Environmental and Experimental Biology (2024) 22: 1–9
http://doi.org/10.22364/eeb.22.01

Original Paper

Environmental and Experimental Biology

ISSN 2255-9582

UNIVERSITY OF LATVIA

Foliar application of commercial humic substances for possible increase of nutrient status and yield of oilseed rape

Anita Osvalde[1]*, Andis Karlsons[1], Gunta Cekstere[1], Laura Āboliņa[1], Solveiga Malecka[2]

[1]Institute of Biology, University of Latvia, Rīga LV–1004, Latvia
[2]Stende Research Center, Institute of Agricultural Resources and Economics, Lībagu Rural Territory, Talsu Municipality, Dižstende LV–3258, Latvia

*Corresponding author, E-mail: anita.osvalde@lu.lv

Abstract

Inconsistent results on the effects of humic substances (HS) on yield and nutrient status of important food crops, including oilseeds, confirm the need for further research on different HS products to match their use to actual field conditions. The aim of this study was to evaluate the effect of foliar application of commercially produced peat- and vermicompost-derived HS preparations on nutrient status and yield of spring oilseed rape (*Brassica napus*). The field experiment was carried out in Stende State Cereals Breeding Institute, Latvia, during the vegetation season of 2012, using the spring oilseed rape cultivar 'Perfect'. Although foliar sprays of HS were applied during the critical stages of crop development from rapid growth to flowering, they were ineffective in improving the supply of the deficient nutrients (N, K, B, Zn, Cu) in leaves. Moreover, both tested HS products caused a decrease in Cu and B content in the seeds, which resulted in a negative trend in oilseed rape seed yield. Therefore, the conducted experiment demonstrated that foliar application of both HS preparations was ineffective to promote nutrient status and did not contribute to spring oilseed rape yield.

Key words: *Brassica napus*, humic substances, leaf analysis, peat extract, seed analysis, thousand seed mass, vermicompost extract.
Abbreviations: HS, humic substances.

Introduction

To implement the European Green Deal (European Commission 2019), the main challenges for plant scientists and agronomists are to improve crop yields in a more efficient and environmentally friendly way. In this aspect, the optimisation of plant mineral nutrition is a particularly important factor in agricultural practice. To achieve and maintain high yields, oilseed rape cultivation requires both adequate and intensive soil and foliar fertilisation (Szczepaniak et al. 2015; Jarecki 2021). Therefore, recent focus has centered on organic inputs, nonconventional means of plant nutrition, biostimulants, including humic substances (HS) (Hemati at al. 2022).

Numerous studies have shown that the application of HS has potentially significant effects on crop agronomic performance and soil quality parameters. HS, derived from decomposed organic materials such as plant, animal, and microbial residues, have biostimulant properties (Canellas et al. 2015; de Hita et al. 2020; Nardi et al. 2021), they improve the nutrient status of plants (Ahmad et al. 2018; Khan et al. 2018), resistance to various abiotic stresses (García et al. 2012; Akladious, Mohamed 2018; Haider et al. 2021) and increase the yield and quality of crops (Dinçsoy, Sönmez 2019; Amiri et al. 2020; Elshamly, Nassar 2023). It is well-documented that HS promote plant growth and physiological processes via both soil and foliar application (de Moura et al. 2023). Due to the overall high cost of commercial HS formulations, foliar application (relatively small amount required) is widely recommended to farmers as an economically sound method to reduce the use of agrochemicals, thus promoting more environmentally friendly agriculture (Lyons, Genc 2016; Monda et al. 2021).

However, some studies have reported limited or no positive responses, and even negative effects, following HS application, particularly on yield traits and quality (Hartz, Bottoms 2010; Osvalde et al. 2012; Leventoglu, Erdal 2014; Osvalde et al. 2016). Considering that the effect of HS on plant growth depends on the source diversity, concentration, dosage and molecular weight of the humus fractions, their effects may differ even for the same plant species (Klavins et al. 2021; Nardi et al. 2021).

Most commercially produced HS are derived from sedimentary materials – lignite, leonardite and coal – due to their extensive deposits (Fatima et al. 2021). Peat and compost from green waste and manure, including vermicompost, are also important sources of HS (Ampong et al. 2022). In Latvia, HS-based products for use in agriculture are extracted from local raw materials, peat and vermicompost, with potassium alkali solution (Bremanis et al. 2013). Latvia has large peat resources (Karofeld et al. 2017), mainly used for horticultural substrate production (Paoli et al. 2022). Vermicompost production in Latvia has significantly developed in recent decades, and it is primarily used as organic fertiliser (Vronskis et al. 2016). The use of peat and vermicompost in producing high-value HS products and their application in agriculture certainly holds innovative potential (Krumins et al. 2021).

Brassica napus L., commonly known as oilseed rape or rapeseed, is the third largest source of vegetable oil in the world (FAO 2021). It is not only an important food source, but also the main feedstock for biodiesel production in Europe (Chmielewska et al. 2021) and a valuable raw material for the polymer industry (de Moura et al. 2023). Oilseed rape has also been a widely cultivated crop in Latvia in recent decades (Fridrihsone et al. 2018). In 2021, the land used for oilseed rape was 11.3% of the total area of agricultural crops in Latvia. In Latvia, similarly as throughout Europe, rapeseed is grown mainly as a winter crop, however, relatively large areas of arable land are also allocated to spring rapeseed, around 15 thousand ha (Agriculture in Latvia 2022).

Inconsistent results on the effects of HS treatments on yield and nutrient status of important food crops, including oilseeds, confirm the need for further research on different HS products, application rates and methods to match the use of a particular HS formulation to real field conditions and plant species.

Therefore, the aim of this study was to evaluate the effect of foliar application of commercially produced peat and vermicompost-derived HS preparations on nutrient status (N, P, K, Ca, Mg, S, Fe, Mn, Zn, Cu, Mo, B) and yield of spring oilseed rape in a field trial. In the research hypothesis, it was assumed that foliar application of HS would have a beneficial effect on spring rape seed yield by stimulating nutrient uptake.

Materials and methods

Experimental design

The field experiment was carried out in the vegetation season of 2012 at the State Stende Cereals Breeding Institute (57°11'22.2"N, 22°33'41.9"E), Latvia. Two commercial HS preparations extracted from peat (K45) and vermicompost (B45) with cavitation technology using KOH at 45 °C, were tested on spring oilseed rape (*Brassica napus*) cv. 'Perfect'. The preparations were produced by scientific research and production company "Intellectual Resources" (Latvia). The chemical characteristics of peat and vermicompost extracts (Bremanis et al. 2013) are presented in Table 1. Foliar treatment with the HS preparation was carried out three times using the manufacturer's specified application rate (1.5 L ha^{-1}, water rate 250 L ha^{-1}) on June 5, July 3 and July 25, 2012. The first HS foliar spray was applied at the 4 to 6 leaf stage, the second at the beginning of flowering and the third at the end of flowering. The experiment was set up with four replications in a randomised block design. The area of each experimental plot was 20 m^2 (2 × 10 m).

Soil conditions and agronomic practices

The soil texture of the experimental field was sod podzolic light loam. Soil agrochemical characteristics, determined from a composite soil sample taken in April before the experiment, are given in Table 2. The soil was characterised as corresponding to rape production in terms of pHKCl (6.48), organic matter (2.60%) content and level of plant available K, P, Ca, and Mg. The detected concentrations of N, S and B in the soil were determined to be low.

As basic fertiliser, 850 kg ha^{-1} of complex fertiliser NPK 16–16–16 (N 140, P 60, K 110 kg ha^{-1}) was incorporated during pre-sowing cultivation. The pre-crop was pea (*Pisum sativum*).

The rape seeds were sown on April 30, 2012, using a sowing rate of 3 kg per ha. All agrotechnical treatments were performed according to the methodology of the State Stende Cereals Breeding Institute. Weeds and pests were controlled throughout the growing season by applying plant protection products – insecticide Proteus and herbicides Butisan and Lontrel.

Oilseed rape was harvested at full seed maturity (September 10, 2012) using a field harvester (Wintersteiger Delta), with seed yield determined for each plot. Seed yield was calculated as t per ha. The mass of 1000 seeds (g) was determined using a Contador Pfeuffer grain counter, weighing each replicate separately on a Scout Pro (200 g) electronic scale with an accuracy of 0.01 g.

Table 1. Chemical characteristics of peat and vermicompost-derived HS extracts (Bremanis et al. 2013). Commercial HS preparation from peat and vermicompost provided by scientific research and production company "Intellectual Resources" (Latvia)

Parameter	Concentration (mg L^{-1})	
HS	960.3	792.3
N	26.29	46.38
P	2.07	4.27
K	595.0	471.0
Ca	11.0	23.0
Mg	0.30	3.40
S	39.51	46.14
Fe	3.90	5.10
Mn	0.02	0.11
Zn	0.36	0.18
Cu	0.02	0.04
B	2.90	4.10
pH	10.35	10.80
Organic matter (%)	0.21	0.17

Table 2. Nutrient concentration, soil pH, electrical conductivity (EC) and content of organic matter in the air-dry soil of the study site, at a depth of 0 to 20 cm. Nutrient sufficiency range in soil (1M HCl extraction) for oilseed rape according to Nollendorfs and Čekstere (2012)

Parameter	Plant-available soil nutrient concentration (mg L^{-1}) in 1M HCl extraction	
	Measured	Sufficiency range
N	30	100 – 160
P	621	140 – 300
K	220	140 – 280
Ca	3360	1600 – 7000
Mg	605	250 – 1100
S	8	40 – 100
Fe	1275	700 – 1500
Mn	185	70 – 180
Zn	12.5	3 – 10
Cu	5.1	4 – 10
B	0.1	0.6 – 1.6
pH_{KCl}	6.4	6.4 – 7.4
EC_{H2O} (mS cm^{-1})	0.33	n.a.
Organic matter (%)	2.6	n.a.

Plant sampling and determination of nutrients

To determine the nutrient status of oilseed rape, leaf material was collected twice: on June 29 (three weeks after the first leaf treatment on July 3, 2012) and on July 18 (two weeks after the second leaf treatment on July 3, 2012). The content of nutrients in the final product (seed) was also estimated.

For leaf samples, approximately 25 of the most recently fully developed leaves were randomly collected from the central part of each plot. For rape seed analysis, samples were taken from the harvested seed material.

In order to determine macro- and micronutrients, the plant material was oven-dried at 60 °C, ground, dry-ashed in concentrated HNO_3 vapours, and the ash was dissolved in 3% HCl solution. Wet digestion in H_2SO_4 and HNO_3 was used for N and S detection, respectively. Atomic absorption spectroscopy, using an acetylene-air flame atomiser (Perkin Elmer AAnalyst 700) was used for the measurement of K, Ca, Mg, Fe, Mn, Zn, and Cu according to the manufacturer's instructions. Levels of P, Mo, N, and B were determined by colourimetry: P by ammonium molybdate in an acid-reduced medium, Mo by thiocyanate in a reduced acid medium, B by hinalizarine in a sulphuric acid medium, N by modified Kjeldal method using Nessler's reagent in an alkaline medium, and S through the turbidimetric method by adding $BaCl_2$, using a spectrophotometer Jenway 6300, as described previously (Cekstere et al. 2020). All the values were expressed as mass % and mg kg^{-1} on a dry matter basis for each macronutrient and micronutrient evaluated, respectively.

Statistical analyses

Nutrient and yield results were analysed with descriptive statistics using the R programming language. A one-way ANOVA with post hoc Tukey HSD test and a Kruskal Wallis test with post hoc Wilcoxon test were conducted to determine statistically significant differences between the effect of HS treatment on nutrient levels, seed yield and seed mass, respectively. Principal component analysis was performed on seed nutrient content and yield data. Significant differences were indicated by $p < 0.05$.

Results

In the vegetation season of 2012, the weather conditions at the trial site were typical for the western part of Latvia and generally suitable for oilseed rape cultivation. Monthly precipitation, temperature and long-term average values of these indicators in the region are shown in Fig. 1. June and August 2012 were rainy and the amount of precipitation in these months was on average 135% compared to long-term observations. Although the weather conditions for plant growth were generally optimal, with sufficient moisture and warmth, the above normal amount of precipitation in August made harvesting conditions difficult.

The effect of foliar application of peat and vermicompost-derived HS preparation on the concentrations of nutrients in rapeseed leaves at the end of June after the first treatment is presented in Table 3. Overall, foliar fertilisation did not alter the macro- and micronutrient content of leaves, as no significant differences were found between treatments and control. The data obtained showed that the content of most nutrients corresponded to the sufficiency range reported for rapeseed leaves (Plank, Tucker 2000; Orlovius 2000; Nollendorfs, Čekstere 2012). The only deviations were found for N, K and B, whose concentrations in leaves were 1.5 to 2 times lower than recommended.

Nutrient concentrations in the rapeseed leaves after the second foliar spray are presented in Table 4. In general, there was no promoting effect of HS application on macronutrient uptake in the rapeseed leaves. Regarding micronutrients, the HS-dependent stimulation effect on

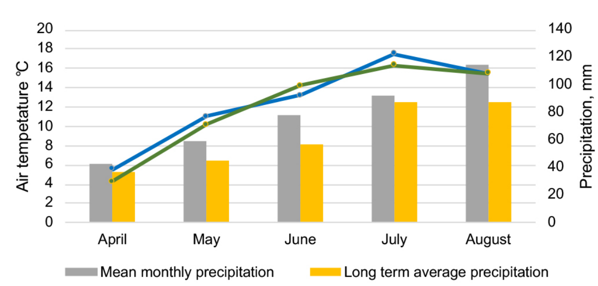

Fig. 1. Weather conditions in the growing season of 2012, including the region's long-term average climatic indicators (Data from Stende Hydrometeorological station, Latvia).

Table 3. Effect of foliar application of peat- (K45) and vermicompost-derived (B45) HS preparation on nutrient concentrations in oilseed rape leaves after the first spray at the 4 to 6 leaf stage (first sampling time on June 29, 2012). Nutrient sufficiency range for oilseed rape leaves (Plank, Tucker 2000; Orlovius 2000; Nollendorfs, Čekstere 2012). There were no significant differences between the treatments *($p > 0.05$)*

Nutrient	Concentration in dried tissue, mean ± SE			Nutrient sufficiency range
	Control	K45	B45	
	Macronutrients (mass %, dry matter)			
N	3.07 ± 0.09 a	2.80 ± 0.21 a	2.90 ± 0.06 a	4.0 – 6.0
P	0.44 ± 0.01 a	0.44 ± 0.02 a	0.45 ± 0.02 a	0.35 – 0.70
K	1.47 ± 0.05 a	1.48 ± 0.10 a	1.51 ± 0.10 a	2.5 – 5.0
Ca	2.46 ± 0.17 a	2.38 ± 0.08 a	2.65 ± 0.18 a	1.0 – 2.5
Mg	0.32 ± 0.01 a	0.31 ± 0.03 a	0.34 ± 0.03 a	0.2 – 0.4
S	0.79 ± 0.10 a	0.74 ± 0.07 a	0.83 ± 0.09 a	0.5 – 0.8
	Micronutrients (mg kg^{-1}, dry matter)			
Fe	75.33 ± 1.33 a	72.00 ± 6.43 a	77.33 ± 1.33 a	50 – 150
Mn	46.67 ± 9.48 a	44.67 ± 11.79 a	43.33 ± 2.91 a	30 – 150
Zn	27.33 ± 1.76 a	26.00 ± 1.15 a	25.33 ± 0.67 a	25 – 70
Cu	6.13 ± 0.71 a	5.53 ± 0.18 a	5.87 ± 0.27 a	5 – 15
Mo	1.49 ± 0.31 a	1.47 ± 0.33 a	1.28 ± 0.22 a	0.4 – 1.2
B	14.00 ± 0.67 a	14.33 ± 0.33 a	14.00 ± 0.58 a	25 – 60

Mo uptake was statistically significant for both treatments and on Fe for the treatment containing vermicompost extract (B45). These changes in the level of micronutrients did not change the nutrient status of rapeseed, as Fe and Mo values found in the control were already in the sufficient or high range, respectively.

Seasonal trends of leaf nutrients revealed a decrease in the concentrations of N, K, Zn and Cu in the second half of July, not falling within the ranges of sufficient values reported in the literature. In contrast, during the vegetation season, a significant increase in B content in the leaves was found for all treatments, generally reaching the level required for rapeseed.

The data showed that foliar application of HS obtained from vermicompost (B45) caused a small but statistically significant increase in P and S content in seeds (Table 5). On the other hand, a significantly lower content of micronutrients Cu and B was found in seeds collected from the experimental plots where K45 or B45 were used.

Regarding seed yield, the results indicated that foliar spraying of HS derived from peat and vermicompost did not significantly affect rape seed yield (Table 6). The average

Table 4. Effect of foliar application of peat- (K45) and vermicompost-derived (B45) HS preparation on nutrient concentrations in oilseed rape leaves after second spray at the beginning of flowering (second sampling time on July 18, 2012). Nutrient sufficiency range for oilseed rape leaves (Plank, Tucker 2000; Orlovius 2000; Nollendorfs, Čekstere 2012). Means with different letters in a row were significantly different between treatments ($p < 0.05$)

Nutrient	Concentration in dried tissue, mean ± SE			Nutrient sufficiency range
	Control	K45	B45	
	Macronutrients (mass %, dry matter)			
N	2.03 ± 0.03 a	2.00 ± 0.06 a	1.98 ± 0.04 a	4.0 – 6.0
P	0.48 ± 0.02 a	0.43 ± 0.01 a	0.47 ± 0.02 a	0.35 – 0.7
K	1.21 ± 0.12 a	1.27 ± 0.06 a	1.21 ± 0.02 a	2.5 – 5.0
Ca	5.47 ± 0.34 a	4.94 ± 0.33 a	5.19 ± 0.21 a	1.0 – 2.5
Mg	0.51 ± 0.05 a	0.45 ± 0.03 a	0.44 ± 0.03 a	0.2 – 0.4
S	1.38 ± 0.06 a	0.92 ± 0.04 b	1.13± 0.07 b	0.5 – 0.8
	Micronutrients (mg kg^{-1}, dry matter)			
Fe	64.67 ± 5.93 a	68.00 ± 8.72 ab	91.33 ± 12.88 b	50 – 150
Mn	59.33 ± 20.41 a	52.00 ± 14.00 a	62.00 ± 14.05 a	30 – 150
Zn	20.13 ± 1.99 a	19.07 ± 0.79 a	21.33 ± 0.67 a	25 – 70
Cu	3.13 ± 0.44 a	3.33 ± 0.29 a	3.07 ± 0.13 a	5 – 15
Mo	2.53 ± 0.58 a	4.05 ± 0.61 b	3.53 ± 0.86 ab	0.4 – 1.2
B	23.67 ± 1.20 a	24.33 ± 1.45 a	24.00 ± 1.15 a	25 – 60

Table 5. Effect of foliar application of peat- (K45) and vermicompost-derived (B45) HS preparation on nutrient concentrations in oilseed rape seeds. Foliar treatments with peat and vermicompost extracts were carried out three times. Means with different letters in a row were significantly different between treatments ($p < 0.05$)

Nutrient	Concentration in seeds, mean ± SE		
	Control	K45	B45
Macronutrients (mass %, dry matter)			
N	2.52 ± 0.07 a	2.43 ± 0.09 a	2.45 ± 0.13 a
P	0.58 ± 0.03 a	0.56 ± 0.03 a	0.66 ± 0.01 b
K	0.89 ± 0.02 a	0.84 ± 0.05 a	0.87 ± 0.03 a
Ca	0.61 ± 0.01 a	0.60 ± 0.01 a	0.60 ± 0.01 a
Mg	0.41 ± 0.03 a	0.39 ± 0.01 a	0.37 ± 0.01 a
S	0.38 ± 0.02 a	0.39 ± 0.01 a	0.44 ± 0.00 b
Micronutrients (mg kg–1, dry matter)			
Fe	70.67 ± 8.74 a	79.33 ± 15.72 a	76.67 ± 2.67 a
Mn	46.00 ± 4.16 a	44.00 ± 6.00 a	48.67 ± 4.67 a
Zn	26.67 ± 0.67 a	23.33 ± 2.40 a	26.61 ± 0.67 a
Cu	4.80 ± 1.33 a	2.87 ± 0.41 b	2.33 ± 0.74 b
Mo	0.71 ± 0.071 a	0.79 ± 0.06 a	0.74 ± 0.02 a
B	7.33 ± 0.17 a	6.83 ± 0.17 b	6.67 ± 0.33 b

seed yield across both HS treatments and control ranged from 2.23 to 2.41 t ha^{-1}. Significant decrease in thousand seed mass was observed for the B45 treatment, when seed yield was 92.5% of that of the control.

Principal component analysis was performed to evaluate the relationships between treatments, accumulation of nutrients in seeds and seed yield. The first three components

Table 6. Effect of foliar application of peat- (K45) and vermicompost-derived (B45) HS preparation on oilseed rape seed yield and thousand seed mass. Foliar treatments with peat and vermicompost extracts were carried out three times (at the 4 to 6 leaf stage, at the beginning of flowering and at the end of flowering). There were no significant differences between treatments for yield data ($p > 0.05$). Means with different letters in a column were significantly different between treatments ($p < 0.05$)

Treatment	Seed yield (t ha^{-1})	Compared to the control (%)	Thousand seed mass (g)
Control	2.41 ± 0.06	100	3.27 a
Peat extract K 45	2.33 ± 0.10	96.7	3.40 a
Vermicompost extract B 45	2.23 ± 0.05	92.5	3.07 b
$LSD_{0.05}$	0.224	n.a.	0.166

with eigenvalues over 1.0 explained 72.18% of the total variance (Fig. 2, Table 7). PC1 had the strongest positive correlations with S, Fe, Mn, and Zn but negative with Cu and seed yield, suggesting that it reflects the micronutrient status of seed material. PC2 had negative values for almost all variables; the strongest correlations were with K, Ca, Mg, and Zn. This mainly measured the macronutrient status. PC3 had the strongest correlations with N, P, and Mg. Nutrients Mo and B were not well represented in the PCA, suggesting they did not significantly contribute to the variance of the analysis.

Principal component analysis confirmed that the

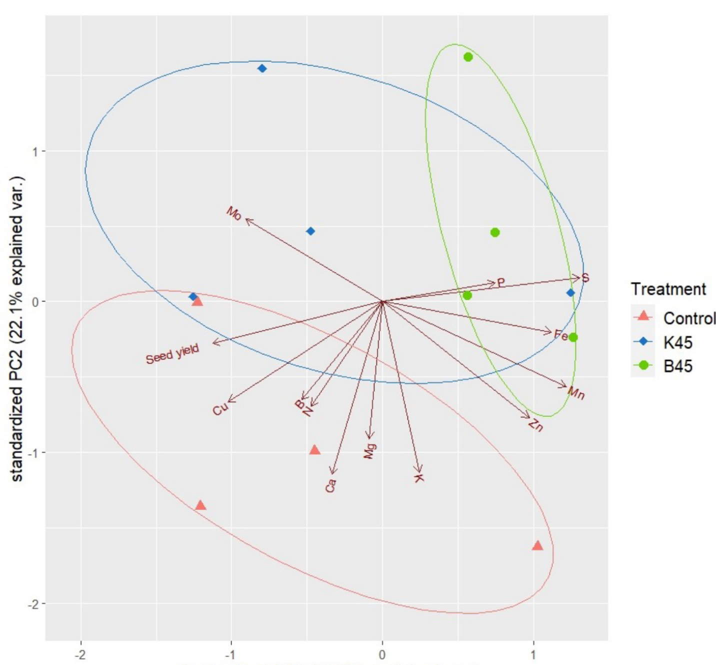

Fig. 2. Principal component analysis of the data set on oilseed rape seed yield and nutrient content as affected by foliar treatment of HS preparation derived from peat (K45) and vermicompost (B45).

Table 7. Eigenvector values of principal component analysis of peat samples describing correlations with the first three components

Variable	PC1	PC2	PC3
N	−0.151	−0.280	0.376
P	0.238	0.050	−0.376
K	0.078	−0.457	0.168
Ca	−0.107	−0.462	−0.281
Mg	−0.029	−0.368	−0.539
S	0.418	0.065	0.081
Fe	0.358	−0.082	0.247
Mn	0.389	−0.228	0.158
Zn	0.311	−0.312	0.233
Cu	−0.327	−0.269	−0.073
Mo	−0.290	0.220	0.269
B	−0.171	−0.262	0.225
Seed yield	−0.360	−0.111	0.211
Explained variance	35.0%	22.1%	15.1%

chemical composition of the seeds of the control plants was generally different from the other treatments. When grouped by treatments, the most important factors in the principal component analysis for the division of individual samples of control variant in the ordination space were Cu, Ca, K, Zn and B levels in seeds, as well as seed yield. Treatment B45 correlated positively with PC1 and PC2, with the lowest variance between samples. The control group had the least common components with the other two treatments.

Discussion

Effect of foliar HS treatments on nutrient uptake in rapeseed leaves

This study was aimed to evaluate the effect of foliar application of commercially-produced peat and vermicompost-derived HS preparation on nutrient status of oilseed rape under conventional growth conditions. When evaluating the difference in nutrient supply, no significant effect was found between the treatments at the beginning of rapeseed growth after the first spray. The analysis of the leaf material showed that macronutrient status, except for N and K, can be described as optimal to high for all treatments. Nitrogen deficiency, especially in the early stage of oilseed rape development, can inhibit vegetative growth and reproductive development, resulting in reduced seed yield (Bouchet et al. 2016; Lin et al. 2020). Potassium performs a variety of physiological activities in plants, such as effective enzyme activation, photosynthesis, resistance to several pests, diseases and environmental stresses (Fageria 2009). Studies show that K availability considerably promotes the accumulation of N, and a significant interaction between these nutrients has been found for oilseed rape (Li et al. 2023). Therefore, optimal K concentration in young plants is necessary not only to achieve a higher seed yield but also for sustainable N management (Szczepaniak 2015). Our study showed that K and N shortage, which was revealed already at the rosette stage, persisted up to flowering. Unfortunately, the application of HS preparations did not result in increased N and K accumulation in rapeseed leaves after the second spray either. For high rapeseed yields, there is a high demand for N and K particularly during the most intensive growth stages from shooting to flowering (Orlovius 2000; Grzebisz et al. 2019). Thus, N and K deficiency in rapeseed leaves during this period indicated a general under-fertilixation and foliar application of HS did not increase nutrient use efficiency.

Regarding micronutrients, at the early stage of crop development, only B deficiency was detected in rapeseed leaves as a mineral nutrition imbalance. According to the results of the current study, foliar application of peat and vermicompost-derived HS preparation did not stimulate the absorption of B in this important timing for the crop. Seasonal trends in micronutrient content revealed an increase in B accumulation in rapeseed leaves during vegetative growth, with all variants reaching levels close to sufficient in the second half of July. It should be noted that Brassica crops have a relatively high B requirement and an adequate supply of B is critical, especially during rapid growth in spring and the flowering phase (Fageria 2009; Ma et al. 2015). Studies on the effect of foliar B application on oilseed rape yield have shown that the increase in seed yield was mainly related to the effect of this micronutrient on seed set in pods and number of pods per plant (Jankowski et al. 2016; Jarecki 2021). Therefore, B fertilisation is recommended in rapeseed nutrient management programs (Wysocki et al. 2007). The beneficial effect of foliar fertilisation with B in combination with biostimulators on rape seed yield was also demonstrated (Sikorska et al. 2020). This suggests that the application of HS preparations in combination with foliar nutrition could be promising and should be investigated further.

The average values of micronutrient concentrations from the control plots revealed a deficiency of Zn and Cu for the second leaf sampling time, which coincided with flowering. However, the two-fold application of HS preparations had no significant effect on the Zn and Cu supply in rapeseed leaves. Zn and Cu levels in leaves had decreased below the widely accepted critical deficiency levels (25 and 4.0 mg kg^{-1}, respectively) for oilseed rape (Plank, Tucker 2000; Fageria 2009). Although oilseed rape is not regarded as a crop that is highly sensitive to low Cu and Zn supply (Orlovius 2000; Rahman, Schoenau 2021; Rahman, Schoenau 2022), insufficient Cu supply limits pollen formation and seed set, while Zn deficiency reduces protein formation (Fageria 2009), which can significantly reduce seed yield and quality. Thus, the application of HS was ineffective in preventing micronutrient (Zn, Cu) deficiencies.

Effect of foliar HS application on seed yield and accumulation of nutrients in seeds

It has been reported that foliar application of HS enhances nutrient uptake and increases the yield and quality of various oilseed crops including rapeseed (Barekati et al. 2019; Amiri et al. 2020; Hemati et al. 2022; de Moura et al. 2023). Our research does not confirm this, as the effect of applied HS preparations on spring rape seed yield was insignificant. Moreover, a negative trend should be noted, as after triple foliar spraying of K45 and B45, the yield of spring rapeseed decreased by 3.3 and 7.5%, respectively. While a significant response of the thousand seed mass to the applied peat-derived preparation K45 was not detected, the use of vermicompost-derived B45 resulted in a decrease in the thousand seed mass of the studied spring rapeseed variety. As the seed yield is largely determined by the thousand seed mass (Szczepanek et al. 2016), the lowest yield was specific to the B45 treatments. This was somewhat surprising as higher efficiency and crop responses have been reported for HS preparations derived from composts and vermicomposts (Adhikary 2012; Balmori et al. 2019; Klavins et al. 2021; Hemati et al. 2022) rather than from peat (Ayuso et al. 1996; Rose et al. 2014; Jindo et al. 2020).

Since the accumulation of nutrients in the seeds can significantly affect the seed yield, the decrease in the content of Cu and B caused by the application of HS preparations could be one of the explanations for the negative results obtained. This was also evidenced by the results of principal component analysis, which showed that seed yield was related to Cu, Ca, K, Zn, B and N levels in seeds. The analysis confirmed that the chemical composition of the control treatment generally differed from others, by having the highest level of these nutrients and seed yield results. The most pronounced differences in seed chemical composition from the control were found for individual B45 samples, and the seed yield was also only 92.5% of that of the control. Our results agree with the findings that foliar fertilisation with biostimulants alone did not significantly affect the rapeseed yield (Sikorska et al. 2020).

The seed yield in the control variant exceeded 2.4 tons per ha and was similar to the average yield for spring rapeseed in Latvia (2.5 t ha^{-1}) reported from 2008 to 2014 (Fridrihsone et al. 2018). This indirectly indicated that a generally suitable spring rapeseed cultivation technology was provided for testing the effectiveness of HS preparations in the experimental field. In recent years, the spring rapeseed yield in Latvia has even decreased: in 2021 it was only 2.0 t ha^{-1} on average (Agriculture of Latvia 2022). There are various reasons for this, including climate change and restrictions on pesticide use; however, the improvement of rapeseed cultivation technology, including innovations in fertilisation, remains an urgent issue. In this regard, one should be aware that the practical benefits of using products containing HS depend on various factors and are highly unpredictable due to inconsistent effects. According to this study, we can conclude that the three-fold foliar application of commercial peat and vermicompost derived HS at typical rates (1.5 L ha^{-1}) was ineffective in promoting yield and nutrient uptake of spring oilseed rape in the open field.

Conclusions

The research results revealed that foliar application of commercial peat and vermicompost-derived HS preparations was ineffective in promoting nutrient status and did not contribute to the spring oilseed rape yield in the field conditions. Although foliar sprays of HS were applied during the critical stages of crop development from rapid growth in spring to flowering, they were ineffective in improving the supply of the deficient nutrients – N, K, B, Zn, Cu – in rapeseed leaves. Moreover, both tested HSs caused a decrease in Cu and B content in the seeds, which resulted in a negative trend in the obtained oilseed rape seed yield. When assessing the application potential of HS products for agricultural production, it is essential to consider additional costs such as labour, fuel, and materials, which may contribute to an increase in overall production expenses. The field experiment results indicate that relying on foliar application of commercial HS to enhance mineral nutrition and productivity of rapeseed is illusory. However, these outcomes are associated with specific growing conditions. Therefore, further experimental studies are needed to elucidate potentially more effective application rates and regimes considering different sources of HS origin, crop types, soil and climate conditions.

Acknowledgements

The research was supported by the European Regional Development Fund project No. 2DP/2.1.1.1.0/10/APIA/VIAA/082 "Study of organic product extracts and their effects in crop production".

References

Adhikary S. 2012. Vermicompost, the story of organic gold: a review. *Agric. Sci.* 3: 905–917.

Agriculture of Latvia. Collection of Statistics. 2022. Central Statistical Bureau of Latvia, Riga, Latvia, 85 p.

Ahmad T., Khan R., Nawaz Khattak T. 2018. Effect of humic acid and fulvic acid based liquid and foliar fertilizers on the yield of wheat crop. *J. Plant Nutr.* 41: 2438–2445.

Akladious S.A., Mohamed H.I. 2018. Ameliorative effects of calcium nitrate and humic acid on the growth, yield component and biochemical attribute of pepper (*Capsicum annuum*) plants grown under salt stress. *Sci. Hortic.* 236: 244–250.

Amiri M., Rad A.H.S., Valadabadi A., Sayfzadeh S., Zakerin H. 2020. Response of rapeseed fatty acid composition to foliar application of humic acid under different plant densities. *Plant Soil Environ.* 66: 303–308.

Ampong K., Thilakaranthna M.S., Gorim L.Y. 2022. Understanding the role of humic acids on crop performance and soil health. *Front. Agron.* 4: 848621.

Ayuso M., Hernández T., Garcia C., Pascual J.A. 1996. Stimulation of barley growth and nutrient absorption by humic substances originating from various organic materials. *Bioresour. Technol.* 57: 251–257.

Balmori M.D., Domínguez C.Y.A., Carreras C.R., Rebatos S.M., Farías L.B.P., Izquierdo F.G., Berbara R.L.L., Calderín García A. 2019. Foliar application of humic liquid extract from vermicompost improves garlic (*Allium sativum* L.) production and fruit quality. *Int. J. Recycl. Org. Waste Agric.* 8: S103–S112.

Barekati F., Hervan E.M., Rad A.H.S., Noor Mohamadi G. 2019. Effect of sowing date and humic acid foliar application on yield and yield components of canola cultivars. *Tarim Bilimleri Dergisi* 25: 70–78.

Bouchet A.S., Laperche A., Bissuel-Belaygue C., Snowdon R., Nesi N., Stahl A. 2016. Nitrogen use efficiency in rapeseed. A review. *Agron. Sustain. Dev.* 36: 1–20.

Bremanis G., Klavinsh M., Purmalis O., Ziemelis R., Malecka S. 2013. Peat humic substances and earthworm biohumus extracts for agricultural applications. *Proc. Latvian Acad. Sci. B* 67: 236–241.

Canellas L.P., Olivares F.L., Aguiar N.O., Jones D.L., Nebbioso A., Mazzei P., Piccolo A. 2015. Humic and fulvic acids as biostimulants in horticulture. *Sci. Hortic.* 196: 15–27.

Cekstere G., Osvalde A., Elferts D., Rose C., Lucas F., Vollenweider P. 2020. Salt accumulation and effects within foliage of *Tilia × vulgaris* trees from the street greenery of Riga, Latvia. *Sci. Total Environ.* 747: 140921.

Chmielewska A., Kozłowska M., Rachwał D., Wnukowski P., Amarowicz R., Nebesny E., Rosicka-Kaczmarek J. 2021. Canola/rapeseed protein–nutritional value, functionality and food application: a review. *Crit. Rev. Food Sci. Nutr.* 61: 3836–3856.

de Hita D., Fuentes M., Fernández V., Zamarreño A.M., Olaetxea M., García-Mina J.M. 2020. Discriminating the short-term action of root and foliar application of humic acids on plant growth: emerging role of jasmonic acid. *Front. Plant Sci.* 11: 493.

de Moura O.V.T., Berbara R.L.L., de Oliveira Torchia D.F., Oliveira Da Silva H.F., van Tol de Castro T.A., Tavares O.C.H., Fernandes Rodrigues N., Zonta E., Azevedo Santos L., Calderín García A. 2023. Humic foliar application as sustainable technology for improving the growth, yield, and abiotic stress protection of agricultural crops. A review. *J. Saudi Soc. Agri. Sci.* 22: 493–513.

Dinçsoy M., Sönmez F. 2019. The effect of potassium and humic acid applications on yield and nutrient contents of wheat (*Triticum aestivum* L. var. Delfii) with same soil properties. *J. Plant Nutr.* 42: 2757–2772.

Elshamly A.M.S., Nassar S.M.A. 2023. Stimulating growth, root quality, and yield of carrots cultivated under full and limited irrigation levels by humic and potassium applications. *Sci. Rep.* 13: 14260.

European Commission. 2019. Delivering the European Green Deal. https://commission.europa.eu/strategy-and-policy/priorities-2019-2024/european-green-deal/delivering-european-green-deal_en.

Fageria N.K. 2009. *The Use of Nutrients in Crop Plants.* CRC Press: Boca Raton, FL, USA, 448 p.

FAO. 2021. Food and Agriculture Organization of the United Nations: FAOSTAT Statistical Database. http://www.fao.org/faostat/en/#home.

Fatima N., Jamal A., Huang Z., Liaquat R., Ahmad B., Haider R., Ali M.I., Shoukat T., ALOthman Z.A., Ouladsmane M., Ali T., Ali S., Akhtar N., Sillanpää M. 2021. Extraction and chemical characterization of humic acid from nitric acid treated lignite and bituminous coal samples. *Sustainability* 13: 8969.

Fridrihsone A., Romagnoli F., Cabulis U. 2018. Life cycle inventory for winter and spring rapeseed production in Northern Europe. *J. Clean. Prod.* 177: 79–88.

García A.C., Santos L.A., Izquierdo F.G., Sperandio M.V.L., Castro R.N., Berbara R.L.L. 2012. Vermicompost humic acids as an ecological pathway to protect rice plant against oxidative stress. *Ecol. Eng.* 47: 203–208.

Grzebisz W., Przygocka-Cyna K., Szczepaniak W., Zawieja A. 2019. Impact of winter oilseed rape nutritional status during vegetative growth on yield. *Plant Soil Environ.* 65: 490–496.

Haider F.U., Liqun C., Coulter J.A., Cheema S.A., Wu J., Zhang R., Wenjun M., Farooq M. 2021. Cadmium toxicity in plants: Impacts and remediation strategies. *Ecotoxicol. Environ. Safety* 211: 111887.

Hartz T.K., Bottoms T.G. 2010. Humic substances generally ineffective in improving vegetable crop nutrient uptake or productivity. *HortScience* 45: 906–910.

Hemati A., Alikhani H.A., Babaei M., Ajdanian L., Lajayer B.A., van Hullebusch E.D. 2022. Effects of foliar application of humic acid extracts and indole acetic acid on important growth indices of canola (*Brassica napus* L.). *Sci. Rep.* 12: 20033.

Jankowski K.J., Sokólski M., Dubis B., Krzebietke S., Zarczynski P., Hulanicki P., Hulanicki P.S. 2016. Yield and quality of winter oilseed rape (*Brassica napus* L.) seeds in response to foliar application of boron. *Agr. Food Sci.* 25: 164–176.

Jarecki W. 2021. The reaction of winter oilseed rape to different foliar fertilization with macro- and micronutrients. *Agriculture* 11: 515.

Jindo K., Olivares F.L., Malcher D.J.P., Sánchez-Monedero M.A., Kempenaar C., Canellas L.P. 2020. From lab to field: role of humic substances under open-field and greenhouse conditions as biostimulant and biocontrol agent. *Front. Plant Sci.* 11: 426.

Karofeld E., Jarašius L., Priede A., Sendžikaite J. 2017. On the after-use and restoration of abandoned extracted peatlands in the Baltic countries: extracted peatlands in Baltics. *Restor. Ecol.* 25: 293–300.

Khan R.U., Khan M.Z., Khan A., Saba S., Hussain F., Jan I.U. 2018. Effect of humic acid on growth and crop nutrient status of wheat on two different soils. *J. Plant Nutr.* 41: 453–460.

Klavins M., Grandovska S., Obuka V., Ievinsh G. 2021. Comparative study of biostimulant properties of industrially and experimentally produced humic substances. *Agronomy* 11: 1250.

Krumins J., Klavins M., Tumilovich A., Irtiseva K., Mironovs V., Lapkovskis V., Ozolins J., Shishkin A. 2021. Potential of Baltic region peat in high added-value products and environmentally friendly applications - a review. *Preprints* 2021100244.

Leventoglu H., Erdal I. 2014. Effect of high humic substance levels on growth and nutrient concentration of corn under calcareous conditions. *J. Plant Nutr.* 37: 2074–2084.

Li J., Zhou Y., Gu H., Lu Z., Cong R., Li X., Ren T., Lu J. 2023. Synergistic effect of nitrogen and potassium on seed yield and nitrogen use efficiency in winter oilseed rape (*Brassica napus*

L.). *Eur. J. Agron.* 148: 126875.

Lin Y., Watts D.B., Torbert H.A., Howe J.A. 2020. Influence of nitrogen rate on winter canola production in the southeastern United States. *Agron. J.* 112: 2978–2987.

Lyons G., Genc Y. 2016. Commercial humates in agriculture: real substance or smoke and mirrors? *Agronomy* 6: 50.

Ma B.L., Biswas D.K., Herath A.W., Whalen J.K., Ruan S.Q., Caldwell C., Earl H., Vanasse A., Scott P., Smith D.L. 2015. Growth, yield, and yield components of canola as affected by nitrogen, sulfur, and boron application. *J. Plant Nutr. Soil Sci.* 178: 658–670.

Monda H., McKenna A.M., Fountain R., Lamar R.T. 2021. Bioactivity of humic acids extracted from shale ore: molecular characterization and structure–activity relationship with tomato plant yield under nutritional stress. *Front. Plant Sci.* 12: 660224.

Nardi S., Schiavon M., Francioso O. 2021. Chemical structure and biological activity of humic substances define their role as plant growth promoters. *Molecules* 26: 2256.

Nollendorfs V., Čekstere G. 2012. Supply of nutrients to rapeseed. *Saimnieks* 8: 46–48. /in Latvian/

Orlovius K. 2000. *Fertilizing for High Yield and Quality. Oilseed Rape.* Basel, International Potash Institute Bulletin No. 16, 125 p.

Osvalde A., Karlsons A., Cekstere G., Malecka S. 2012. Effect of humic substances on nutrient status and yield of onion (*Allium cepa* L.) in field conditions. *Proc. Latvian Acad. Sci., B* 66: 192-199.

Osvalde A., Karlsons A., Cekstere G., Vojevode L. 2016. The effect of vermicompost-derived humic substances on nutrient status and yield of organic potato in field conditions. *Acta Hortic.* 1142: 277-284.

Paoli R., Feofilovs M., Kamenders A., Romagnoli F. 2022. Peat production for horticultural use in the Latvian context: Sustainability assessment through LCA modeling, *J. Clean. Prod.* 378: 134559.

Plank C.O., Tucker M.R. 2000. Canola. In: Campbell C.R. (Ed.) *Reference Sufficiency Ranges for Plant Analysis in the Southern Region of the United States*. Southern Cooperative Series Bulletin 394, p. 9–10.

Rahman N, Schoenau J. 2021. Response of different crop cultivars to micronutrient fertilization and relationship to rhizosphere soil properties. *Commun. Soil Sci. Plant Anal.* 52: 1286–1300.

Rahman N., Schoenau J. 2022. Bioavailability, speciation, and crop responses to copper, zinc, and boron fertilization in south-central Saskatchewan soil. *Agronomy* 12: 1837.

Rose M.T., Patti A.F., Little K.R., Brown A.L., Jackson W.R., Cavagnaro T.R. 2014. A meta–analysis and review of plant–growth response to humic substances: Practical implications for agriculture. *Adv. Agron.* 124: 37–89.

Sikorska A, Gugała M., Zarzecka K. 2020. The impact of foliar feeding on the yield components of three winter rape morphotypes (*Brassica napus* L.). *Open Agric.* 5: 107–116.

Szczepanek M., Wilczewski E., Grzybowski K. 2016. Response of winter oilseed rape (*Brassica napus* L.) on soil applied humus preparation and foliar potassium fertilizer. *Acta Sci. Pol. Agric.* 15: 85–94.

Szczepaniak W. 2015. The mineral profile of winter oilseed rape in critical growth stages-potassium. *J. Elem.* 20: 203–215.

Szczepaniak W., Grzebisz W., Potarzycki J., Łukowiak R., Przygocka-Cyna K. 2015. Nutritional status of winter oilseed rape in cardinal stages of growth as the yield indicator. *Plant Soil Environ.* 61: 291–296.

Vronskis O., Kakitis A., Laukmanis E., Nulle I. 2016. Earthworm biohumus conditioning for pellet production. In: Proceedings of the 15th International Scientific Conference "Engineering for Rural Development", Jelgava, Latvia, 25–27 May, 2016; 15, pp. 997–1002.

Wysocki D.J., Corp M., Horneck D.A., Lutcher L.K. 2007. *Irrigated and Dryland Canola*. Nutrient Management Guide; EM 8943-E; Oregon State University, Corvallis, OR, USA, 8 p.

Received 19 January 2024; received in revised form 2 February 2024; accepted 5 February 2024