

# PROCESS OF COMPOSTING FROM THE PARASITOLOGICAL POINT OF VIEW

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## 1 INTRODUCTION

Animal husbandry, in relation to humans, is connected with whole range of ecological and medical problems. Animals are one of the most frequent sources of environmental contamination. Manure and other wastes from agriculture often contain high concentration not only of animal but also of human pathogens. Animal excrements frequently contain bacteria of the family Enterobacteriaceae, protozoa and eggs or larvae of enteronematodes most of which are of zoonotic character. The most frequent way of transmission of parasitic diseases is through the contact (free-living animals and domestic ones), or through environment contaminated with various developmental stages (oocysts, eggs, larvae).

Utilisation and disposal of wastes generated on animal farms has been the subject of many investigations with regard to contamination of the environment with emissions, toxicity of treated wastes to plants but also potential survival and spreading of pathogenic agents. With regard to potential presence of pathogenic microorganisms in these substrates should be viewed as possible health hazard. One of the suitable methods for recycling organic wastes compatible with the environment is composting (Zhu, 2007; Ogunwande et al., 2008).

The generally known high tenacity of endoparasitic propagative stages and their resistance to various environmental biotic and abiotic factors led us to study the problems of the survival of parasitic germs during the composting process. Only relatively few studies investigated the impact of composting conditions on the survival of parasite stages. The study described here evaluated the effect of aerobic composting of different organic wastes (especially changes in physical and chemical parameters – temperature, -pH, moisture, amount of  $\text{NH}_4^+$ ,  $\text{N}_t$ ,  $\text{P}_t$ ) on the survival of parasitic agents. *A. suum* eggs have been chosen as model eggs. *A. suum* eggs are amongst the helminth eggs most resistant to environmental factors. Their cellular wall is enveloped with an outer layer formed by acid polysaccharides and proteins, central layer consisting of proteins (25%) and lipids (75%, particularly alpha glycosides). Thus this resistant cell wall protects *A. suum* eggs against effects of chemicals and drying (Eckert, 1992). Coccidia oocysts were chosen for comparison because they can also survive for a long time in the environment thanks to their double layered cell wall and are specific for particular species.

## 2 MATERIALS AND METHODS

Investigations were carried out under operating conditions in the pharmaceutical composting factory, brewery and in the poultry farm in the Slovak Republic to determine the effect of composting of different organic wastes on the survival of parasitic germs. The different waste types, potentially contaminated with parasitic and other pathogenic germs, were used for the recycling: poultry excrements, a by-product from Penicillin production (mycelium), sludge from waste water treatment plant, wastes from brewery, straw, sawdust and wood chips.

The composting process proceeded for 28 days in the thermophilic temperature range (from 49°C to 64°C) under operating conditions in the pharmaceutical composting factory. Throughout the experimental period, the ambient temperature ranged from 7.2°C to 8.7°C. A by-product from Penicillin production (mycelium), straw (which can be contaminated with parasite germs) and sawdust were composted in 2 compost channels (2 m wide and 25 m long) which were periodically turned. At the beginning of composting (day 0) and after exposure of substrate to the environment in the channels, samples for parasitological and physical and chemical examination were collected (days 1, 2, 3, 4, 7, 8 and 28 of composting).

The composting process proceeded either for 104 days (winter season) or for 62 days (summer season) in the thermophilic temperature range (from 41°C to 65°C during the winter and from 42.6°C to 71°C during the summer) under operating condition in the brewery. The ambient temperature ranged from -6°C to 12°C during the winter experiment and from 11°C to 31°C during summer experiment. The brewery sludge (sludge produced by

brewery wastewater treatment plant) and some bulky plant material (straw, wood chips and sawdust) were mixed in the ratio 50:10:20:20 and composted in piles. The piles were 50 m long, 1.5 m high and 2 m wide and were aerated mechanically (on days 26 and 98 of composting in winter and on days 26 and 50 in summer). Samples for parasitological and physical and chemical examination were collected from 3 places of the piles (the beginning, the middle and the end) after 0, 1, 4, 5, 6, 7, 36 and 104 days of composting during the winter and after 0, 3, 4, 5, 6, 7, 10, 11 and 62 days during the summer.

Chicken excrements and straw were used in the experiment. Chicken excrements were collected from a poultry farm near Košice. The animals were bred without bedding material. Excrements and straw were mixed and 6 piles (H1-H6) were built (1.5 m high, 5 m long and 2 m wide). The surface of the piles was not covered with any material to imitate natural conditions. The composting process in the piles proceeded from 17 days to 33 days in the thermophilic temperature range (up to 70°C). Piles were periodically turned. The aim of the study was to monitor the effect of aerobic composting of poultry excrements and straw in piles on the survival and development of model nonembryonated *A. suum* eggs.

We used the “artificial contamination of piles” approach to make sure that there is a sufficient number of positive samples in our observations.

Attenuated lines of *Eimeria acervulina*, *E. tenella* and *E. maxima* were used in the form of trivalent vaccine LIVACOX<sup>®</sup> T, which was prepared by Biopharm, Research Institute of Biopharmacy and Veterinary Drugs in the Czech Republic.

*A. suum* eggs were isolated by dissection of a distal uterine part of female pig ascaris. The distal uterine ends were then transferred to a glass homogenizer and processed. The water suspension of eggs was stored in an Erlenmeyer flask in a refrigerator at 4°C.

Coccidia oocysts and model eggs were inoculated by a micropipette into polyurethane carriers prepared according to Plachý & Juriš (1995), at a dose of 1 ml vaccine (30 000 – 50 000 oocysts) or of 1000 eggs per one carrier. The eggs were re-isolated from the inoculated carriers as follows: the carriers were cut into small pieces and washed in a mortar with 3 x 5 ml portions of saline, thoroughly stirred and filtered through a sieve into test tubes. After centrifugation, sediments were transferred to Petri dishes. The viability of exposed nonembryonated *A. suum* eggs was determined by incubation up to the embryonated stage in a thermostat at 26°C for 21 days. Petri dishes with *A. suum* eggs were aerated daily with micropipette. The developmental ability of *A. suum* eggs was compared with that of the control eggs which were kept in distilled water under aerobic conditions. The controls with eggs and oocysts were incubated in distilled water.

### 3 RESULTS AND DISCUSSION

TABLE 1 Damage of *A. suum* eggs and *Eimeria* oocysts during aerobic composting in the pharmaceutical composting factory

Exposure (days)	Damaged ( $\bar{x}$ % $\pm$ SD)	
	<i>A. suum</i> eggs	<i>Eimeria</i> oocysts
0 (control)	16.02 $\pm$ 2.61	9.11 $\pm$ 0.75
1	98.74 $\pm$ 1.18***	100***
2	100***	100***
3	100***	100***
4	100***	100***
7	100***	100***
8	100***	100***
28	100***	100***

\*\*\*Significance at the level P<0.001

In aerobic composting in the compost channels in thermophilic temperature range, high temperatures (up to 70°C) and changes of physical and chemical parameters resulted in total devitalisation of protozoa germs as early as after 24 hours and helminth germs after 48 hours (table 1). Total devitalisation of *A. suum* eggs in compost piles was observed within 7 days (Tables 2-3). The key factors for a viability of parasite eggs are temperature, humidity, changes in physical and chemical parameters of the substrate and a suitable carbon - nitrogen ratio (C:N - 15-30:1).

In contrast to our results Zhu (2007) detected that all roundworm eggs were destroyed after 63 days of composting.

TABLE 2 **Damage of *A. suum* eggs during aerobic composting in piles in winter and summer seasons under operating condition in the brewery**

Exposure (days)	Damaged <i>A. suum</i> eggs ( $\bar{x}$ % $\pm$ SD)	
	Winter season	Summer season
0 (control)	13.80 $\pm$ 2.61	12.62 $\pm$ 2.23
1	64.13 $\pm$ 3.46**	ND
3	ND	16.90 $\pm$ 9.75
4	88.30 $\pm$ 5.47***	45.33 $\pm$ 18.71*
5	92.50 $\pm$ 6.52***	84.36 $\pm$ 18.32***
6	100***	100***
7	100***	100***
10	ND	100***
11	ND	100***
36	100***	ND
62	ND	100***
104	100***	ND

ND – not detected, \*Significance at the level  $P < 0.05$ , \*\*Significance at the level  $P < 0.01$ , \*\*\*Significance at the level  $P < 0.001$

TABLE 3 **Damage of *A. suum* eggs during aerobic composting of chicken excrements in piles**

Exposure (days)	Damaged <i>A. suum</i> eggs ( $\bar{x}$ % $\pm$ SD)
0 (control)	16.02 $\pm$ 2.61
1	28.16 $\pm$ 1.18
2	58.26 $\pm$ 4.63**
3	69.11 $\pm$ 7.22***
4	98.50 $\pm$ 3.65***
7	100***
9	100***
14	100***
17	100***
24	100***

$\bar{x}$  - mean values, SD - standard deviation, \*\*Significance at the level  $P < 0.01$ , \*\*\*Significance at the level  $P < 0.001$

#### 4 CONCLUSIONS

Based on the results it may be stated that aerobic thermophilic composting process can be used for the devitalisation of endoparasite agents of farm animals with zoonotic nature in their excrements as well as human parasitic stages which are potentially present in sludge from waste water treatment plants. From the hygienic and epidemiologic point of view, wastes treated by aerobic thermophilic composting are safe and suitable for further use as fertiliser for direct application on the soil causing no subsequent contamination of surrounding ecosystem with endoparasite agents.

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