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Activity of soil enzymes in constructed wetlands treated with swine wastewater

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ABSTRACT

Continuous application of swine wastewater from lagoons to agricultural land can pose surface and groundwater pollution. Constructed wetland (CW) is an alternate to the lagoon spray field system that reduces the nutrients concentration. One of the biological processes in CW is enzymatic activity which plays a major role in releasing nutrients from organic substances. The objectives of this research were to investigate the activity of soil enzymes in CW treated with swine wastewater and to assess the relationship between the enzyme activity and nutrients concentration. One continuous marsh (CM) and one marsh-pond-marsh (MPM) wetland cells were investigated for enzymatic activity. The activities of dehydrogenase, urease, phosphatase, and β-glucosidase were significantly higher at 0–3 cm than 6–12 cm depth. Enzyme activities were higher in marsh soils of CM than pond soils of MPM. There was no significant difference in enzyme activity between inlet and outlet of CM and pond area of MPM. No significant relationship was found between the enzyme activity and nutrient concentration. Urease, phosphatase, and arylsulfatase activity were correlated with soil C and N, whereas, β-glucosidase activity was correlated with soil C. The results suggest that enzyme activity has aided in detritus decomposition and thus, decreased enzymatic activities may decrease nutrients availability.

1. Introduction

North Carolina ranks second in the nation in swine production after Iowa. These swine operations produce large amounts of waste which needs to be disposed properly without creating environmental hazards. Traditionally, the swine operations flush the waste with water into an aerobic or anaerobic lagoon and spread the lagoon wastewater on agricultural fields. However, such practices may impact surface and ground water quality (Mallin, 2000). Also, continuous application of swine wastewater on land can lead to the accumulation of nitrogen (N) and phosphorus (P) in spray fields. To reduce the contamination of surface and ground water, alternative methods of treating wastewater should be implemented and one of the methods to treat swine waste is the use of constructed wetlands (CWs).

Constructed wetlands provide an efficient ecological system with low maintenance requirements and construction costs to remove nutrients from animal wastewater (Kadlec and Knight, 1996; Hill et al., 1999). Wetlands support large diversity of microbial communities (Dong and Reddy, 2010) which play an important role in nutrients cycling (Wright and Reddy, 2001). Constructed wetlands successfully treat animal wastewater prior to land application and reduce nutrient concentrations applied to crops and pastures (Knight et al., 2000; Reddy et al., 2001). Earlier studies conducted on marsh-pond-marsh (MPM) CWs showed promising results in removing nutrients (Reddy et al., 2001; Hunt et al., 2002; Poach et al., 2004).

Wetland technology removes excess nutrients from wastewater by the process of sedimentation, adsorption, organic matter accumulation, microbial assimilation, nitrification-denitrification, and ammonia volatilization (Johnstone, 1991; Brix, 1993). One of the biological activities is enzymatic approach related to decomposition processes in wetland sediments (Tabatabai, 1982). Enzyme activity depends on both total microbial biomass and enzyme efficiency. Majority of the soluble soil enzymes originate from soil microorganisms where these enzymes are synthesized, secreted, and, in turn they act as generators of signals to induce further enzymes production by other microbes and plants. Enzyme activity in constructed wetlands is affected by many factors, including biological factors (microbial populations, higher taxa, and fauna), soil factors (pH, texture, nutrient composition, depth profiles, organic matter content, etc.), and climatic factors (Kang et al., 1998; Zaman

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et al., 1999; Duarte et al., 2008; Reboreda and Cacador, 2008). Therefore, it is important to understand extracellular enzymes that relate to decomposition process in CWs which will play a vital role as initiators of organic pollutant removal in treatment wetlands (Allison and Vitousen, 2005; Reddy et al., 2010) and many researchers (Krasnits et al., 2009; Aguilar et al., 2008) have recognized their role in interaction between substrate, wetland plants, and microorganisms for wastewater purification.

In this research, we studied the activity of dehydrogenase, urease, phosphatase, and β-glucosidase activity in constructed wetlands receiving swine wastewater. Urease catalyzes the hydrolysis of urea into carbon dioxide and ammonia. The urea in the wastewater originates from swine urine. Phosphatase enzyme catalyzes the organic-P that originates from the indigestible P excretion by swine into inorganic – P. β-glucosidase catalyzes the hydrolysis of sugars resulting in the formation of β-linked monosaccharides and the final product of enzymatic reaction is glucose which is a carbon source for soil microorganisms. Dehydrogenase enzyme oxidizes soil organic matter by transferring electrons and protons from substrates to acceptors and considered as an indicator of microbial activity in soil.

In this study, CWs received nutrients and organic matter from swine wastewater from spring until part of fall and plant residue in winter. Soil enzymes are affected by nutrient loading and available nutrients can potentially decrease their activity (Chrost, 1991; Wetzel, 1991). However, information on enzymatic activity and their relationship to the nutrients concentration in highly nutrient-loaded CWs is sparsely available in the literature. Therefore, in order to predict the nutrient loading impact on nutrient removal efficiency, it is important to better understand the enzymes activity in CWs receiving swine wastewater. The objectives of the study were to (i) determine enzymes (B-glucosidase, dehydrogenase, phosphatase, and urease) activity at different soil depths of constructed wetlands treated with swine wastewater, and (ii) relate their nutrients concentration to the enzymes activity.

2. Materials and methods

2.1. Site and wetland cells design

The experiment was conducted in one continuous marsh (CM) and one marsh-pond-marsh (MPM) cell at the swine research facility of the North Carolina A&T State University farm in Greensboro, NC, USA using surface flow wetlands in a continuous marsh (CM) and marsh-pond-marsh (MPM) design (Fig. 1a). The 11 m wide (W) × 40 m long (L) wetlands were constructed in 1995 (Reddy et al., 2001) and operated since 1998 to treat swine wastewater generated from the university swine farm unit. The CM consisted of 11 m W × 40m L, whereas MPM had 11 m W × 10 m L as marsh sections at both the influent and effluent ends. The water depth of 0.15 m was maintained in marshes of CM and MPM. The pond area of MPM had 11 m W × 20 × m L × 0.75m water depth. The marsh sections of both wetland cell types were planted with *Typha latifolia* L. (broadleaf cattail) and *Schoenoplectus americanus* (American bulrush) (Fig. 1b).

The removal rates of total solids, suspended solids, chemical oxygen demand, and nutrients (N and P) at different hydraulic loads and retention times of these CWs were published by Reddy et al., 2001 and Poach et al., 2004 and microbial diversities were elucidated by Dong and Reddy (2010).

2.2. Soil sampling

An auger having a diameter of 4.4 cm was used to collect soil samples from within each of the two marsh areas and from the pond area of the Marsh-Pond-Marsh system. Four samples were collected at random locations within each of the three sampling areas (marsh inlet, middle, and marsh outlet) in the continuous marsh system and pond area of MPM system. The 0–12 cm samples were sectioned to 0–3, 3–6, and 6–12 cm soil depths. The sampling procedure resulted in a total of 36 samples per cell (four samples × three locations × three depths). We feel that this sampling procedure was sound and the data analysis including measurements of variability demonstrates that the sample numbers were adequate. The wet soil samples were placed in polythene bags, transported in an ice chest to the laboratory, and stored in the refrigerator at 4 °C for further analysis.

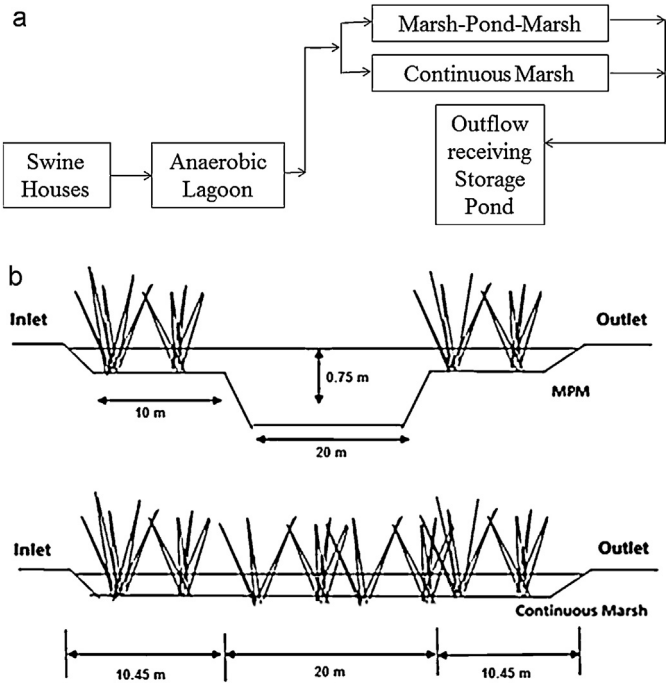

Fig. 1. (a). Flow path of wastewater from the swine houses to the constructed wetlands.
(b). Schematic diagram showing continuous marsh (CM), and pond section separates marsh areas in Marsh-Pond-Marsh (MPM) constructed wetlands.

2.3. Soil analysis procedure

A soil sub-sample was air dried in the laboratory for 2 days. The percent moisture of the original wet sample was determined. The air dried soil samples were crushed and sieved through 2.0 mm mesh to remove any plant or other material, and mixed thoroughly and stored in the refrigerator in plastic bags and used for chemical analysis.

2.4. Soil analysis

The pH of each soil sample was determined using pH meter (Orion 3 star pH bench top), Total carbon and nitrogen were determined using a CHN analyzer (Perkin Elmer series 2 model: 2400), Microbial biomass carbon (MBC) was determined using a fumigation extraction procedure (Vance et al., 1987) with 0.5 M K_2SO_4 extracts being analyzed using a CHN analyzer (Perkin Elmer model 2400). Ammonium (NH_4^+) and nitrate (NO_3^-) extracted with 1.0 M KCL and orthophosphate (PO_4^-) were analyzed using flow injection analyzer (FIA) (Lachat instruments Quick chem. 8500 series 2).