

# Ammonia oxidation, nitrite oxidation, and nitrate reduction rates from R/V Atlantis (AT15-61) cruise in Jan-Feb 2010 and R/V Melville (MV1104) cruise in Mar-Apr 2011 in the Eastern Tropical South Pacific

**Website:** <https://www.bco-dmo.org/dataset/826782>

**Data Type:** experimental, Cruise Results

**Version:** 1

**Version Date:** 2020-10-14

## Project

» [Expression of Microbial Nitrification in the Stable Isotopic Systematics of Oceanic Nitrite and Nitrate](#) (Microbial Nitrification)

| Contributors                         | Affiliation   | Role                   |
|--------------------------------------|---|------------------------|
| <a href="#">Casciotti, Karen L.</a>  | Stanford University                                 | Principal Investigator |
| <a href="#">Santoro, Alyson E.</a>   | University of California-Santa Barbara (UCSB)       | Contact                |
| <a href="#">Gerlach, Dana Stuart</a> | Woods Hole Oceanographic Institution (WHOI BCO-DMO) | BCO-DMO Data Manager   |

## Abstract

Ammonia oxidation, nitrite oxidation, and nitrate reduction rates from cruises R/V Atlantis (AT15-61) in Jan-Feb 2010 and R/V Melville (MV1104) in Mar-Apr 2011 in the Eastern Tropical South Pacific

---

## Table of Contents

- [Coverage](#)
  - [Dataset Description](#)
    - [Methods & Sampling](#)
    - [Data Processing Description](#)
  - [Data Files](#)
  - [Supplemental Files](#)
  - [Related Publications](#)
  - [Parameters](#)
  - [Instruments](#)
  - [Deployments](#)
  - [Project Information](#)
  - [Funding](#)
- 

## Coverage

**Spatial Extent:** N:-9.943 E:-80 S:-20.01 W:-100

**Temporal Extent:** 2010-02-02 - 2011-04-20

## Methods & Sampling

Seawater samples were collected on R/V Atlantis (AT15-61) cruise in Jan-Feb 2010 and on R/V Melville (MV1104) cruise in Mar-Apr 2011. Water samples were collected at discrete depths using Niskin bottle type rosette samplers equipped with either 24 bottles (10L), or 12 bottles (20L), and an SBE9plus conductivity-temperature-depth (CTD) sensor package (SeaBird Electronics, Bellevue, WA). Water for rate incubations was collected into 160 mL glass serum bottles (AT15-61) or 500 mL Tedlar sampling bags (MV1104). See below in Processing Description for further details.

Ammonia and nitrite oxidation rates were determined using  $^{15}\text{N}$  tracer additions. Full methods can be found in the manuscript Santoro et al. (submitted). As described below, incubation methods varied slightly between the first and second cruises.

In Year 1 (2010), rates were determined at four depths at all six stations. Incubations were conducted in 160 mL glass serum vials. Six serum bottles were filled and sealed from each incubation depth, spiked with  $^{15}\text{N}$  tracer (100-200 nM  $^{15}\text{NH}_4\text{Cl}$  or  $\text{Na}^{15}\text{NO}_2^-$ ), and incubated in flowing seawater incubators screened to mimic the *in situ* light environment (euphotic zone samples) or temperature controlled chambers (sub-euphotic zone). Duplicate bottles were sacrificed at timepoints of 0, 12, and 24 h from each incubation depth,  $0.2\ \mu\text{m}$  syringe-filtered into a 60 mL HDPE bottle, and frozen at  $-20^\circ\text{C}$ .

In Year 2 (2011), rates were determined at six depths at five stations. No rates were determined at Stn 5 in Year 2. Incubations were conducted in 500 mL Tedlar bags. Duplicate incubation bags per treatment were filled from the Niskin bottles using silicone tubing, and  $^{15}\text{N}$  tracer (200 nM  $^{15}\text{NH}_4\text{Cl}$  or  $\text{Na}^{15}\text{NO}_2^-$ ) was added via the septum injection port. As in the previous year, bags were incubated at as close to *in situ* light and temperature as possible. At timepoints of 0, 12, 24, and 36 h, 50 mL samples were drawn from each bag through the three-way sampling port using a 60 mL syringe. At each timepoint, incubation water was  $0.2\ \mu\text{m}$  syringe filtered into a 60 mL HDPE bottle tripled rinsed with sample, and frozen at  $-20^\circ\text{C}$ .

Frozen samples were transported frozen to the laboratory, thawed, and prepared for  $^{15}\text{NNO}_2 + \text{NO}_3$  analysis using the 'denitrifier method' (Sigman *et al.*, 2001). For nitrite oxidation rate samples, the added  $^{15}\text{NO}_2^-$  tracer was removed using sulfamic acid addition and subsequent neutralization with NaOH (Granger *et al.*, 2006) prior to sample preparation and analysis. For 2010 samples, where only three timepoints were taken, rates were calculated using the linear fitting method of Dugdale and Goering (Dugdale and Goering, 1967). For 2011, where four timepoints were taken, rates were calculated using a least squares fitting approach that accounts for changes in  $^{15}\text{NNO}_2 + \text{NO}_3$  from co-occurring nitrate uptake (Santoro *et al.*, 2010 and attached analysis files).

Nitrate reduction rates to nitrite were determined in Year 2 using  $^{15}\text{NO}_3^-$  tracer additions ( $>98\ \text{atm}\%$   $\text{Na}^{15}\text{NO}_3$ ).  $^{15}\text{NO}_3^-$  incubations were only conducted in the euphotic zone (three depths). Tedlar incubation bags were prepared and filled as above, and 200 or 400 nM (final concentration) of  $\text{Na}^{15}\text{NO}_3$  was added to each bag using a plastic syringe. Timepoints were sampled and preserved as for the nitrification rate incubations above. In the laboratory, samples were prepared for  $^{15}\text{NNO}_2$  determination using the 'azide method' (McIlvin and Altabet, 2005). Following azide conversion to  $\text{N}_2\text{O}$ , samples and standards ( $\text{N}_23$ ,  $\text{N}_7373$ , and  $\text{N}_10219$ ; Casciotti *et al.*, 2007) were analyzed by IRMS and rates were calculated as described above.

Light inhibition experiments were conducted in Year 2 to test the effect of sunlight on ammonia oxidation, nitrite oxidation, and nitrate reduction. These incubations were conducted at the two shallowest incubation depths, approximating the 1% and 10% light depths at Stations 7, 9, and 11. For these experiments, one set of duplicate incubation bottles for each rate type was incubated at ambient light (bottles 1 and 2 in the data file) and the other in the dark (bottles 3 and 4 in the data file). Tracer addition, subsampling, analysis, and rate calculations were as described above.

## Data Processing Description

MATLAB processing:

The MATLAB script 'etsp\_rates\_2011\_indvT0.m' loads two types of files: initial conditions ('initials...') and isotope data ('data...'). Compiled rates were deposited in a tab-delimited text file. For more information, see Supplemental Files section.

Parameters for Initial Conditions file(s) called "initials"

- AP15N = Starting atom percent of  $^{15}\text{N}$  in the substrate pool;  $[^{15}\text{N}/(^{15}\text{N}+^{14}\text{N})]$ , expressed as a fraction (e.g. 99atm% is 0.99)
- Init\_conc\_No\_15 = Initial concentration of  $^{15}\text{N}$  (micromoles) in the product pool
- Init\_conc\_No\_14 = Initial concentration of  $^{14}\text{N}$  (micromoles) in the product pool

Parameters for the Isotope Data file(s) called "data":

- bottle = bottle (either replicate or treatment)
- time = time duration in hours
- 15R = ratio of  $^{15}\text{N}$  to  $^{14}\text{N}$  in nitrate

BCO-DMO processing:

- Added a conventional header with dataset name, PI name, version date
- Adjusted parameter names to comply with database requirements.

- Combined year, month, day fields into one date field.
- Units removed and added to Parameter Description metadata section.
- Default missing identifier of 'NaN' replaced with 'nd' ('nd' is BCO-DMO system default missing data identifier), but BDL (below detection limit) was preserved.

[ [table of contents](#) | [back to top](#) ]

## Data Files

| File  |
|---|
| <b>TableS2_ETSP_15N_rates.csv</b> (Comma Separated Values (.csv), 8.13 KB)<br>MD5:cd44f916bd80e224737e8a8529738fa5<br><br>Primary data file for dataset ID 826782 |

[ [table of contents](#) | [back to top](#) ]

## Supplemental Files

| File   |
|--|
| <b>etsp_rates_2011_indvT0.m</b> (MATLAB Programming Script (.m), 4.01 KB)<br>MD5:429d25c9444bc3bf1b7b0bef8cb5a1db<br><br>MATLAB script calculates the nitrification rates for six stations in the Eastern Tropical South Pacific in 2011. Calls the function 'nitr_rate_rev.m' and requires the optimization toolbox to use lsqcurvefit.m  |
| <b>initial_conditions_files.zip</b> (ZIP Archive (ZIP), 40.95 KB)<br>MD5:ba02f9983cf4d72be399458094fbbcaf<br><br>Contains 24 files of initial conditions that are used with this dataset's Supplemental Files (listed below) to calculate rates of ammonia oxidation, nitrite oxidation, and nitrate reduction: <ul style="list-style-type: none"> <li>• MATLAB script 'etsp_rates_2011_indvT0.m'</li> <li>• MATLAB function 'nitr_rate_rev.m'</li> <li>• raw_data_files named by station, nitrogen compound, and incubation conditions</li> </ul><br>To be compatible with the script, the data files do not have headers. The columns are: <ol style="list-style-type: none"> <li>1. depth (in meters)</li> <li>2. AP15N (starting atom percent of 15N in the substrate pool expressed as a fraction)</li> <li>3. No_15 (initial concentration of 15N (uM) in the product pool)</li> <li>4. No_14 (initial concentration of 14N (uM) in the product pool)</li> </ol> |
| <b>nitr_rate_rev.m</b> (MATLAB Programming Script (.m), 1.08 KB)<br>MD5:6a581e82b432110ec638492769442107<br><br>MATLAB function defining the change in 15R (15N/14N ratio) over time, used to calculate rates of 15N-labeled substrate production (NO2- or NO3-) in micromoles (uM) per hour given a time series of 15R-NO2- or 15R-NO3-.  |

## File

### raw\_data\_files.zip

(ZIP Archive (ZIP), 9.31 KB)

MD5:90094f1ecc7eec683b9b652a7494abd9

Contains 22 raw data files that are used with this dataset's Supplemental Files (listed below) to calculate rates of ammonia oxidation, nitrite oxidation, and nitrate reduction:

- MATLAB script 'etsp\_rates\_2011\_indvT0.m'
- MATLAB function 'nitr\_rate\_rev.m')
- initial conditions files named by station and nitrogen compound

To be compatible with the script, the data files do not have headers. The columns are:

1. depth (in meters)
2. bottle (replicate and/or treatment)
3. time (duration in hours)
4. 15R-product pool where 15R is 15N/14N ratio in NO<sub>2</sub> or NO<sub>3</sub>

[ [table of contents](#) | [back to top](#) ]

## Related Publications

Casciotti, K. L., Böhlke, J. K., McIlvin, M. R., Mroczkowski, S. J., & Hannon, J. E. (2007). Oxygen Isotopes in Nitrite: Analysis, Calibration, and Equilibration. *Analytical Chemistry*, 79(6), 2427–2436. doi:[10.1021/ac061598h](https://doi.org/10.1021/ac061598h)  
*Methods*

Dugdale, R. C., & Goering, J. J. (1967). UPTAKE OF NEW AND REGENERATED FORMS OF NITROGEN IN PRIMARY PRODUCTIVITY1. *Limnology and Oceanography*, 12(2), 196–206. doi:[10.4319/lm.1967.12.2.0196](https://doi.org/10.4319/lm.1967.12.2.0196)  
*Methods*

Granger, J., Sigman, D. M., Prokopenko, M. G., Lehmann, M. F., & Tortell, P. D. (2006). A method for nitrite removal in nitrate N and O isotope analyses. *Limnology and Oceanography: Methods*, 4(7), 205–212. doi:[10.4319/lom.2006.4.205](https://doi.org/10.4319/lom.2006.4.205)  
*Methods*

McIlvin, M. R., & Altabet, M. A. (2005). Chemical Conversion of Nitrate and Nitrite to Nitrous Oxide for Nitrogen and Oxygen Isotopic Analysis in Freshwater and Seawater. *Analytical Chemistry*, 77(17), 5589–5595. doi:[10.1021/ac050528s](https://doi.org/10.1021/ac050528s)  
*Methods*

Santoro, A. E., Buchwald, C., Knapp, A. N., Berelson, W. M., Capone, D. G., & Casciotti, K. L. (2020). Nitrification and nitrous oxide production in the offshore waters of the Eastern Tropical South Pacific. doi:[10.1002/essoar.10503499.1](https://doi.org/10.1002/essoar.10503499.1)  
*Results*

,  
*Methods*

Santoro, A. E., Buchwald, C., Knapp, A. N., Berelson, W. M., Capone, D. G., & Casciotti, K. L. (2021). Nitrification and Nitrous Oxide Production in the Offshore Waters of the Eastern Tropical South Pacific. *Global Biogeochemical Cycles*, 35(2). Portico. <https://doi.org/10.1029/2020gb006716>  
<https://doi.org/10.1029/2020GB006716>  
*Results*

Santoro, A. E., Casciotti, K. L., & Francis, C. A. (2010). Activity, abundance and diversity of nitrifying archaea and bacteria in the central California Current. *Environmental Microbiology*, 12(7), 1989–2006. doi:[10.1111/j.1462-2920.2010.02205.x](https://doi.org/10.1111/j.1462-2920.2010.02205.x)  
*Related Research*

Sigman, D. M., Casciotti, K. L., Andreani, M., Barford, C., Galanter, M., & Böhlke, J. K. (2001). A Bacterial Method for the Nitrogen Isotopic Analysis of Nitrate in Seawater and Freshwater. *Analytical Chemistry*, 73(17), 4145–

## Parameters

| Parameter         | Description   | Units                       |
|-------------------|---|-----------------------------|
| ISO_Date_Local    | Date of sampling in ISO format (yyyy-mm-dd), Time zone in 2010 was GMT-5, Time zone in 2011 was GMT-4   | yyyy-mm-dd                  |
| Latitude          | Latitude, South is negative   | decimal degrees             |
| Longitude         | Longitude, West is negative   | decimal degrees             |
| Station           | Station number  | unitless                    |
| Cast              | Cast number   | unitless                    |
| Depth             | Sample collection depth   | meters                      |
| Incubation_bottle | Experimental conditions indicator (1=light1, 2=light2, 3=dark1, 4=dark2)  | unitless                    |
| Amox              | Ammonia oxidation rate  | nanomoles per liter per day |
| Amox_SE           | Standard error of fit for ammonia oxidation rate  | nanomoles per liter per day |
| Amox_fitype       | Fit function type for ammonia oxidation rate calculation (1=non-linear least-squares fit to four or more points; 2=linear fit to three or fewer points) | unitless                    |
| Nitox             | Nitrite oxidation rate  | nanomoles per liter per day |
| Nitox_SE          | Standard error of fit for nitrite oxidation rate  | nanomoles per liter per day |
| Nitox_fitype      | Fit function type for ammonia oxidation rate calculation (1=non-linear least-squares fit to four or more points; 2=linear fit to three or fewer points) | unitless                    |
| NO3red            | Rate of nitrate to nitrite reduction  | nanomoles per liter per day |
|                   |   |                             |

|               |   |          |
|---------------|---|----------|
| NO3red_fitype | Fit function type for ammonia oxidation rate calculation (1=non-linear least-squares fit to four or more points; 2=linear fit to three or fewer points) | unitless |
| Year          | Year of sample collection   | unitless |
| Month         | Month of sample collection (local)  | unitless |
| Day           | Day of sample collection (local)  | unitless |

[ [table of contents](#) | [back to top](#) ]

## Instruments

|   |  |
|---|--|
| <b>Dataset-specific Instrument Name</b> | SBE9plus conductivity-temperature-depth (CTD) sensor package   |
| <b>Generic Instrument Name</b>          | CTD Sea-Bird 9   |
| <b>Dataset-specific Description</b>     | SBE9plus conductivity-temperature-depth (CTD) sensor package   |
| <b>Generic Instrument Description</b>   | The Sea-Bird SBE 9 is a type of CTD instrument package. The SBE 9 is the Underwater Unit and is most often combined with the SBE 11 Deck Unit (for real-time readout using conductive wire) when deployed from a research vessel. The combination of the SBE 9 and SBE 11 is called a SBE 911. The SBE 9 uses Sea-Bird's standard modular temperature and conductivity sensors (SBE 3 and SBE 4). The SBE 9 CTD can be configured with auxiliary sensors to measure other parameters including dissolved oxygen, pH, turbidity, fluorometer, altimeter, etc.). Note that in most cases, it is more accurate to specify SBE 911 than SBE 9 since it is likely a SBE 11 deck unit was used. more information from Sea-Bird Electronics |

|   |   |
|---|---|
| <b>Dataset-specific Instrument Name</b> | Thermo Delta Plus XP isotope ratio mass spectrometer  |
| <b>Generic Instrument Name</b>          | Isotope-ratio Mass Spectrometer   |
| <b>Dataset-specific Description</b>     | Thermo Delta Plus XP isotope ratio mass spectrometer is specially designed for measurement of light environmental stable isotopes (2H, 13C, 15N, 18O, 34S) and is a sensitive and selective instrument with applications in hydrology, geology, environmental protection, paleoclimate. |
| <b>Generic Instrument Description</b>   | 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).  |

|   |   |
|---|---|
| <b>Dataset-specific Instrument Name</b> | standard 24-bottle Niskin rosette sampler   |
| <b>Generic Instrument Name</b>          | Niskin bottle   |
| <b>Dataset-specific Description</b>     | standard 24-bottle Niskin rosette sampler   |
| <b>Generic Instrument Description</b>   | A Niskin bottle (a next generation water sampler based on the Nansen bottle) is a cylindrical, non-metallic water collection device with stoppers at both ends. The bottles can be attached individually on a hydrowire or deployed in 12, 24, or 36 bottle Rosette systems mounted on a frame and combined with a CTD. Niskin bottles are used to collect discrete water samples for a range of measurements including pigments, nutrients, plankton, etc. |

[ [table of contents](#) | [back to top](#) ]

---

## Deployments

### AT15-61

|                    |  |
|--------------------|--|
| <b>Website</b>     | <a href="https://www.bco-dmo.org/deployment/58785">https://www.bco-dmo.org/deployment/58785</a>                                    |
| <b>Platform</b>    | R/V Atlantis   |
| <b>Start Date</b>  | 2010-01-29   |
| <b>End Date</b>    | 2010-03-03   |
| <b>Description</b> | See more information at R2R: <a href="https://www.rvdata.us/search/cruise/AT15-61">https://www.rvdata.us/search/cruise/AT15-61</a> |

### MV1104

|                    |  |
|--------------------|--|
| <b>Website</b>     | <a href="https://www.bco-dmo.org/deployment/555585">https://www.bco-dmo.org/deployment/555585</a>                                |
| <b>Platform</b>    | R/V Melville   |
| <b>Start Date</b>  | 2011-03-23   |
| <b>End Date</b>    | 2011-04-23   |
| <b>Description</b> | See more information at R2R: <a href="https://www.rvdata.us/search/cruise/MV1104">https://www.rvdata.us/search/cruise/MV1104</a> |

[ [table of contents](#) | [back to top](#) ]

---

## Project Information

### Expression of Microbial Nitrification in the Stable Isotopic Systematics of Oceanic Nitrite and Nitrate (Microbial Nitrification)

**Coverage:** Eastern Tropical South Pacific

*Description from NSF award abstract:*

Closing the marine budgets of nitrate and nitrous oxide are central goals for researchers interested in nutrient-driven changes in primary productivity and climate change. With the implementation of new methods for oxygen isotopic analysis of seawater nitrate, it will be possible to construct a budget for nitrate based on its oxygen isotopic distribution that is complementary to nitrogen isotope budgets. Before we can effectively use

oxygen isotopes in nitrate to inform the current understanding of the marine nitrogen cycle, we must first understand how different processes that produce (nitrification) and consume (assimilation, denitrification) nitrate affect its oxygen isotopic signature.

In this study, researchers at the Woods Hole Oceanographic Institution will provide a quantitative assessment of the oxygen isotopic systematics of nitrification in the field and thus fill a key gap in our understanding of  $^{18}\text{O}$  variations in nitrate, nitrite, and nitrous oxide. The primary goal is to develop a quantitative prediction of the oxygen isotopic signatures of nitrite and nitrate produced during nitrification in the sea. The researchers hypothesize that oxygen isotopic fractionation during nitrification is the primary factor setting the  $^{18}\text{O}$  values of newly produced nitrate and nitrite. Secondly, they hypothesize that oxygen atom exchange is low where ammonia oxidation and nitrite oxidation are tightly coupled, but may increase in regions with nitrite accumulation, such as in the primary and secondary nitrite maxima. They will test these hypotheses with a series of targeted laboratory and field experiments, as well as with measurements of nitrite and nitrate isotopic distributions extending through the euphotic zone, primary nitrite maximum, and secondary nitrite maximum of the Eastern Tropical South Pacific. The results of these experiments are expected to provide fundamental information required for the interpretation of  $^{18}\text{O}$  isotopic signatures in nitrite, nitrate, and  $\text{N}_2\text{O}$  in the context of underlying microbial processes. A better understanding of these features and the processes involved is important for quantifying new production, controls on the N budget, and  $\text{N}_2\text{O}$  production in the ocean -- which should lead to a better understanding of the direct and indirect interactions among the nitrogen cycle, marine chemistry, and climate.

[ [table of contents](#) | [back to top](#) ]

---

## Funding

| Funding Source   | Award                       |
|--|-----------------------------|
| <a href="#">NSF Division of Ocean Sciences (NSF OCE)</a> | <a href="#">OCE-0961098</a> |

[ [table of contents](#) | [back to top](#) ]