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YIELD OF FRESH MASS AND NITRATE REDUCTASE ACTIVITY IN LETTUCE FERTILIZED BY DIFFERENT NITROGEN FORMS

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ABSTRACT. Fertilization of lettuce by molybdenum and an increase of the pH level in substrates caused an increase of lettuce yield. The ureal form of nitrogen ensured a higher yield of lettuce than the remaining nitrogen forms. With the increase of pH of substrates increased the activity of nitrate reductase and the content of nitrates in lettuce fertilized by molybdenum decreased. The highest activity of nitrate reductase was found in lettuce fertilized by $Ca(NO_3)_2$, and the greatest amount of nitrates was found in lettuce fertilized by $Ca(NO_3)_2$ and NH_4NO_3.

Key words: lettuce, forms of nitrogen, nitrates, nitrate reductase

Introduction

Many genetic and agrotechnical factors have an influence on the content of nitrates in plants. Their recognition may contribute to the diminution of nitrates content and thereby improve the quality of yields. The improvement of yield quality is actually a priority in plant production.

The objective of this work was to investigate the effect of molybdenum, differentiated pH of the substrate and nitrogen forms on the activity of nitrate reductase in lettuce leaves.

A particular role in nitrogen metabolism is played by molybdenum. **Tyksiński** (1992) found that all substrates used in horticulture require fertilization by molybdenum. He determined the lower and the upper critical level for that microcomponent in lettuce growing. He also determined the dosage range of Mo: from 0.5 to 90.0 mg $Mo \cdot dm^{-3}$, where he obtained unified lettuce yields without any symptoms of deficiency or excess of this component.

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One of the most important factors which decides about the nitrate content in the plant is the abundance of nitrate nitrogen in the soil or substrate (**Lisiewska** and **Kmiecik** 1991, **Kozik** 1994).

Material and methods

The experiment was carried out in 2004 in a greenhouse on the area of the Experimental Station Marcelin (Poznań).

Seeds of lettuce, 'Atena' cultivar, were sown on March 8 by the point method in boxes of 10 dm^3 capacity filled with raised peat from Lithuania, limed with $CaCO_3$ in the amount of 5 $g \cdot dm^{-3}$ and enriched with macroelements: N – 150, P – 100 and K – 150 $mg \cdot dm^{-3}$. The boxes were kept in a heated greenhouse.

The substrate for lettuce growing consisted of light medium sand from the arable layer of the field of the Experimental Station Marcelin mixed (on March 22) in 1:1 proportion with high peat from Lithuania. The substrate was paced into 6 dm^{-3} containers after previous addition of $CaCO_3$ (3 or 8 $g \cdot dm^{-3}$) and nutritive components.

Four days after liming, the following macro- and micro-components were applied ($mg \cdot dm^{-3}$):

Macrocomponents	**Microcomponents**	
200 mg N	2 mg Mo (Mo_1 only)	20 mg Zn
150 mg P	2 mg B	40 mg Fe
300 mg K	10 mg Cu	
150 mg Mg	20 mg Mn	

The following variants were applied in the experiment:

- 2 levels of molybdenum:
  - level I (Mo_0) – without addition of Mo,
  - level II (Mo_1) – with addition of 2 mg $Mo \cdot dm^{-3}$,
- 2 levels of liming:
  - level I – (I pH) – 3 g $CaCO_3 \cdot dm^{-3}$ of substrate added,
  - level II (II pH) – 8 g $CaCO_3 \cdot dm^{-3}$ of substrate added,
- 4 nitrogen forms: $(NH_4)_2SO_4$, $Ca(NO_3)_2$, NH_4NO_3 and $CO(NH_2)_2$.

The experiment consisted of 16 combinations and each combination had 4 replications. One replication included one container with 4 lettuce plants.

Lettuce was planted on April 15 into containers in an unheated greenhouse. Containers with lettuce were irrigated with tap water up to 75% of the total water capacity determined by Wahnschaff's cylinders.

On April 28, the plants were top dressed with nitrogen in the amount of 50 mg N per 1 dm^3 of substrate by adequate salts according to the experimental scheme.

Lettuce was cut out on May 10. Plant material was weighed, broken up and dried at 55°C. At the same time, from each combination, 1g of leaves was randomly sampled in order to determine the activity of nitrate reductase.

After harvest of plants, containers with substrates were irrigated to constant weight and then, mean samples of substrate were taken for chemical analyses.

In the substrate, the content of macro- and micro-components was determined by universal method. The results will be presented in a separate publication.

The pH in H_2O of substrates was determined by potentiometric method.

In fresh, frozen plant material, nitrate reductase activity was determined by **Wray's** and **Filner's** method (1970). This analysis was done in the Department of Plant Physiology of Agricultural University of Poznań.

In dried homogenized plant material, the content of macro- and micro-components was determined. Results will be presented in a separate publication.

Nitrates content was also determined by colorimetric method with phenolodisulphonic acid (**Kamińska** et **al.** 1972).

Results and discussion

Growth and development of lettuce

The dominating effect on the appearance of plants was exerted by the pH of substrate.

Mo_0: at level I of pH, plants were significantly lower with a distinct chlorosis particularly visible at $Ca(NO_3)_2$.

Plants fertilized by NH_4NO_3, next to chlorosis showed necrotic spots on older leaves. Plants fertilized by $CO(NH_2)_2$ were bigger with pale-green leaves.

At level II of pH, plants were healthy, without symptoms of chlorosis, but their color had a lighter green shade. Plants were smaller than at level pH II (on Mo_1).

Mo_1: at level I of pH, plants were equalized, distinctly bigger than on Mo_0, with pale-green leaves of lettuce fertilized by $Ca(NO_3)_2$.

At level II of pH, plants were correctly developed with normal green color and high yields.

Lettuce yielding

Both the molybdenum level, pH level and nitrogen form had a distinct effect on lettuce yield. Significantly higher yield was obtained from plants fertilized by molybdenum (Mo_1) as compared with the non-fertilized ones (Mo_0).

Korzeniowska and **Gembarzewski** (1996) also found that deficiency of adequate amount of molybdenum was the main reason of lower yield of maize grown in lower pH soil.

In the discussed experiment, significant differences were also obtained in lettuce yield under the influence of different pH levels. At level II of pH, a higher yield of fresh plant mass was obtained than at pH level I (Table 1).

The nitrogen forms applied in the experiment caused also changes in the yield of lettuce. Yield obtained after fertilization by $CO(NH_2)_2$ was significantly higher than after the application of the remaining nitrogen forms. **Kowalska** (1996) found that nitrogen form had no effect on the yield size of tomato.

The effect of the factors studied in the experiment on the pH of substrate and on the content of molybdenum is shown in Table 2.

Among the nitrogen compounds used in the experiment, $(NH_4)_2SO_4$ decreased the pH of substrates to the highest degree. The content of molybdenum in the substrates on

Table 1

Effect of molybdenum level, pH level and nitrogen form on the yield of lettuce fresh mass (g·container $^{-1}$)

Wpływ poziomu molibdenu, poziomu pH i formy azotu na plon świeżej masy sałaty (g·pojemnik $^{-1}$)

Mo level (A) Poziom molibdenu (A)		pH level (B) Poziom pH (B)		Nitrogen form (C) Forma azotu (C)	
Mo_0	342.2	I	334.5	$(NH_4)_2SO_4$	395.6
Mo_1	455.9	II	463.6	$Ca(NO_3)_2$	373.8
				NH_4NO_3	386.6
				$CO(NH_2)_2$	440.3

Mo level (A) Poziom molibdenu (A)	pH level (B) Poziom pH (B)	Nitrogen form (C) Forma azotu (C)			
		$(NH_4)_2SO_4$	$Ca(NO_3)_2$	NH_4NO_3	$CO(NH_2)_2$
Mo_0	I	207.5	230.0	98.8	362.5
	II	490.0	398.8	478.8	471.3
Mo_1	I	397.5	425.0	481.3	473.8
	II	487.5	441.3	487.5	453.8

$LSD_{0.05}$ for A = 15.8
$LSD_{0.05}$ for B = 15.8
$LSD_{0.05}$ for C = 22.3
$LSD_{0.05}$ for A × B × C = 44.5

$NIR_{0,05}$ dla A = 15,8
$NIR_{0,05}$ dla B = 15,8
$NIR_{0,05}$ dla C = 22,3
$NIR_{0,05}$ dla A × B × C = 44,5

Table 2

pH of substrates after the completion of experiment and the content of molybdenum in the substrates (mg Mo·dm^{-3})

pH podłoży po zakończeniu doświadczenia oraz zawartość w nich molibdenu (mg Mo·dm^{-3})

pH level Poziom pH	Nitrogen compound Związek azotu	Molybdenum level Poziom molibdenu			
		Mo_0		Mo_1	
		pH w H_2O	Mo	pH w H_2O	Mo
I	$(NH_4)_2SO_4$	4.37	0.16	4.40	0.67
	$Ca(NO_3)_2$	4.96	0.16	5.00	0.61
	NH_4NO_3	4.49	0.14	4.63	0.58
	$CO(NH_2)_2$	4.56	0.10	4.59	0.59
II	$(NH_4)_2SO_4$	5.96	0.13	5.95	0.42
	$Ca(NO_3)_2$	6.15	0.13	6.28	0.48
	NH_4NO_3	6.08	0.10	6.41	0.43
	$CO(NH_2)_2$	6.14	0.10	6.29	0.49

Mo_0 in the interval of 0.10-0.16 mg·dm^{-3} showed very distinctly a negative effect on the growth and development as well as on the yielding of lettuce. The content of Mo in the substrate in the amount of a least 0.3 mg·dm^{-3} should be sufficient for lettuce (**Tyksiński** 1992).

Activity of nitrate reductase

The results are illustrated in Figure 1.

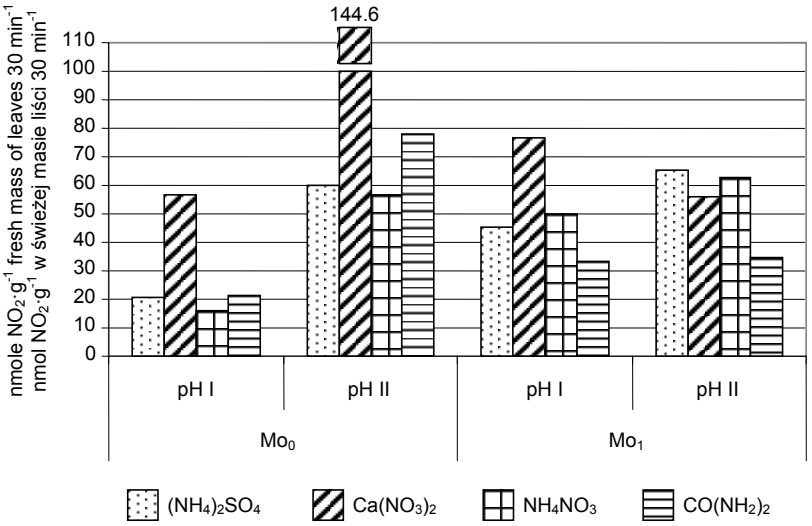

Fig. 1. Activity of nitrate reductase in lettuce leaves after the completion of experiment
Ryc. 1. Aktywność reduktazy azotanowej w liściach sałaty po zakończeniu doświadczenia

Factors studied in the experiment had an effect on the activity of nitrate reductase. Comparison of this activity on both levels of molybdenum revealed that on Mo_1 (in the corresponding combinations) it usually was higher than on Mo_0.

There was a distinct effect of pH level on the activity of nitrate reductase on Mo_0. At level II of pH, it was significantly higher than at level pH I. This dependence was not found on Mo_1.

Also the applied nitrogen compounds differentiated the activity of reductase which was the highest after the application of $Ca(NO_3)_2$. This regularity confirms the study results of **Rożek** et **al.** (1994).

Content of nitrates in lettuce

Nitrates content is shown in Table 3.

The content of nitrates in the leaves of lettuce fertilized by molybdenum was lower than in the lettuce not fertilized by this component, particularly at level I of pH.

Table 3

Content of nitrates in lettuce leaves
Zawartość azotanów w liściach sałaty

pH level Poziom pH	Nitrogen compound Związek azotu	Molybdenum level Poziom molibdenu	
		Mo_0	Mo_1
		% N – NO_3	% N – NO_3
I	$(NH_4)_2SO_4$	1.40	0.46
	$Ca(NO_3)_2$	1.57	1.16
	NH_4NO_3	2.10	0.97
	$CO(NH_2)_2$	1.60	0.87
II	$(NH_4)_2SO_4$	1.14	1.04
	$Ca(NO_3)_2$	1.26	1.12
	NH_4NO_3	1.28	1.09
	$CO(NH_2)_2$	1.08	1.20

Comparison of nitrates content (in the corresponding combinations) between the I and the II levels of pH indicated that it was lower at level II of pH, but only on Mo_0.

Independently of the fertilization with molybdenum and regardless of pH level, the greatest amount of nitrates was found in lettuce fertilized by the nitrate N form, i. e. by Ca $(NO_3)_2$ and NH_4NO_3.

The smallest amount of nitrates was found after the application of $(NH_4)_2SO_4$. The same regularity was found by **Kozik** and **Gleń** (1995). Many authors found a decrease of nitrates accumulation in plants after the application of reduced nitrogen forms (**Rożek** et **al.** 1995, **Sady** and **Rożek** 1995).

Conclusions

1. Factors studied in the experiment had a significant effect on lettuce yields. Molybdenum in the dose of 2 mg $Mo \cdot dm^{-3}$ of substrate increased lettuce yield in comparison with plants grown without molybdenum addition. The second pH level of substrates (5.95-6.41) caused a higher yield of lettuce than the first pH level (4.37-5.00).

The ureal form of nitrogen ensured a higher yield than the remaining studied forms. The yields of lettuce fertilized by $(NH_4)_2SO_4$, Ca $(NO_3)_2$ and NH_4NO_3 did not differ.

2. Fertilization by molybdenum in the dose of 2 mg $Mo \cdot dm^{-3}$ of substrate in relation to the combination without molybdenum:

– increased the activity of nitrate reductase in lettuce, but only at level I of pH,

– significantly decreased the content of nitrates in lettuce at level I of pH, at the second pH level, it also decreased the content of nitrates but in a lesser degree.

3. The pH level and effect on the activity of nitrate reductase and on the content of nitrates:

– in lettuce not fertilized by molybdenum at pH II level, the activity of reductase was 2.6-3.7 times higher than at pH I level; at level II of pH, the amount of nitrates was smaller than at level I of pH,

– in lettuce fertilized by molybdenum, no major effect of the pH level of substrates on the activity of nitrate reductase was found.

At the level of pH II, the amount of nitrates was greater than at level I of pH (except for the combination with $Ca(NO_3)_2$).

4. The highest activity of nitrate reductase was found in lettuce fertilized by $Ca(NO_3)_2$, and the greatest amount of nitrates was found in lettuce fertilized by $Ca(NO_3)_2$ and NH_4NO_3.

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PLON ŚWIEŻEJ MASY I AKTYWNOŚĆ REDUKTAZY AZOTANOWEJ W SAŁACIE NAWOŻONEJ RÓŻNYMI FORMAMI AZOTU

S t r e s z c z e n i e

Wiosną 2004 roku w szklarni Katedry Nawożenia Roślin Ogrodniczych AR w Poznaniu przeprowadzono doświadczenie z sałatą odm. 'Atena', którego celem było zbadanie wpływu molibdenu, zróżnicowanego pH podłoża i czterech form azotu na aktywność reduktazy azotanowej oraz zawartość azotanów w liściach.

Nawożenie molibdenem w dawce 2 mg $Mo \cdot dm^{-3}$ podłoża, wzrost poziomu pH oraz mocznikowa forma azotu spowodowały zwiększenie plonu sałaty.

Nawożenie molibdenem zwiększało aktywność reduktazy azotanowej oraz zmniejszało zawartość azotanów w liściach sałaty.

W sałacie nienawożonej molibdenem na II poziomie pH aktywność reduktazy była większa, a azotanów było mniej niż na I poziomie pH.

W sałacie nawożonej molibdenem nie stwierdzono wyraźnego wpływu poziomu pH podłoży na aktywność reduktazy azotanowej.

Na II poziomie pH więcej było azotanów niż na I poziomie pH, z wyjątkiem $Ca(NO_3)_2$.