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Chapter 36

A synopsis of coral restoration genetics

Iliana B. Baums

Department of Biology, Pennsylvania State University, 208 Mueller Laboratory,
University Park, PA 16802, USA
baums@psu.edu

INTRODUCTION

Reef-building corals are declining world-wide as a consequence of indirect and direct human activities including ship groundings, dynamite blasting, and global warming, leading to coral bleaching. Coral husbandry, the raising of corals in captivity, promises to provide a stock of corals that can be used to supplement local populations without the need to diminish neighboring populations. Additionally, genotypes may be bred in captivity to resist infectious diseases or have higher tolerance towards adverse environmental conditions. In restoration genetics, a new field that combines demographic and genetic approaches to restore natural populations, a theoretical framework is being developed to guide restoration projects. In a recent review, Baums (2008) adapted this framework developed for plants in terrestrial ecosystems to coral husbandry. The following is a short synopsis of that paper. In it, it was proposed that the design of ecologically and evolutionarily sound strategies to restore coral reefs through coral husbandry requires knowledge of adaptive, genotypic and genetic diversity of captive and wild coral populations (Box 1).

Corals employ an astounding array of reproductive strategies. The contribution of asexual and sexual reproductive modes to total recruitment varies (as yet unpredictably) over the range of species (e.g. *Pocillopora damicornis* and *Acropora palmata*) with consequences on local genotypic and genetic diversity (Miller and Ayre, 2004; Baums *et al.*, 2006). In most corals surveyed both inbreeding (including the extreme of selfing) and asexual reproduction contribute to local population structure (Table 1, Baums, in press). On larger scales, coral species often behave as metapopulations that consist of more or less independent subpopulations. Restoration genetics aims to restore the genetic diversity of species on all levels and molecular methods to study these types of diversity are available for an increasing number of coral species. These findings are directly applicable to coral husbandry in that they can guide collection, breeding and out-planting efforts. Initial collections for the purpose of establishing a captive breeding population should capture as much of the genetic diversity of the species as feasible. Breeding and out-planting efforts can have detrimental consequences on the long-term survival of the species mainly through two effects:

i). Inbreeding depression, a reduction in fitness due to mating of relatives, is reduced in naturally inbreeding plant species compared to outbreeders, but remains a concern during late life stages in captive and small restored populations (Husband and Schemske, 1996; Frankham, 2005). Data on inbreeding corals does not exist.

ii). Outbreeding depression can result from mating between distantly related individuals (through the breakdown of co-adapted gene complexes) or mating between individuals that are strongly adapted to local conditions (ecotypes). The likelihood of outbreeding depression decreases with increasing relatedness and increasing similarity of environmental background of parental genotypes ("ecotypes") but may not be detectable until the F2 to F6 generation (Hardner and Potts, 1997; Hammerli and Reusch, 2003). Because of the long life cycle of most coral species direct proof for outbreeding depression may be unobtainable but the risk of outbreeding depression may be reduced by matching parental ecotypes. Studies to date on the existence of ecotypes in corals addressed the holobiont (coral and zooxanthellate symbiont) through reciprocal transplantation experiments of corals. As one might expect, results included detection of species with generalist genotypes (Smith *et al.*, 2007), species that show site-adaptation (D'Croz and Mate, 2004), and species that harbor both generalist and specialist genotypes (Potts, 1984; Vermeij *et al.*, 2007). Future research should aim to separate the heritable contributions of all partners to the performance of the holobiont. To that end, experiments on symbiont-free gametes (Baums, in press) and facultative symbiotic species are promising study systems. In some instances, molecular tools can track captive organisms after reintroduction into the wild through fingerprinting and so assist in collecting data on performance of individuals. Consideration of genetic factors is essential because long-term success of restoration efforts may well be influenced by genetic and genotypic diversity of restored coral populations.

APPLICATION TO HUSBANDRY

The application of molecular tools to coral husbandry may include taxonomic determination of difficult species and the cataloging of genotypic and genetic diversity present in captive stocks. Molecular tools for taxonomic studies are increasingly available for adult (Concepcion *et al.*, 2006) and larval/recruit identification (Shearer and Coffroth, 2006). By utilizing the power of highly variable molecular markers such as microsatellites, identification of clonal replication can be achieved with high certainty (Maier *et al.*, 2001; Le Goff and Rogers, 2002; Le Goff-Vitry *et al.*, 2004; Magalon *et al.*, 2004; Miller and Howard, 2004; Severance *et al.*, 2004; Shearer and Coffroth, 2004; Baums *et al.*, 2005; Underwood *et al.*, 2006; Van Oppen *et al.*, in press). Production of coral larvae in captivity may soon be a reality in zoos and aquaria (several papers, this volume)

Box 1: Genetic and genotypic diversity

Restoration of remnant populations is analog, in some ways, to restoring biodiversity of an ecosystem. The common goal is to resurrect the sites' diversity in both relative and absolute abundance of its members. That is, species richness and evenness are matched to pre-disturbance levels. The building blocks of biodiversity are species with varying degrees of relatedness.

Genetic diversity can be defined on several levels in organisms that have sexual and asexual reproductive modes such as corals and plants (reviewed in Toro and Caballero, 2005). Genetic diversity *sensu strictu* (or gene diversity) refers to the amount of variation on the level of individual genes in a population. Genetic diversity may be expressed as heterozygosity or allelic richness. Genetic variation can be neutral or adaptive and different methods are used to detect and measure these two types of genetic diversity. In contrast, genotypic diversity is defined as the number of unique multilocus genotypes present in a population and varies on the level of whole organisms. A multilocus genotype (genet) may occur several times (ramets) in a population only as a result of asexual replication (identity by descent). The number and relative abundance of ramets from different genets determine the genotypic richness and genotypic evenness, respectively.

It is important to note that repetitive multilocus genotypes resulting from asexual reproduction should appear only once in datasets when analyzing genetic diversity (Halkett *et al.*, 2005); a practice difficult to implement when using markers with relatively low resolution of genotypes such as allozymes (commonly used in the coral literature).

Genotypic and genetic diversity describe fundamentally different processes that have to be managed separately and, as with biodiversity levels, genotypic and genetic diversity should be matched to pre-disturbance levels.

Matching origins between reared and wild stocks is an important goal in restoration genetics to avoid problems with altering gene flow patterns among populations, and inbreeding and outbreeding depression (Hufford and Mazer, 2003; Frankham, 2005; Van Buskirk and Willi, 2006). These potential pitfalls are discussed in the context of genetic studies on corals (Baums, in press).

and molecular tools maybe used to uncover the mode of production of these larvae. Because there are only a limited number of potential parents in the aquarium setting, assignment of parenthood is facilitated. Thus, aquaria provide a unique opportunity to study reproductive behavior of corals.

Captive environments are likely selective; only some genotypes may be suited to survival in aquaria (Petersen, pers. com.) and this may extend to *in-situ* nurseries (see Chapter 33). Light, water quality and settlement density in aquaria can differ substantially from natural conditions and thus may exert selective pressures on captive organisms. Fewer even of the genotypes capable of survival in captivity are able to reproduce sexually under these conditions (see this volume). Differential performance of genotypes in captive environments can be assessed using molecular tools to differentiate among genotypes and track them and their offspring over time. Captive rearing may thus significantly alter the genetic composition of the species under care. According to IUCN guidelines (IUCN, 2002) such selection should be minimized to preserve the genetic viability of the species though it is not clear how this can be achieved.

Species survival plans (www1) management approach for preserving the genetic diversity of threatened or endangered species in captive breeding programs. No plans are as yet developed for threatened corals but should such action become necessary initial collection efforts of brood stock should obtain material from a broad geographic range and from a variety of habitats. Maximizing the genetic variability in the brood stock is important to prevent inbreeding and bottleneck effects and to maintain the genetic integrity of the species.

OUTPLANTING OF CAPTIVE BRED INDIVIDUALS

Coral reef restoration efforts with coral stock that has been nursed either *in-situ* or *ex-situ* are well underway (Shafir *et al.*, 2006; Chapter 41; Chapter 44) and are likely to become more common. Projects proceed in the absence of coral-specific guidelines for transplantation and nursery efforts. If experiences gathered during forest restoration efforts are an indication, unmanaged restoration could lead to long-lasting unwanted changes in the ecosystem (Konig *et al.*, 2002; Lefevre, 2004).

The source of the transplantation stock varies but usually a few animals from nearby reefs are brought into nursery facilities where they are extensively fragmented, mounted on stalks, grown out, and then outplanted. In some instances (see Chapter 41), quarantine procedures and visual health assessments aim to reduce the introduction of pathogens into the native habitat as recommended by IUCN SSC (2002). The efficacy of such visual assessments, while sensible, has not been shown, and further research is needed. We know little about the genotypic and genetic diversity of most coral species, making it difficult to match host and symbionts genetic and genotypic diversity to pre-disturbance levels. Conceivably, planting ramets (generated in the initial phase of a restoration project) of the same genet in close proximity may decrease sexual output as gametes have a reduced chance of encountering non-self gametes for successful fertilization in outcrossing species (Allee effect). It thus would be prudent to track ramets and genets from collection through outplanting. Subsequent monitoring of outplanted colonies for which their clonal identity is known may reveal differential performance of genotypes in the restored environment.

In conclusion, it is recommended that coral specific guidelines be developed because genetic structure of natural coral populations could be unintentionally but irreversibly altered by inadequate restoration and husbandry efforts.

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