Chemosymbiotic species from the Gulf of Cadiz (NE Atlantic): distribution, life styles and nutritional patterns
C. F. Rodrigues, A. Hilário, and M. R. Cunha

This paper samples the species diversity and stable isotope content of symbiotic species around mud volcanoes in the Gulf of Cadiz. The sampling encompasses a wide depth range and varying environmental and chemical characteristics of the sediments. From this, the authors describe the distribution of chemosymbiotic species (which have an unusually high diversity compared with other seep regions of the world) in the context of the known environmental characteristics if the different sites to infer why species may occur at certain places and why this region may support such high diversity. Stable isotope analysis of tissues from select species gives insight into the primary inorganic carbon, nitrogen, and sulfur sources and the mode of carbon fixation used by their symbionts. This work forms a basis for future studies on trophic ecology and resource partitioning among species, especially those that appear ecologically similar, inhabit the same space, but have different isotope contents.

I found the methods, analysis, and conclusions of this paper to be appropriate for the purposes of this study. I have made a few minor comments about the scientific content of the paper and many edits for spelling, grammar, and syntax. Besides the errors due to the authors not being native English speakers, the manuscript is generally clear and well-organized. Though I have pointed out a few of the errors, a more thorough reading by a native English speaker would help to put all of the commas in the right places. I recommend this manuscript for publication in Biogeosciences.

General Comments:

The use of “life styles”. I think it is more appropriate to call this “life histories”, which essentially means the lifestyle of an organism, but is specifically used in a biological (scientific) context. Life history traits of an animal, for example, could be things like the animal lays eggs, lives 12 years, eats algae, etc. Lifestyle (one word) is more often used for people.

Table 2: why not just report the difference between gill and tissue instead of standard error. Standard error does not have much meaning when you are only dealing with two data points and it would be good to show that one tissue is or is not consistently higher or lower in isotope value than the other (using positive and negative signs for the difference). Also, please align the numbers to the right. Ideally the numbers would align by the decimal point.

Figure 2: in panel C, the colors of two of the green shades are not the same in the legend and the figure. Consider using different colors with more contrast and making the symbols in the legend larger. In the caption, saying “mean values” is confusing. I think it is fine to state once that you averaged the value for the two tissues, and thereafter refer to these as individual values instead of mean values. Mean implies averaging among individuals as is the meaning in the second sentence of the caption.

Comments by Line:
Page 17348:
Line 4: change “life styles of those species” to “life histories of the species”
Lines 7-9: Change “Twenty siboglinid and nine bivalve chemosymbiotic species have been identified and were found living in fifteen mud volcanoes during our studies.” to “During our studies, we identified 20 siboglinid and 9 bivalve chemosymbiotic species living in 15 mud volcanoes.” Or “20 siboglinid and 9 bivalve species were identified.”
Lines 14-15: change “Isotopic values found for selected species” to “Tissue stable isotope contents for selected species”
Line 21 and 22: should be “highlights” and “provides”
Line 25: “tubeworms” should be “mussels”. That’s what the Rau and Hedges paper was about and vent tubeworms sometimes have enriched δ13C.

Page 17349:
Line 3: citations?
Line 5: change to “and in exchange the symbionts provide nutrition to the host”
Line 8: DIC doesn’t always come from pore water. Maybe just add “or bottom water”
Line 17: phytoplankton, (comma)
Line 18-19: change to “relative to heterotrophs and animals with methanotrophic symbionts”

Page 17350:
Line 13: “a major influence on the quantity”
Line 15: column, (comma)
Line 20: organization,
Line 21: gill,
Line 22: also have very different life histories
Line 23: “bysate” I have never heard this word before. Do you mean to say “epibenthic” instead of epifaunal?
Line 25: “methane-oxidizing symbionts… and that they have retained”

Page 17351:
Line 3: bottom water, where oxygen
Line 4: poseidonii, which
Line 6: methane is transported
Line 8: their symbionts,
Line 21: sampling devices
Line 22: on board as described in Rodrigues et al. (2011a).
Line 25: “new code names when species hadn’t previously been collected (described?).”
Is this your meaning? Same line change “more” to “most abundant”

Page 17352
Line 3: “freeze-dried” was this really what you did? Or did you dry the samples in a drying oven. Same line: mortar and pestle.
Line 4: tool, and subsamples were separated for… The ground subsample
Line 7 and 10: reground instead of re-grounded
Line 8: shaken not shacked
Line 13-14: change to “are reported in standard delta notation relative to a standard material according to the following equation:”
Line 17: and y the molecular weight of the lighter
identified from always found

multitenticulate (add an L) undetermined not undermined

“...long tubes that continuously cover the crater...”

“on the gill tissue and foot tissue, no difference larger than XX was found between the two tissues”. It is not clear what “significant” means here. Did you do a statistical test? Did you use all species together or each species separately? Might be safer to just say the largest difference instead of using “significant”. I make a note about this for Table 2 as well.

Table 2), which never had $\delta^{13}$C values...” and “-36.8‰” (permil)

values, (27.5‰), showing (not shoing)

Siboglinum lb, which.....MVs,

“with a clear separation of values” might be worth making a figure that shows this visually. There are, after all, only two figures in this manuscript and this spatial difference is interesting.

site, MVs,

“between a eukaryote” Even though eukaryote starts with an e it has a y sound so is preceded by a.

Because the association between an eukaryote and its symbionts can be seen as an adaptation to bridge oxic-anoxic interfaces,” This seems a bit weird to me. I think you mean that “Because the association of a eukaryote and can be seen as an adaptation that allows these organisms to thrive at oxic-anoxic interfaces, the chemistry”

Solemyid, lucinid, and thyasirid

long branching burrows Regarding Vesicomyidae,...sediment,

flexibility: some species are capable of hosting multiple symbioses, including the co-occurrence of sulfur- and methane-oxidizers, and can receive additional nutrition...”

posterior tube and body

only been found in the Porto, Bonjardim, and Carlos Ribeiro MVs, respectively, where...

hydrate not hydrates sustaining a higher biomass... Capt. Arutyonov MV,

found,

settings (no comma)

demand a lower concentration
Line 23: rare,
Line 29: species occur (take out “and may”)

Page 17359
Line 1: surface, while the larger species (no comma) Thyasira
Line 5: the larger species Christineconcha regab was (no parentheses)
Line 20: I believe it is the other way around, biogenic is more negative than thermogenic.
Check Schoell again carefully (the figures are a bit confusing) and see Roberts and Aharon 1994. Hydrocarbon-derived carbonate buildups of the northern Gulf of Mexico continental slope: A review of submersible investigations. This paper is about the Gulf of Mexico, but there is a nice schematic drawing of thermogenic and biogenic methane with the $\delta^{13}C$ ranges.
Line 25: particular (no comma)
Line 27-28: fjords, which are known

Page 17360
Line 1-3: Nevertheless, the values reported here are compatible with methanotrophic nutrition, and ....have already shown...bacteria in...
Line 5: average $\delta^{34}S$ value (below 6‰)
Line 6-7: “it is possible that …may have been pooled” this sounds very uncertain, either they were or not. Instead, write “Because of their small size..., it was sometimes necessary to pool”
Line 8: “B.” mauritanicus, which have highly depleted $\delta^{13}C$ values and $\delta^{34}S$ values lower than 5‰.
Line 13: dependent upon
Line 15-17: “B. childressi, which also harbor a dual symbiosis, but….mauritanicus…..methane-oxidizing bacteria.” B. childressi have only methane-oxidizing symbionts. The other species in the Gulf of Mexico, B. heckerae and B. brooksi, have dual symbioses.

Page 17361
Line 22: enzyme
Line 24: difference not disproportion
Line 25: delete “of the chemoautotrophic symbiosis” from the sentence

Page 17362
Line 7: worth noting
Line 9: Oliver et al. (2011) identified….as the new species A. gadirae.
Line 14: data, the $\delta^{13}C$ values of
Line 25: high values found in (delete “were”) 
Line 27: can exploit different sources of nitrogen (delete “since”) in addition to the nutrition provided by their symbionts, because they may be able to take up and metabolise dissolved organic compounds

Page 17363
Line 2: surficial not superficial
Line 4: possibly not possible
Line 7: in this study results
Line 9: fractionation factors during
Line 13: MVs, which could be due to a higher variation
Line 15: differences may be lower at shallow MVs, because
the variations in environmental setting and AOM…
chemosymbiotic animals
in the Gulf of Cadiz, thiotrophy