DEVELOPMENT OF FATTY WASTE COMPOSTING TECHNOLOGY USING BACTERIAL PREPARATION WITH LIPOLYTIC ACTIVITY

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Abstract. A new composting technology of waste with high fat content was developed in JSC „Biocentras". The composting technology of fat-contaminated waste is based on the use of fat-oxidizing microorganisms. Developed technology is commended for cleaner production/pollution prevention approach as well as meets strict environmental and hygiene requirements. The composting process was investigated for the process optimization by applying the response surface methodology. Values of parameters of composting process were monitored in lab-scale composters. The optimal composition of the composing mixture was determined: the initial fat content – 5%, the concentration of bacterial preparation cells – 10^9 CFU/g, the quantity of structural materials – 9.5%. Fat degradation rate slowed down 3 times if the initial fatty concentration increased from 5% to 20%. Concentrated fatty-waste disposal site prototype was designed. Composting process duration lasted 1 to 1.5 year.

Keywords: fats, oils and grease (FOG), composting, bacterial preparation, biodegradation, mathematical modeling, optimization.

1. Introduction

Sustainable technologies promote the development of green techniques and products in order to live in clean and healthy environment. New effective waste recycling and utilization technologies are introduced. Our goal is to develop an effective, optimized and affordable technology for recovery of fatty pollutants for meat, fish and oil processing enterprises. At the same time, it will reduce the quantity of biodegradable waste disposed in landfills.

Industrial and household fatty waste can be divided into two categories – "yellow" and "brown" fat. The first category fatty waste can be used for the secondary processing of animal feed additives, production of soaps, oils, cosmetics and skin care products. This type of waste can be composted either. The second type of fat is not recycled and must be handled according to the existing waste management requirements. Biodegradable organic waste of this origin can be incinerated, decayed or composted.

According to the Landfill Directive 1999/31/EC, the State's strategic plan for waste management tasks foresaw the quantity of biodegradable waste that can be disposed in landfills. The plan provides that the amount of waste disposed in landfills must be reduced to 75% by 2010 compared with 2000; by 2013 – up to 50%; by 2020 – will have to achieve only 35% of landfill biodegradable waste compared with 2000 (EC directive 2006).

Biodegradable waste is any waste that may be degraded aerobically or anaerobically, eg., garden waste, paper or cardboard (Navickas *et al.* 2007). Composting is the waste management method, when the complex of biological, biochemical and physical processes is acting, i.e. microorganisms, soil animals and enzymes mineralize biodegradable waste transforming it to biogenic elements and mold.

Composting is not only a natural environment–friendly way to reduce organic waste, but also the good way to reuse secondary raw materials or to produce a high-quality product (Das *et al.* 2002). A number of different composting technologies are created. The basis of those technologies is the homogenization, mixing, good aeration and irrigation of biodegradable waste.

Compost systems can be divided according to the historically ordered composting systems (Haug 2000): 1) open system, without a reactor – the system of composting pile, stationary pile system, 2) reactor systems (tanks or closed) – the vertical compost material spreading, horizontal and inclined compost materials spreading, composting in the box.

Open composting system is the cheapest biodegradable waste treatment method, but it has drawbacks and is not suitable for industrial composting. The use of closed-type equipment is popular in composting of industrial waste. They allow reducing unpleasant smells during the composting process, because the resulting gas is released into environment through the bio-filter or membranes. This type of equipment usually has moisture and oxygen support control monitoring possibility, which allows to regulate and to speed the process. Composting process is divided into two

phases – intensive degradation (thermophilic) phase and compost maturation. There are no strict borders between these stages, but the first stage is characterized by a large volume of oxygen consumption and high temperatures, and in the maturation stage the oxygen is consumed in smaller quantities, passive ventilation is sufficient.

The organic waste with high fat content typically is not composted. However, during the lipid degradation much more energy is emitted in comparison with the degradation of other organic components such as carbohydrates. Higher energy levels are advantageous in thermophilic processes, where high temperature is desired to reduce pathogenic microorganisms; the chemical reactions take place faster at higher temperatures also. A thermophilic condition causes changes in the physical properties of fat, such as melting point, dispersion and solubility (Agamuthu 1994). Therefore, the fat can be an excellent source for aerobic composting.

Composting of fat waste from food industry processing plants, slaughterhouses, restaurants deal with some problems: low water retention and solubility, lack of porosity and relatively low biodegradability of some fat. A significant negative influence also has the gas resulting from fat degradation: hydrogen sulfur (H_2S), methyl mercaptan (CH_3SH), ammonia (NH_3) and volatile organic compounds. Special biological additives that promote the production of enzymes and fatty waste biodegradation are used in composting systems for the reduction of strong and unpleasant smell of concentrated gas (Gandhi 1997).

An alternative method for composting could be fatty waste neutralization using soluble lipase preparations. However, this technology is expensive, single-use and does not have wide application in practice (Adewumi *et al.* 2005; Mendes and Castro 2005; Saifuddin and Chuna 2006). Therefore, composting by using bacterial compositions, composed of a mixture of selected microorganism's cultures is more promising and economical method for fatty waste treatment. Microbial strains mixes have strong biodegradation capacity, since the breakdown of complex compounds found in the FOG (fats, oils and greases) waste require genetic information of several organisms. Genetic potential and certain environmental factors such as temperature, pH, and available nitrogen and phosphorus sources determine the speed and extent of biodegradation.

Concentrated fatty waste obtained by mechanical removal, are composted in specially equipped places. These wastes are neutralized using active cultures of microorganisms. In our previous work has shown that the bacterial compositions created by JSC "Biocentras" breakdown effectively the fat in the water and soil and can therefore be applied to speed up composting (Grigiškis *et al.* 2007). Before composting technology can be commercially adopted, it is necessary to determine the grease-rich wastes decomposition rate, time required to compost and optimal composition of the initial compost mixture. Values of parameters of composting process were monitored in lab-scale composters. Developed technology is attractive for cleaner production/pollution prevention approach, as well as meets strict environmental and hygiene requirements.

2. Methods and materials

The fat content, temperature, humidity, pH, total and ammonium nitrogen, phosphorus, total carbon and content of live microorganisms were assessed in compost mixtures during the composting process. The temperature of compost mixtures was measured daily with a digital indicator MASTFCH®MAS838 pinning thermal element to the center of the each reactor. For the determination of humidity 10 g of composted mixture was dried for 40–48 hours in Binder ED 23 (Germany) drying oven in 105 °C ± 1 °C till the constant weight was reached. For the measurement of the pH, the suspension of 1 M potassium chloride solution in the ratio 1: 2.5 (inoLab 720, Germany) was prepared (LST ISO 10390:2005). Fat was extracted by chloroform–methanol mixture. After extraction chloroform layer was poured to the weighting bottle heated to constant weight, dried and weighted. The fat content calculated by mass difference. The total nitrogen content was determined by the Kjeldahl method (InKjel 625 F, S4 Behr, Behr Labor-Technik). Ammonium nitrogen was determined manually by spectrophotometer by extracting with 2 M potassium chloride solution. The content of phosphorus was determined spectrophotometrically, using ammonium molybdate. Spectrophotometrical measurements were made by Shimadzu UV–1601 spectrophotometer (Shimadzu Corporation). The total carbon content was determined by oxidation of a compost mixture in hot potassium dichromate solution (Alef and Nannipieri 1995).

Fats were biodegraded by using bacterial composition, composed of the active microorganisms *Enterobacter aerogenes* E13 (capable of degrading fats), *Arthrobacter sp.* (N3) (capable of degrading aliphatic compounds) and *Bacillus coagulans* (S1) (capable of degrading complex peptide bounds) cultures. The bacterial strains were cultivated in 750 ml Erlenmeyer flasks with 70 ml of nutrient broth ("Oxoid") separately at 30 °C and 200 rpm. in a rotary shaker (Innova 43, New Brunswick Scientific Co.) for 16 h. The cell count was determined by plating serial dilutions of samples on nutrient agar (Oxoid) plates and incubating at 30 ºC for 24 h (Alef and Nannipieri 1995). Reaction surface methodology based on factor experimental was used for optimization of technological parameters of fatty waste composting (Montgomery 2001; Myers and Montgomery 2002; Levisauskas *et al.* 2004). The calculation programs were realized in Matlab/Simulink and the Maple environment.

A new composting technology with high fat content was created (Fig. 1). The composting technology of fat contaminated waste is based on the use of fat oxidating microorganisms. Natural microbiological processes can be accelerated using selected cultures of microorganisms with enhanced activity and facilitating their vital activities. Key features of the proposed composting technology processes are grinding, mixing, incorporation of structural materials and the fertilizer (N–P–K), pH adjustment of composted mixture, spraying of bacterial composition, irrigation and aeration.
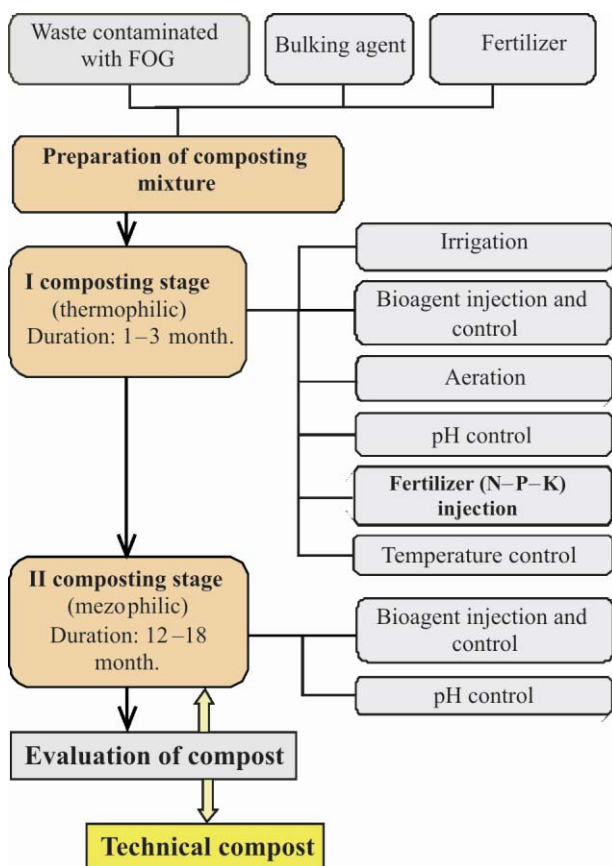
Fig. 1. Composting process diagram

Decanted sewage, collected from the fat traps from meat, fish, vegetable oil processing plants and restaurants and in food preparation businesses have the fat content up to 30–40%. Trapped waste is basically made up of fats and oils.

Saturated and unsaturated fatty acid composition of retained fat varies depending on the company. Retended fat mainly consist of saturated (myristic, palmitic, stearic, eicosanoic, behenic); mono-unsaturated (oleic) and poly-unsaturated (linoleic and linolenic) fatty acids (Coker 2006). The degradation of these fatty acid leads to unpleasant smells.

Animal fat consists predominantly of saturated fatty acids, plant's fat – mainly consist of unsaturated fatty acids. These fatty acids are found in all types of grease, only in various amounts. The model grease fatty substrate, made from equal parts of pork fat, beefy suet and sunflower oil, was chosen for composting experiment. The pork fat is made up of saturated palmitic and stearic fatty acids, beef suet mainly consist of saturated palmitic and stearic fatty acids. Pork fat also has a number of oleic acid. Sunflower oil is mostly composed of polyunsaturated linoleic acid and monounsaturated oleic acid (Gunstone and Padley 1997). Thus, practically all the fatty acids forming fatty waste can be found in the model fat mixture.

Appealing on previous research results, we conclude that the degradation takes place effectively when the fat concentration in soil is 5–15%. Above that level fat degradation rates decrease. Similar results were received by

Erhan and Kleiman (1997) Nakasaki (2004) and Gea *et al.* (2004). Fat degradation increases with increasing time period. In addition, lipid breakdown is dependent on what fatty acids they consist of. The degradation of long chain fatty acids increases with their unsaturation degree and aliphatic chain shortening. It should also be assessed that high concentration of fat has a negative influence on oxygen transport and inhibit the activity of fat-splitting microorganisms (Chipasa and Mędrzycka 2006).

Two series of experiments were carried out for the practical assessment technology being created. The first series of experiments were carried out to assess the initial relationship of the C/N ratio and the temperature of composted mixture. Microorganism's (bacteria, fungi and actinomycetes) content also affects the temperature evolution of composted mixture during the process.

Three composting mixes were prepared and the process was carried out in the 20-liter reactors in the optimal composting conditions recommended by literature (Haug 2000; Bazrafshan *et al.* 2006). The initial concentration of model fat in compost mixtures was (weight ratio) 5%, soil – 40%, peat – 10%, bacterial compositions – 5%, humidity – 40%, pH 6.8, C/N ratio was 30/1, 40/1 and 60/1. C/N ratio was corrected by adding urea to compost mixtures. The temperature of compost mixtures was measured every day.

Three experiments in 20-liter reactors with compost mixtures were carried out in the second series, to choose more correct values of technological parameters which had to be varied in experimental design. Compost mixtures composition is shown in Table 1. Soil and peat were used air-dry. The first compost reactor was supplied with the air at the rate of 0.1 *l*/min for aerobic composting conditions assurance. Compost mixes in the 2nd and 3rd reactors were ventilated naturally during the entire composting process. Compost mixtures were investigated after 2, 4, 8 and 12 weeks. The temperature of composting mixtures was measured in all the reactors every day.

Table 1. Composition of composting mixture

Compost composition	Reactor 1	Reactor 2	Reactor 3 (control)
Fatty substrate, %	10	10	10
Moisture, %	35	35	40
Soil, %	40	40	50
Peat, %	10	10	–
Bacterial preparation,%	5	5	–
CaO, %	0.12	0.12	0.12
Fertilizer, %	0.1	0.1	–

The third series of experiments was carried out to determine the optimal composting conditions of the fat contaminated soil. The goal of optimization was to determine the composition of composting mixture: the quantities of fat, structural materials and bacterial composition, at which the maximum rate of fat degradation is gained. The composting process proceeded in 16 compost boxes with the natural ventilation. Close to D-optimal (B₃) factorial experimental design (Hartmann *et al.* 1974) is shown in Table 2. The manipulated factors were:

X_1 – percentage of fat mixture (5–20%), X_2 – cell concentration of bacterial composition (10^7–10^9 CFU/g); X_3 – percentage of peat (5–15%).

The following technological parameters were fixed during the composting process: temperature – 20 ± 1 °C; pH 6–7; the quantity of added fertilizer ($NH_4NO_3 \cdot KH_2PO_4$) reduced the initial C/N ratio to 60/1; compost mixtures humidity was 50–65%. As with all forms of life involved in the composting process microorganisms needed water. Optimal initial moisture content in composted mixture was 50–65% (Haug 2000; Chipasa and Mędrzycka 2006). With more moisture content, water can fill the pores, and create the conditions for anaerobic composting. Compost boxes were covered with permeable membrane. The temperature of compost mixtures was measured every day (Tremier *et al.* 2004). The additional microorganism culture was sprayed after the 3rd and 6th week. Analysis was performed after 2, 4, 6, 8 and 16 weeks.

Table 2. Experimental design for 16 composting boxes

Box No.	Exsperiment conditions (factor values)					
	X_1		X_2		X_3	
	code	real, %	code	real, CFU/g	code	real, %
1	+1	20	+1	10^9	+1	15
2	−1	5	+1	10^9	+1	15
3	+1	20	−1	10^7	+1	15
4	−1	5	−1	10^7	+1	15
5	+1	20	+1	10^9	−1	5
6	−1	5	+1	10^9	−1	5
7	+1	20	−1	10^7	−1	5
8	−1	5	−1	10^7	−1	5
9	+1	20	0	10^8	0	10
10	−1	5	0	10^8	0	10
11	0	12.5	+1	10^9	0	10
12	0	12.5	−1	10^7	0	10
13	0	12.5	0	10^8	+1	15
14	0	12.5	0	10^8	−1	5
15	0	12.5	0	10^8	0	10
16	0	12.5	0	10^8	0	10

3. Experimental results

3.1. Evaluation of bacterial preparation for composting

The first series of experiments was carried out to evaluate the relationship between compost C/N ratio and temperature (Fig. 2). As the bacteria were breaking down the materials in the compost mixtures, heat was generated and the composts heats up. The temperature changes during the composting process depend not only on the initial C/N ratio, but on the nature of the composted material, the method of composting and compost humidity also. C/N ratio is about 30/1 in ideal composting conditions. The constant temperature rise at the presence of recommended C/N ratio (Fig. 2), the maximum is reached in the 11th day (71 °C). When the C/N ratio is 60/1 the temperature rises slower and settles at 30 °C for longer time. Microorganisms presenting in the bacterial composition are mezophiles, so this temperature range ensures optimum conditions for their vital functions.
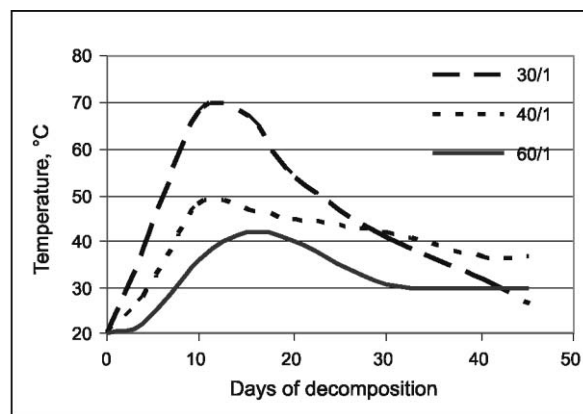
Fig. 2. C/N ratio influence of composted mixture temperature

High C/N ratio shows the lack of nitrogen needed for the optimal growth of microorganism's population. In this case, biodegradation proceeds in relatively low temperatures and not all the organic waste containing carbon is degraded; the process is very slow also. Biological degradation of organic pollutants consists of two processes: the growth of microorganisms and co-metabolism. During the growth of microorganisms organic pollutants are used as the sole carbon and energy source, nitrogen is used for the protein synthesis. It is therefore necessary to maintain a balanced ratio of these elements in a compost mixture.

Biological degradation of fatty waste is complicated by the physical and chemical properties such as their hydrophobisity (insolubility in water). Therefore microorganisms meet difficulties in access and assimilation of fatty wastes. Fatty compounds are different in structures and levels of solubility, therefore enzymes catalyzing fat degradation reactions meet problems in reaching them. Biological degradation of such waste in thermophilic (50–60 °C) conditions is more appropriate than in mezophilic conditions. However, C/N ratio is maintained in high level and the temperature rise relatively slowly at high concentration of fatty substances, therefore the optimal composting proceed at mezophilic conditions (30–37 °C). The destruction of pathogens at high temperature has to be ensured after this stage. In general, nonspore-forming bacteria and vegetative cells or sporulating bacteria are practically destroyed in 5 to 10 min at temperatures of 60–70 °C (moist heat). Composting process management is one of the key factors that lead to weed seeds, disease causing microorganisms and pathogens destruction (Haug 2000).

The initial C/N ratio in composting mixtures was 94 to 101 (Table 3) in the second series of experiments. Fertilizer was added in the 1st and to the 2nd composting reactors to reduce this ratio till 60/1. The optimum moisture content for composting is 55–65%, but the first week of composting it was difficult to ensure the proper moisture content because of the fat properties to repel water. During the composting process moisture retention was improving. The temperature of environment and compost mixture was registered each day (Fig. 3). The temperature rose to 35 °C in the first reactor with air supply after 5 days of composting. The temperature in the reactor

Table 3. Composting results of model fatty pollutant

Reactor No.	Measured parameter*	Composting time, weeks				
		0	2	4	8	12
1	FOG, %	10.0	8.12	4.10	2.81	0.54
	Moisture, %	42.8	45.3	43.8	48.6	46.9
	NH_4^+–N, mg/kg	450.3	191.1	129.4	38.6	21.5
	Total nitrogen (N), mol/kg	0.16	–	–	–	0.70
	Carbon (C), mol/kg	16.2	–	–	–	13.0
	C/N ratio	$101 \rightarrow 60$	–	–	–	18.6
	PO_4^{3-}–P, mg/kg	580.3	281.7	203.6	91.5	78.2
	pH	7.01	5.54	5.26	6.55	6.02
2	FOG, %	10.0	8.62	5.02	3.04	1.68
	Moisture, %	40.5	44.9	45.6	49.2	48.3
	NH_4^+–N, mg/kg	450.3	240.5	136.7	113.1	98.7
	Total nitrogen (N), mol/kg	0.16	–	–	–	0.42
	Carbon (C), mol/kg	16.2	–	–	–	14.2
	C/N ratio	$101 \rightarrow 60$	–	–	–	33.7
	PO_4^{3-}–P, mg/kg	580.1	448.3	316.7	148.0	105.2
	pH	7.06	5.79	5.58	6.70	5.90
3	FOG, %	10.0	8.90	8.06	6.93	6.21
	Moisture, %	38.9	42.8	46.3	45.2	40.4
	NH_4^+–N, mg/kg	40.2	36.3	33.5	24.7	20.4
	Total nitrogen (N), mol/kg	0.121	–	–	–	0.14
	Carbon (C), mol/kg	11.4	–	–	–	8.42
	C/N ratio	94.3	–	–	–	60.1
	PO_4^{3-}–P, mg/kg	50.30	46.0	28.7	23.6	19.6
	pH	7.02	6.27	5.82	6.73	6.39

All results are average of three repeated measurements.

having passive ventilation and the structural material addition rose up only to 30 °C and remained constant for a longer period of time. Temperature rise was not significant in the control reactor with passive (natural) ventilation, but it kept a few degrees higher than room temperature (20 ± 1 °C). The temperature rise in the reactors shows that in all the cases it was mezophilic processes. Reactor with air supply needed more frequent irrigation, because the moisture evaporated because of the forced ventilation.

At the beginning of the composting process, when the population of microorganisms grows and temperature of the compost mixture rises, easily degradable compounds are decomposed. Emitted heat raises the temperature of the compost mixture but pathogens are not eliminated. After 4 weeks when temperature of the compost mixture fell, additional air supply was discontinued and the compost matured further at mezophilic temperature.

Three compost mixtures were treated with heat at 70 ± 1 °C for 12 hours for the reduction of pathogens after 4 weeks of experiment. According to literature data almost all the pathogens are neutralized in this temperature after 1 hour (Haug 2000). Bacterial preparation microbes were not found in the compost mixtures as well. There
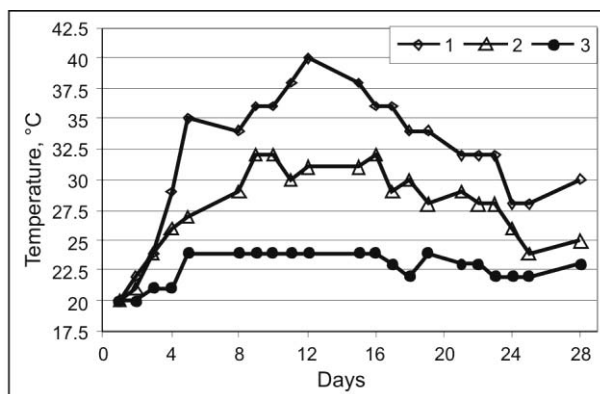
Fig. 3. Temperature of compost and environment (curve 1 – in reactor 1; curve 2 – in reactor 2; curve 3 – in reactor 3 (control))

have not been found bacterial compositions forming microorganisms as well. The demand of re-spraying the bacterial composition into the compost mixture in the mezophilic stage of the compost maturation is obvious. The duration of thermophilic phase must be at least 2 weeks, and maintaining the temperature of compost at 55 °C is essential for the application of this technology to the composting sites (Erhan and Kleiman 1997).

Biodegradation process was very slow and fat content reduced from 10% to 6.21% after 12 weeks with no addition of selected microbial strains and structural material (Table 3). This indicates that wild microflora was able to degrade fatty contaminants at some degree. This reactor had the most unpleasant smell. The smallest drop of pH and the smallest temperature rises was detected in the third reactor indicating a low intensity of fat pollutants degradation processes.

Selected bacterial strains preparations were used in the first and second reactors to accelerate fat degradation process. Comparing contents of degraded fats in the first and second reactors the first reactor with additional aeration had more decomposed fatty pollutants.

The optimal pH for microorganisms involved in composting process is 5.5 to 8.5. Microorganisms forming bacterial composition effectively degrade fatty waste at the pH ranging from 6.3 to 7.5. When microorganisms fragment fatty substances, organic acids appear which reduces the compost pH to 4.5 and below. The activity of microorganisms is slowed down this way, so compost mixture pH is maintained in required level with calcium carbonate.

Sharper fall in pH of the compost mixture, greater amount of emitted energy at the time of biodegradation indicates that the process was more intensive than in the second reactor. It should be noted that these composting reactors almost had no unpleasant odor, and after 4 weeks of composting the mixture corresponded to the smell of compost. According to the developed technology, composted mixtures must be moved to piles for further maturing after thermophilic and mezophilic stage.

Composting process does not stop at a certain point. Compost is stable and can be used after 12–13 weeks of active composting, but the composted material is further being decomposed. C/N ratio of 18.6/1 after 12 weeks was determined in the compost mixture of the first reactor; the third reactor had the C/N ratio of 60/1. It shows the compost is not yet fully formed after 12 weeks of composting. Subsequent compost maturation process continues aerobically degrade resistant materials and compounds. Various invertebrates (protozoa, nematodes, earthworms, insect larvae etc.) at this stage are actively involved in compost ripening process. Compost can mature an indefinite period of time. In many cases, the degree of maturation of compost and composting process end is associated with its physical characteristics – the smell, texture, oxygen consumption, pH, C/N ratio.

3.2. Composting technology optimization

The third series of composting experiments was carried out to define optimum composting conditions of fat contaminated soil. Measured temperatures of 16 compost mixtures were higher than room temperature in the all cases. The number of live microorganisms was set and the ammonium nitrogen and phosphorus content variation was registered (Tables 4 and 5) during the composting process.

Table 4. Change in the number of microorganisms during composting

Reactor No.	Cell concentration, CFU g^{-1}					
	0 weeks	2 weeks	4 weeks	6 weeks	8 weeks	16 weeks
1	$1.1 \cdot 10^9$	$1.5 \cdot 10^6$	$6.5 \cdot 10^7$	$8.7 \cdot 10^6$	$1.5 \cdot 10^6$	$0.9 \cdot 10^6$
2	$1.1 \cdot 10^9$	$2.2 \cdot 10^6$	$3.2 \cdot 10^7$	$6.0 \cdot 10^5$	$2.2 \cdot 10^6$	$1.4 \cdot 10^6$
3	$1.1 \cdot 10^7$	$1.5 \cdot 10^7$	$2.5 \cdot 10^7$	$5.8 \cdot 10^6$	$1.5 \cdot 10^7$	$8.9 \cdot 10^6$
4	$1.1 \cdot 10^7$	$8.7 \cdot 10^6$	$5.0 \cdot 10^6$	$2.0 \cdot 10^6$	$8.7 \cdot 10^6$	$7.7 \cdot 10^6$
5	$1.1 \cdot 10^9$	$1.1 \cdot 10^7$	$7.1 \cdot 10^6$	$9.5 \cdot 10^6$	$1.1 \cdot 10^7$	$9.3 \cdot 10^6$
6	$1.1 \cdot 10^9$	$1.3 \cdot 10^8$	$1.6 \cdot 10^8$	$7.0 \cdot 10^6$	$1.3 \cdot 10^8$	$7.9 \cdot 10^7$
7	$1.1 \cdot 10^7$	$1.5 \cdot 10^7$	$1.5 \cdot 10^7$	$2.4 \cdot 10^6$	$1.5 \cdot 10^7$	$0.7 \cdot 10^7$
8	$1.1 \cdot 10^7$	$2.1 \cdot 10^7$	$1.1 \cdot 10^7$	$6.4 \cdot 10^6$	$2.1 \cdot 10^7$	$1.6 \cdot 10^7$
9	$1.1 \cdot 10^8$	$3.4 \cdot 10^7$	$2.2 \cdot 10^7$	$8.7 \cdot 10^6$	$3.4 \cdot 10^7$	$9.2 \cdot 10^6$
10	$1.1 \cdot 10^8$	$9.3 \cdot 10^6$	$8.2 \cdot 10^6$	$1.2 \cdot 10^6$	$9.3 \cdot 10^6$	$4.8 \cdot 10^6$
11	$1.1 \cdot 10^9$	$5.0 \cdot 10^7$	$2.3 \cdot 10^7$	$1.1 \cdot 10^7$	$5.0 \cdot 10^7$	$2.0 \cdot 10^7$
12	$1.1 \cdot 10^7$	$1.3 \cdot 10^7$	$1.3 \cdot 10^7$	$6.0 \cdot 10^6$	$1.3 \cdot 10^7$	$8.3 \cdot 10^6$
13	$1.1 \cdot 10^8$	$9.1 \cdot 10^6$	$9.0 \cdot 10^6$	$1.5 \cdot 10^6$	$9.1 \cdot 10^6$	$5.3 \cdot 10^6$
14	$1.1 \cdot 10^8$	$3.4 \cdot 10^7$	$4.3 \cdot 10^7$	$6.7 \cdot 10^6$	$3.4 \cdot 10^7$	$9.6 \cdot 10^6$
15	$1.1 \cdot 10^8$	$1.7 \cdot 10^7$	$2.7 \cdot 10^7$	$4.4 \cdot 10^6$	$1.7 \cdot 10^7$	$1.1 \cdot 10^7$
16	$1.1 \cdot 10^8$	$1.4 \cdot 10^7$	$2.4 \cdot 10^7$	$1.8 \cdot 10^6$	$1.4 \cdot 10^7$	$0.9 \cdot 10^7$

A visible reduction in the quantity of microorganisms in composting boxes, where the initial concentration of the mixture of fat was 12.5% and 20% was observed after 2 weeks. Microorganisms require a long adaptation period because fat is heavily degradable substrate. After the adaptation period lipolytic activity arises and fat degradation as well. The maximum fall in pH was detected in the same compost boxes. We can see in Table 5 and Table 6 the decrease of ammonium, orthophosphate and fat content in the same compost boxes indicating the ongoing life processes. The mathematical model for approximation the dependence of the fat degradation rate on the composition of composting mixture (response surface) was developed using the experimental design plan presented in Table 2 and the corresponding experimental data of fat degradation after 2 weeks presented in Table 6.

The second-order multiple polynomial model demonstrates an adequate approximation of the response surface over the investigated region of the factor variations:

$$Y = a_0 + a_1 x_1 + a_2 x_2 + a_3 x_3 + a_4 x_1^2 + a_5 x_2^2 + a_6 x_3^2 + a_7 x_1 x_2 + a_8 x_1 x_3 + a_9 x_2 x_3, \quad (1)$$

where Y is predicted response (percentage of degraded fat), x_1 is percentage of fat mixture, %, x_2 is cell concentration of bacterial composition, CFU/g, x_3 is percentage of peat, %, a_i are the model parameters identified using the least squares method (Montgomery 2001) for normalized factor variation ranges $(-1 \le x_i \le 1)$, $a_0 = 17.3$, $a_1 = -21.9$, $a_2 = 7.33$, $a_3 = 0.47$, $a_4 = 7.62$, $a_5 = 12.0$, $a_6 = -8.88$, $a_7 = -7.19$, $a_8 = 1.62$, $a_9 = -0.732$.

Table 5. Ammonium nitrogen and phosphorus content evolution during composting

Reactor No.	Results of experiments											
	0 weeks		2 weeks		4 weeks		6 weeks		8 weeks		16 weeks	
	NH_4^+–N, mg/kg	PO_4^{3-}–P, mg/kg	NH_4^+–N, mg/kg	PO_4^{3-}–P, mg/kg	NH_4^+–N, mg/kg	PO_4^{3-}–P, mg/kg	NH_4^+–N, mg/kg	PO_4^{3-}–P, mg/kg	NH_4^+–N, mg/kg	PO_4^{3-}–P, mg/kg	NH_4^+–N, mg/kg	PO_4^{3-}–P, mg/kg
1	415.9	295.6	177.6	207.8	138.4	91.9	91.9	138.2	86.3	99.3	91.6	68.1
2	154.3	61.8	106.9	50.6	84.0	41.8	41.8	39.9	15.4	34.3	20.3	20.9
3	428.6	293.8	377.3	269.2	285.8	97.2	97.2	627.3	100.3	545.6	104.3	342.5
4	162.6	147.3	116.4	60.2	42.9	53.5	43.5	23.0	55.7	19.4	51.8	11.6
5	318.7	297.4	200.1	262.8	113.0	27.8	27.9	38.1	23.8	22.6	24.2	14.8
6	159.5	163.7	105.2	49.4	59.2	13.7	13.7	15.1	15.5	13.9	14.8	7.62
7	418.3	159.2	280.9	171.5	116.8	78.0	78.0	177.9	63.9	156.9	51.3	106.9
8	272.4	151.3	109.0	133.7	46.1	15.9	15.9	81.2	13.3	77.6	8.62	28.6
9	435.1	278.9	368.3	752.8	230.2	31.4	31.4	266.4	30.4	249.5	27.8	176.3
10	371.6	241.3	91.5	150.0	27.0	19.0	19.0	26.2	15.2	19.4	7.65	9.32
11	364.3	238.9	129.8	362.0	71.4	57.1	57.1	114.2	10.7	102.5	14.7	95.3
12	379.2	228.7	255.6	164.8	153.5	99.1	99.1	166.6	95.2	151.4	50.5	87.4
13	378,3	245.3	232.7	226.3	197.5	146.6	146.6	181.0	108.3	175.9	73.2	104.1
14	338.1	239.4	191.1	131.8	129.4	38.6	38.6	91.5	40.3	87.5	16.7	35.2
15	378.6	229.8	240.5	248.4	136.7	113.1	113.1	148.1	93.6	132.2	40.1	68.2
16	327.8	184.9	231.5	244.3	90.0	82.3	82.3	147.1	75.2	112.1	25.4	70.2

Table 6. Fat change during composting

Reactor No.	Results of experiments										
	0 weeks	2 weeks		4 weeks		6 weeks		8 weeks		16 weeks	
	FOG conc., %	FOG conc., %	Redused FOG, %	FOG conc., %	Redused FOG, %	FOG conc., %	Redused FOG, %	FOG conc., %	Redused FOG, %	FOG conc., %	Redused FOG, %
1	22.2	21.0	5.38	11.9	46.5	6.80	69.4	3.73	83.2	1.86	91.6
2	5.56	2.08	62.5	1.33	76.0	0.59	89.3	0.05	99.2	0.01	99.8
3	22.2	21.2	4.48	18.4	16.9	14.8	33.2	11.1	49.9	9.64	57.5
4	5.56	3.52	36.6	3.23	41.9	1.57	71.8	0.67	87.9	0.32	94.2
5	22.2	21.6	2.72	14.8	33.5	5.73	74.2	2.46	88.9	0.87	96.1
6	5.56	1.66	70.1	0.58	89.6	0.40	92.7	0.12	97.9	0.05	99.1
7	22.2	21.6	2.63	19.8	10.7	12.0.	46.0	9.44	57.5	7.03	68.4
8	5.56	3.47	37.5	2.05	63.1	0.71	87.1	0.39	92.9	0.20	96.4
9	22.2	19.6	11.9	18.3	17.5	11.0	50.7	7.56	66.0	5.67	74.5
10	5.56	3.35	39.8	2.34	57.8	0.97	82.5	0.32	94.3	0.14	97.5
11	13.9	8.74	37.1	6.28	54.8	1.30	90.6	0.29	97.9	0.02	97.0
12	13.9	10.6	23.3	7.99	42.5	5.92	57.4	0.27	80.3	0.08	88.9
13	13.9	12.0	13.7	9.14	34.2	4.54	67.3	0.18	86.9	0.06	91.6
14	13.9	13.2	5.00	7.89	43.2	3.40	75.5	1.21	91.3	0.07	90.3
15	13.9	11.9	14.3	10.3	25.9	4.23	69.5	1.87	86.5	0.03	95.2
16	13.9	12.1	12.5	10.1	27.2	5.06	63.6	2.14	84.6	0.04	94.3

The model (1) predictions of experimental data at the factorial experiment points are presented in Table 7. The 2 repeated experiments (points 15, 16 in Table 7) were carried out for testing the model adequacy.

The hypothesis concerning the adequacy of model (1) was verified with the F (Fisher) statistic (Myers and Montgomery 2002). The computed value of the F-statistic $F = 27.2$ is less than the critical value $F_{v1, v2, \alpha} = F_{5, 1, 0.05} = 230.2$, therefore, the established model (1) for prediction the percentage of degraded fat is satisfactory.

The response surface model (1) based analysis has shown that the predicted maximum rate of fat degradation (over 73% after 2 weeks of composting) is obtained at the following technological conditions: the fat content – 5.0%, cell concentration in bacterial compositions – 10^9 CFU/g, peat content – 9.5%. A character of the response surface (dependence of the fat degradation rate on the investigated factors) in the vicinity of the predicted maximum percentage point is shown in Fig. 4 by contour plots of the response surface sectional views calculated at the maximum percentage point.

The presented response surface shows that the percentage degradation rate of fat increases with decreasing the initial fat concentration: by decreasing the initial concentration from 20% to 5% (at peat content 9.5% and cell concentration 10^9 CFU/g) the percentage degradation rate rises up to 4 times.

Table 7. Model predictions of experimental data

Number of experiment point in Table 3	Experimental results	Model predictions
1	5.38	7.5
2	62.5	62.6
3	4.48	8.73
4	36.6	35.0
5	2.72	4.82
6	70.1	66.3
7	2.63	3.08
8	37.5	35.8
9	11.9	2.94
10	39.8	46.8
11	37.1	36.6
12	23.3	21.9
13	13.7	8.86
14	5.00	7.91
15	14.3	17.3
16	12.5	17.3
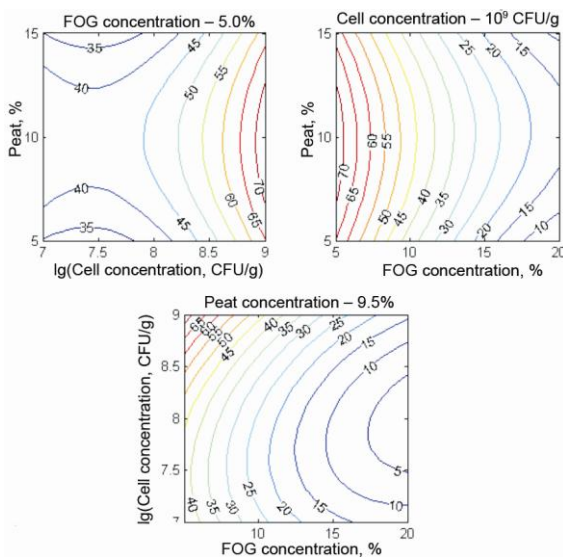
Fig. 4. Dependence of the degraded fat percentage after 2 weeks on the composition of composting mixture (percentage of fat, inoculate cell concentration and the percentage of peat) in the vicinity of maximum degraded fat percentage point

However, in economic terms it is reasonable to treat the compost with higher initial percentage of fat, at which the higher degradation rate over a longer period of time can be achieved in terms of absolute units. At the initial fat concentration 20% in 6 weeks about 70% of total fat was degraded.

3.3. Concentrated fatty waste disposal site prototype

Concentrated fat waste disposal site prototype is designed (Fig. 5). The site is designed on rolled ground with 10 cm of sorbent on the bottom. Compost mixture containing structural material and added bacteria is placed into the closed container with air, bacterial composition and water supply, temperature regulation. The 1st phase of composting lasts 1–3 months. The compost mixture is maintained in thermophilic conditions for the last 2 weeks of treatment to ensure the sanitary-hygienic requirements.
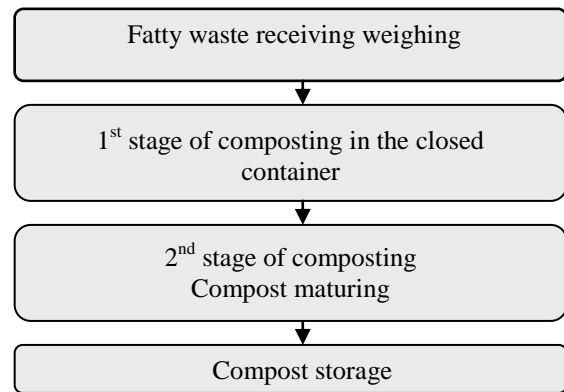

Fig. 5. Concentrated fatty-waste disposal site prototype

At the end of the first phase composting mixture is transferred to the open composting piles. In the second stage compost maturates in mezophilic conditions for indeterminate period of time. Composting pile height is 1.2–1.5 m, the base width is about 2.5–3.0 m, the sides have small rake in order the pile would get narrower to the top and would have the shape of trapezium in the cross–section and the top width would be 0.6–1.0 m. Pile length is 5–6 meters. Composting piles must be mixed 3–4 times during the composting season to access sufficient quantities of oxygen, to get inner and outer layers humidity and fragmentation equal, and to accelerate composting process. Process monitoring consists of regular compost temperature, quantity of microorganisms, pH and moisture measurements, and control tests of samples.

Compost mixture loses about 30–40% of former organic materials in carbon dioxide and water form, therefore the yield of compost represents only 40–50% of the initial volume of compost, and up to a half its initial mass.

Total duration fat waste composting time is 1.0 to 1.5 years. Obtained product is compost, which can be divided into two types. Compost with low–nutrient content can be used for improving soil structure. Compost with enough nutrient's content, can be used as fertilizer in the soil, to supply the plants with the necessary basic nutrients and organic materials. High-quality compost can be used for market needs: in horticulture, gardening, landscape development and agriculture, when the need to improve soil structure exists.

4. Conclusions

Technology of waste with high fat content was developed. The waste is blended with the structural material, sprayed with bacterial composition and enriched with feed elements.

Optimal conditions and composition of composting mixture is determined: the initial fat content – 5%, the concentration of bacterial composition's cells – 109 CFU/g, the quantity of structural materials – 9.5%.

Concentrated fat waste disposal site prototype is designed. Maximal possible fat concentration for this composting technology is 20% and depends on initial waste concentration, and lasts 1 to 1.5 years.

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RIEBALINIŲ ATLIEKŲ KOMPOSTAVIMO TECHNOLOGIJOS SUKŪRIMAS NAUDOJANT LIPAZINIO AKTYVUMO BAKTERINĮ PREPARATĄ

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S a n t r a u k a

UAB „Biocentre" sukurta nauja riebalais užterštų atliekų kompostavimo technologija, pagrįsta riebalus oksiduojančių mikroorganizmų panaudojimu. Ši technologija pažangesnė taršos prevencijos požiūriu, jos privalumas – švaresnė gamyba. Sukurtoji technologija atitinka griežtus aplinkos apsaugos ir higienos reikalavimus. Tyrimai atlikti laboratorinėmis sąlygomis. Riebalinių atliekų kompostavimo procesų technologiniams parametrams optimizuoti taikyta faktoriniais eksperimentais grįsta reakcijos paviršiaus analizės metodologija. Nustatyta optimali eksperimentinė kompostuojamo mišinio sudėtis: pradinis riebalų kiekis – 5 %, biopreparato ląstelių koncentracija – 10^9 KSV/g, struktūrinės medžiagos kiekis – 9,5 %. Padidinus pradinę riebalų koncentraciją nuo 5 % iki 20 %, riebalų skaidymo sparta sulėtėja apie 3 kartus. Suprojektuotas koncentruotų riebalinių atliekų utilizavimo aikštelės modelis. Kompostavimo proceso trukmė 1–1,5 metų.

Reikšminiai žodžiai: taukai, aliejus ir lajus, kompostavimas, biopreparatas, biodegradacija, matematinis modeliavimas, optimizacija.

РАЗРАБОТКА ТЕХНОЛОГИИ КОМПОСТИРОВАНИЯ СИЛЬНОЗАГРЯЗНЕННЫХ ЖИРОСОДЕРЖАЩИХ ОТХОДОВ С ИСПОЛЬЗОВАНИЕМ БАКТЕРИАЛЬНОГО ЛИПОЛИТИЧЕСКОГО ПРЕПАРАТА

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Р е з ю м е

В ЗАО "Biocentras" была создана новая технология компостирования отходов с высоким содержанием жира. Технология компостирования загрязненных жирами отходов основана на использовании микроорганизмов, окисляющих жиросодержащие вещества. Разработанная технология является привлекательной для более чистого производства/предупреждения загрязнения окружающей среды, а также удовлетворяет строгим экологическим и гигиеническим требованиям. Исследован процесс компостирования. Для оптимизации процесса применен метод математического моделирования. Опыты проведены в лабораторных условиях. Определен оптимальный состав смеси: начальное содержание жира – 5%, концентрация бактериальных клеток в препарате – 10^9 КОЕ/г, количество структурных материалов – 9,5%. Деградация жиров замедляется в 3 раза, если первоначальная концентрация жиров увеличивается от 5% до 20%. Разработан и построен прототип площадки для компостирования жировых отходов. Процесс компостирования длится от 1 до 1,5 лет.

Ключевые слова: жиры, растительные и животные масла, компостирование, бактериальный препарат, биоразложение, математическое моделирование, оптимизация.

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