Nitrogen Removal Mechanisms and Pathways in Facultative Waste Stabilization Ponds in the United Kingdom

Eleanor Relton Claire van der Linde

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The candidate confirms that the work submitted is her own and that appropriate credit has been given where reference has been made to the work of others.

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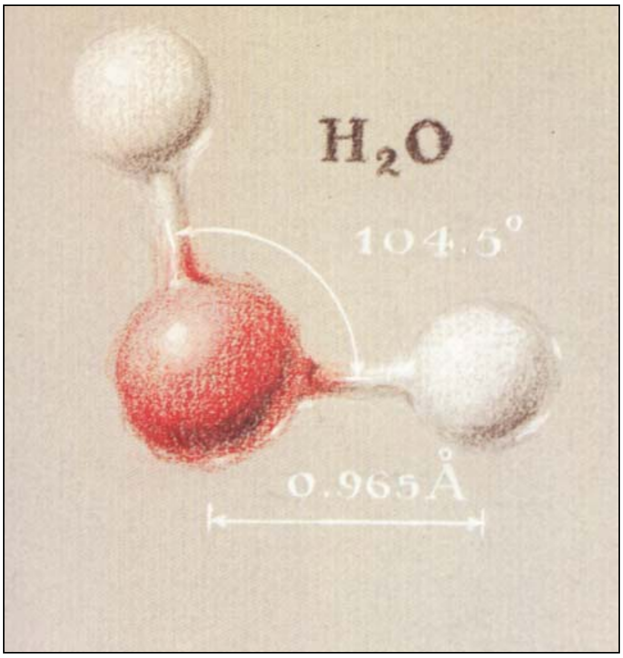
"… Ad majorem Dei gloriam …"

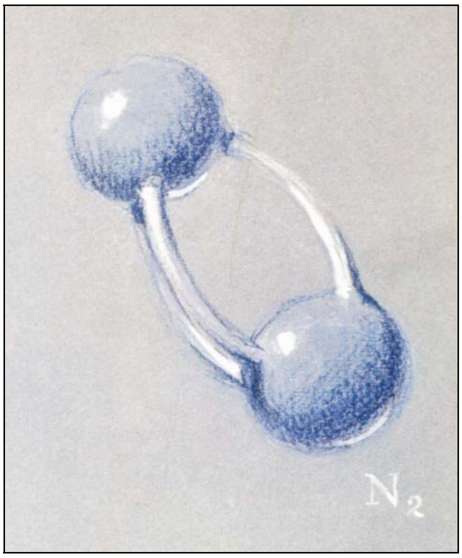


The Water Molecule – taken from Pauling and Hayward, 1964.

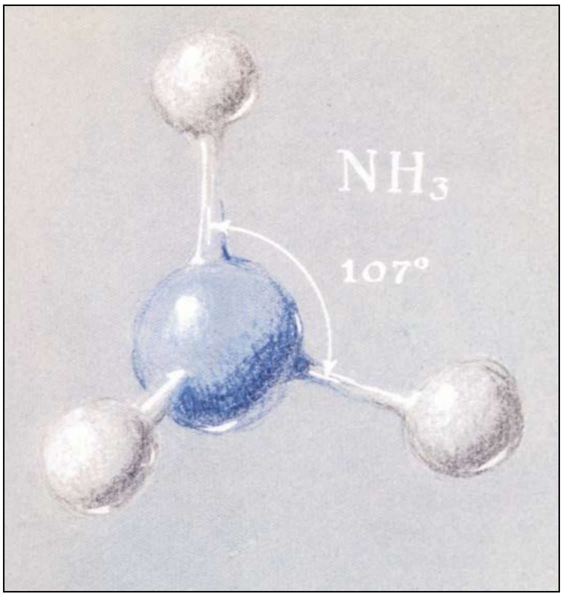
Abstract

The removal of total nitrogen was measured from two pilot-scale primary facultative wastewater stabilization ponds based at Yorkshire Water's Esholt wastewater treatment works in Bradford, UK. Three novel tracer studies, using a stable isotopically labelled spike of ammonium chloride as $^{15}NH_4Cl$, were conducted on both of the ponds, to pinpoint the exact mechanisms and pathways of nitrogen removal, or transformation, within the systems. Each $^{15}NH_4Cl$ spike also incorporated rhodamine WT as a dye tracer to determine the hydraulic characteristics and performance of the ponds. The primary facultative ponds (PFP's) were loaded at the optimum BOD loading of 80 kg/ha d for PFP's in a UK climate. A theoretical hydraulic retention time was set, and maintained by blending the influent wastewater with a freshwater flow, to produce a nominal hydraulic retention (θ_0) time of 30 d. Each experimental period was run for a duration of $3\theta_0$. The concentrations of labelled nitrogen bound as suspended and soluble organic-nitrogen, ammonium-nitrogen, and nitrate-nitrogen were obtained from the analysis of, and partitioning of, daily composite effluent samples. The volatilization of ammonia was also measured, as was sludge accumulation, throughout the duration of each experiment, and appropriate ^{15}N fractions also determined. Stable isotope analysis was conducted using a helium continuous flow isotope ratio mass spectrometer. The molecular microbiological component in this work was undertaken in conjunction with the University of Newcastle, to determine the bacterial speciation of the ponds, with particular emphasis placed on nitrifiers and denitrifiers.

The results revealed that the volatilization of ammonia contributed negligibly to overall permanent nitrogen removal from the ponds. The passage of ^{15}N-ammonium throughout the system was found to approximate the hydraulic flow characteristics of the pond. A large degree of hydraulic short-circuiting was observed in winter operating conditions. Analysis revealed that the predominant form of ^{15}N exiting the pond in summer months was bound in the suspended organic-nitrogen fraction. Both sets of winter results revealed that the largest form of ^{15}N leaving the pond was in the unchanged ammonium ^{15}N fraction. Molecular microbiological analysis, incorporating PCR amplification and DGGE tools, revealed that in many wastewater samples, gathered from all parts of the pond, there was a high presence of bacteria involved specifically with the cycling of nitrogen. Anammox bacteria were only detectable in summer samples, but a wide range of ammonia-oxidising bacteria, archea, and denitrifiers were detected in both winter and summer. Methanotrophs were also found throughout the pond in both periods. Although *in situ* oxidation and reduction rates were not quantified, the presence of these important nitrogen-utilising microorganisms, confirms that simultaneous nitrification and denitrification processes contribute to the transformation of nitrogen via oxidative and reductive processes. The strong presence, and widespread abundance, of denitrifiers throughout the system, revealed that denitrification may play a fundamentally important role in the permanent removal of nitrogen from primary facultative WSP's.



This thesis is dedicated to my amazing parents, John and Felicity van der Linde, who have sacrificed, given, and been through much, to help me to get to where I am. I honour you for that. This work is for you with all of my love, joy, gratitude and appreciation – God bless you.



Dinitrogen and the Ammonia Molecule – taken from Pauling and Hayward, 1964.

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List of Abbreviations and Acronyms

ANNAMOX	Anaerobic ammonium oxidation
AOA	Ammonia oxidising archaea
AOB	Ammonia oxidising bacteria
BOD	Biochemical oxygen demand
COD	Chemical oxygen demand
DGGE	Denaturing gradient gel electrophoresis
DO	Dissolved oxygen
EU	European Union
FC	Faecal coliforms
HRT	Hydraulic retention time
PCR	Polymerase chain reaction
p.e.	Population equivalent
PFP	Primary facultative pond(s)
ORP	Oxidation reduction potential
RTD	Residence time distribution
RWT	Rhodamine WT
SS	Suspended solids
TKN	Total Kjeldahl nitrogen
UK	United Kingdom
UWWTD	Urban wastewater treatment directive
VFA	Volatile fatty acids
WHO	World Health Organization
WSP	Wastewater stabilization pond(s)
WWTW	Wastewater treatment works
θ_0	Theoretical/nominal HRT
θ	Actual HRT
t^{-}	Mean HRT