

## FOLIAR APPLIED BIOPREPARATIONS AS A NATURAL METHOD TO INCREASE THE PRODUCTIVITY OF GARDEN THYME (*Thymus vulgaris* L.) AND TO IMPROVE THE QUALITY OF HERBAL RAW MATERIAL

Cezary A. Kwiatkowski<sup>1</sup>, Małgorzata Haliniarz<sup>1✉</sup>, Elżbieta Harasim<sup>1</sup>, Barbara Kołodziej<sup>2</sup>, Alena Yakimovich<sup>3</sup>

<sup>1</sup> Department of Herbology and Plant Cultivation Techniques, University of Life Sciences in Lublin, Akademicka 13, 20-950 Lublin, Poland

<sup>2</sup> Department of Industrial and Medicinal Plants, University of Life Sciences in Lublin, Akademicka 15, 20-950 Lublin, Poland

<sup>3</sup> Institute of Plant Protection in Priluki, Minsk District, 2 Mira, 223011 Belarus

### ABSTRACT

The aim of this experiment was to determine the effects of three foliar biopreparations applied once or twice (growth stimulant Bio-algeen, fertilizer Herbagreen Basic, and Effective Microorganisms in the form of EM Farming spray) on yield and quality of herbal raw material of organically grown garden thyme (*Thymus vulgaris*). It was proved that the Bio-algeen had an effect on increasing thyme productivity, whereas Effective Microorganisms had no impact at all on herb yield. The beneficial action of biopreparations was more evident under favorable hydrothermal conditions over the study period than under unfavorable conditions. The biopreparations stimulated an increase in the essential oil content in the thyme herb. Bio-algeen, especially when applied twice, had the greatest positive influence on the quality of raw material. Effective Microorganisms were found to have a smaller effect (positive and negative), whereas Herbagreen Basic had positive effect on thyme yield and essential oil content. The effect of Bio-algeen, and to a lesser extent that of Effective Microorganisms, on the content of natural antioxidants (phenolic acids, polyphenols) in the thyme raw material and, moreover, its impact on free radical scavenging and antioxidant activity should be considered to be a particularly valuable finding. Due to application of the Bio-algeen, herbal raw material characterized by the best health-promoting parameters can be obtained.

**Key words:** garden thyme, Bio-algeen, Herbagreen Basic, Effective Microorganisms, herb yield, quality of herbal raw material

### INTRODUCTION

Garden thyme (*Thymus vulgaris* L.) is native to the Mediterranean Sea basin. This plant has a high ability to adapt to environmental conditions and due to this it is grown in temperate climate areas across Europe [Thompson et al. 1998]. Thyme yield and its raw material quality depend on many factors, such as genetic, climatic, soil, and agronomic ones [Shalaby

and Razin 1992, Rey 1993]. There is still a need to continually improve agronomic practices in growing herbal plants, in particular garden thyme which is very popular in Poland and across the world, especially in the context of organic and environmentally friendly methods. We can distinguish here, for example, the introduction of foliar applied biopreparations in

✉ malgorzata.haliniarz@up.lublin.pl

different forms in order to select the most beneficial method that positively affects yield and quality of herbal raw material [Borgen and Davanlou 2000, Przybysz et al. 2010, Kwiatkowski et al. 2014]. Bio-algeen, among others, is a growth biostimulant registered for use in organic farming; this preparation is an extract from sea algae that activates metabolic processes in crop plants, makes them resistant to fungal and viral pathogens, and contributes to better absorption of soil nutrients by the plant. Bio-algeen has been observed to have a positive impact on yield and quality of tomato fruits [Dobromilska et al. 2008] as well as yield of common basil [Kwiatkowski and Juszcak 2011] and garden thyme raw material [Kwiatkowski 2011]. Because NPK mineral fertilizers are not used in organic agriculture, the foliar fertilizer Herbagreen Basic can be used as alternative or supplemental fertilization. It is a latest-generation compound foliar fertilizer produced from natural minerals using advanced turbine technology. Herbagreen Basic is intended for fertilization and stimulation of field- and shade house-grown plants as well as for enhancement of their resistance. It supplies plants with highly available and non-toxic forms of calcium, silicon, magnesium, iron, titanium, manganese, and other elements. It induces plant resistance, detoxifies, increasing the content of antioxidants in the cell sap, and reduces the susceptibility of plants to pest attacks. This foliar fertilizer also stimulates photosynthesis by providing additional CO<sub>2</sub> to the intercellular cavities [<http://www.naturalcrop.com/herbagreen>].

Increased yields and improved quality of herbal raw material can also be achieved by applying Effective Microorganisms. It has been shown that a microbiological preparation (e.g. EM Farming) produces positive results (an increase in yield and in the content of important chemical components) in growing some vegetable and herbal plants [Borgen and Davanlou 2000, Xu 2000, Javaid 2006, Singh 2007]. Microbiological preparations contain, among others, lactic bacteria (*Lactobacillus casei*, *Streptococcus lactis*), photosynthetic bacteria (*Rhodospseudomonas palustris*, *Rhodobacter sphaeroides*), yeasts (*Saccharomyces albus*, *Candida utilis*), actinobacteria (*Streptomyces albus*), and mold fungi (*Aspergillus oryzae*, *Mucor hiemalis*) [Higa 1998, Valarini et al. 2003].

This study **hypothesized** that foliar fertilization with biopreparations (applied once or twice)

would contribute to producing satisfactory yields of high-quality (in particular in terms of antioxidant quality) garden thyme raw material, which would allow us to consider these agents to be suitable for application in organic farming of this herbal plant. An assumption was also made that the efficacy of action of biopreparations could be dependent on weather conditions during the growing season.

**The aim of this study** was to determine the effects of three foliar fertilizers (Herbagreen basic, Bio-algeen, and EM Farming) on yield and selected quality parameters of garden thyme raw material in relation to the control treatment (without application of biopreparations).

## Methods

A field experiment in growing garden thyme (*Thymus vulgaris* L.) was conducted over the period 2014–2016 in Dys, Municipality of Niemce (51°18'57"22°35'06"E), Lubelskie Voivodeship, Poland. The experiment was set up as a split-block design with three replicates and a single plot area of 5 m<sup>2</sup>. The study determined the suitability of garden thyme (cv. 'Słoneczko') for organic cultivation. Garden thyme was grown on podzolic soil (pH in 1 mol of KCl = 6.0–6.3), classified as very good rye soil complex. In the field assigned for the experiment, crops had been grown organically (without using synthetic NPK mineral fertilizers and without application of plant protection chemicals – herbicides, fungicides, insecticides) during the last three years before the establishment of the experiment. Based on tests of soil samples taken each year before the establishment of the experiment, it was found that the soil was characterized by a medium content of available macronutrients (P = 79.2–80.9, K = 84.5–85.7, Mg = 30.2–31.4 mg kg<sup>-1</sup>). The soil humus content was 1.39–1.43% (Tab. 1).

The experiment included the following experimental treatments in the cultivation of garden thyme:

A – without application of foliar sprays (control treatment),

B – foliar spraying with Herbagreen Basic (10.0 g in 1.0 l of water),

C – foliar spraying with Bio-algeen S90 (4.0 ml in 1.0 l of water),

D – foliar spraying with EM Farming EMa – (60.0 ml in 1.0 l of water),

**Table 1.** Characteristics of the soil used in the experiment

Year	pH in 1 mol of KCl	Content			
		P (mg kg <sup>-1</sup> soil)	K (mg kg <sup>-1</sup> soil)	Mg (mg kg <sup>-1</sup> soil)	Humus (%)
2014	6.3	80.9	85.7	31.4	1.43
2015	6.0	79.4	84.5	30.2	1.40
2016	6.1	80.1	84.9	29.7	1.39

**Table 2.** Components of the sprays used in the experiment

Name of spray	Spray composition
Bio-algeen S90	an extract from sea algae; the spray contains 90 groups of chemical compounds, including amino acids, vitamins, alginic acid, and other unidentified active ingredients of seaweeds; the major elements include the following: nitrogen – 0.02%, phosphorus – 0.006%, potassium – 0.096%, calcium – 0.31%, magnesium – 0.021%, as well as boron – 16 mg kg <sup>-1</sup> , iron – 6.3 mg kg <sup>-1</sup> , copper – 0.2 mg kg <sup>-1</sup> , manganese – 0.6 mg kg <sup>-1</sup> ; zinc – 1.0 mg kg <sup>-1</sup> ; moreover, the spray contains molybdenum and selenium
Herbagreen Basic	calcium oxide (CaO) – 36.7%, silicon dioxide (SiO <sub>2</sub> ) – 17.0%, iron trioxide (Fe <sub>2</sub> O <sub>3</sub> ) – 3.4%, magnesium oxide (MgO) – 2.4%, titanium dioxide (TiO <sub>2</sub> ) – 0.5%, potassium oxide (K <sub>2</sub> O) – 0.5%, sodium oxide (Na <sub>2</sub> O) – 0.5%, sulfur trioxide (SO <sub>3</sub> ) – 0.4%, phosphorus pentoxide (P <sub>2</sub> O <sub>5</sub> ) – 0.5%, manganese oxide (MnO) – 0.1%; and trace amounts of boron (1), cobalt (13), copper (26), zinc (34) (mg kg <sup>-1</sup> DM)
EM Farming	anaerobic organisms which release free, chemically uncombined oxygen into the environment during metabolic processes (photosynthetic bacteria, actinobacteria, lactic acid bacteria, fermentation fungi, yeasts) – the percentage contributions of particular microorganism strains in the spray is the manufacturer's secret (patent) and this information is not included in any available data sheets

**Table 3.** Selyaninov's hydrothermal coefficient (K) in individual months of the 2014–2016 growing seasons

Month	Year		
	2014	2015	2016
IV	1.33	1.19	1.21
V	0.92	1.25	1.14
VI	1.24	0.63	0.52
VII	0.80	0.51	0.68
VIII	0.85	0.65	0.59

E – double foliar spraying with Herbagreen Basic (2 × 5.0 g in 1.0 l of water),

F – double foliar spraying with Bio-algeen S90 (2 × 2.0 ml in 1.0 l of water),

G – double foliar spraying with EM Farming – EMa (2 × 30.0 ml in 1.0 l of water) – Table 2.

The previous crop for garden thyme was winter oilseed rape grown for green manure. As regards soil (pre-sowing) fertilization, a mineral fertilizer approved for use in organic farming was applied. Each year, thyme seeds were sown directly into the soil in the third 10 days of April. Seeding was done using a seed drill with a press wheel at a seeding rate of 3.0 kg ha<sup>-1</sup> and with a row spacing of 40 cm. Weed control involved mechanical weed removal (weeder) at 3–5 leaf stage of thyme.

The foliar sprays (treatments B-G) were applied under a pressure of 0.25 MPa using a backpack weed sprayer. In the double spraying treatments, they were applied at 2–3 leaf stage of thyme and at 5–7 leaf stage (following mechanical weed removal). Single application of the sprays was carried out at 5–7 leaf stage of thyme.

Garden thyme was harvested in the second/third 10 days of April with a sickle bar mower, cutting the herb at a height of about 5 cm. After cutting, the herb was dried in a belt dryer at a temperature of about 35°C and subsequently threshed using a Warmianka thresher. The threshed yield was weighed and then the obtained values expressed in kg per plot were converted to t ha<sup>-1</sup>.

Samples were taken from the herbal raw material harvested for chemical composition determinations. Chemical assays were performed at the Vegetable and Herbal Raw Material Quality Laboratory at the Department of Vegetable Crops and Medicinal Plants, University of Life Sciences in Lublin, and the following parameters were assayed:

– essential oil content by pharmacopoeial method [Polish Pharmacopoeia IX 2011],

– chlorophyll A and B by liquid chromatography [Polish Pharmacopoeia VII 2006],

– total polyphenolic content by the Folin-Ciocalteu method (expressed as gallic acid [GAE] equivalents) [Singleton and Rossi 1965, Slinkard and Singleton 1977],

– total phenolic acid content by pharmacopoeial method according to Arnova [Polish Pharmacopoeia VI 2002],

– flavonoid content by the Christ-Müller's method, expressed as quercetin (QE) equivalents [Polish Pharmacopoeia IX 2011],

– 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity [Chen and Ho 1997],

– antioxidant activity by ABTS method reported by Re et al. [1999],

– reducing power of extracts (FRAP) according to Benzie and Strain [1996],

– identification and determination of phenolic acid content by high performance liquid chromatography [Najda 2004],

– carotenoid content by spectrophotometric method at a wavelength of 450 nm [Lichtenthaler and Wellburn 1983].

To determine the temporal and spatial variation of meteorological elements and to evaluate their impact on the growing season of garden thyme, Selyaninov's hydrothermal coefficient (K) was calculated [Bac et al. 1993], dividing the monthly total rainfall by one tenth of the sum of the mean daily air temperatures for a given month (Tab. 3). The calculated hydrothermal coefficient values indicate that during the first months of the thyme growing season plants were well supplied with water, while small water deficits were only recorded in May 2014. But in the summer months (VI–VIII) of 2015 and 2016, on the other hand, minor drought or drought periods were found to occur, except for June 2014. To sum up, the 2014 growing season can be considered to be beneficial for thyme cultivation in terms of water and thermal conditions, whereas in the second part of the growing season in the years 2015–2016 there were water deficits in the soil (drought or minor drought), which had a negative impact on the growth and development of the herbal plant.

The study results were statistically analyzed by analysis of variance and verified by Tukey's test at a significance level of  $\alpha = 0.05$ . Statistica software was used for statistical calculations.

## RESULTS

Garden thyme raw material yield depended on the application of the foliar sprays. Significantly the lowest herb yield was obtained from the control plots and also in the treatments with Effective Microorganisms application. The statistically proven highest yield

was harvested from treatments C and F, and thus in the treatment where the growth stimulant Bio-algeen was used (especially in the case of its double application). When the above-mentioned spray was applied once or twice, thyme herb yield was found to increase by 21–26% relative to the control treatment (A) and compared to treatments D and G. Application of Herbagreen Basic (treatments B and E) also resulted in a significant increase in thyme yield (by 9–14%) compared to treatments A, D, and G. A significantly higher thyme yield was recorded in the year 2014, which was more favorable in terms of hydrothermal conditions in comparison to the years 2015–2016 that were characterized by drought in the summer months. Significantly the highest thyme herb yield (2.21 t ha<sup>-1</sup>) was harvested in 2014 in the treatment with double application of the growth biostimulant Bio-algeen (Tab. 4).

All treatments with foliar spray application included in the experiment had a significant effect on increasing (by 0.51–0.84 percentage point [p.p.]) the essential oil content in the raw material of garden thyme relative to the control (without application of biopreparations). As regards the individual treatments with foliar spray application, single and especially double application of the growth biostimulant Bio-algeen (treatments C and F) as well as double application of Effective Microorganisms (treatment G) had the most beneficial effect on essential oil content. The essential oil content values found in the above-mentioned treatments were significantly higher than those obtained in treatments B and E (where Herbagreen Basic was applied). The essential oil content in the thyme raw material was also affected by year. A significantly higher essential oil content (by 0.20–0.25 p.p.) was recorded in the year 2014, which was favorable for thyme growth compared to the years 2015–2016 (Tab. 5).

Weather conditions during the study period did not modify significantly the content of polyphenols and phenolic acids in the raw material of garden thyme, whereas the foliar sprays influenced differently the above-mentioned quality parameters. Significantly the highest total polyphenols were found after double application of Effective Microorganisms (treatment G) and in the control treatment (A) (without application of biopreparations), in relation to the other treatments. But if we compare the effects of the individual treatments involving biopreparation application, we notice

that both single and double application of the growth biostimulant Bio-algeen and the EM Farming spray contributed to a significantly higher polyphenolic content than application of the foliar fertilizer Herbagreen Basic (Tab. 6).

As far as the phenolic acid content in the raw material of garden thyme is concerned, in most cases the biopreparations were found to have a negative influence compared to the control treatment (A) (without the use of these agents). Only double application of Bio-algeen (treatment F) caused an increase (but statistically non-significant) in total phenolic acids relative to the control. The other foliar spray application treatments contributed to a statistically proven decrease in total phenolic acids relative to treatments A and F, but the use of the Herbagreen Basic spray had a particularly adverse impact on this quality parameter (Tab. 6).

All the biopreparations used in the experiment and their application treatments contributed to a significant decrease in the flavonoid content (by 20.3–32.4%) relative to the control. Application of Herbagreen Basic (treatments B and E) had a particularly adverse effect on flavonoid content, resulting in a significant reduction in this quality indicator also compared to the treatments with application of Bio-algeen (C, F) and Effective Microorganisms (D, G) – Table 7.

In most cases, the biopreparations had a significant negative influence on the carotenoid content in the raw material of garden thyme relative to the control (A). It was only in the treatment with double Bio-algeen application that a similar carotenoid content was found as that for the control plots. Application of Effective Microorganisms (treatments D and G) caused the highest loss in the carotenoid content in the thyme raw material, both in relation to the control (A) and compared to treatments B, C, E, and F (Tab. 7).

When analyzing the data contained in Table 8, we notice significant variations in the effects of the individual treatment combinations with foliar spray application on the thyme herb's ability to scavenge free radicals. EM Farming, in particular in the case of its double application (treatment D and G), and Bio-algeen, also when applied twice (treatment F), showed the most beneficial effect on the parameter in question. Herbagreen Basic, on the other hand, caused a radical decrease in free radical scavenging activity, by 55–70% relative to the control treatment (A) and by

**Table 4.** Dry weight yield of thyme herb (t ha<sup>-1</sup>)

Treatment	Year			Mean
	2014	2015	2016	
A	1.65	1.36	1.41	1.47
B	1.77	1.50	1.56	1.61
C	2.06	1.79	1.72	1.85
D	1.60	1.31	1.38	1.43
E	1.85	1.66	1.62	1.71
F	2.21	1.90	1.86	1.99
G	1.69	1.40	1.47	1.52
Mean	1.83	1.56	1.57	–

LSD (0.05) for: foliar sprays (a) = 0.159, years (b) = 0.151, interactions a × b = 0.147

**Table 5.** Essential oil content in the raw material of garden thyme (% DM)

Treatment	Year			Mean
	2014	2015	2016	
A	2.97	2.70	2.73	2.80
B	3.45	3.33	3.27	3.35
C	3.68	3.47	3.43	3.52
D	3.61	3.43	3.40	3.48
E	3.51	3.23	3.20	3.31
F	3.82	3.55	3.50	3.62
G	3.74	3.67	3.53	3.64
Mean	3.54	3.34	3.29	–

LSD (0.05) for: foliar sprays (a) = 0.168, years (b) = 0.193, interactions a × b = n.s.

**Table 6.** Total polyphenols (mg GAE 100 g<sup>-1</sup>) and total phenolic acids (mg 100 g<sup>-1</sup>) in the raw material of garden thyme – mean for 2014–2016

Treatment	Total polyphenols (mg GAE 100 g <sup>-1</sup> )	Total phenolic acids (mg 100 g <sup>-1</sup> )
A	743.49	192.97
B	482.10	150.51
C	625.67	177.01
D	582.80	168.00
E	565.62	164.34
F	673.60	198.58
G	769.11	168.52
LSD (0.05)	50.234	8.441



**Table 7.** Content of flavonoids (mg QE 100 g<sup>-1</sup>) and carotenoids (mg g<sup>-1</sup>) in the raw material of garden thyme – mean for 2014–2016

Treatment	Flavonoid content	Carotenoid content
A	32.579	0.685
B	22.181	0.316
C	25.956	0.511
D	25.900	0.213
E	22.034	0.458
F	25.572	0.669
G	25.846	0.258
LSD (0.05)	2.3644	0.0214

**Table 8.** DPPH radical scavenging activity (μm TE g<sup>-1</sup>) determined in the raw material of garden thyme

Treatment	Year			Mean
	2014	2015	2016	
A	52.000	51.231	48.692	50.641
B	17.077	15.420	14.652	15.716
C	53.017	51.788	48.916	51.167
D	55.587	52.312	48.697	52.198
E	24.288	22.018	22.455	22.920
F	59.142	57.113	56.876	57.710
G	71.385	67.558	66.915	68.619
Mean	47.499	45.348	43.886	–

LSD (0.05) for: foliar sprays (a) = 2.6124, years (b) = 2.1321, interactions a × b = 2.7663

**Table 9.** ORAC antioxidant activity (μm TE g<sup>-1</sup>) determined in the raw material of garden thyme

Treatment	Year			Mean
	2014	2015	2016	
A	10.98	9.71	8.85	9.84
B	13.11	12.55	12.16	12.60
C	22.59	21.73	21.44	21.92
D	16.91	15.22	15.36	15.83
E	14.21	13.33	13.45	13.66
F	22.64	21.80	21.53	21.99
G	18.22	16.98	17.13	17.44
Mean	16.95	15.90	15.70	–

LSD (0.05) for: foliar sprays (a) = 1.232, years (b) = 1.012, interactions a × b = n.s.

**Table 10.** Reducing power of extracts (FRAP) ( $\mu\text{m TE g}^{-1}$ ) determined in the raw material of garden thyme

Treatment	Year			Mean
	2014	2015	2016	
A	5.727	5.322	5.287	5.445
B	2.246	1.025	1.006	1.425
C	5.984	5.764	5.699	5.815
D	3.951	3.722	3.705	3.792
E	3.144	2.066	2.102	2.437
F	6.599	6.312	6.266	6.392
G	4.347	4.203	4.014	4.188
Mean	4.571	4.056	4.011	–

LSD (0.05) for: foliar sprays (a) = 0.3695, years (b) = 0.4055, interactions a × b = n.s.

**Table 11.** Chlorophyll a and chlorophyll b content ( $\text{mg g}^{-1}$ ) in the raw material of garden thyme – mean for 2014–2016

Treatment	Chlorophyll a content	Chlorophyll b content
A	1.253	0.355
B	1.243	0.345
C	1.122	0.326
D	1.112	0.310
E	0.983	0.252
F	0.942	0.260
G	0.847	0.240
LSD (0.05)	0.1022	0.0493

as much as 290–430% compared to treatment G (double application of Effective Microorganisms). Weather conditions prevailing in the individual growing seasons of garden thyme also modified significantly free radical scavenging activity. The unfavorable Selyaninov's hydrothermal coefficient that characterized months VI–VIII in the years 2015 and 2016 translated into decreased free radical scavenging activity in the thyme herb (by 5–8%) in relation to the year 2014, which was favorable in this respect. Significantly the highest free radical scavenging activity ( $71.385 \mu\text{m TE g}^{-1}$ ) was recorded in 2014 in the treatment with double application of Effective Microorganisms (Tab. 8).

Foliar spray application, regardless of the treatment, resulted in statistically proven higher antioxidant activity of the thyme raw material compared to the control treatment. Application of Bio-algeen (irrespective whether it was applied once or twice) caused a particularly large increase in the antioxidant properties of thyme, whereas foliar spraying with EM Farming (an increase in the ORAC value by 38–44%) and Herbagreen Basic (an increase in the ORAC value by 22–28%) yielded a smaller positive effect. The more favorable hydrothermal conditions that characterized the 2014 growing season had a significant impact on increased antioxidant activity of the thyme herb (by



about 7%) in relation to the years 2015–2016 (Tab. 9).

The foliar sprays exhibited different effects on the reducing power of extracts determined in the raw material of garden thyme. A significant increase in reducing power compared to the control (A) was found when Bio-algeen was used (single application – an increase in reducing power FRAP by about 7%; double application – an increase by about 15%). In the case of the other biopreparations and their application treatments, the reducing power of the extracts was observed to significantly decrease compared to treatments A, C, and F. A particular high (2.2–3.8-fold) reduction in the reducing power of the extracts was found under the influence of Herbagreen Basic application (treatments B and E). The favorable weather conditions during the 2014 growing season contributed to significant improvement in the reducing power of the extracts in the herbal raw material studied compared to the dry years 2015–2016 (Tab. 10).

The data contained in Table 11 illustrate that single application of the biopreparations used in the experiment did not result in changes (increase or decrease) in the chlorophyll a and chlorophyll b content in the thyme herb compared to the control treatment (A) (without foliar spray application). Double application of these sprays, on the other hand, caused a significant decrease in chlorophyll content in relation to the control and treatments B, C, and D by about 15% (Bio-algeen) to 30% (EM Farming).

## DISCUSSION

In the present study, the growth biostimulant Bio-algeen as well as the foliar fertilizer Herbagreen Basic had a positive effect (especially in the treatment with double application) on garden thyme herb yield, which was 1.61–1.99 t ha<sup>-1</sup>. But application of Effective Microorganisms (EM Farming) was practically of no importance for the productivity of this herbal plant – the dry herb yield was about 1.5 t ha<sup>-1</sup>, similarly to the one found in the control treatment. In the literature of the subject, we find different views regarding the significance of foliar biopreparations in determining yield and quality of plant produce. The studies by Kwiatkowski [2011] as well as by Kwiatkowski and Juszcak [2011] regarding single foliar application of growth stimulants (Asahi SL, Bio-algeen, Tytanit)

in garden thyme and sweet basil crops proved that all these preparations contributed to improvement in the quantitative parameters of the herbal raw material (plant height, number of branches, total yield). In turn, the above-mentioned sprays had a negative impact on the chemical composition of the thyme raw material (the content of thymol and macronutrients – N, P, K, Ca) [Kwiatkowski 2011]. In the study by Król [2009], application of Asahi SL, along with Mikosol (foliar fertilizer), also resulted in increased productivity of garden thyme by 13.6%. When conducting a study on application of foliar sprays in the cultivation of chamomile under water stress conditions induced by high air temperature and low soil moisture content, Woropaj-Janczak et al. [2011] found the Asahi SL preparation to be useful (an increase in herb dry matter yield, higher essential oil content yield). However, these authors did not find the biostimulant Bio-algeen and the micronutrient fertilizer Ekolist P to positively affect the quantitative and qualitative parameters of chamomile. The studies by Panajatov et al. [1997], Djanaguirman et al. [2005], and Kołodziej [2008a, 2008b] are confirmation of the positive impact of growth biostimulants on the chemical composition of plant material. The latter author notes that the action of stimulants depends on many factors, such as air humidity, soil moisture content, air temperature, and soil macronutrient availability. Bączek-Kwinta and Seidler-Łożykowska [2004] as well as Bączek-Kwinta et al. [2006] also report that the positive effects of foliar sprays are generally manifested more strongly under plant stress conditions (soil water deficit or too low an amount of available nutrients in the soil). In the present study, the biostimulant Bio-algeen contributed in particular to enhanced antioxidant properties of the thyme raw material and increased reducing power of the extracts as well as to an increase in the content of essential oil and phenolic acids. In earlier studies, Kwiatkowski et al. [2013, 2015] found growth stimulants to beneficially affect the chemical composition of carrot roots.

Kwiatkowski [2015] proved that the growth stimulant Asahi SL exhibited a positive effect on yield and quality of chamomile raw material, while Ekolist P showed such an effect to a smaller extent. Effective Microorganisms had no impact on chamomile productivity and even contributed to a decrease in the flavonoid content in inflorescences. Sulewska and

Ptaszyńska [2005] as well as Martyniuk and Książak [2011] found EM Farming to have an insignificant influence on plant productivity on the example of maize, while Vliet et al. [2006] demonstrated this on the example of grass biomass.

Other authors [Higa 1998, Xu 2000, Shamshad et al. 2001, Condor et al. 2006, Singh 2007] prove that application of the EM Farming spray can cause a distinct increase in yield components, plant biometric characteristics, and some qualitative traits and, as a result, an increase in yield by even 15–20%. In this study, Effective Microorganisms positively influenced the content of essential oil and polyphenols and free radical scavenging activity in the thyme raw material as well as this foliar spray contributed to its greater antioxidant activity. But on the other hand, EM Farming caused a significant decrease in the carotenoid and chlorophyll content and weakened the reducing power of the extracts.

The thyme herb yield and essential oil content obtained in this experiment were similar to the results of other national [Kwiatkowski 2011] and foreign studies [Shalaby and Razin 1992, Heine et al. 2001].

This research proved the positive effect of Bio-algeen, and to a lesser extent in the case of Effective Microorganisms (EM Farming), on increasing the antioxidant properties and activity, phenolic acid content, and free radical scavenging activity in the thyme raw material. On the example of chamomile, Kwiatkowski et al. [2018] showed natural plant fertilization (spent mushroom substrate) to have a positive effect on significant improvement in the antioxidant properties of herbal raw material.

On the example of sweet basil, Onofrei et al. [2017] proved that the production of phenolic compounds and flavonoids was generally stimulated in fertilized basil plants compared to unfertilized ones, consistently for two consecutive years. Similarly, the antioxidant capacity of basil extracts increased as a result of ecological fertilization. Ecological fertilization is therefore recommended for cultivation of basil plants with an increased commodity value and potential phyto-medicinal properties. In a study on fertilization of *Salvia officinalis*, Geneva et al. [2010] noted that application of foliar fertilization and/or mycorrhizal colonization increased shoot and root dry biomass, enhanced the levels of ascorbate, and reduced glutathione and ac-

tivity of guaiacol peroxidase. The applied treatments lowered activities of the antioxidant enzymes CAT, AsPO, and SOD, and changed the relative quantity of essential oil.

A study conducted by Skupień and Oszmiański [2007], Skupień et al. [2008], in turn, demonstrates that application of foliar fertilizers caused multidirectional changes in the physical and chemical properties of the chokeberry fruit. However, the differences between individual fertilization treatments were not significant from the practical point of view (antioxidant properties of the raw material).

## CONCLUSIONS

1. In the light of the obtained results of the three-year study conducted under the soil and climatic conditions of Poland (Lublin region), the suitability of foliar applied biopreparations for in organic farming of garden thyme cannot be determined unambiguously. Their impact on the productivity and quality of thyme raw material, depending on the trait analyzed, was both positive and negative. Generally, the action of the biopreparations was not related to weather conditions during the study period. Nevertheless, the study found a statistically confirmed positive interaction between favorable hydrothermal conditions and the effect of the Bio-algeen spray on thyme herb yield as well as the impact of Effective Microorganisms on free radical scavenging activity in the thyme herb.

2. The foliar sprays Bio-algeen and Herbagreen Basic had a significant effect on the increase in thyme yield, whereas all the biopreparations positively affected the essential oil content in the herb. Moreover, Bio-algeen (in particular in the case of its double application) contributed to a distinct increase in the antioxidant properties, reducing power of the extracts, and phenolic acid content, while EM Farming caused an increase in the polyphenolic content as well as in antioxidant and free radical scavenging activity, which is of essential importance in assessment of thyme raw material from the health-related point of view.

3. All the biopreparations and their application treatments were found to have a negative effect on the content of phenolic acids, flavonoids and carotenoids in the raw material of garden thyme. Furthermore, Effective Microorganisms had a negative influence on

the chlorophyll and carotenoid content in the thyme herb and weakened the reducing power of the extracts. Application of Herbagreen Basic, in turn, contributed to a rapid reduction in free radical scavenging activity and caused a significant decrease in the reducing properties of the extracts.

4. The obtained study results show that, among the group of the biopreparations compared, the growth biostimulant Bio-algeen had the greatest positive importance in organic production of garden thyme, Effective Microorganisms (EM Farming preparation) had no effect on herb yield, while their impact on the qualitative parameters of the herbal raw material was bipolar (generally positive, but also negative), but the foliar fertilizer Herbagreen Basic proved to be least useful.

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