J. agric. Engng Res. (2001) **80** (4), 365–371
doi:10.1006/jaer.2001.0745, available online at http://www.idealibrary.com on IDEAL®
SE—Structures and Environment

Effect of Batch Aeration-Treatment on the Solubility of Phosphorus in Pig Manure

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(Received 24 November 2000; accepted in revised form 26 July 2001; published online 11 September 2001)

Experiments were conducted to investigate the effect of temperature on phosphorus removal from pig manure during batch aeration and the mechanism(s) involved in its removal. Five temperature regimes, 5, 10, 15, 20 and 25°C, were investigated. The resulting effects on pH, volatile fatty acids levels (VFAs), and ortho-phosphate (ortho-P) in the manure were investigated. Results indicated that manure pH was the dominant factor influencing the levels of soluble ortho-P in the manure. Two different mechanisms that effected pH change were identified. Immediately after initiating aeration (*i.e.* during the first 1·5 days—phase I), a rapid rise in pH caused a rapid decline in manure-soluble ortho-P (coefficient of determination R^2 of 0·90). The manure pH beyond the first 1·5 days (phase II aeration) seemed to depend more on the VFAs levels. It was concluded that besides any phosphorus immobilization/release by the microbial biomass in phase II, the VFAs levels in the manure significantly explained (R^2 of 0·83) most of the biologically mediated chemical precipitation/dissolution of ortho-P. Temperature affected soluble ortho-P status only in phase II of aeration. High manure temperatures reduced aeration efficiency but promoted the production of VFAs. The resulting low pH led to the redissolving of ortho-P previously precipitated in the higher pH-regime in phase I; this may potentially reduce the efficiency of removing orthophosphates by subsequent solid–liquid separation treatments.

1. Introduction

Today, most of the problems associated with nutrients from agricultural practices with regard to pollution of surface and ground waters are from non-point sources as opposed to point sources. This is simply because it is easier to treat discharges at point sources than to try to arrest the situation once the pollutants have dispersed (Vinconneau *et al*., 1985). Effective and best management practices dictate that these nutrients are better dealt with at their point sources whenever possible. In this regard, biological processes are gaining interest as effective alternatives to chemical precipitation processes of removing soluble ortho-P from pig manure (Jones *et al*., 1987; Converti *et al*., 1995). The soluble phosphorus species are not only more mobile but are also directly available to algae and, therefore, their quantity in solution is the best measure of phosphorus potential to pollute the environment through a process called eutrophication.

Improved removal of phosphates in wastewater treatments has been accomplished by addition of salts of iron, calcium or aluminium to form sparingly soluble phosphates which are removed by settling (Fuhs & Chen, 1975; Kox, 1981; Maurer *et al*., 1999). On the other hand, substantial accumulation of phosphates in activated sludge has been observed without addition of such salts. These observations have, at times, created confusions regarding the mechanisms of such accumulation, *i.e.* between biological immobilization and chemical precipitation phenomena (Buchan, 1983). A purely physical–chemical model has been suggested by Menar and Jenkins (1970) to explain the latter, in which calcium from naturally hard water forms an insoluble phosphate on the activated sludge floc during aeration when carbon dioxide (CO_2) is expelled, thus raising the pH of the mixed liquor. By contrast, other researchers have argued that aeration promotes the uptake of excess phosphate in excess of immediate need by activated sludge bacteria,

Notation

E_h	redox potential on the standard hydrogen scale, mV
p	calculated probability level of the F-test
R^2	coefficient of determination
X	a negatively charged ion
α	probability level of significance

a phenomenon commonly referred to as 'luxury uptake' (Levin & Shapiro, 1965; Shapiro, 1967). Luxury uptake by bacteria typically occurs when growth is arrested by the lack of a nutrient other than phosphorus, but sufficient energy source is available to actively transfer phosphorus into the cell (Jones *et al.*, 1987). In the activated sludge process, with domestic wastewater as the substrate, bacterial growth is limited by the supply of carbon and energy sources, while all other nutrients are present in excess. This is the same as in the case of pig manure. Since neither phosphorus starvation nor shortage of other nutrients is likely to occur in pig manure, these uptake mechanisms may not play a big role in soluble ortho-P removal.

Other researchers have studied the effects of pH and volatile fatty acids (VFAs) on metabolic release/uptake of soluble ortho-P from microbial biomass. Bond *et al.* (1999) reported that pH affected metabolic phosphate release and also noted that during anaerobic uptake of VFAs, raising the pH of the mixed liquor resulted in increased metabolic phosphate release. Smolders *et al.* (1994) suggested that this extra metabolic phosphate release results from the bacteria requiring more energy at the higher pH for the uptake of acetate ion. In contrast to this suggestion, it has also been hypothesized that the release is a consequence of intracellular acidification of sludge caused by passive diffusion of non-dissociated VFAs across bacterial membranes (Fleit, 1995). Although the explanations as to how the pH and VFAs affect phosphate release/uptake by microorganisms are in conflict, net effects are not in dispute.

The effects of VFAs and other organic acids (produced from microbial degradation) on substrate pH and the potential biologically mediated phosphorus precipitation/dissolution caused by this pH change in pig manure have not been adequately studied. It is a fact that any treatment process that affects the contents of the VFAs and other organic acids will definitely alter the pH of the manure. Pig manure contains substantial quantities of Ca and Fe cations due to mineral supplements in the feeds (Campbell *et al.*, 1997). It therefore follows that any physical–chemical or biochemical processes that affect the pH of the manure are likely to have major effects not only on ortho-P release/uptake by microbes but also on ortho-P dissolution/precipitation.

Previous studies have shown that the solubility of complexes formed between ortho-P and the cations (*e.g.* $Ca_{10}(PO_4)_6X$, where X is an anion such as OH^- or F^-) are affected by temperature (Maurer *et al.*, 1999). Temperature also significantly influences the process rate of biochemical reactions and hence the growth of microorganisms. Jones and Stephenson (1996) reported an increased rate of phosphorus uptake with increasing temperature between 5 and 30°C for activated sludge during aerobic incubation. Understanding the role of VFAs and other associated metabolic products that affect ortho-P precipitation/dissolution is crucial to further improvements of the control and management of ortho-P removal treatment systems. The primary objective of the work presented here was to investigate the effect of temperature on the soluble ortho-P and to evaluate the changes (pH, redox potential and VFAs) in the manure that could help to explain these changes. Specific objectives to achieve were: (1) to relate soluble ortho-P to pH of the manure; and (2) to relate the pH to the VFAs levels in the manure. This study was divided into two phases: phase I was the first 1·5 days of aeration (where purging of CO_2 predominates) while the rest of the period up to 21 days was defined as phase II (where VFAs and other organic and inorganic products were likely to play a bigger role).

2. Methods and materials

2.1. *Equipment and instrumentation*

Figure 1 is a schematic of one unit of the equipment and instrumentation used in this experiment. The reactors were made of clear Plexiglas columns (91·44 cm in height and 15·3 cm in diameter) and were filled with test

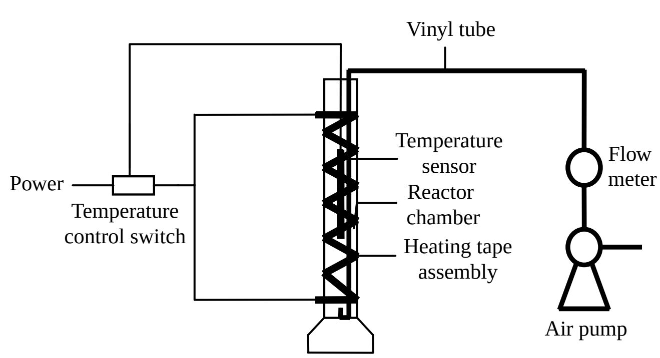

Fig. 1. A unit of the equipment and instrumentation used in the experiment

manure, leaving approximately 15·0 cm headspace to facilitate stirring and to provide room for any frothing created by aeration. To maintain the desired temperature in the manure, a heating tape was used to wrap round the reactor chamber. A temperature control switch determined the 'on-and-off' of the heating tape by the feedback from a temperature sensor positioned at approximately half the depth of manure in the reactor. This configuration maintained the temperature within 1°C of the set temperature. To aerate the manure, a positive pressure air pump (Emerson model 0623-V4-G180DX, Gast MFG Corp.) was used to introduce air into the manure through a vinyl tube (6·35 mm internal diameter) at the bottom of each reactor. A variable area flow meter (Model P-32461-64, Cole-Parmer Instrument Company) was used to regulate the flow in each unit. Five such units were set up to provide manure temperatures of 5, 10, 15, 20 and 25°C, respectively, with a fixed airflow rate of approximately 1·8 $l\,min^{-1}$ (this is the maximum airflow rate that could safely be introduced without causing an overspill of the manure from the reactor). This study was conducted in winter to allow the establishment of the stated temperatures using the heating tapes.

2.2. *Manure collection, loading and sampling*

Pig manure from a finishing barn located at the University of Minnesota Southern Research and Outreach Centre was collected in plastic containers. Prior to filling the reactors, the manure was passed through a 2 mm sieve to screen out large particles for consistency and thoroughly stirred to a uniform mixture. The characteristics of the resulting manure are given in Table 1. In addition, before starting the system, the manure in each reactor was again thoroughly stirred using a motorized paddle-stirrer (Tline Laboratory Stirrer, Model 102, Talboys Engineering Corp.) and a sample was drawn from approximately the mid-depth of each reactor for laboratory analyses. For the first 1·5 days, samples were taken after 2, 4, 6, 10, 24 and 32 h from all the reactors. For the rest of the test, samples were taken every other day. After the determination of pH and redox potential of the manure samples was completed, all the samples were kept in a deep freezer until they were analysed. The pH of the manure was determined using a pH-meter (Orion model 720A, Orion Research Inc.) while redox potential E_h was determined using an oxidation–reduction potential (ORP) meter (DIGI-SENSE, model 5938-52, Cole-Parmer Instrument Company). Frozen samples were thawed and allowed to reach room temperature before starting the laboratory analyses of the chemical, physical and biological properties.

Table 1
Characteristics of raw pig manure used in this experiment

Total solids, $g\,l^{-1}$	33·56
Total volatile solids, $g\,l^{-1}$	23·42
Total suspended solids, $g\,l^{-1}$	21·12
Total volatile suspended solids, $g\,l^{-1}$	16·95
pH	6·72
Total phosphorus, $mg\,l^{-1}$	2291·00
Soluble orthophosphates, $mg\,l^{-1}$	261·00
Redox potential E_h, mV	−97·20
Volatile fatty acids (VFAs), $g\,l^{-1}$	10·83

2.3. *Laboratory analyses*

For soluble ortho-P and VFAs determinations, a well-mixed sample was diluted and then centrifuged at 4000 min^{-1} for 30 min. The centrifuged samples were then filtered using GF/A Whatman filter papers. The ortho-P in the filtrate was determined colorimetrically as the phosphomolybdate complex after reduction with ascorbic acid (APHA, 1998). The VFAs in the filtrates were determined using an esterification method. This method is based on esterification of the carboxylic acids present in the sample followed by colorimetric determination of the esters produced by the ferric hydroxamate reaction. All volatile acids are reported as their equivalent $mg\,l^{-1}$ acetic acid (Hach, 1993).

2.4. *Experiment design and data analyses*

Effects or responses to be investigated in the manure were pH, soluble ortho-P, redox potential and VFAs content. The major parameter of the experiment was temperature at five levels/treatments (5, 10, 15, 20 and 25°C). This design fits the classical one-way or single factor with multiple treatments. A one-way analysis of variance (ANOVA) amongst the treatments was therefore conducted for each of the responses studied. A standard statistical analysis of variance procedure from Excel® was used to analyse the data in this study. The same software was also used for conducting regression analysis between target manure parameters (soluble ortho-P against pH and pH against VFAs). When not stated, statistical significance was implied at a significance level α of 0·05.

3. Results and discussions

3.1. *Phase I aeration*

The variation of soluble ortho-P in the manure within the first 1·5 days at various temperature regimes is

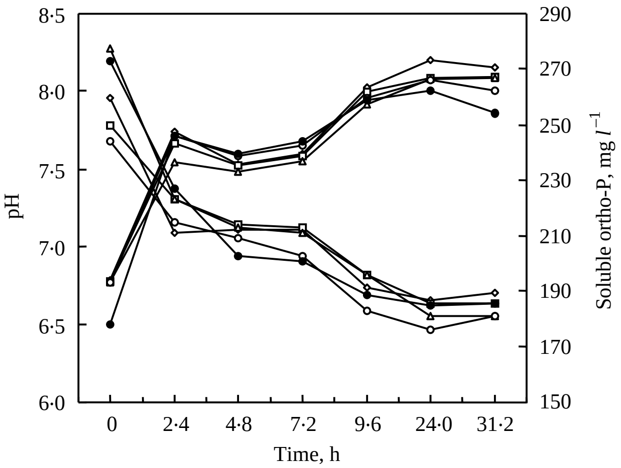

Fig. 2. Changes in soluble ortho-phosphates (ortho-P) (decreasing with time) and pH (increasing with time) in the manure during phase I of aeration at the respective temperatures: ◇, 5°C; □, 10°C; △, 15°C; ○, 20°C; ●, 25°C

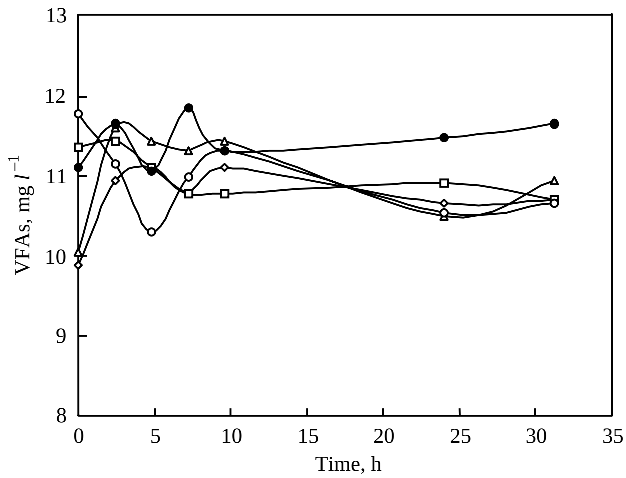

Fig. 3. Changes in volatile fatty acids (VFAs) in the manure during phase I of aeration at various temperatures: ◇, 5°C; □, 10°C; △, 15°C; ○, 20°C; ●, 25°C

presented in *Fig. 2*. The analysis of variance (ANOVA) performed on the data shows no significant differences in ortho-P amongst the five temperatures of the manure at α of 0·05. The reduction in soluble ortho-P in the manure during the first 1·5 days is therefore found to be independent of the temperature of the manure.

The changes in pH of the manure within the first 1·5 days of aeration at the respective temperature regimes are also shown in *Fig. 2*. An ANOVA on the pH within the various temperature regimes gives a value for *P* of 0·99 in the *F*-test. This indicates that at the 5% probability level, the differences in pH of manure during aeration at constant rates are not statistically significant. A rapid rise in pH is, however, observed in all temperature regimes upon aeration. This rapid increase in pH during aeration is attributed to the conversion of bicarbonates to carbonates when carbon dioxide (CO_2) is purged out of solution by the aerating air as reported by past researchers (Menar & Jenkins, 1970; Stevens & Cornforth, 1974; Luo *et al*., 2000).

The variation of pH within this time period seems to follow the inverse of the variation of soluble ortho-P. This suggests an inverse relationship between these two parameters of the manure. A linear regression relationship between pH and the soluble ortho-P for the pig manure obtained by merging and pairing together all the pH and soluble ortho-P data collected during the first 1·5 days of the experiment gives a coefficient of determination of 0·90, which suggests that the pH change in the manure explains approximately 90% of the level of soluble ortho-P in the manure. This observation concurs with that of previous workers who found that raising the pH of the manure would cause the chemical precipitation of soluble ortho-P by metal ions (Ca^{2+}, Fe^{3+}, Al^{3+}) to insoluble phosphates (Kox, 1981; Moore & Miller, 1994, Campbell *et al*., 1997; Maurer *et al*., 1999; Luo *et al*., 2000).

The variations in the VFAs during the first 1·5 days of aeration under different temperature regimes are shown in *Fig. 3*. The changes in the VFAs in this period seem negligible. The ANOVA results indicate that the differences in VFAs among the five temperature regimes investigated are not significantly different at α of 0·05 since a *P* of 0·07 is obtained in the *F*-test. Owing to large increase in pH during this period, it intuitively follows that the pH change cannot have been caused by the changes in VFAs in the manure. In other words, this change in pH cannot be explained by the changes in VFAs of the manure, which is different from the results obtained in phase II aeration (see Section 3.2).

3.2. *Phase II aeration*

The variations of soluble ortho-P in phase II aeration under the same range of temperature regimes are shown in *Fig. 4*. Using an ANOVA procedure to determine the variation of ortho-P with temperature, a value for *P* of less than 0·001 is obtained in the *F*-test. The variation of ortho-P with temperature is therefore found to be statistically significant at α of 0·05, suggesting that the temperature of the manure influences the solubility of ortho-P during aeration.

The pH of the aerated manure at different temperature regimes are presented in *Fig. 5*. An ANOVA for the variation of pH with temperature obtains a value for *P* of

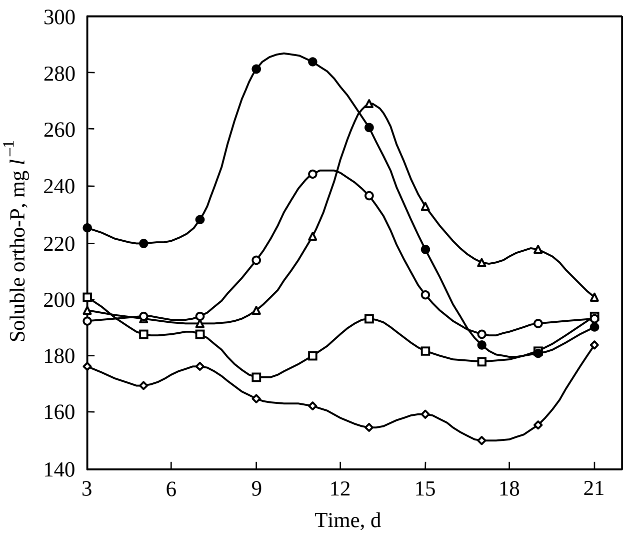

Fig. 4. Variation of soluble ortho-phosphates (ortho-P) with time of aeration under different temperature regimes in phase II of aeration: ◇, *5°C;* □, *10°C;* △, *15°C;* ○, *20°C;* ●, *25°C*

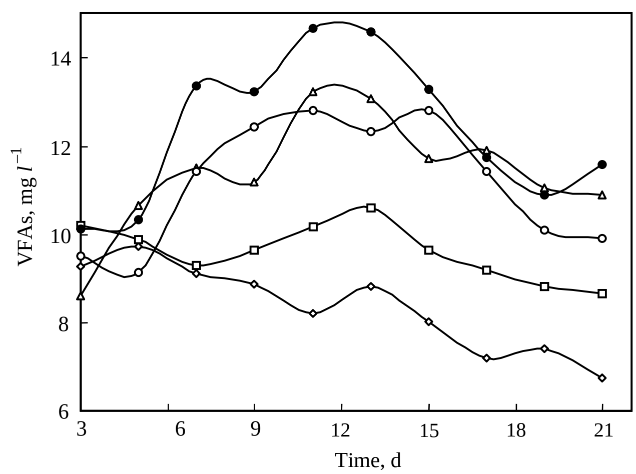

Fig. 6. Variation of volatile fatty acids (VFAs) with time of aeration under different temperature regimes in phase II of aeration: ◇, *5°C;* □, *10°C;* △, *15°C;* ○, *20°C;* ●, *25°C*

less than 0·001 for the *F*-test, suggesting that the pH of manure aerated under different temperature regimes is significantly different at α of 0·05. A comparison of *Figs 4* and *5* reveals the same pattern observed between soluble ortho-P and pH during the first 1·5 days, *i.e.* the plots of pH seem to approximately follow the inverse of the ortho-P plots. A regression analysis between pH and ortho-P yields a coefficient of determination of 0·83, *i.e.* the pH of the manure explains 83% the variation of soluble ortho-P, suggesting that the pH of the manure has a very profound influence on the level of soluble ortho-P in the manure.

Changes in VFAs contents of the aerated manure under different temperature regimes are presented in *Fig. 6*. An ANOVA on their variation with temperature resulted in a value for *P* of less than 0·001 (*F*-test), indicating that the variations of VFAs with temperature are significantly different (α of 0·05). The concentration of VFAs may give an indication of the aerobic or anaerobic status of the reactions taking place in the manure. The falling trend of VFAs in the manure suggest aerobic decomposition while rising levels of VFAs indicate anaerobic degradation (*Fig. 7*). It can therefore be inferred

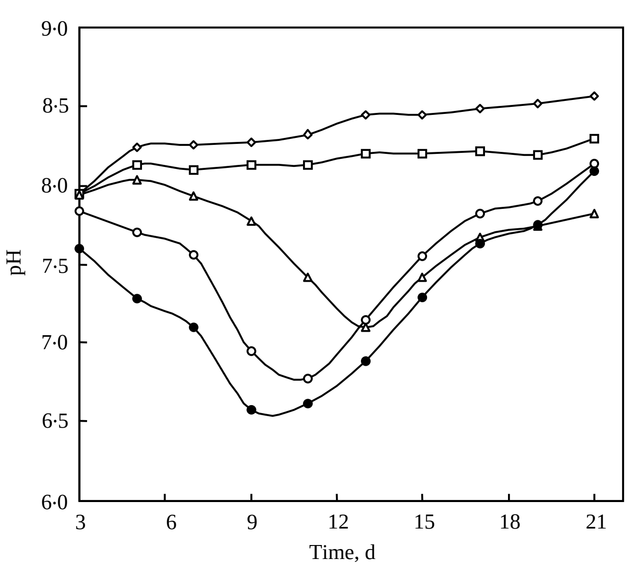

Fig. 5. Variation of pH with time of aeration under different temperature regimes in phase II of aeration: ◇, *5°C;* □, *10°C;* △, *15°C;* ○, *20°C;* ●, *25°C*

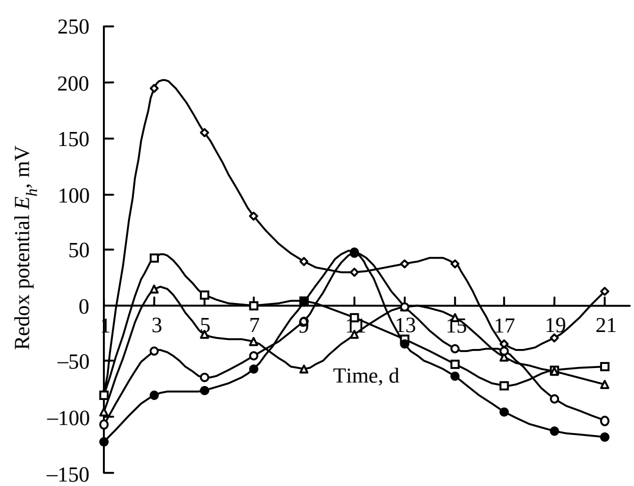

Fig. 7. Variation of manure redox potential E_h with time of aeration under different temperature regimes in phase II of aeration: ◇, *5°C;* □, *10°C;* △, *15°C;* ○, *20°C;* ●, *25°C*

that aerating manure at low temperatures is more effective than at high temperatures. This observation is in accordance with the theory of mass transfer of most gases and for oxygen in particular.

Another observation from *Figs 5* and *6* reveals that the pH of the aerated manure is inversely related to VFAs levels. This observation is confirmed by a linear regression analysis between pH and the logarithm of VFAs, which yields a coefficient of determination of 0·70, suggesting that the production of VFAs by microbial degradation explains the pH change by approximately 70%. It is therefore likely that other processes, such as nitrification, purging of carbon dioxide, and ammonia volatilization, may explain the remaining 30% of the change in pH. From the work of past researchers studying the effect of pH on P release/uptake, raising pH results in the release of P from microbes (Smolders *et al*., 1994; Fleit, 1995; Bond *et al*., 1999). In this phase of aeration, however, lowering pH results in net P-release into solution. It can therefore be inferred that reducing the pH leads to opposing effects of microbial uptake and biologically mediated chemical dissolution; however, the concentration of dissolved ortho-P rises as the microbially mediated chemical effect is greater in this phase II of aeration.

4. Conclusions

(1) There is a rapid drop in soluble ortho-phosphate (ortho-P) during the first 1·5 days of aeration that cannot be explained solely by microbial uptake. This fall is paralleled by a similar gain in the pH of the aerated manure. A good correlation coefficient of 0·95 between these two parameters confirms the dependency of ortho-P on pH that supports the theory of physical–chemical precipitation. There is no change in the volatile fatty acid (VFAs) levels in the pig manure during the phase I aeration. However, a large gain in pH is observed. These observations illustrate that the levels of VFAs cannot explain the changes in pH of the manure. The pH increase in this phase is likely to have come from the purging of carbon dioxide (CO_2) from the manure by the bubbling air as suggested in previous studies.

(2) Temperature is found to have a significant effect on ortho-P levels in the manure during phase II aeration. A strong correlation between soluble ortho-P and pH in the second phase of aeration is also observed. In addition, the pH of the manure is found to be very well correlated (correlation coefficient R of 0·84) to the VFAs levels in the manure, which means that changes in pH can be attributed to the VFAs changes in the manure. It can therefore be inferred that, together with the microbial immobilization/release of ortho-P, the VFAs levels control the amount of biologically mediated precipitation/dissolution of ortho-P in this phase of aeration.

(3) The anaerobic environment favours the production of VFAs, which results in lowered pH, and subsequent dissolution of insoluble ortho-P. Aerobic environment ensures the consumption of available VFAs and discourages further production of these fermentation products. This effectively raises the pH, thus encouraging biologically mediated chemical precipitation/removal of ortho-P.

(4) Besides the previously recognized methods of ortho-P removal by physical–chemical precipitation and microbial uptake, a third mechanism of removal of P from manure previously kept under anaerobic conditions is provided by this biologically mediated chemical precipitation of phosphates.

(5) For the effective removal of phosphorus during aeration, anaerobic conditions must be avoided by the use of a variable-controlled airflow rate to avoid unwanted redissolving of previously precipitated phosphorus.

(6) For a further understanding of all the processes involved in the changes of pH during such aeration treatments, research on the effects of nitrification, ammonia volatilization, and carbon dioxide purging is recommended in future studies.

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