such changes. Nonetheless, the yearly averages of phosphorus content in seeds of sulphur-fertilised plants, irrespective of the dose, recorded significantly higher values than in seeds of unfertilised plants.

Table 4. Content of phosphorus (g·kg^{-1}) in *Chenopodium quinoa* seeds

Wigor S kg S·ha^{-1}	Year			
	2014	2015	2016	2014–2016
0	2.68	3.94	4.37	3.67
20	3.73	3.61	4.15	3.83
40	3.87	3.89	3.85	3.87
60	3.71	3.89	4.37	3.99
Mean	3.50	3.83	4.18	3.84
$LSD_{0.05}$	0.83	ni	0.36	ni

ni – non significant

As for phosphorus, only two years (2014 and 2015) showed a significant effect of sulphur on magnesium content in quinoa seeds (Table 5). In 2014, all the fertilizer doses increased the magnesium content, as compared with the control, but only for 20 and 40 kg S·ha^{-1} the differences were significant (%): respectively; 12.6 and 15.8. In 2015, there was a significant difference in the magnesium content of seeds from plants fertilised with 20 and 60 kg S·ha^{-1}; an increase of 10.6%).

Table 5. Content of magnesium (g·kg^{-1}) in *Chenopodium quinoa* seeds

Wigor S kg S·ha^{-1}	Year			
	2014	2015	2016	2014–2016
0	1.83	2.45	2.57	2.29
20	2.06	2.26	2.65	2.32
40	2.12	2.30	2.53	2.32
60	1.92	2.50	2.58	2.34
Mean	1.98	2.38	2.58	2.32
$LSD_{0.05}$	0.21	0.22	ni	ni

ni – non significant

Similarly to phosphorus and magnesium contents, calcium content in quinoa seeds was significantly differentiated by sulphur application only in two, out of three, study years (2014 and 2016) (Table 6). In 2014, a significant increase in calcium content in seeds, as compared with the control, was achieved by the application of 40 kg S ha^{-1}, while in 2016 – by the application of 60 kg S-ha^{-1}. The respective differences were 15.8 and 24.2 %.

Although for the entire study period (2014–2016) there was no significant effect of sulphur application on the formation of phosphorus, magnesium and calcium contents in quinoa seeds, yet the averaged values for individual doses were always significantly higher than for the control (Table 4–6).

Table 6. Content of calcium (g·kg^{-1}) in *Chenopodium quinoa* seeds

Wigor S kg S·ha^{-1}	Year			
	2014	2015	2016	2014-2016
0	0.90	1.31	1.45	1.22
20	1.03	1.18	1.52	1.24
40	1.14	1.25	1.45	1.28
60	0.92	1.05	1.78	1.25
Mean	1.00	1.20	1.55	1.25
$LSD_{0.05}$	0.23	ni	0.31	ni

ni – non significant

Although in the respective years of the study a significant effect of sulphur on the sodium content of quinoa seeds was found, no directional changes under the influence of the factor were shown, and the average values for respective years and fertilisation levels over the entire study cycle were similar and, generally, amounted to 0.005 g·kg^{-1} (Table 7).

Table 7. Content of sodium (g·kg^{-1}) in *Chenopodium quinoa* seeds

Wigor S kg S·ha^{-1}	Year			
	2014	2015	2016	2014-2016
0	0.006	0.006	0.004	0.005
20	0.004	0.006	0.005	0.005
40	0.004	0.006	0.006	0.005
60	0.005	0.004	0.006	0.005
Mean	0.005	0.006	0.005	0.005
$LSD_{0.05}$	0.001	0.001	0.001	ni

ni – non significant

It could be expected that the consequence of changes in the content of individual macronutrients in quinoa seeds as a result of the application of sulphur would be an effect on the ionic balance of the plant, which would be measured by changes in the values of the equivalent ratios of one-positive to two-positive cations (K^+:Ca^{2+}, K^+:Mg^{2+}, (K^++Na^+):(Ca^{2+}+Mg^{2+})) (Table 8–10). The analysis of their values can provide the basis for inferring the nutritional status of plants (Jakubus and Bakinowska, 2020). The balance between mono- and dipositive cations, according to Kopcewicz and Lewak (2005), has a great influence on maintaining the biocolloids of the cell cytoplasm at the appropriate degree of hydration, and by participating in buffer systems, it determines its pH and osmotic potential. Ion balance can have a big impact on the health humans and animals (Chen *et al*., 2016).

The values of K^+: Mg^{2+} (average – 2.27:1, Table 9) and (K^++Na^+):(Ca^{2+}+Mg^{2+}) (1.72:1, Table 10) obtained in the study of quinoa seeds are within the limits found in the literature, which are respectively: (2–6):1 and (1.6–2.2):1 (Jakubus and Bakinowska, 2020). Slightly higher values compared to the literature data (Chen *et al*., 2016, Filipek-Mazur and Tabak, 2016) were obtained for K^+:Ca^{2+} (average -7.17:1, Table 8), which should be explained by the high abundance of potassium in the soil and the application of a relatively high dose of potassium fertilizer. Comparing the values of the studied indicators with the data of Antonkiewicz (2007), it can be concluded that only the potassium: calcium ratio deviated from the optimal one for animal nutrition.

Table 8. The ionic ratios (K^+:Ca^{2+})

Wigor S kg S·ha^{-1}	Year			
	2014	2015	2016	2014-2016
0	8.03	7.21	6.46	7.23
20	8.08	7.08	6.20	7.12
40	7.66	7.01	6.33	7.00
60	8.25	7.77	5.91	7.31
Mean	8.01	7.27	6.22	7.17
$LSD_{0.05}$	ni	ni	ni	ni

ni – non significant

Table 9. The ionic ratios ($K^+:Mg^{2+}$)

Wigor S kg S·ha^{-1}	Year			
	2014	2015	2016	2014-2016
0	2.48	2.31	2.16	2.32
20	2.43	2.16	2.12	2.24
40	2.46	2.29	2.16	2.30
60	2.36	1.95	2.39	2.23
Mean	2.43	2.18	2.21	2.27
$LSD_{0.05}$	ni	0.33	ni	ni

ni – non significant

Table 10. The ionic ratios ($(K^+ + Na^+):(Ca^{2+} + Mg^{2+})$)

Wigor S kg S·ha^{-1}	Year			
	2014	2015	2016	2014-2016
0	1.89	1.75	1.61	1.75
20	1.87	1.65	1.58	1.70
40	1.86	1.72	1.61	1.73
60	1.83	1.56	1.70	1.70
Mean	1.86	1.67	1.62	1.72
$LSD_{0.05}$	ni	ni	ni	ni

ni – non significant

However, the studies generally did not show a significant effect of sulphur fertilisation on the values of ionic ratios, while differences were observed between the respective years of the study. The highest values of the ratios were recorded for the seeds of plants grown in 2014, with a low precipitation during the quinoa vegetation period (in four out of six months, including the key months for quinoa vegetation – June, July and September, the precipitation was significantly lower than in the multi-year period). Only in 2015 a significant effect of sulphur application on the values of $K^+:Mg^{2+}$ ratio was noted (Table 9). After the application of 60 kg S·ha^{-1}, the seeds of plants showed significantly lower values of this ratio than those of the plants not fertilised or fertilised with 40 kg S·ha^{-1}.

CONCLUSIONS

1. The relationship between the mineral composition of quinoa seeds and weather conditions during the growing season was demonstrated. In 2014 and 2015, with a significantly lower rainfall than 2016, the contents of nitrogen, potassium, phosphorus, magnesium and calcium in the seeds were generally lower. The highest contents of these nutrients were found in 2016, which could have been due to much higher than multi-year average temperature in May and June, as well as relatively high rainfall throughout the growing season.
2. Sulfur fertilization in individual growing years generally significantly increased the nitrogen and calcium contents of quinoa seeds, to a lesser extent it was also true for magnesium. The phosphorus content under these conditions increased in the seeds of plants grown only in 2014, while potassium content generally decreased due to sulphur application.
3. Under the conditions of high soil moisture experienced in 2016, high doses of sulphur (40 and 60 kg S·ha^{-1}) increased the content of most of the elements studied in quinoa seeds.
4. There was no significant effect of sulphur fertilisation on the values of ionic ratios ($K^+:Ca^{2+}$, $K^+:Mg^{2+}$, $(K^+ + Na^+):(Ca^{2+} + Mg^{2+})$), however differences were observed between the respective years of the study. The highest values of the ratios were recorded for the seeds of the plants grown in 2014, with low precipitation during the vegetation period of quinoa.

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ZAWARTOŚĆ I STOSUNKI ILOŚCIOWE MAKROSKŁADNIKÓW W NASIONACH KOMOSY RYŻOWEJ (*Chenopodium quinoa* WILLD.) W ZALEŻNOŚCI OD NAWOŻENIA SIARKĄ

Streszczenie

Opracowanie dotyczy gatunku komosa ryżowa (*Chenopodium quinoa* Willd). Celem badań była ocena wpływu siarki nawozowej na kształtowanie się składu mineralnego i proporcji ilościowych głównych składników pokarmowych w nasionach komosy ryżowej. Do badań wykorzystano odmianę Titicaca komosy ryżowej. Założono jednoczynnikowe doświadczenie polowe w czterech powtórzeniach. Badanym czynnikiem było zróżnicowanie nawożenia siarką. Składnik ten stosowano przedsiewnie wiosną w postaci nawozu Wigor S w ilości: 0 (obiekt kontrolny), 20, 40 i 60 kg $S \cdot ha^{-1}$. Na podstawie przeprowadzonych badań stwierdzono, że w latach, które charakteryzowały się znacznie niższymi opadami, zawartość azotu, potasu, fosforu, magnezu i wapnia w nasionach była na ogół niższa. Najwyższą zawartość tych składników stwierdzono w roku z wysokimi opadami. Nawożenie siarką w poszczególnych latach wegetacji na ogół istotnie zwiększało zawartość azotu i wapnia w nasionach komosy ryżowej, w mniejszym stopniu dotyczyło to również magnezu. Zawartość fosforu w tych warunkach wzrastała w nasionach komosy ryżowej tylko w jednym roku badań, a zawartość potasu pod wpływem aplikacji siarki na ogół obniżała się. Nie wykazano istotnego wpływu nawożenia siarką na wartości stosunków jonowych ($K^+:Ca^{2+}$, $K^+:Mg^{2+}$, $(K^++Na^+):(Ca^{2+}$ i Mg^{2+})), natomiast zaobserwowano różnice w poszczególnych latach badań. Wykazano powiązanie składu mineralnego nasion komosy ryżowej z warunkami pogodowymi w okresie wegetacji. W warunkach dużego uwilgotnienia siedliska wysokie dawki siarki powodowały wzrost zawartości większości badanych pierwiastków w nasionach komosy.

Słowa kluczowe: *Chenopodium quinoa*, nasiona, nawożenie siarką, skład mineralny