Microbial community dynamics during composting od sewage sludge with different additions in bioreactor

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Introduction

With the progress in sewage treatment, there increases the amount of the produced sewage sludges, and their storaging causes a threat for the environment. Therefore, it appears essential to work out effective methods of utilization of these materials which would guarantee the recovery of nutrients for plants and microorganisms deposited in sludges and, at the same time, ensure maximum safety for the environment.

At the present time, a number of methods of sewage sludge utilization are known but most commonly, sewage sludge is composted prior to its introduction into the soil in order to improve its properties, increase nutrient availability and detoxify heavy metals [Droffner et al. 1995]. Following composting processes, wastes containing organic compounds characterised by different sensitivity to decomposition and pathogenic microorganisms are transformed into material which no longer poses hygiene hazards [Hassen et al. 2001].

During the composting process, besides the final product in the form of humus, there are released: heat, compounds of nitrogen, oxygen, phosphorus, CO_2, H_2O, and a significant amount of microorganism biomass is created [Tiquia et al. 2002]. The succession of mesophilic and thermophilic microorganisms in composted material is connected with temperature changes and it is an indicator of the correct course of the composting process [Ishii et al. 2000]. High tempetrature (60-75 °C) reached in the thermophilic phase of composting is a factor which either completely reduces or decreases the number of pathogens [Macgregor et al., 1981]. Composting of sewage sludges favours not only changes in microorganism number, but it also supports changes of enzyme activity levels in composts being regarded as an indicator of a correct course of the composting process [Paleåz et al. 2004].

The objective of the present studies was the determination of the effect of the composting process of sewage sludges with different additions, in controled conditions, on the number of selected microorganism groups and the activity of dehydrogenases.

Materials and methods

Experiment was established in laboratory conditions in 2007. Material used in the studies consisted of sewage sludge originating from the and consisted of wheat and rye straw and sawdust. The microbiological and chemical analyses are presented in tables 1 and 2. Studies were carried out in four bioreactors of 125 dm^3 capacity and equipped with electronic sensors for constant recording of some process parameters (temperature, carbon dioxide, methane, ammonia and oxygen). Materials for studies were thoroughly mixed in a container in weight proportion in relation to dry matter: 75 % of sewage sludge + 20 % of sawdust + 5 % of straw in bioreactor K1 and K2, while in bioreactor K3 and K4, the proportion was: 39 % of sewage sludge + 57 % of sawdust + 5 % of straw.

The experiment was conducted with a constant air flow amounting to 3 L min^{-1} in chambers 1 and 3, and 6 L min^{-1} in chambers 2 and 4.

Material in bioreactors was composted for 668 hrs, while compost samples were taken from all chambers at the same time depending on the actual temperature of the composted material.

On microbiological selective medium, using plate method, the number of colony forming units (cfu) mesophilic, thermophilic bacteria and pathogenic bacteria of *Salmonella, Entreobacteriaceae,* were determined.

Furthermore, in the sampled composted material, the activity of dehydrogenases was identified by spectrophotometric method, using as substrate 1% TTC (triphenyl-tetrazole chloride). Enzyme activity was expressed in mg TPF · kg^{-1} d.m. of compost ·$5h^{-1}$.

Statistical analyses applied in the experiment were used on the basis of Statistica 7.1 program.

Results and discussion

Analysing the changes in the number of choosen groups of bacteria (tab. 1) during the composting process, it was found that the proliferation of the bacteria was subject to significant oscillations connected most probably with temperature changes being characteristic of this process. Miyatake, Iwabuch [2006] informed, that changes in temperature values during composting have an effect on changes in the activity and diversity of microorganisms and thereby on the efficiency of the composting process.

Comparing the number of mesophilic bacteria in composts (tab. 1), between the first two terms, it was found that the proliferation of the discussed bacteria increased over a doze times in the II term in relation to the I term. The reason of this phenomenon was most probably a two-degree increase of temperature and the access to easily decomposing organic matter. Starting with the IV term together with the temperature increase, the number of mesophilic bacteria significantly decreased. The above status was maintained until the VI term. Starting from the VII term, together with the drop of temperature in the composted materials, an increase of bacteria proliferation was recorded. Similar tendencies were found by Hassen et all. [2001].

Analysis of the number of thermophilic bacteria in the studied composts (tab. 1) indicated that in term I of studies, the greatest number of bacteria occurred in compost K2 and it amounted to 28.27 cfu·10^3·g^{-1} d.m.. The composting process which lasted 24 hours caused an increase of temperature in the compost chambers by 11-21°C and at the same time an increase of the number of bacteria in all composts. The bacteria increase and develop in conditions of high temperatures not only because of their specific morphological construction, but also due to the physiologico-chemical properties of their cells, the chemical composition of the substrate and the adequate pH value. Starting with the II term together with the temperature increase, the number of thermophilic bacteria significantly increased.

The above status was maintained until the V term in compost K1-K3 and the VI term in compost K4.

Sanitary studies of sewage sludges and composts detected no presence of bacteria from *Salmonella* genus. The absence of *Salmonella* sp. in sewage sludges could be connected with the efficiency of the method of sludge treatments applied in the sewage treatment plants from which the sludges designed for composting originated.

Analizing the changes in the number of bacteria from *Enterobacteriaceae* family in the composted materials (tab.1) during the whole experiment, it was found that the composting

process contributed to a total reduction of the number of the discussed microorganisms. On the day of the experiment establishment, the highest number of bacteria was recorded in compost K2 – 157.51 cfu·10^2·g^{-1}, while in compost K3, the number was 58.37 cfu·10^2 ·g^{-1} d.m.

Table 1. The number of mesophilic, thermophilic bacteria and bactreia form *Enterobacteriaceae* family in composts (cfu • g^{-1} • d.m. of material)

Kind of compost	Temperature of compost (°C)	Mesophilic bacteria		Thermophilic bacteria		Enterobacteriaceae	
		cfu•105•g^{-1} d.m. of compost	Standard Deviation	cfu•103•g^{-1} d.m. of compost	Standard Deviation	cfu•102•g^{-1} d.m. of compost	Standard Deviation
I date – beginning of experiment							
K1	21	156.70	71.55	21.28	2.74	65.29	11.96
K2	19	185.78	17.13	28.27	3.30	157.51	13.19
K3	19	24.71	12.40	2.92	0.42	58.37	7.94
K4	19	80.17	25.94	24.30	3.44	98.40	39.36
II date - after 24 h							
K1	42	3563.03	1198.90	94.05	52.85	6112.32	601.50
K2	42	2297.51	635.10	356.96	74.72	7048.29	595.68
K3	32	8051.95	1061.17	47.14	17.84	9119.77	837.77
K4	31	6046.46	1804.18	59.87	14.47	10871.59	2486.73
III date - after 32 h							
K1	52	24272.38	1866.02	415552.2	7289.88	7852.83	517.09
K2	50	18337.41	6811.27	30154.85	4034.02	2743.79	226.48
K3	42	10518.96	1295.71	1097.80	373.42	14051.87	2427.59
K4	38	6075.47	1308.29	327.04	77.54	14062.83	1159.96
IV date - after 44 h							
K1	65	9.27	0.97	1212.12	66.70	0.33	0.25
K2	65	22.97	3.49	1896.37	114.88	0.00	0.00
K3	66	4.05	0.50	1056.89	219.80	0.00	0.00
K4	44	1063.41	61.03	3443.68	146.51	9712.66	564.91
V date -after 92 h							
K1	67	1.24	0.16	258.26	73.05	0.00	0.00
K2	63	5.13	0.81	1244.87	262.20	0.00	0.00
K3	66	0.19	0.02	1719.49	187.03	0.00	0.00
K4	50	1.20	0.08	1320.96	606.58	0.00	0.00
VI date - after 164 h							
K1	55	1.20	0.22	168.64	29.26	0.00	0.00
K2	51	3.05	0.39	442.68	69.97	0.00	0.00
K3	44	0.63	0.29	1068.96	330.48	0.00	0.00
K4	51	0.15	0.14	3307.79	238.93	0.00	0.00
VII date - after 668 h							
K1	29	1.77	0.63	124.91	13.35	0.00	0.00
K2	29	13.41	4.22	1341.22	422.42	0.00	0.00
K3	39	65.74	10.80	1503.82	202.41	0.00	0.00
K4	51	20.07	8.03	2186.19	94.58	0.00	0.00

The 20-hour composting process contributed to a violent proliferation of bacteria from *Enterobacteriaceae* family. Such violent proliferation of *Enterobacteriaceae* bacteria was most probably caused by the access of easily degraded organic substances in the initial period of composting and by the optimal temperature for the growth of the discussed bacteria. On the basis of data presented in table 1, we can infer that a further growth of temperature in the composted masses (term IV) contributed to a violent decrease and total

reduce (term V) of the number of the discussed bacteria. Also Vinneras et al. [2003] inform in their studies that the violent increase of temperature to 50-70°C in the composted materials caused the inactivation of pathogenic microorganisms.

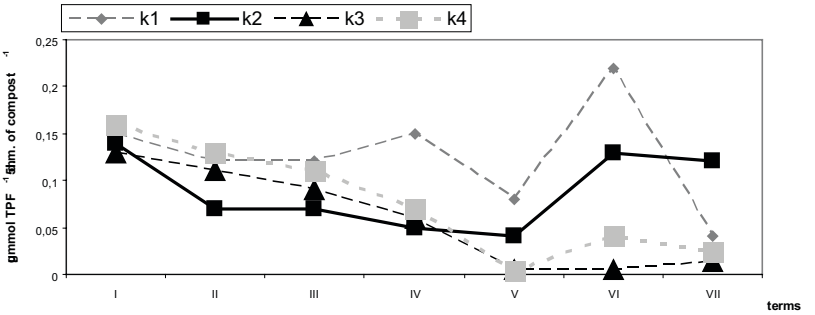

Fig. 1 The changes of dehydrogense activity in composts

An analysis of changes in the activity of dehydrogenases (Fig. 1) during the experiment has indicated that the highest mean level of activity of the studied enzymes occurred in compost K1, while the lowest one was in bioreactor K3.

Conclusions

1. The reduction dynamics of the studied groups of microorganisms (with the exception of thermophilic microorganisms) and changes of dehydrogenase activity in composts was shifted in time, in the majority of cases, according to the temperature increase.
2. The composting process of sewage sludges reduce completely the number of *Enterobacteriacea*.
3. On the basis of the presented studies, it was found that the developed composts met the standards foreseen for composts designed for agricultural purposes **(**Directive European Parlament and Council no 1774/2002).

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