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The Handling of Composted Onion Waste in the Form of Substrates for the Proliferation of the *Trichoderma* sp.

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1. Introduction

Nowadays we can observe an increasingly dynamic development of the cultivation of crops and vegetables, which results in growing amounts of waste. The best way to handle this waste is to re-enter it into the soil environment (Regulation of the Minister of Environment of 20 January 2015), which can be done after proper preparation. The composting process seems to be the safest to the environment and the cheapest method of organic waste handling. The end product of the process is compost, which can be used as an organic fertiliser due to its valuable fertilising properties (Malińska et al. 2014, Starzyk et al. 2015, Czekała et al. 2016, Czekała et al. 2017).

A proper composting process results in compost which contains not only humic substances but also B vitamins and phytohormones. Apart from that, a temperature of 55-75°C at the thermophilic phase makes compost free from animal pathogens and weed seeds (Sánchez et al. 2017).

The assessment of adequacy of the compositing process involves not only the measurement of physical and chemical properties but also the biomass, the count of microorganisms and the enzymatic activity in the compost. These parameters are significant determinants of the degree of mineralisation of organic matter in the composted waste, as they indicate its maturity, stability and possibility of further use (Awasthi et al. 2018, Tiquia 2005).

The product of the composting process may be a good carrier of microorganisms promoting the growth and development of plants, e.g. fungi of the *Trichoderma* sp. genus (Wolna-Maruwka et al. 2016, Wolna-Maruwka et al. 2017). When these microorganisms enter soil, they become adsorbed and grow through the composted organic matter. As a result, they have a greater chance to survive the confrontation with the autochthonous soil microflora. Apart from that, the carrier itself (compost) is a nutrient reservoir.

As results from the studies by Cavalcante et al. (2008), Kancelista et al. (2013) and Smolińska et al. (2014), the adaptive abilities of *Trichoderma* sp. are related both with the type of substrate used as a carrier and the type of strain. The growth and development of these microorganisms are considerably influenced by the pH, humidity and temperature of the substrate as well as the source of carbon and nitrogen.

Moulds of the *Trichoderma* genus are very popular in agriculture because they support the growth of plants and protect them from pathogens (*Fusarium, Alternaria, Rhizoctonia. Botrytis, Sclerotinia*, etc.). They are considered to be biological control agents (BCAs) because of their mycoparasitism and capacity to produce antibiotics and siderophores. Additionally, they are classified as plant growth promoting fungi (PGPF) because they produce phytohormones, provide nutrients to plants and induce systemic resistance (El-Komy et al. 2015, Gajera et al. 2016, Sharma et al. 2017).

What also speaks in favour of applying strains of these microorganisms into soil to promote the growth of plants is the fact that they are resistant to pesticides, phenols and heavy metals. They can be easily isolated from the soil environment. Apart from that, they grow quickly and spore profusely in artificial cultures (Smolińska et al. 2014, Steyaert et al. 2010). There are very few microbial formulations based on *Trichoderma* sp. strains on the Polish market. Only one of them contains Polish strains of these fungi. Microorganisms develop best and exhibit the highest activity in the climate they come from. Therefore, it seems important to develop fertiliser preparations consisting of native Polish strains, which will be applied into soil on an organic carrier, i.e. compost. This will make them more resistant to adverse environmental factors.

The aim of the study was to assess the phytosanitary properties of two strains of *Trichoderma* sp. and their ability to grow and develop in mature compost made from onion waste, rye straw and pig manure.

2. Material and methods

2.1. Experimental design

The experiment was conducted in a laboratory at the Department of General and Environmental Microbiology, Poznań University of Life Sciences, Poland, as part of the project No. UDA-POIG.01.03.01-00-129/09-09 titled 'Polish *Trichoderma* Strains in Plant Protection and Organic Waste Handling', co-financed by the European Union from the European Regional Development Fund under the Innovative Economy Operational Programme.

Mature compost (onion waste compost) made from onion husk (553.8 kg fresh weight), rye straw (21.4 kg fresh weight) and pig manure (21.7 kg) was used in the experiment (Tab. 1).

Table 1. Physicochemical state of compost used in experiments (beginning of experiments)

Characteristic of compost	pH_{H2O}	humidity (%)	C $g{\cdot}kg^{-1}$ d.m.	N $g{\cdot}kg^{-1}$d.m.
Onion compost	8.81	45.11	389.23	24.89

The compost was collected from a 20-tonne prism. It was divided into four equal parts, 10 kg each, and placed in containers with a capacity of 18.8 l. Next, the compost was inoculated with two *Trichoderma* sp. strains, i.e. *T. atroviride* – T1 and *T. harzianum* – T3. In consequence, there were four experimental variants, each in two replicates: 1 – the control sample; onion waste compost (control), 2 – onion waste compost inoculated with *T. atroviride* strain T1 (variant T1), 3 – onion waste compost inoculated with *T. harzianum* strain T3 (variant T3), 4 – onion waste compost inoculated with *T. atroviride* strain T1 and *T. harzianum* strain T3 (variant T1+T3). The *Trichoderma* sp. inoculums came from the collection of strains of the Institute of Horticulture in Skierniewice, Poland. Each of them was applied in the form of conidial spores, at a density of 10^6 per g of fresh mass of compost. The count of spores was measured with a haemocytometer, under a light microscope (Zeiss).

Before the experiment an in vitro test was carried out to exclude antagonism between the *Trichoderma* isolates (Mańka 1974) used in the T1+T3 variant.

The experiment was carried out under controlled conditions in a thermostat, at a constant ambient temperature of 22°C. There were three terms of microbiological and enzymatic analyses: 1st – at the beginning of the experiment, 2nd – on the 30th day of the experiment, 3rd – on the 60th day of the experiment.

2.2. Microbiological analysis

The scope of experiments comprised determination (in five replications) of the total count of moulds and *Trichoderma* sp. The groups of microorganisms were cultured according to the plate method on solid substrates, using appropriate dilutions of soil solutions, expressed as $CFU{\cdot}g^{-1}$ of compost dry matter.

The count of moulds was determined on a medium prepared according to rose bengal agar (Sigma Aldrich) with aureomycin added. Plates were incubated for 6 days at a temperature of 25°C. Colonies of moulds isolated from composts at the end of the thermophilic phase were inoculated to a PDA substrate (Sigma Aldrich) on the day of their inoculation with *Trichoderma* sp. isolates. Next, their systematic position was determined according to mycological keys and the percentage of individual genera in the entire population was calculated (Domsch et al. 1993).

The count of *Trichoderma* sp. strains was determined with the plate method, on a modified rose bengal agar (Sigma Aldrich) with chloramphenicol, streptomycin, metalaxyl and PCNB (pentachloronitrobenzene) added. The plates were exposed to visible light and incubated for 7 days at a temperature of 24°C. In order to confirm the systematic position of *Trichoderma* sp. in the *Trichoderma harzianum* or *Trichoderma atroviride* species the colonies were inoculated to the PDA substrate (Sigma Aldrich). They were initially identified with a microscope. Next, the identification was confirmed by means of in situ hybridisation (FISH), modified according to Amann et al. (1990), where 4% PFA (paraformaldehyde), 0.5% Triton solution, alcohol series (70%, 80%, 96%), 70% formamide solution and two probes, marked at end 3' with marker Cy3 (ACT CCC AAA CCC AAT GTG AA and ATA CCA AAC TGT TGC CTCGG) were applied (Siddiquee et al. 2010).

The rating scale developed by Mańka (1974) was used to determine the type of interactions occurring between strains of autochthonous moulds isolated from the composts at the end of the thermophilic phase and *Trichoderma* sp. isolated applied in the research. The following elements were taken into consideration: the degree to which one colony surrounded another, the width of the inhibitory zone and growth inhibition. A positive result should be interpreted as an effect limiting the development of a particular autochthon (test fungus) by a selected *Trichoderma* sp. isolate (antagonist).

Before the experiment was set up, fungi had been proliferated on a PDA medium (Potato Dextrose Agar). Next, a sterile cork borer was used to cut discs of cultures (10 mm in diameter). The discs were transferred to the PDA medium and spaced at 30 mm from each other. Every day, for 8 days the mycelium diameter was measured in the dual-organism cultures and the type of interaction between the fungi was determined. The cultures were incubated at a temperature of 24°C. Single-organism cultures prepared in an analogical manner were used as control samples. All tests were replicated five times.

2.3. Trends of physicochemical and enzymatic parameters

The dehydrogenase activity (DHA) was determined with the method developed by Camiña (1998) with minor modifications. The compost (5g) was incubated for 24 h with 2, 3, 5-triphenyltetrazolium chloride (TTC) at 30°C, pH 7.4. Triphenylformazan (TPF) was produced, extracted with 96% ethanol and measured spectrophotometrically at a wavelength of 485 nm. The dehydrogenase activity was expressed as μg TPF · g^{-1} DM of compost · $24h^{-1}$.

The chemical analysis of composts was conducted in the laboratory of the Department of Agronomy. The content of carbon was determined with the

Tiurin method. The content of total nitrogen in the fresh weight of composts was measured with the Kjeldahl method by means of a Kjeltec 2200 System analyser (FOSS Tecator). The pH values of compost samples were determined in double distilled water. (pH_{H2O}). The content of dry weight of composts was measured by drying and weighing at a temperature of 105°C in the laboratory of the Institute of Biosystem Engineering.

2.4. Statistical analysis

Statistical analyses were conducted by means of Statistica 12.0 software (StatSoft Inc. 2012). We used two-way analysis of variance to determine the significance of variation in the number of moulds under analysis, depending on the compost combination and term of analysis. Homogeneous subsets of means were identified by means of Tukey´s test at a significance level of $p = 0.05$.

Principal Component Analysis (PCA) was used to illustrate the dependence between the number of microorganisms, DHA activity, pH and moisture content. Simple regression analysis was applied for evaluating the optimal microorganisms growth and DHA activity.

3. Results and discussion

Apart from bacteria and actinobacteria, moulds also take part in the composting process. These microorganisms play a key role in the degradation of organic matter and the formation of humic compounds. Thanks to their activity it is possible to obtain compost of high fertilising value from organic waste, which is often difficult to handle (Wolna-Maruwka et al. 2016, Van Fan et al. 2018).

Before applying *Trichoderma* sp. strains into composts the analysis of the count of moulds showed that there were statistically significant differences already at the first term of analyses. The count ranged from 150 to 340 $\cdot 10^3$ CFU g d.m. of compost, depending on the experimental variant (Fig. 1).

At the subsequent terms of analyses the count of moulds decreased significantly in the variants with *Trichoderma* sp. isolates, especially in the variant with the *T. harzianum* (T3) isolate. The trend based on the daily decrease in the count of moulds fitted the quadratic regression model below (Equ. 1).

Equ. 1. Moulds (combination T3):

$$M= 91.5D^2 - 511.5M + 760$$
$$R^2 = 0.98$$

where:
M – Molds,
D – Days,
R^2 – Coefficient R squared.

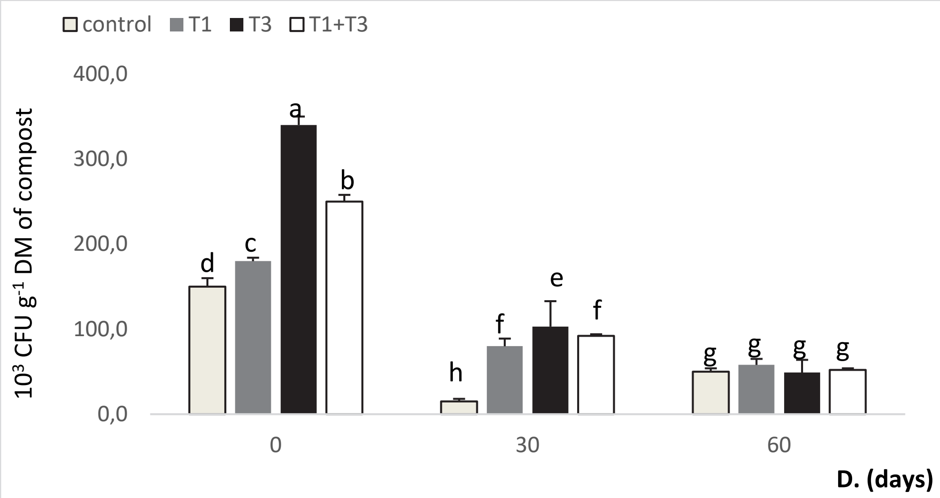

Fig. 1. The changes of total number of moulds in onion composts

Explanation: Means followed by the same letters do not differ significantly at $p = 0.05$; Combination: T1 – onion waste compost inoculated with strain T1, T3 – onion waste compost inoculated with strain T3, T1+T3 – onion waste compost inoculated with strain T1 and T3.

This effect may have been caused both by the addition of *Trichoderma* sp. isolates and by changes in the compost pH and moisture content (Fig. 2), which was confirmed by the principal component analysis (PCA) (Fig. 3).

As results from the study by Steyaert et al. (2010), moulds produce the most spores and develop best when pH ranges from 2.8 to 5.2. In our study the pH of the composts was alkaline, which may have been caused by the gradual decrease in count of moulds as the experiment was incubated.

The substrate moisture is also an important parameter affecting the growth and development of moulds. The study by Natywa et al. (2014) showed that the survival and activity of microorganisms decreased along with the moisture content of the substrate. Moulds became inactive when the moisture content of the environment decreased to 15%. On the other hand, according to Hassen et al. (2001), the development of moulds in compost is inhibited by other factors than pH, humidity or temperature. The development of fungi in compost may be inhibited by microbial antagonism processes, including antibiosis.

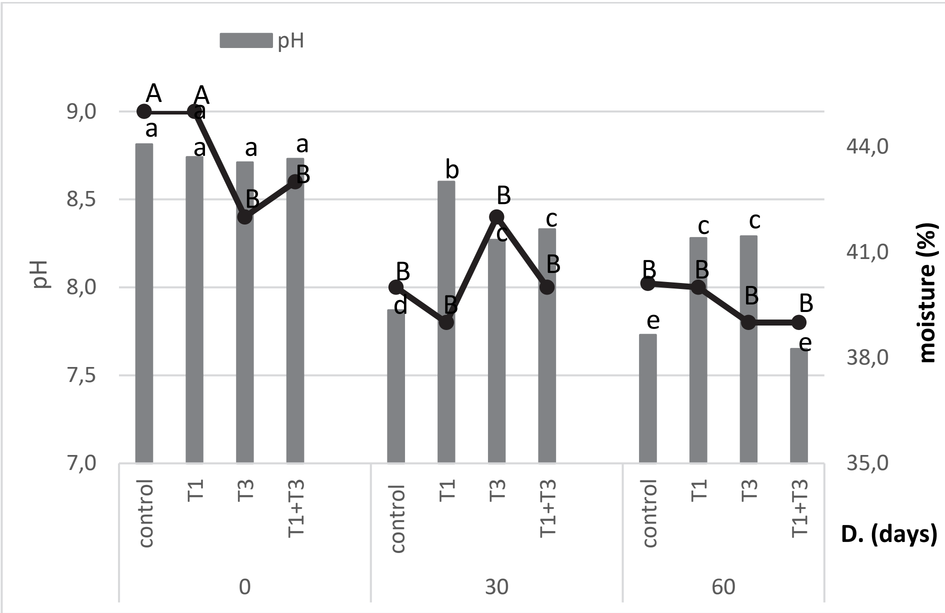

Fig. 2. The changes of pH and moisture value in onion composts

Explanation: Means followed by the same letters do not differ significantly at $p = 0.05$; Combination: OT1 – onion waste compost inoculated with strain T1, OT3 – onion waste compost inoculated with strain T3, OT1+OT3 – onion waste compost inoculated with strain T1 and T3.

This thesis is confirmed by the results of biotic tests shown in Table 2. As can be seen, the *Trichoderma* sp. strains applied into the composts exhibited in vitro antagonism against the autochthonous fungi isolated from the onion compost at the beginning of the experiment.

The results of mycological analyses in Table 3 show that *Penicillium* (31-33%) and *Aspergillus* sp. (4-14%) were the dominant species in the compost. The presence of *Alternaria* sp., *Paecilomyces variotti*, *Rhizopus nigricans* and *Mucor* sp. was also detected. These observations are in agreement with the results of the research by Wolna-Maruwka et al. (2016), where moulds of the *Penicillium* genus were the predominant microflora in compost made from onion waste, wheat straw and pig manure.

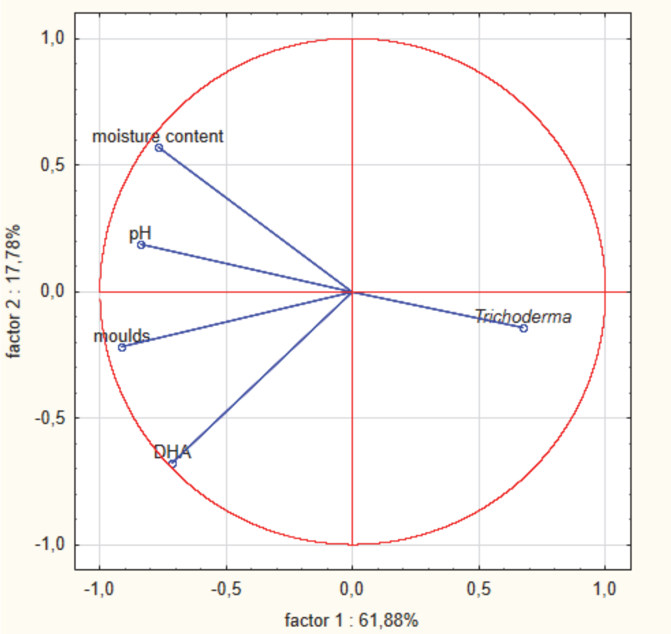

Fig. 3. Dependences between the number of moulds, *Trichoderma* sp, DHA, pH value and humidity (%) of applied in the experimental compost combinations at consecutive terms of analyses (PCA)

The biotic tests (Tab. 2, Phot. 1) showed that the *T. atroviride* strain (T1) exhibited stronger antagonism than the T3 isolate.

Table 2. Results of biotic test in dual cultures

Isolate	*Penicillum* sp. (1)	*Penicillum* sp. (2)	*Aspergillus* sp.	*Aspergillus niger*	*Alternaria* sp.	*Paecilomyces variotti*	*Rhizopus nigricans*	*Mucor* sp.
T1	-6	+8	+4	+6	+6	+6	+8	+6
T3	-6	-4	+4	+4	+6	+4	+6	+6

Explanation: A positive result means that the *Trichoderma* sp. isolate inhibited the growth of the autochthonous colony; 0 – no inhibition, 8 – total inhibition.

Table 3. Isolates of moulds isolated from the compost before inoculation

Isolate	Percentage content (%)
Penicillum sp. (1)	33
Penicillium sp. (2)	31
Aspergillus sp.	14
Aspergillus niger	4
Alternaria sp.	4
Paecilomyces variotti	4
Rhizopus nigricans	5
Mucor sp.	5

Photo 1. Antagonistic relationship between *Trichoderma atroviride* – T1 and *Penicillium* sp. (2)

According to Dłużniewska (2004) and Gajera et al. (2016), *Trichoderma* sp. have numerous antagonistic properties. They are characterised by rapid growth, abundant sporulation, the ability to exist on various substrates, the ability to produce fungitoxic substances, the ease of use of available organic matter and inorganic compounds, and strong parasitic properties resulting from the production of a wide range of antibiotics (alamethicin, tricholine, peptaibols, viridins, gliovirins, gliotoxins, glisoprenin, heptelidic acid, α-pyrrole) and enzymes (cellulase, hemicellulase, xylanase, pectinase, β-1,3-glucanase, chitinase and protease).

According to Perek et al. (2013), the combination of enzymes and antibiotics produced by microorganisms may give a higher level of antagonism than the activity of antibiotics or enzymes alone. The initial degradation of the cell wall by enzymes can facilitate the penetration of antibiotics into the cells of other microorganisms.

However, the interaction tests showed that both *Trichoderma* sp. isolates were susceptible to *Penicillium* sp. strains. According to Wawrzyniak and Waśkiewicz (2014), it resulted from the production of ochratoxin A and citrinin by some *Penicillium* species.

In our research, not only was the count of moulds reduced at the end of the experiment, but also the dehydrogenase activity decreased gradually (Fig. 4). According to Barren et al. (2008), the dehydrogenase activity can be used to monitor the composting process and to assess compost maturity and stability.

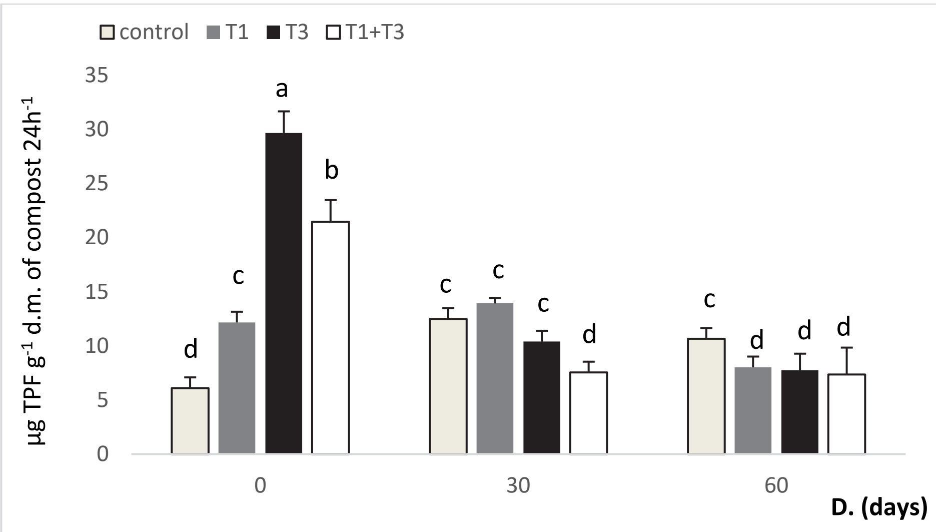

Fig. 4. The changes of dehydrogenase activity in onion composts

Explanation: Means followed by the same letters do not differ significantly at $p = 0.05$; Combination: T1 – onion waste compost inoculated with strain T1, T3 – onion waste compost inoculated with strain T3, T1+T3 – onion waste compost inoculated with strain T1 and T3.

However, this enzyme reaction was mainly observed in the variants where *Trichoderma* sp. isolates were used. Like the count of moulds, especially in the compost inoculated with the T3 isolate, it assumed the trend that matches the quadratic regression model (Equ. 2).

Equ. 2. Dehydrogenase (combination T3):

$$DHA= 8.3113D^2 – 44.2DHA + 65.539$$
$$R^2 = 0.99$$

where:
DHA – Dehydrogenase Activity,
D – Days,
R^2 – Coefficient R squared.

The principal component analysis (PCA) confirmed the negative relation between the count of *Trichoderma* sp. and the dehydrogenase activity. It also indicated the positive correlation between the dehydrogenase activity (DHA) with the pH of composts (Fig. 3).

The reduced dehydrogenase activity during the experiment shows that the active decomposition of organic matter decreased and the compost reached maturity. According to Tiquia (2005), the DHA in mature compost ranges from 12 to 39 μg TPF·g^{-1} d.m. of compost. In our study at the end of the experiment (3^{rd} term), the DHA in the composts amounted to 7.33-10.64 μg TPF·g^{-1} d.m. of compost, that suggest that obtained composts were fully stable and mature.According to the guidelines presented by the abovementioned author, the composts were fully stable and mature.

At the beginning of the experiment the compost samples were analysed mycologically (Tab. 3) for the occurrence of autochthonous *Trichoderma* sp. fungi. As they were not found in any of the experimental variants, it considerably facilitated the control of the survivability of the isolates applied into the composts. 30 days after the application of the strains the highest count of *Trichoderma* sp. was found in the variant containing both T1 and T3 isolates, whereas the *T. atroviride* isolate (variant T1) exhibited the poorest adaptive properties (Tab. 4).

Table 4. The changes of *Trichoderma* sp. (10^2 CFU g^{-1} DM) in onion composts

Combination	D. (day)		
	0 – beginning of experiment	30	60
control	0.00	0.00	0.00
T1	0.00	4.21^e	14.21^c
T3	0.00	9.11^d	24.55^a
T1+T3	0.00	10.01^d	20.11^b

Explanation: Means followed by the same letters do not differ significantly at $p = 0.05$; Combination: T1 – onion waste compost inoculated with strain T1, T3 – onion waste compost inoculated with strain T3, T1+T3 – onion waste compost inoculated with strain T1 and T3.

Within 30 days following the inoculation of the composts the count of the applied *Trichoderma* isolates was reduced by 4 orders of magnitude, from the initial spore density of 10^6 to 10^2 CFU. There was a similar reduction observed in the study by Wolna-Maruwka et al. (2016).

As results from the observations, on the 30th day of the experiment the *Trichoderma* isolates were adapted to the unfavourable environmental conditions, such as the high pH (Fig. 2) and competition with the autochthonous compost microflora (Tab. 2).

According to Kredics et al. (2003), the optimal pH for the growth and development of *Trichoderma* sp. is 4. This finding was confirmed by the statistical analysis (PCA), which indicated a negative relation between the count of these microorganisms and the pH value of the composts (Fig. 3).

However, Vimala Kumari et al. (2014) found that *Trichoderma* isolates collected from agricultural soil were capable of proliferation in an alkaline substrate (pH 7-10).

On the 60th day of compost incubation, the strain survival phase finished and the reproduction phase began. The count of *Trichoderma* sp. increased by more than 100-150%. It is most likely that the variation in the proliferation of the isolates resulted from their different sensitivity to the pH of the substrate. The smallest variation was noted in the variant inoculated with the T3 strain (Fig. 5).

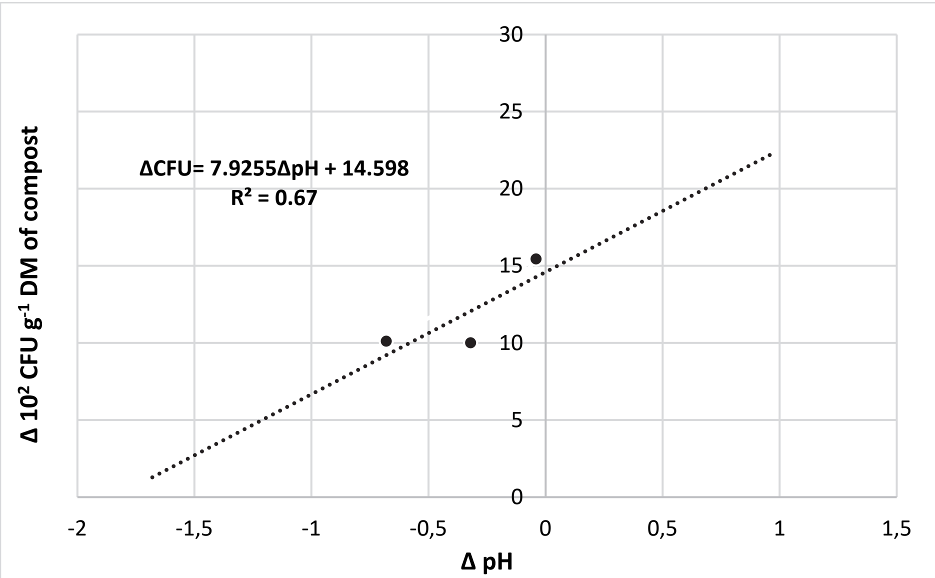

Fig. 5. The effect of the compost pH change during the propagation stage of *Trichoderma* strains on the change of *Trichoderma's* count

As results from the quadratic regression model, the *T. harzianum* strain (variant T3) exhibited the weakest reaction to the change in the compost pH on the 60th day of the experiment. The quadratic regression model shows that the pH decrease of -0.04 from the initial 8.33 resulted in the maximum count of $15.44 \cdot 10^2$ CFU *T. harzianum* g^{-1} d.m. of compost. According to Yedid et al. (1999) and Howell (2003), fungi of the *Trichoderma* sp. genus can regulate the pH of the substrate by producing enzymes such as cellulases, pectinases, xylases and proteases, which increase the pH value. On the other hand, they can reduce the pH of the substrate by producing organic acids.

4. Conclusions

1. The strongest trend in the statistically significant reduction of the count of moulds and dehydrogenase activity to the level showing full maturity of the compost was observed in the variant with the *T. harzianum* isolate (T3).
2. The research showed that *Penicillum* sp. and *Aspergillus* sp. were the dominant strains in the onion waste compost. Apart from that, the antagonism test proved that the *Trichoderma* sp. isolates inhibited the growth and development of most of the autochthonous moulds isolated in the analysis.
3. Both the pH and moisture content of the composts significantly influenced changes in the count of fungi as well as the adaptive properties and the proliferation of the *Trichoderma* sp. isolates applied into the composts.
4. The *T. harzianum* strain (T3) was the least sensitive to the environmental conditions, which resulted in its highest count. The *T. atroviride* isolate (T1) exhibited stronger phytosanitary properties.
5. The research showed that the alkaline onion waste compost enabled the growth and development of antagonistic fungi of the *Trichoderma* spp. genus and that it was a good way of handling nuisance waste.

The research was conducted as part of the project of the National Centre for Research and Development, No. UDA-POIG.01.03.01-00-129/09-09 'Polish Strains of Trichoderma in Plant Protection and Organic Waste Handling'.

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Abstract

In spite of the alkaline reaction of onion waste composted together with rye straw and pig manure, these materials can be used as a substrate for the growth of moulds of the *Trichoderma* genus, with proven phytosanitary properties against plant pathogens. This hypothesis was verified in a laboratory experiment with four variants: a control sample and three variants with two *Trichoderma* sp. strains (*T. atroviride* T1 and *T. harzianum* T3). The isolates were entered into compost individually or simultaneously (T1 + T3). The research included microbial analyses, in which the survivability of *Trichoderma* sp. strains was monitored and the total count of moulds and the dehydrogenase activity were estimated. The analyses were made three times: before applying the strains into compost, one month and two months after the inoculation. Apart from that, the type of interaction between autochthonous fungi isolated from mature compost at the beginning of the experiment and the *Trichoderma* sp. strains was determined.

The count of moulds was most reduced and the lowest dehydrogenase activity was observed in the compost with the *T. harzianum* isolate. Apart from that, the strain also exhibited the strongest adaptive properties to the compost substrate. The *T. atroviride* isolate proved to be the more effective antagonist of autochthonous moulds isolated on the first day of the experiment, including fungi of the *Alternaria* genus. The pH and moisture content of the substrates proved to be significant factors affecting the growth, proliferation and the antagonistic properties of *Trichoderma* sp. isolates.

The simultaneous inoculation of compost with two *Trichoderma* sp. isolates resulted in the worst growth and proliferation of the *Trichoderma* sp. strains, although the two isolates were not antagonistic to each other in vitro.

Keywords:
compost, *Trichoderma*, physicochemical properties, fungi survival test

Zagospodarowanie przekompostowanych odpadów cebulowych w postaci podłoży do namnażania *Trichoderma* sp.

Streszczenie

Przekompostowane odpady cebulowe wraz ze słomą żytnią i obornikiem świńskim pomimo zasadowego odczynu stanowią podłoże umożliwiające wzrost grzybów pleśniowych z rodzaju *Trichoderma*, with proven phytosanitary properties against plant pathogens. Hipotezę tę zweryfikowano na podstawie doświadczenia przeprowadzonego w warunkach laboratoryjnych, w którym zastosowano cztery warianty doświadczalne: kontrolny oraz trzy z dodatkiem dwóch szczepów *Trichoderma* sp. (*T. atroviride* T1 oraz *T. harzianum* T3). Izolaty wprowadzano pojedynczo lub oba jednocześnie (T1+T3) do kompostu. Analizy mikrobiologiczne polegające na monitorowaniu przeżywalności wprowadzonych szczepów *Trichoderma* sp., oszacowaniu ogólnej liczby grzybów pleśniowych oraz aktywności dehydrogenaz przeprowadzono trzykrotnie: przed wprowadzeniem szczepów do kompostu, miesiąc oraz dwa miesiące po inokulacji. Dodatkowo określano rodzaj interakcji zachodzącej między autochtonicznymi grzybami wyizolowanymi z dojrzałego kompostu w momencie zakładania doświadczenia a zastosowanymi szczepami *Trichoderma* sp..

Wykazano, że największą redukcję grzybów pleśniowych oraz najniższą aktywność dehydrogenaz uzyskano w kompoście, do którego wprowadzono izolat *T. harzianum*. Niniejszy szczep wykazał ponadto najsilniejsze właściwości adaptacyjne w kompostowym podłożu. Izolat *T. atroviride* okazał się natomiast skuteczniejszym antagonistą wobec wyizolowanych w dniu zakładania doświadczenia autochtonicznych grzybów pleśniowych, w tym z rodzaju *Alternaria*. Wartość pH i poziom wilgotności podłoży okazały się istotnymi czynnikami warunkującymi wzrost, namnażanie, jak również właściwości antagonistyczne izolatów *Trichoderma* sp.

Stwierdzono, że wprowadzając do kompostu jednocześnie dwa izolaty *Trichoderma* sp. uzyskano najgorszy efekt wzrostu i namnażania się zastosowanych szczepów *Trichoderma* sp., pomimo, że warunkach in vitro izolaty nie oddziaływały na siebie antagonistycznie.

Słowa kluczowe:
kompost, *Trichoderma*, właściwości fizyczno-chemiczne, test przetrwania grzybów