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THE EFFECT OF ZEOLITE ON MICROORGANISM COUNTS IN THE SOLID FRACTION OF PIG EXCRETA UNDER AEROBIC AND ANAEROBIC CONDITIONS

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Summary

The effect of zeolite on microorganism counts has been studied in the solid fraction of pig excreta under aerobic and anaerobic laboratory conditions. Microorganism counts after 24 hours after adding 1 and 2 % zeolite at anaerobic storing ranged within $3.8.10^6$ - $3.7.10^9$. In the control sample at anaerobic storing microorganism counts ranged within $9.9.10^6$- $3.8.10^9$. At aerobic storing after adding 1 and 2 % zeolite microorganism counts ranged within $3.7.10^6$ - $4.8.10^9$. The control at aerobic storing ranged within $6.5.10^6$ - $7.9.10^9$. The largest decrease in microorganism count was observed after 3 weeks in fecal coliform microorganisms. From week 6 onwards, no coliform or fecal coliform microorganisms were seen either at aerobic or aerobic storing. The largest decrease in mesophilic and psychrophilic microorganisms occurred after 3 months from the beginning of the study. Numbers of mesophilic and psychrophilic microroganisms ranged at anaerobic storing within $3.2.10^6$ - $8.0.10^6$ and in the control within $3.5.10^7$ - $3.3.10^7$. Microorganism counts at aerobic storing ranged within $3.3.10^6$ - $9.0.10^6$ and in the control within $4.3.\ 10^7$ - $8.8.10^7$.

Key words: zeolites, coliform, fecal coliform, mesophilic, psychrophilic microorganisms, aerobic, anaerobic storing, compost, pig excreta, solid fraction

Introduction

Large animal farms belong to largest environmental pollutants (Strauch, 1991). Farm animal excreta are by-products of animal production and the largest source of organic compounds that should be returned into soil. Therefore when treating them the attention should be paid to the fact that it is necessary to maintain the circulation of organic compounds and nutrients so that the same amount of utilizable compounds and nutrients would get back

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into soil as with direct application of untreated excreta (Jonáš, Petřiková, 1988). They also contain huge number of microorganisms which decompose organic compounds thus releasing energy needed for their living processes and growth.

Most excreta produced on pig farms are treated by aerobic processes in biological wastewater treatment plants. After treatment, the fluid fraction is discharged into recipient. The solid fraction usually contains high number of microorganisms and therefore it should be further treated. Composting proves to be a way of treatment. In practice, the solid fraction is stored in manure yards where it ripens under anaerobic conditions. At the same time, environment continues to be polluted.

The aim of the study was to investigate the effect of natural zeolite on microorganism counts in the solid fraction of pig excreta.

Materials and methods

Powder zeolite (clinoptilolite), natural aluminosilicate from Nižný Hrabovec, Slovakia was added to the solid fraction of pig liquid manure which was obtained after separation on vibration riddles at the first stage of cleaning treatment (granularity of main component of 0.125 – 0.5mm, pre-dried at 105°C) at the volume amount of 1 and 2 %. The samples were stored in plastic bags at the temperature of 18-22°C for 3 months under laboratory conditions. The temperature at the core of the sample was continuously recorded. During storing, excessive fluid was observed that was flowing out in the first 48 hours of storing. This trial has suggested conditions of practice where solid fraction is stored on manure yards.

Throughout the study, microbiological exminations of samples were focused on the observation of mesophilic, psychrophilic, coliform and fecal coliform microorganisms in samples stored under aerobic and anaerobic conditions in the solid fraction. The standard methods stated in STN (Slovak Technical Norm) 83 0531 and the method of Štěpánek (1982) were employed. For cultivation, MPA and Endo agar (Imuna, Šarišské Michaľany, Slovak Republic) were used. Incubation took place at the temperatures of 20°C, 37°C and 43°C, for 48 hours and 72 hours. The samples were mixed at the ratios 50 kg of solid fraction and 500 g zeolite and 50 kg of solid fraction and 1000 g zeolite. The samples stored under aerobic conditions were taken after mixing and returning into bags. After 24 hours, 3 weeks, 6 weeks and 3 month the samples (n=3) were diluted and inoculated onto solid substrates. Fraction without addition of 1 and 2 % zeolite served as control.

Results and discussion

The effects of 1 and 2 % natural zeolite on microorganism counts in the solid fraction of pig excreta under aerobic and anaerobic conditions are presented in Table 1-4.

Table 1. - MEAN NUMBER OF 3 CONCURRENT COUNTS (CFU/100 G) OF 4 BACTERIA STRAINS IN SOLID FRACTION OF PIG EXCREMENTS AFTER 24 HOURS OF TREATMENT WITH 1% AND 2% NATURAL ZEOLITE

After 24 hours	Mesophilic	Psychrophilic	Coliform	Fecal-coliform
Imput sample	$3.6 \cdot 10^8$	$7.3 \cdot 10^8$	$1.9 \cdot 10^7$	$9.8 \cdot 10^6$
Anaerobic +1% zeolite in bag	$3.7 \cdot 10^9$	$6.8 \cdot 10^8$	$3.2 \cdot 10^7$	$4.9 \cdot 10^6$
Anaerobic +2% zeolite in bag	$7.8 \cdot 10^8$	$3.8 \cdot 10^8$	$4.9 \cdot 10^7$	$3.8 \cdot 10^6$
Control anerobic	$3.8 \cdot 10^9$	$9.4 \cdot 10^8$	$3.9 \cdot 10^8$	$9.9 \cdot 10^6$
Aerobic +1% zeolite in bag	$4.8 \cdot 10^9$	$3.8 \cdot 10^8$	$7.8 \cdot 10^7$	$4.2 \cdot 10^6$
Aerobic +2% zeolite in bag	$5.8 \cdot 10^8$	$4.9 \cdot 10^8$	$6.5 \cdot 10^7$	$3.7 \cdot 10^6$
Control aerobic	$7.9 \cdot 10^9$	$8.8 \cdot 10^8$	$3.9 \cdot 10^7$	$6.5 \cdot 10^6$

Table 2. - MEAN NUMBER OF 3 CONCURRENT COUNTS (CFU/100 G) OF 4 BACTERIA STRAINS IN SOLID FRACTION OF PIG EXCREMENTS AFTER 3 WEEKS OF TREATMENT WITH 1% AND 2% NATURAL ZEOLITE

After 3 weeks	Mesophilic	Psychrophilic	Coliform	Fecal-coliform
Anaerobic +1% zeolite in bag	$7.6 \cdot 10^7$	$3.5 \cdot 10^6$	$4.1 \cdot 10^6$	-
Anaerobic +2% zeolite in bag	$4.3 \cdot 10^7$	$6.5 \cdot 10^6$	$4.1 \cdot 10^6$	-
Control anaerobic	$3.8 \cdot 10^6$	$4.5 \cdot 10^6$	$6.7 \cdot 10^6$	-
Aerobic +1% zeolite in bag	$4.6 \cdot 10^7$	$9.4 \cdot 10^6$	$3.8 \cdot 10^6$	-
Aerobic +2% zeolitet in bag	$9.2 \cdot 10^6$	$3.4 \cdot 10^6$	$4.3 \cdot 10^6$	-
Control aerobic	$7.4 \cdot 10^7$	$9.4 \cdot 10^6$	$3.8 \cdot 10^6$	-

Table 3. - MEAN NUMBER OF 3 CONCURRENT COUNTS (CFU/100 G) OF 4 BACTERIA STRAINS IN SOLID FRACTION OF PIG EXCREMENTS AFTER 6 WEEKS OF TREATMENT WITH 1% AND 2% NATURAL ZEOLITE

After 6 weeks	Mesophilic	Psychrophilic	Coliform	Fecal-coliform
Anaerobic +1% zeolite in bag	$6.6 \cdot 10^6$	$1.0 \cdot 10^7$	-	-
Anaerobic +2% zeolite in bag	$7.2 \cdot 10^6$	$5.2 \cdot 10^6$	-	-
Control anaerobic	$5.6 \cdot 10^7$	$1.2 \cdot 10^7$	-	-
Aerobic +1% zeolite in bag	$3.6 \cdot 10^7$	$6.4 \cdot 10^7$	-	-
Aerobic +2% zeolite in bag	$5.8 \cdot 10^7$	$7.4 \cdot 10^7$	-	-
Control aerobic	$9.2 \cdot 10^7$	$8.6 \cdot 10^7$	-	-

Table 4. - MEAN NUMBER OF 3 CONCURRENT COUNTS (CFU/100 G) OF 4 BACTERIA STRAINS IN SOLID FRACTION OF PIG EXCREMENTS AFTER 3 MONTHS OF TREATMENT WITH 1% AND 2% NATURAL ZEOLITE

After 3 months	Mesophilic	Psychrophilic	Coliform	Fecal-coliform
Anaerobic +1% zeolite in bag	$8.0 \cdot 10^6$	$7.0 \cdot 10^6$	-	-
Anaerobic +2% zeolite in bag	$7.2 \cdot 10^6$	$3.2 \cdot 10^6$	-	-
Control anaerobic	$3.3 \cdot 10^7$	$3.5 \cdot 10^7$	-	-
Aerobic +1% zeolite in bag	$6.4 \cdot 10^6$	$3.3 \cdot 10^6$	-	-
Aerobic +2% zeolite in bag	$3.6 \cdot 10^6$	$9.0 \cdot 10^6$	-	-
Control aerobic	$8.8 \cdot 10^7$	$4.3 \cdot 10^7$	-	-

From the Tables it follows that microorganism counts in input sample prior to the addition of 1 and 2 % zeolite ranged within $9.8.10^6 - 7.3.10^8$. Microorganism counts after 24 hours after adding 1 and 2 % zeolite at anaerobic storing ranged within $3.8.10^6 - 3.7.10^9$. In the control sample at anaerobic storing microorganism counts ranged within $9.9.10^6$- $3.8.10^9$. At aerobic storing after adding 1 and 2 % zeolite microorganism counts ranged within $3.7.10^6$ - $4.8.10^9$. The control at aerobic storing ranged within $6.5.10^6$ - $7.9.10^9$. The largest decrease in microorganism count was observed after 3 weeks in fecal coliform microorganisms. From week 6 onwards, no coliform or fecal coliform microorganisms were seen either at aerobic or aerobic storing. The largest decrease in mesophilic and psychrophilic microorganisms occurred after 3 months from the beginning of the study. Numbers of mesophilic and psychrophilic microroganisms ranged at anaerobic storing within $3.2.10^6$ - $8.0.10^6$ and in the control within $3.5.10^7$ - $3.3.10^7$. Microorganism counts at aerobic storing ranged within $3.3.10^6$ - $9.0.10^6$ and in the control within $4.3.\ 10^7 - 8.8.10^7$.

As concerns the possibility of using natural zeolites in the protection of environment (Amon et al., 1997) and their effect on the survival of pathogenic microorganisms and after studying the available literary data on the effects of zeolitic materials in the process of the treatment of excreta we can say that their effect on microorganisms has been little explored and only little attention is paid to their beneficial effect on the removal of suspended compounds and microorganisms. According to Mumpton and Fishman (1977) the inner structure of zeolite which is formed by linked little canals and cavities does not allow the penetration of bacteria. Our knowledge obtained shows the beneficial effect of zeolite on hygienically important groups of microorganisms, namely on coliform and fecal coliform micro-organisms, also in the composted substrate that we have observed under laboratory conditions.

The temperatures in a composted substrate are determined by the rate of biological processes taking place in it. According to Tiquia (1996) microbial

population in a compost determines the rate of composting, it influences the quality of the product and is responsible for most physical, chemical and microbiological changes in the compost. Temperatures above 55°C support hygienization of compost, temperatures from 45 to 55°C have a beneficial effect on biodegradation rate and those from 35 to 40° C conribute to maximum microbial variability in composting processes (Stentiford, 1996).

The highest temperatures at the core of substrates were recorded in the control substrate throughout the first 6 weeks of the study. In the second half of the study in the substrate with the addition of 1 % zeolite the temperatures were higher throughout the entire period of observation as compared to both control and substrate with 2 % addition of zeolite. The lowest temperature was recorded in the control at the end of the period of observation.

The highest temperature observed at the core of substrates in the control (31.5 °C) is not sufficient for the liquidation of pathogenic microorganisms. It cannot be expected that temperature would be raised to the required level either when composting larger amounts of substrates under similar conditions (Strauch and Ballarini, 1994).

Conclusion

As concerns the possibilty of using natural zeolites in the protection of environment and their effect on the survival of pathogenic microorganisms and after studying the available literary data on the effects of zeolitic materials in the process of the treatment of excreta we can say that their effect on microorganisms has been relatively little explored and only little attention is paid to their beneficial effect on the removal of suspended compounds and microorganisms. Our knowledge obtained indicates the beneficial effect of zeolite on hygienically important groups of microorganisms, namely on both coliform and fecal microorganisms, also in composted substrate that we observed under laboratory conditions.

Microorganism counts prior to the addition of zeolite ranged within $9.9 \cdot 10^6$ - $7.3 \cdot 10^8$. In the control sample at anaerobic storing, microorganism counts ranged within $3.8 \cdot 10^9$ - $9.9 \cdot 10^6$ and at aerobic storing they ranged within $7.9 \cdot 10^9$ - $6.5 \cdot 10^6$.

Our knowledge obtained shows out the beneficial effect of zeolite on hygienically important groups of microorganisms, namely on both coliform and fecal coliform microorganisms and also in composted substrate that we observed under laboratory conditions. From this it follows that under such conditions the solid fraction cannot be applied to agricultural soil without limitations.

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UTJECAJ DODATAKA ZEOLITA U KRUTU FAZU SVINJSKOG GNOJA NA BROJ MIKROORGANIZAMA POD AEROBNIM I ANAEROBNIM UVJETIMA

Sažetak

Istraživan je utjecaj dodataka 1 % i 2 % zeolita u krutu fazu svinjskog gnoja na broj mikroorganizama pod anaerobnim i aerobnim laboratorijskim uvjetima skladištenja. Nakon 24 sata, kod anaerobnog skladištenja i dodataka 1 odnosno 2 % zeolita broj mikroorganizama kretao se od 3.8×10^6 do 3.7×10^{10}. Kontrolni uzorak je pod istim uvjetima sadržavao od 9.9 x 106 do 3.8×10^9 mikroorganizama. Kod aerobnog držanja gnoja i dodataka 1 i 2 % zeolita broj mikroorganizama se kretao od 3.7 x 106 do 4.8×10^9. Broj mikroorganizama u kontroli iznosio je 6.5×10^6 do 7.9×10^9. Dodatak zeolita uzrokovao je smanjenje broja psihrofilnih, mezofilnih, koliformnih i fekalnih koliformnih bakterija, no najveće smanjenje uočeno je nakon tri tjedna kod fekalnih koliforma. Nakon šest tjedana nisu se više mogli dokazati ni ukupni koliformi ni fekalni koliformi, ni kod anaerobnog ni kod aerobnog skladištenja gnoja. Najveće smanjenje broja mezofilnih i psihrofilnih bakterija desilo se nakon tri mjeseca skladištenja gnoja. Njihov se broj kretao od 3.2×10^6 do 8.0×10^6, a u kontroli od 3.5×10^7 do 3.3×10^7. Broj mikroorganizama pri aerobnom držanju iznosio je 3.3×10^6 do 9.0×10^6, a u kontroli 4.3×10^7 do 8.8×10^7. Pokus ukazuje na učinkovito djelovanje zeolita na smanjenje broja higijenski važne skupine mikroorganizama u kompostiranom supstratu.

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