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Effect of adding *Lactobacillus plantarum* and soluble carbohydrates to swine manure on odorous compounds, chemical composition and indigenous flora

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Abstract: Manure odor, which results in the increasing complaints and lawsuits, has increased the tension among swine producers and surrounding residents. The effects of *Lactobacillus plantarum* and different rates of soluble carbohydrates additions to swine manure on odorous compounds, chemical compounds and indigenous flora were evaluated. Additions were calculated on dried manure weight basis. Variables monitored included ammonia (NH_3), hydrogen sulfide (H_2S), odor offensiveness, pH, ammonium nitrogen(NH_4^+-N), volatile fatty acids (VFAs), urease and indigenous flora. The results indicated that the combination of *L. plantarum* and soluble carbohydrates dramatically reduced manure pH. Lower pH resulted in the reduction of NH_3 volatilization (34.6%—92.4%, $P<0.01$), the increases of H_2S ($P<0.05$) and NH_4^+-N (5.3%—17.5%, $P<0.05$). In addition, *L. plantarum* and soluble carbohydrates additions significantly reduced odor offensiveness, those VFAs related to malodor indicators(valeric acids, 12.3%—47.7%, $P<0.05$; *iso*-valeric, 3.5%—23.8%) and the main microorganisms responsible for odor production, with the number of *Eubacteria* in swine manure reducing by 4.9%, 11.6%, 17.4%, 34.1% and 32.2% respectively.

Keywords: *Lactobacillus plantarum*; soluble carbohydrates; swine manure; odorous compounds; indigenous flora

Introduction

Over the last few decades, confined hog feeding operations has increased tremendously. Although the intensification have been proven economically feasible, the production of large quantities of excreta in a small area also generates serious environmental concerns as a result of nitrogen(N) loss, manure odor, surface and ground water pollution and the potential for transmission of pathogens (Jongbloed and Lenis, 1989; Lefcourt and Meisinger, 2001). Odor, which is a sensation stimulation and difficult to quantify, is one concern to which the general public gives the most attention. At present, due to low cost and application flexibility, livestock producers and researchers have focused on the use of chemical and biological additives as alternative methods to control manure odor (Delaune *et al.*, 2004). Biological additives, generally contain mixed cultures of enzymes or microorganisms that enhance the solids and reduce the volatilization of ammonia, hydrogen sulfide and the emission of other odorous compounds. However, due to the complexity of physical and chemical conditions in swine manure, inoculated microorganisms can not grow better than indigenous flora unless environmental conditions are in the optimum growth range for the added bacteria (Zhu, 2000). Although *Lactobacillus plantarum* can produce lactic acid, it is very difficult to flourish in swine manure. In contrast, in addition to offering carbon (C) source and increasing C/N ratio, soluble carbohydrates can reduce manure pH by stimulating the indigenous microorganisms to produce organic acids, whereas it can not sustain the level (Burgess *et al.*, 1998). Thus, *L. plantarum* and soluble carbohydrates combined may be a good remedy for this. The study conducted by McCrory and Hobbs (2001) showed that using *L. plantarum* and glucose combined reduced the pH of pig slurry from 8 to 6. However, no information is available on the effects of these amendments on odorous compounds and biochemical composition in swine manure. In this study, *L. plantarum* and soluble carbohydrates were used as manure amendments to evaluate effects on odorous com- pounds, chemical composition and indigenous flora in swine manure.

1 Materials and methods

1.1 Experimental materials

Fresh swine manure(moisture content: 72%; C/N: 10.2) was collected for testing from Institute of Animal Sciences (IAS), Chinese Academy of Agricultural Sciences (CAAS) swine farm. *L. plantarum* stored in our laboratory was obtained by 12 h fermentation cultivation using APT agar, with the number of effective activated bacteria above 1.8×10^6/ml (Chen, 1995). The composition of the medium is as follows: peptone 10 g/L, beef extract 10 g/L, yeast extract 5 g/L, glucose 5 g/L, sodium acetate

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anhydrous 5 g/L, ammonium citrate dibasio 2 g/L, $MgSO_4 \cdot 7H_2O$ 0.2 g/L, $MnSO_4 \cdot 5H_2O$ 0.2 g/L, $K_2HPO_4 \cdot 3H_2O$ 2 g/L, $CaCO_3$ 20 g/L and Tween80 1 g/L. The pH of the medium was adjusted to 6.8. Chemicals were purchased from Beijing Chemical Regents Company. Cornstalk(moisture: 4%; C/N: 58) were from China Agriculture University Science Park.

1.2 Experimental procedure

After collection, fresh swine manure and 15% cornstalk powder(at 600 g per bottle) were placed in 3 L sterilized triangle bottle, airproofed, incubated at 30—37℃ and allowed to ferment for a period of 18 d. The treatments used for this study were 15% sterilized water control, 15% *L. plantarum* alone, 15% *L. plantarum* plus 5% sucrose, 15% *L. plantarum* and 5% glucose combined, 15% *L. plantarum* with 10% sucrose, and 15% *L. plantarum* in conjunction with 10% glucose. Additions were calculated on dried manure weight basis. There were three replications for each treatment.

1.3 Air sampling and analysis

Aerial ammonia, hydrogen sulfide were monitored at the day 3, 6, 9, 12, 15 and 18 using spectrophotometry at a wavelength of 697 nm and 665 nm respectively (Smith *et al.*, 2004). Ammonia and hydrogen sulfide in the manure bottle were sucked into tubes filled with 0.005 mol/L dilute sulfuric acid and polyvinyl alcohol ammonium phosphate solutions respectively using vacuum pump at a rate of 2 L/min. The inlet of tubes filled with absorbents was connected to the triangle manure bottle. Ammonia and hydrogen sulfide were sucked into absorbents due to the negative pressure created by vacuum pump inside the bottle.

1.4 Odor offensiveness analysis

Due to the complicated component of odorous gas, Schiffman (1998) and Armstrong *et al.* (2000) reported the human nose is an acceptable instrument for sensing odor. In this study, odor offensiveness was rated according to a scale of 1 to 5 in which 1 represents no offensive odor, 2 mildly offensive, 3 moderately offensive, 4 strongly offensive odor and 5 extremely offensive odor (Mackie *et al.*, 1998) at the day 3, 6, 9, 12, 15 and 18. A panel consisting of 10 volunteers was utilized for the evaluation analysis (Otto *et al.*, 2003). As this study did not require volunteers to identify finer differences between the odors, no training was necessary. The tasks of volunteers were to identify the presence of odor and odor offensives. However, a selection criterion was volunteers being familiar with livestock manure odors and the readings of some volunteers were discarded if it was reported they had cold, allergies or had meal less than an hour before a odor evaluation session.

1.5 Manure sampling and analysis

1.5.1 pH and NH_4^+-N

The manure were collected at the day 3, 6, 9, 12, 15 and 18 for determination of pH and NH_4^+-N. The pH was determined with a combination glass-calomel electrode, which was inserted directly into diluted manures (1:1, swine manure to deionized water, w/w) and allowed to stabilize for about 3 min. Manure samples were analyzed for NH_4^+-N using a technicon autoanalyzer and the indophenol blue method(Lefcourta and Meisinger, 2001).

1.5.2 Urease, volatile fatty acids(VFAs) and indigenous flora

At the end of the trials, the manure were collected and variables monitored included urease, VFAs and indigenous flora. Nessler's colorimetry was used for urease analysis (Miller and Varel, 2003). The determination process of VFAs is as follows: 2 g manure samples for each treatment were diluted with 8 ml of distilled water, added two drops of concentrated HCl, mixed, centrifuged at 17500 g for 15 min at 4 ℃, and stored at -20℃ until use. A gas chromatography with flame ionization detector and nitrogen as a carrier gas at a flow rate of 25 ml/min was used. Samples was injected (2 μl) and the temperature for the column, injector, and detector were 60, 250 and 250℃ respectively. A standard solution (10 mmol/L for each VFA) including acetate, proionate, butyrate, isovalerate, valerate was injected at a dose of 2 μl for each VFA. Indigenous flora were composed of *E. coli*, *Lactobacilli*, *Clostridia* and *Eubacteria*. The medium for them were EMB agar, APT agar, Clostrisel agar and ES agar respectively(Li, 1991).

1.6 Statistical analysis

Variance (ANOVA) procedures in SAS was to determine significant effects, with means compared for significance at the 0.05 level using Fisher's protected LSD.

2 Results and discussion

2.1 Odor offensiveness

Odorous compounds are produced under prevailing anaerobic conditions. O'Neil and Phillips (1992) summarized 168 odorous compounds identified in livestock odors including ammonia, amines, volatile fatty acids, sulfur-containing compounds, skatole, phenol, alcohol, and carbonyl compounds. The concentration of specific gases in an odorous air sample may be identified using gas chromatography, mass spectrometry (GC-MS), and other methods. However, an odor is the sensations when a mixture of odorants stimulate the human sensory apparatus-the nose, and determination of any individual odorant conc- entrations are not sufficient to describe character of that odor (Jones *et al.*, 1994). Therefore, besides measurement of specific odorous gases, sensory method was used for rating odor offensiveness. Since a certain time is necessary for inoculated *Lactobacillus plantarum* (LP) to become fully activated and reach dominant levels, only the odor offensiveness data for the last sampling set are presented. Mean odor offensiveness rankings were 2.30, 2.16, 1.24 and 1.13 for manure treated 15% LP plus 5% sucrose, 15% LP and 5% glucose combined, 15% LP plus 10% sucrose, 15% LP along with 10% glucose respectively. Mean odor offensiveness rankings were 3.24 and 3.97 for manure treated with 15% LP alone and 15% sterilized water control. Odor offensiveness rankings for each treatment are reported as log odds ratio relative to the 15% sterilized water control (Fig.1). Compared with the control, all the treated significantly reduced odor offensiveness at a significant level of $P<0.05$ for statistical t test at the end of this study. Moreover, increasing soluble carbohydrates application rates resulted in decreasing odor offensiveness. However, when applied at the same rate, there were not significant difference for odor offensiveness between sucrose and glucose.

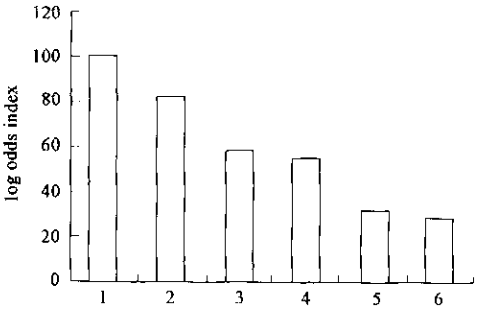

Fig.1 Odor offensiveness of manure. log odds index are relative to the control (index=100) and indicates the likeilhood of increased or decreased odor offensiveness

1. control; 2. LP; 3. LP+5% sourose; 4. LP+5% glucose; 5. LP+10% sucrose; 6. LP+10% glucose

2.2 pH values

The pH of LP and soluble carbohydrates combined remained below 5.2 for the entire test process(Fig.2). The ending pH of manure treated with LP and soluble carbohydrates in combination was significantly lower than the control and manure treated with LP alone, with increasing soluble carbohydrates applications rates resulting in the decrease of pH values. At the end of the trials, LP and 10% sucrose combined reduced manure pH to near 4.0, a reduction of pH 3.8 units over the sterilized control. McCrory and Hobbs (2001) reported that soluble carbohydrates offer a less-hazardous alternative to direct acid addi- tion, inducing a reduction in livestock slurry pH by stimulating the indigenous anaerobic microorganisms to produce organic acids. Subair *et al.* (1995) used several different sucrose concentrations to indirectly reduce the pH of pig slurry with the highest addition rate of 11% reducing slurry pH to 3.5. Theses results agreed with that of this study.

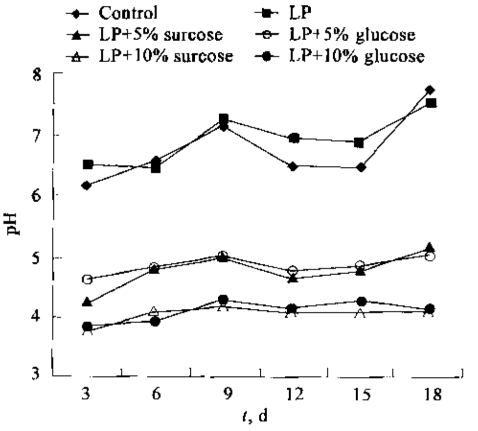

Fig.2 Effect of *Lactobacillus plantarum*(LP) and soluble carhydrates on pH of swine manure

2.3 Aerial ammonia and hydrogen sulfide

LP and soluble carbohydrates in combination additions significantly lowed ammonia (NH_3) volatilization rates. At the end of the study, compared with the control, LP plus 10% sucrose, LP and 10% glucose combined reduced NH_3 volatilization rates by 92.4% and 92.2% respectively (Fig.3). Study(Clemens *et al.*, 2002) showed that the addition of readily degradable organic compounds into the manure could reduce the pH values of manure and this resulted in less NH_3 volatilization.

However, the reduction of manure pH increased hydrogen sulfide(H_2S) emissions(Fig.4). Although the additions of sucrose and glucose at the 10% rate increased H_2S emissions by 45.9% and 48.6% respectively compared with the control, at the end of the trials H_2S emission for manure treated with the high rates of soluble carbohydrates remained below 0.55 mg/m^3.

2.4 Ammonium nitrogen and urease

Both the addition of *Lactobacillus plantarum*(LP) alone and LP plus soluble carbohydrates combined into the manure increased the levels of NH_4^+-N(Fig.5).

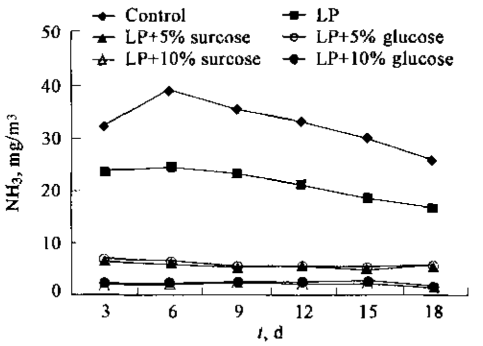

Fig.3 Effect of *Lactobacillus plantarum* (LP) and soluble carbohydrates on NH_3 volatilization of swine manure

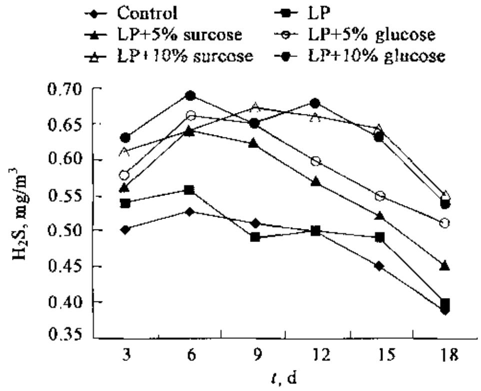

Fig.4 Effect of *Lactobacillus plantarum* (LP) and soluble carbohydrates on H_2S emission of swine manure

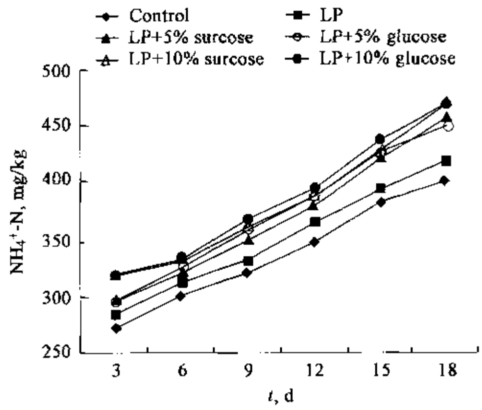

Fig.5 Effect of *Lactobacillus plantarum* (LP) and soluble carbohydrates on NH_4^+-N of swine manure

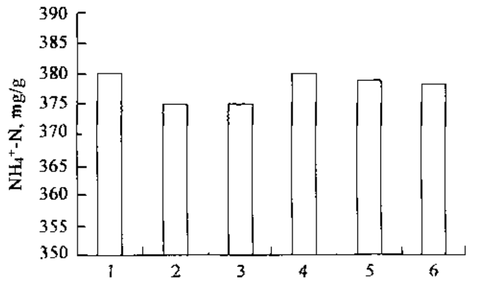

Fig.6 Effect of *Lactobacillus plantarum* (LP) and soluble carbohydrates on urease activity of swine manure
The bars are the same as Fig.1

In comparison with the control, the ending levels of NH_4^+-N increased by 5.3%, 13.5%, 12.5%, 17.5% and 17.0% for manure treated with LP alone, 5% sucrose, 5% glucose, 10% sucrose and 10% glucose respectively. Higher NH_4^+-N also suggested lower ammonia losses from swine manure and thus had a greater overall fertilizer value (Moore *et al.*, 1996). Moore *et al.*(2000) and Otto *et al.*(2003) reported that increases in manure NH_4^+-N were probably due to the acidification of the manure, which converted NH_3 to NH_4^+-N (i.e., NH_4^+-N to NH_3 ratio increasing with an decrease in manure pH) and thus reduced gaseous losses of N.

However, the addition of LP and soluble carbohydrates into the manure did not change urease values of the treated groups significantly according to statistical *t* test (Fig.6). Little is known about the relationship between urease and odor offensiveness, more research is needed to understand the role of urease in manure odor generation.

2.5 Volatile fatty acids and indigenous flora

In general, there are two approaches to measure odor: (1) to measure the concentration of specific gases in an air sample and (2) to use the human nose to perceive odor. Unfortunately, these two approaches are not well correlated. When measuring odor, what one hopes to quantify is the human response to the stimulus. If a chemical compound that is easily measured and correlates well with the response of an odor panel, it can be used as an indicator of odor. Several studies (Lunn and van De Vyver, 1977; Spoelstra, 1980) have showed that ammonia and hydrogen sulfide is a poor parameter in evaluating odor offensiveness. Barth and Polkowski (1974) reported that the volatile organic acids correlated best with the odor offensiveness.

In this study, the additions of *Lactobacillus plantarum* and soluble carbohydrates into swine manure increased the concentrations of acetic acid and propionic acid, but decreased the concentrations of butyric acid, valeric acid and *iso*-valeric acid(Table 1). As compared with the control, the concentrations of valeric acid in the treated manure reduced by 12.3%, 33.9%, 38.5%, 44.6% and 47.7% for LP alone, 5% sucrose, 5% glucose, 10% sucrose, 10% glucose groups respectively at a significance level of $P<0.05$ for *t* test. In addition, LP and 10% sucrose combined addition to swine manure resulted in the lowest concentration of *iso*-valeric acid among all treatments, with a 25.2% reduction over the control. Spoelstra (1980) reported that acetic and propionic acid concentrations have been considered unimportant when investigating odor quality and that reduction in the long-chain and branch-chained VFA has the potential to reduce odor from swine manure. Study conducted by Zhu and Jacobson(1999) and Zahn *et al.* (1997) also showed that VFAs with long carbon chains or branching could be a more suitable odor indicator than short chain acids and the acids in this group include *iso*-butyric, valeric, *iso*-valeric, caproic, and *iso*-caproic acids. In the study, the increasing concentrations of acetic acid and propionic acid did not result in the increasing odor offensiveness. In contrast, reduction in butyric, valeric and iso-valeric acids concentrations leaded to a lower odor offensiveness. The results agreed with that of others researchers (Zhu and Jacobson, 1999; Zahn *et al.*, 2001).

Determination of major malodor indicators for swine manure and the related bacterial genera have been extensively investigated for years. At present, polymerase chain reaction(PCR) which amplified 16S rDNA technology was used as a tool for the isolation of microorganisms responsible for odor production in swine manure (Ouwerkerk and Klieve, 2001). The results of these examinations indicate that the predominant culturable microorganisms from swine manure are obligately anaerobic, low mol percentage G+C Gram-positive bacteria who are members of *Clostridial*, *Eubacterial*, *Lactobacillus* and *Streptococcus* (Whitehead and Cotta, 2001; Cotta *et al.*, 2003). Among all culturable bacterial genera, *Clostridium* (the widest temperature range for growth) and *Eubacterium* (the largest population) are considered as the most likely source of the sickly-sweet nuisance odors (particularly from long carbon chains or branching VFAs (Zhu and Jacobson, 1999; Ouwerkerk and Klieve, 2001). Therefore, it is assumed controlling these bacterial growth will help reduce malodor generation(Zhu, 2000).

In this study, all treated groups reduced the counts of *E.coli* and *Eubacteria*, but increased *Lactobacilli* counts (Table 2). In comparison with the control, the counts of *Eubacteria* reduced by 4.9%, 11.6%, 17.4%, 34.1% and 32.2% for *Lactobacillus plantarum* alone, 5% sucrose, 5% glucose, 10% sucrose, 10% glucose groups respectively.

Table 1 Effect of *Lactobacillus plantarum* (LP) and soluble carbohydrates on the production of the main volatile fatty acids in swine manure

Treatment	Concentration of VFAs, mmol/L				
	Acetic acid	Propionic acid	Butyric acid	Valeric acid	*iso*-Valeric acid
Control	14.42±0.29	5.55±0.20	4.06±0.62	1.30±0.12	1.43±0.05
LP	18.54±0.50	5.76±0.30	4.01±0.67	1.14±0.14	1.38±0.05
LP−5% sucrose	18.87±0.44	6.63±0.32	3.98±0.47	0.86±0.07	1.21±0.07
LP+5% glucose	18.81±0.23	6.89±0.44	3.84±0.42	0.80±0.10	1.24±0.10
LP+10% sucrose	19.96±0.53	7.74±0.59	3.86±0.38	0.72±0.08	1.07±0.07
LP+10% glucose	20.11±0.51	7.90±0.35	3.92±0.36	0.68±0.09	1.09±0.10

Table 2 Effect of *Lactobacillus plantarum*(LP) and soluble carbohydrates on bacterial counts in the swine manure

Treatment	Bacterial genera counts, lg cfu/g			
	Lactobacilli	*E.coli*	*Eubacteria*	*Clostridia*
Control	7.41±0.34	6.56±0.51	10.03±0.37	4.22±0.33
LP	8.07±0.28	6.01±0.42	9.54±0.60	4.98±0.66
LP+5% surcose	8.36±0.43	4.65±0.29	8.87±0.71	4.30±0.42
LP+5% glucose	8.44±0.35	4.47±0.41	8.56±0.50	4.34±0.25
LP+10% surcose	9.47±0.36	4.00±0.44	6.61±0.42	3.62±0.47
LP+10% glucose	9.62±0.28	4.14±0.56	6.80±0.30	3.59±0.54

3 Conclusions

The result of this study indicate that *L. plantarum* and soluble carbohydrates treatment of swine manure can reduce odor offensiveness, manure pH and ammonia volatilization. *L. plantarum* and soluble carbohydrates additions reduced ammonia volatilization significantly (by 34.6%—92.4%). The greatest ammonia volatilization reduction occurred in *L. plantarum* and 10% sucrose combined by reducing the pH of the manure to near 4.0. The reduction of pH resulted in a higher NH_4^+-N (5.3%—17.5%) in the treated manure over the control and thus had a greater overall fertilizer value. Others benefits of *L. plantarum* and soluble carbohydrates treatments are reduction in VFAs related to malodor indicators (valeric acids, 12.3%—47.7%, $P<0.05$; *iso*-valeric, 3.5%—23.8%) and the main microorganisms responsible for odor production, with the greatest reduction in the number of *Eubacteria* (34.1%) occuring in *L. plantarum* and 10% sucrose combined over the control.

The use of *L. plantarum* and soluble carbohydrates amendments to swine manure offers the potential for reducing ammonia emissions, odor offensiveness and related flora responsible for odor production.

References:

Armstrong T A, Williams C M, Spears J W *et al.*, 2000. High dietary copper improves odor characteristics of swine waste [J]. J Anim Sci, 78: 859—864.

Barth C L, Polkowski L B, 1974. Identifying odorous components of stored dairy manure[J]. Trans ASAE, 17: 737—741; 747.

Burgess R P, Carey J B, Shafer D J, 1998. The impact of pH on nitrogen retention in laboratory analysis of boiler litter[J]. Poultry Sci, 77: 1620—1622.

Chen T S, 1995. Manufacture and application of microbial medium[M]. Beijing: China Agriculture Press.

Clemens J, Bergman S, Vandre R, 2002. Reduced ammonia emissions from slurry after self-acidication with organic supplements [J]. Environ Technol, 23(4): 429—435.

Cotta M A, Whitehead T R, Zeltwanger R L, 2003. Isolation, characterization and comparison of bacteria from swine faeces and manure storage pits [J]. Environmental Microbiology, 5: 737—745.

Delaune P B, Moore P A, Daniel T C *et al.*, 2004. Effect of chemical and microbial amendments on ammonia volatilization from composting poultry litter[J]. J Environ Qual, 33: 728—734.

Jones M, Watts P J, Smith R J, 1994. A mobile dynamic olfactometer for feedlot odour studies[J]. Applied Engineering in Agriculture, 10: 417—423.

Jongbloed A M, Lenis N P, 1989. Environmental concerns about animal manure[J]. J Anim Sci, 76: 2641—2648.

Lefcourt J M, Meisinger J J, 2001. Effect of adding alum or zeolite to dairy slurry on ammonia volatilization and chemical composition [J]. J Dairy Sci, 84: 1814—1821.

Li Y L, 1991. Manual of medium[M]. Changcun: Ji Lin Sciences Press.

Lunn F, van De Vyver J, 1977. Sampling and analysis of air in pig house[J]. Agric and Environ, 3: 159—169.

Mackie R I, Stroot P G, Varel V H, 1998. Biochemical identification and biological origin of key odor compounds in livestock waste [J]. J Anim Sci, 76: 1331—1342.

McCrory D F, Hobbs P J, 2001. Additive to reduce ammonia and odor emissions from livestock wastes: A review [J]. J Environ Qual, 30: 345—355.

Miller D N, Varel V H, 2003. Swine manure composition affects the biochemical origins, composition, and accumulation of odorous compounds[J]. J Anim Sci, 81: 2131—2138.

Moore P A, Daniel T C, Edwards D R *et al.*, 1996. Evaluation of chemical amendments to reduce ammonia volatilization from poultry litter[J]. Poult Sci, 75: 315—320.

Moore P A, Daniel T C, Edwards D R, 2000. Reducing phosphorus runoff and inhibiting ammonia loss from poultry manure with aluminum sulfate[J]. J Environ Qual, 29: 37—49.

O'Neil D H, Phillips V R, 1992. Review of the control of odor nuisance from livestock buildings: Part 3: Properties of the odourous substances which have been identified in livestock wastes or in the air around them [J]. Journal of Agricultural Engineering Research, 53: 23—50.

Otto E R, Yokoyama M, Hengemuehle S *et al.*, 2003. Ammonia, volatile fatty acids, phenolics, and odor offensiveness in manure from growing pigs fed diets reduced in protein concentration[J]. J Anim Sci, 81: 1754—1763.

Ouwerkerk D, Klieve A V, 2001. Bacterial diversity within feedlot manure[J]. Anaerobe, 7: 59—66.

Schiffman S S, 1998. Livestock odors: Implications for human health and well-being[J]. J Anim Sci, 76: 1343—1455.

Smith D R, Moore P A, Haggard B E *et al.*, 2004. Effect of aluminium chloride and dietary phytase on relative ammonia losses from swine manure[J]. J Anim Sci, 82: 605—611.

Spoelstra S F, 1980. Origin of objectionable odorous components in piggry wastes and the possibility of applying indicator components for studying odour development[J]. Agric Env, 5: 241—260.

Subair S, 1995. Reducing ammonia volatilization from liquid hog manure by using organic amendments [M]. Montreal: McGill Univ Press.

Sutton A L, Kephart K B, Verstegen M W A *et al.*, 1999. Potential for reduction of odorous compounds in swine manure through diet modification[J]. J Anim Sci, 77: 430—439.

Whitehead T R, Cotta M A, 2001. Characterisation and comparison of microbial populations in swine faeces and manure storage pits by 16S rDNA gene sequence analyses[J]. Anaerobe, 7: 181—187.

Zahn J A, Dispiroto A A, Do Y S *et al.*, 2001. Correlation of human olfactory responses to airborne concentration of malodorous volatile organic compounds emitted from swine effluent [J]. J Environ Qual, 30: 624—634.

Zhu J, Jacobson L D, 1999. Correlating microbes to major odorous compounds in swine manure[J]. J Environ Qual, 28: 737—744.

Zhu J, 2000. A review of microbiology in swine manure odor control[J]. Agriculture, Ecosystems and Environment, 78: 93—106.

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