

The Abundance Ratio of $^{15}\text{N}/^{14}\text{N}$ in Marine Environments*

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Abstract

The ratios of ^{15}N to ^{14}N in the nitrogen bearing substances; dissolved nitrogen gas, ammonia, nitrate, dissolved organic matter and living matters in the western North Pacific Ocean were determined. In the dissolved nitrogen gas, $\delta^{15}\text{N}$ value with respect to the atmospheric nitrogen gas was $+0.9\text{‰}$ which is caused by the isotopic fractionation at the air-sea interface. $\delta^{15}\text{N}$ in nitrate, phytoplankton and sea weeds was $+7\text{‰}$ on an average while δ in zooplankton and fishes were $+10\text{‰}$ and $+15\text{‰}$ respectively.

As to ammonia, $\delta^{15}\text{N}\text{‰}$ in the surface water was -3.5‰ and $+7\text{‰}$ in the deep layer. The above tendency of increasing of $\delta^{15}\text{N}$ values from a simple to complicated substances shows possible effects on the isotopic fractionation along the links in the food chain in the sea.

The change in $\delta^{15}\text{N}$ values among different kinds of matters containing nitrogen in the ocean seems to be mainly due to irreversible changes associated with biochemical reactions and may not be controlled by the isotopic exchange reactions.

The vertical variations of $\delta^{15}\text{N}$ in various forms of nitrogen at different layers in the western North Pacific Ocean were calculated on the basis of the material balance. The mean value of $\delta^{15}\text{N}$ in this sea area was estimated to be $+7\text{‰}$.

Introduction

Hoering (1955) observed the relative amount of nitrogen isotopes in the atmospheric nitrogen and other various nitrogen bearing substances. He found that the range of variation in ^{15}N content was within 13‰ . According to Klots and Benson (1963) $\delta^{15}\text{N}$ in the dissolved nitrogen in distilled water was $+0.85\text{‰}$ at 0°C . Benson and Richard (1961) reported on ^{15}N content in the dissolved nitrogen gas in the open sea water who found that mean $\delta^{15}\text{N}$ value was $+0.13\text{‰}$ with respect to the air. They also found that $\delta^{15}\text{N}$ of the dissolved nitrogen in the anaerobic conditions was about $+0.7$ to $+1.5\text{‰}$ and attributed the increase of ^{15}N to denitrification of nitrate. By using observed values of ^{15}N in the dissolved nitrogen and the nitrogen-argon ratio, they assumed that δ values in organic compounds in the sea would have been from $+20$ to

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+50‰. Using ^{15}N as a tracer, Dugdale *et al.* (1961) found that a few percent of the nitrogen fixation takes place in a lake during summer time.

Hoering (1960) reported that in the nitrogen fixation by several kinds of bacteria the isotopic fractionation was 1.000 ± 0.001 on an average, which suggests that in the biochemical reaction the isotopic fractionation may be small.

In studying the fractionation of nitrogen isotopes in the ocean, it is necessary to know the distribution of nitrogen and its compounds. The results of study on this problem in the western North Pacific will be reported elsewhere. In the present paper results of studies on following subjects will be described.

1. The variation in ^{15}N content in the dissolved gas.
 2. The variation in ^{15}N content in various nitrogen compounds in the sea.
 3. The problem of the isotopic equilibrium of nitrogen in the dissolved nitrogen, nitrite, ammonia and nitrate.
 4. The fractionation factor of nitrogen isotopes among nitrogen compounds.
 6. The material balance of nitrogen isotopes in the sea and the mean δ value of ^{15}N in the marine environment.
 6. The nitrogen isotope ratio in plankton, algae, fishes and sediments.
- Locations of water sampling are shown in Fig. 1.

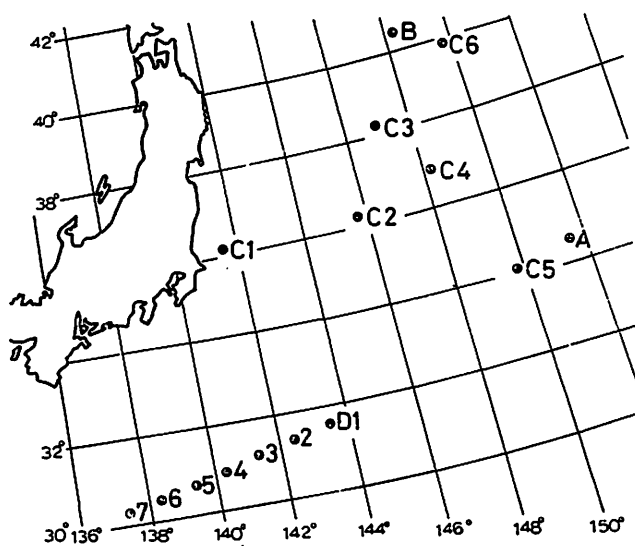


Fig. 1. Locations of sampling.

Preparations of samples for the measurements of nitrogen isotope ratio

The dissolved nitrogen

The dissolved gas in sea water was expelled by CO_2 aboard a ship from which oxygen was removed with a solution of pyrogallol with an apparatus shown in Fig. 2 (Sugawara *et al.*, 1937). To purify the nitrogen gas from the hydrocarbon, CO , CO_2 and O_2 which interfere the mass-spectrometry, an apparatus as shown in Fig. 3 was constructed. A is a quartz tubing in which

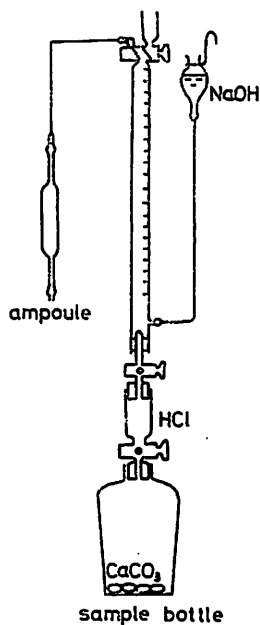


Fig. 2. Apparatus for collecting dissolved gases.

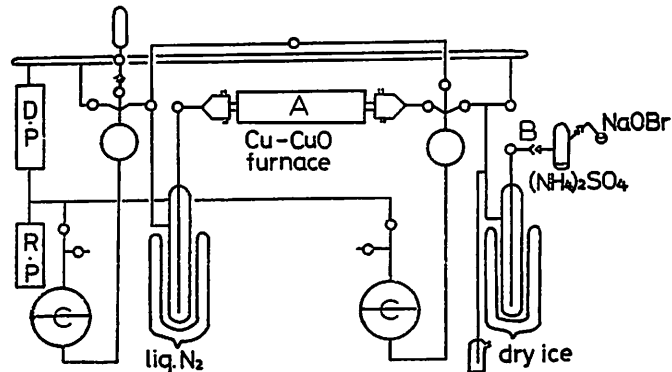


Fig. 3. Apparatus for purification of nitrogen gas.

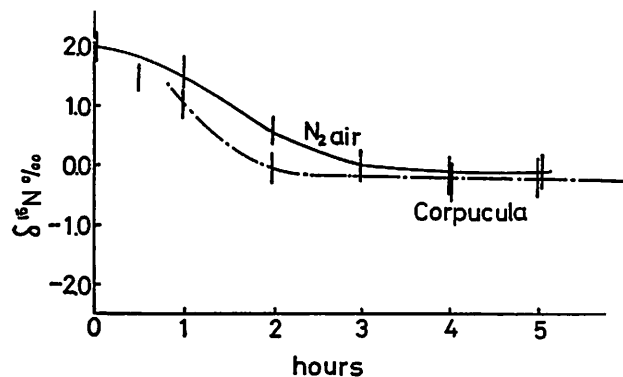


Fig. 4. The variation of $\delta^{15}\text{N} \text{‰}$ with purification time.

Cu and CuO are contained. A sample gas is introduced into the system through the inlet *B* under the pressure of 1.0×10^{-4} mm Hg. A sample gas is circulated through the Cu-CuO furnace at the temperature of 700°C by Toepler pump (C) for 3 to 4 hours to convert the hydrocarbon, CO and O_2 into H_2O and CO_2 which are removed with a liquid nitrogen trap. To examine the purity of the gas, $\delta^{15}\text{N}$ of the atmospheric nitrogen and the nitrogen in *Corpucula japonica* were measured. As seen in Fig. 4, the sufficient purity was obtained in about three hours. Therefore, the purification of sample was continued for 3.5 hours.

Ammonia and nitrate

Sea water was sterilized by adding a small amount of mercuric chloride and stored in a cold room at the temperature -20°C . Water samples were filtered by millipore filters HA type 0.45μ , to remove particulate matters. Ammonia in 100 liter of sea water was concentrated in 20 liter by the consecutive, partial freezing method.

The separation of ammonia was done by the air distillation and it was converted into ammonium sulfate. The trace amounts of ammonia and nitrogen dioxide in the bubbling air were removed by the mixed solution of 0.5 *N* sulfuric acid and *N*-naphthyl-ethylene-diamine-dihydrochloride. After 50–100 ml of 5 *N* NaOH (1:2) solution saturated with K_2CO_3 was added to each portion

(1.5 l) of ammonium sulfate solution, the air was passed in order to distil ammonia which was trapped in 0.4 N sulfuric acid solution of 20 ml. The distillation was continued for about five hours for complete recovery. Then, the ammonium sulfate is oxidized to the nitrogen gas with potassium hypobromite in a vacuum. The nitrogen gas thus obtained was purified in the same way as described above.

Nitrate increases with the depth until about 1,000 m from 0 μg atoms/l to about 40 μg atoms/l. The measurement of nitrogen isotopes in nitrate in sea water of about 20 liters collected in the intermediate and deep layers were done. After concentrated by the partial freezing method, nitrate was further concentrated by evaporation after adding alkali to remove ammonia and the organic matters. The nitrate was reduced completely to ammonia with Devarda alloy. The procedure to convert ammonia to the gaseous nitrogen is the same as above. The recovery was almost 100%.

The organic nitrogen

The decomposition of plankton, algae, fishes and other biological samples was performed by Kjeldahl's method using the mixed powder of HgCl, Se, CuSO₄ and K₂SO₄ as a catalyzer. The ammonia produced was converted to the gaseous nitrogen and was purified as above.

The nitrogen compounds in the sediment

The ammonia and the organic nitrogen are major nitrogen compounds in the sediment. About 500 g of sediment was digested with 3 N HCl solution for several hours under gentle heating. After that the sediment was separated by a centrifuge. The ammonia was first separated from the organic compound by the air distillation. The organic nitrogen remained in the solution was decomposed by Kjeldahl's method.

The mass-spectrometry

The atmospheric nitrogen which was supplied by U. S. National Bureau of Standards was used as the primary standard. The deviation in the relative amount of ¹⁵N in permille from the standard is expressed as follows:

$$\delta^{15}\text{N}\text{‰} = \left\{ \frac{(^{15}\text{N}/^{14}\text{N})_{\text{sample}}}{(^{15}\text{N}/^{14}\text{N})_{\text{std}}} - 1 \right\} \times 1,000. \quad (1)$$

In order to check the fluctuation in readings with time in the mass-spectrometry, two reference standards of ammonium sulfate which differed from each other in ¹⁵N content as much as 7.2‰ were used. The reference standards were calibrated with the primary standard. The nitrogen isotope ratio of the air in Tokyo was also measured which agreed well with the primary standard.

The mass-spectrometer used is RMU-6 manufactured by Hitachi Co. Ltd.,

of the type of 90 degree, single focussing and double collecting. By using Dansgaard's double inlet manifold, a sample and the standard gases are put into the mass-spectrometer through a copper capillary leak of about 50 cm. Following Urey's method, the isotope ratio in a sample is compared with the ratio in the standard at m/e 29 and 28. All measurements were done at the total gain higher than 15V.

There is a possibility of fractionation of isotopes or the contamination during the sample preparation and the measurement. In Winkler's procedure to expel the dissolved nitrogen, about 0.3 ml of the nitrogen gas was introduced from reagents as a blank. The observed value of $\delta^{15}\text{N}$ of the latter nitrogen was $+3.0 \pm 0.5\%$. Therefore, for example, when the volume of the dissolved nitrogen is 14 ml/l and its δ value is $+1.0\%$ the correction factor for the blank will be 0.96. As described above, the air distillation method was used to separate ammonia. The examination for the isotopic fractionation during the distillation must be done. For this purpose, the same reagent of ammonia was distilled five times. The results showed that the fluctuation in $\delta^{15}\text{N}$ was smaller than 0.3%. The isotopic fractionation during the reduction of nitrate to ammonia by Devarda alloy was also examined. For the reduction yields of 73%, 93% and 100%, the relative δ values were respectively 0.0, -0.4 and $+0.3\%$ which were within an experimental error. Therefore, no correction was given to δ values of nitrate.

In the double inlet system, a sample and the standard gas are mixed together. However, this effect is small and no correction was needed. No isotopic fractionation was observed in the concentration method by the partial freezing.

The reproducibility of the isotope measurements was given below:

Name of sample	No. of run	Mean value
Atmospheric nitrogen (N. B. S.)		0.0
Atmospheric nitrogen (laboratory)	10	0.0 ± 0.3
Reference 1. $(\text{NH}_4)_2\text{SO}_4$	15	0.0 ± 0.3
Reference 2. $(\text{NH}_4)_2\text{SO}_4$	20	7.2 ± 0.3

Experimental results

The dissolved nitrogen

Table 1 shows the observed δ values of the dissolved nitrogen in sea water at the stations A, C₅, C₆ and D₇. The values range from -0.9 to $+2.3\%$. The mean δ value is 0.99% which is a little higher than given by Benson and Parker (1961). No correlation of δ values with depth was observed.

Table 1. ^{15}N in the dissolved nitrogen in sea water. The mean ^{15}N , $+0.99\%$.

St. A		St. C ₅		St. C ₆		St. D ₇	
Depth (m)	$\delta^{15}\text{N}\%$	Depth (m)	$\delta^{15}\text{N}\%$	Depth (m)	$\delta^{15}\text{N}\%$	Depth (m)	$\delta^{15}\text{N}\%$
0	0.0	0	-0.1	200	0.9	0	0.4
48	1.2	200	0.8	400	2.3	100	-0.2
96	0.8	1,189	1.5	600	1.8	400	0.2
191	0.3	1,409	1.5	800	1.2	600	1.4
578	1.3	1,633	2.0	970	1.8	800	2.0
772	1.0	1,860	2.2	2,893	1.4	1,000	1.0
1,210	0.7	3,281	1.7			1,250	0.4
1,454	0.8					1,500	0.4

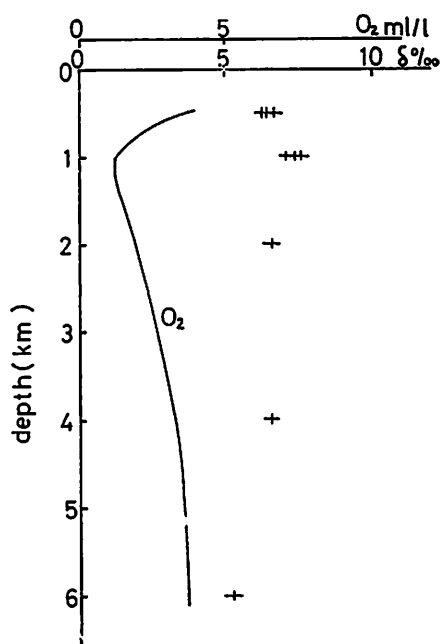
The ammonia and nitrate

Table 2 shows the observed δ values for ammonia and nitrate. It is noticed that the δ values of the surface ammonia was negative, while at 500 meters depth the positive value of about $+7\%$ was observed. Though a number of data is small, the above tendency suggests that there is some difference between the surface and the deeper layers in the process of formation of ammonia.

Observed δ values of nitrate range from $+5$ to $+7.5\%$. In Fig. 5, the vertical variation of δ values in nitrate at D-2 is shown.

Table 2. ^{15}N in ammonia and nitrate in sea water

NH_4^+-N	$\delta^{15}\text{N}\%$
0 m (D-2)	-3.5
500 m (D-2)	6.5
500 m (A)	7.5
NO_3^--N	
500 m (A)	6.3
1,000 m (B)	7.5
500 m (D-2)	6.6
1,000 m (D-2)	7.5
2,000 m (D-2)	6.5
4,000 m (D-2)	6.5
6,000 m (D-2)	5.1
500 m (D-4)	6.3
1,000 m (D-4)	7.0

**Fig. 5.** Vertical variations of δ values in nitrate and dissolved oxygen in the sea (D-2).

The marine plankton

Needless to say, plankton is playing the most important role in the nitrogen cycle in the sea. In Table 3, δ values in plankton are given. It is interesting that zooplankton showed higher δ values than phytoplankton. Among the plankton samples, D_{7-1} and D_{7-2} were collected at D_7 stations at the time interval of several hours. Main species of plankton collected at D_{7-1} are *diatomeae* and *copepoda* and at D_{7-2} , *copepoda*, *diatomeae* and *dinoflagellata*. For this reason, there is a difference in δ value between two samples.

Table 3. ^{15}N in marine plankton

Phytoplankton	$\delta^{15}\text{N}\%$
D-7-1	6.5
D-7-2	3.4
D-16 (33°20'N, 136°32'E)	7.2
X-1	5.3
Zooplankton	
C-2	12.8
X-2 (composite)	9.6

X-1 is a sample which was collected in the Japan Sea consisting mainly of *skeletonema castatum*. To know the change in the nitrogen isotope ratios during the decomposition process of phytoplankton, X-1 which contained a small amount of sea water was divided into two fractions. One was the particulate matter and another was the solution. The particulate matter was filtered with 0.45μ HA millipore filter. It was found that ^{15}N was enriched in the samples in the following order; dissolved < total \setminus particulate. X-2 was a composite sample collected in the frontal area between Oyashio and Kuroshio currents and in the Japan Sea.

Algae

Various kinds of algae were collected at Shimoda, Izu Peninsula, the west of Tokyo, and at the sea shore near Sakai in Tottori Prefecture facing the Japan Sea. The observed δ values in algae are shown in Table 4. Algae were classified into three groups, green, brown and red. Among 15 species, the upper four species in Table 4 belong the green algae, the middle 8 to brown and the last 3 to red. The δ values ranged from 5.2‰ to 9.7‰ with the mean value of 7.5‰. There is no appreciable difference in δ values among green, brown and red algae. Up to now, there is only one available datum for the nitrogen isotope in marine algae given by Hoering (1955). He reported δ value of +8.1‰ in sea weed in Tokyo Bay.

Table 4. ^{15}N in marine algae

	$\delta^{15}\text{N}\%$
<i>Chlorophyceae</i>	
Dictyosphaeria cavernosa	6.5
Codium latum	7.0
Halimeda cuneata	7.6
Boodlea coacta	7.6
<i>Phaeophyceae</i>	
Myelophycus	7.7
Undaria pinnatifida	5.2
Padina arborescens	8.8
Ecklonia cava	8.8
Sargassum ringgoldianum	7.9
Sargassum filicinum	6.7
Sargassum serratifolium	8.3
Culleria cylindrica	5.8
<i>Rhodophyceae</i>	
Condrus ocellatus	9.7
Chondrococcus hornemanni	7.5
Bertholdia japonica	7.2

Fishes

In Table 5 four observed δ values in fishes are presented. In the sample preparation, a special care was paid in decomposing fish bodies completely. The purification of the gas was done for several hours until the constant δ value was reached. As seen in Table 5 fishes show the highest δ values among other nitrogen bearing substances in the sea. In these samples, *Anadara broughtonii* and *Corpucula Japonica* are shell fishes. The δ value of *Corpucula* which was collected at the estuary of a river was the lowest.

Table 5. ^{15}N in marine fishes

	$\delta^{15}\text{N}\%$
Arctoscopus japonicus	20.5
Engraulis japonica	15.6
Anadara broughtonii	17.7
Corpucula japonica	9.9

Marine sediments

Table 6 shows the δ values of nitrogen compounds in the sediments. In the open ocean sediments, δ value of the total nitrogen was about +6.0‰. Samples of sediments were collected at the depths of 1,830 m and 1,910 m respectively at the station D_4 and D_5 in the ocean. Nitrogen compounds in

Table 6. ^{15}N in marine sediment

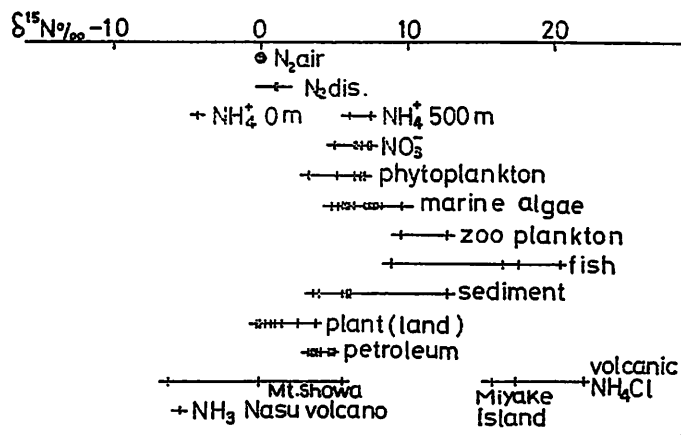
	$\delta^{15}\text{N}\text{‰}$
Pacific Ocean	
D-4 (1,830 m)	
Total	5.8
Org -N	6.0
$\text{NH}_4^+\text{-N}$	5.3
D-5 (1,910 m)	
Total	6.1
Kagoshima Bay	
Total	5.3
Lake Hamana (Brackish)	
Total	3.7
Japan Sea	
Off Niigata	
Total	3.5
Org -N	4.7
$\text{NH}_4^+\text{-N}$	2.9
Yamato Rise	
Total	13.4

some sediments were separated into two components, ammonia and organic matter. In sample D₄, δ value in the organic nitrogen was +6.0‰ and in ammonia, +5.3‰. In a sample which was collected in the Japan Sea off Niigata City, δ value in the organic nitrogen was +4.7‰, and in ammonia 2.9‰.

From the above results, in marine sediments the organic nitrogen seems to have higher δ values than ammonia. A wider range in δ values extending from 4 to 13‰ may be due to the decomposition of organic materials which are derived various kinds of remains of living matters.

A comparison of $\delta^{15}\text{N}$ values of marine samples and land samples

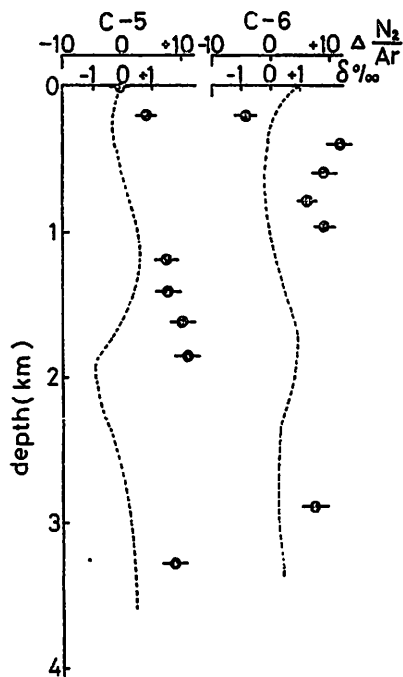
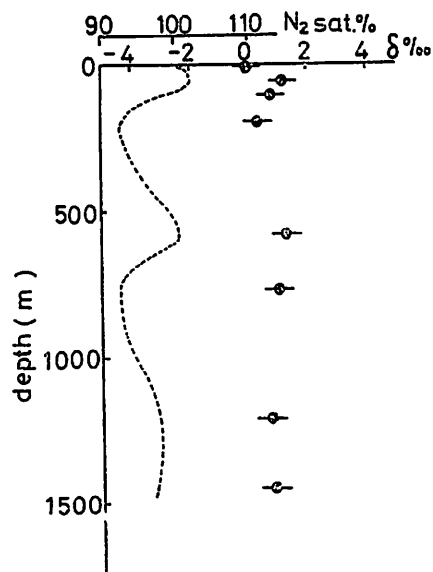
In Fig. 6, δ values in marine and some land materials are compared. In general, ^{15}N seems to be more enriched in marine samples than land samples. Among marine samples, ^{15}N is enriched in substances in the following order; inorganic nitrogen, phytoplankton, algae, zooplankton and fishes. This order suggests the presence of some correlations between isotopic fractionation and links in the food chain in the marine environment.

Fig. 6. ^{15}N abundance in nature.

Discussion

^{15}N in the dissolved nitrogen

There are two possible factors controlling δ values of the dissolved nitrogen in sea water. One is the biological denitrification and the other is the effect of the air-water exchange. In Fig. 7 the vertical variations of the nitrogen-argon ratios and δ values of the dissolved nitrogen at the station C₅ and C₆ are shown. In Fig. 8 the saturation percentage of the dissolved N_2 and the δ values at the station A are shown.

Fig. 7. N_2/Ar and $\delta^{15}\text{N}$ ‰ in dissolved nitrogen.Fig. 8. ^{15}N in dissolved nitrogen and degree of saturation (%) of N_2 dis.

The equilibrium ratio of nitrogen to argon was given by Benson and Parker (1961). Based on the equilibrium ratio, the observed deviation from the equilibrium was calculated as follows:

$$\Delta(\text{N}_2/\text{Ar}) = (\text{N}_2/\text{Ar})_{\text{ob.}} / (\text{N}_2/\text{Ar})_{\text{eq.}} \times 100. \quad (2)$$

The results of calculation shows that there is no correlation between δ values and $\Delta(\text{N}_2/\text{Ar})$ which suggests smaller biological effect on the isotopic fractionation.

On the other hand, as shown in Fig. 9, there is a linear relation between δ value and the sea water temperature. δ value reaches closer to that of the air as water temperature elevates. From this result, it may be concluded that a smaller enrichment of ^{15}N in sea water is mainly due to the isotope effect by the dissolution as suggested by Klots and Benson (1963).

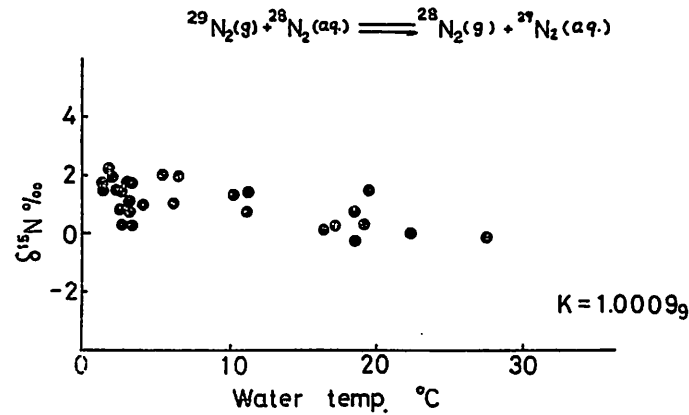
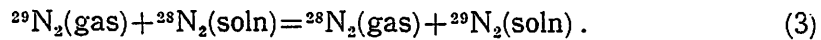


Fig. 9. Fractionation of nitrogen isotope between gas and aqueous solution.

According to them the equilibrium constant (K) in the following exchange reaction is 1.00085 at 0°C .



From δ values obtained by the present authors, the equilibrium constants is estimated to be 1.00099 which corresponds to the enrichment of $+0.99\%$. Richard and Benson (1961) gave a smaller K value of 1.00013 in the dissolved nitrogen in the Atlantic Ocean. The reason for the discrepancy is not known.

Isotope exchange equilibrium

Begun *et al.* (1960) summarized the spectrometric data of the various forms of nitrogen compounds. In Table 7, the partition functions of various forms of nitrogen compounds from 0°C to 25°C are given.

In the following exchange reaction,



Table 7. Partition function ratios of nitrogen compounds at sea water temperature

$$\frac{Q_2}{Q_1} = \frac{\sigma_1}{\sigma_2} \prod_i \frac{U_{2i} \exp(-U_{2i}/2)[1 - \exp(-U_{1i})]}{U_{1i} \exp(-U_{1i}/2)[1 - \exp(-U_{2i})]}$$

$T^\circ\text{K}$ \diagdown Q_1/Q_2	$\frac{^{15}\text{NO}_3^-}{^{14}\text{NO}_3^-}$	$\frac{^{15}\text{NH}_4^+}{^{14}\text{NH}_4^+}$	$\frac{^{15}\text{NO}_2^-}{^{14}\text{NO}_2^-}$	$\frac{^{14}\text{N}^{15}\text{N}}{^{14}\text{N}^{14}\text{N}}$
273.16	1.1727	1.1184	1.0915	1.0886
278.16	1.1677	1.1154	1.0891	1.0866
283.16	1.1633	1.1129	1.0868	1.0847
288.16	1.1589	1.1103	1.0846	1.0829
293.16	1.1546	1.1078	1.0827	1.0811
298.16	1.1059	1.1059	1.0803	1.0794

results of calculation of α show that the heavier isotope is enriched in the nitrogen bearing substances in the following order; molecular nitrogen, ammonia and nitrate.

In Table 8, theoretical values of isotopic fractionation between NH_4^+ and NO_3^- under the assumption that the equilibrium in the isotopic exchange is established are shown. As shown in Table 8 the observed fractionations are very much smaller than the calculated values. This suggests that isotopic fractionation is controlled by some other processes than the isotopic exchange reactions.

Table 8. Comparison between observed and calculated isotopic fractionation factor

$$^{15}\text{NH}_4^+ + ^{14}\text{NO}_3^- = ^{14}\text{NH}_4^+ + ^{15}\text{NO}_3^-$$

$$\alpha_{\text{calc.}} = [Q_2/Q_1]_A / [Q_2/Q_1]_B, \quad \alpha_{\text{obs.}} = \frac{1 + \frac{\delta_A}{1,000}}{1 + \frac{\delta_B}{1,000}}$$

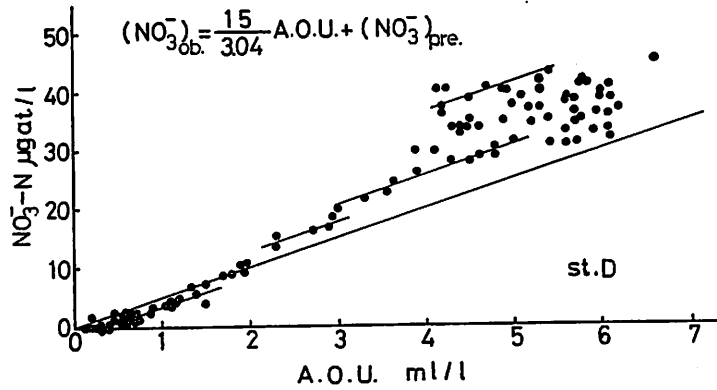
Station	Depth m	Temp. $^\circ\text{C}$	$\alpha_{\text{calc.}}$	$\alpha_{\text{obs.}}$
A	500	11.3	1.044 ₉	0.999
D	500	12.3	1.044 ₆	1.000 ₁

Q ; partition function

The isotope fractionation along the nitrogen cycle

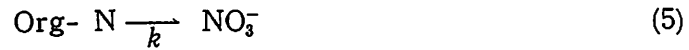
As mentioned above, the isotopic fractionation in the nitrogen cycle in the ocean seems to be not controlled by reversible reactions. The prevailing reactions are supposed to be some irreversible changes associated with biochemical reactions.

In the ocean, the oxidation of organic compounds takes place with formation of nitrate. In the way, ammonia is also formed as an intermediate. As shown in Fig. 10, the linear relation between the concentration of nitrate and the apparent oxygen utilization, indicates that the most part of nitrate in the


 Fig. 10. Relation between nitrate-N and A. O. U. at station D_{1-7} .

deep layer is formed through the oxidation (Sugiura *et al.*, 1964). The variation of $\delta^{15}\text{N}$ values in nitrate may probably be determined by the isotope effect in the oxidation process.

When we consider the oxidation process which may be regarded as the first order reaction as proposed by Grill (1964) in a closed system, the following equations can be obtained



where N and *N are respectively, the light and heavy isotopes of nitrogen, k and k^* are velocity constants of oxidation reactions of organic nitrogen to nitrate. Assuming that there is no nitrate and only the organic nitrogen is present at the beginning and in time 0 to t nitrate is formed by the reaction (5) and (6). The change with time of nitrate concentration at $t=t$ can be expressed as follows.

$$d(\text{NO}_3^-)/dt = k(\text{Org-N}_0 - \text{NO}_3^- - \text{N}_t) \quad (7)$$

$$d(^*\text{NO}_3^-)/dt = k^*(\text{Org-}^*\text{N}_0 - \text{NO}_3^- - ^*\text{N}_t) \quad (8)$$

From the above equations, we can derive the following relations.

$$k/k^* = \ln(1-f)/\ln(1-rf) \quad (9)$$

$$r = (1 - F^{k^*/k})(1 - F) \quad (10)$$

where,

$$F = \text{Org-N}/\text{Org-N}_0$$

$$f = \text{NO}_3^-/\text{Org-N}_0$$

$$f + F = 1$$

$$r = (15/14)_{\text{NO}_3^-} / (15/14)_{\text{Org-N}_0} = 1 + (\delta_{\text{NO}_3^- - \text{N}_t} - \delta_{\text{Org-N}_0}) / 1,000.$$

It has been observed that the rate constant k is larger than k^* so far as the kinetic isotope effect is concerned. The equation (10) can be rewritten as follows:

$$1 + (\delta_{\text{NO}_3^- \cdot \text{N}_t} - \delta_{\text{Org-N}_t}) / 1,000 = (1 - F^{k^*/k}) / (1 - F) \quad (11)$$

If we assume that $\delta_{\text{Org-N}} = 7.0$ and $\delta_{\text{NO}_3^-} = 6.0$ in the range, $0.5 < f < 0.9$, k/k^* can be estimated by the equation (9) to be in the range of 1.0011 and 1.0001. This result is in accord with the value of 1.000 ± 0.001 given by Hoering (1960) in the reaction of nitrogen fixation by azotobacter.

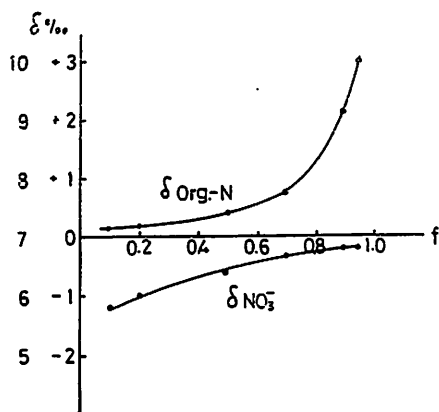
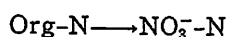


Fig. 11. The variation of δ value in Org-N and in nitrate in the following irreversible reaction under the condition of $k/k^* = 1.001$ in the closed system.



In Fig. 11 and Table 9 the change of δ values in the organic nitrogen and nitrate are given by assuming that the k/k^* is 1.001. In the calculation, the following equations for material balance were used.

$$f\delta_{\text{NO}_3^- \cdot \text{N}_t} + F\delta_{\text{Org-N}_t} = \delta_{\text{Org-N}_t} \quad (12)$$

$$\delta_{\text{Org-N}} - \delta_{\text{Org-N}_t} = f/F(\delta_{\text{Org-N}_t} - \delta_{\text{NO}_3^- \cdot \text{N}_t}) \quad (13)$$

As seen in the figure, it is expected that the δ value of nitrate is -0.2‰ and that of the organic is 2‰ when 90% of the starting organic nitrogen is oxidized to NO_3^- .

Table 9. The variation in δ values in organic nitrogen and nitrate in the following reaction $\text{Org-N} \longrightarrow \text{NO}_3^-$ with $k/k^* = 1.001$ in a closed system.

f	0.1	0.2	0.5	0.7	0.9	0.95
$\delta_{\text{NO}_3^- \cdot \text{N}_t}$	-1.2	-1.0	-0.8	-0.3	-0.23	-0.17
$\delta_{\text{Org-N}_t}$	+0.13	+0.25	0.4	0.1	2.1	3.2

f ; fraction of NO_3^-

On the other hand, in an open system where the δ value of the organic nitrogen in each layer is assumed to be kept constant with time, the relationship between ^{15}N contents of the organic nitrogen and nitrate is determined by following equations.

$$d(\text{NO}_3^-) / dt = k(\text{Org-N}) - c = 0 \quad (14)$$

$$d(^*\text{NO}_3^-) / dt = k^*(\text{Org-N}^*) - c^* = 0 \quad (15)$$

c and c^* , constants.

The equation (15) and (16) give,

$$k/k^* = \frac{\left(\frac{15}{14}\right)_{\text{Org-N}}}{\left(\frac{15}{14}\right)_{\text{NO}_3^- \cdot \text{N}}} = 1 + \frac{\delta_{\text{Org-N}} - \delta_{\text{NO}_3^-}}{1,000} \quad (16)$$

By the equation (17), $k/k^* = 1.001$ gives $\delta_{\text{Org-N}} - \delta_{\text{NO}_3^- \text{-N}} = 1.0$. In the above discussion the variation of δ values in organic materials and nitrate were calculated both in a closed and in an open system. In the sea, the system is complicated due to the various factors such as migration of organic debris, mixing and circulation of water and so on. Therefore, each water mass is considered to be in between two systems, closed and open.

As seen in Table 3, δ values of plankton vary from 3 to 13‰. The population of plankton is different locally in quantity and species. The ecological conditions are also different from place to place. The variation in δ values in nitrate may be partly attributed to the difference in those of the organic matter from which nitrate is formed. The δ value of ammonia in the deep layer which is formed as an intermediate in the oxidation process will be similar to that of nitrate. The negative δ values of NH_4^+ in the surface layer may be a possible result by the kinetic isotope effect through the biological metabolism.

^{14}N passes through bodies of organisms faster than ^{15}N and there is also a trend that ^{15}N is enriched in the compounds with complicated chemical forms.

Geochemical balance of ^{15}N in the sea

It is of interest to estimate δ values of ^{15}N in various chemical forms of nitrogen on the basis of material balance in the ocean. In regard to the nitrogen cycle, the ocean is divided into following three layers.

1. Surface or euphotic layer extending from 0 to 50 m where photosynthesis takes place.
2. The intermediate layer from 50 to 1,000 m where oxidative decomposition of organic matter takes place.
3. The deep layer from 1,000 to 4,000 m in which the nitrate is most abundant among other nitrogen compounds and its concentration is nearly kept constant.

It is also assumed that the oceans are in the stationary state and the concentration of each form of nitrogen compound is nearly constant in each layer.

In the surface the inorganic nitrogen is consumed by the organisms and is transferred into ammonia, nitrite and finally to nitrate again.

In Table 10 the concentration of each form of nitrogen compound in each

Table 10. Concentration of nitrogen compounds at different layers in the sea

Depth (m)	0—50	50—1,000	1,000—4,000
Org -N	10	9	1.8
NH_4^+ -N	3.5	1.3	0.4
NO_2^- -N	0.1	0.05	0.01
NO_3^- -N	0.5	22	36

unit; $\mu\text{g atoms/l}$

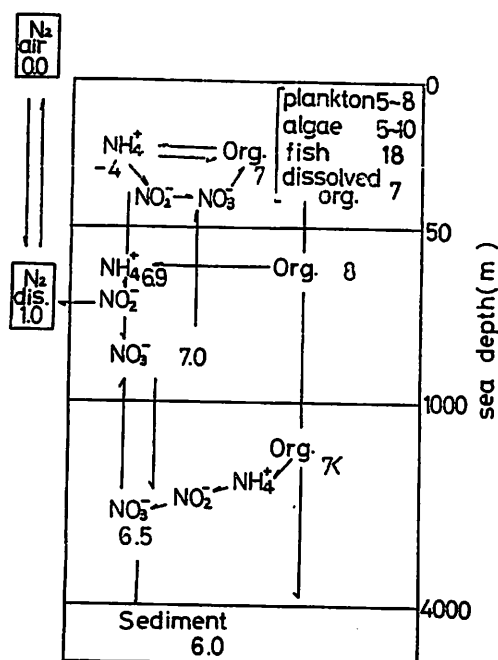


Fig. 12. The variation of $\delta^{15}\text{N}$ ‰ in nitrogen cycle.

2. The intermediate layer

The mean of δ values of the organic nitrogen, ammonia and oxidative nitrate in this layer is $+7.0\text{‰}$. The δ value was often higher near the oxygen minimum layer where it reached up to $+7.5\text{‰}$. However, there was no appreciable difference in δ values among different kinds of nitrogen compounds which suggests the smaller isotopic fractionation takes place in this layer.

3. The deep layer

The δ value of nitrate in this layer is a little lower than that of the intermediate. When $\delta_{\text{NO}_3^-}$ and δ_{mean} are $+6.8$ and $+7.0\text{‰}$ respectively, the δ value of organic forms of nitrogen will be in the range $+7\text{‰}$ to $+11\text{‰}$. The smaller δ value in nitrate causes the higher δ value in the organic nitrogen because of the higher nitrate content in this layer.

4. The marine sediments

There are many factors controlling δ values in sediments. For example δ value of organic debris, the conversion of the chemical forms of the organic materials in marine sediment and so forth. It is found that the organic nitrogen shows higher δ value than that of ammonia in the sediment.

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layer is given.

For the first approximation, the mean value of $\delta^{15}\text{N}$ is assumed to be 7.0‰ throughout the ocean. In Fig. 12 the δ values of various nitrogen compounds in different layers which are calculated on the basis of the material balance are shown.

1. The surface layer

In this layer ammonia and the organic nitrogen are predominant. The observed δ value of ammonia was -3.5‰ which is the minimum value for $\delta^{15}\text{N}$ in the ocean. The sampling was done in the late spring when the biological activity was high. Assuming that δ of ammonia is -3.5‰ , δ value of the organic nitrogen is estimated to be $+10.7\text{‰}$.

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