The effectiveness of aerobes used as manure additives for swine manure odor control

Jun Zhu, PhD, EIT

Summary

Objective: To examine the growth of aerobic bacteria in swine manure when used as manure additives for odor control.

Methods: Hydrogen peroxide was used as an oxygen provider added to the test manure at fixed time intervals to enhance the dissolved oxygen level in manure, simulating intermittent aeration.

Results: Adding hydrogen peroxide failed to establish an aerobic environment in the top liquid. The added aerobes could not outgrow the indigenous anaerobes even when the dissolved oxygen levels in the manure were raised for short intervals on a regular basis.

Implications: The frequency of running intermittent aeration in order to maintain an active aerobic flora requires further study. Without enough aeration, the effectiveness of microbial-based manure additives for odor control purposes in actual manure storage systems is questionable.

Keywords: swine, manure, odor, microbes

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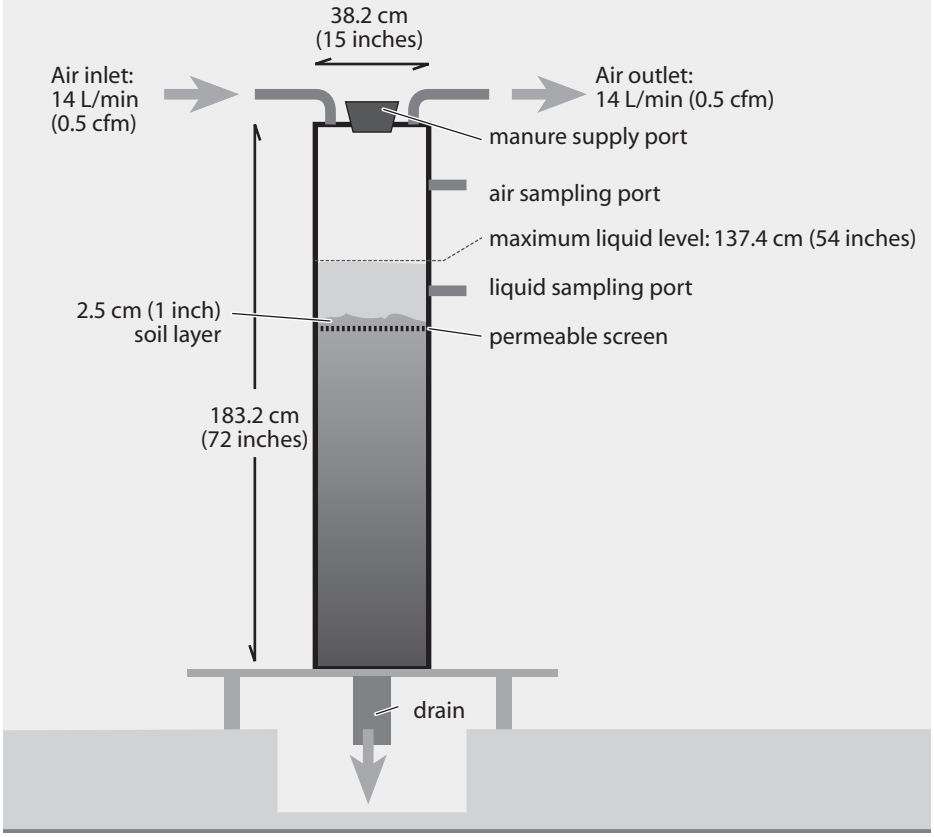
Figure 1: Test column apparatus



The idea of using biological manure additives to control odors was proposed about 20 years ago and a considerable amount of research effort has been expended in this field. Past researchers rarely found pit additive products to be effective in reducing odor levels of swine manure,[1–4] perhaps due to the complexity of odorous components in swine manure. The key hindrance to developing effective manure additive products, however, is our lack of understanding of the biological activities that occur in stored swine manure. Trial-and-error methods to evaluate manure additive products, although common, are both time consuming and fail to measure the growth of the added bacteria. To develop effective additive products, it is important to examine the growth kinetics of microbes added to manure as well as characteristics of the manure environment that may affect the chemical, physiological, and biological processes of additives. The objective of our study was to measure the growth of aerobic bacteria added to test manure under partial aeration. Factors that affect bacterial growth are also discussed.

Materials and methods

Experimental design

Our experiment was conducted over a period of 30 days in an environmentally controlled room with the temperature maintained between 18.3–23.9°C (65–75°F).

The experimental design was created to simulate a surface aerated layer on animal waste storage facilities (Figure 1). The system was composed of a permeable membrane of woven, nondegradable material, which was held at a depth of approximately 30 cm (12 inches) below the liquid surface. The liquid portion above the membrane served both as a substrate for growth of aerobic microorganisms and a semi-perme-

University of Minnesota, Southern Research and Outreach Center, 35838 120th Street, Waseca, Minnesota 56093; zhuxx034@tc.umn.edu

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able barrier for migration of volatile materials from the subsurface layer.

The experimental apparatus consisted of PVC columns 38.2 cm (15 inches) in diameter and 183.2 cm (72 inches) high. Three test columns were assigned to each treatment to obtain three replications. The columns were filled with manure to a depth of 137.4 cm (54 inches) at the beginning of the test and were maintained at that level throughout the entire testing period. Air circulated continuously through the headspace of the column (top 45.8 cm [18 inches]) at a rate of 14 L per minute (0.5 cfm) to simulate a typical ventilation situation in either a deep pit or an outside storage basin.[4] The manure was a mixture of 25% fresh manure with 75% lagoon water to make up to 1% total solids content to mimic the top portion of liquid manure in the storage facilities. Hydrogen peroxide (H_2O_2) was used as oxidizing agent for enhancing the level of dissolved oxygen in the top liquid to simulate aeration.

There were a total of five treatments (four treatments and one control) in this study:

- 50 ppm of H_2O_2 (twice a day: 8:30 AM and 4:30 PM) + microbial additive (ADD),
- 50 ppm of H_2O_2 (twice a day: 8:30 AM and 4:30 PM) + daily catalyst (1:450 dilution) + microbial additive (CATADD),
- 50 ppm of H_2O_2 (twice a day: 8:30 AM and 4:30 PM) ($50H_2O_2$),
- 10 ppm of H_2O_2 (twice a day: 8:30 AM and 4:30 PM) ($10H_2O_2$), and
- control manure.

The biocatalyst and the mixed aerobic microbial strains used in this study were provided by Olympic Environmental Company (Denver, Colorado). The biocatalyst is a water-like substance containing grass extracts with neutral pH and salts of natural groundwater. According to the results of experiments conducted by the manufacturer (personal communication) the biocatalyst is capable of enhancing the availability of oxygen to both aerobic and facultative microbes in a minimal oxygen environment, thereby increasing the growth of these microbes.

Liquid sample collection and analysis

Liquid samples were collected only from the top portion of manure in the columns (above the permeable membrane) and analyzed, at each sampling time, for total anaerobic and aerobic bacterial counts. Methods presented by Zhu, et al.,[5] were used to incubate and calculate bacterial counts.

Concentrations of dissolved oxygen in the top liquid layer were monitored using oxygen probes (OxyGuard Mk I®, Point Four Systems Inc.; British Columbia, Canada). The oxygen probes were calibrated using Zero Oxygen Solution (Cole Parmer Company; Chicago, Illinois). Each column was assigned one probe and the probes were connected with an onsite computer so the concentrations of dissolved oxygen in the liquid were recorded automatically at 1-minute intervals. Recording started immediately after hydrogen peroxide was added and lasted approximately 2.5 hours. This time frame was determined by several trials prior to the test in which a typical descending feature of the concentration of dissolved oxygen was observed; i.e., the H_2O_2-induced increase in dissolved oxygen concentration in the test liquid phased out after approximately 2–3 hours.

Statistical analysis

A complete randomized design with three replications per trial was employed. Student's *t* test at significance level of .05 was used to compare the 10-, 20-, and 30-day samples.

Results

Dissolved oxygen concentrations

Dissolved oxygen (DO) concentrations for all the treatments (except control) increased rapidly after addition of H_2O_2 (Figure 2). The CATADD, ADD, and $50H_2O_2$ treatments reached a maximum mean concentration of approximately 20 mg per L in about 12 minutes and then decreased gradually. As compared to these, the DO increase for the $10H_2O_2$ treatment was much lower. At the end of the recording time (2.5 hours), the $50H_2O_2$ treatment had DO concentrations of approximately 5 mg per L, while the other four treatments each had approximately 2 mg per L. The DO levels for all treatments with H_2O_2 dropped to a range from 0.1–0.3 mg per L before the next addition of H_2O_2.

Bacterial counts

After 10 days, both ADD and $50H_2O_2$ had significantly higher aerobic bacterial counts than the control (Figure 3). Aerobes did not grow better in the CATADD than in the control. Adding aerobic microorganisms to the liquid manure did not necessarily result in an increase in the populations of aerobes (e.g., in the CATADD treatment). In no test columns did aerobic bacteria outgrow anaerobic bacteria after 10 days.

After day 20, the number of anaerobes in all H_2O_2-treated columns was significantly decreased and the aerobic bacterial counts were not significantly different among the treatments containing H_2O_2 (the control had concentrations of 330 for aerobes and 424 for anaerobes) (Figure 3). At the end of the test, the aerobic bacterial counts for the $50H_2O_2$ treatment and the $10H_2O_2$ treatment rebounded significantly as compared to those in the CATADD and ADD treatments (Figure 3).

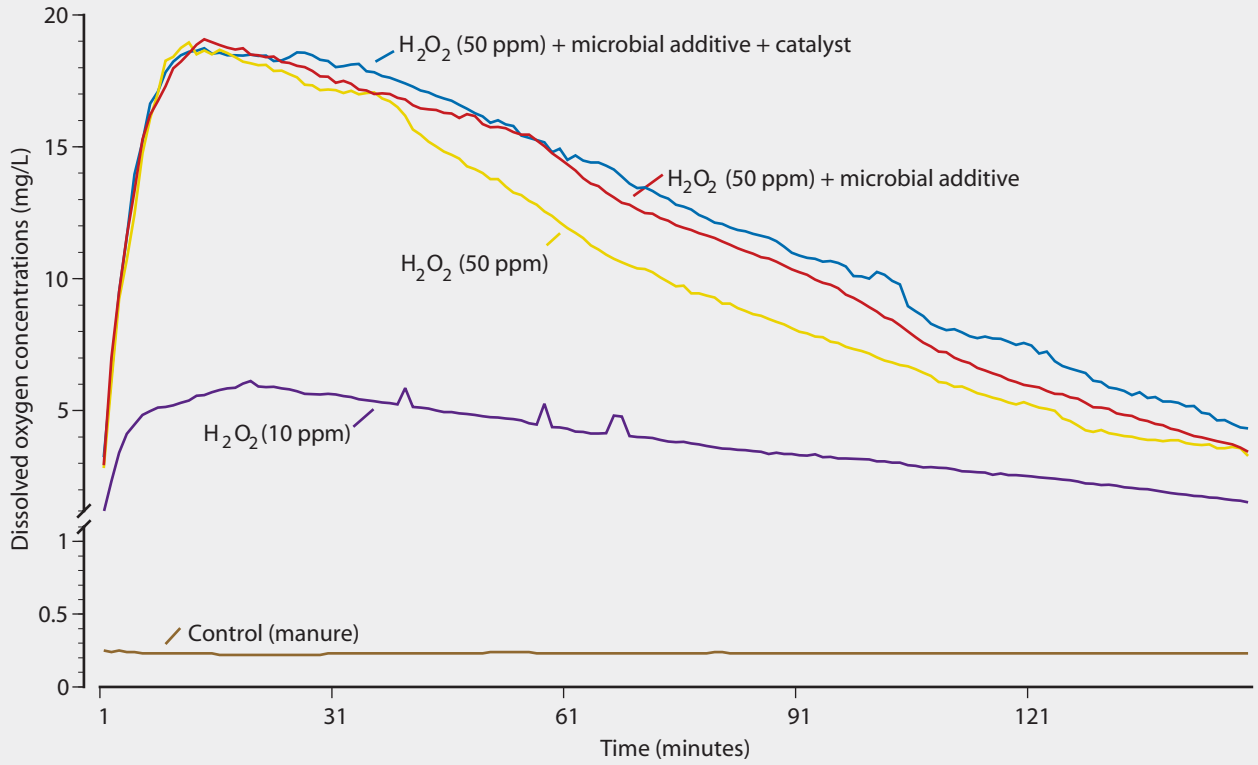
Discussion

Adding H_2O_2 to liquid manure can dramatically raise DO levels within a short time but can not maintain the raised concentration. The ability of liquid to hold oxygen is temperature dependent;[6] under the test environment (approximately 20°C), the saturation concentration of oxygen in manure liquid is around 8–9 mg per L. Thus, it can be assumed that excess oxygen in the manure liquid escaped into the air shortly after being added.

Our observation that aerobic bacteria failed to outgrow anaerobic bacteria after 10 days is consistent with the results obtained by Bourque, et al.,[7] who concluded that inoculated microbes could not become dominant in nonsterilized swine manure samples and that indigenous flora of the manure always grew better than inoculated microorganisms. Goldstein, et al.,[8] have offered this as an explanation for the possible failure of inoculation to enhance biodegradation. There have been numerous studies of the use of microbial additives for swine manure odor control,[1–4] but the success has been relatively limited, as indicated by Ritter.[9] Results from our test showed that externally adding microbes failed to establish an active aerobic flora in liquid manure.

The significantly higher anaerobic bacterial

Figure 2: Averaged dissolved oxygen (DO) concentrations in the aerated layer



counts we observed for most of the treatments after the first 10 days suggests that a fully aerobic environment may not be established by adding H_2O_2 into the liquid. Since most indigenous bacterial genera in swine manure are strict anaerobes, they can always outcompete the added aerobes for nutrients and maintain their growth under anaerobic conditions. It appeared that H_2O_2 had some inhibitory effect on the growth of anaerobes, but this effect was not apparent until after the first 10 days.

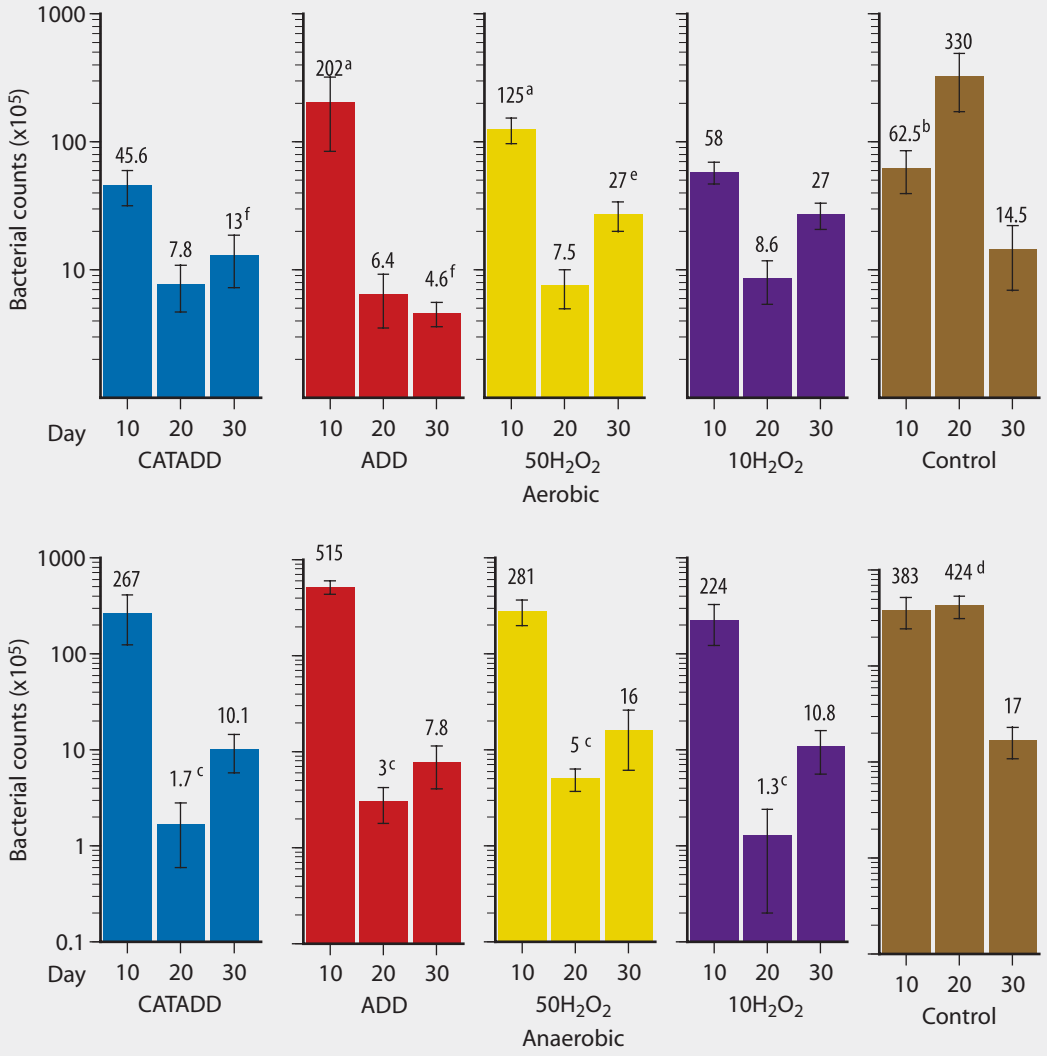
The higher aerobic counts we observed in the control compared to the other treatments could be due to a treatment effect on the growth rates of aerobes. For example, it is possible that the growth of aerobes in the treatments with H_2O_2 was accelerated by the H_2O_2 . The nutrients (such as N, P, K, S, and C) in the top layer of liquid are depleted more rapidly than in the control.[10] In the control, the aerobes grew slowly but steadily, reaching their maximum around day 20. The decrease in aerobe counts in the control after day 20 could also be due to the depletion of nutrients. Perhaps only a small quantity of nutrients could diffuse up from underneath the permeable membrane. However, the overall concentrations of aerobes were low for all the treatments, which implied that the available nutrients were exhausted in the top layer of liquid. This nutrient deficiency probably constricted the growth of both aerobic and anaerobic bacteria, resulting in a reduction in bacterial population. Because nutrients were not analyzed in the manure liquid, we were not able to verify this hypothesis in our study.

According to Grubbs,[11] the key to using bacterial cultures for deodorizing manure is to enable the added bacteria to become the predominant strains. Results from our study showed that merely adding aerobes to manure did not result in a flourishing population of the added bacteria. There are many factors that may affect bacterial growth,[12] few of which are controllable in a real environment. Although attempts have been made to study mechanisms of odorous compound decomposition by aerobic bacteria, past work was mainly focused on determining bacterial functions in digesting odorous compounds under optimum conditions.[13] This usually does not guarantee that bacteria growing well under optimum conditions will also grow well in the field, suggesting that the types of aerobes may also be important in terms of growth and function. In general, microbial species with the fastest growth rate and the ability to utilize most of the available organic matter will be the predominant species.[12] Because the ability of different bacterial genera or species to tolerate the living environment varies, as does their ability to effectively digest odorous organic compounds, it will be critical to identify and select aerobic bacteria specifically for treating swine manure stored in typical storage facilities to develop effective odor controlling additive products. Unfortunately, there is little information to inform these decisions.

The performance of aerobic bacteria in this study raises the question of the feasibility of using microbial additives for odor control. The majority of the bacterial genera used in commercial additives are obligate aerobes, while most storage lagoons, earthen basins, and deep pits are anaerobic (despite claims that some of the outside ponds are aerobic). Deficiency of oxygen in such manure storage systems will kill bacteria in the supplement culture shortly after inoculation, and dominant concentrations of aerobic bacteria will never be achieved. This may explain the limited success of using microbial additives to control odor.

In a number of studies, researchers have tried to forcibly increase the concentrations of aerobic bacteria in the manure by massive inoculation. Ohta and Ikeda[13] reported

Figure 3: Bacterial counts (±SD) at days 10, 20, and 30



By study day, values with different superscripts are significantly different ($P < .05$).

that odorous compounds in swine manure could be greatly decomposed by added bacteria (2 g of bacterial culture per 10 g of swine feces). Another study showed that a dose of about 4.5 kg of bacterial material was needed for each pig marketed to control odor.[14] Obviously, such massive inoculation can be achieved only in the laboratory, not on farms where the volumes of manure to be treated are considerable.

Under the experimental design used in this study, the effect of temporarily raising dissolved oxygen levels in liquid manure on the growth of aerobic bacteria is negligible. To date, no data have been reported regarding the minimal concentration of oxygen that should be maintained in liquid manure to assist aerobic bacterial growth. It appears that, without aeration, the possibility of controlling odor by any of the microbial-based manure additives that have been developed so far is questionable.

Implications

- Adding H_2O_2 at the concentrations used in this study can temporarily raise dissolved oxygen concentration in the manure liquid; however, it contributes little to establishing an aerated layer that can function as a suitable environment for growth of aerobic bacteria. This implies that intermittent aeration, widely recommended to save energy for odor control, may not be able to maintain aerobic activities for a prolonged period.
- Data from this study show that adding aerobic microorganisms alone into swine manure may not necessarily increase the population of aerobic microbial flora, possibly because they were unable to compete with indigenous anaerobes for nutrients. Therefore, it could be inferred that supplemental bacterial culture in an actual manure handling system might not be able to achieve a dominant population. The effectiveness of microbial-based manure additives currently in the market for odor control purpose may be questionable.
- Due to the complex nature of bacterial involvement in swine manure odor reduction and production, research regarding how to control odors using microbial manure additives is still in its infancy. Appropriate aerobic bacterial genera or species that can survive the swine manure storage environment and establish their dominance without aeration to decompose odorous compounds have yet to be found. More research is needed to study the biology of

aerobes, to search for suitable aerobic bacterial species, and, if possible, to develop new species using contemporary recombinant DNA techniques.

- Further research is needed to discover how aeration influences nutrient consumption by aerobes as well as the growth kinetics associated with the depletion of nutrients in liquid manure.

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