Chapter II

EXPERIMENTAL DETAILS

The northern Indian Ocean is an ideal repository of signatures that help us understand various biogeochemical processes operating in the region. The present study aims to address some of these issues that would help in determining upwelling rates in the Arabian Sea, the provenance of sediments being deposited in the Bay of Bengal and to trace the climatic and provenance changes during the past 40-50 kyr. Towards these objectives several vertical profiles of water samples and numerous sediment cores were collected during cruises in the Arabian Sea and the Bay of Bengal.

Sampling Procedures

Seawater sampling

Several depth profiles were occupied in the Arabian Sea during three ocean expeditions in 1994, 1995 and 1997 onboard *FORV Sagar Sampada,*

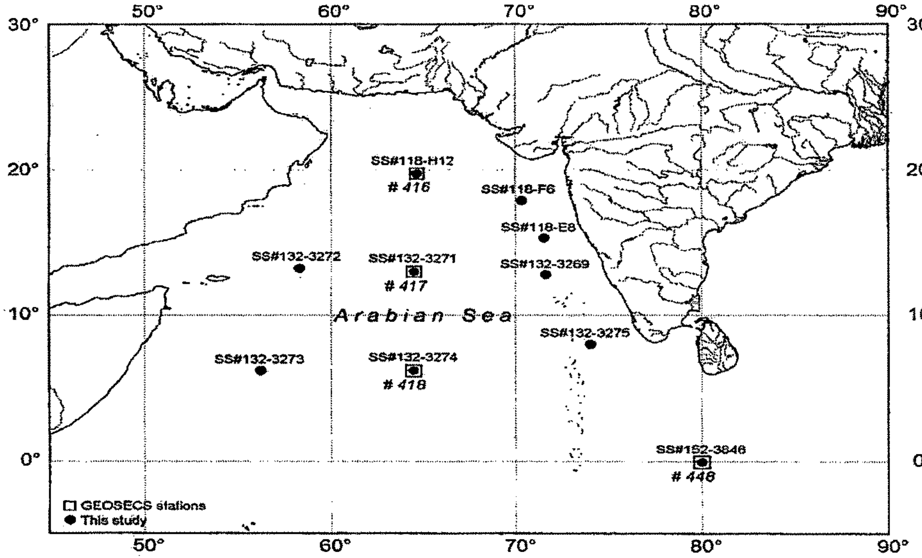

Fig. 2.1: Water Sampling locations in the Arabian Sea and the Equatorial Indian Ocean. GEOSECS station numbers are given after '#'. The boxed filled circles are the reoccupied GEOSECS stations.

oceanographic research vessel belonging to Ministry of Earth Sciences (MOES, earlier Department of Ocean Development (DOD), Government of India) for the collection of seawater at various depths. For the present study, a total of ten sampling stations in the Arabian Sea and one in the equatorial Indian Ocean were identified (Fig. 2.1). For ^{14}C analysis, about ten samples were collected per profile from pre-selected depths, except in the equatorial Indian Ocean station where samples from twenty depths were collected. Nearly 120 lit. of seawater samples were collected from each depth for onboard measurements (laboratory analysis) and processing for various parameters (Narvekar et al., 1997).

Sediment Sampling

Seventeen sediment core samples from the Bay of Bengal were used in the present study (Fig. 2.2). Only the top sections of the cores were analyzed for their chemical and isotopic composition to ascertain their source (provenance). In addition, numerous grab samples from the deltaic regions of rivers draining into the Bay of Bengal were analyzed to understand the source of the sediments.

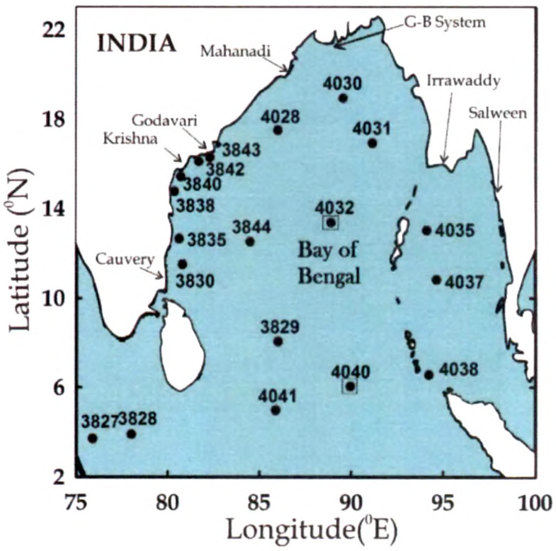

Fig. 2.2: Locations of sediment core samples collected in the Bay of Bengal. Boxed filled circles are the cores studied in detail

Onboard Processing and Measurements

Seawater Measurements

For seawater sampling, 100 lit. Go-Flo bottles with the hydrographic wire were used at various depths at each station. The seawater collected was immediately sub-sampled for dissolved oxygen measurements, dissolved inorganic carbon (ΣCO_2), nutrients (silicate, phosphate and nitrate), salinity and radiocarbon. About 125 ml of the seawater sample was used for dissolved oxygen (measurements done on duplicate samples), 60 ml of the seawater sample was used for ΣCO_2, 500 ml for nutrients, 200 ml for salinity and ~100 l for radiocarbon measurements.

Dissolved Oxygen Measurement

Immediately upon collection of the seawater samples in the BOD bottles, Winkler A and Winkler B were added to the sample to estimate the dissolved oxygen content using the modified Winkler titration method (Carpenter, 1965). The bottles were tightly capped and agitated vigorously. They were agitated again after 30 min and then stored in a dark place for ~8-16 hrs. After this, 0.5 ml of conc. H_2SO_4 was added to liberate iodine. This iodine was titrated with 0.2 N sodium thiosulfate solution by a *Metrohm 655 Dosimat* auto-titrator to the end point using starch as an indicator. Based on repeat analysis of samples, the precision for dissolved oxygen by Winkler titration was ±2 μmolkg^{-1} (1σ).

Estimation of Dissolved Inorganic Carbon (ΣCO_2)

Seawater samples were collected in a 60 ml ground joint bottle and immediately upon collection, two drops of saturated solution of mercuric chloride was added to inhibit organic growth. The sample bottle was capped tight using non-carbon silicone grease and then stored in a refrigerator till analysis. The total dissolved inorganic carbon (ΣCO_2) of seawater is defined as the total molar concentrations of all species of inorganic carbon present in dissolved phase, and is given by,

$$\Sigma CO_2 = [CO_2^*] + [HCO_3^-] + [CO_3^{2-}]$$

where bracket represent total concentrations of these constituents in solution (in mol.kg^{-1}) and [CO_2^*] represents the total concentration of all un-ionized dissolved carbon dioxide (as H_2CO_3 or CO_2). The ΣCO_2 in the seawater samples were determined by coulometric method, following the procedures as outlined by Johnson *et al.* (1985) and given in D.O.E. handbook (1994).

ΣCO_2 was measured in a small aliquot of seawater either onboard the ship or in the laboratory. In the latter case, the samples were poisoned with $HgCl_2$ and refrigerated till the time of measurement. ΣCO_2 measurements were carried out using a Coulometer (model 5012, UIC Inc., Joliet, Illinois). Based on repeat analyses, typical precision of ΣCO_2 measurements was ±3 $\mu mol.kg^{-1}$ (1σ). The accuracy of ΣCO_2 measurements was checked from analyses of certified reference seawater samples (Batch#42, December, 1997), supplied by Prof. Andrew G. Dickson of the Scripps Institute of Oceanography, USA. The mean ΣCO_2 measured during this study for this standard seawater is 1983.2 ± 2.3 $\mu mol.kg^{-1}$ (1σ, n=20), and is in good agreement with its certified value of 1985.1 ± 0.8 $\mu mol.kg^{-1}$ (Dutta, 2001).

Nutrient Measurements

The nutrients measurements for silicate, nitrate and phosphate were done onboard using a *Technicon Auto-Analyzer* (Gordon et al., 1993). The precision ($\pm 1\sigma$) of measurement for silicate, total nitrate and phosphate based on repeat measurements of seawater samples were <2% respectively for all the parameters. Accuracies of silicate and nitrate measurements were about ±5%, as checked by analyzing *CSK* standard solutions (Wako Chemical Industries Ltd., Japan).

Salinity Measurement

The seawater samples were brought to room temperature before the salinity measurement. The salinity of the seawater samples was determined using a *Guildline AutoSal Model 8400A* salinometer. The instrument determines the conductivity ratio (K_{15}) of a sample to that of a potassium chloride (KCl) solution containing 32.4356 g of KCl in 1 kg of solution (Standard Seawater). The software SEACALC was used to calculate salinity from *Autosal* values. The salinometer was calibrated using IAPSO standard seawater. The precision of salinity measurements by *Autosal* was typically ±0.0005 PSU, based on repeat measurements of seawater samples. The CTD salinity values were calibrated using the values measured by Autosal.

Extraction of DIC from Seawater for Radiocarbon Measurement

Nearly 100 l of seawater sample was collected and transferred immediately to a closed circulation system. For CO_2 extraction from seawater, 2N NaOH solution was kept in series with the circulation line along with a 5% HCl solution used as a gas scrubber. The system was connected and a continuous closed system circulation maintained using a *Cole-Parmer* peristaltic pump. Nearly 1 l of H_2SO_4 acid was added slowly through separating funnels to the seawater to extract DIC from seawater. The system was kept in circulation for nearly 8-10 hours to ensure quantitative extraction of CO_2 from the seawater. The extracted CO_2 was trapped in NaOH solution and brought back to the laboratory for analysis. Details of ^{14}C analysis methods in seawater samples followed in this study are described in Bhushan et al. (1994), (2000) and Dutta (2001).

Measurement Procedures

^{14}C measurements in Seawater

Briefly, CO_2 was extracted onboard by acidifying ~100 l of seawater in a closed circulation system trapped in aqueous NaOH solution. In the laboratory, CO_2 was liberated from the alkaline solution upon acidification under vacuum and converted to benzene through formation of Li_2C_2 and C_2H_2, and finally assayed for its ^{14}C activity using low level Liquid Scintillation Spectrometer. For atmospheric ^{14}C analysis, CO_2 was trapped from the air of marine boundary layer by pumping air through NaOH during the cruises using a greaseless pump and assayed for ^{14}C as above. A small aliquot of this CO_2 was used for $\delta^{13}C$ measurements, and the remaining converted to benzene in a Technical and Applied Scientific Knowledge Inc. (TASK) (Athens, Georgia) synthesizer. The benzene was assayed for its radiocarbon activity using a tricarb packard low background liquid scintillation counter (model 2250CA, Packard Instruments Co., Meriden, Connecticut). The counter was calibrated using benzene extracted from NBS Oxalic acid standard (Standard Reference Material 4990–C). Marble blanks were run periodically to check the background and blank levels (Bhushan et al., 1994). Based on repeat analysis of modern standards, typical precision for $\Delta^{14}C$ measurements was ±5‰.

$\delta^{13}C$ measurements of CO_2

The $\delta^{13}C$ measurements were done on about half of the ^{14}C samples using a VG 602D micromass mass spectrometer on a small aliquot of CO_2 liberated in the laboratory during the acidification of NaOH solution containing seawater CO_2 (precision ±0.1‰). $\delta^{13}C$ was measured in an aliquot of CO_2 used for benzene synthesis for isotopic fractionation correction of the ^{14}C results and determination of $\Delta^{14}C$ (Stuiver and Polach, 1977).

Sediment Measurements

AMS Radiocarbon dating

For radiocarbon dating using AMS, nearly 10-15 mg of Planktonic Foraminifera shells of select surface dwelling species (*Globigerina Ruber, Globigerina Sacculifer* etc) were extracted from the sediments through wet sieving and physical picking under a microscope. These shells were acidified to liberate CO_2 and this CO_2 was converted to graphite for AMS measurement. The AMS measurements were done at NSF Facility of AMS at University of Arizona, Tucson, USA using a 10 Mev pelletron accelerator connected to a mass spectrometer.

$CaCO_3$ Estimation

The $CaCO_3$ measurements in the sediments were carried out using *UIC* Coulometer Model 5012 (UIC Inc., Illinois, USA) attached to a CO_2 extraction system. Nearly 10-20 mg of the dried sediment sample was taken in the reaction vessel of the extraction system kept at 80°C. Nearly 3-4 ml of 30% H_3PO_4 was added to the sample and the evolved CO_2 was purged from the extraction system using CO_2 free zero air to the Coulometer. The evolved CO_2 was purged in a monoethanolamine solution in the Coulometer. A three point calibration curve was made using Na_2CO_3 as standard. The measurement precision was 3% based on repeat measurements (Bhushan et al., 2000).

Organic Carbon (C_{org}) Determination

The C_{org} in the samples was estimated based on the difference between the measured total carbon and inorganic carbon. The total carbon along with nitrogen was measured by *FISONS* NA1500 NC Elemental Analyser (*FISONS* Inc., Italy) and the inorganic carbon by *UIC* Coulometer Model 5012 (UIC Inc., Illinois, USA). For the estimation of total carbon and nitrogen using NC elemental analyser, the sediment samples were introduced in small aliquots of 10-30 mg in tin or aluminum cups through an auto-sampler into the combustion tube of the analyzer. The calibration was done using Deer River Black as standard reference material containing 2.53% C and 0.12% N. The average blank concentration measured over a period of about 200 days is 1.31 ± 0.12 μg C. The measurement precision for total carbon is 4% estimated by repeat analysis of Deer River Black Shale over several weeks. The measurement precision in the case of total nitrogen concentrations > 0.1% is 8% (Bhushan et al., 2001).

$\delta^{13}C$ and $\delta^{15}N$ of Organic Matter

For the measurement of $\delta^{13}C$ and $\delta^{15}N$ in the organic matter, sediment samples were decarbonated with 0.5M HCl and rinsed with distilled water. The decarbonated samples were sealed in a quartz tube under vacuum along with CuO, silver fillings and O_2 free high purity Cu wire. After evacuation, sealed tubes were heated at 850°C for nearly 8-10 hours. The evolved CO_2 and N_2 were cryogenically separated and further purified by passing it through a U-tube containing Cu wire at 650°C and collected in a sample-bottle containing molecular sieve. The purified CO_2 fraction was purified for moisture using a cold trap (-90°C) and analyzed for $^{13}C/^{12}C$ ratio using a GEO-2020 mass spectrometer. N_2 was analysed for isotopic ratio on a VG 903 mass spectrometer. Accuracy and precision of the isotopic data were checked using international standard (IAEA-N_2) and replicate analyses of laboratory standard (UBC-ACE; Acetanilide) obtained from the University of British Columbia (Agnihotri, 2001). Carbon and nitrogen isotopic ratios of sedimentary organic matter are expressed as δ values ($\delta^{13}C_{org}$ and $\delta^{15}N$) with reference to V-PDB and atmospheric N_2, respectively. The uncertainties in $\delta^{13}C_{org}$ and $\delta^{15}N$ determinations are ~0.2‰ and 0.3‰, respectively (Agnihotri, 2001).

Major and Trace Elemental Analyses

For major and trace element analyses, the sediment samples were dried and thoroughly homogenized. The samples were digested following conventional HF-$HClO_4$-HNO_3-HCl procedure and brought to solution in 1M HCl medium. The samples were suitably diluted for the elemental measurements using an ICP-AES (*Jobin-Yvon, Model 38S*). The USGS rock standards W-2 and MAG-1 were analyzed to check accuracy and precision of the elemental analyses of sediment samples. Among them, MAG-1 (marine mud) was analyzed extensively in this work as it closely matches with the matrix of sediment samples. In addition, an internal laboratory standard NOVA was prepared and standardized during the analyses of sediment samples. NOVA is a deep-sea clay sediment collected from the North Pacific (NOVA III -13; 3° 55.6' N, 178° 47.3' W) at water depth of 5351 m (Amin et al., 1972). Based on repeat measurements of various check standards, the coefficient of variation was about 2% for major elements and ranged from 3-5% for trace elements (Agnihotri, 2001).

Strontium (Sr) and Neodymium (Nd) Isotopic Measurements

To determine the Sr and Nd isotopic values of the silicate fraction of the sediments, ~1 g samples were decarbonated by leaching it with 0.6M HCl at 80°C with intermittent ultrasonification to remove the carbonate fractions. Further, the samples were heated at 550°C in a furnace to remove the organic material. About 100 mg of the carbonate and organic matter free sediments were transferred to teflon vials and brought to complete solution using ultra-pure HF and HNO_3 (Seastar Chemicals) at ~ 130-140°C in presence of ^{84}Sr and ^{150}Nd tracers. Sr and REEs were separated from the solution following standard ion exchange procedure (Rai and Singh 2007; Singh et al. 2008). From the REE fraction, Nd was separated using HDEHP coated Teflon powder. Sr and Nd concentrations and their $^{87}Sr/^{86}Sr$ and $^{143}Nd/^{144}Nd$ were measured on an Isoprobe-T Thermal Ionization Mass Spectrometer in static multi-collection mode. Mass fractionation corrections for Sr and Nd were made by normalizing $^{86}Sr/^{88}Sr$ to 0.1194 and $^{146}Nd/^{144}Nd$ to 0.7219 respectively. During the course of analyses, SRM987 Sr and JNdi Nd standards were repeatedly measured, these yielded values of 0.710233±0.000012 (1σ, $n = 48$) for $^{86}Sr/^{88}Sr$ and 0.512106±0.000006 (1σ, $n = 7$) for $^{143}Nd/^{144}Nd$ respectively, well within the recommended values. Several Sr and Nd total procedural blanks were measured along with the samples; typical blank for Sr was ~1.6 ng whereas for Nd it was ~300 pg. These blanks are several orders of magnitude lower than the typical total Sr (~7 mg) and Nd (~2 mg) loads analyzed and hence no corrections for blanks were made. Few duplicate samples were processed to check the reproducibility of results.