Interactive comment on “Variation in stable carbon and oxygen isotopes of individual benthic foraminifera: tracers for quantifying the vital effect” by T. Ishimura et al.

T. Ishimura et al.
toyoho@poplar.ocn.ne.jp

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My co-authors and I thank Dr. Fontanier for the constructive comments, which helped to greatly improve the quality of our manuscript. We took into account all the suggestions/comments when revising our paper. Our replies are as follows:

[Dr. Fontanier’s comment] The paper entitled “Variation in stable carbon and oxygen isotopes of individual benthic foraminifera: tracers for quantifying the vital effect” by Ishimura San et al. deals with the applicability of inter-individual delta13C and delta18O distributions (Standard Deviation within species) to reconstruct the bottom water isotopic signatures. This work is based on live and dead (assumed as modern) foraminifera that were sampled at 4 deep-sea sites, off Japan. Different species and genera, with different individual weights (i.e. size), were analyzed individually and their isotope signatures were compared with bottom water delta13CDIC and delta18Oe.c. Within a species, lower delta13C and delta18O values are recorded for smaller individuals. This is in agreement with other published works. Both Ddelta13C and Ddelta18O vary between taxa, what is also consistent with other publications. Finally, the authors show that the average intra-individual delta13C calculated for each species is correlated to the related Standard Deviation – when all species from a same area are plotted on the same graph, a simple equation (a x SDdelta13Cforam) + b = delta13Cforam) can be determined by a linear regression. Then, the authors observe that when SD = 0, the delta13CDIC (= b) is close to the bottom water delta13CDIC. Therefore, they propose that the average intra-individual delta13C and the related Standard Deviation may be relevant and reliable proxy to calculate bottom water delta13CDIC.

[Reply] We will add a taxonomic reference list and SEM pictures in the revised manuscript.

[Dr. Fontanier’s comment] General comments This paper is well written and well illustrated. It is based on a large data set of isotopic measurements that should be published in a peer-review journal. Those high-quality analyses were done on single individuals belonging to taxa which are quite common along the Pacific margin. As a modest taxonomist, I would recommend the authors to provide an appendix with taxonomic references for all taxa which were analyzed in this study. SEM pictures for all taxa would be also necessary for readers who would like to use related taxa for their own investigations.

[Reply] We will add a taxonomic reference list and SEM pictures in the revised manuscript.

[Dr. Fontanier’s comment] Now, when I deal with some interpretations proposed by the authors, I have got some concerns that should be clarified by the authors. For
instance, it seems that the authors have forgotten to use isotopes data of some species (Nonionella globosa, Nonionella labradorica) in figure 5 (in which interpretative linear regressions were drawn). According to me (I may be wrong!), adding those data (with low SD values) would change a large part of the interpretative story.

[Reply] Although we analyzed Nonionella globosa and Nonionella labradorica, there were not enough species analyzed (only two individuals) to discuss inter-individual isotopic variations. We listed the isotopic data merely as informative data (i.e., as “others”), but actually we did not discuss these isotopic data in our discussions. Therefore, we will remove the analytical data regarding Nonionella globosa and Nonionella labradorica from our revised manuscript.

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[Dr. Fontanier’s comment] Moreover, in some cases, the authors have measured isotopes at a genus level (Rutherfordoides and Stainforthia) without considering the potential inter-specific variability. Such a point should be addressed somewhere.

[Reply] We did not discuss the isotopic data on a genus level. Rutherfordoides and Stainforthia did not consist of multiple species (not spp.) but rather single species. We will add the taxonomic name at the species level in the revised manuscript.

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[Dr. Fontanier’s comment] Furthermore, the linear regressions which are proposed by the authors should be tested for their r-value and their p-level significance.

[Reply] We will add the p-value in the revised manuscript.

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[Dr. Fontanier’s comment] Finally, I wonder whether this approach may be relevant and reliable in oligotrophic settings where only few fossilizing shallow infaunal species thrive.

[Reply] Although we are not able to apply our findings to oligotrophic settings right now, as you may know, we think that the inter-individual isotopic variations should be changed by the chemical conditions in sediment, the flux of organic matter, and so on. We will have to study the relationships between those aspects of various sediment settings (e.g., oligotrophic setting) in future work.

On the other hand, we expect that the magnitude of the isotopic disequilibrium (Δδ13C and Δδ18O) in species is also correlated with the inter-individual variation in various settings. This is because isotopic values are determined by:

1) Temperature (almost constant in deep sea)

2) δ13C of DIC / δ18O of H2O of bottom water (homogeneous in deep sea)

3) δ13C of DIC of pore water (heterogeneous in sediment, especially in microscale because of decompositions of organic particles)

4) Microhabitat effect, differences of intracellular pH, and many other factors, including the ‘vital effect’ reported in previous studies (the magnitude of those influences should be different among individuals)

Factors 1 and 2 affect all foraminiferal isotopic compositions equally. The magnitudes of factors 3 and 4 are not always the same but are changeable on an individual level. Therefore, if the effects of factors 3 and 4 are increasing, not only the magnitude of Δδ13C and Δ18O but also the inter-individual isotopic dispersions may increase. If we can collect multiple species having different inter-/intra-species isotopic compositions from the same sample, we expect that we may be able to estimate bottom water DIC by using the method presented in this study.

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[Dr. Fontanier’s comment] For instance, do the authors think that they may rebuild
Our method to estimate $\delta^{13}C$ of bottom water presented in this study is based on the inter-individual isotopic variation of multiple species collected from the same sediment cores at three sites. If only one species was found in a sampling site, it is difficult to estimate $\delta^{13}C$ of bottom water by using our method presented here. If we are able to determine that the inter-individual isotopic variation (SD) of a species (e.g. Epistominella exigua) is almost constant in various sampling sites, we will be able to use them to estimate the $\delta^{13}C$ bottom water by using a generalized relational equation between SD and $\Delta \delta^{13}C$.

[Dr. Fontanier's comment] More generally (and as a respond to the previous question), the authors focus their discussion on the applicability of isotopes to reconstruct bottom water signals. But, for most infaunal foraminiferal species, it seems that the (average) delta13C is strongly constrained by in-sediment processes affecting pore water delta13CDIC (exported productivity, in-sediment organic matter mineralization, cold seeps...) (many publications as references). For instance, the authors should discuss the overall role of microhabitat on the specific (average value) delta13C, before dealing in detail with the inter-individual isotopic variations.

[Reply] We also thought that a main part of the inter-individual isotopic variability in $\delta^{13}C$ is caused by isotopic variability of DIC in sediment pore water. The microscale isotopic heterogeneities caused by the decomposition of organic matter and the wider range of depth habitats of foraminifera may result in large inter-individual variability in their isotopic compositions.

However, even considering the isotopic variation in sediments owing to the decomposition of organic matter and the presence of a geothermal gradient, we cannot account for the extremely negative isotopic values and the large interindividual variation in $\delta^{13}C$ and $\delta^{18}O$. The $\delta^{13}C$ values of most individuals were much lower than $\delta^{13}C$ DIC values of pore water at the sediment depth of which they had been taken. Also we could not explain the variability of $\delta^{18}O$ because the decomposition of organic matter does not change $\delta^{18}O$ of pore water. The $\delta^{18}O$ values of pore water at each sediment-depth show almost homogeneous isotopic values (the magnitude of $\delta^{18}O$ variation among different sediment depths is almost the same as the analytical error). Therefore, as discussed in our manuscript, we concluded that the carbonate ion concentration effect seems to be a more reasonable explanation for the changes of $\delta^{13}C$ and $\delta^{18}O$ of benthic foraminifera in our study.

Meanwhile, most of the smaller species analyzed in this study were not studied in previous isotopic studies. Among thousands of calcareous foraminifera, we could analyze only a small number of species that have a larger shell. In other words, we could not see “the whole image of characteristics of the isotopic disequilibrium of the benthic foraminifera.” As discussed in our manuscript, the complex interactions between many factors and the isotopic composition of biogenic calcite make it difficult to discuss them separately. In this study, we focused on the inter-individual isotopic variations and the whole image of the characteristics of the isotopic signature of benthic foraminifera. Additional in situ biological observations and culture experiments of many kinds of benthic foraminifera should help to clarify the mechanisms responsible for large inter-individual isotopic variations, $\Delta \delta^{13}C$ and $\Delta \delta^{18}O$. We expect also that the analytical method for microscale carbonate will be useful for this purpose.

[Dr. Fontanier's comment] They could do so for the 1208-m depth station where they have pore water data. Indeed, the Ddelta13C of some taxa is sometimes proposed as relevant proxies of environmental parameters (redox conditions in the sediment, exported organic matter flux) that are partly disconnected from bottom water signature.
Yes. The inter-individual isotopic variations will be affected and changed by the environmental parameters of a microhabitat (e.g., redox condition / decompositions of organic matter). Further studies will be needed to discuss the effect of sediment conditions on the inter-individual isotopic variations.

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[Dr. Fontanier’s comment] Specific and technical Comments: p. 6194, line 10. The Nankai Trough and the Sagami Bay are not located in “marginal seas”, are they? The authors should correct this sentence.

[Reply] We will change this sentence in the revised manuscript (“marginal seas” → “continental margin”)

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[Dr. Fontanier’s comment] p. 6194, line 23. “Multicorer” is better than “multiple corers”.

[Reply] We will use the word “multicorer” in the revised manuscript, as you suggested.

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[Dr. Fontanier’s comment] p. 6195, lines 7-10. This paragraph is unclear and should be reformulated. For instance, did the authors use either ethanol or formaldehyde (with Rose Bengal solution) to store sediments before sieving?

[Reply] We did not use ethanol and formaldehyde but filtered seawater for the rose Bengal solution.

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[Dr. Fontanier’s comment] p. 6195, line 11. The authors should precise that they have also analyzed N. labradorica and N. globosa.

[Reply] As mentioned above, we will remove those data from the revised manuscript.

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[Dr. Fontanier’s comment] p. 6195, lines 11-19. As explained in my comments for the figure 2, many taxa that are analyzed in this draft are complex in terms of taxonomic identification. For instance, Nonionella labradorica (name used in this paper) is generally described as Nonionellina labradorica (in most “Japanese” papers that I know). What is the difference between both taxa according to the authors? Nonionella globosa is very close to Nonionella stella, isn’t it? But few taxonomic plates exist as relevant illustration of what a Nonionella globosa looks like. Furthermore, Rutherfordoides and Stainforthia are both tricky genera the species of which may be easily confused. Maybe the authors should be more precise concerning the related species? Rutherfordoides cornuta? Rutherfordoides rotundata? Stainforthia fusiformis? (. . .) Globobulimina presents different species that are very close in terms of morphology (Globobulimina affinis, Globobulimina auriculata, Globobulimina hoeglundi. . .). (. . .) Finally, the authors should add an appendix with taxonomic references for all taxa which were analyzed in this study. If possible, they should precise the species of Rutherfordoides and Stainforthia. SEM pictures for all taxa are necessary for readers who would like to use related taxa for their own investigations.

[Reply] As mentioned above, we will remove the isotopic data on N. labradorica and N. globosa from the revised manuscript. We will add the photograph and SEM pictures and the taxonomic reference list in the revised manuscript.

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[Dr. Fontanier’s comment] p. 6197, line 21. “Authigenic” instead of “authentic”.

[Reply] Thank you for pointing out our error. We will correct this misspelling in the revised manuscript.

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[Dr. Fontanier’s comment] p. 6197, lines 22-25. Can the authors provide the data (with values and graphs) confirming that “interspecies differences in average isotopic values
were not due to the reduced sample size?"

[Reply] We already provide those isotopic values in Table 2, and we showed that “The average single-shell isotopic values approximately corresponded to the average values from five shells analyzed together”. The isotopic values of five shells are within the average isotopic values with SD of single-shell analysis. In addition, the reliability of isotopic analysis for samples over 0.2 µg CaCO3 has already been demonstrated in Ishimura et al. (2004,2008). We believe that “the interspecies differences in average isotopic values were not due to the reduced sample size.”

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[Dr. Fontanier’s comment] p. 6199, lines 13-16. If I look carefully at the table 2 and at the figure 3, a part of the large inter-individual deviation for Brizalina pacifica (MR) and Stainforthia sp. is pulled by measurements performed on very small and dead individuals. Can you trust those values as reliable for primary calcite signals? Don’t the authors think that a part of very low Ddelta13C and Ddelta18O signatures recorded for both taxa may be influenced by secondary calcite (more or less related to cold seeps, for instance)?

[Reply] We believe that the determined isotopic values are original isotopic values of primary calcite. We cleaned all individual specimens using Milli-Q water, and we did not observe any evidence of added errors from foraminiferal sampling (e.g., addition of authentic carbonate, staining by rose Bengal, etc.) or any systematic analytical errors (e.g., leakage of air, isotopic fractionation).

In addition, if we consider the possibility of the addition of secondary calcite to the foraminiferal shell, we could not explain the large isotopic shift of δ18O. The δ18O values of pore water at each sediment-depth show constant isotopic values (the magnitudes of δ18O variation among different sediment depths are almost the same as the analytical error).

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[Dr. Fontanier’s comment] Moreover, the authors have worked with Stainforthia at the genus rank (i.e. without discrimination between different species). In other words, a part of “inter-individual” variability may be related to “inter-species” variability. Right or wrong? If so, related values should be considered with utmost cares and not used for calibration (and regression).

[Reply] All the Stainforthia sp. analyzed in this study belong to a single species. Therefore, it is not a problem to use isotopic data of Stainforthia sp. as “inter-individual isotopic variation” in this study. (We will change “Stainforthia sp.” to “Stainforthia fusiformis” in the revised manuscript.)

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[Dr. Fontanier’s comment] Authors’ interpretations on the relationship between the delta18Oe.c. and foraminiferal signatures are certainly right. However, I don’t think that “species with low inter-individual deviations in (carbon) isotopic composition are more suitable as direct proxies of the bottom water environment”. Indeed, for 90% of taxa there is clear shifts compared to the equilibrium line (bottom water signature) (Fig. 3). . . and it is well-known that those shifts (>1 permile in this study) are not constant for one species and varies in function of in-sediment parameters (organic matter mineralization in the sediment, pore-water oxygenation, alkalinity, methane seepages. . .) (McCorkle et al., 1990, 1997; Schmiedl et al., 2004; Fontanier et al., 2006, . . .). That’s why the Ddelta13C of some taxa is sometimes proposed as relevant proxies of environmental parameters (redox conditions in the sediment, exported organic matter flux) that are either partly or totally disconnected from bottom water signature.

[Reply] One of the important findings in this study is that species with low inter-individual deviations in isotopic composition are “more” suitable as direct proxies of the bottom water environment “in the same sampling site and in the same sediment depth.” We surmise that the ranges of inter-individual isotopic dispersions of certain
species are not always the same among different environmental conditions (redox condition, flux of organic matter, bottom water chemistry, etc.). We have to evaluate the reliability of our results in various environments to make reliable bottom water proxies in future studies.

[Dr. Fontanier’s comment] p. 6199-6200. The paragraph 3.2 and the related illustrations (Figure 5 and Table 3) are slightly confusing. To be honest, I have got some doubts on the related conclusions (i.e. the applicability of inter-individual distributions (SD within species) to reconstruct the bottom water delta13CDIC). Why? (1) I don’t trust in the isotopes values related to either Stainforthia or Rutherfordoides without specific determination. If you consider Uvigerinids for instance, signatures are totally different between U. mediterranea, U. peregrina and U. elongatastriata and they belong all to Uvigerina genus. (2) The authors have forgotten to add isotopes data of Nonionella globosa, Nonionella labradorica and Takayanagia delicata for the Sea of Okthosk in the figure 5. Am I right? If you add those average values (and related SD), it seems that mathematical regressions are suddenly much less convincing for this station. I may be wrong but the authors should discuss this point! (3) If the authors want to draw a convincing regression line, they should have provided the same quality of data for each species at each site (equal number of measurements per species at one site, only living foraminifera). It is not the case in the present study. (4) Coefficients of Determination (R2) are high but it does not mean that the correlation coefficient (r) is statistically significant. The authors should calculate the r-value and the p-level of significance.

[Reply] 1) We did not discuss the isotopic data on a genus level. Rutherfordoides and Stainforthia did not consist of multiple species (not spp.) but rather single species. 2) As mentioned above, we will remove the data on Nonionella globosa and Nonionella labradorica from the revised manuscript. We have plotted the data of Takayanagia delicata for the Sea of Okthosk in the figures. 3) We provided the same quality and quantity of data at each site (an almost equal number of measurements per species at one site). 4) We will add the p-values in the revised manuscript.

[Dr. Fontanier’s comment] p. 6200-6201. Now again, and as explained above, the lower Ddelta13C and Ddelta18O recorded for the smaller and dead individuals belonging to Brizalina pacifica (MR) and Stainforthia sp. might be related to secondary calcite. Have the authors investigate the possibility of authigenic carbonate precipitation that may be related to cold seeps?

[Reply] We did not find any additional carbonate on the shell during microscopic observation of foraminiferal shells and sediments. If the authigenic carbonate affected the isotopic compositions of the foraminiferal shell, we would not be able to explain the variability of δ18O (negative shift larger than the δ18O variation of pore water). For δ13C, as you suggested, we might consider the possibility of authigenic carbonate precipitation, which may be related to cold seeps. In our studied site, no evidence of methane release is found. Also, the statement ‘No systematic difference was observed in isotopic values between living and dead individuals’ supports the idea of a lower possibility of the addition of authigenic calcite to the foraminiferal shell.

[Dr. Fontanier’s comment] Figure 1. According to the caption, the location of both stations A and B in the related map seems wrong and should be checked.

[Reply] We checked them and found that those stations are not wrong. Because Fig. 1 seems to be misleading, we will revise it.

[Dr. Fontanier’s comment] Figure 2. Rutherfordoides sp., Globobulimina affinis (from Sagami Bay), Bulimina aculeata (from Nankai Trough), Stainforthia sp., Nonionella globosa and Nonionella labradorica (from Okhotsk Sea) should be pictured. Indeed, most
of these species/genus are quite complex in terms of taxonomic identification. Therefore, some complementary illustrations would be very useful for readers interested in the related study areas. Moreover, SEM pictures for all taxa (with different views) would be more relevant than normal photographs. Please, don’t use “sp.” in italics for Rutherfordoides sp.

[Reply] We will add the photograph and SEM pictures in the revised manuscript. We will correct the Rutherfordoides “sp.” in Table 2c.

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[Dr. Fontanier’s comment] Figure 3. Figure 4. Where are data for N. globosa and N. labradorica?

[Reply] As mentioned above, we will remove those data from the revised manuscript.

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[Dr. Fontanier’s comment] Figure 4. “R2” is not sufficient. “r” is required with the p-level of statistical significance.

[Reply] We will add the p-values in the revised manuscript.

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[Dr. Fontanier’s comment] Table 1. The presentation of sediment intervals used for pore water analyses is awkward. For instance, what is the meaning of “2”? Is it the sediment interval 1-3 cm? or the sediment interval 0-2 cm? What is the meaning of bottom water?

[Reply] Sediment intervals are +/-0.5 cm (e.g., “2” means “2 +/- 0.5 cm”). Bottom water means the bottom water taken from the multicorer, about 5–10 cm above the sea floor. We will add these explanations in the revised manuscript.

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[Dr. Fontanier’s comment] Table 2a-c. Please, precise in the caption the meaning of “cmbsf”, and also the meaning of “*” used for B. aculeata.

[Reply] We will add the following explanation to the caption: “cmbsf denotes cm below the sea floor.” We did not use “*” for B. aculeata but rather for Cassidulina norvangi in our manuscript. (Results of Cassidulina norvangi reported in a previous study (Ishimura et al., 2004) are denoted by an asterisk.)

We would like to thank you for the helpful comments and suggestions. We trust that the responses to your comments and questions are satisfactory.

Interactive comment on Biogeosciences Discuss., 9, 6191, 2012.