# Dissolved Organic Nitrogen oxidation collected on cruise SAV 17-16 in the South Atlantic Bight aboard the R/V Savannah from 2011 to 2017

Website: https://www.bco-dmo.org/dataset/767048

Data Type: Cruise Results

Version: 1

**Version Date**: 2019-05-08

#### **Project**

» Collaborative Research: Direct Oxidation of Organic Nitrogen by Marine Ammonia Oxidizing Organisms (DON Oxidation)

Contributors	Affiliation	Role
Hollibaugh, James T.	University of Georgia (UGA)	Principal Investigator
Popp, Brian N.	University of Hawaii	Co-Principal Investigator
Biddle, Mathew	Woods Hole Oceanographic Institution (WHOI BCO-DMO)	BCO-DMO Data Manager

#### **Abstract**

This dataset contains the results of analyses related to ammonia oxidation rates, including oxidation rates of 15N supplied as ammonia, urea, 1,2 diamino ethane, 1,3 diamino propane, 1,4 diamino butane (putrescine), arginine and glutamate. Ancillary data including nutrient concentrations and the abundance of ammonia- and nitrite-oxidizing microorganisms are also reported. The samples analyzed to produce the dataset were collected off the coast of Georgia, USA. Most data were collected on one cruise in August 2017, incidental data from 2011, 2013 and 2016 are also reported.

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## Coverage

**Spatial Extent**: **N**:31.99 **E**:-78.765 **S**:30.3175 **W**:-81.356

**Temporal Extent**: 2011-10-04 - 2017-08-19

# **Dataset Description**

Samples were collected from four regions (inshore, midshelf, shelf-break, and oceanic) of the SAB off the Georgia (U.S.A.) coast (Fig. 1; Supporting Information Table S1), with terminology modified from Liu et al. (2018) as follows. "Inshore" stations were within the barrier island complex. "Mid-shelf" stations were outside the barrier island complex to depths < 40 m; due to limited sampling in this zone, no demarcation between "mid-shelf" and "nearshore" stations (as in Liu et al. 2018) was made. "Shelf-break" stations were between 40 m and 500 m depth. While Liu et al. (2018) did not sample waters past the shelf-break, we included deeper stations further offshore (bottom depth > 500 m), which are designated "oceanic" stations. Note that the maximum depth sampled was  $\leq$  500 m due to equipment limitations.

Inshore samples were collected from a dock at Marsh Landing on the Duplin River (Sapelo Island) and the dock at the Skidaway Institute of Oceanography (Fig. 1). Both inshore sites are salt marsh-dominated estuaries. Water from both sites was sampled from a depth ≤ 1 m and was processed immediately at a nearby laboratory (the University of Georgia Marine Institute on Sapelo Island or onboard the R/V Savannah). Water quality data for Marsh Landing samples were collected as part of the Sapelo Island National Estuarine Research Reserve monitoring program. Relevant data from the Lower Duplin ("LD") sonde were downloaded from NOAA/CDMO (http://cdmo.baruch.sc.edu/aqs/, last accessed 22 May 2018).

Most SAB samples were collected in August 2017 on the R/V Savannah (cruise SAV-17-16) along transects across the continental shelf and the Gulf Stream and into the western Sargasso Sea, with sampling focused around the shelf-break (Fig. 1). Water from multiple depths was collected using 12-liter Niskin bottles mounted on a rosette equipped with a Sea-Bird SBE25 CTD. Profiles of salinity, temperature, dissolved oxygen, fluorescence, and photosynthetically active radiation (PAR) were collected using the CTD system as described previously (Liu et al. 2018). PAR attenuation (K<sub>d</sub>) was calculated from plots of ln(PAR) vs. depth as in Liu et al. (2018). Two additional SAB stations were sampled in October 2011 (described previously by Liu et al. 2015 and Tolar et al. 2017) and are referred to as "2011-4" and "2011-12" (Fig. 1). Environmental data and some of the microbial and rate data from 2011 stations are available in other publications (Liu et al. 2015; Tolar et al. 2017; see BCO-DMO dataset qPCR\_Parameters at <a href="https://www.bco-dmo.org/dataset/767141">https://www.bco-dmo.org/dataset/767141</a>).

#### **Acquisition Description**

#### **Nutrient analysis**

Nutrient samples were filtered through 0.22 µm pore size Durapore GVWP filters (Millipore Sigma) and frozen at –20\_C immediately after collection, then stored at –80\_C until analysis. Dissolved nitrate (NO3 –), nitrite (NO2 –), phosphate (PO4 3–), and silicate (SiO4 4–) were measured using a Bran and Luebbe AA3 autoanalyzer as described previously (Wilkerson et al. 2015). Ammonium and urea were measured manually using the phenolhypochlorite method (Solórzano 1969) and the diacetylmonoxime method (Rahmatullah and Boyde 1980; Mulvenna and Savidge 1992), respectively.

#### **Oxidation rate measurements**

We used 15N-labeled substrates (98–99% 15N, Cambridge Isotope Laboratories) to measure the oxidation of N supplied as NH4+, urea, 1,2-diaminoethane (DAE), 1,3-diaminopropane (DAP), 1,4-diaminobutane (putrescine, PUT), L-glutamic acid (GLU), and L-arginine (ARG). 15N oxidation from NH4+, urea, PUT, and GLU were measured extensively, whereas 15N oxidation from DAE, DAP, and ARG was only measured at a subset of stations (Supporting Information Table S1). GLU and ARG were included as a control for remineralization, as their central roles in microbial metabolism leads to rapid catabolism and NH4 + regeneration (Hollibaugh 1978; Goldman et al. 1987). PUT was used in routine assessments of the oxidation of polyamine-N because it is one of the most consistently detected polyamines in seawater (Nishibori et al. 2001a, 2003; Lu et al. 2014; Liu et al. 2015). Although spermine and spermidine are also common in seawater, 15N-labeled stocks of these polyamines were not commercially available. We measured the oxidation of N from DAE and DAP to investigate the effect of aliphatic chain length (which affects pKa) on oxidation rate.

Duplicate seawater samples contained in 1-liter polycarbonate or 250 mL high density polyethylene (HDPE) bottles wrapped with aluminum foil (to exclude light) were amended with 10–50 nM 15N-labeled substrate. Marsh Landing samples were then placed in an incubator held at in situ temperature in the dark. Samples taken at the Skidaway dock were placed in a mesh bag and immersed at the sea surface at the sampling site. Samples collected at sea were incubated in a tank of flowing surface seawater or in an incubator held at 18 C in the dark. Incubation bottles were sampled for 15N analysis immediately after substrate addition and again after a period of ~ 24 h. 15N samples were subsampled into 50 mL polypropylene centrifuge tubes, frozen at –20\_C, and stored at –80\_C until analysis. The 15N/14N ratios of the NO3 – plus NO2 – (NOX) pools (δ15NNOx) in the samples were measured using the bacterial denitrifier method to convert NOX to nitrous oxide (N2O; Sigman et al. 2001). The δ15N values of the N2O produced were measured using a Finnigan MAT-252 isotope ratio mass spectrometer coupled with a modified GasBench II

interface (Casciotti et al. 2002; Beman et al. 2011; McIlvin and Casciotti 2011). Oxidation rates were calculated using an endpoint model (Beman et al. 2011; Damashek et al. 2016). Since the substrates used were uniformly labeled with 15N, the amount of the N added as the 15N spike (in  $\mu$ M) was multiplied by the number of moles of 15N per mole of substrate, which assumes that all of the N atoms have equal probability of being oxidized. This is likely true for urea, DAE, DAP, and PUT, which are symmetrical molecules, but not likely to be true for ARG, which contains 4 N atoms (one in the  $\alpha$ -amino position and three in the guanidine structure of its R-group). Abiotic oxidation of organic N was assessed by measuring 15NOX production following 15N amendment and incubation of 0.22  $\mu$ m filtered seawater (as described above), and potential metabolism of DON by the denitrifying bacteria used to convert NOX to N2O was checked by adding 15N-labeled substrates into the bacterial cultures prior to mass spectrometry.

We were unable to measure the in situ concentrations of the individual components of DON used in oxidation experiments, other than urea. Based on previous measurements made in the SAB (Lu et al. 2014; Liu et al. 2015), we assumed concentrations of 1 nM and 0.25 nM for DAE, DAP and PUT, and 10 nM and 5 nM for GLU and ARG, at inshore and midshelf/shelf-break/oceanic stations, respectively. Rates of polyamine and amino acid oxidation reported below should therefore be considered potential rates, as amendments as low as 10–50 nM are likely to increase substrate concentrations substantially above in situ. Initial substrate 15N activity was calculated using isotope mass balance using the known concentration and 15N activity of the labeled substrates added and assuming the concentrations described above and natural abundance 15N activity (i.e., 0.3663 atom% 15N).

#### **Processing Description**

#### **BCO-DMO Processing Notes:**

- added conventional header with dataset name, PI name, version date
- modified parameter names to conform with BCO-DMO naming conventions
- converted latitude and longitude values from degrees minutes seconds to degrees and appended a new column (lat, lon).
- converted Date column to ISO convention and appended ISO\_Date column for those values.
- replaced "x 10" with "e" to signify power of 10.
- split columns containing "±" into two columns, one for value and one for ± value. Appended
- " sd" to the ± value column.
- converted "nan" and blank cells to "nd".

#### **Related Publications**

Beman, J. M., Chow, C.-E., King, A. L., Feng, Y., Fuhrman, J. A., Andersson, A., ... Hutchins, D. A. (2010). Global declines in oceanic nitrification rates as a consequence of ocean acidification. Proceedings of the National Academy of Sciences, 108(1), 208–213. doi:10.1073/pnas.1011053108

Beman, J. M., Popp, B. N., & Francis, C. A. (2008). Molecular and biogeochemical evidence for ammonia oxidation by marine Crenarchaeota in the Gulf of California. The ISME Journal, 2(4), 429–441. doi:10.1038/ismej.2007.118

Damashek, J., Tolar, B. B., Liu, Q., Okotie-Oyekan, A. O., Wallsgrove, N. J., Popp, B. N., & Hollibaugh, J. T. (2018). Microbial oxidation of nitrogen supplied as selected organic nitrogen compounds in the South Atlantic Bight. Limnology and Oceanography. doi:10.1002/lno.11089

Mincer, T. J., Jensen, P. R., Kauffman, C. A., & Fenical, W. (2002). Widespread and Persistent Populations of a Major New Marine Actinomycete Taxon in Ocean Sediments. Applied and Environmental Microbiology, 68(10), 5005–5011. doi:10.1128/AEM.68.10.5005-5011.2002

Mosier, A. C., & Francis, C. A. (2011). Determining the Distribution of Marine and Coastal Ammonia-Oxidizing Archaea and Bacteria Using a Quantitative Approach. Methods in Enzymology, 205–221. doi:10.1016/B978-0-12-381294-0.00009-2

Santoro, A. E., Sakamoto, C. M., Smith, J. M., Plant, J. N., Gehman, A. L., Worden, A. Z., ... Casciotti, K. L. (2013). Measurements of nitrite production in and around the primary nitrite maximum in the central California Current. Biogeosciences, 10(11), 7395–7410. doi:10.5194/bq-10-7395-2013

Suzuki, M. T., Taylor, L. T., & DeLong, E. F. (2000). Quantitative Analysis of Small-Subunit rRNA Genes in Mixed Microbial Populations via 5'-Nuclease Assays. Applied and Environmental Microbiology, 66(11), 4605–4614. doi:10.1128/AEM.66.11.4605-4614.2000

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#### **Parameters**

Parameter	Description	Units
Cruise_ID	R2R catalog identifier for this cruise	unitless

Sta	Station Identifier. Marsh Landing is on the Duplin River a tidal channel adjacent to Sapelo Island GA.	unitless
Region	Arbitratry assignment of stations to zones in the study area identified by location and water properties: I = Inshore; M = midshelf; S = Shelfbreak; O = Oceanic	unitless
depth	Depth sampled in meters	meters
lon	degrees longitude with positive values eastward	decimal degrees
lat	degrees latitude with positive values northward	decimal degrees
Latitude	Latitude in degrees and decimal minutes N	degrees and decimal minutes N
Longitude	Longitude in degrees and decimal minutes W	degrees and decimal minutes W
Date	Sampling date: MM/DD/YYYY	unitless
ISO_Date	Date following the ISO convention of YYYY-MM-DD	unitless

D : 1 1 400 DNA	0	
Bacterial_16S_rRNA	Concentration of genes for Bacteria 16S rRNA	copies per
	determined by qPCR (see qPCR parameter	liter filtered
	table) in units of copies L-1; Blank Cells = no	
	data; either not sampled or the sample was lost;	
	BLD = Below Limit of Detection. See qPCR	
	parameter table. qPCR data are from single	
	biological replicates; means of triplicate qPCR	
	reactions. Limit of detection is given as copies	
	per mL of template DNA; the limit thus varies by	
	sample depending on volume filtered; extract	
	volume and template volume per per reaction.	
	The limits given are from this sample set	
	assuming the minimum detectable concentration	
	in the template used and 1 uL of template per	
	reaction. They are thus conservative as some	
	reactions were run with 10 uL of template.	
Thaumarchaeal_16S_rRNA	Concentration of genes for Marine Group 1	copies per
i		
	Archaea (Thaumarchaeota) 16S rRNA	liter filtered
	Archaea (Thaumarchaeota) 16S rRNA determined by qPCR (see qPCR parameter	liter filtered
	, , , , , , , , , , , , , , , , , , ,	liter filtered
	determined by qPCR (see qPCR parameter	liter filtered
	determined by qPCR (see qPCR parameter table) in units of copies L-1 Blank Cells = no	liter filtered
	determined by qPCR (see qPCR parameter table) in units of copies L-1 Blank Cells = no data either not sampled or the sample was lost;	liter filtered
	determined by qPCR (see qPCR parameter table) in units of copies L-1 Blank Cells = no data either not sampled or the sample was lost; BLD = Below Limit of Detection. See qPCR	liter filtered
	determined by qPCR (see qPCR parameter table) in units of copies L-1 Blank Cells = no data either not sampled or the sample was lost; BLD = Below Limit of Detection. See qPCR parameter table. qPCR data are from single	liter filtered
	determined by qPCR (see qPCR parameter table) in units of copies L-1 Blank Cells = no data either not sampled or the sample was lost; BLD = Below Limit of Detection. See qPCR parameter table. qPCR data are from single biological replicates; means of triplicate qPCR	liter filtered
	determined by qPCR (see qPCR parameter table) in units of copies L-1 Blank Cells = no data either not sampled or the sample was lost; BLD = Below Limit of Detection. See qPCR parameter table. qPCR data are from single biological replicates; means of triplicate qPCR reactions. Limit of detection is given as copies	liter filtered
	determined by qPCR (see qPCR parameter table) in units of copies L-1 Blank Cells = no data either not sampled or the sample was lost; BLD = Below Limit of Detection. See qPCR parameter table. qPCR data are from single biological replicates; means of triplicate qPCR reactions. Limit of detection is given as copies per mL of template DNA; the limit thus varies by	liter filtered
	determined by qPCR (see qPCR parameter table) in units of copies L-1 Blank Cells = no data either not sampled or the sample was lost; BLD = Below Limit of Detection. See qPCR parameter table. qPCR data are from single biological replicates; means of triplicate qPCR reactions. Limit of detection is given as copies per mL of template DNA; the limit thus varies by sample depending on volume filtered extract	liter filtered
	determined by qPCR (see qPCR parameter table) in units of copies L-1 Blank Cells = no data either not sampled or the sample was lost; BLD = Below Limit of Detection. See qPCR parameter table. qPCR data are from single biological replicates; means of triplicate qPCR reactions. Limit of detection is given as copies per mL of template DNA; the limit thus varies by sample depending on volume filtered extract volume and template volume per per reaction.	liter filtered
	determined by qPCR (see qPCR parameter table) in units of copies L-1 Blank Cells = no data either not sampled or the sample was lost; BLD = Below Limit of Detection. See qPCR parameter table. qPCR data are from single biological replicates; means of triplicate qPCR reactions. Limit of detection is given as copies per mL of template DNA; the limit thus varies by sample depending on volume filtered extract volume and template volume per per reaction. The limits given are from this sample set	liter filtered
	determined by qPCR (see qPCR parameter table) in units of copies L-1 Blank Cells = no data either not sampled or the sample was lost; BLD = Below Limit of Detection. See qPCR parameter table. qPCR data are from single biological replicates; means of triplicate qPCR reactions. Limit of detection is given as copies per mL of template DNA; the limit thus varies by sample depending on volume filtered extract volume and template volume per per reaction. The limits given are from this sample set assuming the minimum detectable concentration	liter filtered

WCA_amoA	Concentration of genes for Archaeal ammonia monooxygenase subunit A from the Water Column A clade; determined by qPCR (see qPCR parameter table) in units of copies L-1; Blank Cells = no data; either not sampled or the sample was lost; BLD = Below Limit of Detection. See qPCR parameter table. qPCR data are from single biological replicates; means of triplicate qPCR reactions. Limit of detection is given as copies per mL of template DNA; the limit thus varies by sample depending on volume filtered; extract volume and template volume per per reaction. The limits given are from this sample set assuming the minimum detectable concentration in the template used and 1 uL of template per reaction. They are thus conservative as some reactions were run with 10 uL of template.	copies per liter filtered
WCB_amoA	Concentration of genes for Archaeal ammonia monooxygenase subunit A from the Water Column A clade; determined by qPCR (see qPCR parameter table) in units of copies L-1; "Blank Cells = no data either not sampled or the sample was lost; BLD = Below Limit of Detection. See qPCR parameter table. qPCR data are from single biological replicates means of triplicate qPCR reactions. Limit of detection is given as copies per mL of template DNA; the limit thus varies by sample depending on volume filtered extract volume and template volume per per reaction. The limits given are from this sample set assuming the minimum detectable concentration in the template used and 1 uL of template per reaction. They are thus conservative as some reactions were run with 10 uL of template.	copies per liter filtered

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Nitrospina_16S_rRNA	Concentration of genes for Nitrospina 16S rRNA determined by qPCR (see qPCR parameter table) in units of copies L-1; Blank Cells = no data; either not sampled or the sample was lost; BLD = Below Limit of Detection. See qPCR parameter table. qPCR data are from single biological replicates; means of triplicate qPCR reactions. Limit of detection is given as copies per mL of template DNA; the limit thus varies by sample depending on volume filtered; extract volume and template volume per per reaction. The limits given are from this sample set assuming the minimum detectable concentration in the template used and 1 uL of template per reaction. They are thus conservative as some reactions were run with 10 uL of template.	copies per liter filtered
N15_added	Final concentration of uniformly 15N labeled test substrates	nanomoles per liter
Ammonia_Oxidation	Ammonia oxidation rate determined from conversion of 15N-labeled ammonium to 15N-labeled nitrite plus nitrate; BLD = Below Limit of Detection. 15N data are mean ± S.E.M. of duplicate biological replicates (see Damashek et al. 2018 for details) . Based on a propagation of error calculation; our conservative estimate of the precision of 15N measurements ±4% for samples at natural abundance and ±5.2% for	nanomoles per liter per day

Ammonia_Oxidation_sd	Deviation of ammonia oxidation rate determined	nanomoles
/ <u>_</u>	from conversion of 15N-labeled ammonium to	per liter per
	15N-labeled nitrite plus nitrate; BLD = Below	day
	Limit of Detection. 15N data are mean ± S.E.M.	
	of duplicate biological replicates (see	
	Damashek et al. 2018 for details). Based on a	
	propagation of error calculation; our	
	conservative estimate of the precision of 15N	
	measurements ±4% for samples at natural	
	abundance and ±5.2% for samples artificially	
	enriched with carrier 15N. Our limit of detection	
	was similar to that reported by Santoro et al.	
	(2013) and Beman et al. (2011).	
N15_ox_from_PUT	Oxidation rate of 15N from putrescine (1;4	nanomoles
	diamino butane) determined from conversion of	per liter per
	the 15N label to 15N-labeled nitrite plus nitrate;	day
	BLD = Below Limit of Detection. 15N data are	
	mean ± S.E.M. of duplicate biological replicates	
	(see Damashek et al. 2018 for details) . Based	
	on a propagation of error calculation our	
	conservative estimate of the precision of 15N	
	measurements ±4‰ for samples at natural	
	abundance and ±5.2% for samples artificially	
	enriched with carrier 15N. Our limit of detection	
	was similar to that reported by Santoro et al.	
	(2013) and Beman et al. (2011).	

N15_ox_from_PUT_sd	Deviation of oxidation rate of 15N from	nanomoles
<u></u>	putrescine (1;4 diamino butane) determined	per liter per
	from conversion of the 15N label to 15N-labeled	day
	nitrite plus nitrate; BLD = Below Limit of	,
	Detection. 15N data are mean ± S.E.M. of	
	duplicate biological replicates (see Damashek	
	et al. 2018 for details) . Based on a propagation	
	of error calculation our conservative estimate of	
	the precision of 15N measurements ±4% for	
	samples at natural abundance and ±5.2% for	
	samples artificially enriched with carrier 15N.	
	Our limit of detection was similar to that reported	
	by Santoro et al. (2013) and Beman et al.	
	(2011).	
N15_ox_from_GLU	Oxidation rate of 15N from L-glutamate	nanomoles
	determined from conversion of the 15N label to	per liter per
	15N-labeled nitrite plus nitrate; BLD = Below	day
	Limit of Detection. 15N data are mean ± S.E.M.	
	of duplicate biological replicates (see	
	Damashek et al. 2018 for details) . Based on a	
	propagation of error calculation; our	
	conservative estimate of the precision of 15N	
	measurements ±4‰ for samples at natural	
	abundance and ±5.2% for samples artificially	
	enriched with carrier 15N. Our limit of detection	
	was similar to that reported by Santoro et al.	
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	I .	
N15_ox_from_GLU_sd	deviation of oxidation rate of 15N from L-glutamate determined from conversion of the 15N label to 15N-labeled nitrite plus nitrate; BLD = Below Limit of Detection. 15N data are mean ± S.E.M. of duplicate biological replicates (see Damashek et al. 2018 for details) . Based on a propagation of error calculation; our conservative estimate of the precision of 15N measurements ±4‰ for samples at natural abundance and ±5.2‰ for samples artificially enriched with carrier 15N. Our limit of detection was similar to that reported by Santoro et al. (2013) and Beman et al. (2011).	nanomoles per liter per day
N15_ox_from_UREA	Oxidation rate of 15N from urea determined from conversion of the 15N label to 15N-labeled nitrite plus nitrate BLD = Below Limit of Detection. 15N data are mean ± S.E.M. of duplicate biological replicates (see Damashek et al. 2018 for details) . Based on a propagation of error calculation; our conservative estimate of the precision of 15N measurements ±4% for samples at natural abundance and ±5.2% for samples artificially enriched with carrier 15N. Our limit of detection was similar to that reported by Santoro et al. (2013) and Beman et al. (2011).	nanomoles per liter per day
N15_ox_from_UREA_sd	Deviation of oxidation rate of 15N from urea determined from conversion of the 15N label to 15N-labeled nitrite plus nitrate BLD = Below Limit of Detection. 15N data are mean ± S.E.M. of duplicate biological replicates (see Damashek et al. 2018 for details). Based on a propagation of error calculation; our conservative estimate of the precision of 15N measurements ±4% for samples at natural abundance and ±5.2% for samples artificially enriched with carrier 15N. Our limit of detection was similar to that reported by Santoro et al. (2013) and Beman et al. (2011).	nanomoles per liter per day

N15_ox_from_DAE	Oxidation rate of 15N from 1;2 diamino ethane determined from conversion of the 15N label to 15N-labeled nitrite plus nitrate; BLD = Below Limit of Detection. 15N data are mean ± S.E.M. of duplicate biological replicates (see Damashek et al. 2018 for details). Based on a propagation of error calculation; our conservative estimate of the precision of 15N measurements ±4% for samples at natural abundance and ±5.2% for samples artificially enriched with carrier 15N. Our limit of detection was similar to that reported by Santoro et al. (2013) and Beman et al. (2011).	nanomoles per liter per day
N15_ox_from_DAE_sd	Deviation of oxidation rate of 15N from 1;2 diamino ethane determined from conversion of the 15N label to 15N-labeled nitrite plus nitrate; BLD = Below Limit of Detection. 15N data are mean ± S.E.M. of duplicate biological replicates (see Damashek et al. 2018 for details). Based on a propagation of error calculation; our conservative estimate of the precision of 15N measurements ±4% for samples at natural abundance and ±5.2% for samples artificially enriched with carrier 15N. Our limit of detection was similar to that reported by Santoro et al. (2013) and Beman et al. (2011).	nanomoles per liter per day
N15_ox_from_DAP	Oxidation rate of 15N from 1;3 diamino propane determined from conversion of the 15N label to 15N-labeled nitrite plus nitrate; BLD = Below Limit of Detection. 15N data are mean ± S.E.M. of duplicate biological replicates (see Damashek et al. 2018 for details). Based on a propagation of error calculation; our conservative estimate of the precision of 15N measurements ±4% for samples at natural abundance and ±5.2% for samples artificially enriched with carrier 15N. Our limit of detection was similar to that reported by Santoro et al. (2013) and Beman et al. (2011).	nanomoles per liter per day

N15_ox_from_DAP_sd	Deviation of oxidation rate of 15N from 1;3 diamino propane determined from conversion of the 15N label to 15N-labeled nitrite plus nitrate; BLD = Below Limit of Detection. 15N data are mean ± S.E.M. of duplicate biological replicates (see Damashek et al. 2018 for details) . Based on a propagation of error calculation; our conservative estimate of the precision of 15N measurements ±4% for samples at natural abundance and ±5.2% for samples artificially enriched with carrier 15N. Our limit of detection was similar to that reported by Santoro et al. (2013) and Beman et al. (2011).	nanomoles per liter per day
N15_ox_from_ARG	Oxidation rate of 15N from L-arginine determined from conversion of the 15N label to 15N-labeled nitrite plus nitrate; BLD = Below Limit of Detection. 15N data are mean ± S.E.M. of duplicate biological replicates (see Damashek et al. 2018 for details). Based on a propagation of error calculation our conservative estimate of the precision of 15N measurements ±4% for samples at natural abundance and ±5.2% for samples artificially enriched with carrier 15N. Our limit of detection was similar to that reported by Santoro et al. (2013) and Beman et al. (2011).	nanomoles per liter per day
N15_ox_from_ARG_sd	Deviation of oxidation rate of 15N from Larginine determined from conversion of the 15N label to 15N-labeled nitrite plus nitrate; BLD = Below Limit of Detection. 15N data are mean ± S.E.M. of duplicate biological replicates (see Damashek et al. 2018 for details). Based on a propagation of error calculation our conservative estimate of the precision of 15N measurements ±4% for samples at natural abundance and ±5.2% for samples artificially enriched with carrier 15N. Our limit of detection was similar to that reported by Santoro et al. (2013) and Beman et al. (2011).	nanomoles per liter per day

Nitrate	Concentration of nitrate determined by cadmium reduction to nitrite followed by subtraction nitrite already present in the sample; Samples run on an autoanalyzer by Francis Wilkerson's lab at San Francisco State University	micromoles per liter
Nitrite	Concentration of nitrite determined by autoanalyzer; Samples run on an autoanalyzer by Francis Wilkerson's lab at San Francisco State University	micromoles per liter
Ammonium	Concentration of ammonium determined by autoanalyzer; Samples run on an autoanalyzer by Francis Wilkerson's lab at San Francisco State University	micromoles per liter
Urea	Concentration of urea determined by the carboxythiazole method; Samples run in Hollibaugh lab at UGA	micromoles per liter
Silicate	Concentration of silicate determined by autoanalyzer; Samples run on an autoanalyzer by Francis Wilkerson's lab at San Francisco State University	micromoles per liter
Phosphate	Concentration of phosphate determined by autoanalyzer; Samples run on an autoanalyzer by Francis Wilkerson's lab at San Francisco State University	micromoles per liter
Temperature	Water temperature in centigrade degrees at the depth sampled measured by the environmental sensing package on the samplling rosette	degrees centigrade
Salinity	Salinity at the depth sampled derived from temperature and conductivity measured by the environmental sensing package on the samplling rosette	PSU
Diss_Oxygen	Dissolved oxygen concentration at the depth sampled measured by the environmental sensing package on the samplling rosette	milliliters per liter

Relative_Fluor  table of contents   back to top		Relative Fluorescence measured by the fluorometer on the Niskin rosette sampler converted to mg Chl a L-1 using a regression equation based on extracted chlorophyll data:  Chl a = 1.7869RF - 2.2541; R <sup>2</sup> = 0.83	relative fluorescence
Instruments Atten_Coeff		PAR attenuation coefficient kz in m-1 calculated as the slope of log(PAR) vs depth	per meter
Dataset- comment specific Instrument Name	Sea-Bird SBE2	identifier for the comment. comment value a signifies Environmental data, bacterial 16S, thaumarchaeal 16S, AOB amoA, ammonia	unitless
Generic Instrument Name	CTD Sea-Bird 2	oxidation, and urea oxidation data were reported previously in Liu et al. (2015) or Tolar et al. (2017);	
Dataset- specific Description	Water from multiple depths was collected using 12-liter Niskin bottles mounted on a rosette equipped with a Sea-Bird SBE25 CTD. Profiles of salinity, temperature, dissolved oxygen, fluorescence, and photosynthetically active radiation (PAR) were collected using the CTD system as described previously (Liu et al. 2018).		
Generic Instrument Description	The Sea-Bird SBE 25 SEALOGGER CTD is battery powered and is typically used to record data in memory, eliminating the need for a large vessel, electrical sea cable, and on-board computer. All SBE 25s can also operate in real-time, transmitting data via an opto-isolated RS-232 serial port.  Temperature and conductivity are measured by the SBE 3F Temperature sensor and SBE 4 Conductivity sensor (same as those used on the premium SBE 9plus CTD). The SBE 25 also includes the SBE 5P (plastic) or 5T (titanium) Submersible Pump and TC Duct. The pump-controlled, TC-ducted flow configuration significantly reduces salinity spiking caused by ship heave, and in calm waters allows slower descent rates for improved resolution of water column features. Pressure is measured by the modular SBE 29 Temperature Compensated Strain-Gauge Pressure sensor (available in eight depth ranges to suit the operating depth requirement). The SBE 25's modular design makes it easy to configure in the field for a wide range of auxiliary sensors, including optional dissolved oxygen (SBE 43), pH (SBE 18 or SBE 27), fluorescence, transmissivity, PAR, and optical backscatter sensors. More information from Sea-Bird Electronics: <a href="http://www.seabird.com">http://www.seabird.com</a> .		

Dataset- specific Instrument Name	Finnigan MAT-252 isotope ratio mass spectrometer
Generic Instrument Name	Isotope-ratio Mass Spectrometer
Dataset- specific Description	The δ15N values of the N2O produced were measured using a Finnigan MAT-252 isotope ratio mass spectrometer coupled with a modified GasBench II interface
Generic Instrument Description	The Isotope-ratio Mass Spectrometer is a particular type of mass spectrometer used to measure the relative abundance of isotopes in a given sample (e.g. VG Prism II Isotope Ratio Mass-Spectrometer).

Dataset- specific Instrument Name	Bran and Luebbe AA3 autoanalyzer
Generic Instrument Name	Bran Luebbe AA3 AutoAnalyzer
Dataset- specific Description	Dissolved nitrate (NO3 –), nitrite (NO2 –), phosphate (PO4 3–), and silicate (SiO4 4–) were measured using a Bran and Luebbe AA3 autoanalyzer as described previously (Wilkerson et al. 2015).
Generic Instrument Description	Bran Luebbe AA3 AutoAnalyzer See the description from the manufacturer.

Dataset- specific Instrument Name	C1000 Touch Thermal Cycler
Generic Instrument Name	qPCR Thermal Cycler
Dataset- specific Description	All reactions (25 µL total volume) were run in triplicate on a C1000 Touch Thermal Cycler equipped with a CFX96 Real-Time System (Bio-Rad), using either the iTaq Universal Green SYBR Mix (Bio-Rad) or the Platinum qPCR SuperMix-UDG (Thermo Fisher).
Generic Instrument Description	An instrument for quantitative polymerase chain reaction (qPCR), also known as real-time polymerase chain reaction (Real-Time PCR).

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# **Deployments**

## **SAV-17-16**

Website	https://www.bco-dmo.org/deployment/767055
Platform	R/V Savannah
Start Date	2017-08-16
End Date	2017-08-19

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# **Project Information**

# Collaborative Research: Direct Oxidation of Organic Nitrogen by Marine Ammonia Oxidizing Organisms (DON Oxidation)

**Coverage**: Coastal waters and the South Atlantic Bight continental shelf from Savannah GA out to the shelf break (SAV 17-16, UNOLS STR \_104733, Marsden Grid 117, Navy Ops NA06), coastal waters around Sapelo Island, Georgia USA

NSF Abstract: Nitrogen is an essential nutrient for phytoplankton that often limits primary production in the ocean, and its availability therefore plays a key role in global ocean productivity. The amounts and form in which nitrogen exist are controlled by microorganisms. One microorganism-mediated process is known as nitrification, which oxidizes ammonia or ammonium to nitrite and nitrite to nitrate, nitrate being the bioavailable form of nitrogen. While this is the well-accepted process of nitrification, preliminary results strongly suggest that a nitrogen-containing compound know as polyamine nitrogen may be directly converted by some microorganisms to nitrate. However, the importance of this process for global biogeochemical nitrogen cycling is unknown. The goal of this study is to evaluate the biogeochemical significance of direct oxidation of polyamine nitrogen, as a model organic nitrogen compound, to nitrification compared to canonical nitrification of ammonia. The project will result in training a postdoctoral researcher and provide opportunities for undergraduates to gain hands-on experience with research on microbial geochemistry and coastal ecosystem processes. Project personnel will also work with the Georgia Coastal Ecosystems Long-Term Ecological Research program to engage a K-12 science teacher in the project. Ammonia oxidation is a key step in the process of converting fixed nitrogen to dinitrogen gas and thus is central to the global nitrogen cycle and to removing excess fixed nitrogen from coastal waters with high concentrations of nutrients. Recent research has shown that Thaumarchaeota play a major role in ammonia oxidation in the ocean. Experiments with enrichment cultures and coastal water samples where ammonia oxidizing archaea are the dominant ammonia oxidizers, show that some forms of organic nitrogen may be oxidized directly to nitrogen oxides without first being regenerated as ammonium. Of the substrates tested, polyamine and particularly putrescine nitrogen appear to be oxidized directly to nitrogen oxides, while amino acid and urea nitrogen is first regenerated as ammonium and then oxidized. The investigators will examine this process in detail over three years using enrichment cultures and experiments conducted with coastal bacterioplankton. Specifically, they will aim to better understand 1) the consequences of this novel process to ocean geochemistry, 2) the fate of the carbon present in polyamines, 3) what organisms are responsible for the direct oxidation, and 4) the chemical characteristics of the organic nitrogen compounds accessible to direct oxidation.

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