### **General Comment**

Although I thought that this paper could be an interesting contribution, as it reports a number of gastropod taxa from different types of environments and depth in the gulf of Cadiz where gastropod fauna is poorly known, this paper has still many weaknesses that required clarifications and analyses.

The paper focus on mud volcano gastropod fauna, but some of them are presently inactive in the Gulf of Cadiz, and therefore the study includes a wide heterogeneity of habitats, much higher than in most of the other cold-seep studies focussing on active seepages. Moreover, data include sampled in "adjacent habitats" mainly represented by carbonate outcrops likely resulting from old seepages. This should be better considered in the different sections of the paper, in order to compare the observed gastropod fauna not only with active seep habitats, but also with other deep-sea environments, including soft and hard substrata.

I am very disappointed to see any consideration to my first comments (short review) with the most important are:

- -<u>Site description</u>: because of the very high heterogeneity at different spatial scales, it is important to give more information about the sites and habitats, even if chemical or visual observation is not available (e.g. did the cores from the mud volcanoes indicated a sulfidic environement: sulphide smell, black sediments?), in which habitats have the experimental devices been deployed.
- -<u>The mixing of results</u> from samples taken in the natural environment and from experiments that should be presented separately before comparison. It is not possible to distinguish both sources of samples in the results (Table 1, Fig 1, Fig 3, Fig4). It is interesting to present together both set of data but it should be clearer of which data is considered for each analysis.
- -<u>The sampling effort</u> has to be estimated in order to validate or discuss more carefully the results (e.g. high % of singletons): was the diversity completely sampled? (rarefaction curves based on area of sampling, or on number of individuals)

Diversity analyses (accumulation, loss and turnover curves representing the species replacement with increasing depth: fig.3) have been performed on

the total dataset of taxa. However, quantitative data are only available for 14 of the 58 samples (USNEL cores) and the sampling effort is highly heterogeneous between subregions, depths and substrata types.

Several conclusions are based on this sampling bias (influence of depth, substrata, subregion, mud volcano...on species turnover, species richness). The diversity cannot be compared if it is not related to sampling effort (sampling area, or at least number of samples or number of sampled individuals).

# **Specific comments**

#### Introduction

## P3709 line 8

If they have been recognized as biodiversity hotspots with enhanced species richness and evenness as well as distinct composition (e.g (Henry & Roberts 2007), coral reefs cannot be considered as "where species richness and diversity rivals that of their tropical counterparts, completely changed our perception of the deep-sea environment

#### P3710 Line 7

Other reference about reproduction and dispersal of seep gastropods could be added:

- Eckelbarger KJ, Young CM (1997) Ultrastructure of the ovary and oogenesis in the methane-seep mollusc *Bathynerita naticoidea* (Gastropoda: Neritidae) from the Louisiana slope. Invertebrate Biology 116:299-312
- Hodgson AN, Eckelbarger KJ, Young CM (1998) Sperm morphology and spermiogenesis in the methane-seep mollusc *Bathynerita naticoidea* (Gastropoda: Neritacea) from the Louisiana slope. Invertebrates Biology 117:199-207
- Young CM, al. (2012) Dispersal of Deep-Sea Larvae from the Intra-American Seas: Simulations of Trajectories using Ocean Models. Integrative and Comparative Biology 52:483-496

#### P3710, line 11

Although discussed later in the paper, the authors present here a very simplified contrasted situation of planktotrophic vs non-planktotrophic species regarding their potential of dispersal (planktotrophs dispersing more than lecitotrophs) and inferred the dispersal capabilities of the different species based on the above simplified statement (lines 20-23)..

Nevertheless this is not always the case in the deep-sea, as said by (Sasaki et al. 2010), or before by Tyler and Young 2009.

There are several hypotheses for long distance dispersal of

lecitotrophic larvae (see Sasaki) particularly for vent and seep species.

#### **Material and Methods**

#### There are several problems in the site description and habitat characterisation:

One is the hard substrate that is sometime attributed to carbonates (that may be related to cold seep)s or corals, and later to the CWC only.

Another one is the description of the three sub-regions: the El Arraiche field (EA) and the deep-water field (DF) are described as mud volcanoes with the most active in the DF region. In contrast, the carbonate province (CP) is presented as a fossil area with carbonates and CWC. However, as seen in Table 1, Mussel beds (Bathymodiolus sp. I assume) have been described from some of the mud volcanoes of this area, so active seeps. I understand that the authors have chosen to separate the sub-regions according to substrate dominance but the occurrence of active seepage could be as important as substrate to understand fauna distribution.

These 3 sub-regions should be indicated in Fig1, where only the mud volcano names are reported

Are these regions corresponding to geographic areas? Fopr some of them it seems (the carbonate province) but for other not (adjacent habitats). This has to be clarified. P3715, line 18

Sub-regions are described as "bathymetric sub-regions". However, adjacent habitats are not included in the first 3 sub-regions and encompasses the whole bathymetric gradient.

The comparison of the number of species, or species composition between sub-regions, or along depth is therefore confusing (p3715 lines17-21)

Other inconsistencies: "flanks" are sometimes considered as "adjacent habitats", sometimes in others (Mvs). E.g. Carlos Ribeiro JC10 54 MC 27.05.07 35º47.30' 08º25.22' 2179 Flank; Coral site NE; Hemipelagic sediments with coral debris Soft is in « Deep field"/ Why not in Carbonate province? And why substrate is classified as "soft"? It seems rather "mixed?"

P3711, lines 24-25

This is unclear if hard samples are only from Cold Water Coral sites or also Carbonates from mud volcanoes: it has to be clarified. There is also a confusion between the Mud Volcano (EA) and the carbonate (CP) provinces

#### **Material & Methods & Results**

The diversity cannot be compared if it is not related to sampling effort (sampling area, or at least number of samples or number of sampled individuals).

- Diversity analyses (accumulation, loss and turnover curves representing the species replacement with increasing depth: fig.3) have been performed on

"the total dataset of taxa". However, quantitative data are only available for 14 of the 58 samples (USNEL cores) and the sampling effort is heterogeneous between sub-regions, depth and substrate type.

These curves highlight that "the three major locations of species replacement are the Mercator MV, the Darwin MV and Captain Arutyunov MV, that are the three sites with the highest sampling effort, what is not really surprising.

- In the same way, the fact that "the major contributors to the species richness in each region were the assemblages from Mercator (EA, 350 m), Darwin (CP, 1100 m) and Captain Arutyunov MVs (DF, 1300 m) with 8, 17 and 11 species, respectively" has to be discussed according to the sampling effort
- Again, the comparison of species richness among substrate type is not valid, for the same sampling bias.

P3714, lines 26...from the dataset, it is observed that "the number of species occurring exclusively in Soft sediments was much higher (22) than the one observed exclusively in Organic (3) or Hard (12) Substrata". However, there are more samples from Soft sediments (34) than from other substrate. This is not surprising that the diversity is higher and then the number of species occurring exclusively in this substrate higher.

The colonisation experiment revealed that some species may have a larger bathymetric range that observed by sampling in natural environment (page 3716, from line 25 to end of the paragraph). These observations have to be considered in discussing the results. May be the favourable habitat is absent, but may be it has not been sampled.

### **Concluding remarks**

From concluding remarks list, those are supported by the data, other not.

- 1. It is expected from its geographical position that Gulf of Cadiz deep-sea gastropod fauna includes species from Atlantic and Mediterranean areas. Our study confirms this hypothesis and gives new evidence for connectivity of deep-sea gastropod communities inhabiting reducing environments. **Yes**
- This work also shows that substrate type, with its associated biotic and abiotic characteristics, has a determinant effect on gastropod distribution in the deepsea, and a diversity of feeding types combined with a variety of dispersal strategies contributes to their successful colonization of deep-sea habitats. NO, as sampling effort inappropriate (too heterogeneous)
- **3.** Moreover, our work suggests that various deep-sea habitats form a network of suitable environments, which enhances diversity and ensures connectivity of deep-sea gastropod populations. Yes, with combination of experimental and sampling approaches