Greenhouse Medium Enrichment with Composted Pig Slurry: Effect on the Rooting of *Pelargonium Peltatum Hort*. Cuttings and Development of Rhizosphere Microflora

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Abstract

The effect of a 25% addition of composted pig slurry on the rooting of *Pelargonium peltatum hort*. in a greenhouse medium consisting of perlite and highmoor peat (1:1 v/v) has been studied. The purpose of this paper is to define relationships between the rooting process and occurrence of microflora in the media without compost (control) and with the addition of that manure, as well as in the rhizosphere of the rooted plants. The development of whole bacterial communities determined by their biomass size (direct method) and the number of colony forming units (CFU) of actinomycetes and fungi (on selective media) were estimated.

The compost added to the greenhouse medium stimulated the rooting of geranium cuttings significantly, increasing their root length by almost 80% and root biomass by over 40%. It also affected a 35% increase in shoot biomass. Except propagative forms of fungi, the medium enrichment with compost simultaneously intensified microflora development, which appeared to be particularly evident in the case of actinomycetes, which belong to bacteria producing phytohormones; the number of these microorganisms in the rhizosphere of plants rooted on the medium with compost was twofold higher than on the medium alone.

Keywords: pig slurry compost, Pelargonium peltatum hort., cutting, rooting, rhizosphere microflora

Introduction

As in chemical fertilizers, the elements P and K in animal waste, pig slurry, occur in the form available to plants [6]. On the other hand, N contained in pig slurry stimulates plant yielding even more strongly than that introduced with manure [8]. On account of the high fertilizing value of that animal waste and the necessity to make it productive, a direct land application happens to be the most popular method for pig slurry disposal. In connection with a substantial increase of animal production for the last decades, it becomes necessary to apply slurry recurrently for a long time on the same fields [11]. Such practice, however, brings about ecological dangers on both local and global scales. A high content of floating parts in pig slurry and its ability to take the form of a gel sedimenting on the soil surface may cause a serious detriment to oxygen conditions in that environment [4]. The deficiency of oxygen at an increase in the level of inorganic N and available C leads to a significant emission of N₂O [10]. This is of particular importance in situations when agriculture actually generates about 70% of anthropogenic emissions of this gas [2]. It has been found

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that pig slurry application also causes an increase in CO_2 emissions [10, 11]. Therefore, direct land application of pig slurry contributes to an increase of CO_2 and N_2O concentration in the atmosphere and thereby to the intensification of the global greenhouse effect.

These unfavourable phenomena may be limited by composting, presenting a simple method of organic waste stabilisation in biofertilizers rich in nutrients required by plants [9]. A mixture of pig slurry and straw is frequently subjected to composting, which not only regulates the C:N ratio, but improves the mechanical structure of compost.

One of the possibilities of using composted pig slurry is its application as an addition to greenhouse media [12].

In our preliminary studies (unpublished data) it has been found that enrichment of a greenhouse medium with pig slurry has a positive influence on the general development of the geranium. The purpose of this work was to state the influence of biofertilizer on the rooting of geranium cuttings. The rate of the rooting of this plant cuttings as well as the abundance of expanding root systems are significantly regulated by phytohormones, which besides plants, can also be produced by plant growth-promoting bacteria occurring in their rhizosphere [11]. In view of this, the effect of compost on the development of microflora in the studied media has also been determined.

Materials and Methods

After drainage a pig slurry was composted for 8 months in a pile consisting of layers of a constant slurry fraction and rye straw added in a 20% amount of entire composted biomass volume.

The greenhouse medium for the rooting of plant cuttings was a mixture of highmoor peat and perlite in the volume ratio 1:1 enriched with 25% (v/v) pig slurry compost. A highmoor peat, pH 4.0, contained 52% of C, 0.2% N and 90% organic matter. Peat reaction was adjusted to pH of 6.5 by the addition of 4 g CaCO₃ per dm³ of peat. Pig slurry compost contained in mg per dm³: 553 N-NO₃, 39 N-NH₄, 500 P, 1990 K, 630 Ca 653 Mg and 359 Cl; the salinity of compost amounted 3.54 g KCl per dm³. As control medium a mixture of a highmoor peat (pH 6.5) and perlite (1:1 v/v) without compost addition was used.

Three-noded cuttings of *Pelargonium peltatum hort*. "Ville de Paris Lila" were rooted during one month at the turn of May-June. The degree of their rooting was evaluated on the basis of biometrical measurements length and biomass of the produced roots. For a more complete evaluation of the added compost influence on the growth of developing plants, the length and biomass of shoots were also determined.

Microbiological analyses of the materials used in the experiments were carried out when laying out the experiment (Term 0) and after rooting. The performed determinations concerned: 1. the total number and biomass of bacteria – by direct (microscopic) method [3]; 2. the number of colony forming units (CFU) of actinomycetes – on selective medium with chitin [14]; 3. the CFU number of fungi – using the modified Warcup method [7]. To

determine the rhizosphere effect when gathering plants, the following medium samples were taken: a) closely adhering to the roots (R_1) , b) remaining on the roots (R_2) , c) non-rhizosphere (NR).

The statistical evaluation of the results consisted of analysis of variance to determine the effect of compost addition to the medium on the rooting of geranium cuttings and on the development of microflora with reference to a rhizosphere effect.

Results and Discussion

Fertilization of greenhouse medium, besides physical factors, such as temperature [15], is significantly important for the rooting of plant cuttings and their further development.

The addition of compost to the medium significantly stimulated the rooting of geranium cuttings (Table 1). Roots of the cuttings growing on the medium with the addition of compost were 78% longer and their biomass was 44% higher in comparison to the cutting roots from the control medium. The addition of compost to the medium increased the shoot biomass increment of rooted cuttings, but had no effect on their length.

	Variable					
Treatment	Root length (cm)	Root weight (g)	Stem length (cm)	Stem weight (g)		
Medium without compost (control)	6.84	0.36	18.67	4.20		
Medium with compost	12.18**	0.52**	19.27 n.s.	5.67**		

Table. 1. Effect of composting pig slurry on rooting and growth of *Pelargonium peltatum hort*. cuttings.

** significant difference at $P \le 0.01$

n.s. non-significant difference at $P \leq 0.05$

The total number of bacteria in the compost in comparison to the greenhouse medium was almost twofold higher, amounting to about 30 x 10^9 and 17 x 10^9 g⁻¹ dry weight of the material, respectively (Table 2). After the rooting period, the number of bacteria ranged from about 27 x 109 in the medium closely adhering to the roots (R_1) to 18 x 10⁹ g⁻¹ in the non-rizosphere (NR) medium in the lack of a distinct differentiation as a result of compost addition to the medium. However, medium enrichment with compost caused bacterial biomass increase from 1.3 mg d. w. (control) to 1.8 mg d.w. $x g^{-1} d.w$. of the material. It should be mentioned that the intensification of bacterial biomass development occurred only in R₁, i.e. in the zone of the strongest influence of plant (Table 2). It should, therefore, be suggested that stimulation of bacterial biomass development caused by the addition of compost resulted mainly from an indirect influence, through plant stimulation for a larger production of root excretions constituting feed for bacteria. This is sup-

Table 2. Total number and biomass of bacteria in composting pig slurry and greenhouse medium used to rooting of Pelargonium peltatum	
hort. cuttings.	

	Time of analyses								
Variable	Time "0" (May 25)			After rooting (June 27)					
	Pig slurry compost	Medium without	t Medium t with	Samples of control medium from:		Samples of medium with compost from:			
		- control		R 1	R2	NR	R1	R2	NR
Number of bacteria $(10^9 \text{ g}^{-1} \text{ d.w. material})$	29.5	16.8	23.4	27.3	20.1	18.4	26.2	18.7	18.8
Biomass of bacteria (mg d.w. g ⁻¹ d.w. material)	3.2	0.7	1.0	1.3	0.9	0.8	1.8	0.9	0.9

Medium: R1 – closely adhering to the roots, R2 – remaining on the roots, NR – non-rhizosphere. Values for kind of medium and rhizosphere effect differ significantly at $P \le 0.01$.

ported by the fact that an increase of the biomass of bacteria resulted not from their higher number, but from the enlargement of average size of cells, which is in close relation with the level of easily available food in the environment. A relatively limited response of bacterial communities to the enrichment of the medium with compost may also be associated with competitive relationships between these organisms and fungi. These suggestions are in agreement with Leżała [5], who found that enrichment of greenhouse medium for geranium culture with composted pig slurry stimulates a little more the development of fungal than bacterial biomass. Rochette et al. [10] noted a rapid increase of microbial biomass and CO_2

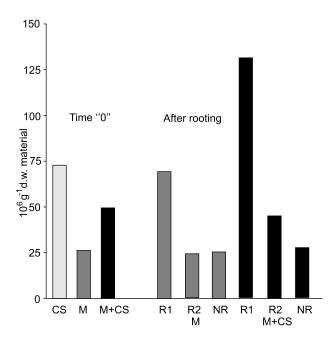


Fig. 1. Colony forming units (CFU) of actinomycetes in compost and greenhouse media used for the rooting of geranium. CS – compost, M – medium (control), M + CS – medium with compost; R1 – medium closely adhering to the roots, R2 – medium remaining on the roots, NR – non-rhizosphere medium.

emission in the soil fertilized directly with pig slurry. Our observations indicate that the addition of previously composted pig slurry to the greenhouse medium may lead to a considerably slower bacterial degradation of organic material contained in it.

Colony forming units (CFU) of actinomycetes (Fig. 1) were almost 3-fold more numerous in the composted pig slurry than in the greenhouse medium alone – their numbers amounted to about 73 x 10^6 and 26×10^6 g⁻¹ d.w. of pig slurry and medium, respectively.

In contrast to whole bacterial communities, actinomycetes responded to medium enrichment with composted pig slurry by intensive development. This was particularly evident in the R_1 zone of a rooted plant, where on average over 130 x 106 CFU of actinomycetes were found in the medium with compost as compared to about 70 x 10⁶ in 1 g d.w. of the control medium. That may prove that composted pig slurry has a positive influence on multiplication of plant growth-promoting rhizobacteria (PGPR), among which one of the most important groups is constituted by actinomycetes [1]. The value of a rhizosphere effect for actinomycetes amounted to 4.8 when geranium was grown on the medium with compost and to 2.7 when geranium cuttings were grown on the medium alone (Fig. 3). This indicated a strong synergic effect of compost and a rooted plant on the development of actinomycetes within its rhizosphere.

Colony forming units of fungi (Fig. 2) occurred in the same numbers in the studied compost as in the medium alone, i.e. about $1400 \times 10^3 \text{ g}^{-1}$ d.w. of the material.

After rooting of cuttings in all the samples of both medium with compost and control medium, the fall of CFU number of fungi was found, the smallest one 30%, in R_1 of geranium. The rhizosphere effect, therefore, was only limited, but did not remove the weakening of propagative fungi in the studied media in the course of the experiment. The observations mentioned above [5] concerning stimulation of fungal biomass development with compost addition in connection with our data prove that compost from pig slurry has a positive influence on the size of biomass of fungi and simultaneously diminishes the activity of their propagative forms in the greenhouse medium for geranium.

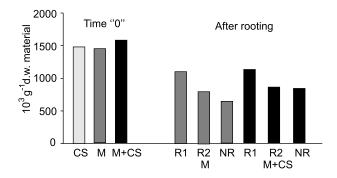


Fig. 2. Colony forming units (CFU) of fungi in compost and greenhouse media used for the rooting of geranium. CS – compost, M – medium (control), M + CS – medium with compost; R1 – medium closely adhering to the roots, R2 – medium remaining on the roots, NR – non-rhizosphere medium.

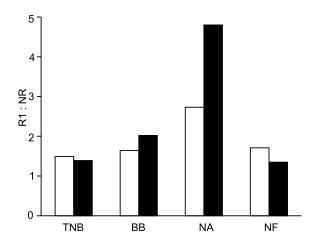


Fig. 3. The rhizosphere effect (R1 : NR) for studied groups of microorganisms in greenhouse media used for geranium rooting. \Box – medium without compost (control), \blacksquare – medium with compost; TNB – total number of bacteria, BB – biomass of bacteria, NA – number of actinomycetes, NF – number of fungi; R1 – medium closely adhering to the roots, R2 – medium remaining on the roots, NR – non-rhizosphere medium.

Bacteria from the genera *Arthrobacter, Pseudomonas, Bacillus* as well as actinomycetes constitute an integral part of bacterial communities determined by the sizes of their biomass. These bacteria display their ability to produce auxins, among others, IAA, and cytokinins [1, 13]. We have found, in composted pig slurry, the population density of bacteria communities is very high. Results of our studies indicate that a positive influence of compost at the beginning of geranium rooting can result in stimulation of root primordium initiation by phytohormones, being products of bacteria introduced in large numbers into subtrate with this biofertilizer. The addition of compost to the medium positively influenced general plant development at further stages of their culture. In the rhizosphere of geranium, the number of actinomycetes recognized as important phytohormone producers [1], simultaneously doubled in comparison to the control. These observations suggest that a positive effect of compost on the rooting of *Pelargonium peltatum hort*. is related to a significant degree with the activity of plant-promoting rhizobacteria, the development of which is stimulated by the addition of that biofertilizer to the greenhouse medium.

In summarizing these results it should be stated that the use of composted pig slurry in growing ornamental plants is a good method of this waste utilisation.

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