1 Symbiodiniaceae conduct under natural bleaching stress during advanced gametogenesis 2 stages of the mesophotic coral *Alveopora allingi*

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11 **Keywords:** Mesophotic Coral Ecosystems (MCEs), Coral Bleaching, Coral Reproduction,
12 Host-Symbiont Interaction, Coral-Algae Symbiosis, Red Sea

13 Abstract

14 The mesophotic coral *Alveopora allingi* from the northern Gulf of Eilat/Aqaba, Red
15 Sea, is affected by year-round partial coral-bleaching events. During these events, the
16 migration of Symbiodiniaceae takes place from the coral-host mesoglea to the
17 developed oocytes in bleached parts of colonies of *A. allingi* but not in the non-bleached
18 parts. Additionally, these oocytes are abnormal, missing part of the structural material
19 of the peripheral areas and are also significantly larger in the bleached areas of the
20 colonies. Hence, we suggest a parasitic behavior of the symbionts or a commensalism
21 relationship which enhance symbionts' needs during bleaching periods and may boost
22 the gametogenesis development in these corals. We propose that evolutionarily, this
23 behavior may greatly contribute to the symbiont community survival throughout the

bleaching period, and it can also be beneficial for the host's persistence and adaptation to bleaching through the acquisition of a specific symbiont community following the bleaching event.

Introduction

Coral reefs have experienced increasing stress over the last four decades due to local anthropogenic perturbations and global climate change, resulting in severe damage to coral populations and their reproduction (Loya 2004,2007; Carpenter et al. 2008; Baird et al. 2009; Harrison 2011; Hughes et al. 2017). The symbiotic relationship between corals and their algal endosymbionts (Symbiodiniaceae) is a key factor in the evolutionary success of hermatypic corals (Wooldridge 2010). This close association between primary producer and consumer enables the tight nutrient recycling that is thought to explain the high productivity of coral reefs (Hoegh-Guldberg 1999; Wooldridge 2013; Muller-Parker et al. 2015). Consequently, environmental and physiological conditions that result in changes in the relationship between animal host and Symbiodiniaceae may have profound ecological and physiological effects (Szmant and Gassman 1990). The loss of Symbiodiniaceae - a phenomenon described as coral bleaching - can cause acute damage to the colony (Glynn 1993; Brown 1997; Loya et al. 2001; Suggett and Smith 2020). Coral bleaching has been observed in response to a diverse range of stressors and is associated with both anthropogenic and natural disturbances (Fitt and Warner 1995; Glynn 1996; Kushmaro et al. 1996; Hoegh-Guldberg 1999; Rowan 2004; Hughes et al. 2017; Hughes et al. 2018). Severe and prolonged bleaching can cause partial to total colony death, resulting in diminished reef growth, transformation of reef-building communities to degraded alternate states, increased bioerosion and, ultimately, the disappearance of reef structures (Glynn 1996). The future state of shallow reefs is glooming and the framework building of coral

communities is expected to shift over to low-relief and less complex entities (Knowlton 2001; Pandolfi et al. 2003; Hughes et al. 2007).

While shallow reefs have recently been intensively affected by mass coral-bleaching (Hughes et al. 2018), very few mesophotic coral ecosystems (MCEs; 30-150 m depth) were reported to have undergone bleaching during these events (Baker et al. 2016; Frade et al. 2018). One explanation could be that of the 'deep reef refuge' hypothesis (DRRH), which suggests that deep ecosystems are more protected from the disturbances that affect shallow reefs, and consequently could provide a viable reproductive source for shallow-reef areas following disturbance (Bongaerts et al. 2010). MCEs too, however, are not immune to bleaching (Frade et al. 2018) or to other disturbances (Rocha et al. 2018; Pinheiro et al. 2019), which in many cases have been overlooked due to technical difficulties. One of the most important questions facing scientists, policy-makers, and the general public is that of why there has been an apparent increase in the incidence of coral bleaching in the last four decades (Goreau and Hayes 1994; Hoegh-Guldberg 1999; Hughes et al. 2017) and what we can do about it?

Nevertheless, although highly impacted by anthropogenic stressors, the shallow corals of the Gulf of Eilat/Aqaba (GoE/A) (northern Red Sea) are considered heat-tolerant and resilient to thermal stress (Fine et al. 2013; Krueger et al. 2017; Osman et al. 2018). They have been described as comprising 'super-corals' that do not experience natural bleaching (Grottoli et al. 2017). Nonetheless, *Stylophora pistillata*, the most abundant depth-generalist coral in the coral reefs of Eilat, suffers from periodic non-fatal bleaching in the summer months in the upper MCEs (Nir et al. 2014), and recent observations have revealed multiple-species bleaching-events on these deeper ecosystems (Eyal et al. 2019).

Symbiotic relationship between the coral host and its Symbiodiniaceae community are complex and essential for the development of reef corals (Trench 1971; LaJeunesse 2020). Nevertheless, these relationships are not necessarily always beneficial for both the host and the symbionts. Parasitism in cnidarian symbiosis under stress conditions was suggested in the past but supported only by indirect evidences (Baker et al. 2018; Peng et al. 2020). Here we use observations from coral reproductive study to present another potential hypothesis of parasitism of the symbionts or/and commensalism relationship between the host and its symbionts.

Sexual reproduction is the most critical part of any living taxa (Kondrashov 1988). Although energetically costly, the benefit of increasing fitness and genetic diversity is priceless. Evolution processes through sexual reproduction and gene-shuffling is the primary adaptive capability to environmental changes and enabling extension to and occupation of new ecological niches (Rundle et al. 2006). In shallow corals, diverse strategies of sexuality were discovered during recent years (Baird et al. 2009; Harrison 2011; Eyal-Shaham et al. 2019; Eyal-Shaham et al. 2020) and references within) but our knowledge of the reproduction of MCE corals remains limited (Shlesinger and Loya 2019).

The purpose of this research was to detect possible natural bleaching effects on the development of gametogenesis in mesophotic corals. Hence, we examine the reproductive ecology of the mesophotic coral, *A. allingi*, under bleaching stress and describe a novel host-symbiont behavior interaction under bleaching conditions.

Methods

Study area, sampling procedures, and histological processes

Coral pairs from the same genets of non-bleached (pigmented) and visually bleached *A. allingi* were sampled in front of the Interuniversity Institute for Marine Sciences in Eilat, Israel (IUI) (29°30'N, 034°55'E), at a depth of 60 m, during the end of their 2011 reproductive season (October 2011). This is a well-developed reef with a wide variety of coral species, and *A. allingi* is one of the most abundant species in the area (Eyal-Shaham et al. 2016). As part of a larger project described in Eyal-Shaham et al. (2016), once a month, during the full moon, nubbins (4–5 cm in length) were randomly sampled from 4–6 healthy colonies of *A. allingi* using SCUBA technical diving and Closed-Circuit Rebreathers (CCR). Only in October 2021 we found partially bleached colonies (two different genet pairs of pigmented and visually bleached nubbins). The sampled colonies were >20 m distant from one another. The collected nubbins were placed in a sealed nylon bag filled with seawater and immediately after collection were fixed in a 4% formaldehyde solution in seawater for 24–48 h. The nubbins were then rinsed in tap water for 15 minutes and transferred to 70% ethyl alcohol. The decalcification process was carried out following the protocol of Eyal-Shaham et al. (2016). A piece of tissue was taken from each decalcified nubbin and 6 µm thick latitudinal histological serial sections were prepared and dyed with Mayer's hematoxylin and Putt's eosin (H&E stain) to highlight the reproduction structures.

Histological and statistical analyses

The histological analysis was performed using a Nikon Eclipse 90i microscope and NIS Elements D 3.2 software (Nikon Instruments Inc.). A detailed study of gonad structure, size, and development was conducted by analyzing the histological cross-sections [see details in Eyal-Shaham et al. (2016)]. Detailed identification of the oocytes (Oc), spermaries (Sp), nucleus and nucleoli (N), and Symbiodiniaceae (Z) were conducted. The examination included size measurements of the oocytes (longest axes), performed

only when the nucleoli were present, and identification of the developmental stages of gametocytes within the polyps. Mean oocyte size of each developmental stage was calculated from all oocytes presented in the samples from the bleached and the healthy groups.

Differences in oocyte sizes within partly bleached colonies were calculated in R language and environment (R Development Core Team 2014) using 1,000 bootstrap samples of the values to indicate the difference between the median oocyte size of the bleached part and that of the control part.

Results and Discussion

During coral bleaching events that occur at 50-60 m depth, the depth-specialist *A. allingi* experiences severe visual bleaching in parts of the branches of otherwise healthy colonies (Fig. 1a, b). Unlike the periodic summer bleaching of *S. pistillata*, also described from the same reef (Nir et al. 2014), and in which the colonies recover after the summer ends, *A. allingi* presents a gradual degradation of the bleached tissue, with mortality of the entire bleached part of the colonies occurring within a few months.

Our analysis of the histological sections of partly-bleached colonies revealed a phenomenon that to our knowledge has never previously been described: Symbiodiniaceae migration to the oocyte cytoplasm [i.e., in the visually bleached part of the colonies, Symbiodiniaceae were observed in the oocytes' cytoplasm, which presented an irregular shape and imperfect cytoplasm with missing parts of the structural material of the peripheral areas, while the healthy (non-bleached) portions of the same colonies presented normally-developed oocytes (Fig. 1c-f)]. Additionally, the bleached coral's oocytes were found to be significantly larger than those of the non-bleached parts (Fig. 2; Mann-Whitney Test, $T=544$, $P<0.001$), which suggests a faster

reproduction development in the impacted parts of these colonies. Although Symbiodiniaceae can be directly transmitted from the parent colony (i.e. vertical transmission) in some species (Lesser et al. 2013; Muller-Parker et al. 2015), the observed change in shape (i.e., larger sizes and imperfect cytoplasm with missing parts of the structural material of the peripheral areas of the oocytes only), suggests that this is not the case in *A. allingi*. Moreover, Symbiodiniaceae were not observed in any mature oocytes of other, healthy, colonies.

We hypothesize that such migration of Symbiodiniaceae into the coral oocyte cytoplasm under severe bleaching conditions potentially enhances the survivorship of the symbiont community, suggesting the existence of a possible mechanism of exploitation of cytoplasmic materials by certain symbionts. An alternative hypothesis is that the partially bleached hosts are in a process of absorbing oocyte lipids as an emergency energetic source. Cell membranes have been disturbed, including the symbiosome surrounding the host cells lining the exterior of the oocyte. This enables symbionts to escape the host cell and invade the oocytes.

The Symbiodiniaceae community in corals is generally dominated by one or two main symbiont types (van Oppen et al. 2005; Ziegler et al. 2017) but can also contain a large number of rarer members of the family, which play important roles in the resilience of the holobiont (Ziegler et al. 2018). Network theoretic modeling predicts that elevated symbiont diversity can increase community stability in response to environmental changes (Fabina et al. 2013). Moreover, the functional role of Symbiodiniaceae in the holobiont is not defined by a single species but rather by the assemblages of symbionts (Ziegler et al. 2018). The rare symbionts, however, associate in several manners with the coral host (Knowlton and Rohwer 2003; Kirk et al. 2013; Baker et al. 2018), which may include non-mutualistic species. Additionally, some Symbiodiniaceae may

transform their mutualistic relationship into a parasitic state during a stressed period, thus parasitizing their hosts for nutrition (Baker et al. 2018). The symbiont benefits are clearly not equal to those of the host, host and this partnership could be lost under times of stress to either the host or the symbionts. In addition to this novel behavior- the phenomenon of symbionts taking advantage of the stressed hosts, might also hold a key to some urgent questions regarding the coral-algae symbiosis breakdown and contribute to our understanding of the mechanisms involved in the bleaching phenomenon. The discovery that Symbiodiniaceae migrate into the oocytes of bleached parts of partially-bleached MCE corals, but not into the healthy (non-bleached) parts, reveals the existence of opportunistic demeanor of the symbionts on their coral host's gametes, which may result in lowered functionality of the affected host parts. From an evolutionary point of view, this behavior is highly beneficial to the symbionts' survival throughout the bleaching period, since expelled symbionts are not expected to survive for long in the environment (Hill and Ralph 2007); but could also be beneficial for the host's persistence and adaptation to bleaching. Selection processes towards bleaching-resistant capabilities at the gamete level of healthy oocytes will result, in the long run, in adapted populations of this coral.

Acknowledgements

This is a preprint of an article published in *Coral Reefs*. The final authenticated version is available online at: <https://doi.org/10.1007/s00338-021-02082-1>

We thank the Interuniversity Institute for Marine Sciences in Eilat for the logistical support, and Barbara Colorni for help with the histological work. The comments of two anonymous reviewers greatly improved the manuscript. This study was supported by the Israel Science Foundation (ISF) No. 1191/16 to YL and by the European Union's

Horizon 2020 research and innovation program under the Marie Skłodowska-Curie post-doctoral grant agreement No. 796025 to GE.

Compliance with ethical standards

All samples were collected and treated according to the Israeli Nature and Parks Authority permit no. 2011/38249.

Conflict of interest

On behalf of all authors, the corresponding author states that there is no conflict of interest.

References

- Baird AH, Guest JR, Willis BL (2009) Systematic and biogeographical patterns in the reproductive biology of scleractinian corals. *Annual Review of Ecology, Evolution, and Systematics* 40:551-571
- Baker DM, Freeman CJ, Wong JC, Fogel ML, Knowlton N (2018) Climate change promotes parasitism in a coral symbiosis. *The ISME Journal* 12:921-930
- Baker EK, Puglise KA, Harris PT (2016) Mesophotic Coral Ecosystems: A Lifeboat for Coral Reefs? United Nations Environment Programme and GRID-Arendal
- Bongaerts P, Ridgway T ,Sampayo E, Hoegh-Guldberg O (2010) Assessing the 'deep reef refugia'hypothesis: focus on Caribbean reefs. *Coral reefs* 29:309-327
- Brown BE (1997) Coral bleaching: causes and consequences. *Coral reefs* 16:S129-S138
- Carpenter KE, Abrar M, Aeby G, Aronson RB ,Banks S, Bruckner A, Chiriboga A, Cortés J, Delbeek JC, DeVantier L (2008) One-third of reef-building corals face elevated extinction risk from climate change and local impacts. *Science* 321:560-563
- Eyal G, Tamir R, Kramer N, Eyal-Shaham L, Loya Y (2019) The Red Sea: Israel Mesophotic coral ecosystems. Springer, pp199-214
- Eyal-Shaham L, Eyal G, Tamir R, Loya Y (2016) Reproduction, abundance and survivorship of two *Alveopora* spp. in the mesophotic reefs of Eilat, Red Sea. *Scientific reports* 6:20964
- Eyal-Shaham L, Eyal G, Sakai K, Nozawa Y, Harii S, Sinniger F, Bronstein O, Ben-Zvi O, Shlesinger T, Loya Y (2019) Repetitive sex change in the stony coral *Herpolitha limax* across a wide geographic range. *Scientific reports* 9:1-12
- Eyal-Shaham L, Eyal G, Ben-Zvi O, Sakai K, Harii S, Sinniger F, Hirose M, Cabaitan P, Bronstein O, Feldman B (2020) A unique reproductive strategy in the mushroom coral *Fungia fungites*. *Coral Reefs*:1-12
- Fabina NS, Putnam HM, Franklin EC, Stat M, Gates RD (2013) Symbiotic specificity ,association patterns, and function determine community responses to global changes: defining critical research areas for coral-Symbiodinium symbioses. *Global Change Biology* 19:3306-3316
- Fine M, Gildor H, Genin A (2013) A coral reef refuge in the Red Sea .*Global Change Biology* 19:3640-3647
- Fitt W, Warner M (1995) Bleaching patterns of four species of Caribbean reef corals. *The Biological Bulletin* 189:298-307

Frade PR, Bongaerts P, Englebert N, Rogers A, Gonzalez-Rivero M, Hoegh-Guldberg O (2018) Deep reefs of the Great Barrier Reef offer limited thermal refuge during mass coral bleaching. *Nature Communications* 9:3447

Glynn P (1993) Coral reef bleaching: ecological perspectives. *Coral reefs* 12:1-17

Glynn PW (1996) Coral reef bleaching: facts, hypotheses and implications. *Global Change Biology* 2:495-509

Goreau TJ, Hayes RL (1994) Coral bleaching and ocean "hot spots". *Ambio-Journal of Human Environment Research and Management* 23:176-180

Grottoli AG, Tchernov D, Winters G (2017) Physiological and Biogeochemical Responses of Super-Corals to Thermal Stress from the Northern Gulf of Aqaba, Red Sea. *Frontiers in Marine Science* 4:215

Harrison PL (2011) Sexual reproduction of scleractinian corals *Coral reefs: an ecosystem in transition*. Springer, pp59-85

Hill R, Ralph PJ (2007) Post-bleaching viability of expelled zooxanthellae from the scleractinian coral *Pocillopora damicornis*. *Marine Ecology Progress Series* 352:137-144

Hoegh-Guldberg O (1999) Climate change, coral bleaching and the future of the world's coral reefs. *Marine and freshwater research* 50:839-866

Hughes TP, Rodrigues MJ, Bellwood DR, Ceccarelli D, Hoegh-Guldberg O, McCook L, Moltschanowskyj N, Pratchett MS, Steneck RS, Willis B (2007) Phase shifts, herbivory, and the resilience of coral reefs to climate change. *Current biology* 17:360-365

Hughes TP, Kerry JT, Álvarez-Noriega M, Álvarez-Romero JG, Anderson KD, Baird AH, Babcock RC, Beger M, Bellwood DR, Berkelmans R (2017) Global warming and recurrent mass bleaching of corals. *Nature* 543:373-377

Hughes TP, Anderson KD, Connolly SR, Heron SF, Kerry JT, Lough JM, Baird AH, Baum JK, Berumen ML, Bridge TC (2018) Spatial and temporal patterns of mass bleaching of corals in the Anthropocene. *Science* 359:80-83

Kirk N, Thornhill D, Kemp D, Fitt W, Santos S (201) Ubiquitous associations and a peak fall prevalence between apicomplexan symbionts and reef corals in Florida and the Bahamas. *Coral Reefs* 32:847-858

Knowlton N (2001) The future of coral reefs. *Proceedings of the National Academy of Sciences* 98:5419-5425

Knowlton N, Rohwer F (2003) Multispecies microbial mutualisms on coral reefs: the host as a habitat. *the american naturalist* 162:S51-S62

Kondrashov AS (1988) Deleterious mutations and the evolution of sexual reproduction. *Nature* 336:435-440

Krueger T, Horwitz N, Bodin J, Giovani M-E, Escrig S, Meibom A, Fine M (2017) Common reef-building coral in the Northern Red Sea resistant to elevated temperature and acidification. *Royal Society open science* 4:170038

Kushmaro A, Loya Y, Fine M, Rosenberg E (1996) Bacterial infection and coral bleaching. *Nature* 380:396

LaJeunesse TC (2020) Zooxanthellae. *Current Biology* 30:R1110-R1113

Lesser M, Stat M, Gates R (2013) The endosymbiotic dinoflagellates (*Symbiodinium* sp.) of corals are parasites and mutualists. *Coral Reefs* 32:603-611

Loya Y (2004) The coral reefs of Eilat—past, present and future: three decades of coral community structure studies *Coral Health and disease*. Springer, pp1-34

Loya Y (2007) How to influence environmental decision makers? The case of Eilat) Red Sea) coral reefs. *Journal of Experimental Marine Biology and Ecology* 344:35-53

Loya Y, Sakai K, Yamazato K, Nakano Y, Sambali H, van Woesik R (2001) Coral bleaching: the winners and the losers. *Ecology Letters* 4:122-131

Muller-Parker G, D'elia CF, Cook CB (2015) Interactions between corals and their symbiotic algae *Coral Reefs in the anthropocene*. Springer, pp99-116

Nir O, Gruber DF, Shemesh E, Glasser E, Tchernov D (2014) Seasonal mesophotic coral bleaching of *Stylophora pistillata* in the Northern Red Sea. *PloS one* 9:e84968

Osman EO, Smith DJ, Ziegler M, Kürten B, Conrad C, El-Haddad KM, Voolstra CR, Suggett DJ (2018) Thermal refugia against coral bleaching throughout the northern Red Sea. *Global Change Biology* 24:e474-e484

287 Pandolfi JM, Bradbury RH, Sala E, Hughes TP, Bjorndal KA, Cooke RG, McArdle D, McClenachan L,
 288 Newman MJ, Paredes G (2003) Global trajectories of the long-term decline of coral reef
 289 ecosystems. *Science* 301:955-958
 290 Peng S-E, Moret A, Chang C, Mayfield AB, Ren Y-T, Chen W-NU, Giordano M, Chen C-S (2020) A shift
 291 away from mutualism under food-deprived conditions in an anemone-dinoflagellate
 292 association. *PeerJ* 8:e9745
 293 Pinheiro HT, Eyal G, Shepherd B, Rocha LA (2019) Ecological insights from environmental disturbances
 294 in mesophotic coral ecosystems. *Ecosphere* 10
 295 R Development Core Team (2014) R: A Language and Environment for Statistical Computing. R
 296 Foundation for Statistical Computing, Vienna, Austria
 297 Rocha LA, Pinheiro HT, Shepherd B, Papastamatiou YP, Luiz OJ, Pyle RL, Bongaerts P (2018) Mesophotic
 298 coral ecosystems are threatened and ecologically distinct from shallow water reefs. *Science*
 299 361:281-284
 300 Rowan R (2004) Coral bleaching: thermal adaptation in reef coral symbionts. *Nature* 430:742-742
 301 Rundle HD, Chenoweth SF, Blows MW (2006) The roles of natural and sexual selection during
 302 adaptation to a novel environment. *Evolution* 60:2218-2225
 303 Shlesinger T, Loya Y (2019) Sexual reproduction of scleractinian corals in mesophotic coral ecosystems
 304 vs. shallow reefs. *Mesophotic Coral Ecosystems*. Springer, pp653-666
 305 Suggett DJ, Smith DJ (2020) Coral bleaching patterns are the outcome of complex biological and
 306 environmental networking. *Global Change Biology* 26:68-79
 307 Szmant A, Gassman N (1990) The effects of prolonged "bleaching" on the tissue biomass and
 308 reproduction of the reef coral *Montastrea annularis*. *Coral reefs* 8:217-224
 309 Trench R (1971) The physiology and biochemistry of zooxanthellae symbiotic with marine coelenterates
 310 III. The effect of homogenates of host tissues on the excretion of photosynthetic products in
 311 vitro by zooxanthellae from two marine coelenterates. *Proceedings of the Royal Society of*
 312 *London Series B Biological Sciences* 177:251-264
 313 van Oppen MJ, Mahiny AJ, Done TJ (2005) Geographic distribution of zooxanthella types in three coral
 314 species on the Great Barrier Reef sampled after the 2002 bleaching event. *Coral Reefs* 24:482-
 315 487
 316 Wooldridge SA (2010) Is the coral-algae symbiosis really 'mutually beneficial' for the partners? *BioEssays*
 317 32:615-625
 318 Wooldridge SA (2013) Breakdown of the coral-algae symbiosis: towards formalising a linkage between
 319 warm-water bleaching thresholds and the growth rate of the intracellular zooxanthellae.
 320 *Biogeosciences* 10:1647-1658
 321 Ziegler M, Eguíluz VM, Duarte CM, Voolstra CR (2018) Rare symbionts may contribute to the resilience
 322 of coral-algal assemblages. *The ISME Journal* 12:161-172
 323 Ziegler M, Arif C, Burt JA, Dobretsov S, Roder C, LaJeunesse TC, Voolstra CR (2017) Biogeography and
 324 molecular diversity of coral symbionts in the genus *Symbiodinium* around the Arabian
 325 Peninsula. *Journal of Biogeography* 44:674-686

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327 Figure 1: Partially-bleached *Alveopora allingi*, with micrographs of gonadal development. (a)
328 general colony view at 60 m depth; (b) partial-bleaching within a single genet; (c) male and
329 female gonads from the non-bleached (pigmented) part of the colony; (d) male and female
330 gonads from the visually bleached part of the colony; (e) magnification of the marked area in
331 section c, showing stage 4 healthy mature oocytes; and (f) magnification of the marked area in
332 section (d), showing several stage 4 severely damaged oocytes with many Symbiodiniaceae

the oocytes' cytoplasm. Scale bars as indicated above the scale-line. Oc: Oocyte, Sp: Spermaries, N: nucleus, Z: Symbiodiniaceae.

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Figure 2: Oocyte sizes of the mesophotic coral *Alveopora allingi* during a partial colony natural bleaching event. (a) Significantly larger stage 4 oocytes in the bleached parts of partly bleached colonies (t-test, $T=544$, $P<0.001$). Centerlines of the boxes represent the medians; box limits indicate the 25th and 75th percentiles; whiskers extend 1.5 times the interquartile range from the 25th and 75th percentiles, notches represent the 95% confidence interval for each median, all data represented by dots. Two different genet pairs of bleached ($n=84$ oocytes) and non-bleached ($n=21$ oocytes) parts were analyzed. (b) The difference in μm between median values of the bleached parts compared to non-bleached parts. The difference in condition is calculated from 1,000 bootstrap samples of the values, indicating the difference between the median oocyte size of the bleached parts and that of the healthy parts. The horizontal black line indicates the 95% confidence interval.