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FORMS OF NITROGEN (NO_3^- -N; NO_2^- -N AND NH_2CONH_2 -N) AND THEIR RELATIONS TO A.O.U. IN THE INDIAN COASTAL WATERS OF ARABIAN SEA

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The distribution of three important dissolved forms of nitrogen, viz. nitrate, nitrite and urea in the surface and bottom water samples collected from 27 selected hydrographic profiles, in the Arabian Sea, along the west coast of India is described. Of the three forms, nitrate concentrations were the highest and comparatively higher concentrations were observed in the bottom water. Decomposition of organic matter resulting in the release of the thermodynamically stable nitrogen species, *i.e.* nitrate, may be the major factor resulting in higher nitrate concentrations at these depths, where the water is also characterized by low values of dissolved oxygen and temperature. The significant positive correlation between A.O.U. and nitrate of the bottom water samples emphasizes the role of oxidative decomposition of organic matter which plays an active role in reducing the oxygen concentrations below the theoretical values since at this depth (≈ 200 m) the net production is taken to be zero. This is also evidenced by the negative correlation of nitrate with dissolved oxygen and temperature, for the bottom samples.

Keywords: Nutrients; Nitrate; A.O.U.; Coastal waters

INTRODUCTION

A nutrient element is defined as one that is functionally involved in the processes of living organisms (Parsons, 1975). Thus anything besides water and carbon dioxide that is required by plants in the synthesis of organic matter or skeletal materials is regarded as a nutrient. Nitrogen and phosphorus are considered to be the main limiting nutrients. Recent ecosystem level nutrient limitation studies support the evidence that phosphorus is the primary limiting nutrient in fresh waters and nitrogen is the limiting nutrient in marine systems (Conley, 2000). Most organisms can only use nitrogen in the 'combined' forms such as nitrate and ammonia which are less abundant in nature than organic N is but are more easily assimilated (Naqvi and Jayakumar, 2000).

The major forms of nitrogen in the marine environment are interrelated by a series of biogeochemical reactions known as the 'nitrogen cycle'. The difference in temperature, length of

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growing season, the solar irradiance can be expected to influence primary production and in turn the whole 'nitrogen cycle'. Factors like tidal range, climate and the vegetation dominating the coastal zone have not been taken into account, although it is presently known that activity of photosynthetic organisms dramatically affects nitrogen dynamics (Sundback and Snoeijs, 1991). Human activities have markedly altered the Earth's 'nitrogen cycle' by doubling the natural rate of nitrogen fixation and causing atmospheric N decomposition to increase by three fold to more than ten fold compared to preindustrial times (Kronvang *et al.*, 1993; Bonsdorff *et al.*, 1997). The increase in nutrient levels in many coastal marine systems following to anthropogenic impacts is well documented (Ryther and Dunstan, 1971; Kronvang *et al.*, 1993; Bonsdorff *et al.*, 1997). High levels of nutrients (N and P) can cause eutrophication in fresh waters and coastal zones (Owens, 1993; Oslo and Paris Commission, 1993; Stanners and Bourdeau, 1995; Van Dijk *et al.*, 1995). Nitrogen loading which enters coastal waters can be lost via 3 processes. (1) Transport to outer sea area during seasons of low primary production. (2) Accumulation into the bottom sediments (3). Denitrification, which is known to be the main sink for nitrogen especially in many coastal areas (Gran and Pitkanen, 1999).

In this paper, the distributions of three important forms of nitrogen, *viz.* nitrate, nitrite and urea, have been correlated with the distribution of hydrographical parameters like salinity, temperature, pH, dissolved oxygen and apparent oxygen utilization, in the Arabian Sea. Nitrogen is present in sea water as (a) molecular nitrogen (b) fixed inorganic salts, such as nitrate, nitrite and ammonia (c) a range of organic nitrogen compounds, *e.g.*, amino acids and urea and (d) particulate nitrogen. In order to satisfy their nitrogen requirements, phytoplankton are normally dependent on fixed nitrogen, with a preference to nitrate, nitrite and ammonia. This process takes place in the euphotic zone and some of the nitrogenous nutrients are also released in soluble form within this zone. The remainder are transported out via sinking particulates, and a large fraction of this nitrogen is released back into the solution at depth in the water column by remineralisation of the organic material, mainly via bacterial mediation, the final inorganic end product being nitrate.

Combined inorganic nitrogen is added to the oceans by terrestrial drainage and most river waters contain substantially greater amounts of combined nitrogen than does sea water. Biological processes are dominant in the interconversion of the various forms of nitrogen in the sea, and in turn, all marine organisms are dependent directly or indirectly upon the store of inorganic nitrogen in the sea for the synthesis of many of their essential components.

Nitrogen exists in nine oxidation states (-3 to $+5$) all of which have significant role in the marine environment (Martin, 1970). Nitrate-nitrogen is the thermodynamically most stable form of combined inorganic nitrogen in well-oxygenated waters and the variation in concentration of nitrate and of the more reduced inorganic nitrogenous compounds is predominantly the result of biologically activated reactions. Nitrate-nitrogen is the intermediate oxidation state between ammonia and nitrate, and as such it can appear as a transient species in both the oxidation of ammonia and the reduction of nitrate. Urea is a low molecular weight organic compound, weakly exothermic on hydrolysis and less attractive for "sensu stricto" heterotrophic organisms; but as a nitrogen compound, it is easily hydrolysable for autotrophic organisms under limiting conditions (Nair *et al.*, 1994). A considerable amount of urea is found in some coastal and marine environments. It may originate from fresh water discharge, especially through sewage from densely populated areas, since man and other terrestrial vertebrates represent a primary source of urea. Drainage from agricultural regions, where urea

based fertilizers are used, contributes heavily also in the concentration of urea (Remsen, 1971).

MATERIALS AND METHODS

Water samples were collected during cruise No.162 of FORV SAGAR SAMPADA during the pre-monsoon period. Analyses were done for both the surface and bottom water samples collected from 27 selected hydrographic profiles lying between latitudes $7^{\circ} 10' - 20^{\circ} 40' N$ and longitudes $70^{\circ} 40' - 77^{\circ} 22' E$ in the Arabian sea, along the west coast of India (Fig. 1). The sampling was done using a Niskin sampler (Model 1010) and samples were filtered through a Whatman GF/C ($0.45 \mu m$). The filtered samples were stored in polythene bottles by deep-freezing for subsequent nutrient analyses. Hydrographical parameters like salinity and tem-

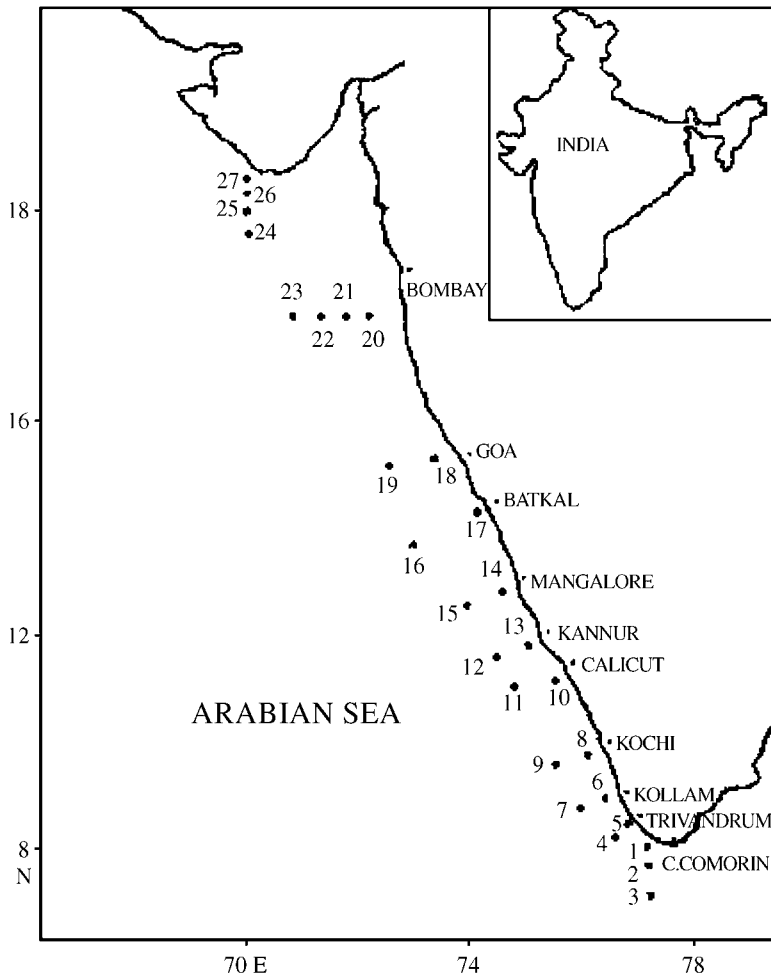


FIGURE 1 Location map of stations sampled.

perature were obtained from *in situ* measurements using Conductivity Temperature Depth Profile (CTD). pH was measured using a digital pH meter. The chemical estimation of dissolved oxygen was done on board by the Winkler method (Grasshoff, 1983).

Nitrate, nitrite and urea were determined by spectrophotometric analyses using a Hitachi UV-VIS spectrophotometer.

Urea was determined by the diacetyl monoxime method (Grasshoff, 1983). The principle of this method is that in strongly acidic solution and in the presence of a weak oxidant, urea forms a condensation product with diacetyl monoxime. This product interacts with semicarbazide to form a magenta coloured complex with an absorption maxima at 520 nm.

Spectrophotometric determination of nitrite in sea water was based on the reaction of nitrite with an aromatic amine, sulphanilamide hydrochloride, leading to the formation of a diazonium compound which, when coupled with a second aromatic amine, N-(1-naphthyl)ethylenediamine dihydrochloride, leads to the formation of a pink coloured azo dye (Grasshoff, 1983) at a λ_{\max} of 540 nm.

The method adopted for nitrate determination was the reduction of nitrate to nitrite by passing through a column containing copper-coated cadmium granules (Grasshoff, 1983) and the resulting nitrite was analysed as described above. The reduction efficiency of the column was checked to be 98%.

RESULTS AND DISCUSSION

The results obtained for the analyses of hydrographical parameters like temperature, salinity, pH, dissolved oxygen and A.O.U. of surface and bottom water samples are given in Table I.

TABLE I Hydrochemical characteristics of the surface (S) and bottom (B) water samples collected from different stations along the west coast of India

| Station No. | Temperature (°C) | | Salinity ($\times 10^{-3}$) | | Dissolved Oxygen ($ml l^{-1}$) | | pH | |
|-------------|---------------------|-------|----------------------------------|-------|-------------------------------------|-------|------|------|
| | S | B | S | B | S | B | S | B |
| 1 | 29.26 | 28.06 | 33.93 | 33.73 | 4.648 | 4.183 | - | - |
| 2 | 29.35 | 13.91 | 33.94 | 35.06 | 4.533 | 0.397 | - | - |
| 3 | 29.33 | 17.30 | 34.07 | 35.04 | 4.841 | 0.582 | - | - |
| 4 | 29.40 | 13.55 | 34.02 | 35.12 | 4.823 | 0.556 | 8.40 | 7.86 |
| 5 | 29.38 | 29.28 | 34.07 | 34.07 | 4.534 | 4.491 | 8.33 | 8.31 |
| 6 | 29.23 | 29.24 | 34.59 | 34.60 | 4.626 | 4.501 | 8.15 | 8.21 |
| 7 | 29.19 | 12.90 | 34.29 | 35.20 | 4.694 | 0.481 | 8.37 | 8.28 |
| 8 | 29.07 | 14.30 | 34.26 | 35.22 | 4.901 | 0.478 | 8.56 | 8.32 |
| 9 | 29.05 | 29.06 | 34.69 | 34.69 | 4.793 | 4.543 | 8.40 | 8.39 |
| 10 | 29.60 | 29.13 | 35.03 | 35.02 | 4.708 | 4.544 | 8.41 | 8.40 |
| 11 | 29.08 | 13.72 | 34.62 | 35.27 | 4.578 | 0.308 | 8.51 | 8.23 |
| 12 | 28.96 | 13.78 | 35.35 | 35.31 | 4.879 | 0.349 | 8.58 | 8.12 |
| 13 | 28.26 | 28.15 | 34.77 | 35.06 | 4.960 | 4.313 | 8.52 | 8.45 |
| 14 | 27.66 | 27.50 | 35.37 | 35.35 | 4.784 | 4.303 | 8.49 | 8.43 |
| 15 | 28.41 | 14.34 | 35.59 | 35.42 | 4.966 | 0.351 | - | - |
| 16 | 25.76 | 25.75 | 35.55 | 35.55 | 5.051 | 4.898 | 7.52 | 7.38 |
| 17 | 26.52 | 26.49 | 35.72 | 35.74 | 4.891 | 4.908 | 7.53 | 7.36 |
| 18 | 27.12 | 26.26 | 37.86 | 35.96 | 5.320 | 3.561 | 7.57 | 7.43 |
| 19 | 26.74 | 25.81 | 35.90 | 36.06 | 5.096 | 3.404 | 7.50 | 7.46 |
| 20 | 23.41 | 23.38 | 35.41 | 35.40 | 4.473 | 3.751 | 7.53 | 7.49 |

Figure. 1 shows the location map of selected hydrographic profiles. The station wise and depth wise distribution of nitrate, nitrite and urea are given in Figures 2 and 3 respectively. The comparisons of theoretical dissolved oxygen values and A.O.U. of the surface and bottom water samples are given in Figures 4 and 5. The significant correlations with respective hydrographical parameters are given in Figures 6 and 7.

In the results presented here, the surface values of nitrate-nitrogen showed a variation of $0.79\text{--}6.58\ \mu\text{mol l}^{-1}$ whereas the bottom concentrations ranged from $0.64\text{--}13.2\ \mu\text{mol l}^{-1}$. Higher concentrations were reflected in the bottom water samples compared to the surface with the exception of Mumbai stations (21, 23–27) which displayed higher nitrate concentra-

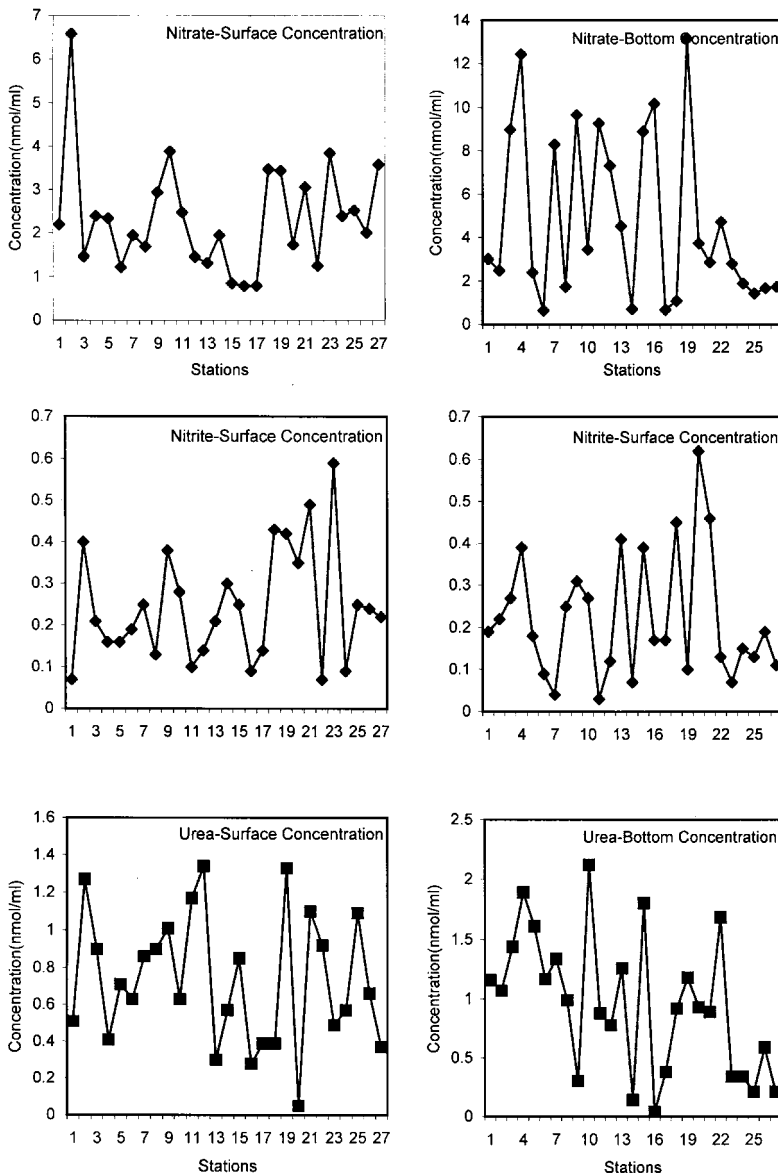


FIGURE 2 Distribution of nitrate, nitrite and urea in the surface and bottom (30 and 200 m depths) water samples in the Indian coastal waters of Arabian Sea.

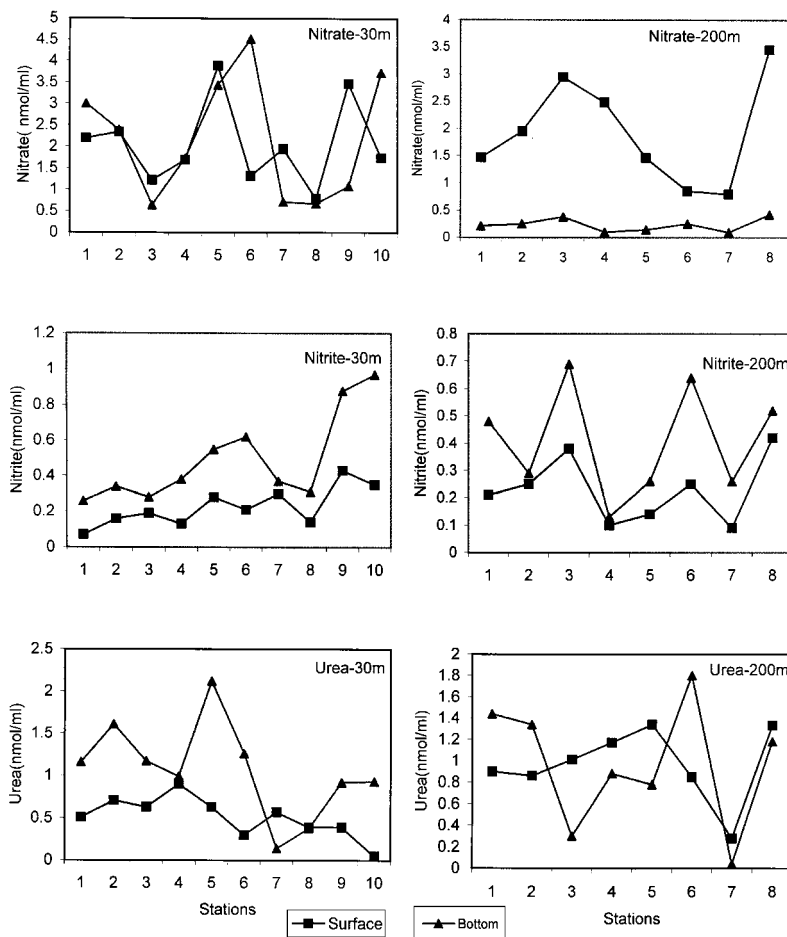


FIGURE 3 Distribution of nitrate, nitrite and urea in the surface and bottom water samples at depths 30 and 200 m in the Indian coastal waters of Arabian Sea.

tion in the surface compared to the bottom. The highest surface concentration was reported in the southern part of the west coast (Station 2) and the lowest value in the central region (Stations 16 and 17). The highest bottom concentration was observed at Station 19 (central region of west coast) and the lowest value at Station 6 (southern part of the west coast).

As in the case of nitrate, nitrite concentrations were higher in the bottom waters compared to the surface. The surface variation was from 0.07 – $0.59 \mu\text{mol l}^{-1}$ with the highest value in the northern part of west coast (Station 23) and the lowest value at Stations 1 and 22 (southern and northern regions respectively). For bottom water samples the highest value was in the northern region (Station 20) and the lowest in the southern region of west coast (Station 11).

The study revealed the surface concentration of urea to be varying from 0.05 – $1.34 \mu\text{mol l}^{-1}$ and the bottom values from 0.04 – $2.12 \mu\text{mol l}^{-1}$. The highest concentrations in the surface as well as bottom water samples were observed in the southern part of west coast (Stations 12 and 10 respectively). The lowest surface concentration was observed in the northern part (Station 20) and the lowest bottom concentration in the central region (Station 16). Among the three forms of dissolved nitrogen, nitrate concentrations were comparatively higher and urea exhibited much lower concentrations.

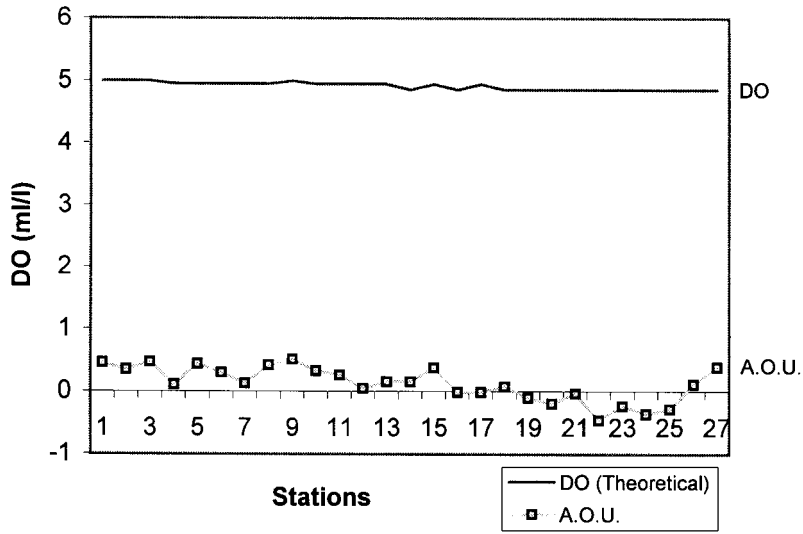


FIGURE 4 Comparison of DO (Theoretical) and A.O.U. in the surface samples.

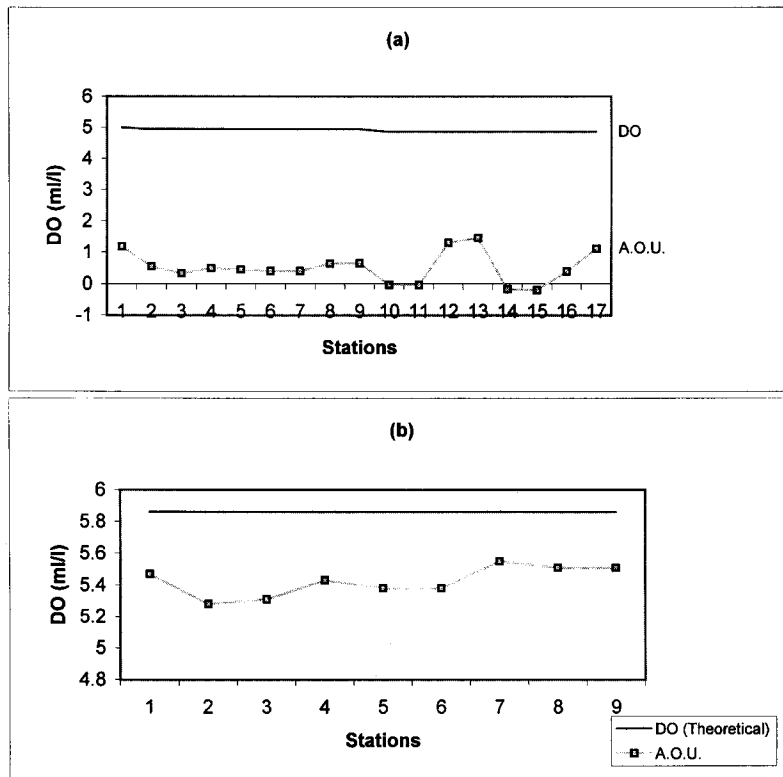


FIGURE 5 Comparison of DO (Theoretical) and A.O.U. in the bottom water samples from depths 30 m (a) and 200 m (b).

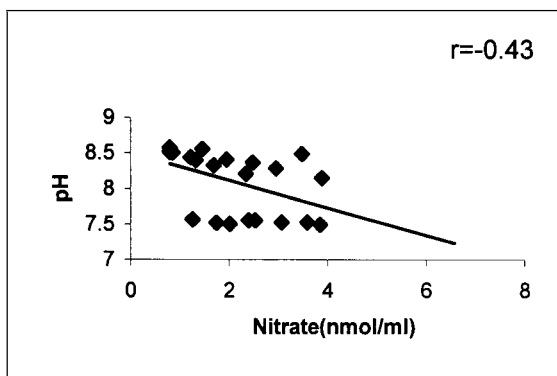


FIGURE 6 Correlation between nitrate and pH in the surface waters.

For nitrate, the Figure showing the distribution at lower depth is a merging type, whereas clear distinction between surface and bottom concentrations was observed at higher depth *i.e.* at ≈ 200 m depth where the bottom values displayed a significant difference from the respective surface values.

The higher values of nitrate - nitrogen observed in the deeper layers compared to the surface are an indication of higher nutrient consumption in the surface layers leading to increased photosynthetic activity along with the regenerative process which plays an active role in the nutrient enrichment of bottom layers. First stage in this organic decomposition process will be the oxidation of ammonia to nitrite and to nitrate with the utilization of available oxygen. In accordance with this, the results presented here showed higher concentrations of nitrate in the bottom water samples compared to the other two forms of nitrogen, namely, nitrite and urea. Direct dissolution from the atmosphere and primary production increases the oxygen concentration in the surface water while the organic decomposition depletes the oxygen in the bottom layers (Unnithan *et al.*, 1975).

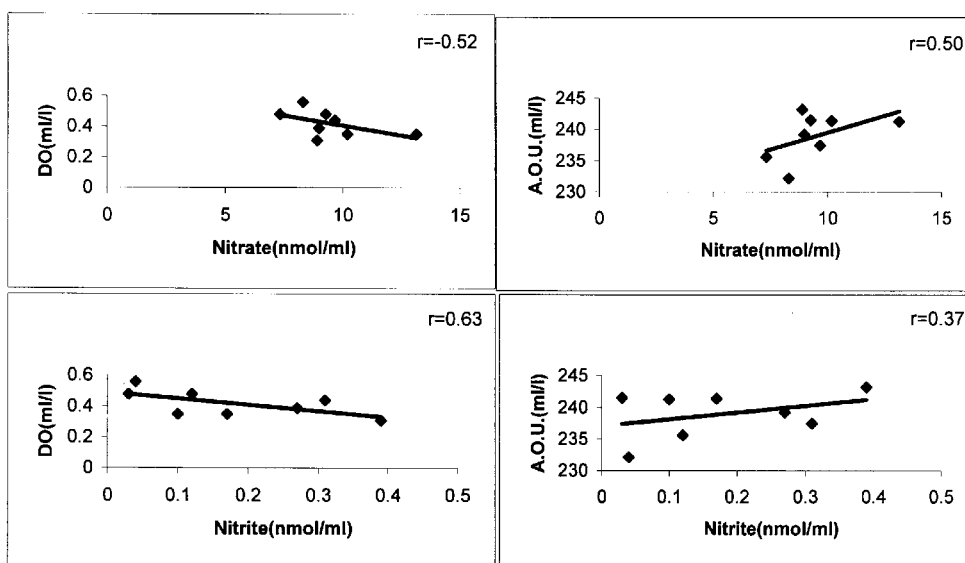


FIGURE 7 Correlation of nitrate and nitrite with DO and A.O.U. in the bottom water samples (200 m Depth).

Earlier studies on the northern basin of Arabian sea (Sen Gupta *et al.*, 1976) showed nitrate-nitrogen values ranging from $2.5\text{--}10\ \mu\text{mol l}^{-1}$. Similar studies conducted in the western Indian Ocean (Shirodkar and Jayakumar, 1990) displayed a variation of $0\text{--}29.37\ \mu\text{mol l}^{-1}$. Sen Gupta *et al.* (1976) have found nitrite values in the northern basin ranging from $0.25\text{--}0.75\ \mu\text{mol l}^{-1}$. Nitrite-nitrogen concentrations in the range of $0.31\text{--}3.12\ \mu\text{mol l}^{-1}$ have been observed in the western Indian Ocean (Shigenobu Takeda *et al.*, 1995). In the Arabian sea, a relatively low N/P ratio in inorganic nutrients in the water column implies that nitrogen would be depleted first by the biota if the deep water was upwelled into the photic zone (Pollenhne *et al.*, 1993). Earlier studies have shown that urea concentration increases towards central west coast of India. The studies on the distribution of urea in the coastal and oceanic waters around India showed that urea forms a significant nitrogen source for marine phytoplankton off the west coast of India (Verlencar, 1980).

On comparing the interrelationship between the nitrogen species (nitrate, nitrite and urea) and various hydrographical parameters, the following observations could be drawn. The surface values of nitrate, nitrite and urea displayed no significant correlation with the hydrographical parameters such as salinity, temperature, DO and A.O.U. (Apparent Oxygen Utilization). The surface values of nitrate showed a negative correlation with pH ($r = -0.45$, Fig. 6) whereas nitrite and urea showed no significant correlation with pH in the surface samples. The bottom water samples collected from 200 m depth displayed significant correlation only for nitrate and nitrite with their respective dissolved oxygen values and A.O.U. For these samples no significant correlation was observed for nitrate, nitrite and urea with other hydrographical parameters such as pH, temperature and salinity.

For phytoplankton to be productive, they require an optimum range of temperature. The regeneration of nutrients by the decomposition of organic matter resulting in the release of thermodynamically most stable form of nitrogen species *i.e.* nitrate may be the major factor resulting in higher nitrate concentrations in the samples collected from the bottom at 200 m depth which are characterized by low values of dissolved oxygen and temperature. The temperature recorded for the bottom samples were very low ($12.9^{\circ}\text{--}14.4^{\circ}\text{C}$) compared to the surface. One of the comparatively significant correlations was the interrelationship between nitrate-nitrogen and dissolved oxygen in the bottom water samples (200 m) with a coefficient of correlation $r = -0.52$ (Fig. 7) and between nitrite-nitrogen and dissolved oxygen ($r = -0.63$, Fig. 7). As observed, low dissolved oxygen values at these depths were associated with higher concentration of nitrate-nitrogen. Dissolved oxygen is an important parameter in assessing water quality. In aquatic systems, oxygenation is the result of an imbalance between the process of photosynthesis, degradation of organic matter and reaeration (Granier *et al.*, 2000). The surface replenishment by atmospheric exchange and the photosynthetic activity and the subsurface consumption of oxygen for the oxidation of organic matter make the dissolved oxygen content of surface waters slightly higher than that of subsurface waters. The changes in concentrations will be affected by photosynthetic activity and it will be more pronounced in the euphotic surface layers. Dead organisms and other organic matter undergo bacterial decomposition process as they fall from the surface and this regenerative process is active throughout the water column. The oxidative process removes oxygen from these waters and the end products of decomposition accumulates in the deeper layers. As a consequence of nutrient loading, detrimental effects have been observed on the health of coastal ecosystems leading to, amongst other things, excess accumulation of phytoplankton

biomass (Malone *et al.*, 1986), episodes of noxious blooms, reductions in aquatic macrophyte communities (Sand-Jensen and Borum, 1991; Duarte, 1995) and the depletion of dissolved oxygen in bottom waters. Similar comparisons between nutrients and dissolved oxygen in the upper 1000 m of equatorial Indian Ocean showed a good negative correlation (Shirodkar and Jayakumar, 1990). Another study conducted in the Lakshadweep waters also displayed a negative correlation (Singh *et al.*, 1990).

CORRELATION WITH APPARENT OXYGEN UTILIZATION

Apparent oxygen utilization is a useful parameter, which can be estimated from the difference between the calculated oxygen saturation value at the *in situ* temperature and salinity and the actual concentration measured. The biologically induced changes in oxygen concentrations, which have taken place after the water has ceased to be in direct exchange with the atmosphere, are usually expressed in terms of A.O.U. (Riley, 1971).

In these results, the surface samples (Stations 16, 17, 19–25) were characterized by negative A.O.U. values, whereas the bottom samples showed positive A.O.U. values with exception of a few stations. The negative A.O.U. values exhibited by some surface stations indicates that the measured oxygen values are in excess in comparison with the theoretical ones. It agrees with the concept that the surface layers are characterized by higher dissolved oxygen contents due to increased photosynthetic activity and they have a chance for direct reoxygenation from the atmosphere. The productivity of a water column will be highest within the upper 10 m, where light conditions permit maximum photosynthetic activity. As a result of these the concentration of nutrients will be low and the dissolved oxygen will be high in the surface compared to the bottom. In this result a positive correlation was reflected in the comparison between the A.O.U. values of the bottom water samples (200 m) and the respective nitrate concentrations ($r = 0.50$, Fig. 7) and also with the respective nitrite concentration ($r = 0.38$, Fig. 7). The positive A.O.U. values in the bottom samples indicate that the measured oxygen concentrations are lower than the theoretical ones. The bottom samples were collected from a depth of about 200 m (below the compensation depth) where net production is taken to be zero and in this zone decomposition of organic matter dominates photosynthesis. From these observations it can be concluded that the oxidative decomposition of organic matter plays an active role in bringing down the oxygen values much lower than the expected ones. First stage in this decomposition process is the oxidation of ammonia to nitrite and to nitrate using the available oxygen. Any anomaly between the assumed and measured oxygen concentrations is ascribed to photosynthesis or to the biological oxidation of organic matter. Hence A.O.U. should be related to nutrient changes which accompany these processes (Spencer, 1975). This is further emphasized by the good negative correlation between nitrate-nitrogen and dissolved oxygen which was an exclusive feature of the samples collected from approximately 200 m depth.

No significant correlations were reflected in the comparison of A.O.U. values with urea which may be due to its comparatively low thermodynamic stability.

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