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EFFECTS OF TREATMENT OF APPLE TREES WITH VARIOUS BIOPRODUCTS ON TREE GROWTH AND OCCURRENCE OF MYCORRHIZAL FUNGI IN THE ROOTS

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ABSTRACT

The aim of the study was to evaluate selected bioproducts in terms of their effects on tree growth and the presence of mycorrhizal fungi in the roots of trees of two apple cultivars, 'Topaz' and 'Ariwa', grown under greenhouse conditions. The trees were planted in rhizoboxes and grown under the following fertilization regimes: 0 – control, NPK control, manure, Micosat F, Humus UP, Humus Active + Aktywit PM, BF Amin, BF Quality, Tytanit and Vinassa. The results showed that the biostimulants: Humus UP, Humus Active and Aktywit PM produce beneficial effects on the morphological characteristics of the root system, and Micosat F on the extent of mycorrhiza formation in the roots. The bioproducts used are a safe, effective and economically viable method of fertilizing plants, limiting the use of chemical means of production and thus helping to protect the environment.

Key words: plant growth, AMF fungi, *Malus × domestica*, biopreparations

INTRODUCTION

The development of organic methods of plant cultivation, as a safe and environmentally friendly way of agricultural production, has resulted in a significant increase in the number of farms in the world [Willer and Kilcher 2012]. The search for new biofertilizers is due to the reduced availability of the traditional means of production such as manure, composts and organic mulches [Derkowska et al. 2015b]. The use of organic waste from plant production, e.g. from the production of wine or oil products, is a valuable source of organic matter and minerals for plants and is a safe material for the production of biofertilizers [Polverigiani et al. 2014a]. Biofertilizers may contain in their composition, among other things, plant ex-

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isms that increase the availability of minerals from soil organic matter, which are the source of food for plants [Bhattacharjee and Dey 2014]. The biofertilizers and biostimulants of natural origin used on fruit crops increase plant productivity by acting on their physiology, e.g. by stimulating the biochemical processes that are important in the growth and yielding of plants. Natural biopreparations are friendly to people and the environment, which leads to sustainable crop production [Wally et al. 2013, Mishra and Dash 2014]. The use of biofertilizers of organic origin is conducive to the development of symbiotic soil microorganisms, including bacteria and mycor-

tracts (humic compounds) and beneficial microorgan-



rhizal fungi, which beneficially affect the growth and development of plants [Cameron 2010, Raviv 2010]. The growing of plants by ecological means precludes the use of mineral fertilizers and other chemical means of plant production (pesticides, herbicides), and has a beneficial effect on the biodiversity of mycorrhizal fungi in the rhizosphere of plants [Tuck et al. 2014]. By contrast, excessive use of mineral fertilizers and plant protection products adversely affects the occurrence of arbuscular mycorrhizal fungi (AMF) and their species biodiversity [Turrini and Giovannetti 2012]. The ecological methods of plant fertilization ensure the safety of the crops produced and help maintain the soil at the required fertility level, by, e.g. increasing the occurrence and biodiversity of arbuscular mycorrhizal fungi [Verbruggen et al. 2012, Hazard et al. 2013, Jansa et al. 2014]. The worldwide research in this regard is aimed at determining the effectiveness of the use of biofertilizers, including the effectiveness of fertilization of particular crop species [Quilty and Cattle 2011]. The aim of the study was to evaluate selected bioproducts in terms of their effects on tree growth and the presence of mycorrhizal fungi in the roots of trees of two apple cultivars, 'Topaz' and 'Ariwa', grown under greenhouse conditions.

MATERIALS AND METHODS

The experiment was conducted in the glasshouse complex of the Research Institute of Horticulture in Skierniewice in the years 2009–2016. Trees of two apple cultivars, 'Topaz' and 'Ariwa', grafted onto M26 rootstocks, were planted in rhizoboxes measuring $60 \times 50 \times 8$ cm, which were filled with 17 kg of a podsolic soil per rhizobox. The soil had been taken from the experimental orchard of the Institute in Dąbrowice. The soil had a pH of 5.5 and the following composition: organic matter – 1.5%, N – 0.07% of air dry mass, C – 0.88% of air dry mass, P – 15 mg·100 g⁻¹ of soil, K – 11 mg·100 g⁻¹ of soil, Mg – 6 mg·100 g⁻¹ of soil, Ca – 1145 mg·kg⁻¹, B – 4.9 mg·1000 g⁻¹ of soil, Cu – 6.5 mg·1000 g⁻¹ of soil, Fe – 701 mg·1000 g⁻¹ of soil, Mn – 127 mg·1000 g⁻¹ of soil, Zn – 6.9 mg·1000 g⁻¹ of soil. The plants were grown under greenhouse condi-

tions, with a photoperiod of 16/8 h (day/night) and light intensity of 70 μ M m⁻² s⁻¹, at 25/20°C during the growing season and 5/8°C in the winter, and air humidity of 50%. The plants in every treatment of the experiment were grown in five replications (each represented by one rhizobox with one plant) in the following combinations:

1. Control 0 (no-treatment) – unfertilized podsolic soil (with soil content characterized above);

2. Control NPK – standard NPK soil fertilization: 4 g NH₄NO₃ per plant, 3 g triple superphosphate per plant and 6g K₂SO₄ per plant (70 kg N·ha⁻¹ as NH₄NO₃, 60 kg P·ha⁻¹ as P₂O₅, and 120 kg K₂O·ha⁻¹ as K₂SO₄);

3. Dry granulated bovine manure, suitable for organic farming (Doktor O'grodnik), containing: 55% C, 1% N, 0.3% P and 1% K; the product contains also microelements and soil microorganisms $(110^{6} \text{ CFU} \cdot \text{g}^{-1})$; it was applied to the soil, near the root system (1 g per plant), at planting;

4. Micosat F (CCS Aosta s.r.l.) – a mixture of beneficial soil fungi and bacteria containing: five species of AM fungi: *Glomus mosseae* Taxtersensu Gerd. & Trappe, *G. intraradices* Schenk & Smith, *G. caledonium* (Nicolson et Gerdemann) Trappe et Gerdemann, *G. viscosum* Nicolson and *G. coronatur* (Giovannetti); *Trichoderma viride* Pers.; three rhizosphere bacteria species (*Bacillus subtilis, Pseudomonas fluorescens, Streptomyces* spp.) with a total concentration of 10^6 CFU·g⁻¹ of substrate. The product contains 40% C, 0.15% N, 431 mg·kg⁻¹ P and 9558 mg·kg⁻¹ K; it was applied to the soil, near the root system (10 g per plant), at planting and then, 2 weeks after the first application, Micosat F MS 200 was applied to the leaves at (20 g per plant);

5. Humus UP (Ekodarpol) – an extract from a vermicompost containing 0.65% C, 0.03% N, $30.8 \text{ mg} \cdot \text{kg}^{-1}$ P and 4535 mg $\cdot \text{kg}^{-1}$ K; the product was first applied to the soil at planting as a 2% solution (5 ml \cdot 500 ml per plant) and then three times during the growing period (1% solution, 2.5 ml \cdot 500 ml per plant);

6. Humus Active + Aktywit PM (produced by Ekodarpol; Humus Active – an extract from organic manure produced with the help of Californian earthworms; Aktywit PM – an activator of microbial life,

produced on the basis of molasses); Humus Active was applied to the soil as a 2% solution at (2% solution, 10 ml \cdot 500 ml per plant) a in conjunction with a 1% solution of Aktywit PM applied to the soil at (1% solution, 5 ml \cdot 500 ml per plant);

7. BioFeed Amin (produced by Koppert; an extract containing plant amino acids); the preparation was applied to the soil as a 0.5% solution at 2.5 ml per plant;

8. BioFeed Quality (produced by Koppert; a seaweed extract containing humic and fulvic acids); the preparation was applied to the soil as a 0.5% solution at 2.5 ml per plant;

9. Tytanit (produced by Intermag); the stimulant was applied to the leaves as a 0.05% solution at 2,5 ml per plant;

10. Vinassa – molasses residue from yeast production containing 12.0% C, 1.86% N, 949 mg·kg⁻¹ P, 17615 mg·kg⁻¹ K; the product was applied to the plants five times during the growing period as a 0.5% solution (50 ml per plant).

After the completion of the experiment in February 2016, the plant material was collected to measure plant growth characteristics, root growth characteristics, and the degree of mycorrhizal association in the roots of 'Topaz' and 'Ariwa' apple trees.

Measurements and observations

Assessment of tree growth characteristics. The growth characteristics of the trees were determined over the years 2009–2016 by measuring the diameter of the tree trunk and the length and number of annual shoots (new growth of more than 5 cm). Each year, after the vegetation period, the diameter of the trunks was measured at a height of 30 cm above the soil surface with callipers, followed by calculations of the trunk cross-sectional area (TCSA). Also performed were measurements of the number of annual shoots, their total and average length. Each year after the vegetative growth of the trees, all the lateral shoots were cut off, and then their number and length were determined by the classical methods (tabs 1 and 2).

Determination of root growth characteristics. The root system of the apple trees was collected in February 2016 during the liquidation of the experiment in order to determine the morphological characteristics of the roots. The collected roots were rinsed free of soil particles with tap water, and after drying their fresh weight was determined. The roots were then scanned with an EPSON EXPRESSION 10000 XL root scanner. The growth characteristics of the roots of the apple trees, i.e., length, surface area, diameter, volume, and the number of root tips, were determined using the WinRhizo software [Arsenault et al. 1995]. The roots were then dried at 55°C for 7 days, and based on weight measurements, the dry weight of the roots was determined (tabs 3 and 4).

Assessment of root colonization by arbuscular mycorrhizal fungi. The roots of apple trees (10 g from each replication), collected in February 2016, were stained according to the method developed in the Rhizosphere Laboratory of the Research Institute of Horticulture [Derkowska et al. 2015a]. Next, microscopic specimens were prepared and examined with a Nikon 50i microscope (objectives with magnifications of $20\times$, $40\times$, $60\times$, $100\times$), and photographic records of the observed mycorrhizal structures were produced. The assessment of the degree of colonization of the roots by arbuscular mycorrhizal fungi was performed by the Trouvelot method [Trouvelot et al. 1986]. Based on the results, mycorrhizal frequency (F%) was calculated using the computer program MYCOCALC, available from the website: http://www2.dijon.inra.fr/mychintec/Mycocalc-prg/ /MYCOCALC.EXE (tab. 4).

Statistical analysis. The results were statistically analyzed using multivariate analysis of variance in the system of random blocks. Multiple comparisons of the means for the combinations were performed with Tukey's test, at a significance level of $\alpha = 0.05$, using STATISTICA v. 10 software package (StatSoft, Inc. 2011).

RESULTS

Compared to the other biopreparations, Vinassa and mineral fertilization significantly increased the number of shoots of apple trees of the cultivar 'Topaz'. The largest cumulative length of shoots and average length of an annual shoot were recorded for the trees of the cultivar 'Ariwa' resulting from the use of standard NPK fertilization. The standard NPK

Treatment	Number of shoots			Total	length of s (cm)	shoots	Average	e length of shoots (cm)		Trunk cross-sectional area (TCSA) (cm ²)		
	'Topaz'	'Ariwa'	$\overline{\mathbf{X}}$	'Topaz'	'Ariwa'	$\overline{\mathbf{X}}$	'Topaz'	'Ariwa'	$\overline{\mathbf{X}}$	'Topaz'	'Ariwa'	X
Control 0	12 b-d	11 a-d	12 b	232 b-f	300 d-g	266 а-с	20 а-е	28 e-h	24 ab	18 b	15 a	17 a
Control NPK	14 d	11 a-d	13 c	327 e-g	366 g	347 c	24 b-g	34 gh	29 c	19 b	15 a	17 a
Manure	12 b-d	12 b-d	12 b	231 b-f	249 d-g	240 а-с	19 a-e	22 a-f	23 ab	19 b	16 a	18 a
Micosat F	8 ab	11 a-d	10 a	111 ab	217 а-е	164 a	15 a-c	21 a-f	18 a	19 b	15 a	17 a
Humus UP	11 a-d	11 a-d	11 ab	122 а-с	228 b-е	175 ab	11 a	20 а-е	16 a	22 c	16 a	19 ab
Humus Active + Aktywit PM	7 a	10 a-d	9 a	100 a	268 d-g	184 ab	13 ab	28 e-h	21 ab	19 b	15 a	17 a
BF Quality	12 b-d	9 a-c	11 ab	200 a-d	213 а-е	207 ab	17 а-е	25 c-h	21 ab	20 b	15 a	18 a
BF Amin	12 b-d	11 a-d	12 b	197 a-d	301 d-g	249 а-с	16 a-d	27 d-h	22 ab	19 b	14 a	17 a
Tytanit	13 cd	10 a-d	12 b	242 c-g	354 fg	298 bc	19 a-e	37 h	28 bc	19 b	15 a	17 a
Vinassa	14 d	10 a-d	12 b	271 d-g	329 e-g	300 bc	20 а-е	32 f-h	26 b	19 b	14 a	17 a

Table 1. Effect of biofertilizers on the growth characteristics of 'Topaz' and 'Ariwa' apple trees. Greenhouse experiment,2009–2016

Means in columns marked with the same letter do not differ significantly at p = 0.05 according to Tukey's multiple test

Table 2. Effect of fertilization on the number and the total length of annual shoots, and the average length of an annual
shoot of apple tree cultivars 'Topaz' and 'Ariwa'. Greenhouse experiment, 2009-2016

Treatment		verage number annual shoo			rage total lei inual shoots	C	Average length of an annual shoot (cm)			
	'Topaz'	'Ariwa'	$\overline{\mathbf{X}}$	'Topaz'	'Ariwa'	$\overline{\mathbf{X}}$	'Topaz'	'Ariwa'	$\overline{\mathbf{X}}$	
Control 0	11 ab	9 a	10 a	182 ab	229 bc	206 b	17 ab	26 c	22 ab	
Control NPK	13 b	9 a	11 a	198 b	230 bc	214 bc	16 a	27 cd	22 ab	
Manure	12 ab	9 a	11 a	182 ab	206 b	194 ab	17 ab	24 bc	21 a	
Micosat F	12 ab	10 a	11 a	198 b	205 b	202 b	14 a	24 bc	19 a	
Humus UP	10 a	10 a	10 a	186 ab	185 ab	186 a	11 a	20 b	16 a	
Humus Active + Aktywit PM	11 ab	9 a	10 a	169 ab	209 b	189 a	14 a	26 c	20 a	
BF Quality	11 ab	10 a	11 a	109 a	233 с	171 a	15 a	23 b	19 a	
BF Amin	10 a	10 a	10 a	128 a	219 b	174 a	16 a	25 bc	21 a	
Tytanit	9 a	10 a	10 a	152 ab	231c	192 ab	19 ab	25 bc	22 ab	
Vinassa	11 ab	10 a	11 a	139 a	248 d	194 ab	17 ab	25 bc	21 a	

Means in columns marked with the same letter do not differ significantly at p = 0.05 according to Tukey's multiple test

Treatment	Root length (cm·plant ⁻¹)				t surface and m ² ·plant ⁻¹)			oot diamet nm∙plant⁻¹				
	'Topaz'	'Ariwa'	$\overline{\mathbf{X}}$	'Topaz'	'Ariwa'	$\overline{\mathbf{X}}$	'Topaz'	'Ariwa'	$\overline{\mathbf{X}}$	'Topaz'	'Ariwa'	$\overline{\mathbf{X}}$
Control 0	2480 ab	3844 c	3162 a	1541 gh	1295 c-g	1418 ab	1.53 bc	1.13 ab	1.33 a	42 b-h	36 a-f	39 a
Control NPK	3448 a-c	3185 a-c	3317 a	1132 а-е	1146 b-e	1139 a	1.48 a-c	1.17 ab	1.33 a	56 gh	34 а-е	45 b
Manure	2881 a-c	2649 а-с	2765 a	1377 d-h	1169 b-f	1273 a	1.52 bc	1.41 a-c	1.47 ab	41 b-h	52 e-h	47 b
Micosat F	3386 a-c	3384 a-c	3385 ab	1523 gh	1113 a-d	1318 a	1.46 a-c	1.06 ab	1.26 a	55 f-h	29 a-d	42 ab
Humus UP	3495 a-c	3532 а-с	3514 b	1616 h	1258 c-g	1437 ab	1.47 a-c	1.16 ab	1.32 a	60 h	37 a-g	49 b
Humus Active + Aktywit PM	2708 а-с	3607 bc	3158 a	1657 h	1175 b-f	1416 ab	2.12 d	1.04 ab	1.58 b	89 j	31 a-d	60 c
BF Quality	3531 a-c	3332 а-с	3432 ab	1472 f-h	1022 а-с	1247 a	1.33 а-с	0.98 a	1.16 a	49 d-h	25 а-с	37 a
BF Amin	3663 bc	2844 а-с	3254 a	1443 e-h	839 a	1141 a	1.25 a-c	0.97 a	1.11 a	45 c-h	20 a	33 a
Tytanit	3702 bc	2787 а-с	3245 a	1380 d-h	886 ab	1133 a	1.19 ab	1.02 ab	1.11 a	41 b-h	23 ab	32 a
Vinassa	3602 bc	2250 a	2926 a	1390 d-h	1146 b-e	1268 a	1.23 a-c	1.73 cd	1.48 ab	43 b-h	48 d-h	46 b

Table 3. Assessment of the morphological characteristics of the roots of 'Topaz' and 'Ariwa' apple trees under the influence of biofertilizers. Greenhouse experiment, 2009–2016

Means in columns marked with the same letter do not differ significantly at p = 0.05 according to Tukey's multiple test

Table 4. Effect of biofertilizers on the number of root tips, root fresh and dry weight, and mycorrhizal frequency in the
roots of 'Topaz' and 'Ariwa' apple trees. Greenhouse experiment, 2009-2016

Treatment	Number of root tips (per plant)				t fresh we (g∙plant ⁻¹)	0	Root dry weight (g·plant ⁻¹)			Mycorrhizal frequency (F%)		
	'Topaz'	'Ariwa'	$\overline{\mathbf{X}}$	'Topaz'	'Ariwa'	$\overline{\mathbf{X}}$	'Topaz'	'Ariwa'	$\overline{\mathbf{X}}$	'Topaz'	'Ariwa'	X
Control 0	19128 a-d	16281 ab	17705 a	365 ef	304 b-d	335 a	191 b-f	157 а-е	174 a	10 d	10 d	10 b
Control NPK	17521 а-с	17880 a-c	17701 a	409 fg	300 b-d	355 ab	221 d-g	141 ab	181 a	7.78 bc	3.33 a	5.56 a
Manure	17052 ab	15876 ab	16464 a	446 gh	342 de	394 c	260 gh	190 b-f	225 c	8.89 cd	10 d	9.45 ab
Micosat F	18837 a-d	17958 a-c	18098 ab	472 h	278 а-с	375 b	291 h	144 a-c	218 b	15.56 f	17.78 g	16.67 c
Humus UP	17252 ab	20139 b-e	18696 ab	449 gh	325 с-е	387 b	251 gh	171 a-f	211 b	14.45 f	15.55 f	15 c
Humus Active + Aktywit PM	17816 a-c	23434 с-е	20625 ab	426 gh	257 ab	342 ab	238 f-h	137 ab	188 a	14.44 f	14.45 f	15 c
BF Quality	18471 a-d	24899 e	21685 b	414 f-h	271 а-с	343 ab	233 f-h	156 a-d	195 ab	6.67 b	6.67 b	6.67 a
BF Amin	20795 b-e	16614 ab	18705 ab	399 fg	222 a	311 a	238 f-h	112 a	175 a	12.22 e	7.78 bc	10 b
Tytanit	24047 de	15322 ab	19685 ab	399 fg	267 а-с	333 a	230 e-h	136 ab	183 a	10 d	4.44 a	7.22 ab
Vinassa	23371 с-е	13677 a	18524 ab	401 fg	252 ab	327 a	214 c-f	111 a	163 a	8.89 cd	7.78 bc	8.34 ab

Means in columns marked with the same letter do not differ significantly at p = 0.05 according to Tukey's multiple test

fertilization also caused an increase in the average number of annual shoots in the cultivar 'Topaz'. The use of the fertilizer Vinassa significantly increased the average total length of annual shoots of the 'Ariwa' trees and the biostimulant Tytanit stimulated the growth of shoots in length in apple trees of the cultivar 'Ariwa'. The greatest trunk cross-sectional area (TCSA) was shown by the trees of the cultivar 'Topaz' treated with the bioproduct Humus UP, compared to the control trees (tabs 1 and 2). However, a comparison of the biopreparations used, regardless of the cultivar, does not indicate any beneficial effect of any of them on the growth of the aboveground parts of the plants (tabs 1 and 2).

The tested biopreparations had a favourable effect on the growth characteristics of the roots of apple trees. The bioproducts Humus Active and Aktywit PM significantly increased the surface area, diameter and volume of the roots of 'Topaz' apple trees, compared to the control trees fertilized with NPK. The greatest length of roots was shown by the control trees of the cultivar 'Ariwa', but these differences were not statistically significant (tab. 3). However, a comparison of the applied biopreparations, regardless of the cultivar, indicates other dependencies. The biostimulant Humus UP significantly influenced the increase in root length and root surface area, while Humus Active and Aktywit PM stimulated the growth in diameter and volume of the roots of the apple cultivars studied (tab. 3).

The use of the biopreparation BF Quality increased the number of tips of the roots of the 'Ariwa' trees, compared to the NPK control trees. Applications of the biofertilizer Micosat F contributed to a significant increase in fresh and dry weight of roots of the cultivar 'Topaz', compared to the roots of the control trees without fertilization (Control 0) and trees fertilized with NPK (tab. 4). Comparing the biopreparation used, regardless of the cultivar, it was found that the application of BF Quality and also Humus Active and Aktywit PM stimulated the plants to produce more root tips, while manure, Humus UP and Micosat F increased the fresh and dry weight of the roots of the apple cultivars studied (tab. 4).

The biopreparations also had a positive effect on the degree of mycorrhizal association in the roots of 'Topaz' and 'Ariwa' apple trees. The use in the experiment of the biofertilizer Micosat F increased to the greatest extent the formation of mycorrhizal structures in the roots of the apple cultivar 'Ariwa'. The biopreparations Humus UP and Humus Active and Aktywit PM contributed to increasing mycorrhizal frequency in the roots of apple trees of the cultivar 'Topaz', compared to the control trees (Control 0) and those fertilized with NPK (tab. 4). This relationship was also observed when comparing the applied biopreparations irrespective of the cultivar. The application of the biopreparation Micosat F, biostimulant Humus UP, and Humus Active and Aktywit PM stimulated, to the greatest extent, the formation of mycorrhizal structures in the tree roots of the apple cultivars studied (tab. 4).

DISCUSSION

There was no significant impact (regardless of the cultivar) of the applied bioproducts on the growth of the aboveground parts of the apple trees, even though the bioproducts selected for the individual cultivars, Vinassa and Humus UP for 'Topaz' and Tytanit for 'Ariwa', showed a beneficial effect on the plants. These results differ from those obtained by other authors. Mehta and Bharat [2013] studied the effect of native mycorrhizal fungi of the genus *Glomus* spp. in container-based cultivation of apple trees planted in a soil affected by the replant disease. They observed that inoculation of apple trees with the isolate AMFS-2 of the mycorrhizal fungus Glomus fascicu*latum* contributed to an increase in plant height, trunk diameter, length of internodes, leaf surface area, fresh and dry weight of shoots and roots, compared to the non-inoculated trees. Positive effects of the bioproducts used in our experiment had been reported by Grzyb et al. [2014], who demonstrated a beneficial after-effect of the use of bioproducts in nursery production on the subsequent vegetative growth and yielding of apple trees under orchard conditions. They found that the products Humus Active + Aktywit PM, BF Quality, BF Amin, Vinassa and Tytanit

used in the nursery stimulated the growth and yielding of apple tree cultivars 'Topaz' and 'Ariwa' after they had been planted in the orchard. Gastoł and Domagała-Świątkiewicz [2015] evaluated the effects of various biofertilizers (granular and liquid mycorrhizal inocula, extracts of marine algae and from organic soils) on the growth of 'Topaz' apple trees grafted onto M26 rootstocks, growing in an orchard in a soil with the replant disease. They observed that the strongest growth was shown by the trees inoculated with mycorrhizal preparations MicoPlant M and MicoPlant S. Grzyb et al. [2013] studied the quality of maiden apple and sour cherry trees fertilized, in an organic nursery, with the granular fertilizer Florovit. They found that the applied organic fertilizer influenced the increase in tree height, the number of lateral shoots, and the trunk cross-sectional area (TCSA).

The applied bioproducts had a positive impact on the growth characteristics of the roots of plants the tested apple cultivars. The results showed that the biostimulator Humus UP and also Humus Active and Aktywit PM had a beneficial effect on the morphological characteristics of the root system. These results are also confirmed by other researchers. Beneficial effects of organic fertilizers on the growth of the root system were observed by Polverigiani et al. [2014a], who studied the rate of release of minerals, shoot growth, nutritional status, and biomass and morphology of the roots after the application of organic fertilizers in the cultivation of apple trees grafted onto M9 rootstocks. They found that the use of a preparation consisting of waste from wine-growing increased the length of the roots of apple trees. Polverigiani et al. [2014b] studied the growth of the root system of M9 apple rootstocks in a replanted soil taken from five sites in Central Europe. Their research showed that apple trees growing in soils that were wasteland (fallow) showed the highest dry mass of roots, and that the longest length of roots was shown by trees growing in a replant soil. These results are consistent with previous results/research on the impact of bioproducts on root growth and mycorrhizal frequency in the roots of two apple cultivars 'Topaz' and 'Ariwa' in the field [Derkowska et al. 2014]. Inoculation of the roots with the biopreparation Micosat F (containing an inoculum consisting of five species of mycorrhizal fungi) contributed to an increase in the number of root tips in trees of the cultivar 'Topaz'.

The biopreparations had a positive impact on the colonization of the roots of the tested apple cultivars by arbuscular mycorrhizal fungi. Micosat F exerted the greatest beneficial influence on the formation of mycorrhizae in the roots of apple trees of the cultivars studied. A positive effect of mycorrhizal inoculation of the roots of apple trees was also shown by Gastoł and Domagała-Świątkiewicz [2015], who studied the effects of granular and liquid mycorrhizal inocula, extracts from marine algae and from organic soils on the growth of 'Topaz' apple trees grafted onto M26 rootstocks, growing in an orchard with the replant disease present. The apple tree roots inoculated with mycorrhizal preparations in liquid form were reported to show more intense formation of mycorrhizal structures, compared to the roots of plants inoculated with solid mycorrhizal substrates in granular form. All the mycorrhizal preparations contributed to an increase in the degree of colonization of roots by AMF fungi. Meyer et al. [2015] studied the effect of conventional and organic methods of growing 'Cripps Pink' apple trees, grafted onto M7 rootstocks, on the presence of mycorrhizal fungi in the roots of plants and in the soil. They observed that in the organic cultivation of apple the roots were colonized to a greater extent than the roots of apple trees grown by conventional methods, and that they were characterized by a higher number of spores in the soil. Our previous studies had also suggested a beneficial effect of the applied biofertilizers on the formation of mycorrhizal structures in the roots. Derkowska et al. [2014], while studying the effects of bioproducts on the growth of the root system and the degree of mycorrhizal association in the roots of two cultivars of apple trees growing in the field, had found that treatments of 'Topaz' and 'Ariwa' apple trees with the biopreparations Micosat F, Humus UP, Humus Active + Aktywit PM, BioFeed Amin, Vinassa, Florovit Eko and with the organic fertilizer Florovit Pro Natura, exerted a positive impact on tree growth and mycorrhizal frequency in the roots of those apple trees.

Studies on the biodiversity of beneficial soil microorganisms in fruit crops bring new knowledge, with the possibility of it being applied in horticultural practice. The development of environmentally friendly technologies for the growing of fruit plants will contribute to improvements in fruit quality and fertility of arable and degraded soils in Poland.

CONCLUSIONS

Among the bioproducts tested, the greatest influence on the growth and occurrence of mycorrhizal fungi in the roots of trees of two apple cultivars 'Topaz' and 'Ariwa' grown under greenhouse conditions was exerted by Humus UP, Humus Active and Aktywit PM, and Micosat F. None of the biopreparations used, regardless of the cultivar, produced a clearly beneficial effect on the growth of the aboveground parts of apple trees. Application of Humus UP, Humus Active and Aktywit PM has a beneficial effect on the morphological characteristics of the root system of apple trees. Application of the mycorrhizal preparation Micosat F has a positive influence on the formation of mycorrhizae in the roots of 'Topaz' and 'Ariwa' apple trees. The beneficial effects of the tested preparations on tree growth and the degree of mycorrhizal association in the roots of apple trees, as well as field studies in this area, will contribute to their implementation into fruit-growing practice, restrictions on the use of chemical means of production, and consequently to the protection of the environment and human health. Long-term use of biofertilizers, organic mulches and microbiologically enriched composts has a positive influence on the growth and presence of mycorrhizal fungi in the roots of apple trees.

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REFERENCES

- Arsenault, J.L., Poulcur, S., Messier, C., Guay, R. (1995). WIN-RHIZO a root-measuring system with a unique overlap correction method. HortScience, 30, 906.
- Bhattacharjee, R., Dey, U. (2014). Biofertilizer, a way towards organic agriculture: A review. Afr. J. Microbiol. Res., 8(24), 2332–2342. DOI: 10.5897/AJMR2013.6374.
- Cameron, D.D. (2010). Arbuscular mycorrhizal fungi as (agro)ecosystem engineers. Plant Soil, 333, 1–5. DOI: 10.1007/s11104-010-0361-y.
- Derkowska, E., Sas Paszt, L., Harbuzov, A., Trzciński, P., Bogumił, A. (2014). The effect of biopreparations on root growth and microbiol activity in the rhizosphere of apple trees. Acta Sci. Pol. Hortorum Cultus, 13(6), 127–137.
- Derkowska, E., Sas Paszt, L., Dyki, B., Sumorok, B. (2015a). Assessment of mycorrhizal frequency in the roots of fruit plants using different dyes. Adv. Microbiol., 5(1), 54–64. DOI: 10.4236/aim.2015.51006.
- Derkowska, E., Sas Paszt, L., Trzciński, P., Przybył, M., Weszczak, K. (2015b). Influence of biofertilizers on plant growth and rhizosphere microbiology of greenhouse-grown strawberry cultivars. Acta Sci. Pol. Hortorum Cultus, 14(6), 83–96.
- Gąstoł, M., Domagała-Świątkiewicz, I. (2015). Mycorrhizal inoculation of apple in replant soils – enhanced tree growth and mineral nutrient status. Acta Sci. Pol. Hortorum Cultus, 14(4), 17–37.
- Grzyb, Z.S., Piotrowski W., Sas Paszt, L., Pąśko, M. (2013). Badania wstępne nad wpływem różnych biopreparatów na zmiany odczynu i zawartość składników w glebie i liściach okulantów jabłoni i wiśni. J. Res. Appli. Agric. Eng., 58 (3), 198–203.
- Grzyb, Z.S., Piotrowski, W., Sas Paszt, L. 2014. Treatments comparison of mineral and bio fertilizers in the apple and sour cherry organic nursery. J. Life Sci., 8(11), 889–898.
- Hazard, C., Gosling, P., van der Gast, C., Mitchell, D., Doohan, F., Bending, G. (2013). The role of local environment and geographical distance in determining community composition of arbuscular mycorrhizal fungi at the landscape scale. ISME J., 7, 498–508. DOI: 10.1038/ismej.2012.127.
- http://www2.dijon.inra.fr/mychintec/Mycocalc-prg/ /MYCOCALC.EXE

- Jansa, J., Erb, A., Oberholzer, H.-R., Smilauer, P., Egli, S. (2014). Soil and geography are more important determinants of indigenous arbuscular mycorrhizal communities than management practices in Swiss agricultural soils. Mol. Ecol., 23, 2118–2135. DOI: 10.1111/mec.12706.
- Mehta, P., Bharat, N.K. (2013). Effect of indigenous arbuscular – mycorrhiza (*Glomus* spp.) on apple (*Malus domestica*) seedlings grown in replant disease soil. Ind. J. Agric. Sci., 83(11), 1173–1178.
- Meyer, A.H., Wooldridge, J., Dames, J.F. (2015). Effect of conventional and organic orchard floor management practices on arbuscular mycorrhizal fungi in a 'Cripp's Pink'/M7 apple orchard soil. Agric. Ecos. Envir., 213, 114–120. http://dx.doi.org/10.1016/j.agee.2015.07.026.
- Mishra, P., Dash, D. (2014). Rejuvenation of Biofertilizer for Sustainable Agriculture and Economic Development. (SAED) Consilience J. Sust. Develop., 11(1), 41–61. http://dx.doi.org/10.7916/D8FQ9W9H.
- Polverigiani, S., Kelderer, M., Lardschneider, E., Neri, D. (2014a). Organic wastes use in horticulture: influences on nutrient supply and apple tree growth. Int. J. Plant Soil Sci., 3(4), 358–371. DOI: 10.9734/JJPSS/2014/8351.
- Polverigiani, S., Kelderer, M., Neri, D. (2014b). Growth of 'M9' apple root in five Central Europe replanted soils. Plant Root, 8, 55–63. DOI: 10.3117/plantroot.8.55.
- Quilty, J.R., Cattle, S.R. (2011). Use and understanding of organic amendments in Australian agriculture: a review. Soil Res., 49,1–26. http://dx.doi.org/10.1071/SR10059.
- Raviv, M. (2010). The use of mycorrhiza in organicallygrown crops under semi arid conditions: a review of benefits, constraints and future challenges. Symbiosis, 52, 65–74. DOI: 10.1007/s13199-010-0089-8.

- Trouvelot, A., Kough, J.L., Gianinazzi-Pearson, V., 1986. Mesure du taux de mycorhization VA d'un systeme radiculaire. Recherche de methods d'estimation ayant une signification fonctionnelle. In: Physiological and genetical aspects of mycorrhizae, Gianinazzi-Pearson, Gianinazzi, S. (eds). INRA, Paris, 217–221.
- Tuck, S., Winqvist, C., Mota, F., Ahnström, J., Turnbull, L.A., Bengtsson, J. (2014). Land-use intensity and the effects of organic farming on biodiversity: a hierarchical meta-analysis. J. Appl. Ecol., 51, 746–755. DOI: 10.1111/1365-2664.12219.
- Turrini, A., Giovannetti, M. (2012). Arbuscular mycorrhizal fungi in national parks, nature reserves and protected areas worldwide: a strategic perspective for their in situ conservation. Mycorrhiza, 22, 81–97. DOI: 10.1007/s00572-011-0419-6.
- Verbruggen, E., van der Heijden, M., Weedon, J., Kowalchuk, G., Röling, W. (2012). Community assembly, species richness and nestedness of arbuscular mycorrhizal fungi in agricultural soils. Mol. Ecol., 21, 2341– 2353. DOI: 10.1111/j.1365-294X.2012.05534.x.
- Wally, O.D., Critchley, A., Hiltz, D., Craigie, J., Han, X., Zaharia, L.I., Abrams, S., Prithiviraj, B., (2013). Regulation of phytohormone biosynthesis and accumulation in *Arabidopsis* following treatment with commercial extract from the marine macroalga *Ascophyllum nodosum*. J. Plant Growth Regul., 32, 324–339. http://dx.doi.org/10.1007/s00344-012-9301-9.
- Willer, H., Kilcher, L. (2011). The World of Organic Agriculture – Statistics and Emerging Trends 2011. IFOAM and FiBL, Bonn. https://shop.fibl.org/fileadmin/ documents/shop/1546-organic-world-2011.pdf#page=26.