Loss of coral reef habitat is often associated with transitions to benthic communities dominated by noncalcareous benthos \((4, 8)\). The results of Kayanne et al. (1) reveal metabolic measures to be sensitive indicators of reef health \((9)\), but the findings more likely reflect the condition of the local sampling site than the normal behavior of healthy reef communities.

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Response: We are grateful for the comments we have received about our report \(1\). They bring up points that we did not fully discuss in the report. Gattuso et al. stressed that our measured values of productions are remote from the common reef production rate. His reference of ‘‘standard performance’’ of \(P/I=1.0 \pm 0.1\) \((2)\). The standard performance of coral reefs are a first-order estimation; it is not a precise indicator with regard to the CO2 sink versus source problem. The threshold between CO2 sink or source is very sensitive to precise organic and calcium carbonate production rates, and thus direct measurement of \(P_{CO2}\) changes provides better information. We should obtain more direct measurements of \(P_{CO2}\) changes to better address the reef sink versus source problem. Moreover, the idea that \(P/I=1\) is partly based on the fact that the tropical ocean is depleted in nutrients for supporting net organic production. However, as we stated in our report, recent studies have revealed the importance of nitrogen fixation in coral reefs, which provides new nutrients to coral reefs.

As we stated in our report, different reef zones might act differently as to CO2 fluxes. The whole-reef production values in the region by Kinsey \(3\) were mainly estimated from lagoon water chemistry. In our report, we presented an example that shows how a reef flat might act as a sink of CO2, and suggested that reefs, in general, may not be sources of CO2.

On the basis of our earlier investigations, together with those by many other reef scientists on community structure of corals and other benthic organisms, we chose Shiraho reef to represent the common features of fringing coral reef. If one considers the pattern of community distribution and flow regime on Shiraho reef, then it is clear that our measurement of \(P_{CO2}\) changes shows average reef metabolism on this reef.

Buddemeier questions the health of our study site by referring to Kühmann’s review of Ishigaki Island reefs \(4\). Kühmann surveyed Ishigaki Island as a whole, but focused on the worst part of it in this review \(4\). In his original paper \(5\), however, he showed stepped stages of human impact and found that living coral coverage of our study site on north of Shiraho reef in table 1 in \(5\) was 70%, which represents the highest value of all the sites he investigated. He classified Shiraho reef as one of the ‘‘reefs with no signs of deterioration.’’ Planck et al. investigated our study site thoroughly and concluded that the Shiraho coral reef ecosystem is ‘‘world-class with respect to its diversity of corals and fishes’’ \(6\). Veron identified 359 species in Ishigaki and nearby islands (Yaeyama Islands), almost the same number as that found in the Philippine Islands. He furthermore ranked North Shiraho reef as preserving a high diversity of corals \(7\).

With regard to nutrient input which may alter reef community structure, we have measured inorganic nitrogen concentrations on the reef flat water of Shiraho reef \(8\). The concentrations of nitrate, nitrite, and ammonium were less than 0.4, 0.08, and 0.45 \(\mu M\), respectively, which contradicts Buddemeier’s statement that Shiraho reef might be strongly eutrophicated. On the basis of these studies, we think that Shiraho reef shows the typical community structure of healthy fringing coral reefs.

The other point Buddemeier raises is that the small amount of organic material preserved in reef sediments refutes the idea that reefs might act as a CO2 sink. However, the fate of organic products in coral reefs is not only sediment burial, but also transport to the outer ocean and biomass increase. We have measured a positive net primary production on the reef flat, which includes consumption and respiration. We think that most of the net organic products are washed out into the outer ocean. However, the fate of organic materials in the ocean is beyond the scope of our report. Its purpose was to point out that the reef flat at Shiraho—the most productive zone in reefs—acts as a sink of CO2. However, we cannot comment on the statement ‘‘export of DOC/POC from reef to ocean or export by downslope sediment transport would not provide a credible mechanism for differential sequestration’’ without conducting actual measurement of these fluxes.

Gattuso et al. also question the representativeness of our study site. We agree that our report did not provide adequate explanation of which part of the reef community metabolism our measurements represented in relation to the flow regime. During high tides, our current measurements showed that water comes steadily from north-northeast with a current speed over 5 cm s\(^{-1}\) [figure 2 in our report \(1\)]. This water comes from the outer ocean over the reef crest north of the study site and runs over turf algae and living coral communities [figure 1B in \(1\)]. During low tide stagnant periods, we observed a slow current with an average of 3 to 4 cm s\(^{-1}\), which mixed well the water around the study site surrounded by living coral patches. Therefore, our measurements likely show the metabolism of corals and algae on the reef flat at Shiraho. We are carrying on further measurements of currents at other points to clarify the flow regime.

Gattuso et al. also point out that errors of the mean daytime and nighttime \(P_{CO2}\), values overlapped with the offshore value. The purpose of this estimation was to convert the visually conspicuous relations of \(P_{CO2}\) and light intensity into a quantified discussion, but the data were too small to make a statistically rigorous conclusion; we agree that we should obtain more data in the future. However, we would like to point out that the light intensity in March, when we made the observations and estimations, is relatively low compared to other months in this island (mean light intensities are 552 and 864 \(\mu mol\ m^{-2} s^{-1}\) in March and in August, respectively). Productions depend primarily on light intensity, and we have obtained higher productions in August.

Gattuso et al. point out the discrepancy between our estimates of production and those of Nakamori et al. \(9\). Their estimates of organic production and calcium carbonate production, however, depend only on one daytime (4-hour) set and one nighttime (3.5-hour) set of measurements of pH and alkalinity changes. From these data, they estimated the whole day net production on the basis of the tentative assumption that the daytime length is 7.5 hours. After the publication of their study, we have accumulated numerous measurements on organic production and calcium carbonate production of Shiraho reef and have related them with actual change in light intensity. In addition to the conventional pH-alkalinity method to estimate the productions, we computed them from more measured alkalinity and \(P_{CO2}\) changes, six times through \(P_{CO2}\) observation.

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Self-Fertilization, Linkage Disequilibrium, and Strain in Plasmodium falciparum

We are impressed by the elegant data presented by R. E. L. Paul et al. (1) on the high rate of self-fertilization in Plasmodium falciparum, the agent of the most malignant form of malaria. This study contributes significantly to our knowledge on this pathogen's basic biology. Nevertheless, we find that there is a major logical gap in the conclusions arrived at by Paul et al. They appropriately argue that frequent selfing in P. falciparum is a medicinally relevant feature, for it should favor the maintenance of “multi-locus phenotype associations,” in particular those governing virulence, drug resistance, or variant surface antigen polymorphism. This is quite logical: Self-fertilization, by inhibiting genetic recombination, leads to a situation of actual clonality (2) (offspring genotypes that are identical to the parental cells), which should help stabilize those multi-locus associations that are elsewhere favored by natural selection. Then they state that “there was sufficient outbreeding to disrupt any linkage disequilibrium” (linkage disequilibrium is the non-random association between genotypes scored at different loci). These two proposals taken separately are conceivable, but they are incompatible to each other.

If self-fertilization, as evidenced by studying the three loci MSP-2, MSP-1, and GLURP, was unable to maintain any multi-locus association between these loci (as shown by lack of linkage disequilibrium at the three loci), it is not tenable that it could significantly help in stabilizing any other multi-locus combination. Two possibilities can be entertained. First, selfing can play in itself a significant role in maintaining multi-locus association, and this should be observed with the MSP-2, MSP-1, and GLURP loci. Second, the natural selection has the dominant role in stabilizing those multi-locus phenotypes associated with virulence, drug resistance, or “immunologically sensitive” variant antigens (a statement that is consistent with the observation of linkage equilibrium at other loci). In the latter case of variants mainly maintained by natural selection, the role of self-fertilization would appear consequently limited.

Another concern in the approach used by Paul et al. lies in the difficulty of evidencing any linkage disequilibrium with their data. Each of the three loci under study exhibits considerable allelic variation. The expected frequency of each possible multi-locus combination is therefore low, which proportionally lowers any possibility of evidencing linkage, even with exact statistical tests. This situation leads to a large type II error risk (to see no significant linkage while linkage does exist). If a conservative model is taken, in which five equiprobable alleles (much less than actually recorded in these data) are segregating at each locus, the probability of any multi-locus combination does not exceed 0.23^3 = 0.008. This renders difficult to evidence any significant linkage, unless considerable sample sizes are used, which is not the case in this study.

Although the discovery of high-rate self-fertilization in P. falciparum is a major breakthrough in our knowledge of the agent of malaria, its actual impact on this parasite's population structure in humans still has to be clarified by classical population genetic means that depend on linkage disequilibrium analysis. The notion of strain in microbiology relies on the existence of stable multi-locus associations (especially, of course, those combinations dealing with medically relevant characters), and if no such multi-locus associations are found in P. falciparum, the notion of strain has to be held in abeyance for this parasite. Should this be verified, any efforts for individualizing multi-locus genotype (that is, strain characterization) in P. falciparum may not be successful, for these genotypes will appear as most unstable. The only approach that remains possible in this case is the typing of individual genes.
Response: Coral Reefs and Carbon Dioxide
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