Syntrophic nitrogen exchange between zooxanthellae and host corals as viewed from amino acid nitrogen isotopes

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The reef-building coral is one of the most prominent animal-plant symbiotic systems, exhibiting extremely high primary production in oligotrophic tropical oceans. As an essential factor behind this, the existence of conservative internal recycling pathways of nutrients such as nitrogen (N) and phosphorus has been supposed; however, exact mechanisms of nutrient recycling still remain to be clarified. In normal animal-plant interactions, animals acquire N from plants through grazing, and plants can reuse N once animals excrete N as urea or ammonium. Are similar trophic linkages operating between zooxanthellae and host corals within the symbiotic associations? To address this question, we used compound-specific N isotope analysis of amino acids. Using amino acid N isotope signature (d$_{15}^{N}$-AA) of animal tissues, one can evaluate simultaneously the apparent trophic level (ATL) of the animal, and the d$_{15}^{N}$-AA of plants at the basis of food chain on which the animal depends. From the latter, the d$_{15}^{N}$ of dissolved inorganic nitrogen (DIN) on which the plants depend can also be estimated and used for N source evaluation. We collected specimens of Acropora pulchra and some other hermatypic corals from fringing reefs around Ishigaki Island and Sekisei Lagoon, southwestern Japan, separated zooxanthella cells and host coral tissues using centrifugation, and measured bulk and amino acid d$_{15}^{N}$ values using EA/IRMS and GC/C/IRMS, respectively. The d$_{15}^{N}$-AA patterns of zooxanthellae and host tissues from single colonies were quite similar to each other, and ATL mostly ranged between 0.9 and 1.5 (ATL of primary producers being defined as 1.0) with no significant difference between zooxanthellae and the host. There was no evidence that host corals graze on zooxanthellae. The bulk d$_{15}^{N}$ values of both zooxanthellae and host tissues were low in pristine sites and got higher in polluted sites, indicating that they directly reflected d$_{15}^{N}$ of external N sources available at each site. While pieces of A. pulchra colonies were incubated with Artemia as food source for 2 weeks, the bulk d$_{15}^{N}$ and d$_{15}^{N}$-AAs gradually increased towards those of Artemia, and the increase rates were similar between zooxanthellae and host tissues. ATL also increased from 0.97 to 1.33 (zooxanthellae) and 1.37 (host tissues), which indicates that zooxanthellae did not simply reuse excreted metabolites from the host. Our results suggest that zooxanthellae and the host share a common reservoir of amino acids, from which they synthesize proteins for their biomass. Sources of amino acids for this reservoir would depend on origins: DIN may be incorporated and synthesized into amino acids primarily by zooxanthellae, while external food sources may be caught and digested into amino acids primarily by the host. Irrespectively of origins, the most part of acquired N seems to be stocked in the common amino acid reservoir before being used further for biomass synthesis. Thus, N exchange pattern between zooxanthellae and the host coral would be different from those found in non-symbiotic plant-animal interactions, and can not be regarded as simple recycling. It rather seems to be an effective system for sharing resources from several different origins between zooxanthellae and the host, to enhance their survival and growth under high-energy, low-nutrient conditions.

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