Response to reviewer's general comments:

Reviewer's Comment #1: *I have doubts that the manuscript serves the purpose of a review paper (in Biogeosciences) because in its present state it does not cover well the broad range of studies available in the literature about this topic.*

Author's Response #1: I totally agree. This is a perspective essay, whose intent is to test/challenge the consistency of the previously proposed CO_2-limitation induced warm-water bleaching model (viz. Wooldridge, 2009); with a particular focussed attention in this case (i.e. the present article), on the importance (or not) of the demographic parameters of the algal symbionts to the proposed bleaching sequence. It is thus not, and was never intended to be an exhaustive review of all the literature on coral bleaching. The Editorial staff @Biogeosciences need to assist in recommending the correct category for the manuscript given their current classification scheme.

Reviewer's Comment #2: *Many other studies have tested and given evidence for a tight link between the disruption of photosynthesis and the onset of bleaching.....The resultant (so-called) photoinhibition model of coral bleaching.....cannot be ignored in such a review focusing on the onset of bleaching. Impaired symbiont photosynthesis is a commonly accepted factor determining bleaching-susceptibility and should have been put in the right context along this review.*

Author's response #2: This paper contributes to previous efforts to demonstrate the consistency of the CO_2-limitation bleaching model with the considerable (but fragmented) understanding that exists for both biophysical and ecological aspects of warm-water coral bleaching. The present essay thus becomes most beneficial when considered from the fact that the proposed model has already been discussed in terms of its close alignment to the observed/predicted response characteristics arising from the well-established 'photoinhibition model' of coral bleaching (see, Wooldridge, 2009); i.e., the proposed CO_2-limitation bleaching model is in complete agreement with the downstream outcomes/expectations of the photoinhibition bleaching model, which has algal

photoinhibition, oxidative damage and host-cell disruption as underlying processes (Gates et al., 1992; Lesser, 1996; Jones et al., 1998; Warner et al., 1999). However, the CO_2-limitation bleaching model goes beyond the photoinhibition model to propose that in some (natural) cases, the photoinhibition response is initially triggered by a failure of the coral host to maintain a sufficient supply of CO_2 for its endosymbiont partner, particularly during periods of excess solar radiation when the photosynthetic demand for CO_2 is maximal.

As outlined by Wooldridge (2009) and summarised by Figure 1 (present paper), theoretical considerations do permit CO_2-limitation within the 'dark reactions' of photosynthesis to be proposed as a potential trigger for the classic bleaching sequence of photoinhibition, oxidative damage and zooxanthellae expulsion. In this case: (i) lack of CO_2 substrate required for the 'dark reactions' can reduce the rate of consumption of the products of photosynthetic electron transport (ATP and NADPH), subsequently causing the photosynthetic electron transport components of the 'light reactions' to become blocked (Takahashi and Murata, 2006); (ii) continued funnelling of excitation energy into the over-reduced electron transport chain can then trigger the onset of photoinhibition (Jones and Hoegh-Guldberg, 2001), damage essential photosynthetic components, (principally photosystem II, PSII), and generate damaging reactive oxygen species (ROS) (Lesser, 1996; Warner et al., 1999); and (iii) the excess production of ROS beyond the antioxidant defence strategies of the coral host (and zooxanthellae) has been linked to the host-cell necrosis and detachment that underpins zooxanthellae expulsion (Gates et al., 1992; Dunn et al., 2002). Importantly, this sequence of events is consistent with the understanding that the bleaching process begins with impairment of the CO_2-fixation mechanism within the zooxanthellae and that the severity of the bleaching impact is a direct function of light intensity (Jones et al., 1998).

Importantly, the outlined possibility for CO_2-limitation to be the upstream trigger of the photoinhibition response takes some of the onus off the algal photosynethic machinery (per se) as the 'weak-link' in the thermal bleaching sequence. This suggestion may explain why the majority of expelled zooxanthellae from thermally stressed corals remain

photosynthetically competent (Ralph et al., 2001). Indeed, when viewed from the perspective provided by the CO_2-limitation model, the easily identified 'Achilles' heel' of the bleaching response is the vulnerability of the supply chain of CO_2 for the zooxanthellae; which as explained by Wooldridge (2009, 2010) and summarised in Figure 1 (present paper) is heavily reliant on a tight-cycling of autotrophic carbon/energy.

Reviewer's Comment #3: *The whole concept of the study comes about based on the observation that after an initial expulsion of algal symbionts there is more photosynthetically fixed carbon being directed into algal cell multiplication.....in consequence, this reduction in photosynthate transfer will disrupt the carbon concentration mechanism of the host, leading to CO_2-limitation and ultimately leading to symbiont expulsion through a series of events that are downstream of those..... To start with, please allow me putting a very naive question.....if this linkage would exist, wouldn't symbiont cell densities increase just before bleaching occurs?*

Author's response #3: It is important to keep in mind that CO_2-limitation can occur because of: (i) an enhanced demand for CO_2, and/or (ii) a reduced supply of CO_2. Assuming constant Chl-a per algal cell, CO_2 demand will largely be a function of both zooxanthellae density and irradiance levels (i.e. high zooxanthellae densities combined with high irradiance levels equates to maximal CO_2 demand for carbon fixation). At high irradiance levels, Muscatine et al. (1989) explain that respiratory CO_2 is insufficient to meet the demands of the symbiosis, and dictates that the CO_2 supply chain becomes heavily reliant on host carbon concentrating mechanisms (CCMs) to convert abundant seawater (external) HCO_3^- into readily diffusible CO_2. Moreover, as outlined in Wooldridge (2009), the cellular operation of the host CCMs requires continuous energy in the form of ATP, which ultimately derives (over short time periods ~minutes) from the supply of fixed-carbon from the zooxanthellae. In this way, a tight-cycling of fixed carbon is crucial for a stable endosymbiosis.

The CO_2-limitation bleaching model (Wooldridge, 2009) implicates both an initial increase in demand of CO_2 (principally via enhanced irradiance levels) and a progressive (self-enhancing) decrease in CO_2 supply (via an ATP-dependent disruption of the CCMs) as the

triggers for CO_2 limitation and zooxanthellae expulsion. In this way, the narrative for a natural thermal bleaching event may be explained as follows:

Step 1: 'Doldrum' weather conditions leading to high irradiance levels (= high CO_2 demand) and calm conditions (= reduced passive supply of HCO_3^-/CO_2 across diffusive boundary layer; Smith and Walker, 1980) act to initiate CO_2-limitation within the zooxanthellae population (which will be exacerbated by any ambient condition(s) that has promoted an enlarged population, e.g. eutrophication, elevated pCO_2).

Step2: The intracellular CO_2-limitation will trigger an initial zooxanthellae expulsion which will continue only until CO_2 demand re-equilibrates with CO_2-supply (i.e. dynamic expulsion leading to smaller zooxanthellae population will act to lower CO_2 demand).

Step 3: However, as outlined in detail within the present paper, the extent to which such an expulsion process continues (and leads to mass bleaching) is ultimately govern by any associated disruption in the CCMs (i.e. bulk CO_2 supply mechanism). The proposed problem arises for situations in which a large number of zooxanthellae are expelled (per day) and then subsequently produced (per day). In this case, the increased respiratory cost of regrowth may lead to a negative autotrophic carbon balance (P/R <1) when integrated across the diurnal irradiance cycle. Such a situation implies a reduction (or cessation) in photosynthate transfer to the coral host, which may occur even when the zooxanthellae population density appears stable (= net zero growth). This inverse relationship between photosynthate transfer and symbiont MI(%) has been documented in corals (McGuire and Szmant, 1997), sea anemones (Verde and McCloskey, 1996) and jellyfish (Sachs and Wilcox, 2006).

Step 4: Any disruption (reduced efficiency) in the CCMS will trigger further zooxanthellae expulsion until the intracellular CO_2 demand is commensurate with the new (reduced) level of supply.

Step 5: During this process, however, the reduced zooxanthellae population would be exposed to a new set of symbiotic conditions that further reinforce the expulsion process.

First, due to reduced self-shading and greater skeletal reflection (Enriquez et al., 2005), the reduced population becomes exposed to higher per zooxanthellae irradiance, effectively lowering the external irradiance level at which destructive photoinhibition is initiated (Franklin et al., 2006). Second, the reduced population may experience a greater availability (i.e. less competition) of the essential nutrients (e.g. NH_4^+), which typically limit zooxanthellae growth rates. In combination with the prevailing elevated temperature, a nutrient surfeit would permit a greater proportion of the remnant zooxanthellae population to undergo mitosis, further reducing the already diminished transfer of photosynthates to the host. These symbiotic conditions would ensure that upon the next completion of cytokinesis (~9–12 h) (Wilkerson et al., 1983), the zooxanthellae demand for CO_2 will once again exceed the supply capacity of the symbiotic association, thereby initiating further 'pulsed' release from an already reduced zooxanthellae population. The dynamics of this reinforcing, stepwise reduction in zooxanthellae is consistent with the observational evidence showing that: (i) the MI(%) of the remnant zooxanthellae population increases despite a progressive reduction in absolute density (Jones and Yellowlees, 1997; Strychar et al., 2004), and (ii) mass bleaching does not occur as a single simultaneous release of the zooxanthellae population, but rather proceeds as a series of 'pulsed' releases that initiate a stepwise reduction in zooxanthellae numbers, with a ~9–12-h interval between the stepped pulses (Strychar et al., 2004).

Step 6: Once initiated, the destructive downward spiral is predicted to continue until the only zooxanthellae remaining *in symbio* are those that are maintained by a passive CO2 supply route, as evidenced by the fact that even corals that appear completely bleached white still maintain a zooxanthellae population ~10% of the normal density (Jones and Yellowlees, 1997). Yet, this remnant zooxanthellae population is expected to transfer only limited caloric benefit to the coral host. Apart from the reduced numbers, the continued high MI(%) will ensure limited transfer of photosynthate until the resumption of cooler (= lower division) temperatures. This leaves the coral host with limited resources, principally catabolism of its somatic tissue reserves (Szmant and Gassman, 1990), to maintain function and combat starvation.

Reviewer's Comment #4: *The correlation claimed (in this paper) to exist between bleaching thresholds and MI could as easily be explained by both parameters being a consequence of the same cause (e.g., thermal susceptibility of the photosynthetic apparatus).*

Author's Response #4: I hope that the above mentioned details of the CO_2-limitation model (as previously outlined in Wooldridge 2009) helps explain why I believe that the dynamics of the bleaching response are commensurate with a strong involvement of algae growth (MI) dynamics – not just as a secondary consequence. Indeed, I would go so far (as outlined in Wooldridge 2010) to suggest that the key inference arising from the CO_2-limitation bleaching model is that the maintenance of the coral-algae symbiosis is conditional on a continuous tight cycling of autotrophic energy, which in turn requires the algal symbionts to incur a 'fitness cost' in terms of their specific growth rate and population density. Wooldridge (2010) outlines the evidence to suggest that this fitness cost is enforced by the coral host, rather than benignly conferred by cooperating algal symbionts. Far from being unequivocally mutualistic, such symbiotic functioning is best explained in terms of a controlled parasitism whereby the coral host actively 'farms' its domesticated zooxanthellae in order to optimise the receipt of autotrophic energy. In this way, the thermal breakdown of the symbiosis (i.e. coral bleaching due to photodamage of algal photosynthetic apparatus) is reposed as a breakdown in the exploitative and captive measures of the coral host. In essence, this suggests that the photodamage of algal photosynthetic apparatus is 'a consequence leading to a cause' of coral bleaching; a subtle but important detail.

Reviewers Comment #5: *There is relevant evidence for an important role of the host.... It has been demonstrated that feeding can prevent heated corals from bleaching (see., Ferrier Pages et al. 2010, JEMBE). These and other studies suggest that there is a more advanced interdependence between host and its symbionts than conceptualised in the present paper, and imply that a reduction of photosynthate translocation from the symbionts to the host could likely be initially compensated by the host.*

Authors Response #4: Wooldridge (2009) explains the possibility (indeed likelihood) that at low to moderate levels of autotrophic disruption the host may retain the capacity to utilise stored tissue (e.g. lipid) reserves and/or heterotrophic feeding to help maintain the CCMs and thus forestall the onset of mass zooxanthellae expulsion (i.e. enhance bleaching resistance). This would help to explain results which highlight that feeding can help heated

corals to resist bleaching (see, Borell and Bischof, 2008; Ferrier Pages et al. 2010, JEMBE). However, apart from those coral species that are particularly well adapted for heterotrophic feeding, continued autotrophic disruption quickly leads to the depletion of tissue reserves (Szmant and Gassman, 1990; True, 2005). This fact is consistent with the natural thermal bleaching sequence for a population of massive *Porites* spp. in which *mass* expulsion of zooxanthellae only occurred upon depletion of tissue reserves below a common lower-threshold (True, 2005). A similar pre-bleaching sequence has also been noted for a branching *Acropora* spp. (Ainsworth et al., 2008). Indeed, this phenomenon may underpin empirical bleaching relationships that are characterised by specific temperature duration relationships (see e.g. Berkelmans, 2002). In this case, the enhancing impact of temperature on zooxanthellae MI(%) and subsequent declines in autotrophic capacity dictate that as SSTs rise, progressively less time is required before the host's energy storage reserves fall below the level that triggers the onset of bleaching. Intuitively, this effect will be tempered by the amount of storage material maintained by the coral, and may contribute towards the explanation for why thick-tissued corals (e.g. massive *Porites* spp.) are typically more resistant to thermal stress (Loya et al., 2001). For the extreme and rapid thermal stress that characterises many laboratory experiments, mass zooxanthellae expulsion appears to precede independently of host storage reserves, and may indicate: (i) the inability of the host to quickly mobilise its stored energy reserves; and/or (ii) the concerted action of the coral host to re-allocate the use of its energy stores towards other homeostatic processes.

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