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Wind-Driven Surficial Oxygen Transfer and Dinitrogen Gas Emission from Treatment Lagoons

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Surficial oxygen transfer plays an important role, when analyzing the complex biochemical and physical processes responsible for ammonia and dinitrogen gas emission in animal waste treatment lagoons. This paper analyzes if currently known nitrogen biochemical pathways can explain the enigmatic dinitrogen gas emissions recently observed from the treatment lagoons, based on the amount of wind-driven oxygen that can be transferred through the air-water interface. The stoichiometric amounts of the maximum dinitrogen gas production potential per unit mass of O_2 transferred were calculated according to three most likely biochemical pathways for ammonia removal in the treatment lagoons—classical nitrification-denitrification, partial nitrification-denitrification, and partial nitrification-Anammox. Partial nitrification-Anammox pathway would produce the largest N_2 emission, followed by partial nitrification-denitrification pathway, then by classical nitrification-denitrification pathway. In order to estimate stoichiometric amount (i.e., maximum) of N_2 emission from these pathways, we assumed that heterotrophic respiration was substantially inhibited due to high levels of free ammonia prevalent in treatment lagoons. Most observed N_2 emission data were below the maximum N_2 emission potentials by the classical nitrification-denitrification pathway. However, one value of observed N_2 emission was much higher than that could be produced by even the partial nitrification-Anammox pathway. This finding suggests yet unknown biological processes and/or non-biological nitrogen processes such as chemodenitrification may also be important in these treatment lagoons.

Key Words: Oxygen transfer; Denitrification pathways; Dinitrogen; Treatment lagoons.

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INTRODUCTION

Anaerobic lagoons are commonly used to treat wastewater, which is flushed from livestock operations. This treatment produces a substantial reduction in the nitrogen, which is added from animal excrements. The lagoons are generally high in biochemical oxygen demand, ammonia, and bicarbonates. They tend to be slightly alkaline and highly buffered.[1] While specific lagoon characteristics vary with design, geographic location, time of year, and loading rates; conditions are generally favorable for some level of ammonia volatilization and denitrification. Ammonia volatilization has been thought to be substantially related the reduction of nitrogen, because the lagoons were perceived to be 1) highly suitable for ammonia volatilization and 2) limited in oxygen necessary for nitrification/denitrification.[2]

However, recent results have presented somewhat of an enigma for emissions from anaerobic lagoons—lower than expected ammonia and higher than expected dinitrogen gas emission rates from the treatment lagoons.[3,4] A similar departure of actual results from the expected ammonia volatilization was observed for constructed wetlands treating swine wastewater.[5 6] Harper et al.[3] postulated that the unexpected high emissions of dinitrogen gas were due to chemical denitrification of ammonia with iron in the lagoons. In supporting their hypothesis, Harper et al.[4] reported a negative value of the Gibbs free energy for the chemical conversion of ammonia to dinitrogen gas (chemodenitrification). Although the negative value of the Gibbs free reaction energy implies that the chemodenitrification is thermodynamically feasible, it does not conclusively prove their postulate.

Other researchers suggest that new microbial pathways, such as anaerobic ammonia oxidation (Anammox), heterotrophic and autotrophic denitrification may be wholly or partly responsible for the unusual emissions of ammonia and dinitrogen gas.[7–10] After reviewing these new microbial pathways, Jones et al.[2] claimed that the amount of oxygen absorbed into the top surface layer of the lagoons should be adequate to convert the ammonia to dinitrogen gas. They used the oxygen mass transfer coefficient values ranging from 0.05 to 5.0 m/d[11,12] in estimating the amount of oxygen that could be absorbed into the lagoons. The oxygen absorption rate into the treatment lagoons associated with this range of the oxygen transfer coefficient was from 4.4 to 440 kg O_2 $ha^{-1}d^{-1}$, a 100-fold range. Jones et al.[2] were unable to either narrow the oxygen absorption rate or more quantitatively evaluate the biological dinitrogen gas emission rate.

The objectives of this paper are to estimate wind-driven surficial oxygen absorption rate into treatment lagoons based on the existing knowledge in the literature, and to analyze if currently known N transformation pathways can explain the enigmatic N_2 gas emission data with the new insight on oxygen transfer into treatment lagoons.

WIND-DRIVEN SURFICIAL OXYGEN TRANSFER

Although earlier research endeavors had attempted to quantify the oxygen absorption rate through the oxygen mass-transfer coefficient (K) correlations, the lack of agreements among these correlations makes it very difficult to use the information.[13] Fortunately, substantive gas transfer data for stationery water body systems, in which wind is the single most important turbulence agent, have been published during the last 5 decades. Ro et al.[13] synthesized a new unified oxygen transfer coefficient correlation based on the compiled data published for the last 5 decades as:

$$K_L(\text{cm/h}) = 170.6 \cdot Sc^{-1/2} U_{10}^{1.81} \left(\frac{\rho_a}{\rho_w}\right)^{1/2} \quad \text{for} \quad \text{U}_{10} > 0 \tag{1}$$

where K_L = mass transfer coefficient (cm/h), Sc = Schmidt number (v/D), U_{10} = wind speed measured at 10 m (m/s), v = kinematic viscosity of water (m^2/s), D = molecular diffusivity of gas (m^2/s), ρ_a = air density (kg/m^3), and ρ_w = water density (kg/m^3).

The new equation predicts the transfer coefficient of oxygen as well as other sparingly soluble gases (i.e., liquid-side controlled gases) based on molecular properties of the gas and weather conditions. The mass flux rate of atmospheric oxygen absorbing into the treatment lagoon through its water surface can be estimated based on the lagoon size and the operating dissolved oxygen concentration of the lagoon.

$$+J_{O2} = K_L(C_S - C) \tag{2}$$

where J_{O2} = surficial oxygen absorption rate ($\text{kg/m}^2/\text{s}$), K_{L} = oxygen transfer coefficient (m/s), C_S = saturation dissolved oxygen concentration (kg/m^3), and C = bulk dissolved oxygen concentration (kg/m^3).

To estimate the surficial oxygen absorption rate, the oxygen mass transfer coefficient was first calculated using Equation 1 under varying wind speeds and temperatures (Fig. 1). The transfer coefficient increases with temperature. Film-based or turbulent boundary layer mass transfer theories[13] also predict the increase in transfer coefficient with temperature because molecular diffusivity also increases with temperature. At an average wind speed of 6 m/s, the oxygen transfer coefficient increases from 5.1 to 7.4 cm/hr for the increase in temperature from 10 to 25°C, respectively.

After the oxygen mass transfer coefficient was calculated, the maximum surficial oxygen flux into the lagoon surface was then estimated assuming the bulk dissolved oxygen concentration was zero in Eq. 2. The maximum surficial oxygen flux increased with wind speed: The oxygen absorption rate increased from 20 kg $\text{ha}^{-1}\text{d}^{-1}$ to 103 kg $\text{ha}^{-1}\text{d}^{-1}$ at wind speed 2 to 5 m/s, respectively. As discussed before, the oxygen transfer coefficient increased with temperature; however, the solubility of oxygen decreased with temperature. As a result, the

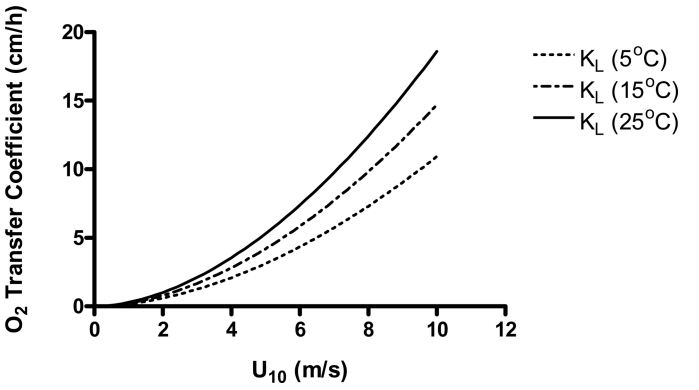

Figure 1: Oxygen mass transfer coefficient under various wind speeds and temperatures.

surficial oxygen flux into a lagoon becomes rather insensitive to temperature (Fig. 2).

N_2 EMISSION AND THE SURFICIAL O_2 TRANSFER IN THE TREATMENT LAGOONS

Valuable insights on different biological nitrogen pathways in the treatment lagoons can be obtained by analyzing the stoichiometric amounts of dinitrogen gas that can be produced per mass of atmospheric oxygen transferred. In subsequent discussion, we (1) determined the stoichiometric oxygen requirements for ammonia removal and dinitrogen gas production in the treatment lagoons based on current understandings of most likely biochemical pathways; (2) determined maximum N_2 emission potentials, i.e., the stoichiometric dinitrogen

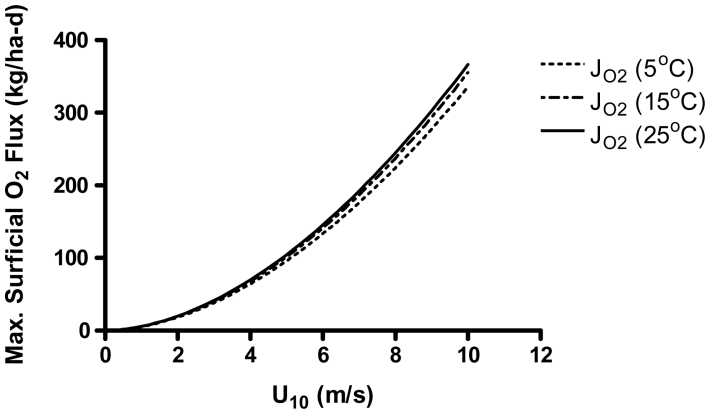

Figure 2: Maximum surficial oxygen transfer rate at different wind speeds and temperatures.

gas production rates from different pathways per maximum surficial oxygen mass transfer rates; and (3) compared the maximum N_2 emission potentials with published N_2 emission data.

The Stoichiometric O_2 Requirements for Various N Biochemical Pathways

Classical Nitrification-Denitrification Pathway. The classical biological nitrogen removal pathway involves a two-step process. In the first nitrification step, ammonia is oxidized to nitrate with intermediate formation of nitrite. Aerobic ammonia oxidizers such as the well-known genera *Nitrosomonas* and *Nitrosospira* oxidize ammonia (not ammonium) to hydroxylamine by the enzyme ammonia monooxygenase (AMO), which is further oxidized to nitrite by the enzyme hydroxylamineoxidoreductase (HAO). The classical biological nitrogen removal mechanism assumes further oxidation of nitrite to nitrate by a different group of nitrifying bacteria, *Nitrobacter* species. The stoichiometric equations for these energy-yielding ammonia oxidation steps are:

$$\begin{array}{l} NH_3 + 1.5O_2 \rightarrow NO_2^- + H^+ + H_2O \\ NO_2^- + 0.5O_2 \rightarrow NO_3^- \\ \hline NH_3 + 2O_2 \rightarrow NO_3^- + H^+ + H_2O \end{array} \qquad (3)$$

The nitrate formed in the nitrification process is then reduced to molecular nitrogen in the denitrification step. Hetrotrophic denitrifying bacteria reduce nitrate to nitrite and then eventually to dinitrogen gas under anoxic conditions. According to this classical nitrification-denitrification pathway, 4.6 kg of O_2 is required to completely oxidize 1 kg of total ammonia nitrogen (TAN) to nitrate and subsequently reduce to dinitrogen gas (based on the energy-yielding oxidation reactions, Eq. 3). If the above energy-yielding oxidation steps and the cell synthesis reactions are combined, the overall nitrification reaction can be represented as:[14]

$$NH_4^+ + 1.83O_2 + 1.98HCO_3^- \rightarrow 0.021C_5H_7O_2N + 0.98NO_3^- + 1.04H_2O + 1.88H_2CO_3 \qquad (4)$$

If the overall reaction (Eq. 4) is considered, the oxygen requirement would be 4.2 kg O_2/kg TAN, which was the value we used for the later analysis. Although other nitrogenous gases such as nitric and nitrous oxides may be released when oxygen level is so high that the denitrifying enzymes are not completely expressed,[15] the final end product of the nitrification-denitrification process is dinitrogen gas.

Partial Nitrification-Denitrification Pathway. A significant body of new knowledge about the nitrogen removal pathways has been accumulated

during the last 2 decades:[16] The above classical nitrogen removal mechanism may not be the dominant mechanism in the treatment lagoons receiving high-strength animal wastewater with high levels of ammonia and COD. Ammonia can be partially nitrified to nitrite (partial nitrification), followed by denitrification or anaerobically oxidized (Anammox) to dinitrogen gas and nitrate. These new pathways represent significant benefits to the wastewater treatment process because less oxygen is required. The treatment process is also faster, because the nitrite reduction rate is about 1.8 times faster than the rate of the nitrate reduction.[17]

The partial nitrification might result from the fact that high levels of unionized ammonia inhibit the growth of *Nitrobacter*. Unionized ammonia concentration greater than 0.1–1.0 mg/L inhibited *Nitrobacter* species in laboratory and pilot plant studies.[18] In anaerobic swine waste treatment lagoons, the typical range of total ammonia concentration (TA = NH_3 + NH_4^+) is from 280 to 650 mg TA/L:[1] At pH 7.7, the free ammonia concentration ranges from 7.6 to 17.7 mg/L. This level of unionized ammonia will likely inhibit the *Nitrobacter* species, and the nitrite will be the final product for the nitrification step. Although a small amount of the nitrite can be denitrified by some nitrifying bacteria such as *Nitrosomonas eutropha*,[19] the majority of the nitrite will be quickly reduced to dinitrogen gas by heterotrophic denitrifiers. Regardless of whether the nitrite is denitrified by heterotrophs or autotrophs, the partial nitrification-denitrification pathway represents 25% less oxygen requirements than the complete nitrate nitrification-denitrification pathway, i.e., 3.1 kg-O_2/kg TAN based on Eq. 5.

Partial Nitrification-Anammox Pathway. The anaerobic ammonia oxidizers (Anammox) reduce the nitrite with ammonia as an electron donor. Schmidt et al.[20] reported the following overall reaction:

$$NH_4^+ + 1.32NO_2^- + 0.066HCO_3^- + 0.13H^+ \rightarrow 1.02N_2 + 0.26NO_3^- + 0.066CH_2O_{0.5}N_{0.15} + 2.03H_2O \quad (5)$$

The ratio of ammonia to nitrite consumed by the Anammox bacteria is about 1:1.3. The excess nitrite is further oxidized to nitrate anaerobically; and the electrons derived from this oxidation are used for fixation of CO_2.[16,21] Although a more detailed biochemical pathway of the Anammox bacteria is not fully understood at this time, it has been postulated that ammonium ion (not un-ionized ammonia) is oxidized by hydroxylamine to produce hydrazine, which is a volatile and toxic intermediate. An enzyme similar to HAO from the Anammox bacteria further oxidizes hydrazine to dinitrogen gas.[22–24] The Anammox process requires a preceding partial nitrification step for oxidizing only about 57% (i.e., 1.32/2.32) of the total ammonia that has been removed. Therefore, the oxygen requirement removing ammonia with the

complementary partial nitrification-Anammox pathway is further reduced to about 1.8 kg O_2/kg TAN (based on Eq. 5).

Heterotrophic nitrification pathway. Heterotrophic nitrification is another pathway in which the oxidation of ammonia, hydroxyl amine or organic nitrogen compounds are carried out under low oxygen conditions by heterotrophs. Because these bacteria preferentially occur under acidic environments and the nitrification rates of the heterotrophic nitrifiers are low compared to autotrophic bacteria, this may not be a significant factor in the treatment lagoons.[16] The high level of alkalinity in the treatment lagoons preventing pH decrease further reduces the likelihood of the heterotrophic nitrification as an important N pathway.

Maximum N_2 Production Potential. Several researchers observed that heterotrophic growth was inhibited and the associated oxygen utilization rates were greatly reduced by high unionized ammonia concentration.[25,26] In the top oxic layer of treatment lagoons, the heterotrophic oxygen utilization would likely be limited due to high level of free ammonia typically found in treatment lagoons. Thus, most of the oxygen absorbed from the air would be consumed by the autotrophs for ammonia oxidation. The nitrate and/or nitrite formed from the nitrification process at the top oxic layer would then diffuse into the anoxic layer below. There nitrite/nitrate would be reduced to dinitrogen gas by denitrifiers. Whereas the denitrification reaction goes to completion, and its rate is faster than the nitrification rate; there would be no accumulation of nitrate/nitrite in the lagoons.

If we assume that the heterotrophic respiration was substantially inhibited due to high levels of free ammonia prevalent in treatment lagoons, the maximum potential of producing N_2 gas via classical nitrification-denitrification, partial nitrification, and the complementary partial nitrification-Anammox pathways would simply be reciprocals of the oxygen requirements for nitrification; 0.24, 0.32, and 0.56 kg N_2 per kg-O_2, respectively. These maximum N_2 production potentials from the three N pathways were plotted against U_{10} as shown in Figure 3. For a given mass of the atmospheric oxygen transferred to treatment lagoons at wind speed 5 m/s, the ammonia removal process undergoing the partial nitrification-Anammox pathway would produce the most N_2 gas (58 kg/ha-d), followed by the partial nitrification-denitrification pathway (33 kg/ha-d), and the least by the classical nitrification-denitrification pathway (25 kg/ha-d).

Comparison with Published N_2 Gas Emission Data

There are only a few reports of N_2 gas emission observation from swine waste treatment lagoons. Harper et al.[3] measured N_2 gas emission from a four-stage swine waste treatment lagoon system in Georgia. An average wind speed of 4.0 $\pm$ 3.4 m/s from 1994 to 1996 (measured at 1.6 m above

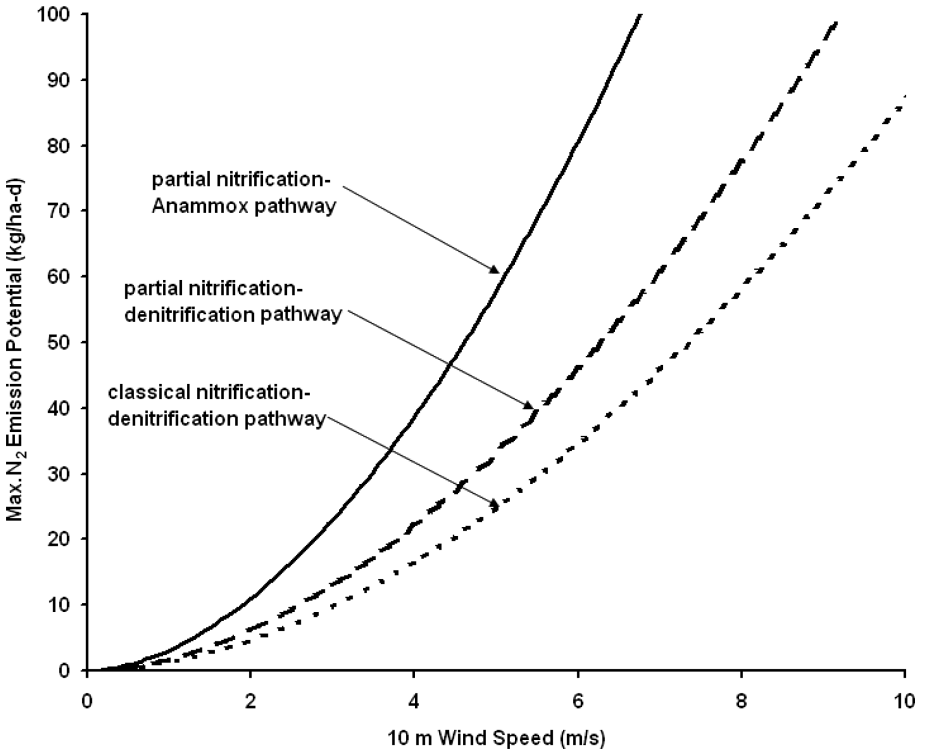

Figure 3: Maximum N_2 production potential from swine waste treatment lagoons via different N pathways.

water surface) and the average air and water temperatures of 21.4 ± 6.5°C and 23.3 ± 7.6°C, respectively, were reported. The N_2 fluxes from the four lagoons were 23.1, 11.3, 11.8, and 11.8 kg $N_2 ha^{-1} d^{-1}$. We compared these N_2 emission data with the previously determined maximum N_2 production potential curves (Fig. 4). The average wind speed was converted to U_{10} using the seventh-root law.[13] All observed N_2 flux values were below the maximum N_2 production potential predicted from the classical nitrification-denitrification pathway. Notwithstanding previous unsubstantiated assumptions, it suggests that these observed N_2 emission may be explained by the classical nitrification-denitrification pathway.

Harper et al.[4] later reported additional dinitrogen gas emission observations based on the systems nitrogen balance approaches. The average N_2 emissions from lagoons of North Carolina farrow-to-finish (FF), farrow-to-wean (FW), and Georgia FF farms were 85.6, 14.4, and 23.1 kg-N_2 $ha^{-1} d^{-1}$, respectively (Table 8 of their paper). The median wind speeds of these farms (Table 2 of Harper et al., 2004) were normalized to 10-m wind speeds. Two of the three observations again fell below the maximum N_2 production potential line of the classical nitrification-denitrification pathway. However, the large

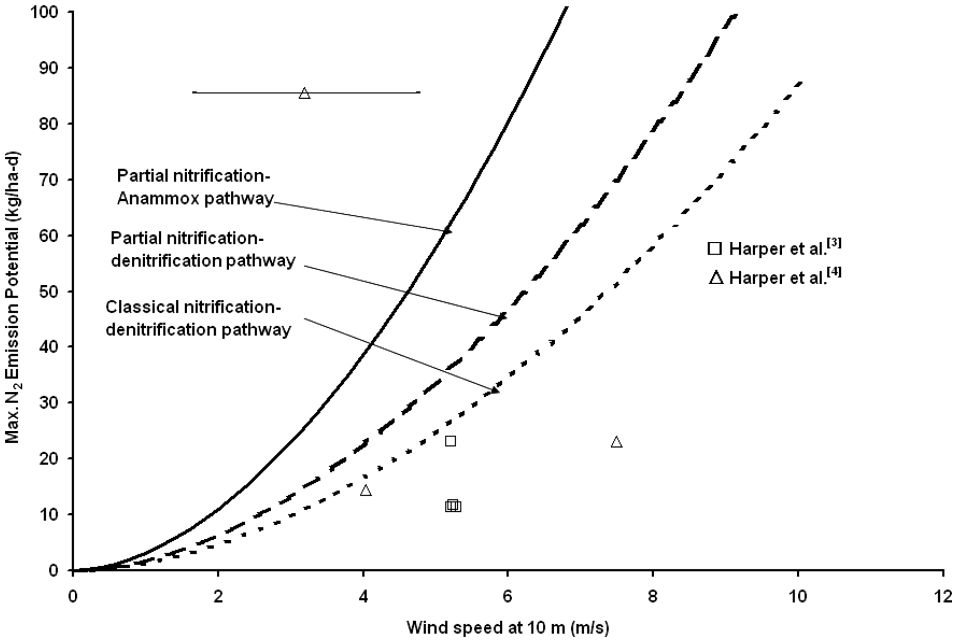

Figure 4: Comparing existing N_2 emission data with the max. N_2 potential curves.

emission value of 85.6 kg-N_2 $ha^{-1}d^{-1}$ (North Carolina FF) was far greater than the maximum N_2 production potential theoretically possible by the most N_2-yielding partial nitrification-Anammox pathway even without any competition from heterotrophs for the limited oxygen.

To produce this much N_2, an average wind speed of about 10 m/s was required to supply enough oxygen for the classical nitrification-denitrification microbial reactions. Even for the least oxygen-demanding pathway of the partial nitrification-Anammox pathway, the rate of 85.6 kg-N_2 $ha^{-1}d^{-1}$ requires more than 6 m/s of prevailing wind speeds around the lagoons. We plotted the range of wind speeds (i.e., minimum to maximum) of the North Carolina FF farm to see if this range overlapped any of the maximum N_2 production potential lines; but the maximum N_2 production potentials of this range were still much smaller than the observed value of 85.6 kg-N_2 $ha^{-1}d^{-1}$. Therefore, it appears that the alternative chemical denitrification or a yet-defined microbial consortium may indeed play an important role in these treatment lagoons as suggested by Haper et al.[4] and Megonigal et al.[27]

CONCLUSION

In our attempt to clarify the enigmatic observations of high dinitrogen gas emission rates in treatment lagoons, we used the new unified oxygen transfer

coefficient equation to estimate the maximum surficial oxygen flux into the treatment lagoons; the oxygen flux increased with wind speed, but it was rather insensitive to temperature changes (10 to 25°C). We assumed that the atmospheric oxygen transferred into the top oxic layer of the treatment lagoon supports the nitrification process producing nitrites/nitrates, which are necessary precursors for subsequent denitrification reaction. Heterotrophic respiration was assumed to be inhibited, due to high levels of free ammonia prevalent in treatment lagoons. With this assumption, the stoichiometric amounts of the maximum dinitrogen gas production were calculated based on three most-likely biological pathways for ammonia removal in the treatment lagoons—classical nitrification-denitrification, partial nitrification-denitrification, and partial nitrification-Anammox.

We then compared the maximum stoichiometric dinitrogen gas production potentials, that could be supported by the given mass of atmospheric oxygen transferred into the treatment lagoons, with the observed N_2 emission data. These N pathways supported by the surficial oxygen transfer appear to explain the most of the observed N_2 emission data. However, one N_2 emission data set with a much higher value than can be supported by the three established biochemical pathways suggests the possibility that chemodenitrification or a yet-defined microbial consortium may also be important in these treatment lagoons.

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REFERENCES

1. Bicudo, J.R.; Safley, Jr.,L.M.; Westerman, P.W. Nutrient content and sludge volumes in single-cell recycle anaerobic swine lagoons in North Carolina. Trans. ASAE **1999**, *42* (4), 1087–1093.

2. Jones, M.L.; Liehr, S.K.; Classen, J.J.; Robarge, W. Mechanisms of dinitrogen gas formation in anaerobic lagoons. Adv. Environ. Res. **2000**, *4*, 133–139.

3. Harper, L.A.; Sharpe, R.R.; Parkin, P.B. Gaseous nitrogen emissions from anaerobic swine lagoons: ammonia, nitrous oxide, and dinitrogen gas. J. Environ. Qual. **2000**, *29*, 1356–1365.

4. Harper, L.A.; Sharpe, R.R.; Parkin, T.B.; De Visscher, A.; van Cleemput, O.; Byers, F.M. Nitrogen cycling through swine production systems: ammonia, dinitrogen, and nitrous oxide emissions. J. Environ. Qual. **2004**, *33*, 1189–1201.

5. Knight, R.L.; Payne, Jr.,V.W.E.; Borer, R.E.; Clarke, Jr.R.A.; Pries, J.H. Constructed wetland for livestock wastewater management. Ecol. Engr. **2000**, *15*, 41–55.

6. Poach, M.E.; Hunt, P.G.; Sadler, E.J.; Matheny, T.A.; Johnson, M.H.; Stone, K.C.; Humenik, F.J.; Rice, J.M. Ammonia volatilization from constructed wetlands that treat swine wastewater. Trans. ASAE **2002**, *45* (3), 619–627.

7. Bodelier, P.L.; Libochant, J.; Blom, C.W.P.M.; Laanbroek, H.J. Dynamics of nitrification and denitrification in root-oxygenated sediments and adaptations for ammonia-oxidizing bacteria to low-oxygen or anoxic habitats. Appl. Environ. Microbiol. **1996**, *62* (11), 4100–4107.

8. Goreau, T.J.; Kaplan, W.; Wofsky, S.; McElory, M.; Valois, F.; Watson, S. Production of NO^{2-} and N_2O by nitrifying bacteria at reduced concentrations of oxygen. Appl. Environ. Microbiol. **1980**, *40* (3), 526–532.

9. Muller, E.B.; Stouthamer, A.H.; Verseveld, H.W. Simultaneous NH_3 oxidation and N_2 production at reduced O_2 tensions by sewage sludge subcultured with chemolithotrophic medium. Biodegradation **1995**, *6*, 339–349.

10. Van de Graaf, A.A.; Mulder, A.; Bruijin, P.D.; Jetten, M.; Robertson, L.A.; Kuenen, G. Anaerobic oxidation of ammonium is a biologically mediated process. Appl. Environ. Microbiol. **1995**, *61* (4), 1246–1251.

11. Kadlec, R.H.; Knight, R.L. *Treatment Wetlands*; Boca Raton FL, Lewis Publishers: 1996.

12. Bowie, G.L.; Mills, W.B.; Porcella, D.B.; Campbell, C.L.; Pagenkopf, J.R.; Rupp, G.L.; Johnson, K.M.; Chan, P.W.H.; Gherini, S.A. Tetra Tech. Rates, constants, and kinetic formulations in surface water quality modeling (second edition). EPA/600/3-85/040 Athens, Georgia, **1985**.

13. Ro, K. S.; Hunt, P. G. New unified equation for wind-driven surficial oxygen transfer into stationary water bodies. Submitted to Trans. ASABE, 2005.

14. Metcalf & Eddy, Inc.. *Wastewater Engineering Treatment, Disposal, and Reuse*; McGraw-Hill, Inc.: New York, 1991.

15. Korner, H.; Zumft, W.G. Expression of denitrification enzymes in response to the dissolved oxygen level and respiratory substrate in continuous culture of *Pseudomonas stuzeri*. Appl. Environ. Microbiol. **1989**, *55*, 1670–1676.

16. Schmidt, I.; Sliekers, O.; Schmid, M.; Bock, E.; Fuerst, J.; Kuenenm, J.G.; Jetten, M.S.M.; Strous, M. New concepts of microbial treatment processes for the nitrogen removal in wastewater. FEMS Microbiol. Rev. **2003**, *27*, 481–492.

17. Wett, B.; Rauch, W. The role of inorganic carbon limitation in biological nitrogen removal of extremely ammonia concentrated wastewater. Water Res. **2003**, *37*, 1100–1110.

18. Anthonisen, A.C.; Loehr, R.C.; Parkasam, T.B.S.; Srinath, E.G. Inhibition of nitrification by ammonia and nitrous acid. J. WPCF **1976**, *48* (5), 835–852.

19. Poth, M. Dinitrogen production from nitrite by a *Nitrosomonas* isolate. Appl. Environ. Microbiol. **1986**, *52*, 957–959.

20. Schmidt, I.; Sliekers, O.; Schmid, M.; Cirpus, I.; Strous, M.; Bock, E.; Kuenen, J.G.; Jetten, M.S.M. Aerobic and anaerobic ammonia oxidizing bacteria—Competitors or natural partners?. FEMS Microbiol. Rev. **2002**, *39*, 172–181.

21. Corti, B.; Fetzner, S. Bacterial metabolism of n-alkanes and ammonia under oxic, suboxic, and anoxic conditions. Acta Biotechnol. **2002**, *22*, 299–336.

22. Lindsay, M.R.; Webb, R.I.; Strous, M.; Jetten, M.S.M.; Butler, M.K.; Forde, R.J.; Fuerst, J.A. Cell compartmentalization in planctomycetes: Novel types of structural organization for the bacterial cell. Arch. Microbiol. **2001**, *175*, 413–429.

23. Schalk, J.; Vries, D.S.; Kuenen, J.G.; Jetten, M.S.M. Involvement of a novel hydroxylamine oxidoreductase in anaerobic ammonium oxidation. Biochemistry **2000**, *39*, 5405–5412.

24. Van de Graaf, A.A.; Bruijn, P.D.; Robertson, L.A.; Jetten, M.S.M.; Kuenen, J.G. Metabolic pathway of anaerobic ammonium oxidation on the basis of N-15 studies in a fluidized bed reactor. Microbiol. UK **1997**, *143*, 2415–2421.

25. Lee, S.-M.; Jung, J.-Y.; Chung, Y.-C. Measurement of ammonia inhibition of microbial activity in biological wastewater treatment process using dehydrogenase assay. Biotechnol. Lett. **2000**, *22*, 991–994.

26. Yang, S.-F.; Tay, J.-H.; Liu, Y. Inhibition of free ammonia to the formation of aerobic granules. Biochemical Eng. J. **2004**, *17*, 41–48.

27. Megonigal, J.P.; Hines, M.E.; Visscher, P.T. Anaerobic metabolism: Linkages to trace gases and aerobic processes. Pages 317–424. in Schlesinger, W. H., (Ed.). *Biogeochemistry*. Elsevier-Pergamon: Oxford, UK, 2004.