Stable carbon and oxygen isotope study on benthic foraminifera: Implication for microhabitat preferences and interspecies correlation

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Stable isotopes of benthic foraminifera have widely been applied in micropalaeontological research to understand vital effects in foraminifera. Isotopic fractionations are mainly controlled by ontogeny, bottom/pore water chemistry, habitat preference, kinetic effect and respiration. Discontinuous abundance of a species for isotopic analysis has forced us to select multiple species from down-core samples. Thus standardisation factors are required to convert isotopic values of one species with respect to other species. The present study is pursued on isotopic values of different pairs of benthic foraminifera from the Krishna–Godavari basin and Peru offshore to understand habitat-wise isotopic variation and estimation of isotopic correction factors for the paired species (*Cibicides wuellerstorfi*–*Bulimina marginata*, *Ammonia* spp.–*Loxostomum amygdalaeformis* and *Bolivina seminuda*–*Nonionella auris*). Infaunal species (*B. marginata*, *Ammonia* spp. and *N. auris*) show a lighter carbon isotopic excursion with respect to the epifaunal to shallow infaunal forms (*C. wuellerstorfi*, *L. amygdalaeformis* and *B. seminuda*). These lighter $\delta^{13}\text{C}$ values are related to utilisation of CO_2 produced by anaerobic remineralisation of organic matter. However, enrichment of $\delta^{18}\text{O}$ for the deeper microhabitat (bearing lower pH and decreased CO_3^{2-}) is only recorded in case of *B. marginata*. It is reverse in case of *N. auris* and related to utilisation of respiratory CO_2 and internal dissolve inorganic carbon pool. Estimation of interspecies isotopic correction factors for the species pairs ($\delta^{13}\text{C}$ of *C. wuellerstorfi*–*B. marginata*, *L. amygdalaeformis*–*Ammonia* spp., *N. auris*–*B. seminuda*) and $\delta^{18}\text{O}$ of *C. wuellerstorfi*–*B. marginata* are statistically reliable and may be used in palaeoecological studies.

Keywords. Benthic foraminifera; oxygen and carbon isotopes; microhabitat; isotopic correction factors.

1. Introduction

Foraminifera, a group of unicellular eukaryotic protozoa, have widely been used in palaeoclimatic

and palaeoceanographic reconstructions. Most of the foraminiferal taxa bear calcitic tests (except agglutinated and organic walled varieties) and

test-morphology itself records their preferred environmental conditions (Corliss 1985). The stable carbon (^{12}C and ^{13}C) and oxygen (^{16}O and ^{18}O) isotopes present within the calcitic tests of foraminifera are widely used as key proxies for palaeoenvironmental interpretations (Shackleton 1974, 1977; Rathburn *et al.* 1996; Moodley *et al.* 2000; Cooke and Rohling 2001; Zachos *et al.* 2001; Fontanier *et al.* 2006; Ravelo and Hillaire-Marcel 2007; Bhaumik *et al.* 2011; Ishimura *et al.* 2012). The use of stable oxygen and carbon isotope values of foraminiferal tests was strengthened by the pioneering efforts of Urey (1947), McCrea (1950), Epstein *et al.* (1953), Emiliani (1955) and Shackleton and Opdyke (1973). These landmark studies revealed that the $\delta^{18}\text{O}$ signal mainly reflects sea water temperature and fluctuation of global ice volume whereas the $\delta^{13}\text{C}$ value reflects palaeoproductivity and age of the water-mass (Shackleton and Opdyke 1973; Shackleton 1977; Zachos *et al.* 2001). However, several recent studies demonstrated that benthic foraminiferal oxygen isotope is also influenced by deep water hydrographic changes and thus might not always be used as a good indicator for ice volume estimation (Shackleton 2000; Skinner and Shackleton 2006; Elderfield *et al.* 2012; Rohling *et al.* 2014). Both carbon and oxygen isotopes may be measured on different foraminiferal carbonate species (calcite and aragonite). It is believed that fractionation of oxygen and carbon isotopes from sea/pore water to biogenic carbonate are controlled by thermodynamics and $\delta^{13}\text{C}$ of dissolved inorganic carbon/carbonate in sea/pore water, respectively (Cooke and Rohling 2001; Fontanier *et al.* 2006; Hoogakker *et al.* 2010; Ishimura *et al.* 2012).

Isotope values of benthic foraminifera (bottom dwellers) are important tool to understand past global climate as well as their preferred microhabitat. Studies suggest that deviation in equilibrium of isotopes in foraminiferal carbonates from the sea water (vital effect) may be biased by the effects of ontogeny, respiration, symbiont photosynthesis, microhabitat, climatic variability, bottom and pore water temperature, pore water chemistry, diagenesis, pH of water, and changes in CO_3^{2-} (Shackleton 1974; Cooke and Rohling 2001; Saraswati *et al.* 2004; Fontanier *et al.* 2006; Schmiedl and Mackensen 2006; Hoogakker *et al.* 2010; Ishimura *et al.* 2012; Marchitto *et al.* 2014). Some species of benthic foraminifera prefer to live within the uppermost centimetre of sediments (epifaunal), some

within the top two centimetres (shallow infaunal) and some below two centimetres of sediments (deep infaunal) (Corliss 1991; Jorissen *et al.* 1995). Differences in their habitation as well as their generic divisions cause disequilibria in isotopic fractionation within their biogenic calcitic tests with respect to sea water (Rathburn *et al.* 1996; Fontanier *et al.* 2006; Hoogakker *et al.* 2010; Ishimura *et al.* 2012). This disequilibrium in isotopic fractionation in benthic foraminifera is mainly controlled by the kinetic and metabolic effects, pore water chemistry, food supply and pH of the sea/pore waters (McConnaughey 1989; Cooke and Rohling 2001; Fontanier *et al.* 2006; Hoogakker *et al.* 2010; Ishimura *et al.* 2012).

Numerous studies have been undertaken for the interspecies conversion of isotopic values by adding or subtracting vital offsets (Shackleton 1974; Graham *et al.* 1981; Katz *et al.* 2003; Fontanier *et al.* 2006; Mackensen 2008; Ishimura *et al.* 2012; Marchitto *et al.* 2014). This conversion is known as standardization of isotopic values of one species with respect to another. Standardization is required when a selected species for isotopic analysis does not continuously show down core abundance. In such a case, isotopic analysis of two or more other species are required and need standardization with respect to the earlier selected species. Such standardization of isotopic values with sea water or other species will give us a continuous down core isotopic record and help us to infer palaeoceanography and pore water chemistry. In this way, the isotopic values of *Cibicides wuellerstorfi*, *Oridorsalis umbonatus* and *Uvigerina peregrina* are commonly used in palaeoclimate study (Shackleton 1974; Woodruff *et al.* 1980; Bemis *et al.* 1998; Haug and Tiedemann 1998; Fontanier *et al.* 2006; Hoogakker *et al.* 2010; Bhaumik *et al.* 2011).

The present study is pursued on the carbon and oxygen stable isotopic values of paired species *Bulimina marginata*–*Cibicides wuellerstorfi*, *Ammonia* spp.–*Loxostomum amygdalaeformis* (from NGHP-Hole 10D, Krishna–Godavari Basin, Bay of Bengal) and *Bolivina seminuda*–*Nonionella auris* (from ODP Hole 680B, Peru offshore margin) to estimate isotopic offset (vital effect) as well as isotopic adjustment (standardisation) of paired species. Furthermore, comparison among the paired species isotopic values taken from the same down-core samples will enable to infer the microhabitat preference of one species with respect to the other.

2. Location and oceanographic settings

National Gas Hydrate Program (NGHP) Site-10D (15°51.8609'N, 81°50.0749'E; water depth 1038 m) is located ~36 km off the eastern coast of India in the Krishna–Godavari (K–G) Basin within the Bay of Bengal, Indian Ocean (Collett *et al.* 2007, figure 1). A total of 97 core samples were analysed from 200 m long core of Quaternary age. Seismic study together with pore water salinity and core sediment observations showed that this site has maximum gas hydrate saturation between 27 and 160 m below sea floor (mbsf) (Collett *et al.* 2007).

The east Indian coastal current (EICC) flows along the eastern coast of India flooding up to a depth of ~300 m (Mukherjee *et al.* 2014). The mid-to-deep water column of the Indian Ocean can be divided vertically into a number of water masses; the Antarctic Intermediate Water (AAIW) up to ~1200 m (Tchernia 1980), a mixture of well oxygenated North Atlantic Deep Water (NADW) and North Indian Deep Water or NIDW between 1200 and 2000 m (Wyrski 1971; Tchernia 1980; GEOSECS 1983; Bhaumik *et al.* 2014) and the Antarctic Bottom Water (AABW), which is cold, nutrient-rich and lies below 3000 m (Tchernia 1980;

Bhaumik *et al.* 2014). The water mass between 2000 and 3000 m is a mixture of NADW and AABW (De and Gupta 2010; Bhaumik *et al.* 2014).

Ocean Drilling Program (ODP) Hole 680B, Leg 112 (11°03.90'S, 78°04.67'W, water depth 252.5 m) is located in the Salaverry Basin, off Peru continental margin of the east Pacific Ocean (Suess *et al.* 1988; figure 2). A total of 300 samples (late Pliocene to Recent) were analysed, out of which only 119 samples are productive in terms of foraminiferal population. The Peru continental margin or Andean continental margin is configured by both tectonic activity and coastal upwelling (Suess *et al.* 1988). The Peru Current and the Peru Counter Current are the two oceanic currents that dominate over the studied site (Suess *et al.* 1988; Oberhänsli *et al.* 1990). The Peru continental margin represents one of the best coastal upwelling regimes in the world and is rich in organic carbon (Suess *et al.* 1988).

3. Datasets and methods

Foraminiferal separation from the core samples was carried out following the technique described

Figure 1. Location map of NGHP Hole 10D in the Krishna–Godavari Basin, Bay of Bengal (modified from Collett *et al.* 2007).

Figure 2. Location map of ODP Hole 680B, offshore Peru (Suess *et al.* 1988).

in Bhaumik *et al.* (2011). Specimens of benthic foraminiferal species *Cibicides wuellerstorfi*, *Bulimina marginata*, *Ammonia* spp. (including *A. beccarii*, *A. tepida*, *A. perkinsoniana* and two other unidentified forms resembling *Ammonia*) and *Loxostomum amygdalaeformis* were taken from fraction range between 125 and 300 μm from NGHP Hole 10D, K–G Basin (figure 3). Specimens of *Ammonia beccarii* are considered as primary species for isotopic analysis to minimise possibility of inter species variation within *Ammonia* group. However few specimens of other species of *Ammonia* (*A. tepida*, *A. perkinsoniana*) were also picked in some cases to achieve minimum requirement of specimens for isotopic analysis. None of the specimens contain any earthy colour or polished or abrasion/worn out indicating no signature of transportation or reworking. At least eight specimens of each species were picked for isotopic analysis and cleaned in an ultrasonic bath using methanol. All the required species (*C. wuellerstorfi*, *B. marginata*, *Ammonia* spp. and *L. amygdalaeformis*) are not documented in each down core samples. Specimens of different species were picked based on their availability in each down core sam-

ples. In this way specimens of *B. marginata* and *C. wuellerstorfi* show their coexistence only in 10 down-core samples (Appendix) out of total 97 samples. Similarly 19 down-core samples documented coexistence of *Ammonia* spp. and *L. amygdalaeformis* (Appendix).

Approximately 30 specimens of *Bolivina seminuda* and 18 specimens of *Nonionella auris* were picked from the same fraction size (between 125 and 300 μm) from ODP Hole 680B, Peru offshore (figure 3). Coexistence of these species with significant numbers for isotopic analysis are observed only in 27 down-core samples (Appendix). All the specimens were ultrasonically cleaned with methanol prior to isotopic analysis. No transported/reworked specimens were used for isotopic analysis.

Stable carbon and oxygen isotopic analyses of benthic species from the K–G Basin were carried out at the Mass Spectrometry Laboratory of the Indian Institute of Technology, Kharagpur. Isotopic compositions of foraminiferal carbonates were measured using a continuous flow Delta^{PLUS} XP stable isotope ratio mass spectrometer equipped with an automated carbonate preparation system

Figure 3. SEM images of foraminiferal species used for isotopic analysis. Bars represent 100 μm size. 1–2: *Ammonia beccarii*, 3–4: *Ammonia perkinsoniana*, 5–8: *Ammonia tepida*, 9–12: *Ammonia* spp., 13–14: *Bulimina marginata*, 15–17: *Bolivina seminuda*, 18–20: *Cibicides wuellerstorfi*, 21–22: *Loxostomum amygdalaeformis*, 23–25: *Nonionella auris*.

(Gas-Bench-II). Around 200 μg sample was transferred into glass vials and flushed with 99.999% pure He gas. Samples were reacted with 100% H_3PO_4 at 72°C. The released CO_2 was purified by removing water and other trace gases and isotopic ratios were measured. A routine precision of $\pm 0.1\text{‰}$ (2σ) for both $\delta^{13}\text{C}$ and $\delta^{18}\text{O}$ was monitored by analysing two in-house CaCO_3 standards as Z-Carrara (procured from Physical Research Laboratory, Ahmedabad) and BDH (procured from University College London) calibrated against NBS-19.

Samples from the Peru offshore zone were analysed in a Stable Isotope Ratio Mass Spectrometer (Delta V plus model from Thermo Fisher) at the Wadia Institute of Himalayan Geology, Dehradun, following the same procedure as described above. For day-to-day measurement, secondary laboratory standards were used which were calibrated against NBS-18. Repeat measurements of the secondary laboratory standards show a precision and accuracy better than $\sim 0.1\text{‰}$ (1SD) for carbon and oxygen isotope measurements. All isotopic results from both the sites are expressed in terms of parts per mille (‰) normalised to VPDB standards.

The ordinary least-squared (OLS) regression method was performed to estimate the isotopic offsets between each paired species. The OLS regression method is applied on the isotopic datasets to estimate the true population relationship between a continuous response variable (y) and a continuous explanatory variable (x) as the datasets are small and lie within control limits. A similar study has earlier been carried out by Katz *et al.* (2003) on isotopic values of some early Palaeogene benthic foraminifera. Katz *et al.* (2003) used the least median of squares (LMS) based on the method of moment (MM) estimator because they used large datasets in their study.

The OLS regression is a generalised linear modelling technique that is used to model a single response variable which has been recorded on at least an interval scale. At a very basic level, the relationship between a continuous response variable (y) and a continuous explanatory variable (x) may be represented using a line of best-fit, where y is predicted, at least to some extent, by x (Ryan 1997).

4. Results

In case of the species pair *C. wuellerstorfi*–*B. marginata*, both the carbon and oxygen isotopes show reasonably positive correlation ($R = 0.57$

and $R = 0.64$, respectively) with relatively low p-values (figure 4, table 1). Reasonably positive correlation ($R = 0.60$) with significant low p-values ($p = 0.007$) for $\delta^{13}\text{C}$ is observed in the pair *Ammonia* spp.–*L. amygdalaeformis* (figure 4, table 1). But the $\delta^{18}\text{O}$ values for the same pair do not show good correlation (figure 4, table 1). A good correlation ($R = 0.76$) with very low p-value is also observed for $\delta^{13}\text{C}$ of the species pair *B. seminuda*–*N. auris*, but the same pair does not show good correlation ($R = 0.17$) for $\delta^{18}\text{O}$ (figure 4, table 1).

The downcore variations of isotope data for all the above-mentioned paired species (*C. wuellerstorfi*–*B. marginata*, *Ammonia* spp.–*L. amygdalaeformis* and *B. seminuda*–*N. auris*) are also considered for comparison (figure 5). *Cibicides wuellerstorfi* reliably records $\delta^{13}\text{C}$ of dissolved inorganic carbon in sea water as it precipitates its test close to the oxygen isotopic equilibrium of sea water with a constant offset of 0.64‰ (Shackleton *et al.* 1984; Cooke and Rohling 2001; Katz *et al.* 2003). Thus addition of 0.64‰ with the $\delta^{18}\text{O}$ values of *C. Wuellerstorfi* (i.e., adjusted values) gives the oxygen isotopic values of sea water (figure 5). However specimens of *C. wuellerstorfi* is not recorded at Hole 680B.

Derived equations obtained by the OLS regression methods are presented with R and p-values in table 1 for the prediction of estimated isotopic offsets of the paired species. The table exhibits the degree of percentage variations of data for the pairs *C. wuellerstorfi*–*B. marginata* (for $\delta^{13}\text{C}$ and $\delta^{18}\text{O}$), *Ammonia* spp.–*L. amygdalaeformis* (only for ^{13}C), and *B. seminuda*–*N. auris* (only for ^{13}C) are $>55\%$. The p-values for all the above cases show that the paired datasets are statistically significant at 90% confidence interval. The derived OLS regression equations for the paired species $\delta^{18}\text{O}_{\text{Ammonia spp.}} - \delta^{18}\text{O}_{\text{L. amygdalaeformis}}$ and $\delta^{18}\text{O}_{\text{B. seminuda}} - \delta^{18}\text{O}_{\text{N. auris}}$ show less percentage variation of data with high p-values.

5. Discussion

5.1 Factors for fractionation of foraminiferal carbon and oxygen isotopes

Isotopic analyses of each paired species were performed on specimens picked from the same sediment samples (same depth for each pair of data). Thus it is presumed that the effect of climate variability, bottom and pore water temperatures, pore

Figure 4. Scatter plots of isotopic values for the paired species *Cibicides wuellerstorfi*–*Bulimina marginata*, *Ammonia* spp.–*Loxostomum amygdalaeformis* and *Bolivina seminuda*–*Nonionella auris*. Correlation and p-values are given within each panel for every pair. Scatter $\delta^{18}\text{O}$ plots of *Ammonia* spp.–*Loxostomum amygdalaeformis* and *Bolivina seminuda*–*Nonionella auris* do not show significant correlation and hence are not considered for OLS regression analysis.

Table 1. Equations with dependent and independent variables as derived by OLS regression analysis.

Variable (y)	Variable (x)	N	Regression equation y on x	Regression equation x on y	R	p-value
$\delta^{13}\text{C}$ <i>C. wuellerstorfi</i>	$\delta^{13}\text{C}$ <i>B. marginata</i>	10	$y = 0.75x + 0.7$	$x = 0.43Y - 0.94$	0.57	0.0868
$\delta^{18}\text{O}$ <i>C. wuellerstorfi</i>	$\delta^{18}\text{O}$ <i>B. marginata</i>	10	$y = 0.66x - 0.29$	$x = 0.61Y + 1.5$	0.64	0.0476
$\delta^{13}\text{C}$ <i>Ammonia</i> spp.	$\delta^{13}\text{C}$ <i>L. amygdalaeformis</i>	19	$y = 0.39x - 0.96$	$x = 0.91Y + 1.1$	0.60	0.007
$\delta^{13}\text{C}$ <i>B. seminuda</i>	$\delta^{13}\text{C}$ <i>N. auris</i>	26	$y = 0.5x - 0.25$	$x = 1.1x - 1.2$	0.76	0.000

R and p-values are included in the table to understand statistical significance of the dataset. The first regression equation predicted the value of y when x is known and the second regression equation predicted the value of x when y is known.

water chemistry, pH of water and diagenesis will be equally effective for both the paired species picked from single depth. Variations in microhabitat (epifaunal, infaunal and deep infaunal), ontogeny and respiration effects are considered as important factors influencing the fractionation of isotopes of each paired species (Cooke and Rohling 2001; Ishimura *et al.* 2012). Studies showed that ontogenic effect is more intense during juvenile stages (Moberly 1968; Berger *et al.* 1978). Relatively larger size fraction (125–300 μm) bearing adult forms are used in this study. Thus it may be assumed that ontogenic effect is minimum in used specimens.

Fractionation of carbon isotope in benthic foraminiferal calcitic test mainly depends on the food habits and microhabitat within the sediments (Fontanier *et al.* 2006; Mackensen 2008). It is already suggested that decomposition of sedimentary organic matter along the down core is responsible for the depletion in $\delta^{13}\text{C}$ within pore water. This isotopic depletion is reflected by the infaunal to deep-infaunal benthic foraminifera calcified in pore water (Fontanier *et al.* 2006; Mackensen 2008). However, the epifaunal taxa reflect the bottom water isotopic composition and calcified in equilibrium with the ocean water (Bemis *et al.* 1998; Fontanier *et al.* 2006). Thus richness of organic carbon within the sediments and its intake by foraminifera is mirrored by the carbon isotopic fractionation. CO_2 and dissolved inorganic carbonate (DIC) within cell of foraminifera produced during metabolism are used for test calcification and thus shows a direct relation in fractionation of carbon isotopes (de Nooijer *et al.* 2009).

There are two school of thoughts regarding the biasness of $\delta^{13}\text{C}$ in foraminiferal calcitic test in methane rich environment. One group of workers suggests that presence of methane may cause depletion in foraminiferal $\delta^{13}\text{C}$ in a tune of -5 to -55‰ (Hill *et al.* 2003, 2004; Barbieri and Panieri 2004;

Martin *et al.* 2007) whereas others proposed foraminiferal distribution as well as foraminiferal carbon isotopes are unbiased by the methane seepages (Torres *et al.* 2003; Lobegier and Sen Gupta 2008). None of the specimens used in this study bears such depleted $\delta^{13}\text{C}$ values and thus it may be considered that the faunal assemblages are unaffected by methane seepage. Studies showed that shallow infaunal to deep infaunal species generally bear depleted $\delta^{13}\text{C}$ values at the tune of -2 to -4‰ (Ravelo and Hillaire-Marcel 2007; Mackensen 2008).

On the other hand, fractionation of oxygen isotope in foraminiferal calcitic test depends on numerous factors such as kinetic effect (temperature dependent), ontogenesis, symbiosis, photosynthesis, respiration, gametogenesis and change in pH (Lane and Doyle 1956; Grossman 1987; Wefer and Berger 1991; Spero and Lea 1996; Hoogakker *et al.* 2010; Ishimura *et al.* 2012; Marchitto *et al.* 2014). Studies showed that $\delta^{18}\text{O}$ enrichment in calcitic tests of benthic foraminifera is also directly linked to the depth preference owing to the kinetic effect (Bemis *et al.* 1998; Hoogakker *et al.* 2010; Marchitto *et al.* 2014).

5.2 Microhabitats of paired species

5.2.1 Paired *Cibicides wuellerstorfi*–*Bulimina marginata*

Bulimina marginata has been reported from surface samples of the North Sea (Jarke 1961; Gabel 1971), whereas some workers inferred that these species prefer to live within the top 1 cm of sediments (Stefanelli *et al.* 2005; Murray 2006; Basak *et al.* 2009). Study of Brückner and Mackensen (2008) showed that low mean $\delta^{13}\text{C}$ values of *B. marginata* reflect a shallow infaunal habitat of this species. Carbon isotopic distribution pattern of both the species (*C. wuellerstorfi*–*B. marginata*) in the present study shows almost

parallelbreak relationship throughout the studied interval with lighter $\delta^{13}\text{C}$ values of *B. marginata* (figure 5). Lighter $\delta^{13}\text{C}$ values of *B. marginata* than *C. wuellerstorfi* (figure 5), thus indicate infaunal habitat in organic carbon rich environment. The depletion in $\delta^{13}\text{C}$ of *B. marginata* could be explained by release of ^{13}C depleted CO_2 owing to high anaerobic remineralisation rate of organic materials at depth (Cooke and Rohling 2001; Fontanier *et al.* 2006; Hoogakker *et al.* 2010). The released ^{13}C depleted CO_2 then used to build calcitic tests of this species (de Nooijer *et al.* 2009) which in-turn is documented by lighter $\delta^{13}\text{C}$ values.

Seven out of ten $\delta^{18}\text{O}$ values of *B. marginata* show very close relationship with adjusted $\delta^{18}\text{O}$ values of *C. wuellerstorfi* (equilibrated with sea water). The small deviations in $\delta^{18}\text{O}$ indicate that *B. marginata* calcified its test almost in equilibrium with the sea water (figure 5). However, the deviations in $\delta^{18}\text{O}$ are more when compared with unadjusted oxygen isotopic values of *C. wuellerstorfi*. This enrichment in $\delta^{18}\text{O}$ of *B. marginata* is also indicative of infaunal habitat. Bemis *et al.* (1998) observed mild $\delta^{18}\text{O}$ enrichment in infaunal *Uvigerina* than epifaunal *Cibicidoides* owing to low pore-water pH and decreased CO_3^{2-} in deeper sediments. Study of Spero *et al.* (1997) also inferred that $\delta^{18}\text{O}$ in foraminiferal carbonate decreases with increasing CO_3^{2-} .

5.2.2 Paired *Ammonia* spp.–*Loxostomum amygdalaeformis*

Studies showed that *Ammonia* group of species prefer infaunal microhabitat (Debenay *et al.* 1998; Murray 2006). On the other hand habitat preference of *L. amygdalaeformis* is not well established. However, shape of these species reflects shallow infaunal habitat (Bernhard *et al.* 1997; Hottinger 2000). Comparison between carbon isotopic plot of *Ammonia* spp. and *L. amygdalaeformis* shows significant difference in their $\delta^{13}\text{C}$ values (figure 5). Depleted carbon isotopic values of *Ammonia* spp. as compared to *L. amygdalaeformis* thus clearly indicate its infaunal habitat (preference to greater depth). This depletion in $\delta^{13}\text{C}$ of *Ammonia* may be related to high anaerobic remineralisation rate of organic materials as well as utilisation of released CO_2 to build calcitic tests as stated in case of pair *C. wuellerstorfi*–*B. marginata*.

A comparison of oxygen isotope values of both the species reveals that $\delta^{18}\text{O}$ of *Ammonia* shows

enrichment in 11 points, depletion in 3 points and bears almost similar values in 5 points (figure 5). Thus oxygen isotope values do not provide a clear picture about the difference in their microhabitat.

5.2.3 Paired *Bolivina seminuda* and *Nonionella auris*

In general, *B. seminuda* is considered as an epifaunal to shallow infaunal species (Gooday *et al.* 2000; Duijnste 2001) whereas *N. auris* is considered to be deep-infaunal (Corliss and Emerson 1990; Wefer *et al.* 1994). Continuous $\delta^{13}\text{C}$ plot of *N. auris* and *B. seminuda* shows that *N. auris* always bear lighter $\delta^{13}\text{C}$ values than *B. seminuda* within the sequence (figure 5). Depletion in $\delta^{13}\text{C}$ thus indicates deep-infaunal habitat of *N. auris* than *B. seminuda* which supports the observations of earlier researchers who assigned deep infaunal habitat to *N. auris* (Corliss and Emerson 1990; Wefer *et al.* 1994). As stated earlier, depletion in $\delta^{13}\text{C}$ of *N. auris* could be related to release ^{13}C depleted CO_2 owing to anaerobic remineralisation of organic matter (Cooke and Rohling 2001; Fontanier *et al.* 2006; Hoogakker *et al.* 2010) and intake of these CO_2 to build the test of this species (de Nooijer *et al.* 2009).

Previous works and this study show that *N. auris* prefers more depth (deep infaunal) than *B. seminuda* (Corliss and Emerson 1990; Wefer *et al.* 1994; Gooday *et al.* 2000; Duijnste 2001). Studies also showed that enrichment in $\delta^{18}\text{O}$ of benthic foraminiferal tests increases with depth owing to the change in kinetic effect (Bemis *et al.* 1998; Hoogakker *et al.* 2010; Marchitto *et al.* 2014). Thus it is expected that infaunal *N. auris* should bear enriched $\delta^{18}\text{O}$ values than *B. seminuda*. However, comparison in $\delta^{18}\text{O}$ values of *N. auris* with respect to *B. seminuda* shows depletion instead of enrichment in most of the depth points (figure 5). Thus, the depletion in $\delta^{18}\text{O}$ of *N. auris* with respect to *B. seminuda* is unexpected and does not relate to its microhabitat preference. Rather it may be linked to preference of food and oxygen. *N. auris* prefers to live in high organic flux environment (Wefer *et al.* 1994; Rathburn *et al.* 2000) than *B. seminuda*, the latter is more influenced by oxygen (Sen Gupta and Machain-Castillo 1993). Thus it may be presumed that larger body volume bearing *N. auris* requires more utilisation of food to maintain metabolic activity and produced isotopically lighter oxygen (^{16}O) bearing more CO_2 as a by-product of metabolism. This generated metabolic

Figure 5. Comparison in continuous plots of carbon and oxygen isotopic values for the paired species *Cibicides wuellerstorfi*–*Bulimina marginata*, *Ammonia* spp.–*Loxostomum amygdalaeformis* and *Bolivina seminuda*–*Nonionella auris*. Plots of adjusted δ¹⁸O of *C. wuellerstorfi* to sea water is presented by red colour.

Table 2. Isotopic correction factors for paired species.

Correction factors when <i>y</i> is dependent variable	Correction factors when <i>x</i> is dependent variable
$\delta^{13}\text{C}$ of Bm = $(\delta^{13}\text{C}$ of Cw - 0.7)/0.75	$\delta^{13}\text{C}$ of Cw = $(\delta^{13}\text{C}$ of Bm + 0.94)/0.43
$\delta^{18}\text{O}$ of Bm = $(\delta^{18}\text{O}$ of Cw + 0.29)/0.66	$\delta^{18}\text{O}$ of Cw = $(\delta^{18}\text{O}$ of Bm - 1.5)/0.61
$\delta^{13}\text{C}$ of La = $(\delta^{13}\text{C}$ of Am + 0.96)/0.39	$\delta^{13}\text{C}$ of Ab = $(\delta^{13}\text{C}$ of La - 1.1)/0.91
$\delta^{13}\text{C}$ of Na = $(\delta^{13}\text{C}$ of Bs + 0.25)/0.5	$\delta^{13}\text{C}$ of Bs = $(\delta^{13}\text{C}$ of Na + 1.2)/1.1
It is important to note that equation for conversion of isotopic values of one species to other species and <i>vice versa</i> are different as regression coefficients are different. La = <i>L. amygdalaeformis</i> , Am = <i>Ammonia</i> spp., Bm = <i>B. marginata</i> , Cw = <i>C. wuellerstorfi</i> , Na = <i>N. auris</i> and Bs = <i>B. seminuda</i> .	

CO₂ with internal DIC pool together might be used to build the test of *N. auris* (Lane and Doyle 1956; Grossman 1987; de Nooijer *et al.* 2009). Thus the utilisation of isotopically lighter oxygen and carbon isotopes together leads to the depletion in $\delta^{18}\text{O}$ as well as $\delta^{13}\text{C}$ in the test of *N. auris*.

5.3 Interspecies isotopic correction factors (vital effects)

It is considered that the isotopic offset of a species with respect to equilibrium or other species is constant through time (Shackleton 1974; Woodruff *et al.* 1980; Marchitto *et al.* 2014). Each and every species has its own preferred microhabitat, ontogeny, and respiration rate which probably deviates the fractionation of isotopes from equilibrium (Cooke and Rohling 2001). Isotopic values of species like *Cibicides wuellerstorfi*, *Oridorsalis umbonatus* and *Uvigerina peregrina* are widely used for palaeoceanographic reconstruction. Paucity of some of these species forced us to determine the isotopic values of other species for palaeoceanographic reconstruction, and hence there is a need to estimate isotopic offset of other species with some known species for better understanding of palaeoceanography. OLS regression study is pursued on both $\delta^{13}\text{C}$ and $\delta^{18}\text{O}$ values of paired species *C. wuellerstorfi*–*B. marginata* and the correction factors are presented in table 2. However, the OLS regression study is only pursued on $\delta^{13}\text{C}$ data of pair species *Ammonia* spp.–*L. amygdalaeformis* and *B. seminuda*–*N. auris* and related correction factors are tabulated in table 2. The OLS regression study is not performed on other cases as the correlation values are low.

This study shows difference in oxygen isotopic correction factor from the estimation of Katz *et al.* (2003) for the pair species *C. wuellerstorfi*–*B. marginata* (table 2). Katz *et al.* (2003) show an adjustment relationship for oxygen isotope as $\delta^{18}\text{O}$

of *Bulimina* spp. $-0.28 = \delta^{18}\text{O}$ *Cibicidoides* spp. whereas present data show the factor as $(\delta^{18}\text{O}$ of *B. marginata*–1.5)/0.61 = $\delta^{18}\text{O}$ *C. wuellerstorfi*. This study is performed on relatively less number of samples ($N = 10$) and thus scope of further investigation still exists. This study also suggests that test calcification of *B. marginata* is almost in equilibrium with sea water as observed by Shackleton *et al.* (1984) whereas other species considered in this study do not show such equilibrium with sea water.

6. Conclusions

Study from the Krishna–Godavari Basin and offshore Peru shows that benthic foraminiferal species *Bulimina marginata*, *Ammonia* spp., and *Nonionella auris* prefer greater depth (infaunal) whereas *Cibicides wuellerstorfi*, is raised epifaunal, and *Loxostomum amygdalaeformis* and *Bolivina seminuda* are epifaunal to shallow infaunal in nature. Fractionation of carbon isotope in these foraminiferal tests is related to depth of microhabitat as well as incorporation of ^{13}C depleted CO₂ released by anaerobic remineralisation of organic materials. However, oxygen isotope fractionation shows direct relation with depth preference for the pair species *C. wuellerstorfi*–*B. marginata*. Depleted $\delta^{18}\text{O}$ of *N. auris* (infaunal) indicates more utilisation of metabolic CO₂ to build test of *N. auris*.

Estimated isotopic correction factors obtained from the OLS regression methods for the species pairs ($\delta^{13}\text{C}$ and $\delta^{18}\text{O}$ of *C. wuellerstorfi*–*B. marginata*, $\delta^{13}\text{C}$ of *L. amygdalaeformis*–*Ammonia* spp., *N. auris*–*B. seminuda*) are statistically reliable as the degree of percentage variations of data are > 55%. The p-values for these paired datasets are also statistically significant at 90% confidence level and thus may be used as isotopic correction factors within the upper bathyal zone.

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Appendix

Table A1. Carbon and oxygen isotope dataset with sample depth used in this study.

Depth (mbsf)	$\delta^{13}\text{C}$ <i>C. wuellerstorfi</i>	$\delta^{13}\text{C}$ <i>B. marginata</i>	$\delta^{18}\text{O}$ <i>C. wuellerstorfi</i>	$\delta^{18}\text{O}$ <i>B. marginata</i>
15.36	−0.32	−1.13	2.62	3.97
16.86	−0.61	−1.57	0.12	4.22
31.64	0.04	−0.53	1.40	2.13
63.05	0.12	−0.62	2.40	3.32
110.46	0.38	−1.37	−1.44	−0.48
121.23	0.23	−0.47	0.06	3.05
146.11	0.79	−0.86	−0.16	0.18
149.11	0.67	−0.45	−0.40	−0.05
160.71	−0.68	−1.03	3.52	3.73
179.51	−0.59	−1.31	3.94	2.51

Depth (mbsf)	$\delta^{13}\text{C}$ <i>A. baccri</i>	$\delta^{13}\text{C}$ <i>L. amygdalaeformis</i>	$\delta^{18}\text{O}$ <i>A. beccarii</i>	$\delta^{18}\text{O}$ <i>L. amygdalaeformis</i>
88.43	−1.65	−0.29	0.29	−0.49
89.16	−1.73	−1.69	0.16	−1.01
93.38	−1.63	0.04	0.09	0.10
94.3	−1.08	−0.74	−0.10	−0.77
98.29	−0.59	−0.26	0.08	−0.26
101.01	−1.26	0.35	−0.74	−1.06
102.3	−0.31	0.59	−0.69	−1.29
109.11	−0.78	0.02	−0.77	−1.11
110.46	−0.38	0.37	−0.52	−2.05
111.96	−0.52	0.41	−0.84	−0.62
113.46	−0.33	0.56	−0.56	−0.64
117.21	−0.64	0.57	−1.37	−0.62
119.91	−0.41	1.26	−0.40	−0.29
123.49	−1.05	0.71	−0.89	−0.76
130.61	−0.74	−0.07	−0.58	−0.46
146.11	−0.57	0.64	−0.17	−0.80
147.61	−1.30	0.66	1.84	−1.90
149.11	−0.62	1.07	−0.62	−1.28
151.63	−0.60	1.25	−1.78	−0.40

Depth (common)	$\delta^{13}\text{C}$ <i>B. seminuda</i>	$\delta^{13}\text{C}$ <i>N. auris</i>	$\delta^{18}\text{O}$ <i>B. seminuda</i>	$\delta^{18}\text{O}$ <i>N. auris</i>
0.6	−0.47	−2.48	1.9	1.71
5.1	−1.8	−1.65	2.04	0.26
7.9	−1.68	−3.08	1.7	1.36
8.2	−4.02	−4.66	1.58	0.8
8.5	−2.59	−4.17	1.2	1.56
8.8	−2.48	−4.83	1.58	1.12
9.1	−2.32	−5.39	1.43	1.07
9.4	−1.71	−2.3	1.85	1.41
9.7	−1.73	−2.53	2.03	1.72

Table A1. (Continued.)

Depth (common)	$\delta^{13}\text{C}$ <i>B. seminuda</i>	$\delta^{13}\text{C}$ <i>N. auris</i>	$\delta^{18}\text{O}$ <i>B. seminuda</i>	$\delta^{18}\text{O}$ <i>N. auris</i>
13.9	−1.96	−3.33	1.13	1.34
16.8	−2.17	−2.62	1.8	1.07
17.7	−1.6	−1.67	2.1	1.69
18.3	−1.55	−2.34	1.53	0.11
18.6	−2.23	−2.66	1.37	1.35
22.2	−2.14	−4.56	1.18	0.79
22.5	−2.71	−3.42	1.47	1.15
22.8	−2.89	−5.55	1.46	1.37
26	−1.24	−1.36	1.79	1.11
26.3	−1.02	−2.39	1.65	0.3
26.6	−1.45	−3.77	1.66	0.81
26.9	−1.31	−2.17	1.25	0.91
27.8	−1.21	−2.4	1.64	−0.01
28.1	−1.14	−2.28	1.5	0.86
28.4	−0.9	−1.85	1.82	1.14
28.7	−0.95	−1.85	1.91	0.65
29	−0.81	−1.88	1.7	0.39
29.9	−0.92	−1.8	1.98	1.2

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