

Production mechanism and global budget of N₂O inferred from its isotopomers in the western North Pacific

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[1] We measured the intramolecular distribution of ¹⁵N as well as conventional nitrogen and oxygen isotope ratios in oceanic nitrous oxide (N₂O) in the western North Pacific for the first time. In contrast to a nearly homogeneous vertical distribution of a bulk nitrogen isotope ratio, a wide variation in site preference for intramolecular ¹⁵N distribution was found, suggesting the subsurface and deep source mixing and the production mechanism in the ocean. The oceanic composition of N₂O isotopomers quantitatively outlines the ocean as the most significant source after the terrestrial one. *INDEX TERMS*: 0312 Atmospheric Composition and Structure: Air/sea constituents fluxes (3339, 4504); 1615 Global Change: Biogeochemical Processes (4805); 4820 Oceanography Biological and Chemical: Gases; 4870 Oceanography Biological and Chemical: Stable isotopes; 9355 Information Related to Geographic Region: Pacific Ocean

1. Introduction

[2] Nitrous oxide is a greenhouse gas subject to the mitigation targets in the Kyoto Protocol. While the major sink of N₂O is photochemical reactions in the stratosphere, most of its recognized sources exist in the earth's surface and include: microbial production in natural and agricultural soils and water, industrial formation as a byproduct from adipic acid production, fossil fuel combustion, and so on [Prather *et al.*, 1994]. Although a number of studies have been conducted using various approaches, there is still great uncertainty about the estimated magnitudes of global sources and sinks.

[3] The isotopic signature of N₂O is an alternative tool for deducing the global N₂O cycle, since it reflects its precursor materials and subsequent physico-chemical processes, and a global budget can be depicted by isotopic mass balance [Yoshida and Matsuo, 1983; Kim and Craig, 1993; Rahn and Wahlen, 2000]. Previous observations in the subarctic, subtropical and tropical Pacific, and the Arabian Sea showed that ¹⁵N and ¹⁸O in N₂O are generally depleted in near surface water and enriched in the deeper water column [Yoshida *et al.*, 1989; Dore *et al.*,

1998; Kim and Craig, 1990; Yoshinari *et al.*, 1997; Naqvi *et al.*, 1998]. However, the origin of excess N₂O in the ocean has not been clearly identified and mass balance analyses using ¹⁵N–¹⁸O relationships are still ambiguous about the role of the ocean.

[4] Yoshida and Toyoda [2000] found that ¹⁵N is more enriched at the center site (to which we refer as the α site) of the asymmetric NNO molecule, than at the end (β) site, and that the variation in composition of tropospheric N₂O isotopomers can be explained by the mixing of sources. Here, we analyzed the isotopomers of N₂O in the subarctic western North Pacific, one of the most productive oceanic regions, to obtain its characteristics and to assess the contribution of the ocean to the atmospheric N₂O.

2. Experiments

[5] Seawater samples were collected at station KNOT (Kyodo North Pacific Ocean Time series, 44°N, 155°E) during several cruises of three research vessels between July 1999 and October 2000 (N. Tsurushima *et al.*, Seasonal variations of carbon dioxide system and nutrients in the surface mixed layer at station KNOT in subarctic western North Pacific, submitted to the *Deep-Sea Research*, 2001). During the cruise of October 2000, we also sampled water at 500 km southeast of sta. KNOT (sta. KN14, 41°N, 159°E), to examine the effect of the subtropical water mass, since sta. KNOT is just north of the subarctic front.

[6] Dissolved N₂O was extracted in a similar manner to dissolved methane analysis [Popp *et al.*, 1995; Tsunogai *et al.*, 1998] and introduced into the preconcentration/gas chromatograph/isotope ratio mass spectrometer system [Yoshida and Toyoda, 2000]. Details of the procedures will be shown elsewhere (H. Yamagishi *et al.*, manuscript in preparation, 2001). The isotopomer ratios of ¹⁵R^{bulk}, ¹⁸R, and ¹⁵R^α were determined and ¹⁵R^β was obtained by the relationship of $^{15}R^{bulk} = (^{15}R^{\alpha} + ^{15}R^{\beta})/2$, where $^{15}R^{\alpha} = [^{14}N^{15}N^{16}O]/[^{14}N^{14}N^{16}O]$, $^{15}R^{\beta} = [^{15}N^{14}N^{16}O]/[^{14}N^{14}N^{16}O]$, $^{18}R = [^{14}N^{14}N^{18}O]/[^{14}N^{14}N^{16}O]$ [Toyoda and Yoshida, 1999]. Isotopomer ratios of a sample (R_{sample}) are expressed as per mil deviation from ¹⁵N/¹⁴N and ¹⁸O/¹⁶O ratios of the standard materials (R_{std}), atmospheric N₂ and standard mean ocean water (SMOW), respectively: $\delta X = (R_{sample}/R_{std} - 1) \times 1000$ where X = ¹⁵N^{bulk}, ¹⁵N^α, ¹⁵N^β or ¹⁸O. Typical analytical precision is 0.6‰, 0.9‰, 1.5‰, and 0.9‰ for $\delta^{15}N^{bulk}$, $\delta^{15}N^{\alpha}$, $\delta^{15}N^{\beta}$, and $\delta^{18}O$, respectively.

3. Results and Discussion

[7] We inferred from a small time-variation in the profiles of N₂O concentration and isotopomer ratios that the processes deter-

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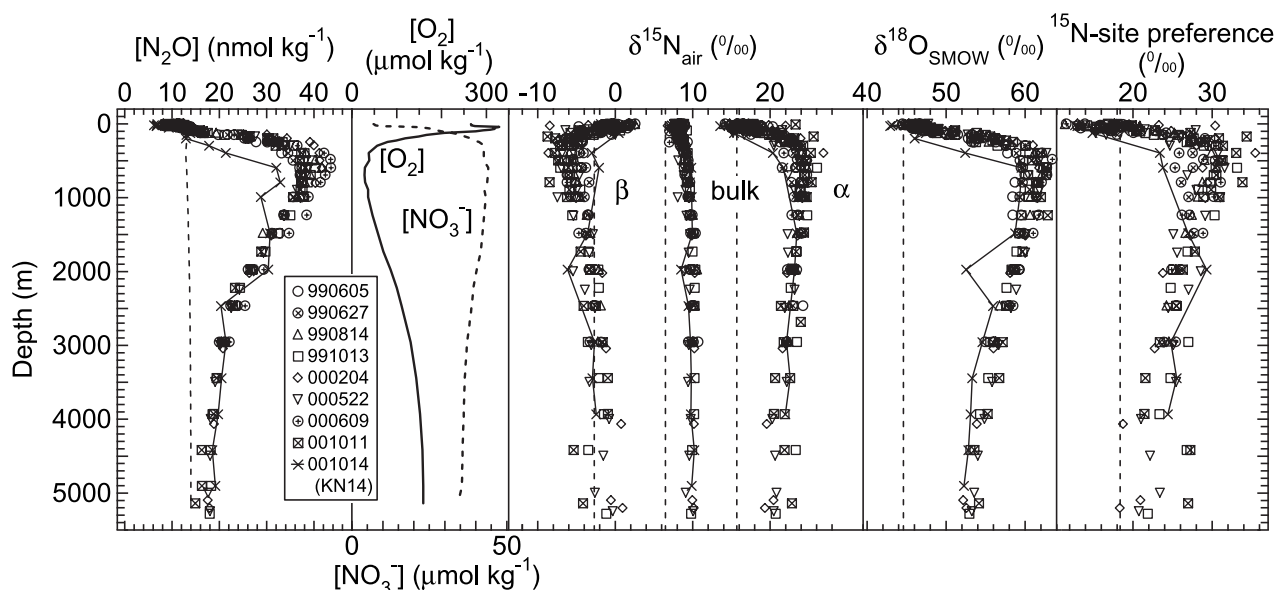


Figure 1. Vertical profiles of the concentration of dissolved N₂O, O₂, and NO₃⁻, isotopomer ratios ($\delta^{15}\text{N}^{\text{bulk}}$, $\delta^{15}\text{N}^{\alpha}$, $\delta^{15}\text{N}^{\beta}$, and $\delta^{18}\text{O}$) and ¹⁵N-site preference ($\delta^{15}\text{N}^{\alpha} - \delta^{15}\text{N}^{\beta}$) of N₂O observed at stations KNOT (8 profiles) and KN14 (one profile). Only typical profiles are shown for [O₂] (solid line) and [NO₃⁻] (dotted line). The legend shows sampling dates with a format of “yyymmdd”. The dotted line with the [N₂O] plot indicates calculated N₂O concentration at 100% air saturation, overlapping with observed value shallower than 100 m. The dotted lines with isotopomer-ratio plots show the respective values in maritime air.

mining them are relatively steady (Figure 1). The N₂O concentration profile is similar to that previously observed at a nearby station [Yoshida *et al.*, 1989] and has an inverse relationship with dissolved oxygen. Bulk nitrogen and oxygen isotope ratios of N₂O are about 1‰ higher in the surface water than in the atmosphere, which agrees with the equilibrium isotope fractionation [Inoue and Mook, 1994]. Compared with the observations in the subtropical North Pacific [Dore *et al.*, 1998], the profiles of $\delta^{15}\text{N}^{\text{bulk}}$ and $\delta^{18}\text{O}$ show no subsurface minimum, and their maximum appear at shallower depth with $\delta^{15}\text{N}^{\text{bulk}}$ exhibiting only a slight increase with depth. In contrast, isotopomer ratios $\delta^{15}\text{N}^{\alpha}$ and $\delta^{15}\text{N}^{\beta}$ show characteristic depth profiles. The $\delta^{15}\text{N}^{\alpha}$ increases with depth between the surface and about 500 m and gradually decreases in deeper regions, whereas the profile of $\delta^{15}\text{N}^{\beta}$ is almost a mirror image of that of $\delta^{15}\text{N}^{\alpha}$. The most striking features are that the site preference for ¹⁵N, expressed as $\delta^{15}\text{N}^{\alpha} - \delta^{15}\text{N}^{\beta}$, shows a quite similar profile to the $\delta^{18}\text{O}$ and N₂O concentration profiles, and their respective maxima coincides.

[8] The isotopic profile of oceanic N₂O is determined by three factors: isotopic composition of the substrates in production processes, kinetic isotope fractionation during production or consumption of N₂O, and mixing of N₂O from several sources. The last two factors are discussed here since we have not measured isotopic composition of the substrates.

[9] The kinetic isotope effect has often been applied to characterize biologically produced and consumed N₂O by low and high isotope ratios, respectively. However, the vertical gradient of $\delta^{15}\text{N}^{\text{bulk}}$ near the N₂O concentration maximum is smaller than that expected from the large isotope fractionation reported for production and consumption of N₂O by nitrifying and denitrifying bacteria [e.g., Yoshida, 1988; Barford *et al.*, 1999]. Even if we consider that the kinetic isotope effect can be masked when substrates are limited, the observed $\delta^{18}\text{O}$ profile is not consistent with that of $\delta^{15}\text{N}^{\text{bulk}}$.

[10] These contradictory results for the bulk isotope ratios can be resolved when we look at the ¹⁵N-site preference and consider the intermediates in the production or consumption processes. If we exclude the possibility that different chemical species are

combined to produce N₂O, and note that the recognized precursors of N₂O contain only one nitrogen atom in their molecules (e.g., NH₂OH, NO, NO₂⁻), the site preference is seen to be independent of the $\delta^{15}\text{N}$ value of the precursors and is determined by the reaction step where the chemical equivalence of two nitrogen atoms is lost. The most probable such step is an N-O bond breakage of the intermediate, hyponitrite [⁻ONNO⁻], which is considered to exist as a free species or a complex with an enzyme [e.g., Weeg-Aeressens *et al.*, 1988]. We propose that this reaction be a rate-determining step and that the preferred dissociation of the ¹⁴N-O bond in [⁻O¹⁴N¹⁵NO⁻] results in the observed enrichment and depletion in ¹⁵N at the α and β sites, respectively. An experimental result, that the ¹⁵N-site preference of N₂O formed by inorganic reaction is about 30‰ for both oxidation of NH₂OH and reduction of NO₂⁻ (S. Toyoda, unpublished data, 2001), supports this mechanism. If there exist equilibrium steps in the microbial formation of N₂O, a maximum site preference of 46‰ at 25°C can be estimated from a theoretical calculation for equilibrium fractionation between the isotopomers [Richet, 1976]. If ¹⁵N-containing isotopomers are fractionated during the reaction of an intermediate, an observed enrichment in ¹⁸O can be explained by a concomitant isotope effect in the same reaction step. In summary, we suggest that N₂O near the concentration maximum is mainly produced by nitrification under substrate (NH₄⁺)-limited conditions, and that the formation of N₂O from a hyponitrite-like intermediate is probably the rate-determining step.

[11] Mixing effect can be separated into horizontal and vertical one. Except for $\delta^{15}\text{N}^{\text{bulk}}$, which shows only a small fluctuation, the variation in concentration and isotopomer ratios near the N₂O maximum suggests horizontal mixing of subtropical water from a comparison of the profiles at stations KNOT and KN14 (Figure 1). The vertical mixing between the sea surface and N₂O maximum is examined in Figure 2. Near-surface water down to 100 m below the surface is well mixed and in equilibrium with the atmosphere, giving almost constant isotopomer ratios. By contrast, between 100 and 200 m (the region between the two vertical broken lines in Figure 2), the isotopomer ratios show a linear correlation with the inverse concentration, which indicates mixing of two end-members,

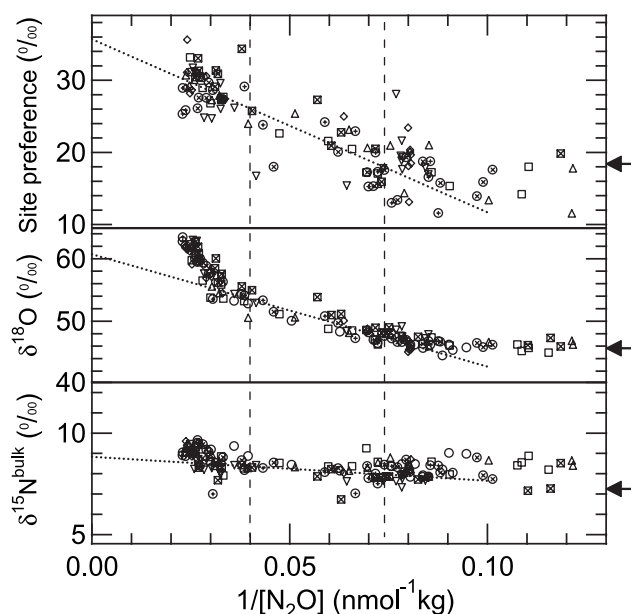


Figure 2. Analysis of mixing effect on isotopomer ratios and ^{15}N -site preference in N_2O between the surface and about 500 m. Each symbol corresponds to that of Figure 1 (data at KN14 are excluded). The dotted lines are mixing lines obtained from linear regression of the data between the two vertical broken lines. The arrows on the right axis indicate the isotopomer ratios of dissolved N_2O in equilibrium with atmosphere [Inoue and Mook, 1994], although the site preference is assumed to be constant in the equilibrium.

being atmospheric N_2O and biologically-produced N_2O , whose isotopic signature corresponds to the y -intercept of the dotted line in Figure 2. However, the $\delta^{15}\text{N}^{\text{bulk}}$ and $\delta^{18}\text{O}$ data between 200 m and concentration maximum (500–600 m) appear to be on another mixing line, suggesting two biological end-members exist at 200 m and the N_2O maximum. This is not evident from the site preference data that show a fluctuation larger than that expected from analytical precision. Neither the difference in processes occurring in the two sources nor the factors causing the variation in the site preference can be determined in the present study. Assuming that the N_2O emitted from the shallower source is transported to the surface by simple diffusion and contributes to the net sea-to-air flux of this gas, we obtain isotopomeric signatures of $\delta^{15}\text{N}^{\text{bulk}} = 8.8\text{‰}$, $\delta^{18}\text{O} = 60.7\text{‰}$, and a site preference = 35.7‰ for the source, and estimate the vertical net flux to be about $1.6 \pm 0.4 \mu\text{mol m}^{-2} \text{d}^{-1}$ from the one-dimensional vertical mixing coefficient [Li *et al.*, 1984] and the measured gradients of N_2O concentration. The flux is comparable to that estimated in the subtropical North Pacific [Dore *et al.*, 1998], but its $\delta^{15}\text{N}^{\text{bulk}}$ and $\delta^{18}\text{O}$ are higher than those reported in other regions of the ocean by 3–6‰ and 10–20‰, respectively [Dore *et al.*, 1998; Kim and Craig, 1990; Naqvi *et al.*, 1998; Yoshinari *et al.*, 1997]. As for the site preference, a slightly different vertical profile has been observed at Station ALOHA in typical subtropical oligotrophic region (B. Popp *et al.*, submitted to the *Global Biogeochemical Cycles*, 2001). Hence, further studies are needed to determine the global isotopomeric flux of oceanic N_2O .

[12] Figure 3 shows the N_2O isotopomers' signatures in the ocean and those of other reservoirs, sources, and sinks reported so far, including present results. In a conventional $\delta^{15}\text{N}^{\text{bulk}} - \delta^{18}\text{O}$ plot (Figure 3a), the oceanic end-member estimated in this study is close to the “isotopically enriched oceanic source” proposed by Rahn and Wahlen [2000], which can be balanced with isotopically depleted terrestrial source suggested by Pérez *et al.* [2000].

[13] In contrast, the site preference– $\delta^{15}\text{N}^{\beta}$ plot (Figure 3b) gives us information on the contribution of the ocean to the atmospheric N_2O independently of oxygen isotope ratios. The distribution of oceanic and tropospheric N_2O has a similar trend, but the range of the former is about twice that of the latter, indicating an effect of the oceanic source on tropospheric isotopomer ratios of N_2O . If we assume that the average of the isotopomer ratios of N_2O from

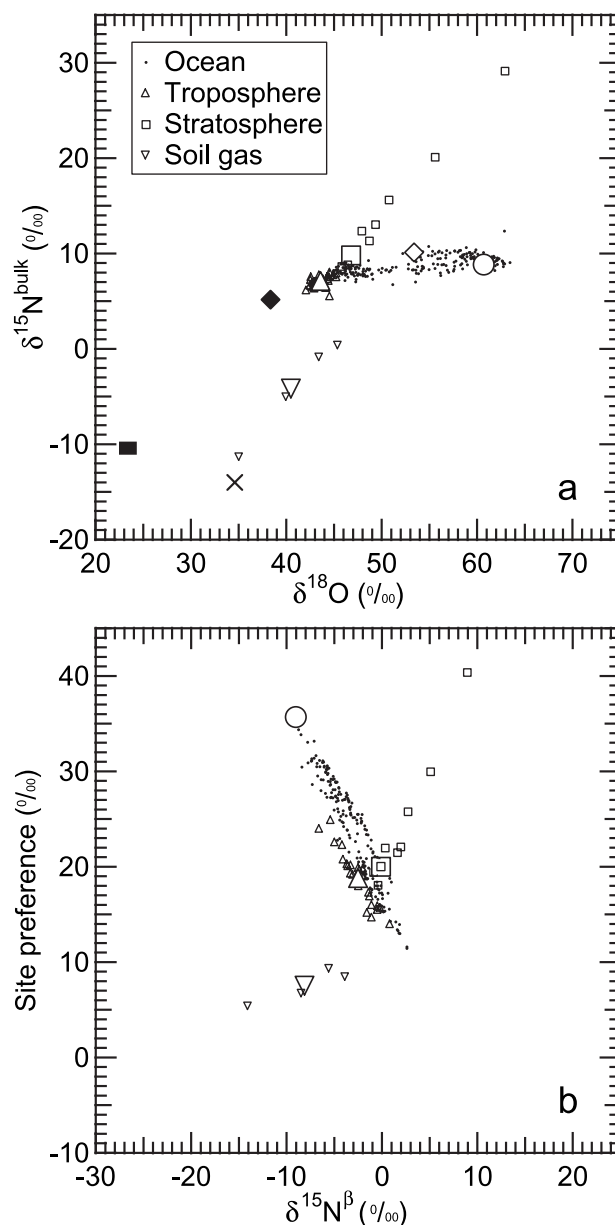


Figure 3. Isotopomeric composition of N_2O observed in the ocean (black dots, this work), troposphere (open triangles) [Yoshida and Toyoda, 2000], stratosphere (open squares) [Toyoda *et al.*, 2001], and that emitted from fertilized grassland (open inverted triangles) [Yamulki *et al.*, 2001], plotted in (a) $\delta^{15}\text{N}^{\text{bulk}}$ vs. $\delta^{18}\text{O}$ and (b) site preference vs. $\delta^{15}\text{N}^{\beta}$ diagrams. The larger open circle, triangle, square, and inverted triangle indicate the estimated oceanic source, the tropospheric average, the stratospheric weighted average, and the average for grassland data, respectively. Open and filled diamonds show estimated oceanic sources for two scenarios [Rahn and Wahlen, 2000]. The filled square and cross indicate soil-emitted N_2O reported by Pérez *et al.* [2000] (flux-weighted average) and Kim and Craig [1990], respectively.

fertilized grassland [Yamulki *et al.*, 2001] equals the flux-weighted average of those from terrestrial and industrial sources as a first approximation, the relationship between the estimated isotopomer ratios for three end-members and the tropospheric average [Yoshida and Toyoda, 2000] is clearly found in Figure 3b: the oceanic source is characterized by large site preferences, whereas terrestrial and industrial ones are distinguished by little preference, with the stratospheric N₂O [Toyoda *et al.*, 2001], which contributes to the back flux into the troposphere, showing a relatively close isotopomeric signature to that of tropospheric N₂O. Thus, we can calculate the contribution of the oceanic source to the total surface source by applying a simple one-box mass balance model [Yoshida and Toyoda, 2000] to those estimated values for end-members. The fraction of ocean flux was calculated independently by three methods using $\delta^{15}\text{N}^{\text{bulk}}$, $\delta^{18}\text{O}$, and the ¹⁵N-site preference, giving respectively, values of 0.23 (-0.29 – +0.45), -0.30 (-0.67 – +0.06), and 0.27 (0.22 – 0.31), with the ranges in the brackets corresponding to the 1 σ uncertainty of the average of the grassland N₂O data. Although the results obtained from $\delta^{15}\text{N}^{\text{bulk}}$ and the site preference agree with each other and are comparable to the relative ocean flux of 0.06–0.31 estimated by Prather *et al.* [1994], the calculation using $\delta^{18}\text{O}$ gives an unlikely value, presumably because the $\delta^{18}\text{O}$ values assigned for the terrestrial and industrial sources or oceanic one were not truly representative of them. Therefore, we can reduce the uncertainty in the oceanic N₂O flux by determining three independent parameters, $\delta^{15}\text{N}^{\text{bulk}}$, $\delta^{18}\text{O}$, and site preference, for the surface sources.

[14] Isotopomers of N₂O in the ocean contain biogeochemical information on its production, consumption and transportation. The ¹⁵N-site preference, in particular, can give us more sensitive information on its origin than a bulk nitrogen isotope ratio. We believe that this parameter will help us to estimate the terrestrial N₂O emission, the most significant source, as well as the production and consumption mechanisms of N₂O after the accumulation of further investigations such as the characterization of various sources, time-series observations of reservoirs, determinations of fractionation of isotopomers in formation/decomposition reactions, and the modeling of global circulation.

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