

## Interactive comment on "16S rRNA gene metabarcoding reveals a potential metabolic role for intracellular bacteria in a major marine planktonic calcifier (Foraminifera)" by Clare Bird et al.

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- The title is misleading as it is not supported by any result presented in this study, and should be changed.
- Synechococcus clade I also contains green-light specialists (e.g. strains ROS8604 or SYN20, see Pittera et al. 2014, ISME J, doi 10.1038/ismej.2013.228). Similarly, only two strains of clade IV have been characterized for pigmentation. The claim that all clade I and IV Synechococcus are chromatic acclimaters (discussion 4.1.5 and 4.2) is thus false for clade I and should be moderated for clade IV.

C1

- In paragraph 4.1.1, you claim that PCR amplification of 16S RNA and rbcL "provides additional evidence that Synechococcus DNA [is] not grossly degraded by nucleases". However, you do amplify 16S marker gene for metabarcoding of bacteria that you claim are digested by the host. You thus use the same result to draw diametrically opposed conclusions.
- In paragraph 4.1.3, you observe 100% identity between 16S RNA and rbcL markers between "endobionts" and clade IV Synechococcus CC9902. However, strains CC9902 and BL107 share 100% and 98.9% nucleotide identity for these markers, yet share an average nucleotide identity of 91.3% for the core proteins (see Dufresne et al 2008, Genome Biology, doi 10.1186/gb-2008-9-5-r90), lower than the threshold value commonly used for bacterial species definition (94%). The conclusion "This strongly supports a strategy of horizontal rather than vertical transmission" should thus be moderated, as two strains exhibiting high degrees of similarity for these markers can be quite divergent for the rest of their genome: these markers thus provide evidence but are not sufficient to totally exclude genetic drift between internal and free-living Synechococcus.

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