Moving Corals Reef-based Coral Larval Restoration – Standard Operating Procedure

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Location	**Traditional Owner Group**
Heron Island, One Tree Island	Bailai, Gurang, Gooreng Gooreng, Taribelang Bunda
Palm Islands, Townsville	Manbarra, Bindal
Lizard Island	Ngurruumungu, Dingaal

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1 Executive Summary

The Moving Corals Sub-Program of the Reef Restoration and Adaptation Program (RRAP) is a reef-based research and development project that was established to increase the scale and efficiency of coral larval restoration outcomes. The research aimed to develop innovative methods, while also adapting and testing existing methods, to increase larval supply and enhance settlement and recruitment of corals on damaged reef areas on the Great Barrier Reef. We achieved these outcomes by collecting wild coral spawn, mass culturing >180 million competent genetically diverse larvae, and deploying millions of coral larvae to increase settlement and recruitment in a range of reef environments. Innovative methods developed during the Moving Corals project include high resolution hydrodynamic modelling and particle tracking to optimise spawn slick collections and larval releases, drone operations for searching and locating spawn slicks, industrial scale spawn sucker collections, larval staining for tracking larval deployments, larval seedbox development and application for delaying larval release close to the benthos and increasing the scale of impact of settlement, and micro-imaging to automate workflows and detect new settlers directly on reefs. Results from this project can now be applied to enhancing ecologically meaningful restoration outcomes at larger scales on damaged reefs in future.

Specifically, the Moving Corals program focused on reef-based collections of spawned gametes and embryos from wild coral spawn slicks and mass culture of larvae for restoration by developing and optimising methods to:

1. Efficiently collect large quantities of gametes and embryos in open reef systems.
2. Mass culture larvae at high densities to maximize the production of competent larvae that are ready to settle.
3. Deploy larvae at scale using cost-effective approaches including free larval releases and targeted releases, and settlement onto devices to increase settlement rates prior to deployment on reefs.

This report presents details of the methods and equipment used during the five phases of larval restoration including:

1. Rapid gravid coral checks to predict coral spawning periods and innovative hydrodynamic modelling to optimise spawn slick collection on source reefs
2. Collection of many millions of spawned gametes and early embryos from surface slicks after coral spawning events using active and passive collection methods
3. Mass culture of many millions of genetically diverse reef-coral larvae in floating larval culture pool systems and in tanks on vessels
4. Deployment of larvae onto a range of reef areas with reduced coral cover using a range methods including innovative larval seedboxes, and settlement of larvae onto limestone tiles and other devices in larval pools to increase settlement rates prior to deployment of settlers onto reefs (coral seeding)
5. Monitoring of initial larval settlement and longer-term monitoring to determine coral recruitment outcomes using newly developed macrophotogrammetry methods.

This report presents guidelines for reef-based coral larval restoration procedures that can be used by a wide range of restoration practitioners, reef managers, Traditional Owners and other key stakeholders to scale up larval-based coral restoration and enhance recovery of reef-coral assemblages on damaged reef systems with insufficient larval supply and recruitment for natural recovery.

2 Background

The ongoing and accelerating loss of foundation reef corals on many reefs around the world, including areas of the Great Barrier Reef (GBR), has severely depleted breeding populations, leading to reduced reproductive output and larval supply (e.g., Hughes et al. 2019) – both critical for natural reef recovery. Therefore, enhancing larval supply to increase larval recruitment is increasingly important for restoring reefs that are experiencing insufficient natural larval supply for recovery (reviewed in Omori 2019, Randall et al. 2020, Banaszak et al. 2023, Harrison 2024a). Furthermore, culturing larvae from corals that have survived recent bleaching events is likely to increase the environmental tolerance of their offspring and enhance resilience in restored populations (Quigley 2024). Understanding when and how reef corals reproduce, and the biology and ecology of their larvae, are essential for developing effective restoration methods and for managing coral reef ecosystems, so a brief introduction to coral life cycles and sexual reproduction is provided below.

2.1 Introduction to reef coral sexual reproduction and life cycles

Scleractinian reef-building corals have a biphasic life cycle consisting of a microscopic planktonic and dispersive planula larval stage that settles and metamorphoses into a polyp form, from which the predominantly sessile and colonial adult corals grow until they become large enough to develop gametes for sexual reproduction and larval production (Harrison and Wallace 1990, Richmond 1997, Gleason and Hoffman 2011, Harrison 2024a). The majority of reef corals that have been studied so far are broadcast spawning hermaphrodites (producing both eggs and sperm within the same polyps and colonies), with fewer gonochoric (separate sexes) broadcast spawning corals known, and a smaller number of brooding corals that develop planula within their polyps recorded globally (Baird et al. 2009, Harrison 2011, Harrison 2024a). On the GBR, more than 90% of the nearly 200 coral species studied to date are broadcast spawners and most of these are hermaphrodites (Harrison 2024a), with most but not all of these species spawning during synchronous multispecies mass coral spawning periods on the GBR that occur during a few nights after full moon periods from October to December each year (Harrison et al. 1984, Willis et al. 1985, Babcock et al. 1986, Oliver et al. 1988, Baird et al. 2021).

Spawned gametes are often released in highly buoyant egg-sperm bundles that float to the sea surface where the bundles break apart enabling cross-fertilization with gametes from other colonies of the same species (Oliver and Babcock 1992, Willis et al. 1997, Mumby et al. 2024), and occasionally hybrids form from cross-fertilization of eggs and sperm from genetically compatible corals that are traditionally classified as separate species (Willis et al. 1997, Chan et al. 2019, Lamb et al. 2024). During mass coral spawning events large surface slicks can form at the sea surface that include billions of gametes and fertilized embryos, and these coral spawn slicks can be kilometres long and may persist until the following morning in periods of calm weather (e.g., Oliver and Willis 1987, Wolanski et al. 1989, Doropoulos et al. 2019a). The embryos develop over about 24 hours until they develop cilia at which stage they are termed planula larvae (Harrison and Wallace 1990). Most coral larvae are planktonic and are dispersed by currents during their 3-6 day development period until they become fully formed and competent to settle, by which time many larvae have been dispersed away from their natal reefs and may settle on other reefs downcurrent, or be entrained in eddies and settle on natal reefs, or die before they can settle (Harrison and Wallace 1990, Jones et al. 2009, Figueiredo et al. 2013, Hock et al. 2019, Randall et al. 2024). In addition, some coral larvae remain competent to settle for many weeks and up to 2-4 months after development, providing increased potential for longer-distance dispersal and metapopulation connectivity between separate reef systems (Richmond 1987, Nozawa and Harrison 2002, Wilson and Harrison 2005, Graham et al. 2008, Randall et al. 2020, 2024, Kininmonth 2024).

Competent larvae search for suitable settlement sites and then attach and metamorphose into coral polyps and develop a ring of tentacles around the polyp mouth for feeding and start to secrete their calcium carbonate exoskeleton (Harrison and Wallace 1990, Randall et al. 2020). Many and sometimes most of the newly settled coral polyps die, due to process of competition, predation, sedimentation, and density-dependent processes (e.g., Doropoulos et al. 2016, 2017, dela Cruz and Harrison 2017, Wakwella et al.

2020). Survival of settled polyps increases substantially when they acquire microscopic Symbiodiniaceae photosymbionts (often referred to as zooxanthellae) that provide energy for increased growth and survival. When the settled polyps acquire sufficient energy and grow large enough, they begin to divide by asexual budding to form new polyps, and the colonies gradually increase in size and complexity until they become visible recruits after about 9-14 months after settlement (Gleason and Hoffman 2011, Doropoulos et al. 2015, 2016, dela Cruz and Harrison 2017). If colonies continue to survive and grow they become sexually mature within 2-4 years for fast-growing branching corals and over longer periods for slower growing massive corals, thereby completing the life cycle (dela Cruz and Harrison 2017, Doropoulos et al. 2019a, Randall et al. 2020, Harrison et al. 2021, Banaszak et al. 2023).

Coral larval restoration processes align with these sexual reproductive stages and can be grouped into five contiguous phases (Omori & Iwao 2014, dela Cruz and Harrison 2017, Doropoulos et al. 2019a, Doropoulos 2022, Roff et al. 2023, Harrison 2024a) consisting of:

1. Predicting coral spawning periods and identifying breeding corals through gravid coral checks and
2. Collecting coral spawn
3. Rearing coral larvae
4. Settling coral larvae
5. Monitoring settlement, growth and survival of polyp settlers, juveniles and recruits through to adult breeding stages.

This report provides guidelines for successful reef-based coral larval restoration methods in each of these five phases that can be used by a wide range of stakeholders to cost-effectively collect wild spawn, mass culture many millions of genetically diverse coral larvae, and release and settle larvae for increasing larval supply and recruitment of foundation reef-building corals on damaged reefs that lack sufficient larval supply to ensure natural recovery. These methods include innovative approaches developed during the *Moving Corals Sub-program* as well as the adaptation of some existing methods to scale up larval production and delivery. To place these outcomes in perspective, a brief history of reef-based larval restoration research and the evolution of different methods and equipment is provided below.

2.2 Development of reef-based coral larval restoration

The reef-based coral larval restoration concept was first conceived in the early 1980s, following the discovery of the mass coral spawning phenomenon on the Great Barrier Reef in 1981 (Harrison et al. 1984) and the first recorded mass coral bleaching event that impacted the Central GBR a few months later in early 1982. Mass coral spawning provided predictable access to billions of genetically diverse gametes enabling efficient collection of many millions of eggs and embryos for larval rearing, and the subsequent mass bleaching event highlighted the ecological fragility of reef-building corals to stress and rapid environmental changes. An initial trial rearing larvae from samples of coral spawn in a conical larval culture plankton net system supported within an inflated truck inner tube at Yunbenun (Magnetic Island) in the early 1980s showed that reef-based larval culture was feasible (Harrison, unpubl.), but funding for research to test and develop this approach were not available until much later.

In 1997, Heyward et al. (2002) completed the first published study on Ningaloo Reef in Western Australia, rearing coral larvae from wild spawn slick samples containing about 4 million *Acropora* spp. eggs in 2 x 2 m floating culture ponds, with ~5% larval survival after 6 days. Competent larvae were then released from the pools via a pipe into 1.8 x 1 m tent enclosures on the reef, resulting in 100-fold greater settlement on terra cotta tiles in the larval seeded plots compared with natural control plots when monitored ~6 weeks later (Heyward et al. 2002).

Beginning in 2002, research in Japan tested various coral larval culturing methods using laboratory tanks and floating larval ponds (Omori and Iwao 2014, Omori 2019). Early at-sea trials used modified 2 × 2 m fish farming ponds to rear ~420,000 larvae per unit, releasing ~1.6 million larvae into net enclosures, resulting in 2,000–5,000 settlers per block and 34–60 juvenile survivors after 18 months (Omori et al. 2004, 2007). Suzuki et al. (2012) cultured larvae of two *Acropora* species in tanks, transferred them in plastic bags to reefs, and found initial settlement increased with larval density, though survival remained low after six months. From 2012–2018, Suzuki et al. (2020) developed a 'larval cradle' system combining spawn

collection and larval rearing, enabling the capture of millions of eggs and high larval survival within mesh nets. Settlement onto artificial substrates yielded 20% survival after six months when placed on elevated reef racks.

Larval restoration research was initiated in Palau in 2008, using ~1 million ex situ reared *Acropora digitifera* larvae which were settled onto concrete pallet balls enclosed in fine mesh nets (Edwards et al. 2015). Significantly higher densities of settled larvae were recorded on tiles on the larval seeded treatments compared with controls without larvae. However, subsequent monitoring after 30 weeks and 13 months after settlement showed no significant differences in densities of juvenile *Acropora* between larval seeded and control treatments (Edwards et al. 2015).

Extensive reef-based larval restoration research in the Philippines began in 2012, with early trials demonstrating significantly enhanced settlement of *Acropora tenuis* larvae reared ex situ and deployed under mesh enclosures on degraded reefs (dela Cruz and Harrison 2017). Longer-term monitoring confirmed that larval enhancement led to higher densities of sexually mature adult corals after three years, unlike control sites, with similar results observed for *A. loripes* (dela Cruz and Harrison 2017, 2020). A high-density trial in 2016 using ~1.6 million larvae—sourced in part from the first spawning of earlier restored colonies—resulted in high initial settlement, rapid growth to maturity, increased coral cover, and greater abundance of reef fishes in larval restoration plots (Harrison et al. 2021). Since 2015, large-scale methods for spawn collection and larval rearing have been developed using floating larval pools, net systems, and boom-based spawn catchers, enabling cost-effective mass production of larvae from multiple coral species (Harrison et al. 2021, 2024; Harrison and dela Cruz 2022). These efforts have restored diverse coral assemblages on degraded reefs, with restored populations now supplying larvae to nearby areas enabling natural recruitment (Harrison et al. 2021, 2024).

Larval-based restoration trials began on the Great Barrier Reef (GBR) in 2016, with initial deployments of ~1 million *A. spathulata* larvae showing successful settlement, growth to sexual maturity within five years, and increased coral cover in high-density treatments (Harrison 2017, Harrison et al. unpublished). Subsequent research introduced improved floating booms and larval pools for mass culturing tens of millions of genetically diverse larvae from wild spawn slicks, significantly enhancing larval settlement compared to controls (Harrison and dela Cruz 2022, McLeod and Hein et al. 2022, Harrison 2024b). These programs also built local capacity through training with Marine Parks staff, Traditional Owners, and tourism operators (Harrison et al. 2020). Large-scale innovations at Heron Island in 2018 included the first ever vessel-based industrial scale larval culture systems (Doropoulos et al. 2019a,b). Robotic vehicles were developed and tested for spawn collection and targeted larval delivery across multi-hectare reef areas (Harrison and Dunbabin 2019; Dunbabin et al. 2020; Mou et al. 2022). Since 2021, new equipment and small-vessel based spawn collection methods have further enhanced larval culture and delivery efficiency, with these techniques now applied across projects in the GBR, Philippines, and Maldives (Harrison 2024b).

Building on from and identifying critical gaps from previous work and current work occurring globally, the RRAP Moving Corals Subprogram has enabled the development, testing and application of a range of innovative methods. These are largely focussed on operational scaling and conservation decision making, including high resolution hydrodynamic modelling and particle tracking to optimise spawn slick collections and larval release strategies, drone operations for searching and locating spawn slicks, industrial scale spawn sucker collections, larval staining for tracking larval deployments, larval seedbox development and application for delaying larval release close to the benthos and increasing the scale of impact of settlement, and micro-imaging to automate workflows and detect new settlers directly on reefs. Together, these methods significantly enhance large-scale larval restoration outcomes that can be applied to future reef restoration projects. The reef-based coral larval restoration method is now mature, highly cost-effective and scalable, and is adaptable to a wide range of reef environments and use by diverse stakeholder groups and organisations.

3 Objectives and Scope

This report provides detailed guidance on standard operating procedures and equipment that are used for reef-based coral larval restoration. The target audience includes coral and reef restoration practitioners, reef managers, Traditional Owners and local communities, reef tourism operators, marine industries, and other relevant stakeholders that are involved with or planning to do coral restoration on the Great Barrier Reef and in other reef regions where damaged and degraded coral reefs require enhanced larval supply to enable recovery of foundation coral assemblages.

The SOP is divided into five sections corresponding to the five main phases of larval restoration (Omori & Iwao 2014, Doropoulos et al. 2019a, Harrison et al. 2021, Doropoulos 2022, Roff et al. 2023, Harrison 2024a). Each Phase has a number of component procedures that are numbered sequentially to enable practitioners to identify relevant components in each of the restoration phases. Within each component, information is provided on Background rationale, Procedure, Prerequisites, Risks and Hazards, Equipment and Materials and the Implementation steps. Greater levels of information are provided for innovative methods newly developed during the Moving Corals project, and key details are provided where standard operations have used previously developed processes and equipment from other projects that are described elsewhere (see Harrison and dela Cruz 2022, and Harrison 2024b for further details).

4 Phase 1 Gravid coral checks and hydrodynamic modelling to predict optimal locations for coral spawn collections on source reefs

4.1 Gravid coral checks to determine source reefs for coral spawning

4.1.1 Background and rationale

Coral larval restoration relies on access to spawned gametes for fertilization and mass larval culture, therefore identifying source reefs with healthy reproductive broadcast spawning corals and assessing the maturity of their eggs and sperm are needed to predict likely periods of spawning for spawn collection. Reef corals exhibit a range of gametogenic cycles, but most broadcast spawning species studied so far have a single annual cycle of egg and sperm development each year with eggs developing over about 5-9 months and sperm usually develop over shorter periods (Harrison and Wallace 1990, Randall et al. 2020, Harrison 2024a). During the latter stages of egg development, the maturing oocytes often become increasingly pigmented and in the last few weeks before maturation and spawning they often become pink to red coloured or may exhibit other colours including yellow, orange, green, purple or sometimes blue etc. (Harrison et al. 1984, Babcock et al. 1986, Roff et al. 2023, Harrison 2024a). Strongly pigmented eggs indicate that spawning is likely to occur within the current or next lunar cycle and often occurs after the full moon period on the GBR, therefore gravid coral checks of many replicate colonies of different species enable the likely spawning period and extent of spawning to be predicted. However, it is important to note that some corals spawn white or slightly cream coloured mature eggs (Babcock et al. 1986), and pigmentation patterns can vary within and among populations of different species, requiring repeated monitoring of gravid corals to best predict spawning periods.

4.1.2 Procedure

Gravid coral checks involve systematic surveys of healthy corals at different reef sites to quantify the numbers of colonies in different populations that are reproductive (gravid), the size and any colouration of eggs and spermaries within broken sections of polyps, and usually also involve photo-identification records to identify the coral taxa sampled. Gravid coral checks involve sampling coral colonies by carefully breaking branches or fracturing small areas of the coral to reveal the interior areas of the polyps including the mesenteries where the gametes develop (Harrison and Wallace 1990).

4.1.3 Prerequisites

Knowledge of coral taxa is helpful for identifying coral species underwater, but if good quality photographs showing distinctive taxonomic features of the coral are taken during sampling, many coral taxa can be identified from the images.

4.1.4 Risks and Hazards

Gravid coral surveys are done in situ underwater and usually require scuba diving and/or snorkeling if the reef site is relatively shallow. All diving and snorkel operations have associated risks that require personnel to be fit and competent for diving and/or snorkeling, and Australian organisations have minimum standards required for these activities including ADAS certification of the dive leader/coordinator, annual commercial dive medicals and servicing of dive gear, first aid training and experience with reef work by the dive team. Gravid coral checks can be done on snorkel in shallow reef areas but the risks of shallow water blackout need to be considered carefully, and in most cases gravid coral surveys of large numbers of colonies are more efficiently completed using scuba diving operations which also allow careful sampling of coral colonies to minimize stress and damage.

4.1.5 Equipment and Materials

For branching corals and species with relatively fragile skeletons, sampling can be done using surgical bone cutters, side cutters, long-nose pliers or small masonry cold chisels to carefully break branches or small sections of the colony to reveal the tissues and developing oocytes and/or spermaries within the coral polyps. For massive corals sampling is usually done using a fine wedge-shaped masonry cold chisel and hammer. A small waterproof camera with macro- or microimaging (e.g. Olympus TG 6/7 series compact camera) is also important so that images of each sampled colony and the size and colours of the developing oocytes/spermaries can be recorded and saved for permanent storage. If the sampled coral colonies are planned to be repeatedly monitored, waterproof numbered colony tags (e.g. cattle tags, stainless steel tags) and cable ties or nails can be used to temporarily tag and identify each colony or be attached onto on the reef substrata adjacent to the colony. An underwater notebook or recording sheet can also be useful for recording additional data.

4.1.6 Implementation steps

The following standard operations are usually used for routine gravid coral surveys and to determine source reefs with healthy breeding coral populations that can be used for collecting coral spawn.

1. Identify healthy breeding sized coral colonies that are known to broadcast spawn gametes. The size and age at sexual maturity varies between coral species so it is best to sample colonies larger than ~15-20 cm diameter that are more likely to be sexually mature (Harrison and Wallace 1990, Randall et al. 2020, Roff et al. 2023). In addition, it is important to sample areas away from rapidly growing branch tips or close to colony margins as these regions may be sterile or have lower fecundity (Wallace 1985).
2. Where possible identify and record the species/genus or lowest taxonomic category of coral to be sampled and take photographs of the colony prior to sampling and close-up photos of areas of the coral that show the size and characteristic features of the polyps and surrounding areas and underlying skeletal morphology that can be used to identify the species or genus.
3. For branching corals, carefully insert bone cutters or pliers or other sampling instruments down into the lower part of the branch and gently twist or cut the branch to reveal the skeleton and polyp sections (Fig. 4.1.1).

Figure 4.1.1: Moving Corals team member sampling gravid Acropora branching coral colony with long-nosed pliers to record details of developing eggs and spermaries within the polyps (Photo: D. McShane SCU)

4. Examine the broken section of the branch on the colony and record/photograph with microscope mode, the broken section to confirm the presence/absence of developing eggs and spermaries and their colour (Harrison et al. 1984, Babcock et al. 1986). Examine the base of the broken section of the branch to confirm details.

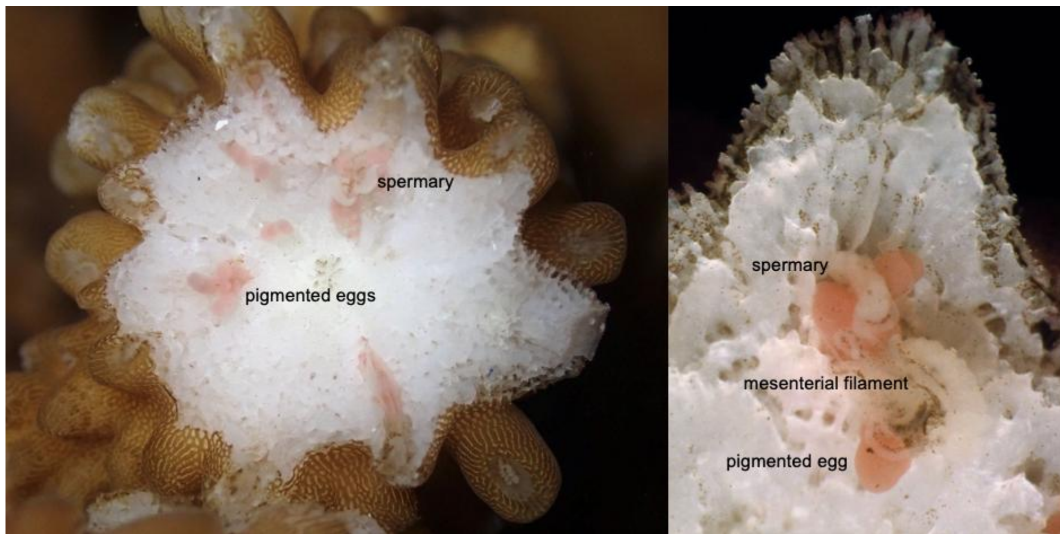


Figure 4.1.2: Broken section of gravid Acropora branching coral colonies showing the arrangement of mature pigmented pink and red eggs on mesenteries and large cream-coloured spermaries on adjacent mesenteries and associated mesenterial filaments within the polyps (Photos: P. Harrison, from Harrison 2024b).

5. Carefully reinsert the broken section of the branch into the colony branch matrix to allow the tissues to regrow and fuse so that damage is minimised and spawning biomass is retained (or samples can be collected for fecundity, taxonomic and genetic analyses).
6. If no obvious developing gametes are present in the first sampled branch, sample up to two additional branches of the colony to confirm the reproductive status of other areas of the colony.
7. Continue sampling a minimum of 10-20 colonies of the same species to quantify the reproductive status of the population.

Sampling massive corals

8. Sampling colonies of massive species requires careful planning to minimise damage, and sampling areas with irregular growth can enable small sections ~2-3 cm diameter of tissues and skeleton to be removed more easily than from other areas.
9. Identify the colony to be sampled and/or photograph the colony prior to sampling and take close-up photos of areas of the coral that show the size and characteristic features of the polyps and surrounding areas and underlying skeletal morphology that can be used to identify the species or genus.
10. Massive corals typically have dense skeletons and therefore sampling with a fine wedge-shaped masonry cold chisel and hammer is best to remove the sample for assessing reproductive status.
11. Examine the broken sections of polyps on the colony and on the sample removed and record/photograph with microscope mode the broken sections to confirm the presence/absence of developing eggs and spermaries and their colour (Fig. 4.1.3).
12. Carefully reattach the sample to the colony to enable tissues to repair and regrow to reduce stress and avoid the loss of spawning biomass from the colony, or collect the sample for fecundity, taxonomic and genetic analysis.
13. Continue sampling a minimum of 10-20 colonies of the same species to quantify the reproductive status of the population.

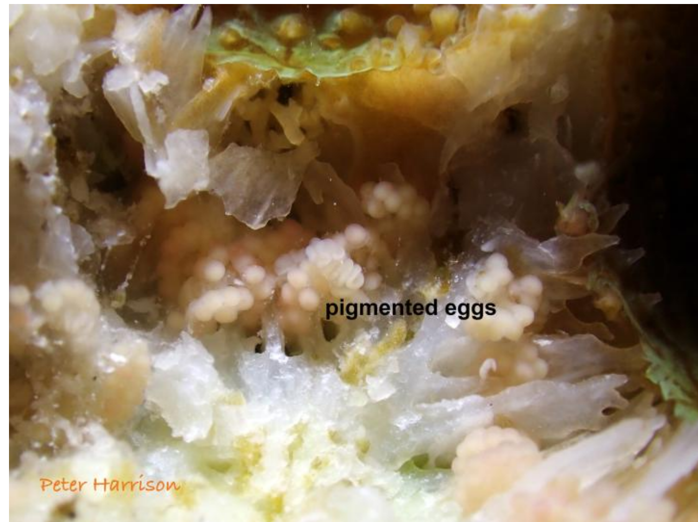


Figure 4.1.3: Broken section of gravid merulinid brain coral showing clusters of maturing lightly pigmented developing oocytes on mesenteries (Photo: P. Harrison).

Once gravid coral checks are completed, collate the data to assess the reproductive status of the coral assemblages sampled and the likelihood of spawning during the current or subsequent lunar cycles based on the size and colouration of the developing oocytes and spermaries, and therefore the potential for the sampled reef area to be used as a source reef for the collection of coral spawn. The methodology for gravid coral sampling is mature and used by many organisations internationally.

4.2 Hydrodynamic modelling to optimise coral spawn collection

4.2.1 Background and rationale

Following coral spawning the egg-sperm bundles and other buoyant gametes typically aggregate at the sea surface where they are subject to wind and wave action and currents that can cause them to disperse or aggregate in convergence zones. Predicting where coral spawn slicks are likely to occur in dynamic and complex reef environments subject to fluctuating weather and hydrodynamic condition is important for optimizing efficient coral spawn collection for larval rearing. Therefore, the Moving Corals team developed an innovative approach using 2D or 3D hydrodynamic models (EMS 2023) and particle tracking tools (e.g., CONNIE, OceanParcels), as well as in situ current measurement using tilt current meters, to identify probable coral spawn slick convergence zones. Details are provided in Gouezo et al. (2025a).

The methods are usually applied in the days and weeks leading up to predicted mass spawning events.

- System design: Use of spatial datasets (e.g., coral cover) coupled with coral spawn release simulation models (https://github.com/marine-ecologist/reefspawn), to predict particle (spawn/larvae) movement over 12–24 hours.
- Frequency: Annually during coral spawning season.
- Duration: Each application spans approximately 3–10 days including pre- and post-event fieldwork.
- Maturity level: Mature for model-based predictions; Demonstrated for field validation.
- Adoption level: Multi-organisational use within RRAP; methods are transferrable to national and international coral restoration efforts.

4.2.2 Procedure

The procedures outlined here support site selection, timing, and success assessment of coral larval-based restoration interventions. They are based on spatial modelling of gamete dispersal and larval retention following mass spawning events. The approach combines predictive hydrodynamic simulations with field validation using current meters, providing a data-informed foundation for coral spawn collection and larval seeding strategies.

4.2.3 Prerequisites

- **Existing documents**:
  - Modelling Guidelines for Larval Dispersal (Langlais et al. 2024)
  - LOWEL Tilt Current Meter User Guide
  - Gouezo et al. (2025a)
- **Software and tools**:
  - GIS software: QGIS or ArcGIS
  - Hydrodynamic models (eg. RECOM)
  - Particle tracking tools: OceanParcels, CONNIE
  - Data analysis: R or Python
  - Domino software for tiltcurrentmeter set up and data extraction
- **Other requirements**:
  - Personnel must be trained in spatial analysis, basic programming, and underwater deployment procedures.
  - Knowledge of local coral spawning events timing
  - Awareness of site-specific environmental conditions (e.g., prevailing currents, wind exposure).
- **Standards**:
  - Use of GPS with <5 m accuracy

4.2.4 Risks and Hazards

Model-based methods:

- **Data quality risk**: Inaccurate coral cover or hydrodynamic data may compromise predictions
- **Operational risk**: Model complexity may lead to computational delays if not streamlined

Field validation methods:

- **Instrument loss or damage**: Ensure all loggers are secured with backups and clearly tagged

Emergency procedures:

- Carry VHF radio or satellite communication device during field deployments.
- Follow site-specific dive safety protocols (e.g., buddy system, surface support)

4.2.5 Equipment and Materials

<u>Procedure equipment</u>

- Computer with hydrodynamic model access
- GPS device with <5 m accuracy
- GIS software (QGIS or ArcGIS)
- R or Python for data processing

- LOWEL Tilt Current Meters or equivalent
- Dive gear and underwater slates
- Secure mounting materials (rope, cable ties, dive weights)

Personal protective equipment (PPE) and other safety equipment

- Dive-certified PPE (e.g., wetsuits, gloves, weight belts)
- Surface buoy, dive flag, and underwater communication signals
- First aid kit and emergency O_2
- VHF or satellite phone for remote site communication

4.2.6 Implementation steps

Predicting spawn slicks convergence zone for spawn collection using dispersal models

Note: This method requires access to a 2D or 3D hydrodynamic model and a particle dispersal tool such as CONNIE or OceanParcels. Lower-resolution models (made of grid cells 100m and lower) are often better at capturing reef-scale hydrodynamics.

This section is adapted from Gouezo et al. (2025a) and provides step-by-step guidance on predicting where coral spawn (eggs and sperm) will travel locally ~12-16hr after mass spawning events, helping identify areas where coral embryos can be collected. This information is especially useful for planning collection efforts.

Step 1: Map Coral Reef Areas with Live Coral Cover >10%

Using **GIS software** (e.g., QGIS or ArcGIS):

- Create a .shp file with polygons outlining all reef areas where **live coral cover is greater than 10%**.
- Recommended data sources:
    - **Allen Coral Atlas**: https://allencoralatlas.org/atlas/
    - **AIMS Long-Term Monitoring (LTM) Program** (e.g., data from 2021) https://apps.aims.gov.au/reef-monitoring/reefs

Figure 4.2.1: Example of coral cover estimates in 2021 around Lizard Island

Step 2: Estimate Gamete Production

Coral spawn production is related to both **live coral cover** and **reef size**.

For each reef polygon:

1. Use the formula (adapted from Gouezo et al. 2025a) to calculate reproductive output per square meter of reef (assuming the original relationship is based on a 50 m² reef):

Reproductive output per m² = ((808,820.7 × coral cover %) – 173,407) / 50

2. Multiply the result by the polygon's area (in m²) to get **total gamete production**.

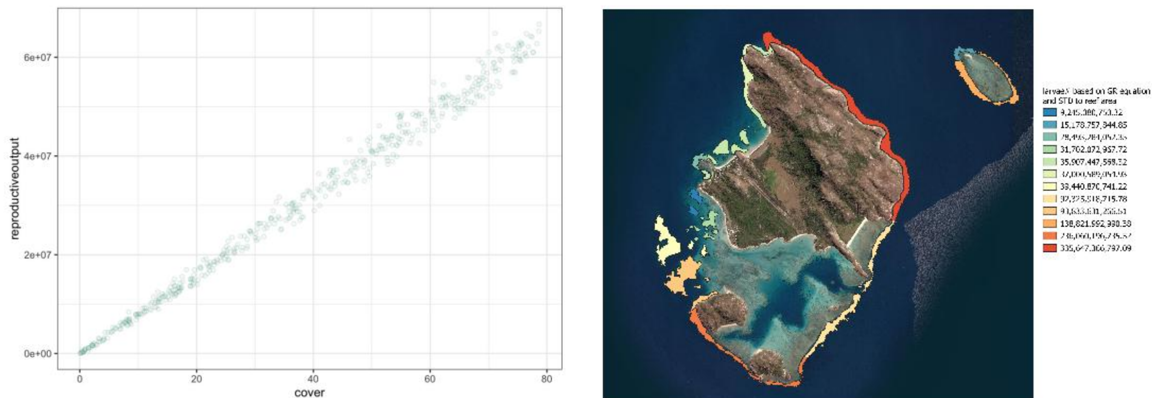


Figure 4.2.2: Relationship between reproductive output and coral cover (left) and estimated overall reproductive output around Lizard Island

For more technical details, refer to https://github.com/marine-ecologist/reefspawn and Appendix S2 in Gouezo et al. (2025a).

Step 3: Scale Total Output for Particle Release

Because gamete numbers can be extremely high:

- Scale all values so the total number of released particles across all polygons is between 500,000 to 1 million.
- This makes the simulation manageable while still providing reliable predictions of where particles (representing coral spawn) will go.

Figure 4.2.3: Relative reproductive output around Lizard Island for dispersal simulation

Step 4: Standardize Particle Release per Polygon

To make the model consistent:

- Adjust the number of particles so each polygon releases the same number, e.g., 5,000 particles per polygon.

Figure 4.2.4: polygons split into equal number of particles for dispersal modelling

Step 5: Set Simulation Timing for Spawning Events

- Identify the key nights of coral spawning (usually after full moons during specific seasons).
- Set the release time for particles between 19:00 and 21:00 near the surface (e.g., at 0.25 m depth).
- Track the particles for 16 hours to simulate how long coral spawn typically floats before sinking.

Step 6: Apply Wind Forcing (if needed)

- If the spawning night is windy (e.g., >15 knots), apply a 3% wind forcing to the model to simulate how wind pushes floating spawn.
- If conditions are calm or the area is in a wind shadow, wind forcing can be omitted.

Step 7: Analyze and Visualize Particle Movement

- Once the simulation is complete:
    - Export the results as .tiff raster files showing particle concentrations over time.
    - Import these files into QGIS and sum them to visualize total particle concentrations.
- Areas with high concentrations (often in dark red) indicate spawn slick convergence zones—ideal targets for monitoring or collection.

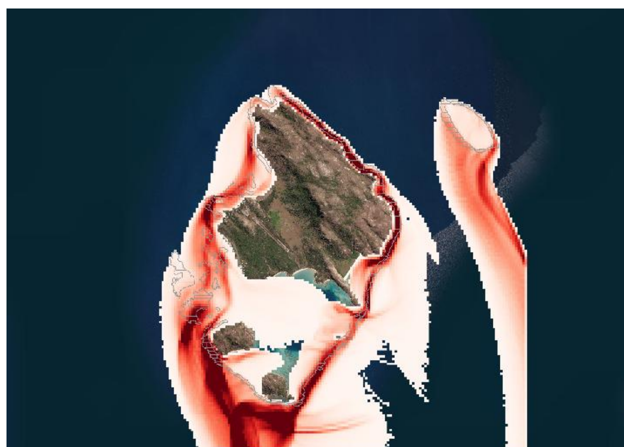


Figure 4.2.5: map showing overall particles concentrations during 16hrs following spawning event

Step 8: Ground-Truth the Predictions

- During reef-based collection of coral spawn:

- Visit the predicted convergence zones
- Use **GPS** to mark locations of dense spawn slicks
- Compare observed data to model predictions to assess accuracy

4.3 Optional drone operations and helicopter surveys to identify slicks for additional embryo collections

4.3.1 Background and rationale

In addition to completing gravid coral checks to predict the timing of coral spawning on source reefs (4.1) and hydrodynamic modelling to identify likely areas of spawn slick aggregation during spawning (4.2), collections of coral spawn can potentially be supplemented using drone operations and optional helicopter surveys early in the early morning after major coral spawning events to locate additional sources of embryos in slicks for sampling if required.

Figure 4.3.1: Example of early morning drone flight at Eyrie Reef in 2023 showing extensive coral spawn slicks the morning after mass coral spawning (Photo: G. Roff)

4.3.2 Procedure

This procedure provides guidance for drone operations for scientific operations over water. It outlines a standard procedure for planning a flight through to safely completing and recovering a drone over water. It also covers the standard procedures used for searching for coral slicks and recording and measuring them effectively.

4.3.3 Prerequisites

Drone operations must comply with relevant CASA Regulations and the operations must be compliant with the regulations and the rules set out in the Remote Operators Certificate (ReOC) for each organisation. The following five 'golden' rules apply to all drone operations, whether flying privately or for an organisation, and there may be other restrictions applicable to the flight plan, which the pilot must take into account.

1. The maximum altitude for drone flights is 120m. Operators can apply to CASA for an exception to this rule for a specific job if required.
2. Drones can only be flown during daylight hours. Operators can apply to CASA for an exception to this rule for a specific job if required.

3. The operator must be able to always see your drone during flight, but can apply to CASA for an exception to this rule for a specific job if required.
4. Do not fly near controlled airspaces, heliports etc. Airports have a restricted 5.5 km radius. Operators must be familiar with these regulations and appropriate flight boundaries.
5. Never fly over populated areas with crowds, such as streets, parks or beaches. Maintain at least 30m distance from other people.

4.3.4 Risks and Hazards

A range of risks and hazards are associated with drone operations over water and these are integrated into the information provided below for the implementation steps.

4.3.5 Equipment and Materials

Drone suitable for over-water flights and experienced operator/s with approved flight plans.

4.3.6 Drone flight implementation steps

1 Flight Planning

Before taking off, there are numerous checks to be undertaken to ensure the flight is safe and successful. These can be broken down to Flight Plan Checks and Aircraft Checks.

<u>Flight Plan Checks</u>

Complete the AVCRM at least a few days prior to the flight to ensure the flight is safe, the flight has a high chance of success and safe return, and that all relevant regulations have been followed. All of the following must be considered:

1. Airspace Restrictions. Will the flight intrude in any restricted airspace? Operators must keep clear of airports and helipads and also keep clear of populated areas for safety reasons. Use the OpenSky App to quickly check the proposed flight area.

2. Local Restrictions. Will the flight be over any special zones like National Parks? If this is the case, the operator may have to approach them directly to apply for permission. Different States in Australia operate differently, but generally commercial flights are forbidden in National Parks unless permission is sought. This also applies to Marine National Parks.

3. Will the flight comply with the Five Golden Rules? Will it remain below 120m, be able to keep line of sight, and be on the ground before the sun sets?

4. What will the weather be like? If it is too windy or raining then the flight must be delayed.

5. Battery life. Will the flight have enough battery to get home after the mission is complete? Make sure that a generous buffer is planned in case of headwinds on the return flight, or to take time with a difficult landing on a boat etc.

<u>Aircraft Checks</u>

1. Are there enough batteries for the flight? Make sure all batteries are fully charged the night before, including the batteries in the remote.

2. Are the DJI firmware for the aircraft, remote and the batteries up to date?

3. Is there sufficient phone coverage to connect the remote to the internet in the flight area? If not, then make ensure the remote has the relevant maps downloaded in advance.

4. Will the drone get wet on the boat? It is best to purchase a Pelican or similar case that is waterproof and impact proof to keep the drone and controller dry and safe while on the boat and not in the air.

5. Power up your aircraft and check all parts including operation of propellors, camera gimbal and LEDs. Make sure the camera settings are correct for the requirements.

Pre-Flight

Once on site and ready to go, make the following checks before getting airborne.

1. Prior to take off, do a final check of the aircraft batteries, camera etc.

2. What is the weather like? Consider delaying if the wind is too strong or if rain is present. Consider wind direction, especially on the flight home; flying into the wind will drain more battery so if the flight home is going to be against the wind, then factor in a generous time buffer.

3. Are there birds around? Seabirds especially are a common hazard, especially around islands where they nest. Nesting sites are often restricted airspace. Make sure you keep clear of birds wherever possible and have a plan to avoid them if your drone is attacked.

4. Is it sunny and bright? Fit a ND filter to the camera as appropriate to get the best vision.

5. Ensure there is a clear take off and landing zone. Preferably this is a large flat area and people can be kept clear of this space. If on a vessel then create a space paying attention to superstructure, antenna etc.

6. Alert other people nearby before take off. If necessary, have the Master of the vessel stop in the water and turn to make any wind come from a favourable direction.

7. Launch the drone and fly clear of the vessel. Once clear, pause before starting the mission. Check the drone's responsiveness. Take a breath and calm yourself after the excitement of clearing the vessel.

2. During Flight

Once airborne, conduct the mission as planned but keeping in mind the following.

1. Set the drone's home point to the controller immediately. If the vessel is underway, then update the home point every five minutes or so. Many drones have been lost because operators forgot to update their home point and the drone then tries to land where the vessel was some time ago. Update the home point before any landing operation.

2. Be aware of the battery life. The drone should Return to Home within a safe level to ensure safe landing.

3. Even over open ocean, do not neglect standard drone flight awareness. Be aware of hazards including birds, the whip antenna on vessels that may not be easily seen etc.

3. Post Flight

After a successful landing:

1. Shut down the drone and change the battery if there are more flights to do.

2. Change the SD card even if there is plenty of space on it to avoid losing data if problems occur during the next flight.

3. Secure the drone. Place it back inside the Pelican Case or other waterproof container before getting underway.

4. Considerations for Flights over water

These are general recommendations for conducting flights from vessels large and small:

1. Be conscious of the wind. It is often stronger offshore and can impact battery performance. Wind will also push the boat around and necessitate more frequent updating of the Home Point.

2. For take off and landing, work with the vessel operator to hold the vessel stationary and into the swell to reduce as much roll as possible.

3. Be aware of the altitude. The ultrasonic sensor that informs the operator of elevation will not be as accurate over water, especially if there is a swell.

4. Having the controller on a lanyard around the operator's neck is useful to avoid risks of it dropping during boat movements, and to enable both hands to be available while catching the drone if needed.

5. Hand catching. Use a glove to protect fingers from propellor cuts. Put the drone into sports mode to turn off the obstacle avoidance, so that the drone won't keep flying away from the operator when they reach for it. Bring the drone to eye level and reverse it towards the operator, grabbing it from underneath. As soon as the operator has it grasped firmly, turn it upside down quickly to force the rotors to switch off.

5. Obligations for Pilots

Pilots should check that all necessary certifications and requirements are up to date. Different institutions may have different obligations and requirements and should be checked and communicated before a drone operation is carried out. For Example, CSIRO has a comprehensive RPAS management system that is controlled by CSIRO's Chief Pilot. All pilots who fly for CSIRO must hold a current RePL registered with CASA, and operate under CSIRO's ReOC at all times. CSIRO pilots must fly at least every three months to maintain the validity of their licence under CSIRO's ReOC. Flights must be logged in advance through AVCRM; the online job management portal, for approval. Each job must contain all the flight specifics, as well as relevant safety documents. As each job commences and is completed, relevant checklists must be completed.

4.3.7 Helicopter Operations (optional)

Helicopter operations can support locating spawning slicks early in the morning after major coral spawning events, if resources are available to support this approach. Helicopter operations require the availability of a helicopter company willing and able to assist with spawn slick location. It is essential to have good communication procedures with the pilot of the helicopter to ensure slicks are located in a timely manner.

1. Find and communicate with a helicopter company and pilot willing to assist with project.

2. Organize helicopter-to-boat communication and share project timelines and plans.

3. Coordinate and commence dawn flights to locate slicks early in the morning after major spawning events.

4. Record spawn slick locations and estimate their sizes with GPS, and relay information to vessel-based team(s) on the water who can be guided to the slick location for additional collections of embryos for culturing.

Figure 4.3.2: Example of a coral spawn slick near Lizard Island located during an early morning helicopter survey for slicks in 2022 (Photo: P. Harrison)

5 Phase 2 Collecting coral spawn

Coral spawn can be collected using a range of active vessel-based methods or passively using spawn catcher nets, and the standard operational methods, and details of new processes developed in the Moving Corals project are detailed below.

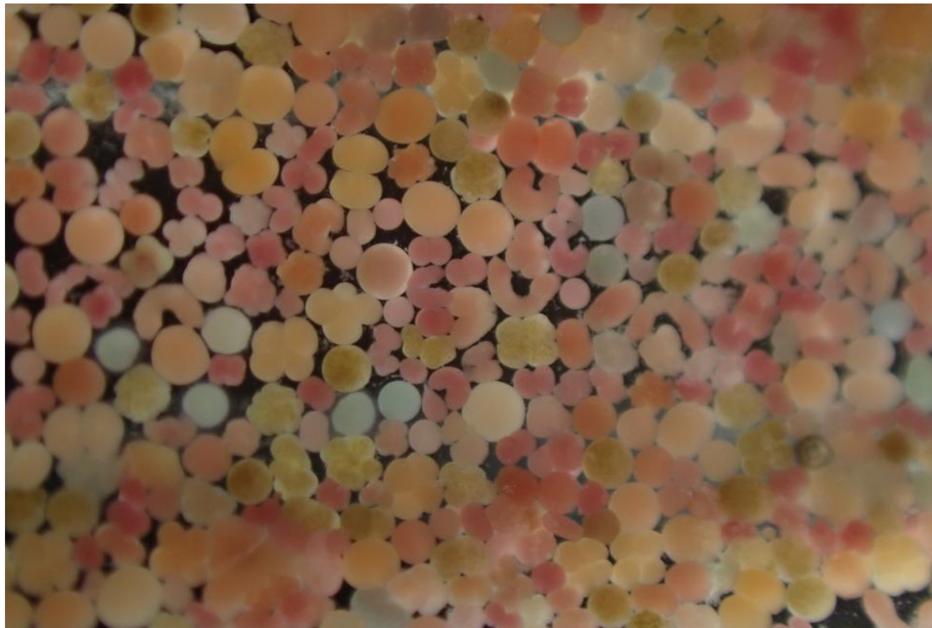


Figure 5.1 Diverse assemblage of early developmental phases of coral embryos being cultured that were sourced from wild slicks (Photo: L. Hardiman)

5.1 Vessel-based spawn collections

5.1.1 Background and rationale

The main operating procedures employed to collect most of the coral spawn for larval culturing in the Moving Corals project are based on deploying small teams of people on multiple small research vessels that are operational at night. These mobile vessel-based catcher groups enable the teams to initially search different reef areas and then rapidly relocate to reef areas where coral spawn aggregations and denser slicks were observed to enable efficient harvesting of gametes and early embryos for mass larval production. In addition, a larger 'spawn sucker' vessel-based method has developed during the Moving Corals project, providing additional large-scale operational flexibility for coral spawn collections.

5.1.2 Procedure

Collections of coral spawn involve night operations on small- to medium-sized vessels and transfer of collections into fertilization tubs where high rates of cross-fertilization can occur to maximise genetic diversity of embryos and larvae to enhance the resilience of restored populations. The fertilized eggs and early-stage embryos are then transferred into the culture pools for mass larval rearing (Phase 3).

5.1.3 Prerequisites

Knowledge of coral spawning periods is important for reef-based operations and the major spawning nights can be predicted based on the gravid coral checks (Phase 1) to identify sources of coral spawn from healthy breeding corals and prior knowledge of coral spawning patterns. Hydrodynamic modelling can help predict likely aggregation sites for coral spawn and slick formation based on changes in weather and hydrodynamic conditions (Gouezo et al. 2025a, Phase 1) to increase the efficiency of spawn collections. In addition, experienced crew who can identify scleractinian reef coral spawn slicks and distinguish these from spawn

of soft corals or floating rafts of *Trichodesmium* cyanobacteria at the sea surface are important so that collections focus on spawned gametes of reef corals to avoid contaminating the cultures with potential competitors and toxic phytoplankton.

5.1.4 Risks and Hazards

Most reef corals spawn at night, therefore reef-based operations need to be safely conducted at night around reef areas. Standard vessel safety measures such as confirmed vessel seaworthiness, experienced skipper and crew, vessel safety and emergency gear, radio and mobile or satellite phone communications and GPS tracking to identify hazards are essential for safe operations at night around reef areas. In addition, all crew members need to wear PFDs (personal floatation devices) to ensure their safety while operating over water. Regular planned radio communications with a night operations controller and communications between vessels are important to monitor all night operations.

5.1.5 Equipment and Materials

Vessel-based spawn collections require access to small vessels with space for a skipper and at least two crew members for spotlighting and collecting spawn from the sea surface. In addition, powerful LED spotlights are important for visual surveys to locate corals spawn slicks. Standard collections are done using handheld spawn collection scoop nets on extendable poles that are modified from commercial pool cleaning nets (Harrison 2024b). The collected spawn samples are then transferred into fertilization tubs or nally bins onboard the vessels for fertilization.

5.1.6 Implementation steps

1. Carefully plan safe night operations with detailed briefing for all crews prior to embarking on vessels, and prepare and check all safety, lights, communications and collecting equipment is available and fully operational for each evening.
2. A few coral species are known to spawn at dusk but most reef corals spawn after dark between 20:00 and 22:45 hr hence night operations should commence prior to 20:00 hr and continue until 23:00 unless sufficient coral spawn has been collected before that time each night. Vessel crews depart and maintain radio communications and begin searching for surface aggregations of egg-sperm bundles that are characteristic of broadcast spawning hermaphrodites including *Acropora*, merulinid brain corals and many other species. Some species also release gametes in clouds so individual eggs and clouds of sperm are also sometimes evident at the sea surface. It is important to avoid collecting spawn mixed with soft coral spawn or *Trichodesmium* slicks which are toxic.
3. Once sufficient coral spawn or denser spawn slicks are located, radio other collecting teams to alert them to the location and begin collecting spawn using surface scoop nets with fine mesh conical nets and a collection jar at the cod end (Fig. 5.1.1 Harrison 2024b). If spawn slicks are very dense, millions of eggs can be simply collected using buckets to collect samples alongside the vessel.

Figure 5.1.1 Modified spawn collection nets with collection jars enable very efficient collection of coral spawn from the sea surface at night (Source Harrison 2024b).

4. When sufficient coral spawn has been collected in the nets etc, transfer the contents into a large fertilization tanks on board the vessel and carefully mix the samples to maximise cross-fertilization.
5. Continue collecting until sufficient spawn has been collected and then gently stir the contents of the fertilization tank to maximise fertilization and cross-fertilization to enhance genetic diversity within the cohort.
6. If the spawn collection sites are distant from the larval culture pools and if weather and sea conditions are rough, then baffle balls can be added to the culture tub to reduce seawater and spawn spilling out of the tank during transit to the culture pools (Fig. 5.1.2 Harrison 2024b).

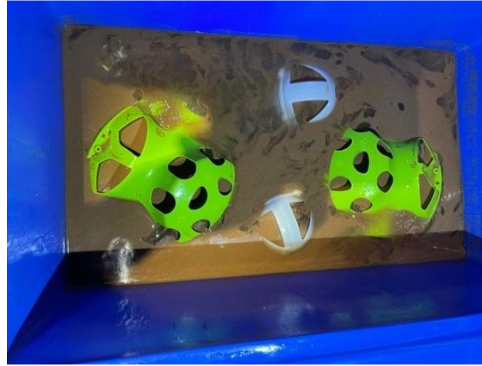


Figure 5.1.2 Coral spawn collections are transferred into fertilization tanks to optimise cross-fertilization rates and where necessary baffle balls can be added to reduce excess water movements during transit (Source Harrison 2024b).

7. Although fertilization may occur within a few minutes of eggs and sperm being combined in the fertilization tank some eggs are fertilized later, and the first cleavage of fertilized eggs is usually not evident for at least 45-60 minutes later. Therefore, keeping eggs and sperm combined for at least an hour is important to optimise fertilization rates and cross-fertilization.
8. It is important to keep sperm densities high during the fertilization period, but sperm should then be removed to avoid polyspermy (Willis et al. 1997) and reduce the risk of oxygen depletion as sperm stop swimming and accumulate at the bottom of the tank where they may begin to degrade and lower water quality. Therefore, after 1-1.5 hours sperm washing is done to remove sperm from the fertilization tank. This can be achieved by opening the tap at the bottom of the tank to remove excess sperm and seawater and then slowly replace seawater to refill the tank for maintaining embryo health during transit (Harrison 2024b).
9. Total abundance of eggs and embryos can then be calculated by carefully and thoroughly mixing the contents of the tank and then taking up to 5 replicate samples of the contents which are then counted under stereo microscopes to determine fertilization rates, and quantify densities of eggs and embryos per mL which this can be used to calculate the total collection abundance taking into account the measured volume of the seawater in the fertilization tank.
10. Collected eggs and embryos in clean seawater are then carefully skimmed off the surface of the tanks and slowly transferred into the larval pools for culturing (Phase 3).

The vessel-based mobile coral spawn collection methodology is mature and is used by many organisations internationally and is being applied in different regions of the GBR (Harrison 2024b).

5.2 Vessel collections using 'Spawn Sucker' methods

The techniques covered include the use of the pump-driven spawn collector (termed "spawn sucker"), which is a vessel mounted collection scoop for operation at sea. The spawn sucker was designed to optimise bulk collection of slicks at low densities, and to facilitate transfer of slicks into larval culture ponds and/or ship-based rearing facilities, or culture directly in the large collection tanks on the vessel. While the methods are technically mature and validated at local scales, their broader application may require vessel-specific customization. Limitations include reliance on the large infrastructure and suitable field equipment.

5.2.1 Background and rationale

As the first step of larval restoration, collection of coral larvae in large densities is a critical step in the process. While collection of coral larvae from high density slicks using hand-scoops and nets is the primary method of collection for RRAP protocols (see 5.1 collections and 6.1 pool cultures), a vessel-based approach to collecting and condensing coral larvae was developed during the RRAP fieldwork. The collection vessel (termed "spawn sucker") was developed after several trial methods during coral spawning and is optimised to collect and concentrate larvae at large scale from low-density surface slicks.

- **System design**: Vessel specific mounted operation for field settings.
- **Frequency**: In-situ collections during coral spawning and in the early morning following spawning
- **Duration**: 2-6 hr periods
- **Maturity level**: demonstrated in field settings
- **Adoption level**: validated (organisational use within RRAP), likely limited due to vessel constraints

5.2.2 Procedure

Deployment of the spawn sucker collection device requires assembly on a suitable vessel such as a small barge, and operations at night to collect coral spawn from the sea surface.

5.2.3 Prerequisites

Knowledge of likely coral spawning periods and reef areas where coral spawn is likely to accumulate.

5.2.4 Risks and Hazards

Field Activities:

- **Physical hazards**: Risk of injury during boat operations, heavy lifting of equipment – apply appropriate marine operations safety and PPE procedures.
- **Fatigue**: operate with sufficient crew members, ensure distribution of workloads.
- **Weather-dependent operations**: Field activities must consider sea state and tides.

Control Measures:

- Follow vessel safety protocols and PPE
- Maintain clear data working space
- Document spawn procedures, maintain notes on operations

5.2.5 Equipment and Materials

Procedure equipment

Core Pumping System

- Primary Diaphragm Pump – e.g. *Paddock SPDP80HX* or equivalent, ≥250 L/min, ≥8 m suction lift
- Backup Diaphragm Pump – identical or compatible for redundancy
- Fuel Container (20 L minimum) – compliant jerry can with funnel
- Spare Parts Kit – diaphragms, seals, gaskets, bolts
- Pump Tools – wrench set, screwdrivers, seal puller
- Pump Manual (printed)

Plumbing and Fittings

- Camlock Fittings (4") – male/female, A/B/C/D adapters
- Gaskets and O-rings (spares) – compatible with camlocks
- Hose Clamps and Zip Ties – stainless steel, marine-grade

- Thread Tape / PTFE Tape

Transfer and Suction Hoses

- Transfer Hose (4”) – 10–20 m sections, rigid, UV-resistant
- Suction Hose (4”) – reinforced, clear or marine-grade, 5–8 m

Biological Protection

- Larval Holding Tank – Rainwater tank (~2000 litre) with overflow protection
- Insulated Transport Containers – ~100 litres with lids
- Sample collection jars for larval counts

Field Ops & Monitoring

- Field Operation Lights ≥10,000 lumens – waterproof, portable, battery or 12V
- Power Bank / Battery Pack (≥20Ah) – USB and 12V out
- Field Notebooks – waterproof, hard cover
- Pencils / Alcohol Markers
- GPS Device – handheld or vessel-mounted
- Compass / Backup GPS App
- Time-Synced Watches

<u>Personal protective equipment (PPE) and other safety equipment</u>

- Level 100+ Life Jacket (PFD Type 1 or 150N) – AS 4758.1 compliant
- EPIRB (Emergency Position Indicating Radio Beacon) – where required
- Marine Radio (VHF or HF) – handheld if no fixed unit
- High-visibility Clothing – waterproof, with reflective strips
- Non-slip Footwear – closed-toe deck boots or shoes
- Sun Protection for early morning operations only:
  - Broad-brimmed hat or legionnaire cap
  - Polarised sunglasses (AS/NZS 1067 compliant)
  - Long-sleeved sun-protective clothing
  - SPF 50+ sunscreen

5.2.6 Implementation steps

The spawn sucker system is a frame attached to the front of a shallow 12 m barge that allows for scooping and pumping of spawn into storage tanks. The vessel is driven through surface slicks, which are concentrated and pumped directly into 2,000 L round tanks that are held on the vessel. Once full, the tanks are drained from valves on the bottom, retaining the surface floating spawn. The spawn sucker can be run through several cycles of collection and concentration, and when full the material can be transferred into the culture ponds or surface based aquaculture systems, or cultured directly in the large onboard collection tanks.

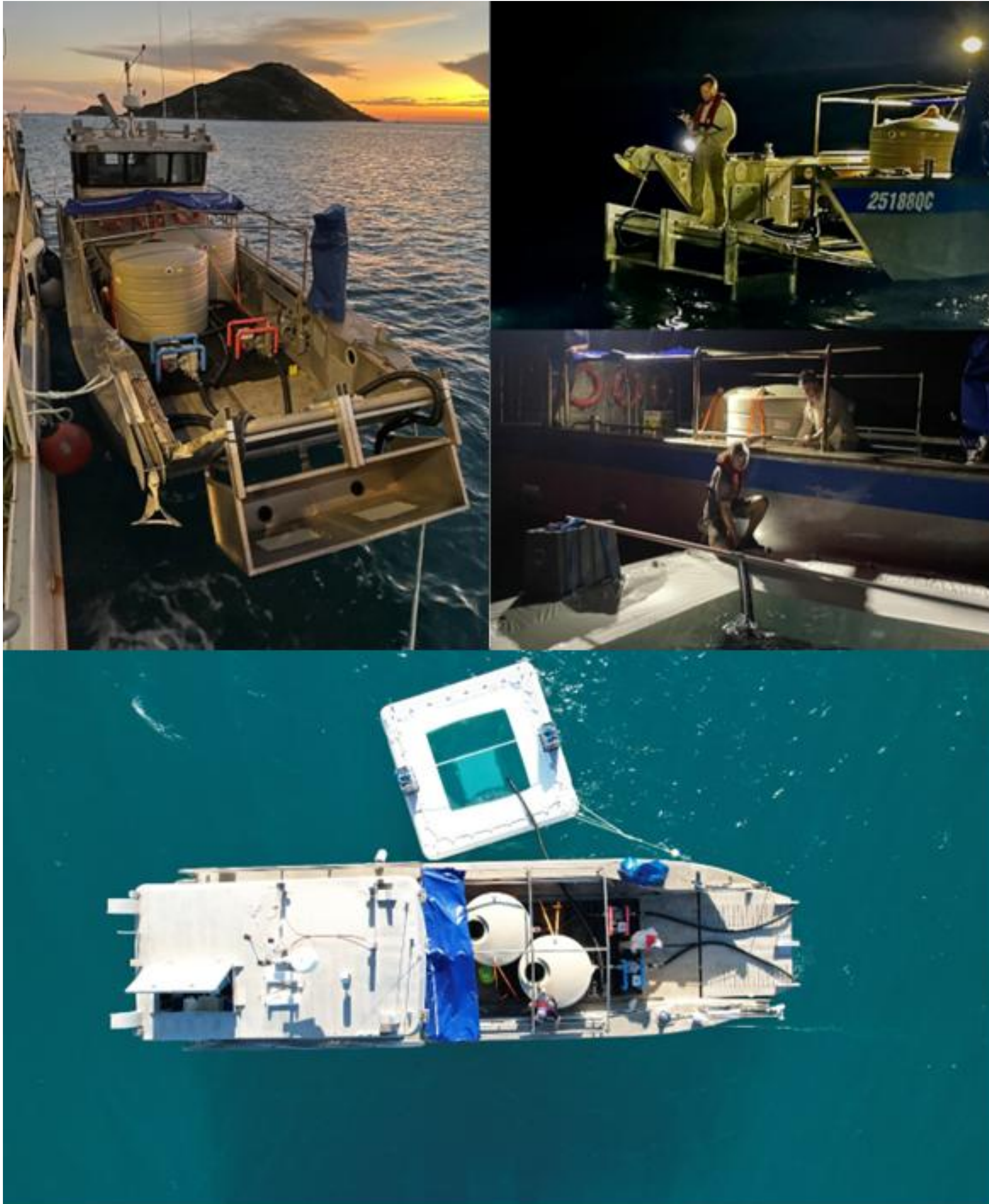


Figure 5.2.1: Spawn sucker during field collections at Lizard Island in 2022 collecting and transferring coral larvae (Photos: C. Doropoulos, G. Carlin)

Figure 5.2.2: Spawn sucker with updated floating collection mouth during collection, and rainwater tanks full of spawn following collection during field collections at Eyrie Reef in 2023 (Photos: G. Carlin, C. Doropoulos)

5.2.7 Coral spawn collection (spawn sucker)

1. Setup and design

- Design and fit spawn sucker to vessel.
    - Operations at Lizard Island utilised an Aluminium Marine Tri Hull barge vessel (12m length, “Spoonbill”).
    - Ensure vessel has suitable deck space for operations (e.g. 6*3m) for deployment of pumps and rainwater collection tanks
    - Ensure shallow draft (<0.5m) for operations in shallow coral reef environments during collection
    - 0.5 m2 plankton mesh panels are built into the bottom of the scoop to concentrate the sample prior to transfer from the scoop to the tanks

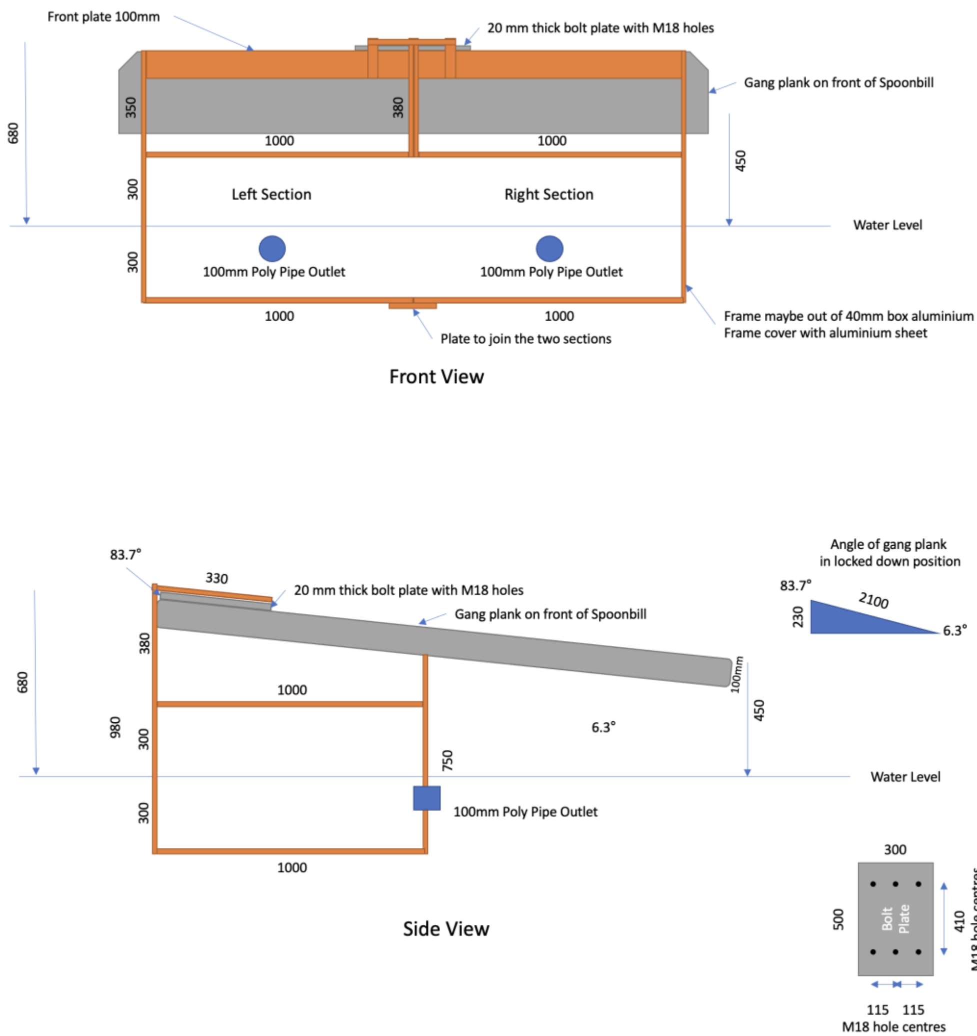


Figure 5.2.3: Design specification for the spawn sucker.

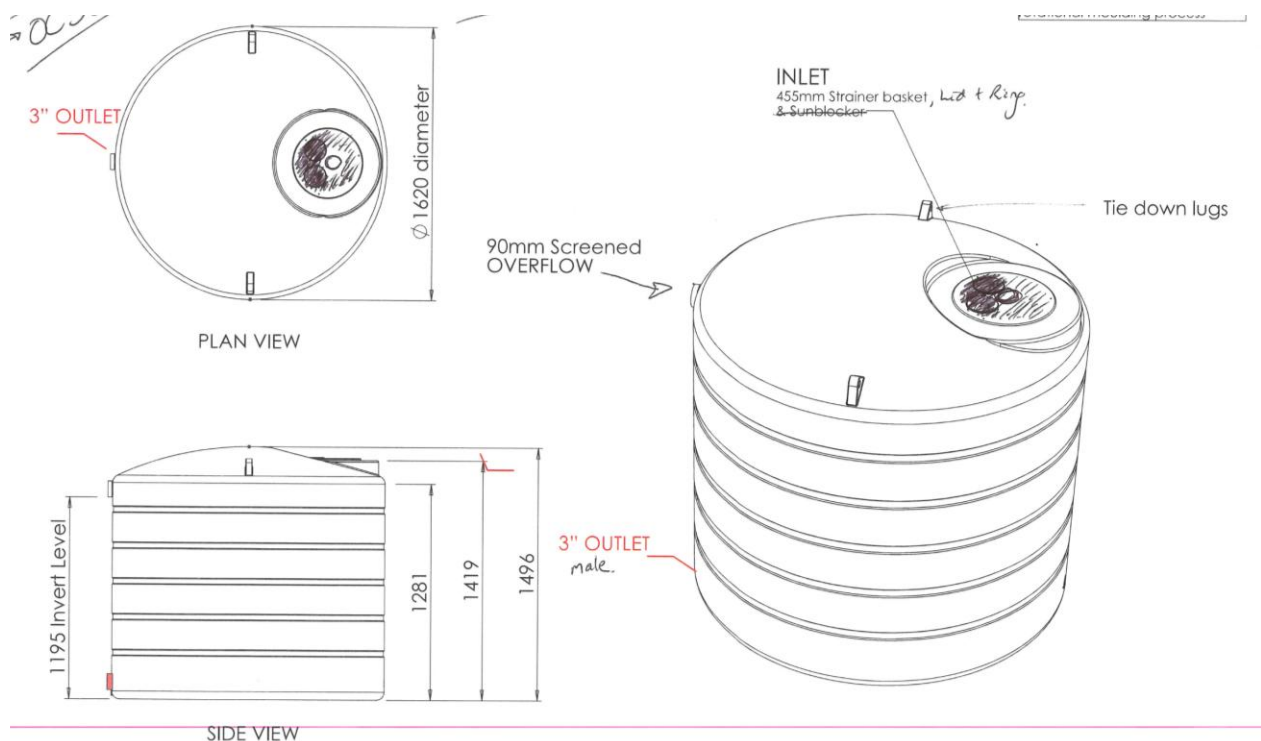


Figure 5.2.4: Design and setup for rainwater tanks

1. **Setup pumping and hoses**

- Ensure 4" camlock fittings on pump intakes and spawn sucker plumbing
- Use rigid transfer hose (transparent to view contents)
- Setup collection tanks (minimum 2000 litres) with stopcock drains on the base of the tank
- Connect spawn sucker collection frame – diaphragm pump – rainwater tanks

Figure 5.2.5: Setup for camlock fittings and pressure gauge on pumps

2. **Field operations:**
- Scoop is suspended ~10–40 cm below water surface using the drawbridge mechanism
- Water depth/angle of scoop adjusted dynamically via drawbridge winch control
- Pumps operate in parallel, each running at ~150 L/min
- Each pump fills a tank in ~13 minutes at the specified flow rate

3. **Pump Operation Strategy**
- Low flow rates are maintained to:
    - Reduce turbulence and mechanical shear
    - Improve survival rates during transfer
- Pumps must not run dry
- Pumps are started only after scoop is submerged and stabilised
- Priming valves checked before deployment to ensure smooth startup

Personnel & Communication

- *Chief Operator (front deck):*
    - Monitors scoop position, depth, and water clarity
    - Communicates real-time adjustments to:
        - Vessel Captain (speed, heading, position)
        - Pump Assistant (flow rate, engagement timing)
- *Vessel Captain:*
    - Drives barge along pre-surveyed reef transect
    - Adjusts drawbridge depth to keep scoop at optimal level
    - Maintains slow, consistent speed (~0.5–1.0 knot recommended)
- *Pump Assistant (deck):*
    - Engages/disengages diaphragm pumps on command
    - Monitors pump rate, sound, and hose pressure
    - Watches for clogs, vibration, or flow reduction
    - Coordinates filling time for each tank

4. **Monitoring and Adjustment**
- Flow rate and fill time logged per tank
- Scoop submergence depth logged (initial + every 5 min)
- Operators visually check for:
    - Larval density buildup on mesh
    - Foam or debris accumulation
    - Mesh integrity/damage
- If larval density declines, vessel speed or scoop depth may be adjusted

5. **Shutdown and Rinse Protocol**
- Once all tanks are filled:
    - Pumps disengaged while scoop still submerged (prevents backflush)
    - Transfer hoses flushed with clean seawater
    - Scoop rinsed externally, then internally via hose pressure
- Mesh panels checked for:
    - Damage
    - Debris
    - Biological residue (scraped gently with soft brush if needed)

5.3 Passive spawn collections using spawn catchers

5.3.1. Background and rationale

Floating spawn catchers provide a very efficient method for larger-scale collections of tens to hundreds of millions of spawned coral gametes when moored downcurrent from healthy reefs with abundant gravid during spawning periods (detailed in Harrison et al. 2021, Harrison and dela Cruz 2022, Harrison 2024b). Deployment of spawn catchers was optimised in the Moving Corals project by identifying reef sites with high proportions of gravid corals with mature eggs and sperm (Phase 1) and predictions of spawn aggregation locations from hydrodynamic modelling (Gouezo et al. 2025a).

5.3.2. Procedure

Spawn catchers consist of inflatable pontoons with vinyl and plankton mesh nets that can be opened to allow spawn to float into the net (Fig. 5.3.1). Spawn catcher systems are deployed prior to predicted coral spawning periods either near coral assemblages with abundant gravid corals with mature gametes, or in reef locations that are predicted to have aggregations of coral spawn and slicks. Spawn accumulates in the open net and the net is then closed to contain the spawn for larval rearing.

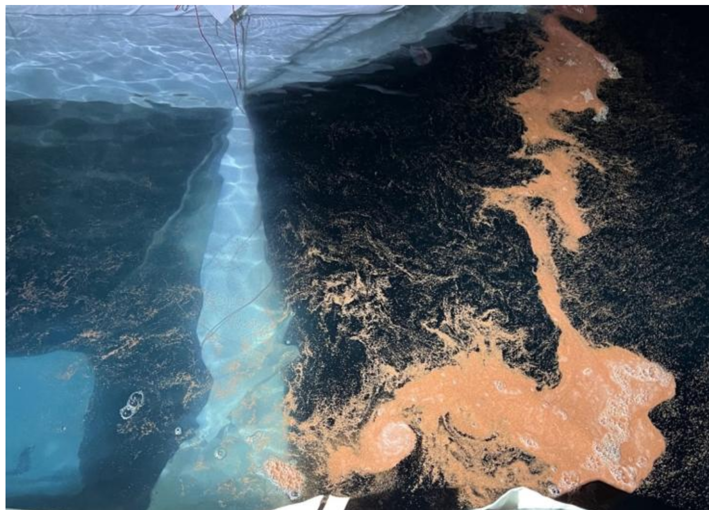


Figure 5.3.1: Millions of egg-sperm bundles and fertilized eggs collect in the floating spawn catcher net systems following mass coral spawning periods (Source Harrison 2024b).

5.3.3. Prerequisites

Spawn catchers are best deployed in reef locations where abundant corals with mature gametes are present and where the currents and local hydrodynamic conditions are likely to carry the spawn into the catcher net system.

- **Existing documents**:
  - Designs and demonstration of reef-based spawn catcher deployments and effectiveness at passive collection of hundreds of millions of eggs and early embryos (Harrison et al. 2021, Harrison and dela Cruz 2022, Harrison 2024b)

5.3.4. Risks and Hazards

People operating the pumps to inflate the spawn catcher pontoons need to be trained to ensure they are using the pumps and inflation ports on the pontoon correctly. High pressure scuba tanks should not be used to inflate the pontoons due to the risks of rupturing the pontoon structure. Crews need to be experienced and trained in the safe operations of deploying the anchor systems to temporarily moor the

spawn catchers near reefs, targeting sand patches to avoid direct damage to any live coral. When deploying the anchor, consideration needs to be given to possible changes in wind and current directions that may cause the spawn catcher and submerged net to move in an arc around the anchor, to avoid the net entangling on nearby reef areas. Attaching the open-closing net system needs to be done safely so that staff are not at risk from the ropes and weights used to deploy the net.

5.3.5. Equipment and Materials

The spawn catcher system consists of an inflatable pontoon to provide the floating structure, an open-closing vinyl and plankton mesh collection net, and an anchor system attached to a bridle on the pontoon (further details are available in Harrison 2024b).

5.3.6. Implementation steps

1. Optimise the location of the spawn catcher system on a reef area downcurrent from predicted spawning source reef with abundant gravid corals (Phase 1), or in areas predicted to have accumulations of coral spawn based on hydrodynamic modelling forecasts (Gouezo et al. 2025a).
2. Set up the anchor system with an anchor of appropriate size and holding capacity, and length of chain and anchor rope of sufficient length and capacity to hold the catcher system in place during adverse weather.
3. Deploy the anchor system safely making sure that staff are clear of the ropes as the anchor and chain and attached rope is slowly lowered onto the sandy habitat near the reef area.
4. Inflate the pontoon using the supplied low pressure airpump until the pontoon is rigid and then attach the spawn collection net along the surface of the pontoon (Harrison 2024b).
5. Attach the anchor line to the rope bridle system of the pontoon and when secured open the spawn catcher net in preparation for collection at night.
6. During coral spawning operations at night, vessel crews monitor spawning activity and check on spawn collecting in the net. When sufficient spawn has been collected, the net is closed and the spawn can then be retained in the catcher net for additional larval rearing capacity, or the spawn can be skimmed off the sea surface for transfer to larval culture pools (Phase 3).
7. At the end of the larval culturing period when competent larval cohorts have been deployed or sued for larval settlement on devices (Phase 4), the net is removed from the pontoon, the pontoon system is deflated and collected, and the anchor system is carefully removed from the reef area. All components need to be cleaned with high pressure hoses to clean fouling organisms off the equipment, and then thoroughly dried before packing and storage ready for the next deployment.

The methodology for large-scale passive spawn collection using floating spawn catchers is mature and is used by multiple organisations internationally and in different regions of the GBR (Harrison 2024b).

6 Phase 3 Mass larval culture

The third phase of the reef-based larval restoration process involves mass culture of diverse coral larvae that are reared from the coral spawn collected on reefs. Most of the larvae reared during the Moving Corals project were cultured in floating inflatable pool and bespoke net systems (developed and described in Harrison 2024b), with additional larvae reared in vessel-based aquaculture tank systems in 2023 that were adapted and optimised from earlier designs (see Doropoulos et al. 2019b for details).

6.1 Reef-based floating inflatable larval culture pools

6.1.1. Background and rationale

Scaling up coral larval restoration requires cost-effective and logistically efficient methods for mass culture and routine production of many millions of coral larvae. The simplest and most efficient method for mass larval rearing is to collect samples from the immense volumes of coral spawn that are produced during annual peak spawning periods on reefs and transfer the spawn into floating larval culture pools for development until they are competent to settle 4-6 days later (Harrison 2024a). Previous research enabled the development of various designs for larval culture pools that can be deployed on reefs (see Section 2.2), and recent work has developed and established a standard 4 x 4 m inflatable pontoon and 3 x 3 m bespoke culture net system that has been used to successfully rear hundreds of millions of larvae at up to 70% production efficiency (Harrison 2024b). This pool system was adopted for routine use in the Moving Corals project to scale up larval production and optimise mass larval culture by testing larval rearing and production efficiencies in different reef areas and using different spawn stocking densities.

6.1.2. Procedure

Reef-based larval culture involves setting up and deploying floating larval culture pools on reefs for mass larval production for restoration. Coral spawn collected in Phase 2 operations is transferred into larval pools for development until larval cohorts are competent to settle, usually after 4-6 days. The competent larvae are then released onto reef systems or settled onto devices in larval pools (Phase 4).

6.1.3. Prerequisites

Successful larval culture requires an understanding of how and when to collect coral spawn, and how to set up the larval pool systems to ensure that larvae are cultured successfully for deployment.

- **Existing documents**:
    - Designs and demonstration of reef-based inflatable larval pool and culture net systems deployments and effectiveness at mass culturing tens of millions of healthy competent larvae (Harrison et al. 2021, Harrison and dela Cruz 2022, Harrison 2024b)

6.1.4. Risks and Hazards

As with the deployment of the spawn catcher pontoon and net systems, people operating the pumps to inflate the larval pool pontoons need to be trained to ensure they are using the pumps and inflation ports on the pontoon correctly. High pressure scuba tanks should not be used to inflate the pontoons due to the risks of rupturing the pontoon structure. Practitioners also need to be experienced and trained in the safe operations of deploying the heavy anchor systems to temporarily moor the pools safely in lagoon or passage areas or near reefs. When deploying the anchor in sandy habitats, consideration needs to be given to possible changes in wind and current directions that may cause the pool and submerged net to move in an arc around the anchor, to prevent the net contacting or entangling on nearby reef areas. Attaching the culture net system needs to be done safely so that staff are not at risk from the ropes and weights used to deploy the net.

6.1.5. Equipment and Materials

The standard inflatable larval culture pool and net systems consists of a 4 x 4 m inflatable pontoon to provide the floating structure, a 3 x 3 m tapered vinyl and plankton mesh culture net, and an anchor system attached to a bridle on the pontoon (further details are available in Harrison 2024b).

6.1.6. Implementation steps

1. Locate the larval pool/s in an area with sufficient water depth and free from reef areas and other hazards that may entangle the net if it moves around in an arc on the anchor system during the larval culture period, and in sandy reef habitats where the anchor and chain will not damage sensitive marine life.
2. Set up the anchor system with an anchor of appropriate size and holding capacity, and length of chain and anchor rope of sufficient length and capacity to hold the culture pool system safely in place during adverse weather.
3. Deploy the anchor system carefully onto sandy habitat areas making sure that staff are clear of the ropes as the anchor and chain and attached rope is slowly lowered onto the sandy habitat for anchoring.
4. Inflate the pontoon using the supplied low pressure air pump until the pontoon is rigid and then attach the larval culture net along the surface of the pontoon using the ropes and holders (Fig. 6.1.1, Harrison 2024b).

Figure 6.1.1: Standard 4 x 4 m larval culture pools (Harrison 2024b) containing many millions of developing embryos for routine mass larval culture on reefs and temporarily moored in sandy habitats (Photo: P. Harrison)

5. Attach the anchor line to the rope bridle system of the pontoon and when secured confirm that the anchor system is safely set. A second pool can be attached to the anchored pool if the mooring location is relatively calm and sheltered and weather conditions are appropriate (Fig. 6.1.1.)
6. When sufficient coral spawn has been collected by the vessel-based mobile collection teams, fertilization and sperm washing stages are completed and density samples have been taken (Phase 3), the spawn can be carefully skimmed off the surface of the fertilization tanks and gently transferred into the larval culture pools.
7. Larval densities can be monitored throughout the development period to determine survival rates over time, and larval development rates leading up to settlement competency. The contents of the larval pool should be stirred and then up to five replicate density samples taken using a Niskin bottle plankton sampler in midwater, or by inverting small sampling containers while submerged within the larval pool. Larvae within each sample are counted under dissecting microscopes in plankton counting trays to standardise the larval density estimates per mL, and the total densities of larvae can be estimated by accounting for the total volume of seawater and larvae within the culture pool net.

8. At the end of the larval rearing period, the net is removed, the pontoon system is deflated and removed, and the anchor system is carefully removed from the reef area. The net, pontoon and anchor systems must be thoroughly cleaned with high pressure hoses to clean algae and any other fouling organisms off the equipment. These components then need to be thoroughly dried before packing and storage ready for the next larval culture period.

The methodology for large-scale larval culture in inflatable pontoon and culture net systems is mature and is now being used by many organisations internationally and in different regions of the GBR (Harrison 2024b).

6.2 Vessel-based larval culture in tanks

6.2.1 Background and rationale

Vessel-based larval culture allows practitioners to collect spawn slicks during natural spawning events, culture larvae to settlement competency en masse in controlled conditions for transport across long distances for targeted reef delivery. This avoids the need for intensive land-based infrastructure, removes any handling of parent colonies, and adds a long-distance transfer system that is simpler than transferring in and out of in situ culture pools. In previous trials (Doropoulos et al. 2019b), testing the culturing and settlement of corals using a 50,000 L culture system built on a tugboat was conducted. That work demonstrated that it is feasible to concentrate, maintain, and grow coral larvae onboard. The current project aimed to optimise the previous design and deliver larvae directly from the on-board culture tanks directly to the reef.

6.2.2 Procedure

The on-board floating aquaculture system consists of 4 x 10,000 L rainwater tanks, each measuring 3 m diameter x 1.6 m height. Utilising wider and shorter tanks a) increases surface area to facilitate higher stocking density on collection when gametes are positively buoyant, b) makes an easy working height for internal tank access when necessary, and c) reduces water height from the deck for vessel stability. Each tank has two hatches cut into it to allow access into the tanks and air flow through the tanks. The aquaculture system is flow-through with filtered seawater, achieving flow rates of >4,000 L per hour per tank. There is a single intake hose that feeds into the skid system, which consists of a 100 µm cartridge filter that flows into a 5 µm cartridge filter and then to a UV filter. Filters need to be changed and cleaned regularly depending on water quality. Following filtration, the filtered seawater flows through a central pipe to each individual tank. Each tank has a manual flow adjustment valve and associated flow meter. Flow into the tanks is via a manifold on the top of each tank, with each manifold having four down pipes with outlets running from top to bottom on one side of each pipe, allowing flow to drive circulation in the tank when each pipe was orientated in the same direction. Inside each tank is a central standpipe for outflow, which is surrounded by a plankton mesh cover that has an air-curtain surrounding it. The external standpipe utilises a gated system to allow adjustment of the internal water heigh of each tank to 100%, 35%, and 5% capacity. All outflows run into a single pipe that flows off the stern of the vessel. This outflow point is also utilised for direct larval delivery from the tanks on the vessel to the reefs via 3” hosing and camlock fittings.

6.2.3 Prerequisites

Successful larval culture requires an understanding of how and when to collect coral spawn, and how to set up an aquaculture system to ensure that larvae are cultured successfully for deployment. Careful planning with maritime engineers is also required to incorporate vessel weight and power supply limits.

6.2.4 Risks and Hazards

As with collections using the spawn sucker methods, risks and hazards should consider:

Field Activities:

- **Physical hazards**: Risk of injury during boat operations, heavy lifting of equipment – apply appropriate marine operations safety and PPE procedures.
- **Fatigue**: operate with sufficient crew members, ensure distribution of workloads.
- **Weather-dependent operations**: Field activities must consider sea state and tides.

Control Measures:

- Follow vessel safety protocols and PPE
- Maintain clear data working space
- Document culturing procedures, maintain notes on operations

6.2.5 Equipment and Materials

Intake

- Spa pumps
- Hosing

Filtration

- Canister filters (100 um, 5 um)
- UV filter

Air

- Air pumps
- Air curtains
- Air lines

Distribution and regulation

- PVC piping, elbows, caps
- Ball valves, gate valves
- Hosing for larval release

Culturing

- Large tanks (e.g., 2,000 - 10,000 L rainwater tanks)
- Standpipes
- Phytoplankton mesh

Figure 6.2.1: CAD design drawing and images of the flow-through vessel-based aquaculture system on the aft deck of Toanui in 2023. Each tank holds 10,000 L of seawater (Photos: C. Doropoulos, North Marine)

6.2.6 Implementation steps

1. Design aquaculture system based on weight capacity and space available on vessel. If not familiar with aquaculture systems in general or specific requirements for coral larvae, work with experts for advice. Vessel types can include powered, shallow hull vessel such as Toanui (pictured above), or powered deep hull vessels such as the tugboat utilised in Doropoulos et al. (2019b). Tugboats tend to have higher weight and space limits but are more restricted in how close they can travel to the reef. Alternative vessels include powered- and dumb-barges.

2. Install the aquaculture system onto the vessel. Allow 3 days for installation with a team of 3 experienced personal. Note that the vessel operator team will also need to be involved during the installation for use of HIAB for lifting equipment from dock to vessel, electronic supply, and testing balance and safety of the vessel under the weight distribution of the system when full.

3. During coral spawning, collect from wild coral spawn slicks (described in previous sections) and stock tanks on board the vessel with contents. Stock tanks with ~30-50% cover of coral spawn slick.

4. Conduct larval density sampling to monitor larval cultures (described in previous sections).

5. At the end of the larval culturing period, larvae are:
    a. Slowly released directly from the tanks to the reef restoration area for settlement onto the reef via hosing that is fixed from the tank outflows to the reef using stakes
    b. Settled onto settlement substrata that are placed into the tanks, after which they are deployed to the reef.

6. The system is cleaned, flushed, dried, packed, and stored ready for the next spawning operation.

7 Phase 4 Larval settlement and deployment on reefs

Mass culture of many millions of genetically diverse coral larvae produces very large competent larval cohorts that can be deployed onto reefs or settled onto devices in larval settlement pools to increase larval settlement and recruitment on reefs that lack sufficient natural supply to recovery from disturbance impacts including mass coral bleaching events. A range of larval release and settlement methods have been tested in different reef habitats during the Moving Coral project including the development of some innovative methods for staining larvae to identify cultured larvae after release (Doropoulos and Roff 2022), hydrodynamic modelling to predict periods of reduced current flow over target reef sites to optimise larval release periods and increase the likelihood of larvae being retained near the release sites (Gouezo et al. 2025b), and slow releases of larvae from larval seedboxes to increase larval supply over longer periods and therefore maximise settlement around the release sites (Doropoulos et al. 2025).

7.1 Staining coral larvae

Colouring coral larvae with vital stains provides a simple, effective, and rapid method for visually distinguishing different larval cohorts. When appropriately refined, staining is visible to the naked eye and can support field applications and experimental differentiation without complex imaging tools. The larval staining method is (i) procedurally simple and rapid; (ii) easily detectable in field settings; (iii) easily scalable to large numbers (10^6 to 10^9) of larvae; (iv) nontoxic in the marine environment; (v) uses widely available stains; and (v) cost effective. Our experimental results (see Doropoulos & Roff 2022 for complete methodological details) demonstrate that two vital stains—Nile blue and neutral red—can be used successfully with minimal impacts on larval viability and settlement capacity, provided concentration and exposure times are carefully controlled.

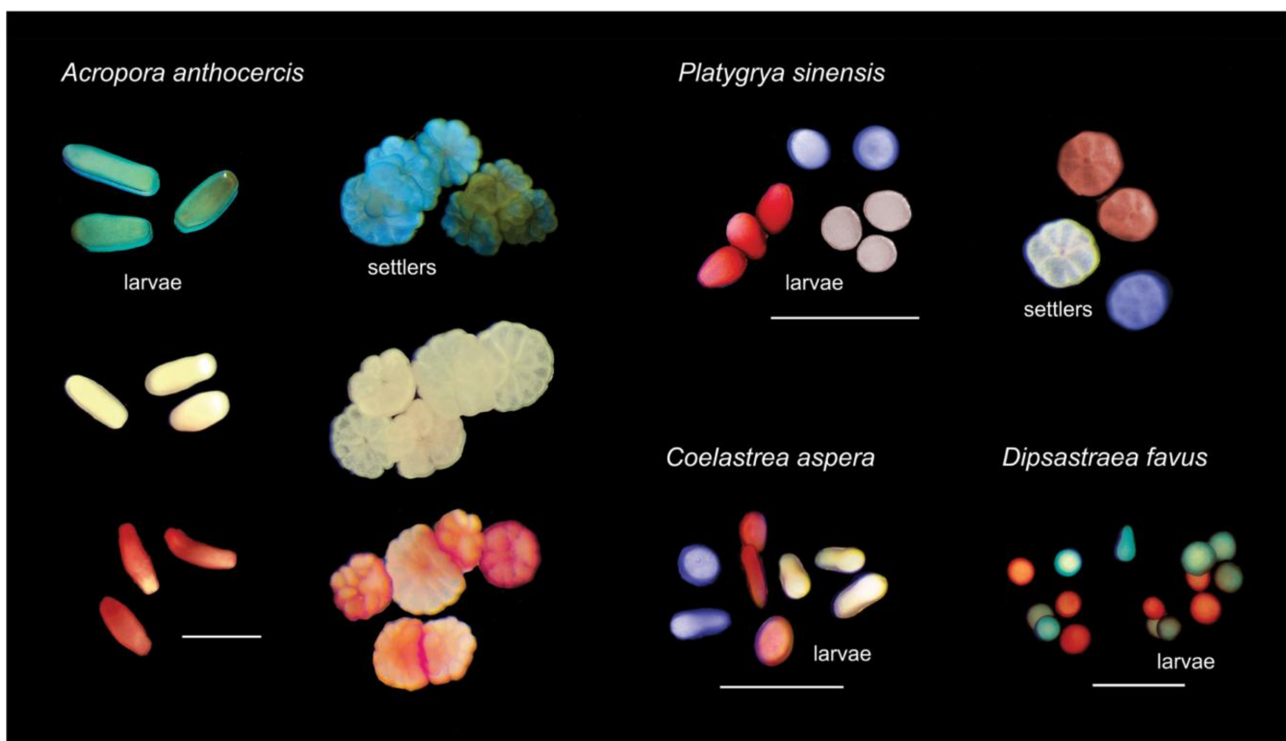


Figure 7.1.1: Representative images of free-swimming and newly metamorphosed larvae (Nile blue, unstained, neutral red) from A. anthocercis, P. sinensis, C. aspera*, and mixed Nile blue and neutral red stained* D. favus *larvae. White scale bars = 1 mm. (Source: Doropoulos & Roff 2022)*

7.1.1 Background and rationale

Vital staining of coral larvae is a mature and demonstrated methodology that enables rapid, visual identification of individual larval cohorts for both laboratory and field applications. The technique involves the use of biocompatible vital stains Nile blue and neutral red which are absorbed by coral larvae and allow for simple discrimination from naturally pigmented individuals. It has been validated through multi-species experiments under controlled and natural conditions and has shown no significant negative impact on larval survival or settlement when applied under optimized concentrations and exposure times.

The methodology is fully developed, with large-scale deployment demonstrated in field environments involving over 100,000 larvae. The technique has been successfully applied using standard laboratory equipment, such as 6-well culture plates or scintillation vials, and requires only basic microscopy or visual inspection for scoring. Its simplicity and low-cost support widespread adoption, and it is suitable for integration across institutional, organisational, and international research or restoration programs. Adoption is currently at the organisational to multi-organisational level, with the potential to scale to national and international standardisation due to its reliability, portability, and replicability across taxa and environments.

7.1.2 Procedure

The procedure involves a validated, reliable, and scalable methodology for staining coral larvae using vital dyes (specifically Nile blue and neutral red) to enable visual differentiation of larval cohorts in both laboratory and field settings. The protocol is applicable to a wide range of coral species across multiple developmental stages and has been demonstrated at research and restoration scales. The procedure involves preparing stains, handling larvae, exposure protocols, survival and settlement assessment, and field deployment.

7.1.3 Prerequisites

This section outlines the prerequisite documentation, equipment, datasets, and standards required to effectively implement this SOP for vital staining of coral larvae. This includes procedural dependencies, technical resources, environmental considerations, personnel requirements, and regulatory alignment. These dependencies ensure standardisation, reproducibility, and safe operational execution in both laboratory and field contexts.

Data, Tools, and Software Required

- **Experimental Metadata Log** (e.g., Excel, FieldBook App)
  - For recording treatment combinations, exposure durations, larval counts, survival, settlement scores.

- **Image Capture Software**
  - *ImageJ* for post-processing of larval photographs.

  - **Safety Data Sheets (SDS)**
  - For all stain compounds used (e.g., Nile Blue A, Neutral Red). These must be accessible in the laboratory or field site.

Environmental considerations

Disposal of Stain Solutions

Vital stains are considered non-toxic in dilute concentrations, but disposal should comply with institutional and environmental regulations to avoid reef contamination. Use absorbent waste or activated charcoal traps where possible.

Marine Debris Prevention

All disposable plastics (pipettes, vials, well plates) should be captured, labelled, and disposed of in designated marine waste systems.

Lighting & Temperature Control

Incubations should be done in the dark at ambient water temperatures (typically 26–28°C). Light exposure can interfere with stain uptake.

Seawater Source

Filtered (0.2 µm) or UV-treated seawater is required to maintain sterility and avoid microbial interference.

Personnel Prerequisites and Training

Core Competencies Required:

- Experience in coral spawning and larval handling.
- Basic microscopy and larval identification skills.
- Familiarity with chemical safety and handling of biological materials.
- Proficiency in R or equivalent software for analysis (optional but recommended).

Training Resources:

- SeaSim Coral Husbandry Course or equivalent institutional training.
- Online modules via Reef Restoration and Adaptation Program (RRAP).
- Internal lab inductions including larval staging and morphology.

Alignment with International Standards

While there is no formal ISO standard for coral larval staining, the procedure aligns with best practices from:

- Great Barrier Reef Marine Park Authority (GBRMPA) protocols
- Australian Institute of Marine Science (AIMS) SeaSim protocols
- The Coral Restoration Consortium (CRC) best practice manuals
- UNESCO IOC protocols for marine biological experimentation

7.1.4 Risks and Hazards

This procedure involves handling marine larvae, chemical stains, and working in controlled aquaria or field settings. The following hazards, risks, and control measures must be considered before, during, and after implementation. Partner organisations should integrate this information with their institutional risk management frameworks and complete formal risk assessments prior to field or lab work.

Chemical Hazards

Hazard	Risk	Control Measures
– **Nile Blue & Neutral Red**	Eye, skin, and respiratory irritation; staining of skin and surfaces; unknown	Use PPE (gloves, safety glasses, lab coat); prepare solutions in fume hood or ventilated area; avoid

	long-term ecotoxicological impact at high concentrations	direct contact; label all containers clearly; dispose of according to institutional chemical waste procedures
Solution Spillage	Environmental contamination or lab surface staining	Use spill trays during preparation; clean spills immediately with paper towel and dispose in designated chemical waste
Chemical Storage	Degradation of stain quality or contamination	Proper labelling and segregation of chemicals to prevent cross-contamination. Storage in appropriate containers (e.g. amber bottles for light-sensitive stains) that are tightly sealed and chemically compatible. Controlled storage environment – maintain recommended temperature, humidity, and protection from light to preserve chemical stability. Regular monitoring and inventory checks to identify expired or degraded chemicals. Use of secondary containment to contain leaks or spills that could lead to contamination

Biological and Environmental Risks

Hazard	Risk	Control Measures
– Live Coral Larvae	Cross-contamination between species or tanks; mortality due to poor water quality	Use species-specific vials/wells; sterilise containers between uses; use filtered (0.2 µm) or UV-treated seawater; monitor temperature and DO regularly
Release of Stained Larvae	Accidental introduction of larvae or chemicals into natural reef without permits	Ensure secure netting or enclosures in field trials; collect all settlement tiles post-trial; follow permit conditions strictly
Wastewater Disposal	Discharge of chemical-stained water into sensitive environments	Pre-treatment of wastewater to remove or neutralise hazardous chemicals before disposal. Use of dedicated chemical waste containers to collect and segregate stained or contaminated water from general wastewater. Adherence to environmental regulations and disposal guidelines for chemical effluents, including obtaining relevant permits if required. Regular inspection and maintenance of drainage and disposal systems to prevent accidental discharge. Training of personnel on correct disposal procedures and environmental risks associated with improper wastewater handling.

Operational and Handling Risks

Hazard	Risk	Control Measures
– Microscopy and Scoring	Eye strain, fatigue, bias in scoring	Work in 20-minute intervals with breaks; rotate observers if possible; standardise scoring criteria; use same observer across replicates if feasible
Manual Pipetting and Vial Handling	Repetitive strain, dropped vials, mislabelling	Use pipette aids or multichannel pipettes; label vials clearly and double-check before adding larvae; use staging trays
Fieldwork (at-sea deployment)	Drowning, sunburn, dehydration, boat-related injuries	Mandatory use of personal protective equipment (PPE) such as life jackets, hats, sunglasses, and sunscreen. Comprehensive pre-deployment safety briefings covering emergency procedures, vessel safety, and environmental conditions. Access to sufficient drinking water and shaded rest periods to prevent dehydration and heat-related stress. First aid kits and trained personnel available on board to respond to injuries or medical emergencies. Use of vessel safety protocols, including maintaining communication with shore, checking weather forecasts, and ensuring proper vessel maintenance and operation.

Emergency Procedures

- **Chemical Exposure (Skin/Eyes):**

  Flush affected area with water for at least 15 minutes. Remove contaminated clothing. Seek medical advice immediately. SDS documentation should be present onsite.

- **Larval Spillage or Cross-Contamination:**

  Remove any larvae outside designated containers. Sterilise affected surfaces using 70% ethanol or autoclaving if required. Report if any permit-sensitive species are affected.

- **Field Emergency (Weather, Injury):**

  Abort deployment if weather conditions exceed operational limits. Contact base via VHF or satellite comms. Follow institution fieldwork emergency protocol.

- **Waste Mismanagement:**

  Report immediately to the safety officer. Isolate contaminated area and recover any leaked solution for proper disposal.

Note: All researchers must complete institutional chemical safety training and read the SDS for each stain prior to handling. Emergency contacts, first aid kits, and fire extinguishers must be available in all laboratories and field sites.

7.1.5 Equipment and Materials

1. Procedure equipment

Table 7.1.1: Procedural equipment required

Item	Description / Notes	Approximate Quantity
− Vital Stains	Nile Blue, Neutral Red (analytical grade, powder form)	~100 mg–1 g per experiment depending on concentration
Scintillation Vials (10 ml)	Glass vials for incubation of individual larval batches	100–200 units
6-well Culture Plates	For grouped larval staining and incubation	10–20 plates
Filtered Seawater (0.2 µm)	Pre-filtered seawater for staining, rinsing, and holding larvae	~20–40 L per experiment
Pipettes & Tips	Manual or electronic pipettes (0.1–10 ml range)	2–3 pipettes, 100–200 tips
Digital Timer / Clock	To synchronise staggered incubation times	1 unit
Dissecting Microscope	For scoring larval stain intensity and survival	1–2 units
Imaging Software	e.g. *Toupview* (ToupTek) for documenting stain intensity	1 install per workstation
Settlement Tiles / Substrates	Preconditioned tiles (e.g., 5 × 5 cm or Porolithon chips)	20–50 depending on replicate size
Glass Measuring Cylinders & Beakers	For preparation and dilution of stain solutions	2–3 per stain type
Plastic Transfer Pipettes	For transferring larvae between containers	50–100 units
Water Bath or Controlled-Temperature Room	Optional: maintain constant temperature during incubation	1 unit (optional)
Labeling Tape & Markers	For clearly marking replicates and controls	1 roll + 1 marker
Camera (optional)	For in situ field documentation of stained larvae	

2. Personal protective equipment (PPE) and other safety equipment

Table 7.1.2: PPE required

Stain	Typical Concentration Range	Incubation Time Range
– Nitrile Gloves	Prevent skin contact with stains	All users handling stains or larvae
Safety Glasses	Protect eyes from accidental splashes	All users handling liquids
Lab Coat or Apron	Protect clothing and skin from staining	Required in all lab settings
Fume Hood or Ventilated Work Area	Recommended during stain preparation	Optional but preferred
Chemical Waste Containers	For disposal of staining solutions and contaminated water	Minimum 2 containers (labelled)
First Aid Kit	Standard emergency first aid supplies	1 per lab/field site
Spill Kit	Absorbent material, gloves, bags for chemical spill response	1 kit per site
Fire Extinguisher	CO_2 or dry powder extinguisher if working with electronic imaging	1 per lab room (per regulations)
Sharps Container	If using glass pipettes or broken vials	Optional

7.1.6 Implementation steps

1 Procedure Preparation

- Ensure all equipment and PPE listed in Section 6 are available and functional.
- Label all containers clearly with stain type, concentration, and timepoint.
- Pre-condition seawater (filtered to 0.2 µm) to match larval culture temperature.
- Prepare stain stock solutions using analytical grade powders. Handle using gloves and eye protection in a ventilated area.

2 Larval Collection and Preparation

- **Step 1: Larval Collection**
    - Collect larvae using larval concentrators such as:
        - Pool scoops lined with 125 µm mesh
        - PVC concentrator tubes fitted with plankton mesh

- Gently transfer larvae to temporary culture containers (e.g., 50 L Nally bins or aquaria) using wide-bore pipettes or beakers.
- Maintain mild aeration or water circulation using an aquarium air pump.

3 Larval Staining Protocol

- **Step 2: Staining Setup**
  - Pre-mix stains (e.g., Nile blue, neutral red) in clean filtered seawater to required concentrations (1–1,000 mg L^{-1}).
  - Add larvae to the staining containers at the defined start times so that all exposures finish simultaneously.
  - Maintain temperature at ambient rearing conditions.
  - Provide gentle flow or aeration to ensure oxygenation during incubation.
- **Step 3: Staining Execution**
  - Incubate larvae for prescribed durations based on the stain (typically 10–120 minutes).
  - Observe larval condition periodically to detect any adverse responses.
  - At completion, carefully remove larvae and immediately rinse 2–3 times in fresh filtered seawater.

Table 7.1.3: Staining guidelines

Stain	Typical Concentration Range	Incubation Time Range	Notes
Nile Blue	1–1,000 mg L^{-1}	10–120 minutes	Strong visibility, low toxicity
Neutral Red	1–100 mg L^{-1}	~10 minutes	Strong visibility, higher concentrations reduce survival

7.5 Scoring of Stain Intensity and Survival

- Step 4: Scoring Protocol
  - Subsample 10–20 larvae per replicate into a petri dish.
  - Use a dissecting microscope under consistent light and magnification.
  - Score using an ordinal staining scale:

    1 = No stain

    2 = Light

    3 = Medium

    4 = Strong
  - Record number alive vs. dead to quantify staining-related mortality.

- All observations must be made by the same observer where possible.

7.6 Larval Imaging

- Step 5: Visual Documentation
    - Use a digital dissecting microscope with consistent light settings (e.g., Toupview software).
    - White balance and contrast settings should be standardized for cross-replicate comparison.
    - Photograph representative stained larvae and settlement surfaces post-deployment.

7.7 Notes and Special Considerations

- Always include unstained controls for baseline comparisons.
- Minimize handling time and avoid temperature shocks.
- Maintain uniform incubation duration, stain concentration, and rinsing procedures across all treatments.
- Prior to any large-scale or novel species application, confirm stain compatibility in a pilot trial.
- Ensure water exchange is sufficient to avoid anoxic conditions, particularly for >60 min incubations.
- Store unused stain solutions according to safety guidelines (see Section 7.1.3).

7.2 Hydrodynamic modelling to optimise larval retention during larval releases

7.2.1 Background and rationale

When larvae are freely released into the water column they are rapidly dispersed by currents. Therefore, to optimise larval settlement on target reef areas selected for larval restoration, it is important to understand likely retention periods around different reef areas and the influences of hydrodynamic conditions and weather conditions. Using the model outputs, larval releases onto different reef sites can be planned during extended periods of low flow and increased retention to reduce larval dispersal and increase larval settlement after release.

7.2.2 Procedure

The procedures outlined here are based on an innovative approach developed during the Moving Corals project using 2D or 3D hydrodynamic models and particle tracking tools (e.g., CONNIE, OceanParcels), as well as in situ current measurement using tilt current meters to identify larval release site selection, timing of releases, and likely success assessment of coral larval-based restoration interventions (Gouezo et al. 2025b). They are based on spatial modelling of larval retention that combines predictive hydrodynamic simulations with field validation using current meters, providing a data-informed foundation for coral spawn larval restoration strategies.

The methods are usually applied in the days leading up to larval competency periods and planning for larval releases.

- System design: Use of spatial datasets (e.g., coral cover), coupled with simulation models to predict particle (larvae) movement over 12–24 hours.
- Frequency: Annually during coral spawning season.
- Duration: Each application spans approximately 3–10 days including pre- and post-event fieldwork.
- Maturity level: Mature for model-based predictions; Demonstrated for field validation.

- Adoption level: Multi-organisational use within RRAP; methods are transferrable to national and international coral restoration efforts.

7.2.3 Prerequisites

- **Existing documents**:
    - Modelling Guidelines for Larval Dispersal (Langlais et al. 2024)
    - LOWEL Tilt Current Meter User Guide
    - Gouezo et al. (2025a)
- **Software and tools**:
    - GIS software: QGIS or ArcGIS
    - Hydrodynamic models (eg. RECOM)
    - Particle tracking tools: OceanParcels, CONNIE
    - Data analysis: R or Python
    - Domino software for tiltcurrentmeter set up and data extraction
- **Other requirements**:
    - Personnel must be trained in spatial analysis, basic programming, and underwater deployment procedures.
    - Knowledge of local coral spawning events timing
    - Awareness of site-specific environmental conditions (e.g., prevailing currents, wind exposure).
- **Standards**:
    - Use of GPS with <5 m accuracy

7.2.4 Risks and Hazards

Model-based methods:

- **Data quality risk**: Inaccurate coral cover or hydrodynamic data may compromise predictions
- **Operational risk**: Model complexity may lead to computational delays if not streamlined

Field validation methods:

- **Instrument loss or damage**: Ensure all loggers are secured with backups and clearly tagged

Emergency procedures:

- Carry VHF radio or satellite communication device during field deployments.
- Follow site-specific dive safety protocols (e.g., buddy system, surface support)

7.2.5 Equipment and Materials

Procedure equipment

• Computer with hydrodynamic model access

• GPS device with <5 m accuracy

• GIS software (QGIS or ArcGIS)

• R or Python for data processing

• LOWEL Tilt Current Meters or equivalent

• Dive gear and underwater slates

• Secure mounting materials (rope, cable ties, dive weights)

Personal protective equipment (PPE) and other safety equipment

• Dive-certified PPE (e.g., wetsuits, gloves, weight belts)

• Surface buoy, dive flag, and underwater communication signals

• First aid kit and emergency O_2

• VHF or satellite phone for remote site communication

7.2.6 Implementation steps

Predicting the residence time of larvae at restoration sites using dispersal models

Note: This method requires access to a 2D or 3D hydrodynamic model with forecasting capabilities at least 2 days in advance, and a particle dispersal tool such as CONNIE or OceanParcels**.** Lower-resolution models (with grid sizes of 100 m or less) are often better at capturing reef-scale hydrodynamics.

This method is adapted from Gouezo et al. (2025a) and outlines step-by-step guidance for predicting how long larvae remain near restoration sites following release. The goal is to help identify the locations and times with the highest potential for local larval retention, maximizing the chances of successful local coral settlement.

Step 1: Map Candidate Restoration Sites
Using GIS software such as QGIS or ArcGIS:

- Create a .shp file with equal-sized polygons (e.g., 1-hectare circles or rectangles) to represent candidate restoration sites.

Step 2: Determine Potential Larvae Delivery Dates

- Based on known coral spawning dates, estimate larval delivery windows by adding 4 to 8 days post-spawning.

Step 3: Run Dispersal Simulations

- For each candidate site, release a set number of particles (e.g., 5,000) during each hour starting from 6:00 AM to 5:00 PM over the selected delivery dates.
- Track particle movement for 24 hours per simulation.
- This will generate 12 dispersal simulations per site per day.

Step 4: Identify Optimal Times for Larval Delivery

- Using the simulation outputs (often formatted as a data frame with GPS locations over time), calculate how many particles remain within 1 hectare of the release site each hour after release. Example R code available: [Larval Delivery Sim GitHub Repository]
- Summarize the data into a table showing particle counts over time for each delivery scenario.
- Calculate the Particle Residency Time (PRT) using the e-folding time method:
    - E-folding time is the number of hours it takes for particle count to drop to 1/e (~36.8%) of the original number.
    - For 5,000 released particles, the threshold is approximately 1,800.
    - This is a common metric to estimate how long larvae stay near the release site (Gouezo et al., 2021).
- Use graphs or heatmaps to visualize which sites and delivery times yield the highest PRT. These times are ideal for planned larval releases.

Example Interpretation

The graph below shows particle concentration within 1 hectare of the release site. Each row is a delivery date; each column is an hour of the day.

- Black bars show particle concentration each hour after delivery.
- Dashed line = e-folding threshold (1/e × 5,000 = 1,800)

Example scenarios:

- 🔴 **Day 11652, 6:00 AM**: Particle count drops below 1,800 after **3 hours** → **PRT = 3 hrs**
- 🟢 **Day 11656, 1:00 PM**: Particles remain above 1,800 for **13 hours** → **PRT = 13 hrs** (high local retention!)
- 🟠 **Day 11655, 12:00 PM**: Particles stay near the site for 3 hours, then leave and briefly return at lower concentrations → **PRT = 3 hrs**

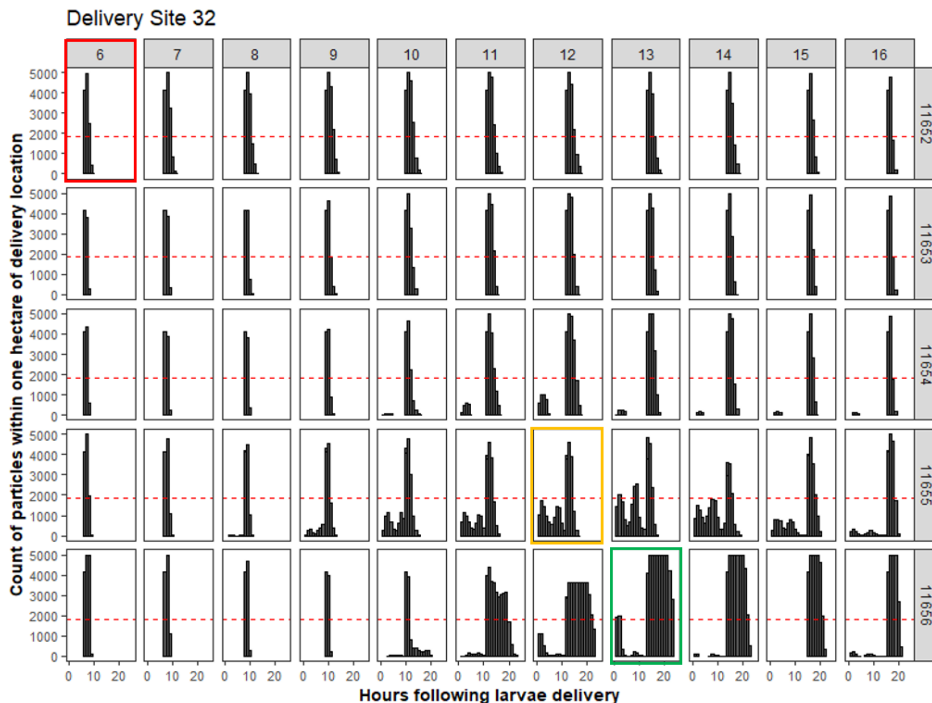


Figure 7.2.1: Multi plots showing the number of particles within one hectare of delivery locations for each dispersal simulation through time (release time as column) and day# as row at one delivery site

Investigating local retention characteristics of restoration sites using tilt current meters

In situ instruments that measure currents provide valuable data to ground truth dispersal models and are also useful when models are unavailable. These instruments give information on current speed and direction at regular time intervals (e.g., every minute). They are particularly useful for identifying 'slack current' conditions, which typically last between 1 to 3 hours on coral reefs, but may last up to 12 hours at specific times and locations. Understanding these timeframes is crucial for maximizing local larval retention on restored reefs.

Further reading on this topic can be found in Gouezo et al. (2025a).

Instrument Example: LOWEL Tilt Current Meters

The example below uses the **LOWEL tilt current meters**. Before following the steps below, please read the instructions manual to understand how to set up the loggers:
LOWEL TCM-1 Tilt Current Meter User Guide (Download the **'Universal User Guide'**).

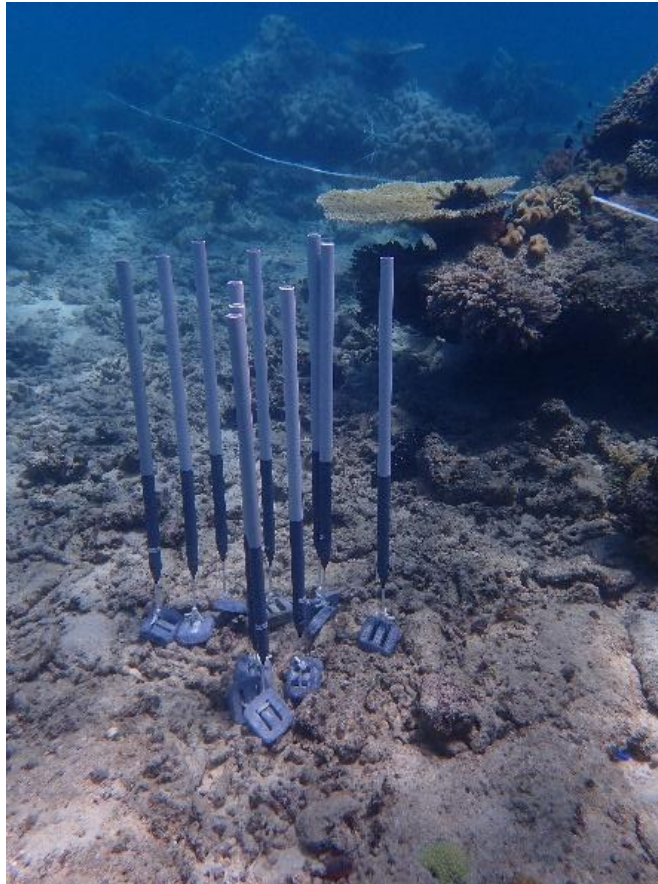


Figure 7.2.2: Photo showing tilt current meters set up on dive weight

Step 1: Prepare Logger for Deployment

- Follow the user guide to set up the logger for data collection.

Step 2: Deploy Logger on the Reef Using One of Two Options

1. **Option 1**: Find a strong crevice on the reef and tie the logger using a bowline knot. Ensure that the distance between the knot and the end of the logger is XX cm, as advised in the user guide.
2. **Option 2**: Attach the logger to a dive weight by tying a bowline knot or securing it with a cable tie.

- Record the following:
    - **Serial number of the logger**
    - **GPS location** of deployment
    - **Depth** where the logger is tied
    - **Date and time** of deployment
- Leave the logger in place for the necessary period.

Step 3: Retrieve Current Data the Day Before Planned Larvae Delivery

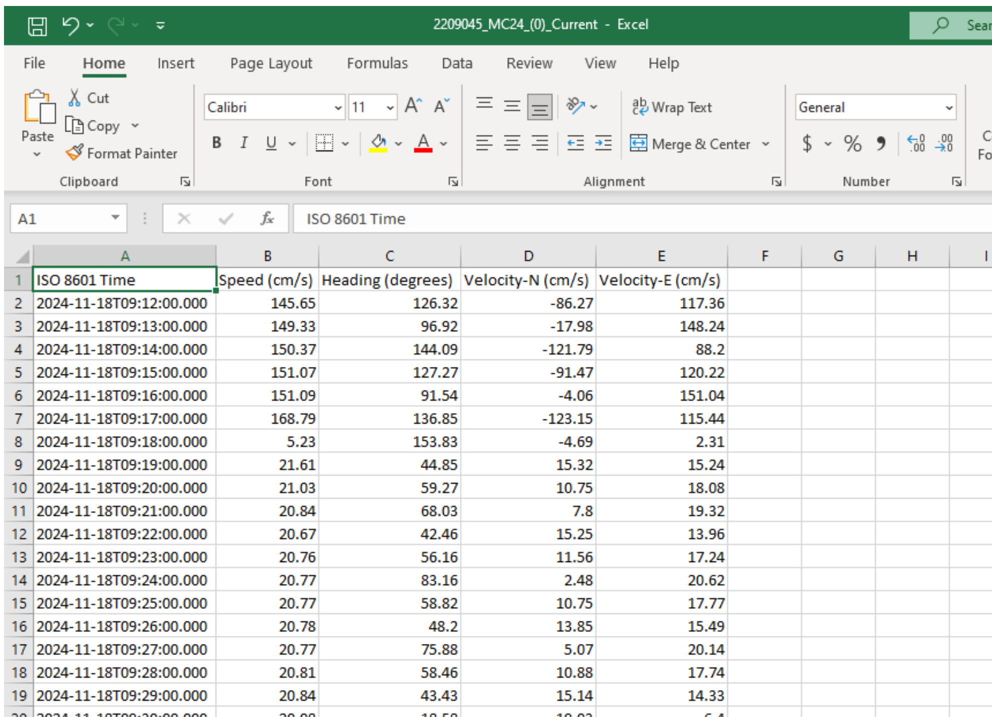
- At 4-5 PM the day before the intended larvae delivery:
    - Exchange the logger with a new one.
    - Record the date and time of retrieval, and the date, time, site, and depth information of the new logger.

Step 4: Download the Data

- Download the data using Domino software, as detailed in the user guide.
- Convert the data into a .csv file for easier analysis.

Step 5: Explore the Data

- The data will be provided at minute intervals and in cm/s



	A	B	C	D	E
1	ISO 8601 Time	Speed (cm/s)	Heading (degrees)	Velocity-N (cm/s)	Velocity-E (cm/s)
2	2024-11-18T09:12:00.000	145.65	126.32	-86.27	117.36
3	2024-11-18T09:13:00.000	149.33	96.92	-17.98	148.24
4	2024-11-18T09:14:00.000	150.37	144.09	-121.79	88.2
5	2024-11-18T09:15:00.000	151.07	127.27	-91.47	120.22
6	2024-11-18T09:16:00.000	151.09	91.54	-4.06	151.04
7	2024-11-18T09:17:00.000	168.79	136.85	-123.15	115.44
8	2024-11-18T09:18:00.000	5.23	153.83	-4.69	2.31
9	2024-11-18T09:19:00.000	21.61	44.85	15.32	15.24
10	2024-11-18T09:20:00.000	21.03	59.27	10.75	18.08
11	2024-11-18T09:21:00.000	20.84	68.03	7.8	19.32
12	2024-11-18T09:22:00.000	20.67	42.46	15.25	13.96
13	2024-11-18T09:23:00.000	20.76	56.16	11.56	17.24
14	2024-11-18T09:24:00.000	20.77	83.16	2.48	20.62
15	2024-11-18T09:25:00.000	20.77	58.82	10.75	17.77
16	2024-11-18T09:26:00.000	20.78	48.2	13.85	15.49
17	2024-11-18T09:27:00.000	20.77	75.88	5.07	20.14
18	2024-11-18T09:28:00.000	20.81	58.46	10.88	17.74
19	2024-11-18T09:29:00.000	20.84	43.43	15.14	14.33

- For better visualization, you may want to average the data at 2-minute or 5-minute intervals.

Important: When averaging, always average the velocity components (u and v vectors) first. Averaging the direction directly, as it is circular data, will lead to incorrect results.

- Once the u and v components are averaged, you can recalculate the 5-minute average speed and direction.
- **R Code Example**: The provided R code shows how to create a plot showing the current speed and direction, with arrows indicating the direction of flow using:
    1. non-averaged data at minute intervals.
    2. Average data at chosen interval: ie. 5-minute, 10 minutes etc…

```
# 1. Plot non-averaged current data at minute intervals

setwd("XXXX")

library(tidyverse)
library(lubridate)
library(scales)
library(ggnewscale)

data_f<-read.csv("XXXX.csv")  ## add name of .csv file from the currentmeter

#define date range of when the currentmeter was recording current
Start_date="2024-11-26 00:01"
End_date="2024-11-28 11:00"

names(data_f)
```

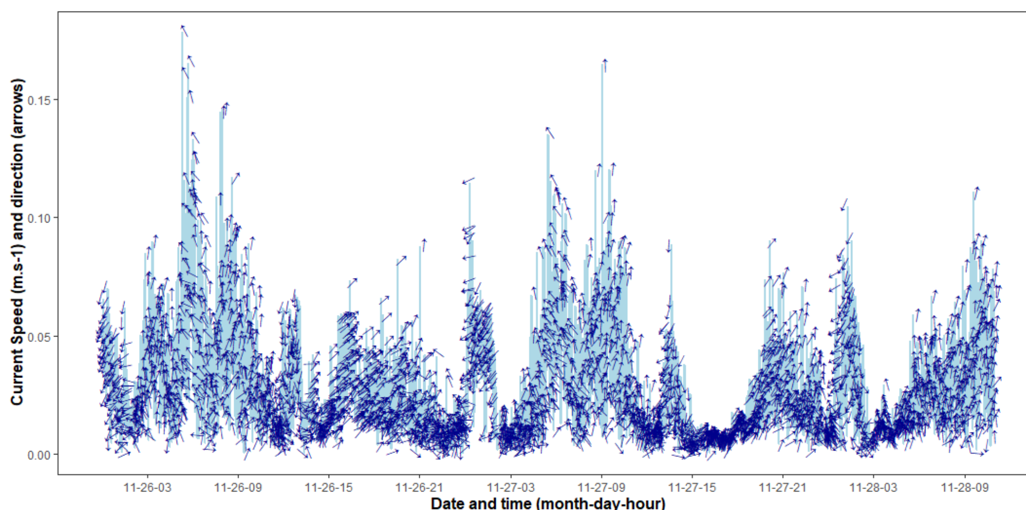
```
str(data_f)

#convert speed in m.s-1 and rename
data_f$speed_f<-data_f$Speed..cm.s./100
data_f$direction_f<-data_f$Heading..degrees.
data_f$u_f<-data_f$Velocity.E..cm.s/100
data_f$v_f<-data_f$Velocity.N..cm.s/100

#sort out time/date format
library(stringr)
data_f$time<-str_sub(data_f$ISO.8601.Time, start=-12,end=-5)
data_f$date<-as.POSIXct(data_f$ISO.8601.Time, tz="Australia/Queensland")
data_f$date<-as.POSIXct(paste(data_f$date, data_f$time), format="%Y-%m-%d %H:%M",
tz="Australia/Queensland")
str(data_f)

#filter dataset to correct start and end date/time of current recording
data_f<- data_f %>%
   filter(date > as.POSIXct(Start_date,  tz="Australia/Queensland"),
          date < as.POSIXct(End_date,  tz="Australia/Queensland"))

p_tiltC<-ggplot() +
  #plot current speed and direction using 12min averages
  geom_line(data=data_f, aes(x=date, y=speed_f), color="lightblue",linewidth=1)+
  # Compute the direction of the current using decimal degrees. Assuming that you want 0 degrees
to be North (arrow point up), use the following
  geom_text(data=data_f,aes(angle=-direction_f+90,x=date, y=speed_f),color="darkblue", label="→",
size =4,fontface="bold")+ # the label should be an arrow pointint East: →
  labs(x = "Date and time (month-day-hour)",
       y = "Current Speed (m.s-1) and direction (arrows)",
       color = "Legend")+
  scale_x_datetime(labels = date_format("%m-%d-%H", tz="Australia/Queensland"),
                   date_breaks = "6 hours")+
  theme_bw()+
  theme(axis.text=element_text(size=9), axis.title=element_text(size=12, face = "bold"))+
  theme(panel.grid.major = element_blank(), panel.grid.minor = element_blank())+
  theme(panel.grid.minor.x = element_blank())
p_tiltC
```



 # 2. Plot 5min-averaged current data

```
head(data_f)

# Round the time to nearest 5-minute interval
data_f$date_rounded <- round_date(data_f$date, unit = "5 minutes")

# Group by rounded time intervals and calculate mean of u_f and v_f
data_f_5min <- data_f %>%
```

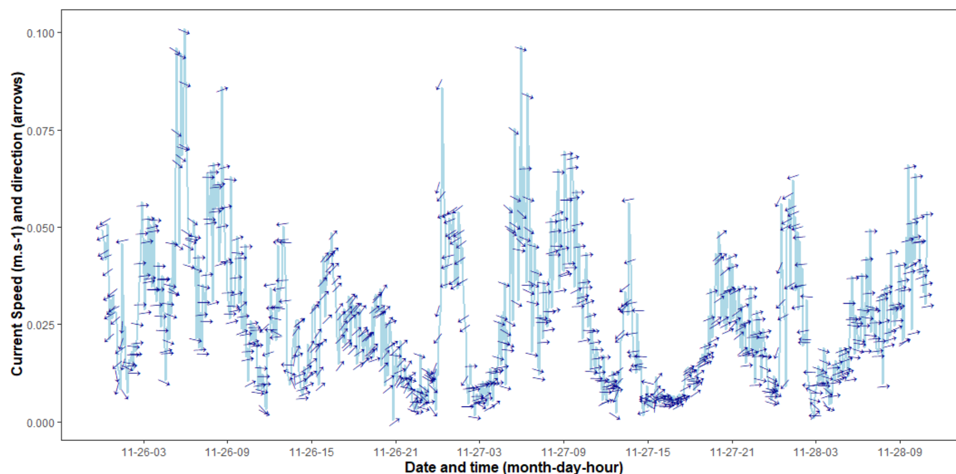
```
  group_by(date_rounded) %>%
  summarise(u_f = mean(u_f),
            v_f = mean(v_f))

# Calculate speed_f and direction_f
data_f_5min <- data_f_5min %>%
  mutate(speed_f = sqrt(u_f^2 + v_f^2),  # Calculate speed_f using Euclidean norm
         direction_rad = atan2(v_f, u_f),  # Calculate direction in radians
         direction_f = (atan2(v_f, u_f) * (180 / pi) + 360) %% 360)  # Calculate direction_f in
degrees


# View the first few rows of the new dataset
head(data_f_5min)

#filter dataset to correct start and end date/time of current recording
data_f_5min<- data_f_5min %>%
  filter(date_rounded > as.POSIXct(Start_date,  tz="Australia/Queensland"),
         date_rounded < as.POSIXct(End_date,  tz="Australia/Queensland"))


p_tiltC<-ggplot() +
  #plot current speed and direction using 12min averages
  geom_line(data=data_f_5min, aes(x=date_rounded, y=speed_f), color="lightblue",linewidth=1)+
  # Compute the direction of the current using decimal degrees. Assuming that you want 0 degrees
to be North (arrow point up), use the following
  geom_text(data=data_f_5min,aes(angle=-direction_f+90,x=date_rounded,
y=speed_f),color="darkblue", label="→", size =4,fontface="bold")+ # the label should be an arrow
pointint East: →
  labs(x = "Date and time (month-day-hour)",
       y = "Current Speed (m.s-1) and direction (arrows)",
       color = "Legend")+
  scale_x_datetime(labels = date_format("%m-%d-%H", tz="Australia/Queensland"),
                   date_breaks = "6 hours")+
  theme_bw()+
  theme(axis.text=element_text(size=9), axis.title=element_text(size=12, face = "bold"))+
  theme(panel.grid.major = element_blank(), panel.grid.minor = element_blank())+
  theme(panel.grid.minor.x = element_blank())
p_tiltC
```



7.3 Larval releases onto reefs

7.3.1 Background and rationale

One of the methods employed by Moving Corals to deliver larvae to the reef substrate are “free releases”, which is delivery from either vessel-based tanks or from larvae concentrated from culture pools delivered down a transfer pipe to the reef substrate. The rationale for a free release can either be deployment of

larvae onto the substrate during a slack current to maximise larval retention, or during a changing tide to maximise dispersal of larvae across a reef area.

7.3.2 Procedure

Larvae are delivered to the reef from the surface through a transfer hose (typically 4” diameter) onto the reef substrate. Larvae are either from i) concentrated larvae from reef culture pools which are deployed using pipes attached to the bottom of the vinyl funnel section at the base of the larval culture nets (see Harrison 2024b) or transferred to the delivery site in large tubs (e.g., 250 litre), or ii) direct from vessel based rearing tanks. The transfer hose is attached to the reef substrate via stakes, and deployment occurs by slowly decanting the larvae through the transfer hose to maintain dispersal onto the reef substrate and avoiding high volumes of larvae being forced through the pipe, which results in “clouds” that disperse throughout the water column.

7.3.3 Prerequisites

High numbers of competent larvae from the mass larval rearing phase (Phase 3) are a prerequisite to larval delivery to ensure localised settlement. As a prerequisite, assess the competency of the larval cultures using rapid settlement assays (e.g., Heyward & Negri 1999). For delivery, if larvae are cultured in pools, the pool can be towed over the delivery site, or the larvae can be concentrated and collected into a transfer tub prior to delivery if deployment occurs in a habitat located away from the pools (see section 6.1). If the larvae are deployed from vessel-based aquaculture, then the correct hose adapters (camlock fittings) and hoses (4” transfer hose) are required to allow the tanks to drain from the base.

Knowledge of tide and current flow around the delivery site is critical to the delivery stage, noting that speed and direction of surface currents can differ from that above the reef substrata. Hydrodynamic modelling can help predict likely dispersal sites based on changes in weather and hydrodynamic conditions (Gouezo et al. 2025a; Section 7.2) to increase the efficiency of delivery.

7.3.4 Risks and Hazards

Standard vessel safety measures such as confirmed vessel seaworthiness, experienced skipper and crew, vessel safety and emergency gear, radio and mobile or satellite phone communications and GPS tracking to identify hazards are essential for safe operations around reef areas. In addition, all crew members need to wear PFDs (personal floatation devices) to ensure their safety while operating over water.

7.3.5 Equipment and Materials

Access to small vessels with space for a skipper and at least two crew members with snorkelling equipment to attach the transfer hose to the reef. The transfer hose is ideally 4” in diameter to facilitate flow, and up to ~12 m length depending on depth of the deployment target sites. If deploying from vessel or tanks, ensure correct camlock fittings/couplings (typically either A or B) to attach the transfer hose to the tank. A surface float is used to maintain buoyancy of the transfer hose at the surface, and stakes/cable ties are used to attach the end of the pipe to the reef. If delivering from concentrated larvae, 100-250 litre tubs and a handscoop.

7.3.6 Implementation steps

1. Prior to concentration and transfer of larvae to the delivery site, a 4” transfer hose (typically PVC, ideally transparent) is attached to the reef substrate by a snorkel pair (if shallow delivery <6m). The direction of the end of the pipe is oriented parallel to the reef so that larvae are delivered across the reef substrate, and not either onto or angled away from the substrate, which can result in plumes of larvae being advected away from the substrate and to the water column.
2. Ensure the delivery of larvae matches the required tidal conditions (e.g. prior to the peak of low tide for optimal larvae retention during slack conditions, or with increasing current flow if dispersing larvae across a habitat is required). If possible, use tilt meters or similar instruments to measure current direction and strength prior to and during larval release.

3. At arrival at delivery site, anchor where possible. If delivery from vessel, attach camlock fittings from transfer pipe to tanks. If delivering from the boat with concentrated larvae, align the transfer hose with the side of the boat. If delivery from larval culture pools, position the larval pool over or near the delivery site and attach the transfer pipe to the funnel at the base of the culture net (Harrison 2024b).
4. Deliver larvae slowly down the transfer pipe – either using a handscoop or small bucket for concentrated larvae from transfer tubs, or by regulating the flow from the larval culture pools by slowly drawing the net up to concentrate the larvae into the base of the net and down the funnel into the pipe (Harrison 2024b), or from the tank using the flow valve. A surface snorkeller can be used to communicate to the person in charge of delivery to visually confirm that larvae are being delivered onto the reef substrata at a low flow rate to avoid a "cloud" that disperses throughout the water column. When the final larvae are deployed, avoid slowly add clean seawater to rinse the remaining larvae from the pipe system but avoid high flow rates while rinsing with seawater.
5. Once larvae are released, to avoid turbulence and disturbance from the vessel and motors, either i) keep the vessel anchored onsite for 1-2hrs following release, or ii) allow the vessel to drift from the anchor location prior to engaging engines (assuming within visual site and communications with shore or vessel team and additional boat support can be deployed if required).

7.4 Larval seedbox controlled releases

7.4.1 Background and rationale

On coral reefs, the experimental deployment of competent coral larvae using restrained approaches – such as larval enclosures – has successfully enhanced coral settlement rates (Heyward et al. 2002, Edwards et al. 2015, dela Cruz and Harrison 2020), and in some cases, has restored breeding populations of mature corals (dela Cruz and Harrison 2017, Harrison et al. 2021). These experiments have demonstrated that directly settling larvae onto reefs can be effective in restoring degraded or larvae-limited systems. However, their application so far been mainly limited to relatively small areas (e.g. 7-96 m^2) due to the logistical challenges of deploying tents and nets underwater. To address the issue of scalability, the concept of large-scale coral reef restoration through unrestrained releases of sexually produced coral larvae was modelled as a viable strategy by Doropoulos et al. (2019a). This approach aims to mimic natural broadcast spawning reproductive processes by releasing competent larvae onto degraded reef areas en masse to settle across broad areas of reef through targeted placement of releases. Despite its theoretical potential, there are currently no published empirical studies demonstrating the success of unrestrained larval releases in reseeding reefs at scales required to be ecologically meaningful (>1ha). The success of such releases is further complicated by species-specific larval competency windows interacting with the complex hydrodynamics of coral reef systems, which can disperse larvae unpredictably and reduce the likelihood of successful recruitment. Given the complex physical and biological interactions affecting larval propagules, slowing down the release and dispersal of competent larvae while maintaining their proximity to the benthos should increase both the likelihood of larvae encountering suitable substrate following their release, and the proportion of larvae competent to settle. This strategy leverages natural hydrodynamic processes to reduce larval loss to advection and improve retention near target restoration areas, while enhancing local retention through an extended release allows for a wider window of larval competency in multi- species wild spawn collections.

- **System design**: field-based larval seedboxes for deployment post-spawning
- **Frequency**: single or multiple deployments after larval cultivation
- **Duration**: 2-48 hr deployments
- **Maturity level**: demonstrated in field settings
- **Adoption level**: validated (organisational use within RRAP), intended uptake at national and international levels.

7.4.2 Procedure

The procedure involves collecting cultured coral larvae (Phase 3) and transferring them into larval seedboxes to increase gradual larval releases on reef deployment sites over extended periods.

7.4.3 Prerequisites

Prior to implementation, all personnel should be trained in coral spawning timing, larval rearing, and safe fieldwork operations on reefs. Site selection must be conducted based on reef accessibility, wave exposure, and substrate composition. Experimental setup requires prefabricated seedboxes and access to competent coral larvae.

Relevant documentation includes protocols on:

- Coral larval culture and competency testing
- Waterproofing and securing field experimental devices
- Deployment planning using tide and weather forecasts

Tools required include:

- GPS units for precise placement
- Diving equipment for seedbox positioning and retrieval
- Data sheets for environmental and experimental condition logging

Personnel should be competent in underwater navigation, coral handling, and reef-safe anchoring methods.

7.4.4 Risks and Hazards

Risks associated with larval seedbox deployment include physical injury during boat-based or underwater work, entrapment or entanglement with lines or devices, and potential damage to coral habitat. Equipment failure, such as detachment of seedboxes, can result in data loss or habitat disturbance.

Environmental risks include altering water flow or shading resident benthic organisms. Larval mortality may occur due to prolonged exposure to air, high temperatures, predation following transfer, and possible oxygen depletion if water flow is too low and larval concentrations are too high.

Controls include:

- Use of buddy systems during all diving operations
- Pre-deployment checklists for gear and seedbox seals
- Using biodegradable or reef-safe materials for all attachments
- Field testing anchor points before full deployment

7.4.5 Equipment and Materials

Procedure equipment

- Seedboxes, dimensions standardized per study design
- Anchoring devices (e.g., sand screws or weighted bases) for securing boxes
- Coral larvae (competent stage, concentration determined by pilot testing)
- Surface marker buoys to indicate seedbox locations
- Deployment platform (e.g., boat or barge)

- Labelling tags (engraved or waterproof)
- Backup seedboxes and larvae in case of field losses
- GPS units for georeferencing each seedbox
- Underwater slates for dive data recording

Personal protective equipment (PPE) and other safety equipment

- Dive gear including fins, mask, snorkel, BCD, and regulators
- Gloves for handling equipment and sharp coral substrate
- Full wetsuits for protection against stings and abrasions
- Safety knives or line cutters for entanglement risks
- Life jackets for all personnel on boats
- First aid kit (marine-specific)
- Emergency communication device (e.g., VHF radio or satellite phone)

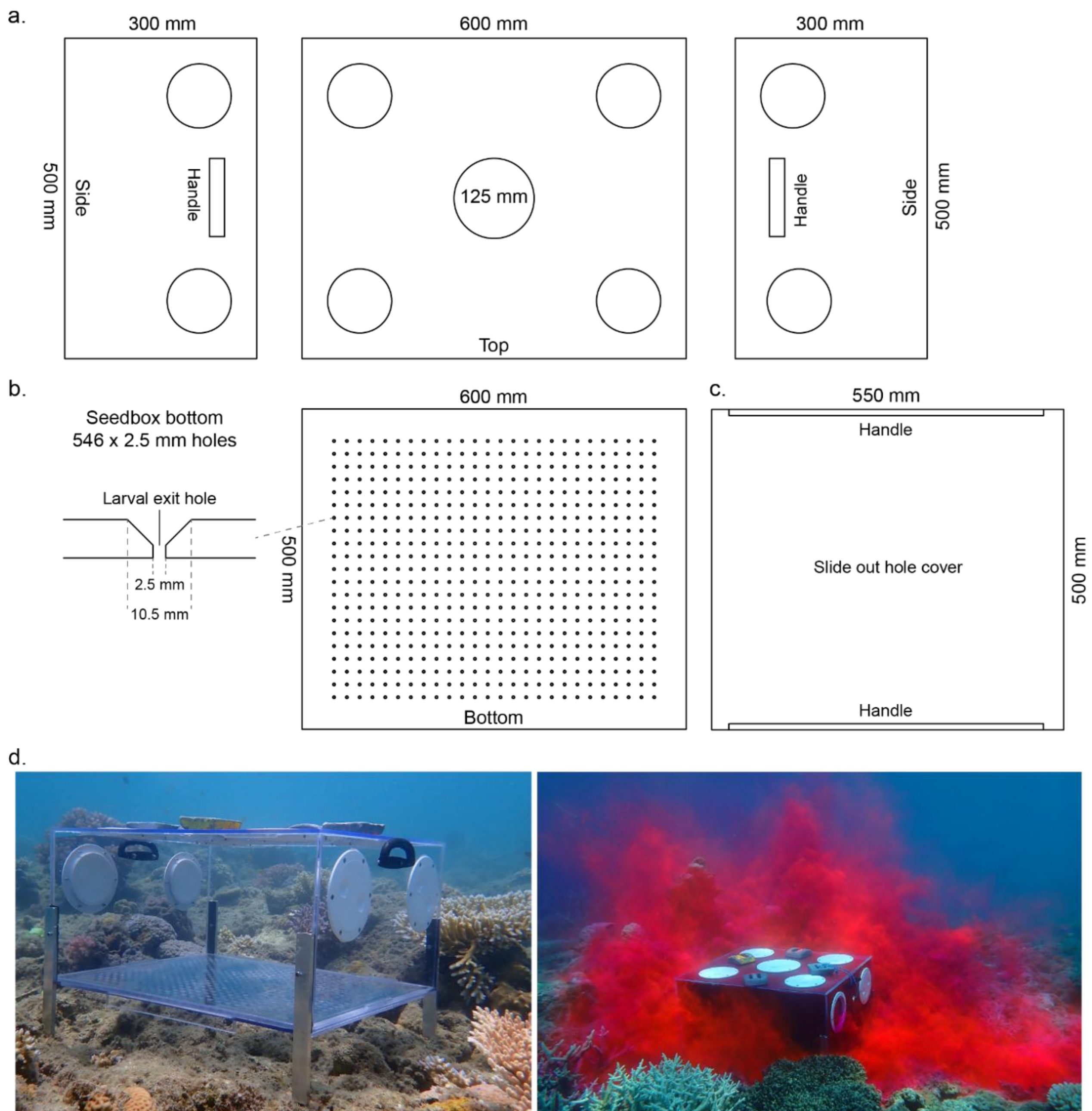


Figure 7.4.1: Top and sides of a larval seedbox showing placement of eight, 100 mm holes and a central 125 mm hole on top for inspection ports. (b) Bottom side showing the drill pattern for the 546 holes, each tapering from 10.5 mm to 2.5 mm to encourage larval escape, and (c) the hole cover guides on the base and a 550 x 500 mm cut section of 6 mm polycarbonate sheet that slides over and covers the holes in the base. (d) Larval seedbox (left) prior to larval deployment showing it raised 50 mm from the substrate on aluminium legs and (right) showing the properties of the rhodamine dye coming out from the bottom and dispersing laterally and vertically (Source: Doropoulos et al. 2025)

7.4.6 Implementation steps

Step 1: Pre-deployment Setup

Confirm that coral larvae have reached competency through prior laboratory culture and monitoring. Prepare all seedboxes under shaded and ventilated conditions to prevent thermal or UV stress. Ensure mesh, inlet ports, and anchor points are intact and functional.

On the day of deployment, load seedboxes onto the vessel with appropriate cushioning to prevent damage during transit. Verify all gear using a pre-departure checklist. Assess sea conditions and confirm

environmental suitability for deployment. Secure GPS devices, pre-labelled seedboxes, and backup materials. Establish tile arrays if using to assess efficacy of deployments.

Step 2: Transport and Placement
At the reef site, use GPS coordinates to navigate to pre-assigned seedbox positions. Divers descend in teams to place seedboxes at designated substrate locations, avoiding live coral or sensitive benthic fauna. Each seedbox should be placed to optimize exposure to water flow while minimizing disturbance.

Use reef-safe anchors to secure seedboxes in place. Attach surface marker buoys with visible tags to aid later retrieval. Record placement coordinates and take photographs for post-deployment verification.

Step 3: Larval Introduction
Carefully transport larval containers to the deployment site in insulated containers. Add larvae to each seedbox via designed entry points, using pipettes or graduated containers to ensure equal volume per replicate.

Minimize larval exposure to air, UV, and temperature fluctuations during transfer. Record key metadata including larval batch ID, time of introduction, water temperature, and volume introduced per box. Gently mix water to encourage uniform dispersal inside the seedbox.

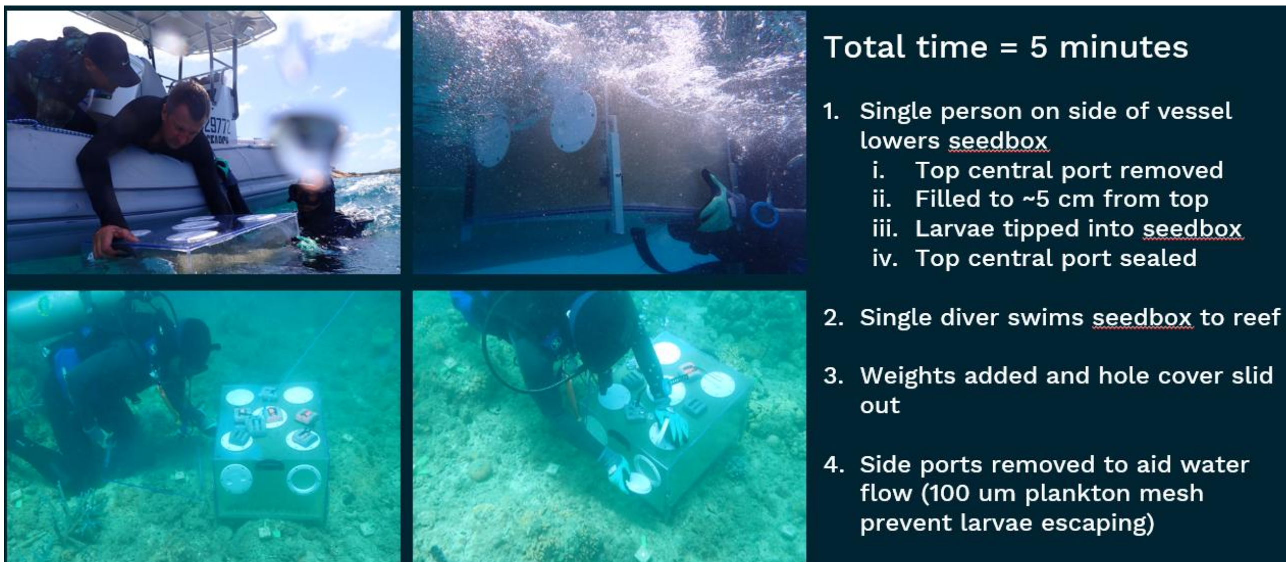


Figure 7.4.2: Larval introduction steps and larval seedbox deployment workflow (Photos: G. Roff)

Step 4: Monitoring and Maintenance
Inspect seedboxes daily or at scheduled intervals to ensure structural integrity. Check for physical displacement, damaged mesh, or sediment accumulation.

Log visual observations on larval activity and box condition. If environmental parameters (e.g., water flow or turbidity) change substantially, record deviations and consider repositioning or pausing deployment. Backup seedboxes may be deployed if failures are observed.

Step 5: Retrieval and Processing
After a defined settlement period (typically 48–120 hours), retrieve seedboxes and surrounding settlement tile array using dive teams. Secure tiles in insulated containers filled with ambient seawater for transport back to shore or lab. Avoid jostling or flipping tiles during recovery.

In a controlled environment, transfer tiles to microscope-compatible trays. Quantify settled larvae per tile using stereomicroscopy or image-based analysis. Score settlement success by treatment group and reef zone. Retain representative specimens for imaging or preservation.

Step 6: Data Logging and Reporting
Log all retrieval metadata, including time, location, box ID, environmental observations, and larval counts. Use spreadsheets or field data apps for digital entry. Conduct quality checks to flag inconsistencies or missing records.

7.5 Larval settlement in larval culture pools and on reef tables

7.5.1 Background and rationale

Optimising larval settlement to enhance recruitment is essential for ensuring the success of larger-scale coral larval restoration outcomes on reefs. Planktonic larvae tend to be dispersed in currents when freely released onto reefs which reduces the chances of settlement in target restoration sites, therefore settling larvae onto biologically conditioned settlement devices in controlled environments can significantly increase settlement success and settler production rates from mass larval cultures. Larval settlement onto tiles and other settlement devices within larval culture pools moored on the reef, or in larger-scale aquaculture tanks on board vessels (Doropoulos et al. 2019b), provides a very effective way of maximising settlement outcomes prior to release of settlers onto reefs for enhancing recruitment and recovery of depleted coral populations in degraded reefs (Harrison 2024b). Mass settlement of larvae onto limestone tiles and ceramic CAD devices within larval culture pools was successfully trialled during the Moving Corals project with settled spat on devices then deployed onto different reef areas to monitor survival and growth (Waters et al. 2025). In addition, we successfully tested mass settlement of ~1 million larvae onto large numbers of CADs and tiles on a table enclosed in a bespoke designed plankton mesh enclosure, which was deployed on sand in Lizard Island reef lagoon.

7.5.2 Procedure

Reef-based larval settlement involves initially deploying settlement tiles, CADs and other devices onto suitable reef areas for biological conditioning some weeks prior to their use in settlement trials. Deploying larval culture pools on reefs and collecting coral spawn are then required to produce millions of competent larvae for settlement (Phase 3). When larval cohorts contain sufficient numbers of competent larvae, settlement devices can be placed into larval culture pools or on tables enclosed in nets to optimise production of settled spat which can then be transferred onto reef areas for subsequent growth and recruitment.

7.5.3 Prerequisites

Successful larval settlement requires an understanding of coral larval settlement processes and biological conditioning processes required to induce large numbers of competent larvae to settle onto natural and artificial settlement surfaces. In addition, knowledge of how and when to collect coral spawn (Phase 2), and how to set up the larval pool systems to ensure that millions of healthy larvae are cultured successfully (Phase 3) for optimising settlement rates is required.

- **Existing documents**:

  Details of reef-based larval culture pool systems and mass culture of healthy competent larvae, plus larval settlement within culture pools (Harrison et al. Cruz 2021, Harrison and dela Cruz 2022, Harrison 2024b, Waters et al. 2025).

7.5.4 Risks and Hazards

Deployment of settlement tiles and devices on the reef requires scuba diving to set up the posts to attach the devices onto during the biological conditioning period (see Fig. 8.1.1 below). Therefore, standard safe research diving procedures including ADAS certification, commercial dive medical, First Aid and Oxygen therapy certification and regular certified inspections of diving gear are required. Practitioners also need to

be experienced and trained in the safe operations of inflating the larval culture pools and attaching the culture nets, and deploying the heavy anchor systems to temporarily moor the pools safely in lagoon or passage areas or near reefs. Diving or snorkeling operations are required to retrieve the conditioned settlement devices from the reef for transfer into larval pools, and subsequently for deploying settled spat on devices.

7.5.5 Equipment and Materials

Biological conditioning of settlement devices requires deploying large numbers of limestone tiles, CADs or other devices onto frames or racks that are set up on reef areas to enable microbial and CCA settlement for enhancing larval settlement. Larval production requires larval culture pool and net systems to be set up on sandy areas near reefs (Harrison 2024b). Equipment required includes replicate travertine limestone tiles cut into 5x5x1 cm sizes with a 6 mm hole drilled in the centre and a small hole drilled on the top left to attach a numbered tag (see Harrison 2024b). The ceramic CAD settlement devices were designed and provided by AIMS (see Randall et al. 2021). The tiles and CADs are threaded onto stainless steel rods interspersed with spacers and deployed on star pickets arranged in a square to optimise deployments (see Fig. 8.1.1 below).

Deploying settlement devices within the larval pools requires the devices to be attached onto frames or racks which are then submerged into the larval culture pools to encourage larval settlement. Monitoring of initial settlement rates requires the settlement devices to be carefully collected and transferred to monitoring stations on the vessel or in the laboratory so that the settlers can be counted under stereo microscopes and mapped and photographed before deploying the devices with settlers onto target reef areas.

7.5.6 Implementation steps

The following standard operations are used for routine deployment of settlement devices for biological conditioning on reefs, transferring conditioned devices into reef culture pools for larval settlement, and monitoring settlement rates before deploying the settlers on devices onto reef areas for further growth and development.

1. Set up tiles and devices onto metal rods with spacers between each device.
2. Deploy sets of tiles and devices onto frames or racks temporarily set up on rubble reef areas taking care not to damage corals and other sessile biota (see Fig. 8.1.1 below). Suitable reef areas for device conditioning include habitats with moderate current flow and high CCA cover to facilitate development of appropriate biota on the surfaces of the devices to encourage larval settlement. Devices are usually deployed for ~4-8 weeks but the conditioning periods can vary depending on the timing of reef trips and the rate of conditioning.
3. Retrieve the conditioned settlement devices just prior to deployments in larval pools and where possible monitor or photograph the devices to identify and map locations of wild settled spat prior to their use.
4. Attach the groups of conditioned devices onto racks and carefully submerge them into culture pools to encourage larval settlement (Fig. 7.5.1, Harrison 2024b).

Figure 7.5.1: Example of biologically conditioned tiles on metal rods attached to a settlement rack for deployment in larval culture pools (Photo: P. Harrison)

5. Settlement devices can be submerged for periods of 6-24 hours depending upon the densities of larvae in the culture pool and settlement rates required, then the devices are carefully retrieved and submerged in seawater for transfer to monitoring stations or for deployment on reef sites.
6. Settled spat are counted and mapped under dissecting microscopes to quantify initial larval settlement rates, and tile surfaces can be photographed using high resolution macro-photographs (see section 8.1 below).
7. Tiles and CADs with settled spat are then returned to reef sites for longer term deployments to monitor survival and growth over time.
8. At the end of the larval settlement period and after remaining larvae have been deployed, the net is removed, the pontoon system is deflated and removed, and the anchor system is carefully removed from the reef area. The net, pontoon and anchor systems must be thoroughly cleaned with high pressure hoses to clean algae and any other fouling organisms off the equipment. These components then need to be thoroughly dried before packing and storage ready for the next larval culture period.

The methodology for settling large numbers of competent coral larvae onto tiles and other settlement devices within larval pools is mature and is used by multiple organisations internationally and in different regions of the GBR (Harrison 2024b).

Larval settlement on devices on reef racks

In addition to the standard larval settlement methods within larval culture pools described above, the Moving Corals project enabled an innovative mass larval settlement method to be developed and tested using ~1 million larvae that were settled onto 1,000 CADs and 300 limestone tiles within a fine mesh enclosure attached to a reef rack (Fig. 7.5.2). This process was designed to increase the scale of pre-deployment larval settlement to provide options for future mass larval settlement at industrial scales on reefs.

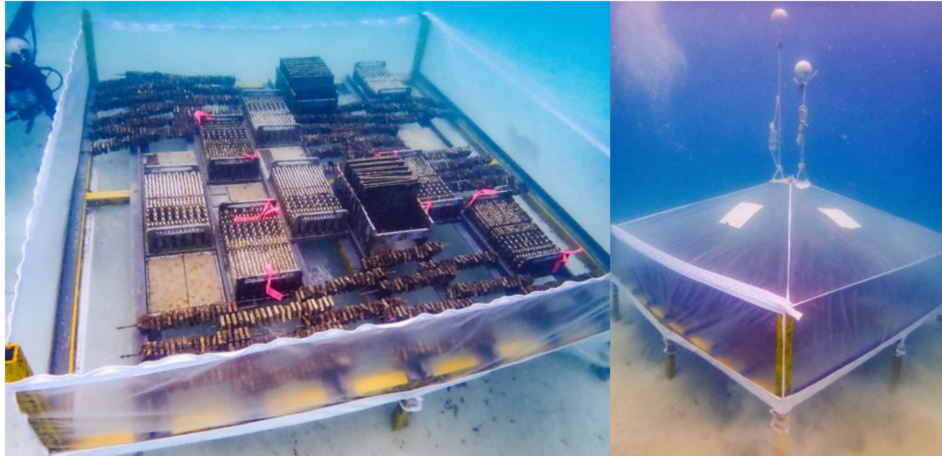


Figure 7.5.2: Large scale larval settlement trial on CADs and limestone tiles on a reef rack enclosed in a zippered plankton mesh tent enclosure designed for this project (Photo: D. McShane SCU Media)

The experimental methods used were as follows:

1. Thread groups of tiles onto metal rods with spacers between each tile, and insert CADs into racks with different packing densities.
2. Deploy sets of tiles and CADs on the reef table in the lagoon on sandy substrata (Fig. 7.5.2).
3. Condition the settlement devices for 6 weeks to establish biofilms and CCA on the exposed surfaces.
4. Once competent larvae are available from a larval pool, slowly move the larval pool over the reef rack ready for larval transfer.
5. Attach the base of the plankton mesh tent underneath the base of the reef rack then slowly pull up the net and close the zipper (Fig. 7.5.2). Attach buoys to the top of the tent enclosure to maintain the desired shape

9. Attach a transfer pipe from the base of the pool net and into the portal opening on the top on the tent enclosure.
10. Carefully stir the larvae within the culture pool and take replicate larval density samples to calculate the total number of larvae present prior to deployment.
11. Slowly draw up the larval culture net and allow the larvae to flow down through the base of the net into the pipe and into the larval settlement enclosure. Flush the remaining larvae from the transfer pipe system by slowly pouring seawater down the pipe.
12. Allow larvae to settle for 24 hours and then open the net enclosure to allow seawater to flow freely over the settlement devices to maintain healthy conditions.
13. Collect representative subsamples of the tiles and CADs and transfer them in seawater to monitoring stations to quantify larval settlement rates.
14. Count and map larval settlers under dissecting microscopes to quantify initial larval settlement rates, and settlement surfaces can be photographed using high resolution macro-photographs (see section 8.1 below).
15. Tiles and CADs with settled spat are then returned to the reef rack for longer term monitoring of survival and growth over time.
16. At the end of the larval culturing and settlement period, the nets are removed, the pool pontoon is deflated and removed, and the anchor system is carefully removed from the reef area. The nets, pontoon and anchor systems must be thoroughly cleaned with high pressure hoses to clean algae

and any other fouling organisms off the equipment. These components then need to be thoroughly dried before packing and storage ready for the next larval culture period.

The methodology for mass settlement of competent larvae onto settlement devices enclosed in a mesh tent is mature and can be scaled up for use by multiple organisations in a wide range of reef regions in future.

8 Phase 5 Monitoring larval settlement and recruitment

8.1 Monitoring larval settlement and post-settlement survival

Monitoring larval settlement rates after release and detailed monitoring of post-settlement survival and growth of settlers to recruitment are important aspects of the larval restoration process as the outcomes from different operations can be assessed and adapted to continually improve the approaches for larger-scale deployments in future. This section provides detailed methodologies for assessing coral larval settlement and post-settlement survivorship following larval delivery interventions. It covers a range of approaches from the deployment and retrieval of conditioned settlement tiles to advanced image-based monitoring workflows using high-resolution photography and photogrammetry. These methods allow practitioners to detect initial settlement signals, quantify early recruitment, track individual coral recruits through time, and assess survivorship and growth both in aquaria and in situ. The procedures were developed by the Moving Corals Team, drawing on published techniques from Gouezo et al. (2023, 2025b), Doropoulos et al. (2025), and others. While some components (e.g., tile deployment) are well-established, others (e.g., image annotation for recruit tracking) represent recent innovations with high reproducibility and automation potential.

8.1.1 Background and rationale

The success of coral larval restoration techniques relies on quantifying settlement rates and tracking post-settlement performance. This SOP integrates four core techniques:

1. **Deployment of pre-conditioned tiles** to measure larval settlement shortly after delivery.
2. **Photography monitoring** in aquaria to quantify settler abundance and survivorship over time.
3. **Photogrammetry workflows** to detect and monitor coral recruits in situ on reef substrate.
4. **Visual quadrat-based surveys** for monitoring juvenile coral populations on reef substrate

These methods can be applied sequentially or independently based on project scope. They are particularly useful within 2–14 days post-delivery for detecting early settlement, and from 2–12 months post-settlement for survival and growth monitoring.

- **System design**: Modular methods applicable to both lab and field settings.
- **Frequency**: following each larval release; regular monitoring at 3-6 month intervals and when possible
- **Duration**: 3–10 days during interventions, with long-term monitoring extending up to two years following intervention.
- **Maturity level**:
  - Settlement tile methods – *mature*
  - High-res imaging + annotation workflow – *demonstrated*
  - Photogrammetry + juvenile surveys – *validated*
- **Adoption level**: Multi-organisational and National (RRAP-wide implementation), with potential for international replication.

8.1.2 Procedure

The procedures consist of four components

1. Pre-conditioning and deployment of settlement tiles
2. Retrieval and microscopy-based processing
3. High-resolution photo documentation and batch editing using Python
4. Recruit annotation and tracking using CVAT and Python scripts
5. In situ reef-based photogrammetry and visual quadrat surveys

8.1.3 Prerequisites

The final monitoring phase builds on the outcomes from the first four phases of the larval restoration processes. This final phase is designed to assess the outcome of larval delivery interventions through standardized monitoring of early settlement and subsequent coral recruit survival and growth.

8.1.4 Risks and Hazards

Field Activities:

- **Physical hazards**: Risk of injury during diving, star picket deployment, or use of hammers – apply appropriate dive safety and PPE procedures.
- **Equipment loss**: Tagging and GPS tracking is essential to prevent loss of deployed baseplates or tiles.
- **Weather-dependent operations**: Field activities must consider sea state and tides.

Lab Activities:

- **Handling of settlers**: Improper handling of tiles during retrieval or photography can damage recruits.
- **Data integrity**: Risk of mislabelling or losing photo metadata; backups are essential.
- **Manual fatigue**: High annotation workloads may lead to reduced accuracy, rotate annotators and perform spot checks.

Control Measures:

- Follow dive safety protocols
- Use appropriate tools (hex drivers, tile jigs, camera sliders)
- Maintain clear data workflows with naming conventions and backups
- Always label tiles and datasets clearly during every step

8.1.5 Equipment and Materials

Procedure equipment

- Travertine tiles (5×5×1 cm) + stainless steel rods, bolts, nuts
- Custom baseplates and star pickets
- baskets for transport
- Tagging materials (bird bands, cable ties)
- Microscopes with powerful lights
- Underwater camera + housing (Sony Alpha R IV + LAOWA probe lens)
- Tripod and slider rails for photography
- Flow-through tanks
- Laptops with Anaconda, JupyterLab, Imaging Edge, or Metashape software
- Annotating equipment (PC/tablet, mouse, CVAT account)
- Dive lights ≥2000 lumens
- Quadrats and tagging supplies (cow tags, nails)

Personal protective equipment (PPE) and other safety equipment

- Dive-certified gear (wetsuits, gloves, knives, etc.)
- Waterproof notebooks, slate and pencils

- Sun protection (hat, sunscreen) for field teams
- Safety signage and first aid kit for lab and field settings

8.1.6 Implementation steps

Part 1 Assessing settlement signal following larval delivery using tiles

In order to understand whether the delivery of larvae onto the reef was successful, assessing the settlement signal can be done by deploying and monitoring settlement tiles placed on the reef. This method allows for quantifying larval settlement and early recruitment following intervention.

Step 1: Condition Tiles on the Reef Before Intervention

Timing

- Tiles must be conditioned on the reef **1–3 months prior** to the intervention.

Materials Needed

- Travertine tiles (5×5×1 cm) with a 6 mm hole in the centre
- M6 stainless steel rods
- Spacers (6 mm PVC hose, cut into 1–2 cm pieces)
- cable ties
- Star pickets

Instructions

- Assemble tiles and spacers onto the stainless-steel rods.
- Select a reef with:
    - Good water flow,
    - Easy access,
    - A rubble area to deploy the tile "kebabs" between star pickets.
    - Record the exact GPS location
- To minimise the footprint and maximise the deployment, star pickets can be arranged in a square grid

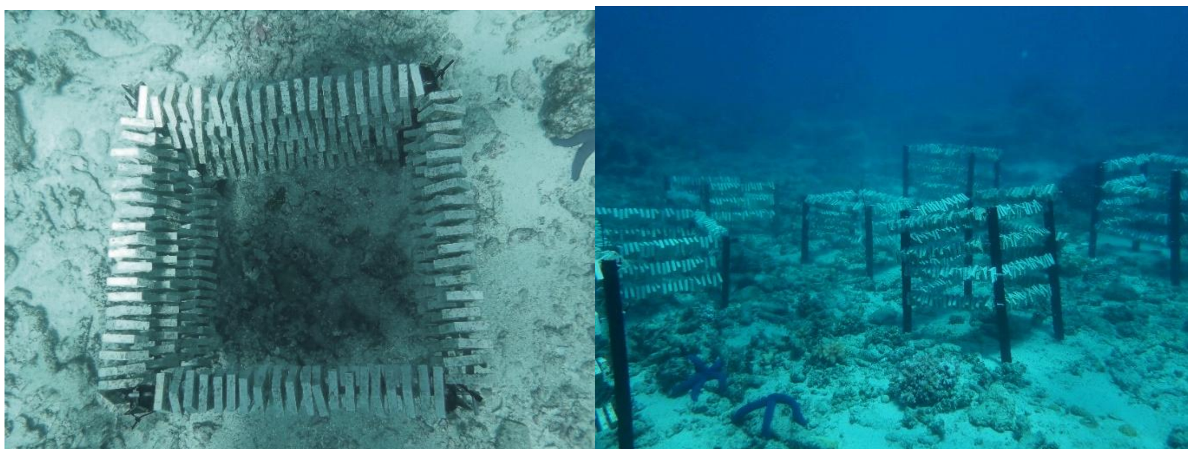


Figure 8.1.1: Settlement tiles assembled on rods with spacers, arranged between star pickets on rubble beds for conditioning

Step 2: Collect Conditioned Tiles and Assemble for Baseplate Deployment

Materials Needed

- Conditioned tiles
- M6 stainless steel (SS) 30 mm bolts
- Stainless steel washers
- Stainless steel nylon lock nuts
- Stainless steel bird bands with engraved tag numbers
- Socket wrenches

Instructions

- Assemble tiles onto the bolts in the following order:
  Bolt → Washer → Tile → Washer → Tag → Lock Nut
- Tighten securely using socket wrenches.

Figure 8.1.2: Settlement tile assembled on bolt with tag

Step 3: Deploy Baseplates on the Reef

Materials Needed

- Custom-made stainless steel 'mini baseplates'
- 2.8 mm galvanized nails
- Hammer

Instructions

- On the reef, locate areas with hard substrate.
- Place the baseplate on the substrate and nail it into place using the hammer and galvanized nails.
- Thread the bolt–tile assembly into the baseplate.

Figure 8.1.3: Settlement tile 'mini' baseplate deployment

Additional Notes

- Tiles can be deployed in spatial arrays (e.g., regular grids, circular arrays) to capture the spatial footprint of settlement, following methods such as Doropoulos et al. (2025).
- An underwater map must be created by recording the tag number and location of each tile.

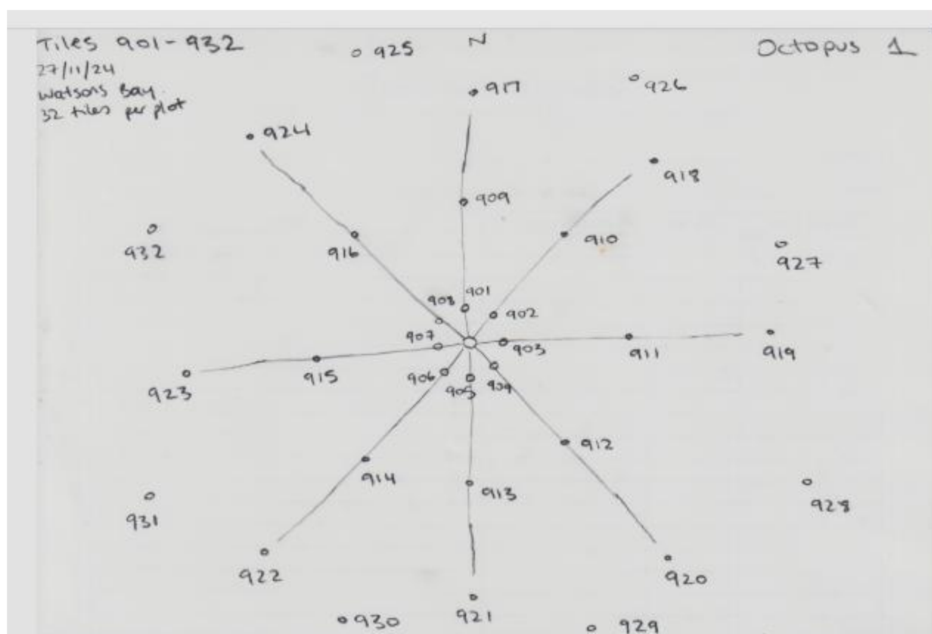


Figure 8.1.4: Example of an underwater map layout

Step 4: Retrieve Tiles 2–3 Days After Larvae Delivery

Instructions

- Retrieve tiles carefully:
  - Use a hex screwdriver when possible.
  - Alternatively, hold tiles gently by the corner to minimize handling effects on settlers.

Figure 8.1.5: Securing tile onto the bolt tile assembly

Transport Instructions

- Secure each tile into threaded inserts in transport baskets (one basket holds ~15 tiles).
- Stack 4–5 baskets inside each other and place them in a nally bin filled with seawater.
- In the aquarium:
  - Place baskets in a flow-through system at a tilted angle (use bricks) to prevent sediment accumulation on tile surfaces.

Notes

- Tiles can also be placed loosely in baskets; however, this may increase handling damage during transport.

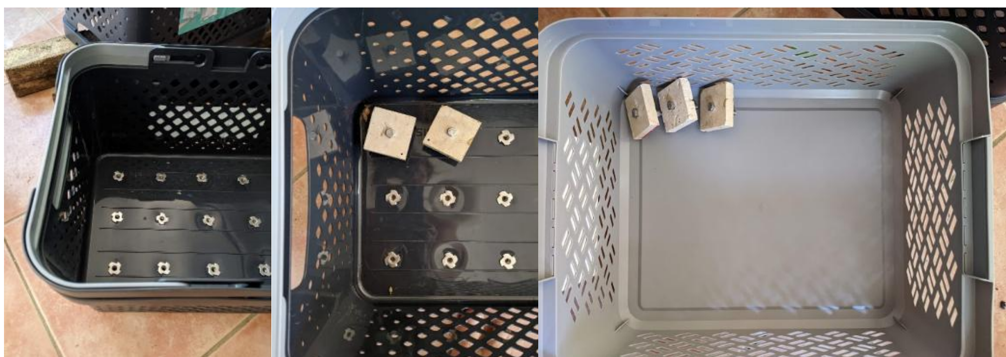


Figure 8.1.6: Baskets set up to retrieve settlement tiles carefully after interventions

Step 5: Process Settlement Tiles Under the Dissecting Microscope

Materials Needed

- Stereomicroscope
- Strong microscope light
- Deep Tupperware
- Custom aluminium tile jig (to hold tiles at the same plane during microscope inspection)
- Waterproof datasheets
- Pencils
- Clickers

Instructions

- Retrieve a tile from the flow-through system and record:
    - Tag number and any other relevant information onto the waterproof datasheet.
- Place the tile onto the jig under the microscope.

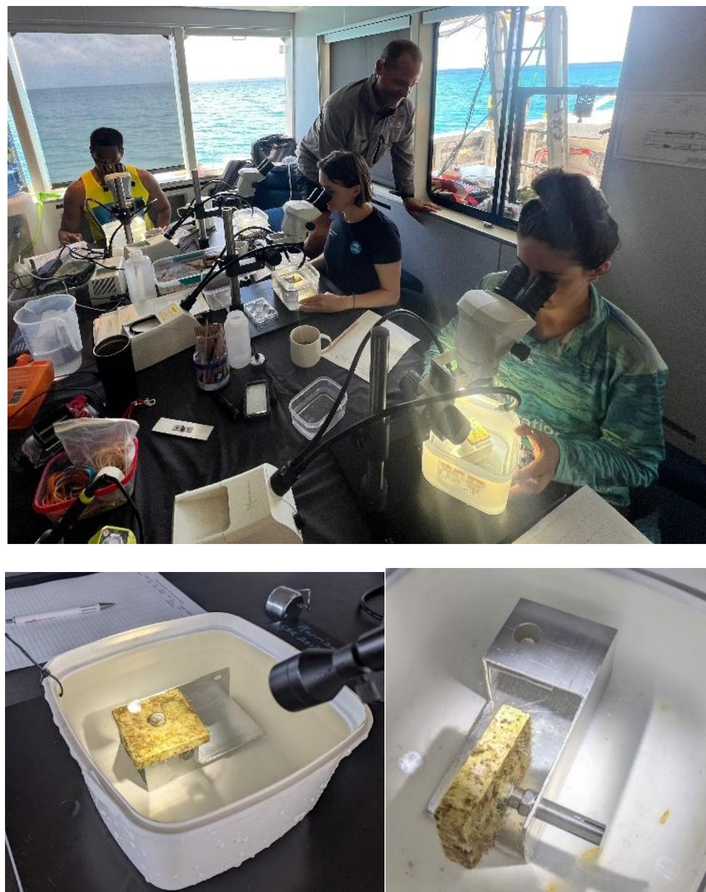


Figure 8.1.7: Microscope set up with custom 'jig' to inspect all faces of tiles

- Adjust the microscope focus and inspect the tile surface carefully:
    - Use a "mow the lawn" pattern to ensure thorough inspection of:
        - Top,
        - Bottom
        - Sides
- Record the total count of settlers per tile face on the waterproof datasheet. (Example datasheet provided below)

Project Name: RRAP MovingCorals Experiment Name: SlowRelease_Mermaid

Site location (GPS coordinates): -14.6667°, 145.4667°

Date Tile collection : 3/12/2024 Date Tile processing: 3/12/2024

Observer	PlotID	DeliveryMethod	TileTagID	TileFace	SettlersCount	Comments
Marine	25	Octopus	102	Top	2	
Marine	25	Octopus	102	Bottom	28	Lots of sediments trapped in turf
Marine	25	Octopus	102	Sides	8	
Marine	25	Octopus	123	Top	0	
Marine	25	Octopus	123	Bottom	5	5 soft corals
Marine	25	Octopus	123	Sides	2	
…..						

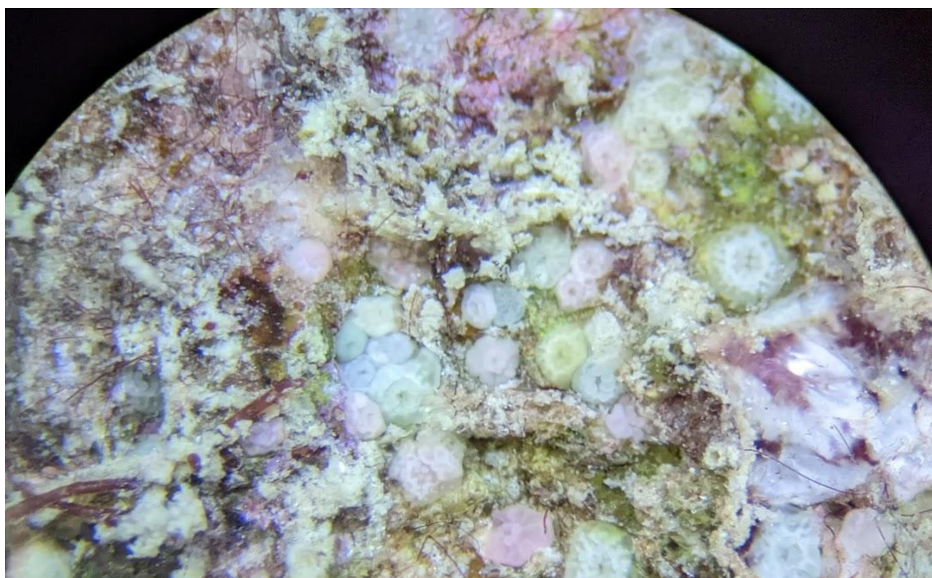


Figure 8.1.8: Diverse assemblage of 2-day old coral settlers on a tile following intervention

Step 6 (Optional): Photograph and Redeploy Tiles to Monitor Survivorship and growth

Instructions

- For tiles with more than one settler, photograph each tile to document settler size, position, and number (see instructions in the next section).
- Redeploy photographed tiles back onto the reef to monitor post-settlement survival and growth over time by securing them to baseplate
- Retrieve unused baseplates from the reef.

Part 2 Monitoring post-settlement survival and growth of coral recruits on tiles using high-resolutions photographs

Goal
Photograph tiles in a flow-through tank using the **LAOWA probe lens** and analyse the images for settler assessment.

Step 1. Set up the camera and tripod

Equipment Required

- 2 Tripods
- Camera slider with bracket
- Sony Alpha R IV camera
- LAOWA 24mm f/14 2X Macro Probe lens
- Power bank + USB-C cable (to power LED light on the probe lens)
- PC tablet (optional, for remote connection and better display)
- Square aluminum bar with 6mm holes to hold tiles
- Slate, pencils, waterproof datasheet

Set-Up Instructions

- Assemble the camera on the slider between the two tripods over a **flow-through aquarium** or a **nally bin** filled with seawater.
- Secure **8–10 tiles** in a row on the **aluminum bar** by inserting bolts into the threaded inserts.
- Record the corresponding **TagID** for each tile in order on an **underwater datasheet** placed next to the tiles for easy reference.

Figure 8.1.9: Photography set up to take high resolution photographs of tiles

Step 2: Take Photos and Record Metadata

Camera and Software Set-Up

- Connect the Sony camera to the PC tablet using a USB cable.
- On the camera:
    - Go to MENU → Network → Transfer/Remote → PC Remote Function → PC Remote → ON
    - Set PC Remote Connect Method to USB.
- Open Imaging Edge 'Remote' software on the tablet.
- In the Remote software, adjust the following settings:
    - Manual Mode

- Aperture: manually set directly on the lens to ~f/30 (tape the ring to avoid movement)
- Shutter Speed (SS): 1/60
- ISO: 800 or Auto
- Drive Mode: 2-second self-timer
    - If not available in the Remote software, set via Camera Menu → Drive Mode → Single Shooting + 2-sec Self-Timer.

Photography Instructions

- Switch on the LED ring light of the probe lens (powered by the external power bank).
- Center the tile in the middle of the frame.
- Adjust the focus manually on the lens OR remotely (using a motorized focusing device if available).
- Use X5.9 magnification to zoom into the image, adjust focus carefully, then take the photo.
- Important:
    - Do not touch the setup while shooting.
    - Even slight motion will cause blur.
- Move the camera along the rail to the next tile.
- Check and adjust focus before each photo.

After Photographing One Row (Top Side)

- Flip all tiles to photograph the underside face.
- Repeat the same photography procedure for the underside, maintaining tile order.
- Record photo filename next to the Tile ID and Tile Face on the datasheet.

Efficiency Tip

- A second person can prepare the next batch of tiles while photographing is ongoing.

Metadata Entry

- After imaging, enter the photo filename, tile tag number, and face (top or bottom) into an Excel spreadsheet as shown below:

date	location	RAWfileID_1	RAWfileID	orientatio	isTag	tagId
2022-12-16	Watsons_	1612_00072	1612_K149_T	T	0	149
2022-12-16	Watsons_	1612_00073	1612_K136_T	T	0	136
2022-12-16	Watsons_	1612_00074	1612_K115_T	T	0	115
2022-12-16	Watsons_	1612_00075	1612_K076_T	T	0	76
2022-12-16	Watsons_	1612_00076	1612_K132_T	T	0	132
2022-12-16	Watsons_	1612_00077	1612_K149_B	B	0	149
2022-12-16	Watsons_	1612_00078	1612_K136_B	B	0	136
2022-12-16	Watsons_	1612_00079	1612_K115_B	B	0	115
2022-12-16	Watsons_	1612_00080	1612_K076_B	B	0	76
2022-12-16	Watsons_	1612_00081	1612_K132_B	B	0	132

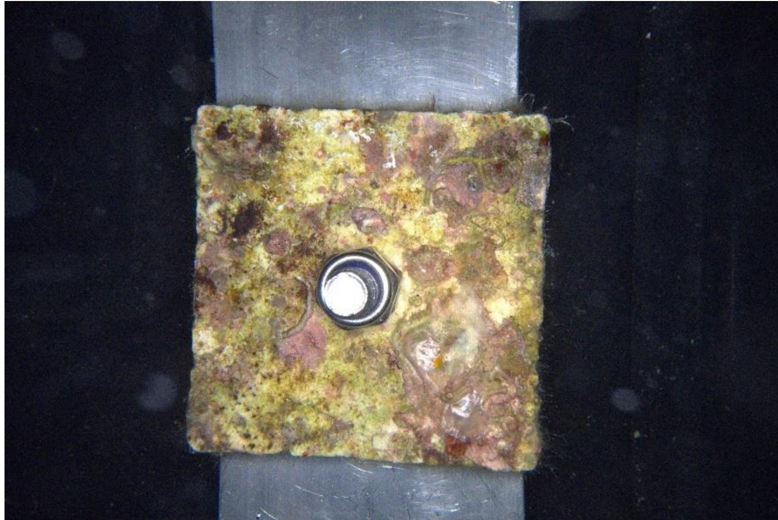


Figure 8.1.10: Example of a good photo (RAW format, before batch editing step below)

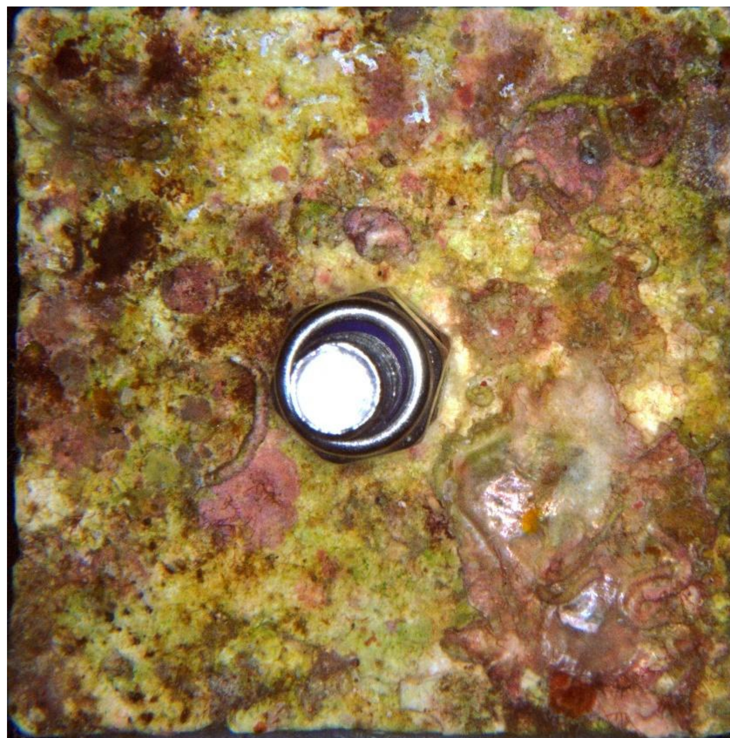


Figure 8.1.11: Example of the same photo after the batch editing process (see Step 3 below)

Step 3: Batch Edit Photos Using Python Workflow

This workflow was developed by George Roff, Marine Gouezo, Magda Guglielmo, and Brint Gardner (Gouezo et al. 2025b)

1. Install Python and Set Up

Install Anaconda

- Follow the prompts to install Anaconda from: https://docs.anaconda.com/free/anaconda/install/windows/

Download Code

- Copy the folder **coralsettlement-errfp_CODES_MC** into your Documents folder. (*Recommended: Do not change the location of this folder!*)

Create a New Environment

1. Open **Anaconda Navigator**.
2. Go to the **Environments** tab.
3. At the bottom left, click the **Create** icon.
4. Name your environment (e.g., imaging).

Activate Your Environment

1. Open **Anaconda Prompt** (search for it on your computer).
2. Type:
3. conda activate imaging

You should see (imaging) at the start of your command line instead of (base).

Install Required Packages

1. Change directory (cd) to your code folder. Example:
2. cd C:\Users\mgouezo\OneDrive - Southern Cross University\Documents\Python Scripts\coralsettlement-errfp_CODES_MC
3. Install the required packages:
4. pip install -r requirement.txt
   - **Note:** If this gives an error, first install pip:
   - conda install pip

Then try again.

5. You can check all installed packages in Anaconda Navigator under your environment (imaging).

Install JupyterLab

- In Anaconda Navigator:
  - Switch to the **Home** tab.
  - Make sure the **imaging** environment is selected.
  - Install and **Launch JupyterLab**.
    It will open in your web browser.

2. Workflow to Batch Edit Photos

Prepare Your Photos

- Organize your photos into folders (preferably on a hard drive, not the cloud).
- Example folder structure:
- E:\Ret_Exp_2022_TilesPhotos\DEC_2022\1p1
- Each folder should have:
  - **Raw photos**
  - A **.csv file** containing:
    - Sampling Date (yyyy-mm-dd format)
    - Location
    - Raw File ID

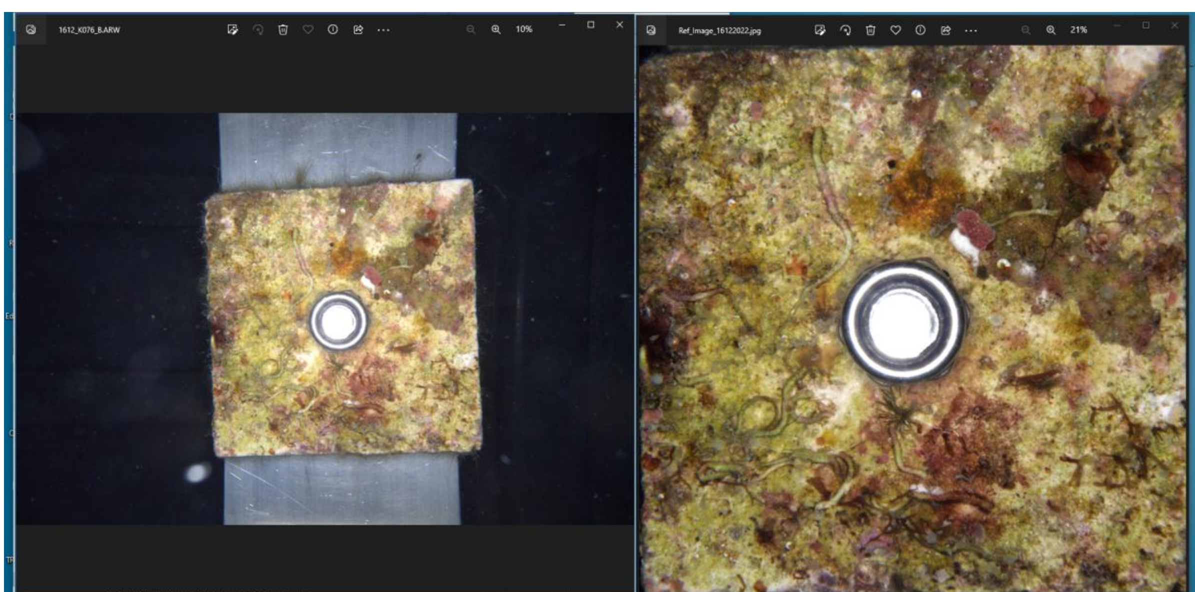
- Tile Orientation (T for top or B for bottom)
- Tile Tag ID
- Column isTag with all values = 0

IMPORTANT:
Make sure all dates in the CSV are formatted as yyyy-mm-dd.
(Format cells → Custom → enter yyyy-mm-dd)

	A	B	C	D	E	F	G
1	date	location	RAWfileID_1	RAWfileID	orientatio	isTag	tagId
2	2022-12-16	Watsons_	1612_00072	1612_K149_T	T	0	149
3	2022-12-16	Watsons_	1612_00073	1612_K136_T	T	0	136
4	2022-12-16	Watsons_	1612_00074	1612_K115_T	T	0	115
5	2022-12-16	Watsons_	1612_00075	1612_K076_T	T	0	76
6	2022-12-16	Watsons_	1612_00076	1612_K132_T	T	0	132
7	2022-12-16	Watsons_	1612_00077	1612_K149_B	B	0	149
8	2022-12-16	Watsons_	1612_00078	1612_K136_B	B	0	136
9	2022-12-16	Watsons_	1612_00079	1612_K115_B	B	0	115
10	2022-12-16	Watsons_	1612_00080	1612_K076_B	B	0	76
11	2022-12-16	Watsons_	1612_00081	1612_K132_B	B	0	132

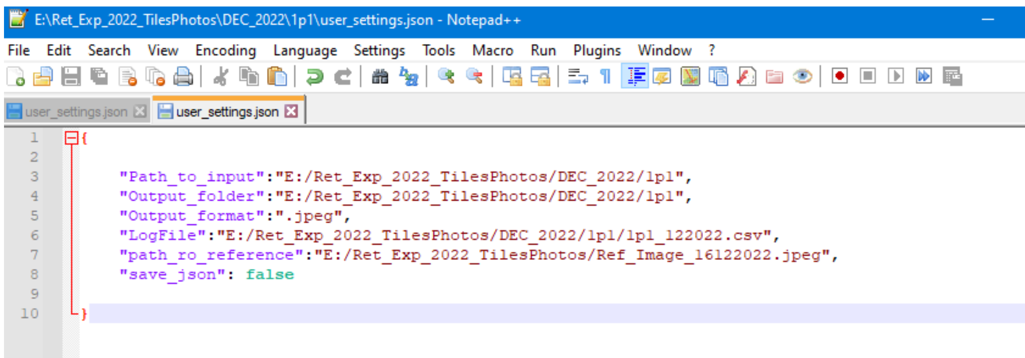
Create Your Reference Photo

- Choose one photo from the folder.
- Manually crop and edit it (use any software like *Affinity Photo* or *Photoshop*).
- Save the edited reference photo as:
- Ref_Image_16122022.jpeg
- Save it in the same input folder as your raw photos.
- Keep the resolution and exposure consistent for all photos in the time series



3. Edit User Settings

1. In JupyterLab, open the folder coralsettlement-errfp_CODES_MC.
2. Edit the file user_settings.json:
    - Update the paths:
        - Input folder (where raw photos are located)
        - CSV file
        - Reference image



Run the Image Editing Script

1. In JupyterLab, open:
2. Convert_Crop_Edit_Label_Images.ipynb
3. Follow the instructions inside the notebook.

Interactive steps for each photo:

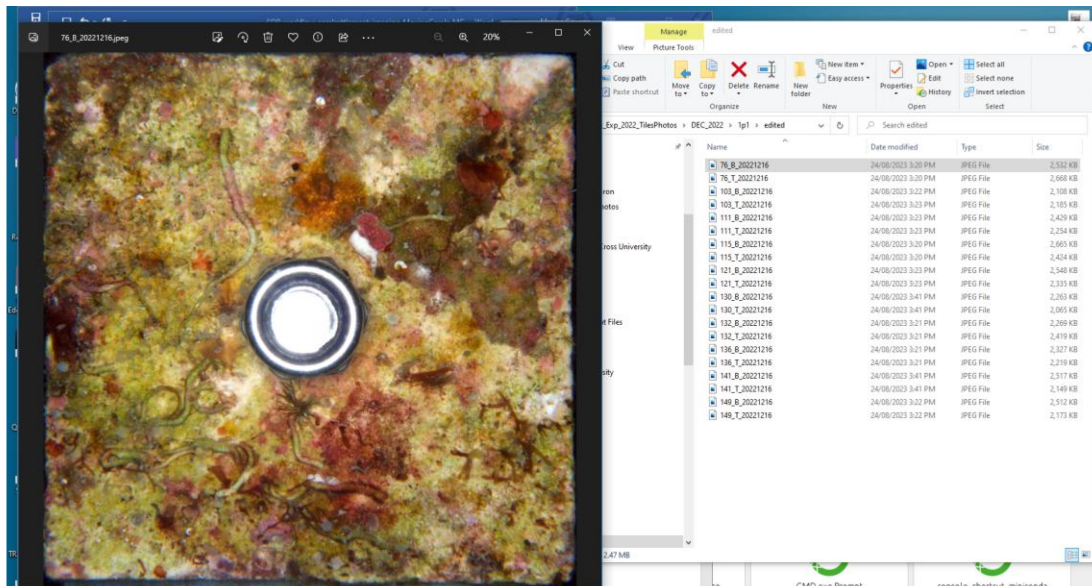
- Left click to select 4 corners (important: 4 points required!).

- Double right click to delete points.
- Press ESC when done.

- Press ESC without selecting points to skip that image.

Images will be:

- Cropped
- Aligned as a square
- Edited to match the reference image
- Relabelled based on the metadata in the .CSV file
- Saved as high-quality .jpeg files named:
- tagID_orientation_YYYYMMDD.jpeg



Additional Notes

- If you need to stop:
  Press CTRL-C in the terminal — the script will resume from the last processed image when you rerun.
- Once finished, your processed photos will be in a new folder (output folder created automatically).
- Once all time points are processed, group tile replicates belonging to the same treatment into a common folder if needed

Step 4: Annotate recruits on photos of the same tile over two time points

This workflow involves annotating coral recruits on tile images, exporting the annotation files, and processing these annotations to classify recruits as **alive**, **dead**, **chimera**, or **new natural recruits** based on their position within the tile grid over time.

1. **Annotate Images Using CVAT**

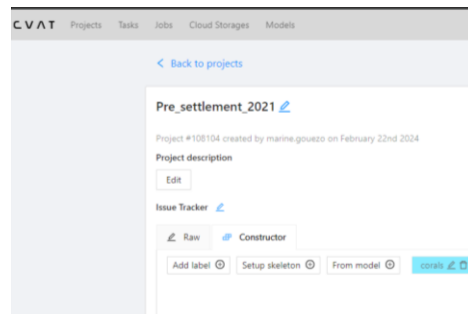
Set Up a CVAT Account

- Create your free CVAT account here:
  https://www.cvat.ai/

Create a New Project

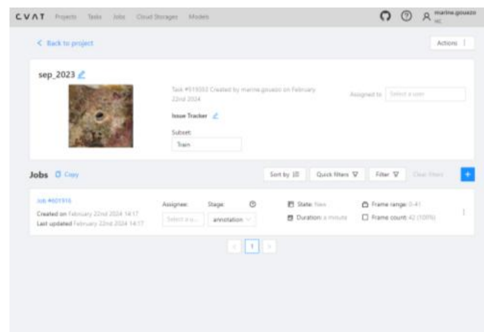
- Start a new project (e.g., Pre_Settlement_2021).
- Add Labels:
  - Create a label called Corals (cyan color, Polygon type).

- Click Continue and Submit.



Create a Task

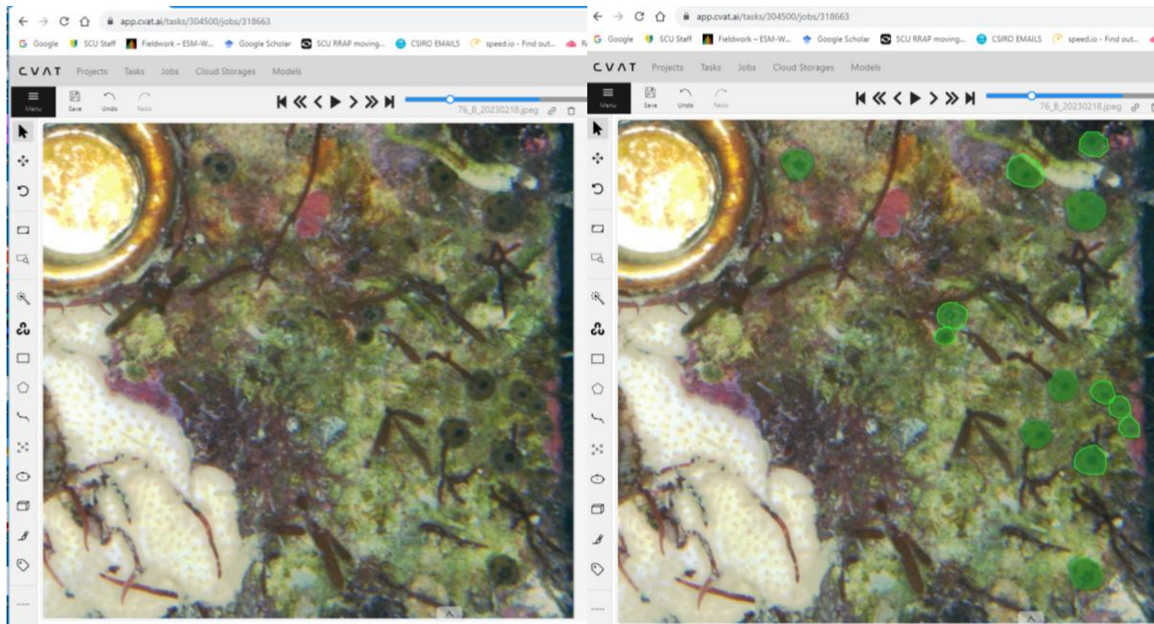
- Inside the project:
  - Click + to create a New Task (e.g., Sep_2023).
  - Subset: Choose "Train".
  - Upload all photos related to the treatment and time period.
  - In Advanced settings, increase image quality to 100%.
  - Click Submit and Open



Start Annotating

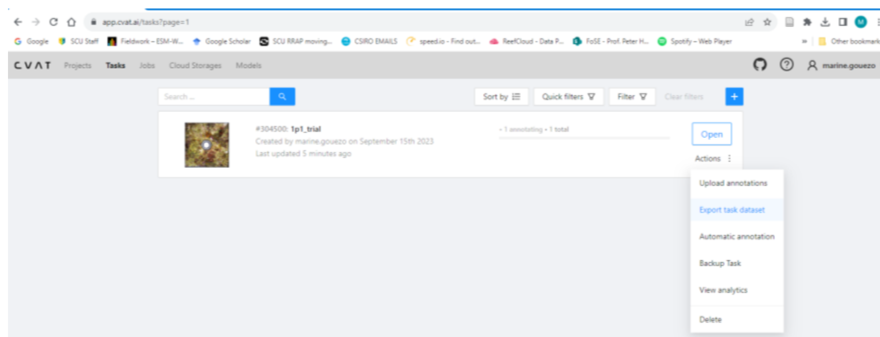
- Open your task under the Tasks tab.
- Find the Job Section → Click the Job Number ID in blue.
- Annotate Images:
  - Select the Polygon Tool on the left.
  - Select the correct label (e.g., Corals).
  - Select Shape (NOT Track).
  - Draw around each recruit.
    (Tip: hold Shift for smoother polygons.)
  - After finishing a shape, press 'N' to save it.
- Save frequently
- You can rotate images if needed before annotating.

See example below, before and after annonating



2. Export Annotations from CVAT

- Go back to the Tasks tab.
- Export task datasets:
  - Save only the annotation file (.xml).
  - Do not save images.
- Save the exported annotations.xml file inside the folder where your images are located.



3. Process Annotations to Classify Coral Recruits

Overview: What the Code Does

- For each polygon in an image:
  - Calculates contour area, perimeter length, and centroid (all in pixels).
- Tracks polygons through two time points, If two or more recruits merge → classified as a **Chimera** and provide details on which contour ID from previous time points are.

Important Notes

- The code requires at least one annotation per tile (cannot handle empty tiles).
- The code processes only two time points at a time:
  - Run separately for T1–T2, T2–T3, T3–T4, etc.

Running the Processing Script

Open Python Environment

- Open Anaconda Navigator.
- Activate the environment: imaging.
- Launch JupyterLab.

Navigate to Scripts

- Navigate to the folder:

coralsettlement-errfp_CODES_MC/Chimera

- Open a new Python notebook.
- Copy and paste the following into a cell, adjusting paths to your files:

```
# Define paths

input_path = r'D:\CVAT\Tiles_032024'

output_path = r'D:\CVAT\Tiles_032024\Output'

annotation_file = r'D:\CVAT\Tiles_032024\annotations.xml'

# Run the processing script

%run Process_Annotations.py --input_path {input_path} --output_path
{output_path} --annotation_file {annotation_file}
```

Run the Script

- Press the Play button to execute.

Outputs

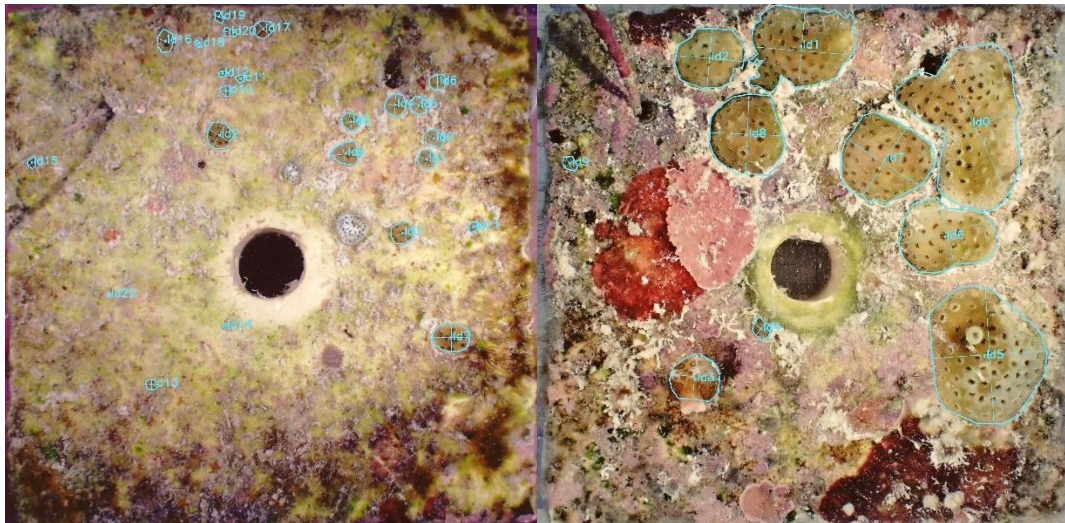
See the provided folder called 'Example' within the 'Chimera' folder for sample outputs and structure.

- .csv files containing:
  - Tile ID
  - Contour ID
  - Contour perimeter
  - X and Y centroid coordinates (in pixels)
  - 'Chimera' column indicating if a previous-time-point recruit merged with another.

- **3 output images** for visual validation.

One named as the tile sample that shows the overlap of polygons through time:

One image for each time points labelled as TileID_Face_MMYYYY_labelled



Part 3 Monitoring post-settlement survival and growth of coral recruits on the reef using macrophotogrammetry methods

Following interventions, post-settlement survival and growth can be monitored directly on the reef substrate starting 2–3 months after larval delivery using macrophotogrammetry techniques.

Requirements

- High-resolution camera with underwater housing
- Powerful dive lights >= 2000 lumens and quadrat
- Computer with strong processing capabilities dedicated to processing models
  (or access to a High Performance Computing (HPC) facility through your institution or collaborators)

- License for Agisoft Metashape photogrammetry software

Workflow Reference

- The full workflow, including all field and processing steps, is detailed in the open-access study: Gouezo et al. (2023): https://doi.org/10.1111/2041-210X.14175

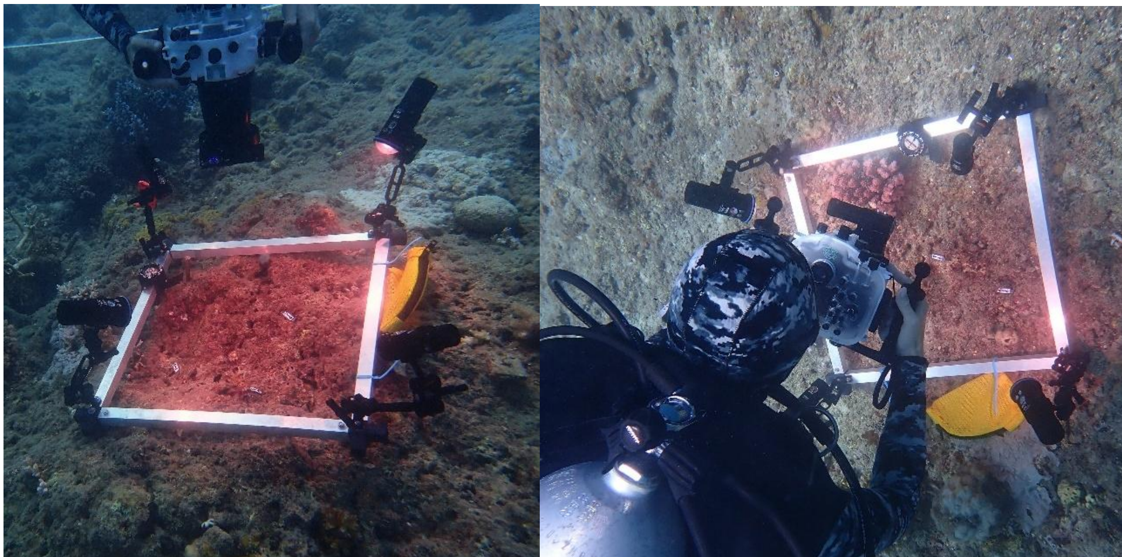


Figure 8.1.12 Diver scanning the small ~0.2 m² by taking macrophotographs (~230-300) at different angles around the scenes

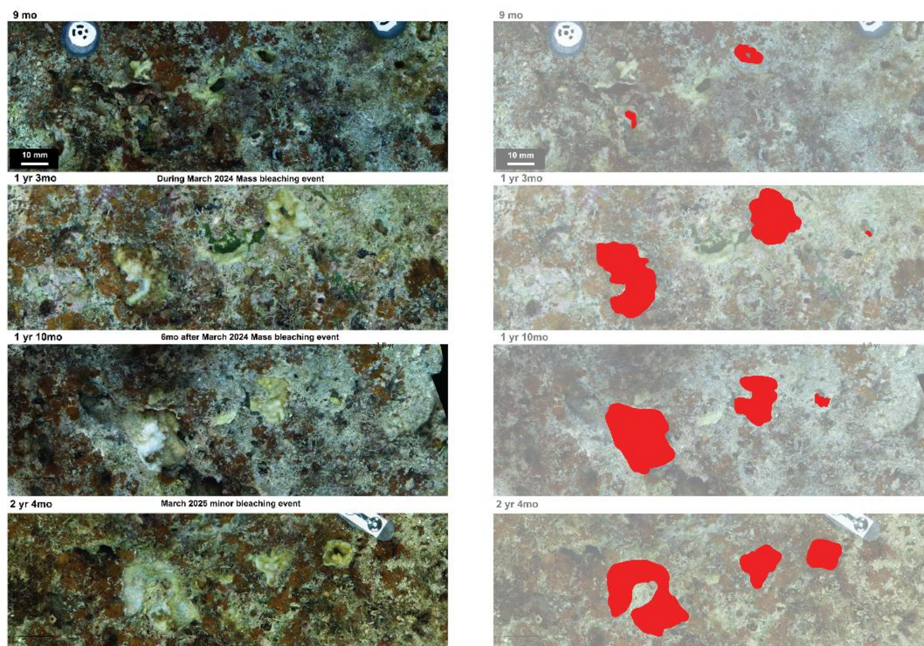


Figure 8.1.13 Example of time series of the same area of the reef through time showing 3 Acropora recruits growing and shrinking

Part 4 Monitoring juvenile corals populations on the reef using quadrat-based visual surveys

Juvenile corals can also be surveyed visually on the reef using common methods (e.g. Roth and Knowlton 2009). The workflow below describes the approach used by both the **EcoRRAP** and **Moving Corals** subprograms within RRAP.

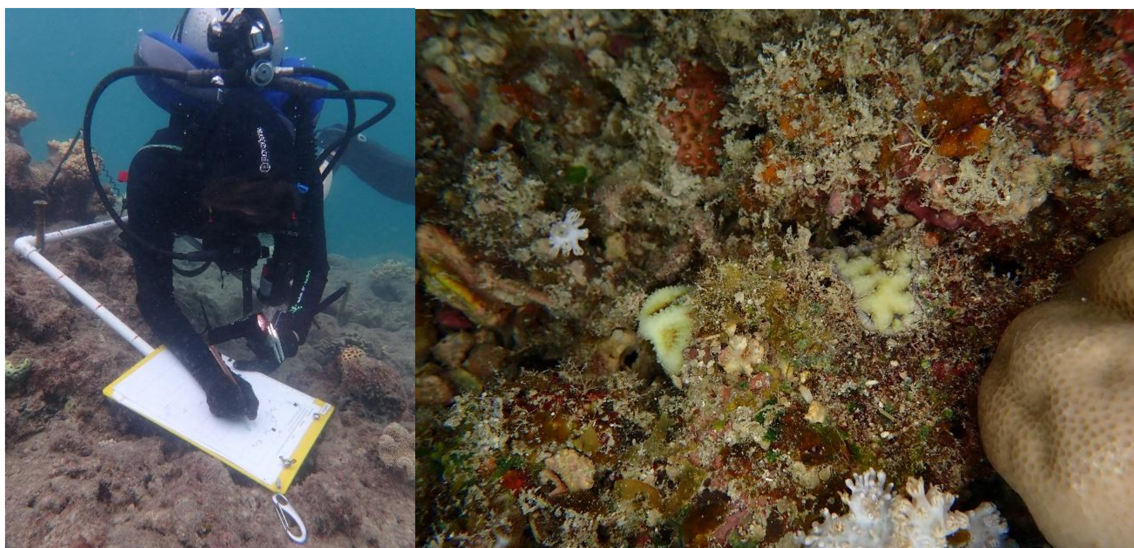


Figure 8.1.14 Diver surveying juvenile coral community within a quadrat and close up photographs with coral recruits on the reef

Step 1: Permanently Mark an Area for Monitoring

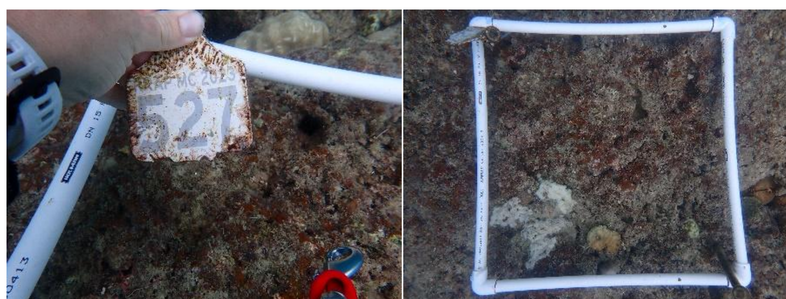
Instructions

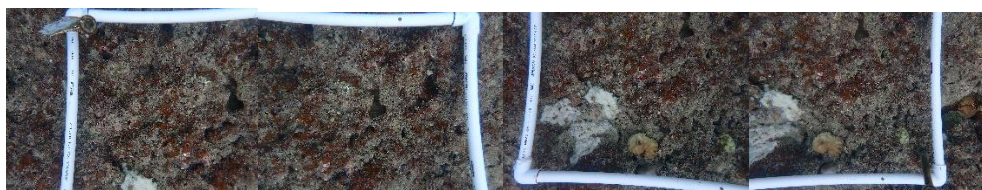
- Select reef surfaces that are:
    - Free of fouling organisms and sediment.
    - Rich in microhabitat spaces (e.g., crevices, small reef ledges).
    - Avoid flat area is turbid environment as they will accumulate sediment
- Place a 0.5 m × 0.5 m quadrat at the desired location.
- Nail a deck nail at each corner of the quadrat.
- Tag the quadrat with a laser-printed cow tag and cable tie it to the nail.

Step 2: Photograph the Quadrat

Instructions

- Take:
    - One photo to the cow tag
    - One photo of the entire quadrat, with the cow tag showing in the upper left corner
    - One photo of each quarter of the quadrat for more detailed records.

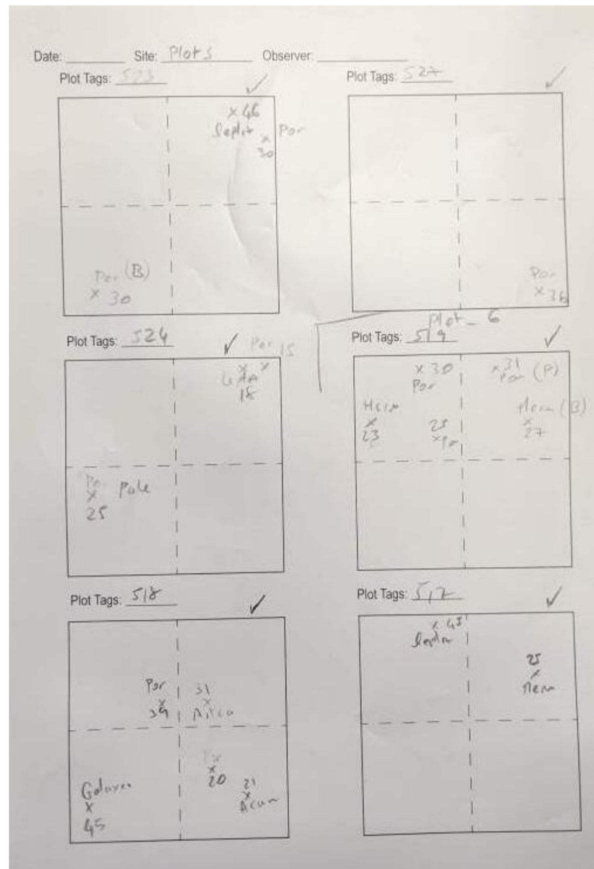




Step 3: Record Juvenile Coral Data (Baseline Survey)

Instructions

- Using a printed blank quadrat diagram (similar to below), record the location, maximum diameter, and taxon of all visible juvenile corals (<40 mm in diameter).



- Survey method:
    - Always start with the corner where the tag is located.
    - Use a mow-the-lawn search pattern to systematically cover the quadrat.
- Tools:
    - Dive torch (especially for deeper or overcast conditions).
    - Use a Caliper for accurate coral measurements.

- If the taxon is unknown:
    - Take a photo of the datasheet drawing and a close-up photo of the recruit for later identification.
- **Backup**:
    - Photograph all datasheets at the end of each dive day.

Subsequent Surveys

- For later time points:
    - Print the annotated quadrat diagram from the previous survey.
    - This saves time when updating the dataset through time.

(See the next step for how to transpose juvenile coral data onto 2D images.)

Step 4: Transpose Juvenile Coral Data onto 2D Images Using Photoshop

Stitch Photos Together

- Open all quadrat photos in Photoshop.
- Go to File → Automate → Photomerge.
- Select Add Open Files (ensure Blend Images Together is ticked) → OK.
- Use the Crop tool (C) to crop the final stitched image to the quadrat area.
- Save the new stitched image.

Prepare for Annotation

- Open the stitched quadrat image in Photoshop.
- Unlock the background layer (double-click or drag the padlock to the bin).
- Crop further if needed.
- Adjust brightness, exposure, and contrast as needed.

Add a Reference Mark

- Draw a small black square in the corner where the tag is located.

Annotate Juveniles

- Create a new layer (Ctrl + Shift + N) and name it "text".
- Zoom in (Alt + Scroll) for better precision.
- Use the Magnetic Lasso Tool (L) to outline each juvenile coral.
- For each recruit:
  - Edit → Stroke → set:
    - Location: Outside
    - Width: 8 pixels
  - Add a label using the Text Tool (T):
    - Font size ~36–48 pt (adjust as needed).
- **Special case**:
  - If a juvenile on the hand-drawn map is not visible on the image:
    - Outline its location with the Lasso tool.
    - Stroke it as normal.
    - Add a “*” next to its label.

Create Coordinate Grid

- Stay on the "text" layer.
- Use the Line Tool (U) to divide the quadrat into 4 × 4 smaller squares (16 cells).
- Create a new layer (Ctrl + Shift + N) named "coordinates".
- Add text labels to the grid:

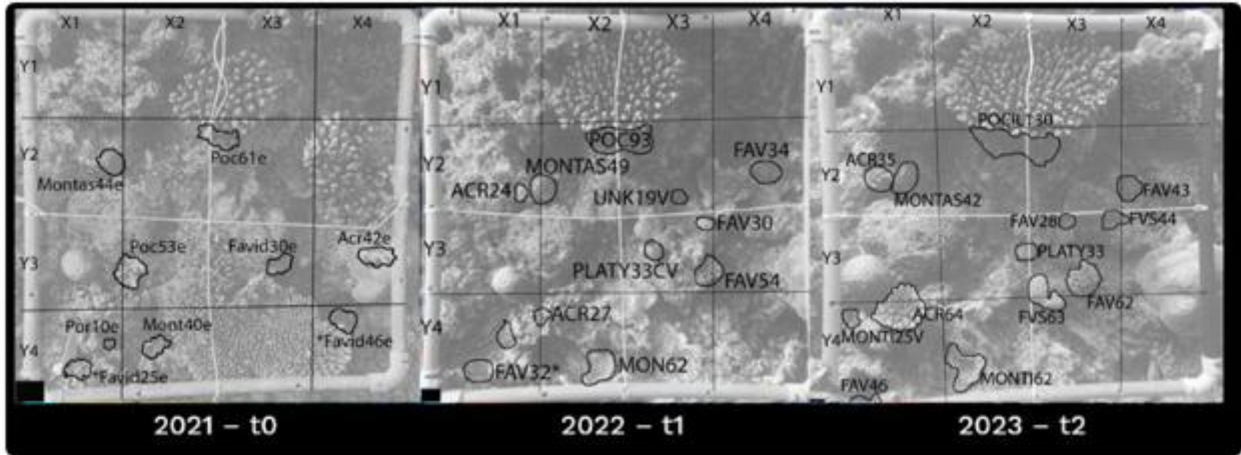
X1Y1	X2Y1	X3Y1	X4Y1
X1Y2	X2Y2	X3Y2	X4Y2
X1Y3	X2Y3	X3Y3	X4Y3
X1Y4	X2Y4	X3Y4	X4Y4

Record Juvenile Coral Coordinates

- Record each recruit's position (e.g., X2Y3) in a Excel spreadsheet using the coordinate system (see example below).

Create Final Versions

- Duplicate Layer 0 and rename it "Layer 0 B&W".
- Convert to Black & White:
  - Go to Image → Adjustments → Black and White.
  - Set opacity to 60%.
- Save the Photoshop file as .psd (e.g., Quadrat01_20230922.psd).
- Export final annotated images as .jpeg if needed
  (turn off "Layer 0" before exporting if necessary).



- Enter the data into Excel similar to as below:

QuadratID	xy	CoralID_T0	CoralID_T1	Size_20231207	Status_20231207	Size_202403	Status_20240315
557	X1Y1	Acropora	NA	22	alive	NA	dead
557	X1Y1	Favites	NA	27	alive	NA	dead
557	X2Y2	Dipsastreae	NA	41	alive	NA	dead
557	X2Y2	Unknown	Merulinidae	15	alive	15	alive
557	X4Y2	Diploastrea	NA	82	alive	NA	dead
557	X4Y2	Acropora	NA	32	alive	NA	dead
557	X4Y2	Acropora	Acropora	14	alive	45	alive
557	X4Y2	Acropora	NA	30	alive	NA	dead
557	X4Y4	Montipora	NA	25	alive	NA	dead
557	X4Y4	Porites	Porites	47	alive	45	alive
557	X4Y4	Acropora	NA	46	alive	NA	dead

Notes:

- When a new recruit appears during a later survey:

Categorize it as alive_new in the Status column of your metadata spreadsheet.

- Coral identification (CoralID) can change through time:

As corals grow, their features become more apparent, allowing for more precise taxonomic identification.

Differences may also occur due to observer bias between surveys.

- Important:Always record CoralID separately for each time point:

Use CoralID_T0 for the initial survey.

Use CoralID_T1 for the later survey.

This method ensures that changes in identification are properly tracked and can be reviewed later without confusion.

9 Conclusions

The RRAP Moving Corals project has been highly successful in developing, testing and applying a range of innovative methods to enhance larval restoration outcomes in a range of reef locations, as well as adapting and testing some previously developed methods for capturing coral spawn slicks for reef-based mass larval rearing in culture pools and tanks. The Moving Corals innovations include developing high resolution hydrodynamic modelling and particle tracking to (a) predict spawn slick aggregations for optimising collections of coral spawn in complex reef environments, and applying these models to (b) predict optimum locations and times for larval releases to minimise losses through larval dispersal in currents. We have also developed standardised drone operations and helicopter surveys to search for and locate spawn slicks, industrial scale spawn sucker collection methods, and novel larval staining methods for tracking larval deployments after releases. Additional significant innovations include the development of a new larval seedbox method for unprecedented scaling of larval restoration through delayed free-release methods that dramatically expand the spatial impact of larval based restoration without the need for physical containment, micro-imaging to automate workflows for monitoring settlement and survival on tiles, and a new macrophotogrammetry technique enabling detection of new coral settlers directly on reefs for the first time.

Together, these methods significantly enhance larger-scale reef-based larval production and settlement outcomes on reefs, which will enable coral restoration outcomes to be applied at much larger scales in future reef restoration projects. The reef-based coral larval restoration method of collection, culture, and deployment from wild coral spawn slicks is now mature, highly cost-effective and scalable, and is adaptable for use in a wide range of reef environments. Several of these approaches are being operationalised by a broad range of stakeholders—including restoration practitioners, reef managers, Traditional Owner groups, reef-based tourism operators, and environmental organisations—across the Great Barrier Reef and internationally, to support the strategic scaling of larval-based coral restoration and facilitate ecological recovery in reef systems where natural larval supply and recruitment processes are no longer adequate to sustain resilience processes.

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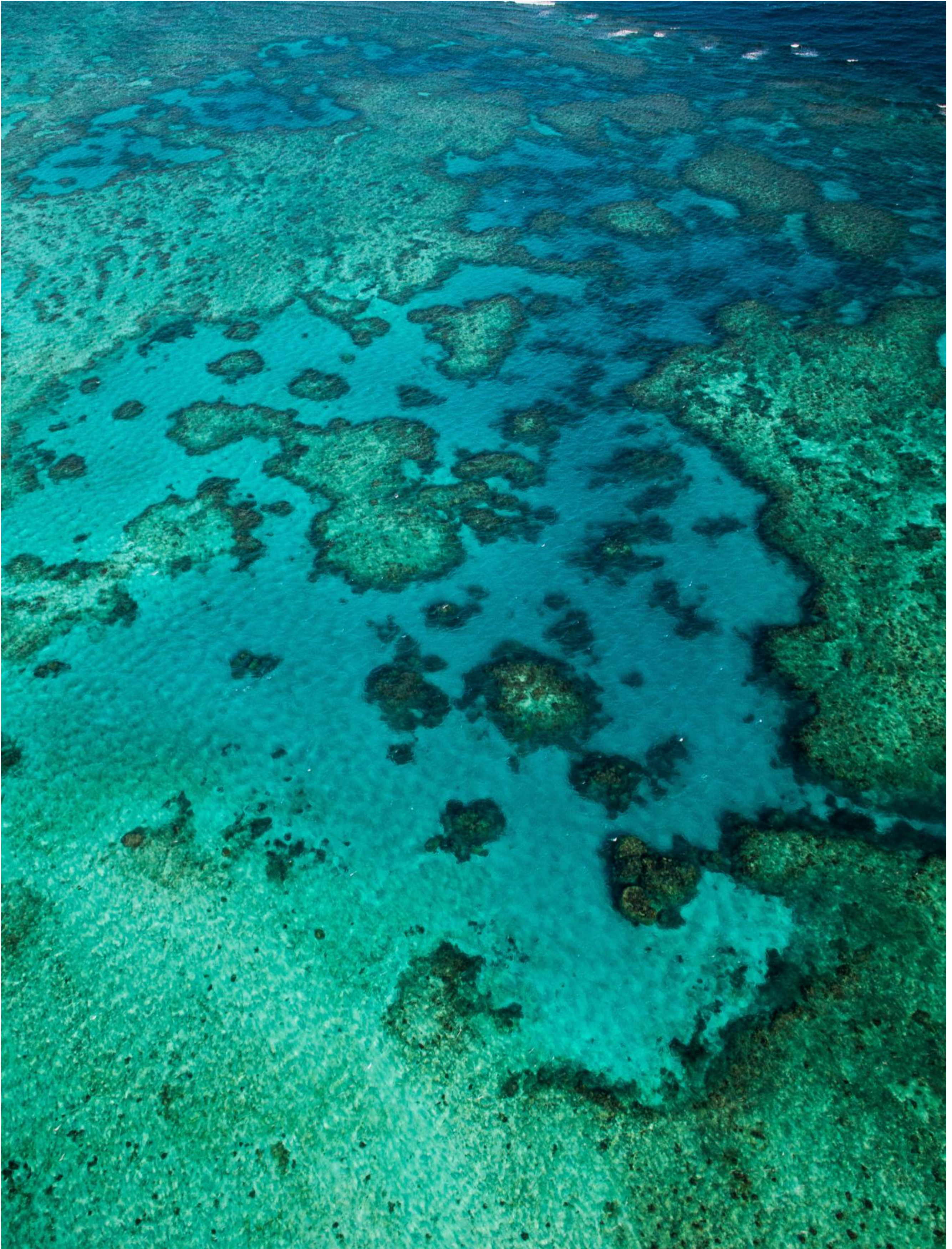

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