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Biogeosciences Discussions

Interactive comment on “*Blastodinium* spp. infect copepods in the ultra-oligotrophic marine waters of the Mediterranean Sea” *by* C. Alves-de-Souza et al.

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Anonymous Referee #1 Received and published: 4 April 2011

This paper contributes valuable information on morphotypes of Blastodinium, copepod infections, SSU rDNA gene sequence analyses and oligonucleotidic probes to describe abundances of of Blastodinium dinospores in three stations of a cruise. For me, this represents very valuable data for this manuscript. When authors infer about dinospore distributions and their ecological relevance in oligotrophic waters, they have to take in mind several points: -the authors recognise that your work likely underestimates the genetic diversity of Blastodinium, since several morphotypes could not be amplified

with cluster-specific oligonucleotidic probes. -the parasites presented highly seasonality -there is no data on copepod abundance and distribution in the sampling stations since sampling were performed by a net These points raise some questions: -does dinospores morphology reflect the different clusters defined in this study based upon the SSU RDNA gene analyses?

Answer: No. Each cluster grouped together very different sequences. We only described the most important morphotypes occurring in our samples (main size of all individuals, theca thinness), but diverging morphotypes were also observed inside each clusters. This was yet described in the result part: “Most of these cells had a relatively large transversal cingulum. Thecae were relatively thick for cells targeted by probes BCON2 and BMANG1. This was not the case for majority of BLA2-targeted cells, which exhibited thinner thecae (Fig. 5C). The smallest cells were detected using the probe BMANG1 (7-10 μm in length and 5-10 μm in width), whereas larger cells were observed using the probe BCON2 (11-18 μm in length and 9.5-13 μm in width). Beside these general characteristics, a given probe was associated with several distinct morphotypes, especially within the BMANG1 cluster (Fig. 5B).” To clarify that point also in the discussion, we added the following sentence:: “Although the three different probes were associated to relatively resembling individuals (by their mean size for example), several different morphotypes were in fact observed inside each cluster. This is congruent with the huge genetic diversity recorded, leading to the conclusion that each cluster cannot be simply reduced to a single morphotype”.

-does the authors expect similar results in winter conditions?

Answer: No, this was yet discussed in the manuscript. See the following sentence: “Blastodinium occurrences are reported to have a marked seasonality, with highest prevalences observed during warmer period of the year in the Mediterranean Sea (Chatton, 1920; Skovgaard and Saiz, 2006). Concomitantly, Chatton (1920) also reported slower sporulations at low temperature. Thus, the summer conditions during the BOUM cruise were probably likely favourable for Blastodinium spp..

-the abundance and distribution of the copepods is not known, so is the paragraph of dinospore-host distribution in oligotrophic waters going beyond to the infections data?

Answer: We agree that there is no available data on the VERTICAL distribution of ADULT copepods during this study. However, vertical distributions of copepods in the Mediterranean sea (particularly during summer periods) are particularly well documented (see Paffenhöfer and Mazzocchi, 2003 and Peralba and Mazzocchi, 2004). We used these references to discuss congruence with the observed dinospore distributions.

There are good points in this work since results will raise several hypothesis to be tested. For e.g. why high values in one stations and in two groups (Corycaeida and Calanoida Group) were recorded. Other example is about the Blastodinium infections, that are supposed to be initiated during the early stages of copepod development however,it seems there is a drastic partitioning between nauplii and dinospores in that cruise. Does the authors have any explanation for the first stage of infection by Blastodinium?

Answer: Our hypothesis was yet exposed in the discussion of the manuscript: "Blastodinium infections are supposed to be initiated during the early stages of copepod development (Chatton, 1920). However, there is a drastic partitioning between nauplii and dinospores (with a significant negative correlation). In other part, nauplii are known to largely consume prey items smaller than the Blastodinium dinospores. In contrast, copepodites do feed on prey of dinospore-size, 10-20 μm (Wilson, 1973). Based on these considerations, it appears likely that copepodites are the first stage infected by Blastodinium spp."

Anonymous Referee #2 Received and published: 30 May 2011 Unicellular organisms parasitic of crustaceans are well known as a powerful factor controlling host mortality and fecundity in freshwater ecosystems (e.g. Green J, 1974; Burns C.W., 1985, 1989; Ebert D. et al. and some others). Prevalence values up to 80- 90% are not too rare

in highly productive systems with dense populations of plankton crustaceans. It is not like this in marine pelagic ecosystems. Moreover, these systems are rather poorly investigated in respect to host-parasite interactions. The results obtained for oligotrophic and ultra-oligotrophic waters of the Mediterranean Sea (16% of all copepods and even more, 51% of Coryceidae were infected with gut living parasites Blastodinium spp.) are very impressive and, at the first glance, the figures look too high in the situation of low biological productivity and low densities of populations.

Answer: Comparison between both ecosystems is effectively very interesting. We added this comment in the discussion part.

It seems that interacting populations of copepods and free-swimming stages (dinospores) of Blastodinium spp. should be aggregated and spatially overlapped. What hydrophysical (meso- and microscale) and biological (behavioural) mechanisms maintain such an overlapping? To understand these mechanisms, more coordinated experimental and field studies on copepods and parasites behaviour and microdistribution are required. Vertical structure of the water column, thin-layer patterns of hydrophysical parameters, and microcirculation should be a valuable addition to biological and ecological studies. However, these are the points for future investigations.

Answer: The three stations considered during this study are located inside anticyclonic eddies. In the text, we added two valuable references describing influence of eddies upon microbial communities from this special issue (Christaki et al. and Crombet et al.). How the formation of such mesoscale structures interacts with parasitic interactions is effectively an interesting question. Presence of deep intense local diatom blooms at station C argued for thin-layer patterns forced by hydrophysical mechanisms. Similar processes may favor the host-parasite encountering rate. However, we agree with the reviewer that more discussion on that direction remains exclusively speculative. To open this discussion, we added the following sentences in the conclusion: "The free-living stages (or dinospores) of the parasite Blastodinium spp. occur in the water column and can be detected by FISH technique. They formed relatively dense communities located at the DCM or slightly below, a vertical distribution similar to that of their copepod hosts. Interestingly, deep intense blooms of diatoms were also detected during the BOUM transect, especially at station C (Crombet et al., this issue). Such phenomenon may be favoured by hydrophysical meso- and microscale mechanisms. The trophic mode of the dinospores and their ability to persist outside a copepod host at relatively high density are among the many questions which remain concerning these organisms."

The discussed paper contains valuable information on the morphological and genetic diversity of Blastodinium spp., their distribution among copepod hosts and within the water column. The suggested technique (FISH – fluorescent in situ hybridization method) for assessment of abundance and diversity of free-living dinospores allows to fill the gap in our knowledge of the life cycle of these widely distributed and ecologically important parasites. At this stage of Blastodinium studies, it is plausible to combine data on different aspects of parasite biology, ecology and phylogeny in one paper. Deeper, more specific investigations on phylogeny and diversity of these parasites, on the one hand, and ecology, behaviour and spatial distribution, on the other hand, require to tackle these problems separately. The paper is thoroughly prepared, properly structured and well written.

I suggest only few small corrections: 1. P. 2578, l. 11-14 – negative correlation between dinospores and total eucaryotes is indicated, but RS = 0.60 is given in l. 14. Should be "-0.60".

Answer: Changed in the text

2. P. 2578, l. 24 – "typical of" instead of "typical for"

Answer: Changed in the text
