

## ***Interactive comment on “Light-dependent calcification in Red Sea giant clam *Tridacna maxima*” by Susann Rossbach et al.***

**Susann Rossbach et al.**

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"The manuscript investigates a common yet little known question on the light dependency of iconic giant clams. Using experiments, Authors were able to show interesting results that showed congruence to the species' natural depth distribution. Some explanations were provided regarding their results, but I think there is scope to expand their Discussion (e.g. how symbiotic species may play a role in affecting depth distributions and affect calcification). The current manuscript depth peer review, as well as to provide more details on the mechanism of light-induced calcification and why there is a maximum light threshold before calcification rates drop could be further expounded."

We thank the reviewer for the comments, which were very constructive and helpful.

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Below, we provide a “Response to reviewer” document detailing, point-by-point, the actions taken to address each comment. The original comments are marked with quotation signs "", and our responses refer to line numbers of the revised manuscript whenever possible, so that the changes can be easily assessed.

Specific comments:

1) "Abstract, Line 8: Tridacninae is the subfamily for giant clams. Please amend the first sentence."

Answer: Abstract, Line 8 was changed to: 'Tropical giant clams of the subfamily Tridacninae, including the species *Tridacna maxima*, are unique among bivalves as they live in a symbiotic relationship with unicellular algae and generally function as net photoautotrophic.'

2)"Introduction, Line 15: Suggest to use Neo et al., 2017 - a review of species status, instead of Neo et al., 2015 that looks at ecological roles. Reference citation: Neo ML, CCC Wabnitz, RD Braley, GA Heslinga, C Fauvelot, S Van Wynsberge, S Andréfouët, C Waters, AS-H Tan, ED Gomez, MJ Costello & PA Todd (2017) Chapter 4. Giant clams (Bivalvia: Cardiidae: Tridacninae): A comprehensive update of species and their distribution, current threats and conservation status. In: Hawkins SJ, Evans AJ, Dale AC, Firth LB, Hughes DJ, Smith IP (eds.), *Oceanography and Marine Biology: An Annual Review*, Volume 55. Pp. 87–388. CRC Press: Boca Raton, FL."

Answer: We agree with the reviewer and changed the text accordingly:

Page 1, Line 13-16: 'Currently, all giant clam species are listed in the IUCN Red List of Threatened Species (IUCN, 2016) and protected under Appendix II of the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES). Most of them are considered under a lower risk / conservation dependent status, however the IUCN status of tridacnine species, is in need of updating according to Neo et al. (2017).'

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3) "Discussion, Section 4.1: Suggest Authors to look at the following papers to compare how locations of giant clams (sheltered versus exposed sites) can affect distribution.

References: Militz TA, J Kinch & PC Southgate (2016) Population Demographics of *Tridacna noae* (Röding, 1798) in New Ireland, Papua New Guinea. *Journal of Shellfish Research* 34(2): 329-335. Neo ML, L-L Liu, D Huang & K Soong (2018) Thriving populations with low genetic diversity in giant clam species, *Tridacna maxima* and *T. noae*, at Dongsha Atoll, South China Sea. *Regional Studies in Marine Science* 24: 278–287."

Answer: We thank the reviewer for this comment and added the following paragraph to the discussion:

Page 10, Line 17-33:

'Explanations for the observed contrasts in numbers of clams per m<sup>2</sup> at both reefs could lay in the probable differences in abiotic environmental conditions at the surveyed sites. For instances, giant clams at the exposed reef are potentially more at risk from high wave action than at the sheltered reef site, which could impact the initial settlement (Jameson, 1976) as well as the survival of juveniles (Foyle et al., 1997), as both have been shown to be influenced by geographical factors (Foyle et al., 1997). While a previous study (Militz et al., 2015), in which abundances of giant clam species in French Polynesia were examined, report similar patterns for *T. crocea*, opposite patterns were observed for abundances of *T. maxima* in that region. In the reefs surveyed by Militz and colleagues (2015), *T. maxima* showed higher abundances at reef sites with a high exposure, in comparison to those with low exposure levels. However, additional factors such as temperature and local geomorphology might also have an influence on giant clam densities. Therefore, it is not possible to confidently identify the underlying causes for the observed differences by considering exposure alone. For example, *T. maxima* specimens from our study, which were located at the more southern reef, could be possibly also exposed to higher surface water temperatures due to location

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of this reef at lower latitudes. Mean seasurface annual temperature of the Red Sea have been shown to increase towards lower latitudes and can be as high as 33 °C in the Central and Southern Red Sea (Chaidez et al., 2017). Further, the local geomorphological features of each reef could influence the light availability of benthic habitats. Consequently, differences in the local topography could have led to different angles of incident light and shading conditions, which would then result in differences between reefs even though the examined depths are identical.

4) "Discussion, Section 4.2: Suggest Authors to refer to LaJeunesse et al., 2018 (Systematic Revision of Symbiodiniaceae Highlights the Antiquity and Diversity of Coral Endosymbionts) and symbiont-related papers on giant clam(e.g. DeBoer et al., 2012; Ikeda et al., 2017; Lim et al., 2019), and make inferences on how symbiont species may affect depth distribution with respect to light.

References: DeBoer TS, AC Baker, MV Erdmann, Ambariyanto, PR Jones & PH Barber (2012) Patterns of Symbiodinium distribution in three giant clam species across the biodiverse Bird's Head region of Indonesia. *Marine Ecology Progress Series* 444: 117-132. Ikeda S, Yamashita H, Kondo S-n, Inoue K, Morishima S-y, Koike K (2017) Zooxanthellal genetic varieties in giant clams are partially determined by species-intrinsic and growth-related characteristics. *PLoS ONE* 12(2): e0172285. Lim SSQ, D Huang, K Soong & ML Neo (2019) Diversity of endosymbiotic Symbiodiniaceae in giant clams at Dongsha Atoll, northern South China Sea. *Symbiosis*."

Answer: We thank the reviewer for this valuable comment and added the following paragraph to the manuscript:

Page 12, Line 11-25: 'Giant clams, including *T. maxima* can potentially harbour multiple genera of Symbiodiniaceae simultaneously (DeBoer et al., 2012; Ikeda et al., 2017), including Symbiodinium, Cladocopium and Durusdinium (previously referred to as clade A, C and D (LaJeunesse et al., 2018)) (DeBoer et al., 2012). The composition of these associated algal symbionts might therefore also impact the susceptibility to (high) light

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levels, as different genera of Symbiodiniaceae (in symbiosis) exhibit different physiological and ecological patterns, including sensitivity to light and temperature (Rowan et al., 1997; Berkelmans and Van Oppen, 2006). However, a previous study on Red Sea giant clams and their associated Symbiodiniaceae (Pappas et al., 2017), report that *T. maxima* in the region exclusively associated with *Symbiodinium* spp. (previously clade A) which was thus assumed to represent an optimal group for the local environmental conditions. Yet, the reliance of calcification of host organisms (e.g. *T. maxima*) on their relationship with symbiotic algae could provide an explanation for the significant decrease in net calcification rates at the highest light treatment (1061  $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ ). These diminished rates could be the result of photoinhibition and even photo-damage of the associated unicellular algae, when exposed to these high incident light levels. This would be also supported by the pronounced decrease in gross primary production rates at this light treatment. High incident light level, especially high level of UV radiation in shallow waters, have been previously shown to be correlated with decreased calcification rates in other marine calcifiers such as stony corals, e.g. *Porites compressa* (Kuffner, 2001).

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