

BOX 2 | Experimental considerations for future OA and reproduction studies

Improve field research capability

- Spawning timing, duration, and intensity in natural settings are not easily predictable, and we lack baseline reproduction knowledge for most marine species. Additionally, physiological and reproductive responses of laboratory-raised organisms may be compromised by stress associated with captivity.
- Research initiatives should collect baseline knowledge. Technologies should be developed and integrated with biophysical models to track gametes and larvae in the natural environment.

Measure multiple reproductive traits simultaneously

- Reproduction is a complex process. It is important to examine multiple reproductive traits simultaneously to get a better understanding of overall reproductive performance.
- Reporting measures of reproductive success that integrate multiple traits will improve the predictability of demographic models (ex. fecundity and survival of mother, survival of offspring).

Incorporate diversity in test subjects and experimental design

- Studies should maximize genetic variation, as organism origin (ie. field site, hatchery) and genotype can affect sperm and egg compatibility and confound findings.
- Future work should incorporate increased taxonomic diversity and multiple stressors to expand the breadth of species and conditions studied

Account for length of gametogenic cycles

- Gametogenic cycles vary between species. Many studies only expose organisms to OA when gametes are almost fully developed.
- Accounting for gametogenic cycle length improves understanding of OA effects on gamete development and parental effects.

Test multiple sperm concentrations

- The probability of detecting a treatment effect may be dependent upon sperm concentrations. “Optimal” sperm concentration may be higher than ecologically relevant concentrations. Low sperm concentrations can exacerbate OA effects.
- Experiments should use multiple sperm concentrations and gamete incubation times, as these factors may alter the probability of fertilization and/or polyspermy.

Understand limitations of intergenerational studies

- Artificially-induced fertilization methods (ie. strip spawning, chemical/hormonal injection, etc.) used in intergenerational studies may cause organisms to release gametes before complete gametogenesis.
- Many intergenerational OA studies inherently include adult exposure during reproduction, so those studies formally measure and report reproduction metrics.
- Future research should differentiate between within-generation carryover effects, cross-generational impacts (F0-F1), and multigenerational plasticity (F1-F2+), and determine how reproductive conditioning may impact each facet of plasticity.

See previous reviews for additional challenges in the design and execution of ocean acidification experiments (Albright et al. 2011, Grazer and Martin 2012, Przeslawski et al. 2017).