^{15}N ABUNDANCE IN THE DRY VALLEY AREA, SOUTH VICTORIA LAND, ANTARCTICA: ECO-PHYSIOLOGICAL IMPLICATIONS OF MICROORGANISMS

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Abstract: Stable nitrogen isotope ratio in nitrogen-bearing substances from the Dry Valley area, south Victoria Land, was reported. The lowest $\delta^{15}N$ value of around -50‰ was found for the epibenthic algae collected from nitrate-rich saline ponds in the Labyrinth. In Lake Vanda, nitrate in its maximum layer (57–59 m) was significantly rich in ^{15}N (10.3–13.4‰) as compared with the sedimentary organic nitrogen (-4.6‰). The variation of $\delta^{15}N$ seemed to probably result from nitrogen isotope fractionation associated with nitrate assimilation by algae.

In the east lobe of Lake Bonney, the $\delta^{15}N$ value in nitrate increased with depth from 4.9 to 31.2‰. Dissolved N_2 gas in the west lobe exhibited the highest $\delta^{15}N$ value (1.5–2.5‰) among those observed in anoxic layers of various aquatic systems. The latter results from denitrification of nitrate with high $\delta^{15}N$ value at temperatures lower than 0°C.

1. Introduction

It is widely recognized that stable isotope ratio of nitrogen is one of the most promising indicators to trace a matter cycling in natural ecosystems as well as to study ecophysiology of microorganisms concerned. KAPLAN and his co-workers have employed elementary compositions and isotope ratios of C, N, H and S for tracing the source of sedimentary organic matter (PETERS *et al.*, 1978; SWEENEY *et al.*, 1978). The nitrogen isotope ratio was emphasized to be a likely parameter for dietary analysis and for determining a trophic level of animals in natural environments (MINAGAWA and WADA, 1984). WADA and HATTORI (1976) have found that the $\delta^{15}N$ values of marine phytoplankton are closely correlated with the form of nitrogenous compounds used for their growth. The nitrogen isotope fractionation factor is inversely correlated with a growth rate constant in the process of nitrate assimilation by a marine diatom (WADA and HATTORI, 1978).

The Dry Valley area in south Victoria Land is the largest polar desert containing numerous saline lakes and ponds. The presence of a large amount of nitrogenous compounds together with epibenthic algae in these aquatic systems and in their surrounding areas is of particular interest from the nitrogen isotopic point of view. Considerable attention has been focused on the distributions of nutrient salts in several saline lakes such as Lakes Vanda, Bonney and Fryxell. An individual lake manifests its own distribution pattern of nitrogenous compounds in a well stratified water colunm together with discrete bands of microbial communities (TORII *et al.*, 1975; VINCENT *et al.*, 1981). The evaporites and soils around the lakes also contain a large amount of nitrate involving sodium niter (JOHANNESSON and GIBSON, 1962; MORIKAWA *et al.*, 1974).

WADA *et al.* (1981) first reported the nitrogen isotope ratio in soil nitrate and epibenthic algae from the McMurdo Sound region. According to their data, nitrate in Antarctic soil is extremely depleted in ^{15}N as compared with biogenic nitrogen in the temperate zone, and algae collected from a nitrate-rich saline pond and from a penguin rookery exhibit, respectively, the lowest and the highest δ^{15}N values among terrestrial biogenic nitrogen. In this paper, the nitrogen isotope ratio in epibenthic algae, and nitrate and dissolved N_2 in the Dry Valley area, south Victoria Land, Antarctica are reported. Possible factors controlling the variation of δ^{15}N in the Antarctic aquatic ecosystems are discussed with emphasis on the eco-physiology of microorganisms concerned.

2. Materials and Methods

Samplings were performed in December 1979 and in December 1980. Epibenthic algae were collected from unnamed saline ponds in the Labyrinth, near the terminus of the Wright Upper Glacier. Water samples in Lake Vanda were collected from the nitrate maximum zone at a mid-lake deep water site (maximum depth, 68 m). At the sampling site a hole was drilled through 3 to 4 m permanent lake ice by using a SIPRE ice-auger. Water temperature and electric conductivity were measured at every 50 cm by a thermistor and an electric conductance meter. Water sampling was carried out by using a 1 *l* Kitahara-type polyacrylate sampler. Water sampling was also made at the east and west lobes of Lake Bonney.

Unless otherwise specified, all of the analytical method were those described by TORII *et al.* (1975) and WADA *et al.* (1981) as follows: for dissolved oxygen, Winkler method; for chloride, mercuric thiocyanate and mercuric nitrate method; for ammonium, phenol-hypochlorite method; for nitrite, diazotization method and for nitrate, cadmium reduction method.

Extraction of dissolved gases was carried out in the following way. Lake waters were introduced into 250 m*l* glass bottles with a stopcock, then sterilized with 1 m*l* saturated $HgCl_2$ solution and brought back to Japan. The dissolved gases were extracted *in vacuo* in the laboratory by using a vacuum system with a Toepler pump. Procedures for δ^{15}N assay were described elsewhere (WADA and HATTORI, 1976). The nitrogen-argon ratio and the nitrogen isotope ratio were measured using a Hitachi RMU-6R mass spectrometer fitted with a dual inlet system and a double collector for ratiometry. The nitrogen isotope ratio was expressed in permil deviations from atmospheric nitrogen as defined by the following equation:

$$\delta^{15}\mathrm{N}(‰)=\frac{(^{15}\mathrm{N}/^{14}\mathrm{N})_{\mathrm{sample}}-(^{15}\mathrm{N}/^{14}\mathrm{N})_{\mathrm{air}}}{(^{15}\mathrm{N}/^{14}\mathrm{N})_{\mathrm{sample}}}\times 1000.$$

Table 1. Distribution of $\delta^{15}N$ in various nitrogen-bearing substances from the Dry Valley area in south Victoria Land. WT: Water temperature, SG: Specific gravity at 25°C, ER: Evaporation residue at 180°C (g/kg), unit of inorganic nitrogenous compounds: μg atom N/l.

Sample	Date	$\delta^{15}N$ (‰)	Remarks
Epibenthic algae (unnamed saline pond in the Labyrinth)			
L–7	20 December 1980	−45.2	WT 1.2°C pH 7.0 SG 1.003 ER 6.41 Cl 2.75 g/kg
L–4	20 December 1980	−42.6	WT −1.8°C pH 7.0 SG 1.038 ER 51.9 Cl 18.1 g/kg
L–8	21 December 1980	−44.0	WT 2.0°C pH 6.0 SG 1.021 ER 27.8 Cl 11.2 g/kg
Nitrate in lake waters			
Lake Vanda 52 m	30 November and	10.3	WT 14.8°C Cl 4.94 g/kg NH_4^+ 1.0 NO_2^- 0.39 NO_3^- 84.5
54 m	1 December 1979	10.9	WT 17.3°C Cl 15.9 g/kg NH_4^+ 1.0 NO_2^- 1.03 NO_3^- 179
56 m		13.4	WT 19.7°C Cl 24.6 g/kg NH_4^+ 2.3 NO_2^- 1.63 NO_3^- 209
Lake Bonney, east lobe			
20 m	4 December 1979	4.9	WT 5.3°C Cl 106.7 g/kg NH_4^+ 1.73 NO_2^- 12.2 NO_3^- 670
25 m		25.1	WT 2.3°C Cl 134.8 g/kg NH_4^+ 0.57 NO_2^- 45.5 NO_3^- 260
30 m		32.2	WT −0.5°C Cl 140.4 g/kg NH_4^+ 1.92 NO_2^- 42.7 NO_3^- 240
Don Quixote Pond	5 December 1979	−7.0	Cl 57.94 g/kg NH_4^+ 11.3 NO_2^- 21.5
Reduced form of nitrogen			
Lake Bonney, west lobe			
Bottom sediment	20 December 1980	7.3	under anaerobic condition
Lake Vanda			
Bottom sediment (68.3 m)	17 December 1980	−4.6	under anaerobic condition
Lake Fryxell			
Ammonium (15 m)	22 November 1979	6.2	NH_4^+ 200 NO_2^- 0.02 NO_3^- 0.0

3. Results

Table 1 gives a summary of $\delta^{15}N$ obtained in the present investigation. Epibenthic algae exhibited quite low $\delta^{15}N$ values (-42 to -45‰). The minimum value of -45.2‰ was found for epibenthic algae collected from an unnamed small pond in the Labyrinth near the terminus of the Wright Upper Glacier. On the other hand, nitrate in the lake waters from Lake Vanda and the east lobe of Lake Bonney gave very high $\delta^{15}N$ values between 4.9 and 32.2‰. The $\delta^{15}N$ value in nitrate increased with depth examined so far in both lakes. The highest value was found at the depth immediately beneath the nitrate maximum layer. On the other hand, the $\delta^{15}N$ value for nitrate from the Don Quixote Pond in the North Fork was fairly low (-7.0‰). The pond contained very high concentration of nitrate up to 24.8 mg at. N/*l* (TORII *et al.*, 1975). Reduced forms of nitrogen such as ammonium and sedimentary organic nitrogen sometimes indicate a representative $\delta^{15}N$ value in aquatic ecosystems, because of their large pool size of nitrogen. The $\delta^{15}N$ value of the reduced nitrogen was variable among three lakes so far examined (Table 1).

Lake Bonney consists of the east and west lobes. Chemical variables are quite different from each other (Fig. 1 and Table 2). The east lobe is supersaturated with oxygen in the upper 13 m. It decreases down to 1–2 m*l*/*l* below 20 m, while the bottom

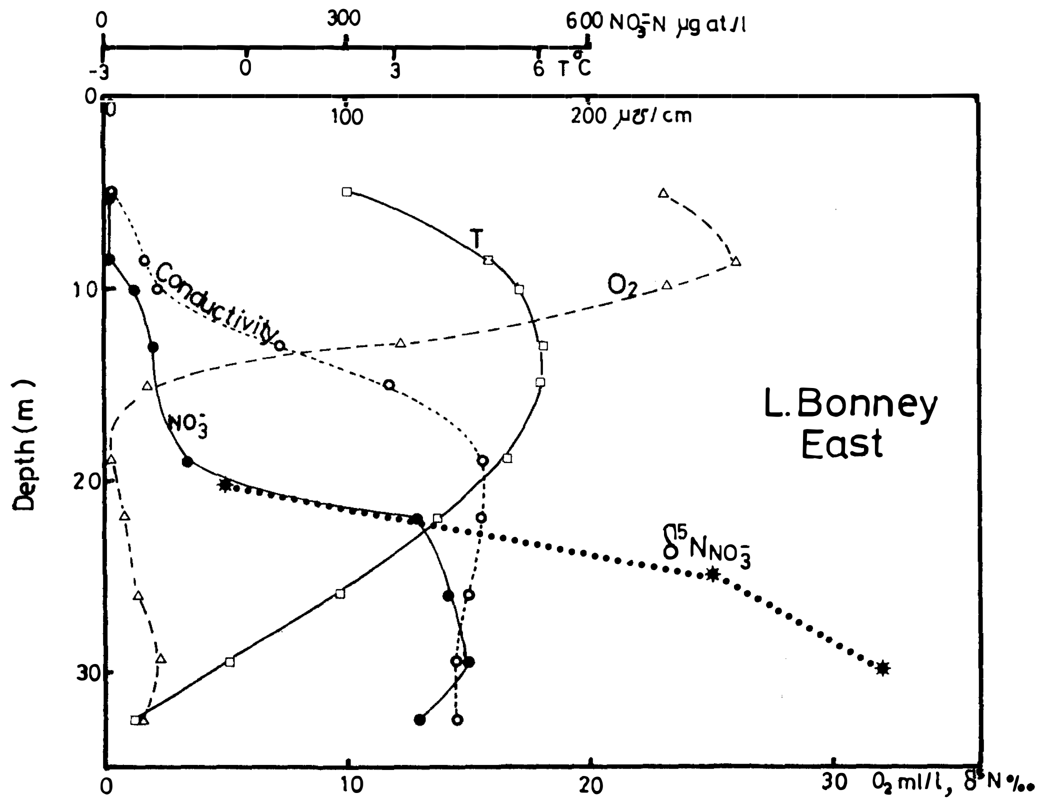

Fig. 1. Distribution of physical and chemical variables (after TORII et al., 1975) and $\delta^{15}N$ in nitrate at a mid-lake deep water site in the east lobe of Lake Bonney on 4 December 1979.

layer of the west lobe became completely anaerobic below 10 m (TORII *et al.*, 1975). Nitrate concentration in the bottom layer (below 20 m) of the east lobe is one order magnitude higher than that in the west lobe. $\delta^{15}N$ for nitrate in the east lobe remarkably increased with depth from 4.9 to 32.2‰ irrespective of the amount of dissolved oxygen.

The $\delta^{15}N$ value for dissolved nitrogen gas in the west lobe of Lake Bonney ranged from 1.5 to 2.5‰; these values were significantly higher than those (0.3–0.8‰) equilibrated with atmospheric nitrogen (0.00‰). Vertical distribution of the nitrogen/argon ratio varied significantly with depth. The low values of 21 to 27 were obtained in the upper layers above 15 m, while the ratio became higher than 35 below 25 m (Table 2).

Table 2. Vertical profiles of the N_2/Ar ratios and $\delta^{15}N$ of dissolved N_2 gas in the west lobe of Lake Bonney. Sampling date: 26 December 1980.

Depth m	Water temperature °C	Cl g/kg	Sp.Gr. (25°C)	pH	NH_4^+	NO_2^-	NO_3^-	N_2/Ar	$\delta^{15}N$ ‰
					μg at. N/l				
5	0.2	0.14	1.001	8.40	7.4	0.34	14.3	25.25	1.9
10	2.6	3.01	1.003	7.46	3.5	0.26	12.1	21.29	2.1
15	0.1	45.40	1.066	6.13	1.1	0.09	12.7	26.49	2.5
20	−2.6	60.09	1.071	5.85	173	0.06	13.1	N.D.*	N.D.*
25	−3.7	72.11	1.094	5.65	57	0.30	11.0	35.66	2.1
30	—	76.25	1.100	5.72	192	0.05	1.7	35.17	1.5

* Not determined.

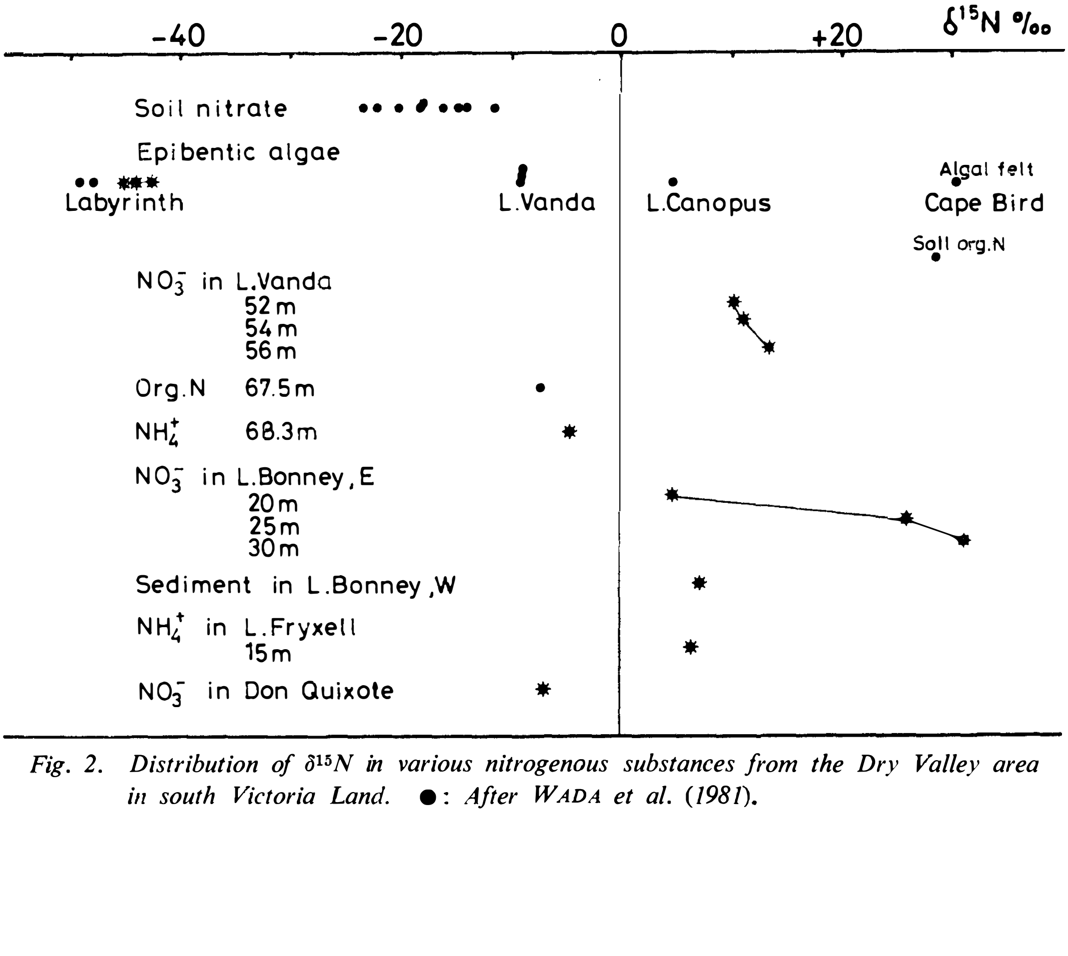

Fig. 2. Distribution of $\delta^{15}N$ in various nitrogenous substances from the Dry Valley area in south Victoria Land. ●: After WADA et al. (1981).

4. Discussion

Figure 2 summarizes the $\delta^{15}N$ values in various nitrogen compounds from the Dry Valley area. Data reported by WADA *et al.* (1981) were also included. The nitrogen isotope ratio of soil nitrate ranged from -11 to $-24‰$ and the lowest value of $-23.4‰$ was found for soil materials collected in the elevated areas near Lake Vanda, where soil nitrate seemed to originate from atmospheric precipitation. Epibenthic algae gave a wide range of $\delta^{15}N$ values from -49 to $4.0‰$. The variation resulted likely from difference in $\delta^{15}N$ of source nitrogen as well as in physicochemical and microbial nitrogen transformation in each ecosystem (WADA *et al.*, 1981). The epibenthic algae collected from saline ponds in the Labyrinth gave the lowest $\delta^{15}N$ value down to $-49‰$ in the biosphere.

The occurrence of the lowest $\delta^{15}N$ value in the algae was schematically explained in Fig. 3. Gaseous ammonia ($-10‰$) and nitrogen dioxide ($-9.3‰$) in the atmosphere

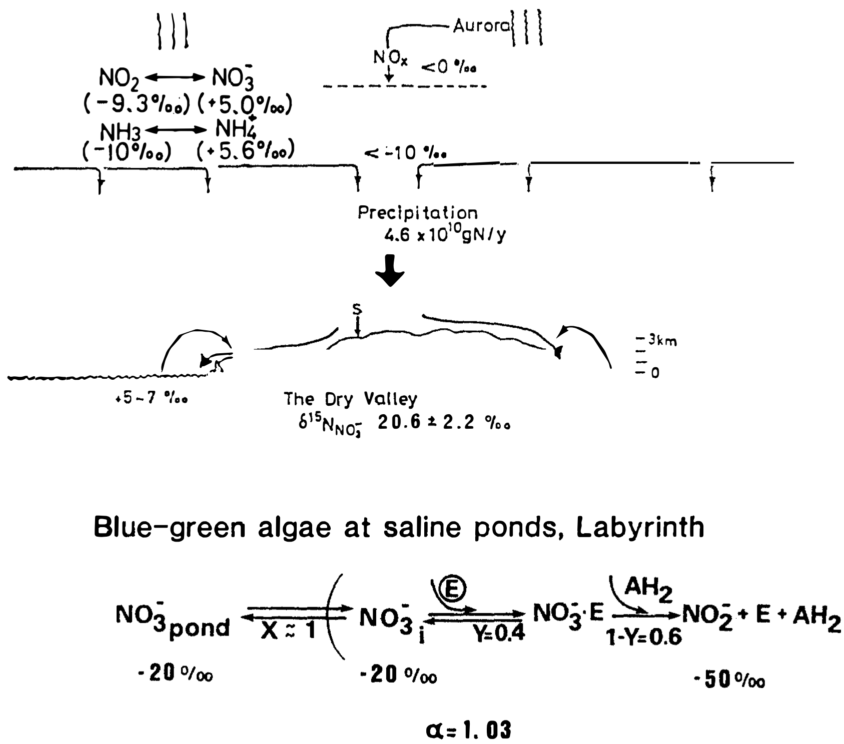

Fig. 3. Schematic model for the occurrence of ^{15}N depleted epibenthic algae in the saline ponds at the Labyrinth. Top figure illustrates the origin of ^{15}N depleted nitrate via atmospheric precipitation (see text). Bottom figure indicates nitrate reducing system of the algae. $NO_3^-{}_{pond}$: nitrate in saline ponds, $NO_3^-{}_i$: nitrate in algal cell, NO_3^--E: enzyme substrate complex, AH_2: electron donor such as NADH or NADPH, X and Y: fraction of flux of backward reaction to forward reaction. According to WADA (1980), the overall fractionation factor (α: $NO_3^-{}_{pond}$-algal cell) is given by the equation: $\alpha=1+\Delta k_2XY$, where Δk is the fractionation factor of N-O bond cleavage. In case $\alpha=1.03$, X and Y become 1 and 0.4, respectively and $\delta^{15}N_{algae}=-20‰-30‰=-50‰$.

in the temperate regions exhibited more negative $\delta^{15}N$ values than those of ammonium and nitrate in aerosols (around 5‰), because of the isotope exchange equilibria between gaseous and condensed phases (MOORE, 1977). The successive removal of the condensed phase during air mass transportation to the polar region can enhance ^{15}N depletion in nitrogenous compounds which precipitate in the Antarctic region. NO_x depleted in ^{15}N can be also produced by photochemical reactions as well as by auroral activities (PARKER *et al.*, 1978). In fact an average $\delta^{15}N$ value of −20.6‰ was obtained for soil nitrate from the elevated areas in the Wright Valley (WADA *et al.*, 1981). This value is, thus, adopted for nitrate in the saline ponds in the Labyrinth where most of nitrogenous compounds are supplied via atmospheric precipitation. According to WADA and HATTORI (1978), the nitrogen isotope fractionation during nitrate assimilation by a marine diatom is inversely related to its growth rate under light-limited growing conditions. A fractionation of 1.03 has been obtained as an maximum. Saline ponds in the Labyrinth contain a large amount of nitrate up to 10 mg at. N/*l*. Growth of the epibenthic algae at high nitrate concentration and low light intensity may yield a large isotope fractionation. The lowest value of the epibenthic algae can, thus, be explained by the combination of low $\delta^{15}N$ in source nitrate and the large isotope fractionation associated with the nitrate assimilation.

The mixolimnion in Lake Vanda extends from the surface down to 40 m (Fig. 4). Water temperature gradually increases up to 25°C below 40 m, providing an optimal condition for microbial activities even in the polar region. Dissolved oxygen is supersaturated above 55 m, and then decreases rather sharply with depth. The bottom layer below 59 m is anaerobic. Rates of nitrification, photosynthesis and denitrification were highest at 52.5–55 m, 55–59 m and 59.5–62.5 m, respectively (VINCENT *et al.*, 1981). Increase in $\delta^{15}N$ of nitrate between 52 and 56 m, thus, results mainly from an isotope fractionation during the assimilation of nitrate which is produced via nitrification. It is suggested that the nitrate reduction of phytoplankton at the depth of 55–59 m

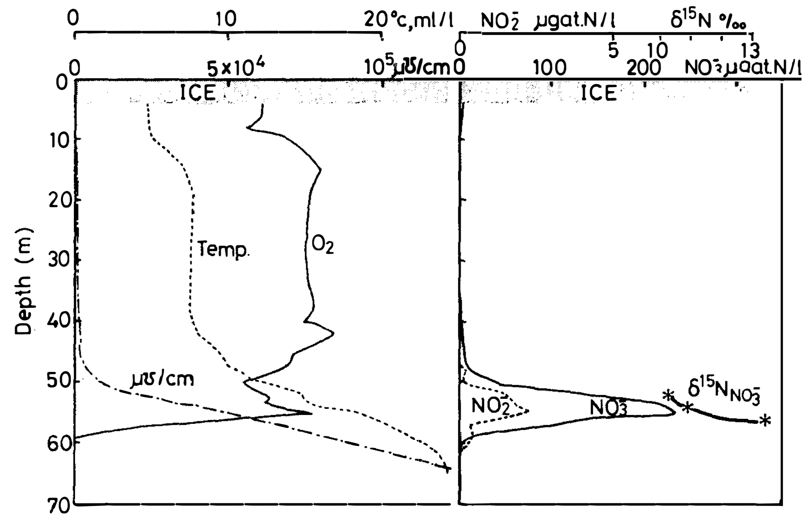

Fig. 4. Distribution of physical and chemical variables (after TORII *et al., 1975;* VINCENT *et al., 1981) and $\delta^{15}N$ in nitrate in Lake Vanda on 30 November and 1 December 1979.*

proceeds under the light limited condition, that is, the rate of nitrate assimilation is controlled by the supply of electron donor, *e.g.*, NADH, NADPH, or reduced feredoxin.

As indicated in Fig. 1, nitrate was highly enriched in ^{15}N in the bottom layers of the east lobe of Lake Bonney. Intensive microbial investigation has never been carried out for this part of the Lake. Some biological activities relating to nitrate reduction and/or ammonia volatilization may cause the observed $\delta^{15}N$ variation in nitrate, as no other process can produce this magnitude of variation in $\delta^{15}N$. However, at present, the dissolved oxygen content in the east lobe showed a marginal content with which bulk denitrification cannot proceed. The development of anaerobic condition in bottom layers is generally related to the supply of organic material. It remains unclear whether some organic aggregate in the middle layers can provide an anaerobic microsite where denitrification takes place with a large isotope fractionation or not. Alternative explanation takes some historical events into considerations. According to MATSUBAYA *et al.* (1979), Lake Bonney was once shallow–salt deposit system and water depth has been increasing during the geological time. If so, there was some stage when active algal community had flourished and enhanced the ^{15}N content of the total nitrogen in Lake Bonney as is the case with Lake Canopus where ^{15}N enrichment of epibenthic algae might have taken place through the isotope fractionation associated with ammonia volatilization (WADA *et al.*, 1981).

Distribution of dissolved oxygen in the west lobe of Lake Bonney clearly indicated the occurrence of denitrification in the anoxic layers. The $\delta^{15}N$ values of dissolved N_2 gas exhibited the highest value so far reported in the literature (Fig. 5). Average $\delta^{15}N$ value of 2.0‰ can appear in case about 100 μg at. N/*l* of nitrate with 20‰ was converted to molecular nitrogen. Comparison of the west lobe with the east lobe

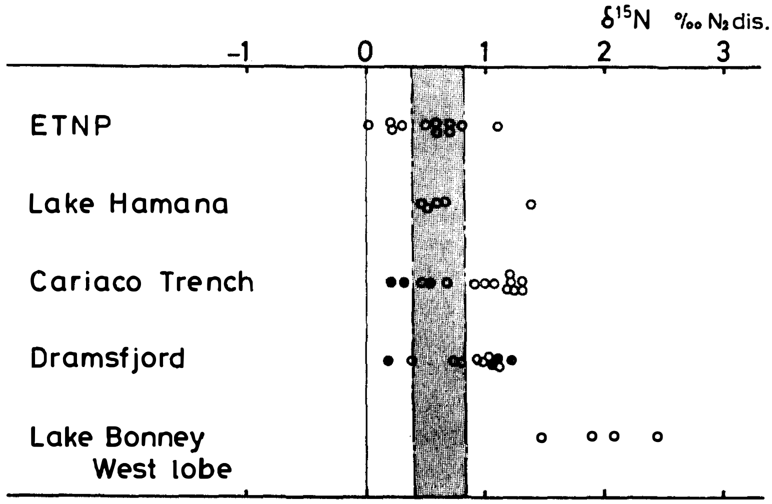

Fig. 5. Summary of $\delta^{15}N$ in dissolved nitrogen gas in various anoxic waters (○). Shaded area stands for the ^{15}N values of dissolved nitrogen gas in equilibrium with atmospheric nitrogen. Data for oxic waters were also included (●). ETNP: the eastern tropical North Pacific Ocean (after CLINE and KAPLAN, 1975). Lake Hamana: after WADA et al. (1970). Cariaco Trench and Dramsfjord: after RICHARDS and BENSON (1961).

suggested the likely occurrence of this type of denitrification. Supersaturation of oxygen in the upper layer of the west lobe may result from the oxygen supply by living algal mat as was reported in Lake Fryxell (WHARTON *et al.*, 1982). Bubbling of O_2 from the bottom may lower the nitrogen/argon ratios in the upper layers of the west lobe (Table 2).

Reduced form of organic nitrogen exhibited more negative value in Lake Vanda (−4.6‰) than those in Lakes Fryxell (6.2‰) and Bonney (7.3‰), suggesting the different origin of nitrogen in these lakes.

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