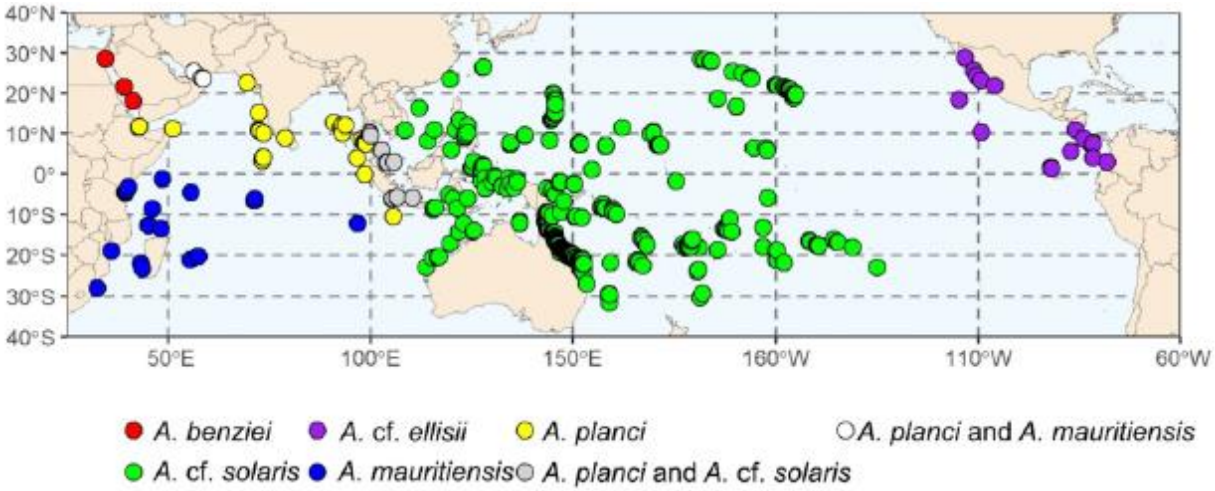
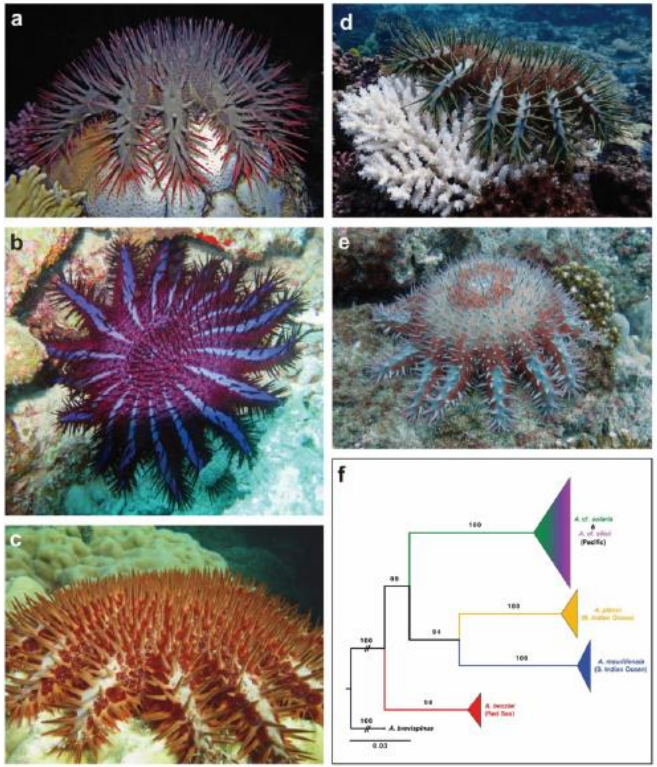
Early detection of the 5th Crown-of-Thorns Seastar outbreak on the GBR using species specific eDNA markers

Sven Uthicke, Jason Doyle, Maria Gomez Cabrera, Frances Patel

AIMS: Australia's tropical marine research agency.

Introduction: Crown of Thorns Seastar Species





CoTS Outbreaks on GBR

Australian Government | Australian Institute of Marine Science

4 outbreaks since the 1960

Last to start ~ 2010

CoTS Initiation Box (possibly further N)

Divers culling CoTS on the GBR

Active culling program on the GBR

> 40% of coral decline attributed to CoTS

Osborne et al (2011). PloS one 6(3): e17516.
De'ath et al. (2012). *Proc. Natl. Acad. Sci. USA*, *9*, 17995-17999.

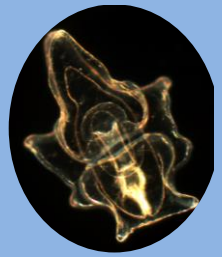
CoTS Survey Methods

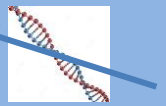
Main Current method: Manta tow

- Used by AIMS LTMP since 1984
- Also by CoTS control program
- Great large-scale method, but only higher densities can be detected
  - **Goal: Develop an eDNA method to detect early outbreaks allowing early intervention and Understanding of outbreak causes**

Applications using CoTS – specific primers

1 Larvae



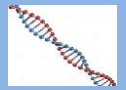


Plankton sample

Uthicke et al. (2015). Scientific Reports
Doyle et al. (2017). Marine Biology
Uthicke et al. (2019). Marine Biology

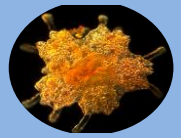
2 Juveniles and adults

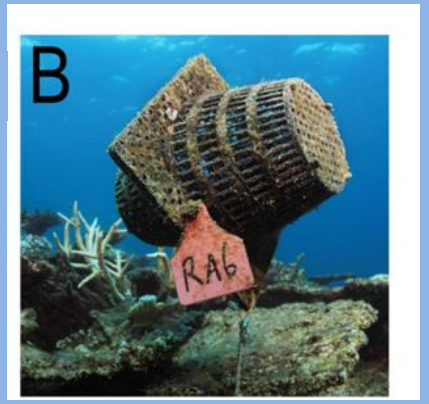
Water sample (2L)



Uthicke et al. (2018). Coral Reefs
Doyle and Uthicke (2020). Environmental DNA
Kwong et al. (2021). Marine Biology
Uthicke et al. (2022). Science of the Total Environment

3 Young recruits





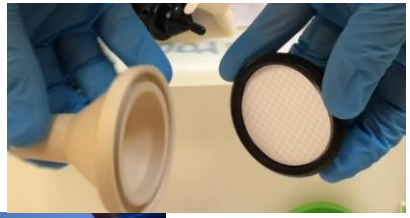
Doll et al. (2021). The Biological Bulletin **241**(3)

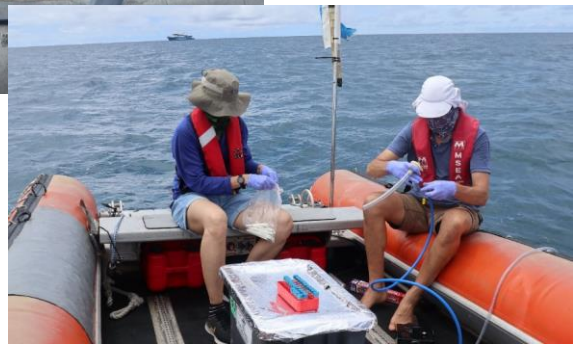
4 CoTS predators (gut contents analyses)

Fish: Kroon et al. (2020). Scientific Reports **10**(1): 1-14.
Invertebrates: Wolfe et al. (in press), PNAS;
Desbiens et al. (2023). *Coral Reefs, 42, 579-591.*

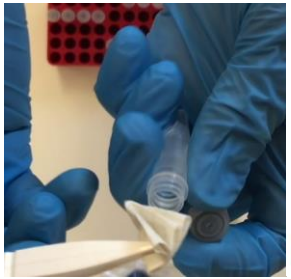
CoTS eDNA General Workflow

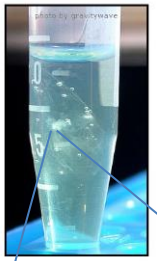
1. filtering surface water through Cellulose Nitrate filter (1µm)—fix in ATL



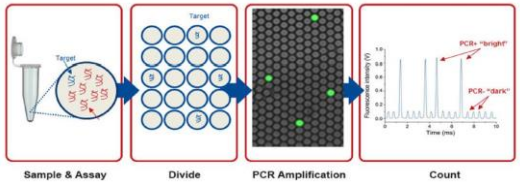


2. Extract DNA





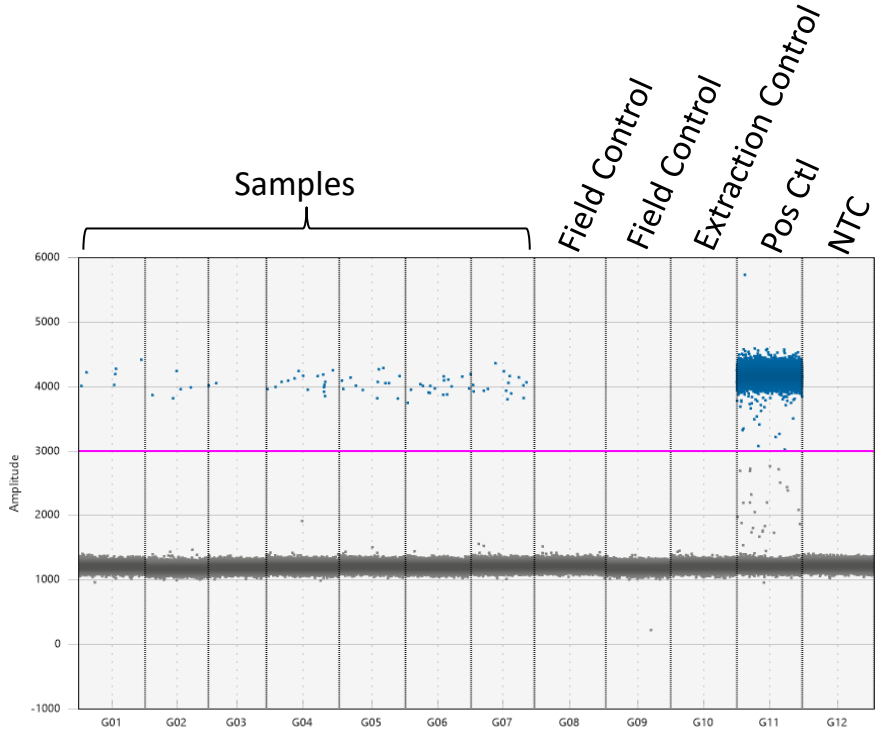
3. Analyse DNA for CoTS using digital PCR



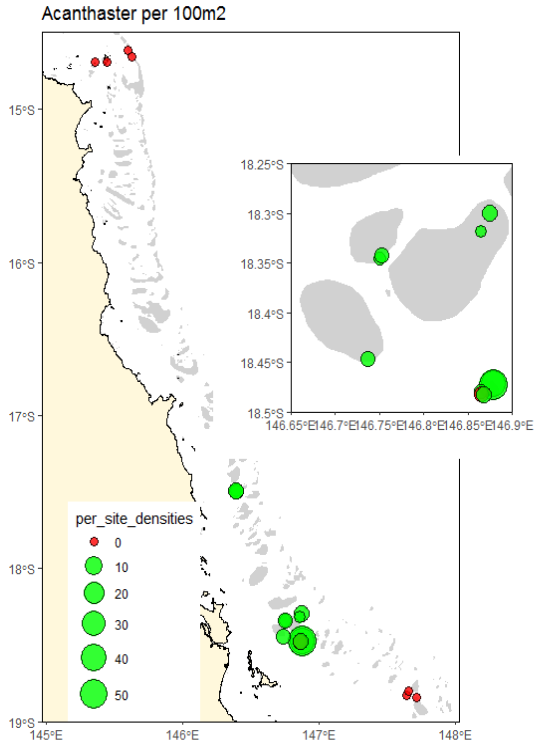
Eble, J. A., et al. (2020).
doi.org/10.1016/bs.amb.2020.01.001

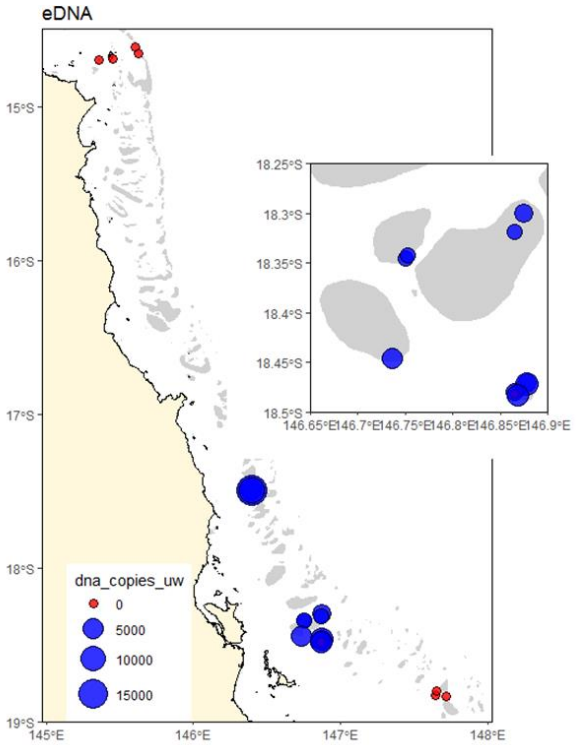
Digital Droplet PCR (ddPCR)

- TaqMan probe assay
- Auto droplet generation
- Minimal hands on (combined with Qiacube) - ↑ automation
- Lab workflow ‘normalised’ across users
- Low copy number detection – important for cryptic animals in marine environment



Adult detection through eDNA: previous work

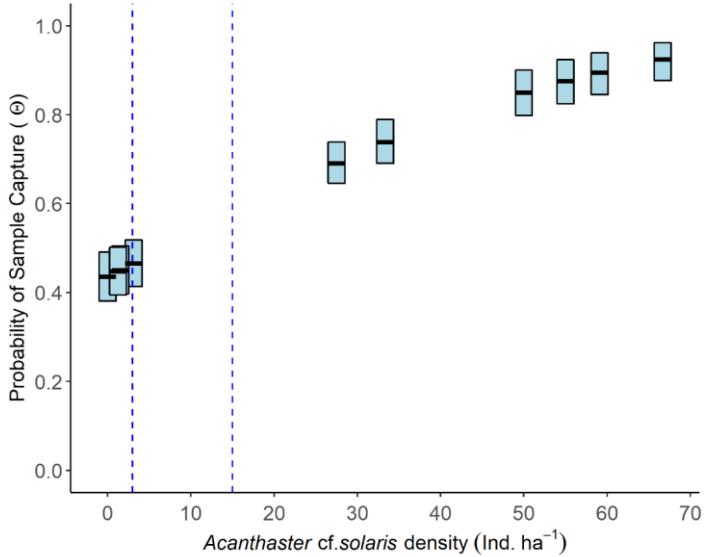




Uthicke, S., M. Lamare and J. R. Doyle (2018). Coral Reefs 37: 1229–1239

Adult detection through eDNA: previous work

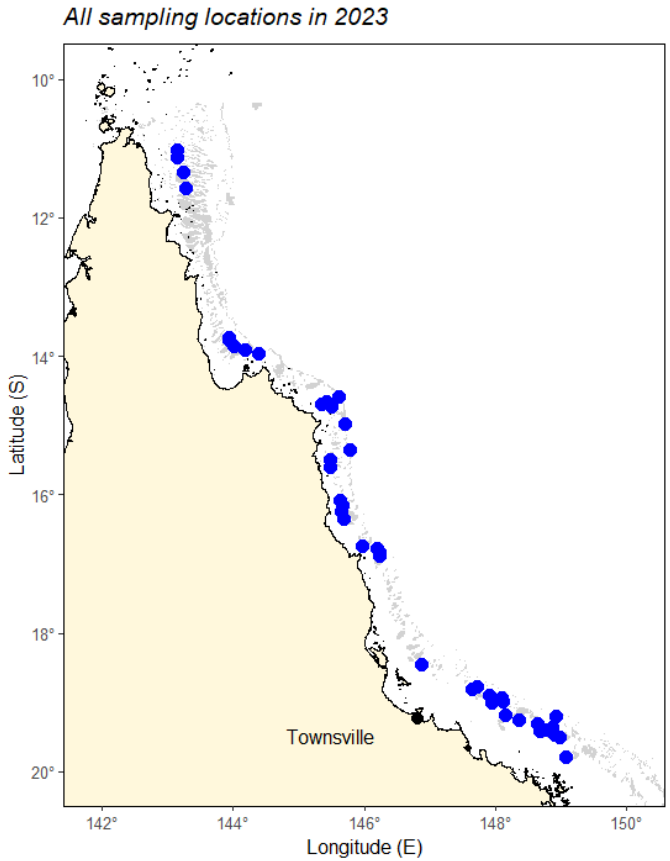
Sample capture corresponds to CoTS at low densities



→ Clear relation between densities and proportion of positive samples

Uthicke, S., Robson, B., Doyle, J. R., Logan, M., Pratchett, M. S., & Lamare, M. (2022). Science of the Total Environment, 158143.

Methods: Operationalising monitoring. 1) Large scale patters



Reefs surveyed:

22 in 2021
26 in 2022
42 in 2023

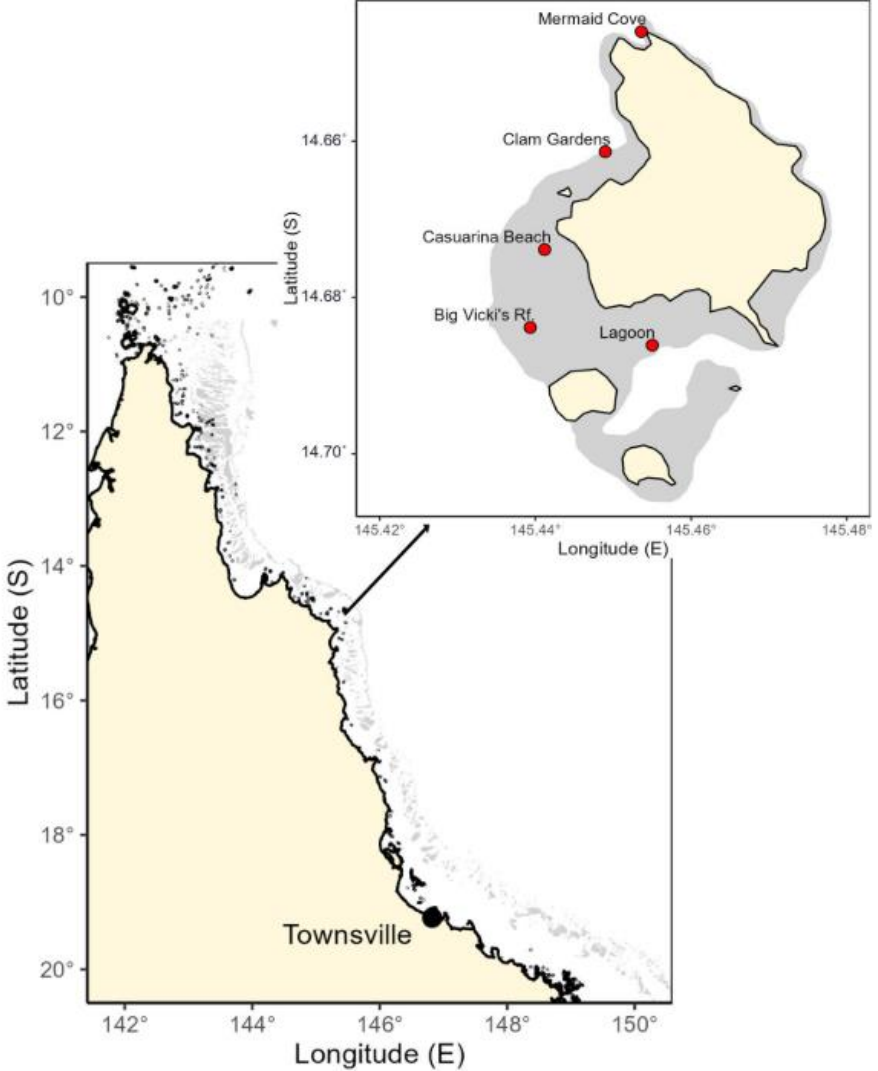
Each reef: 3 sites and 12 samples per site

Operationalising monitoring
2) Sentinel Station at Lizard island

Establish a sentinel station and test **small scale patterns**

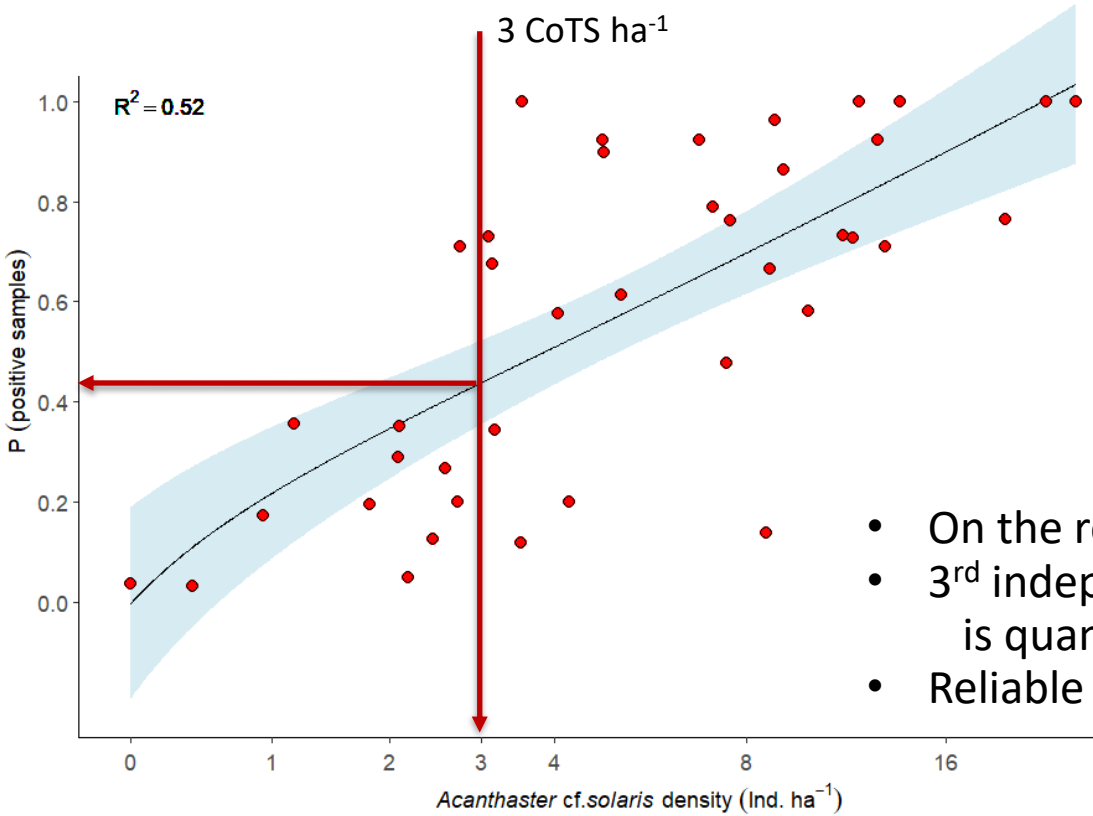
- 2x15 samples at 5 sites, annually
- Now fully analyzed 5 years of data

- Here (and at several other reefs) comparison to **scooter surveys**



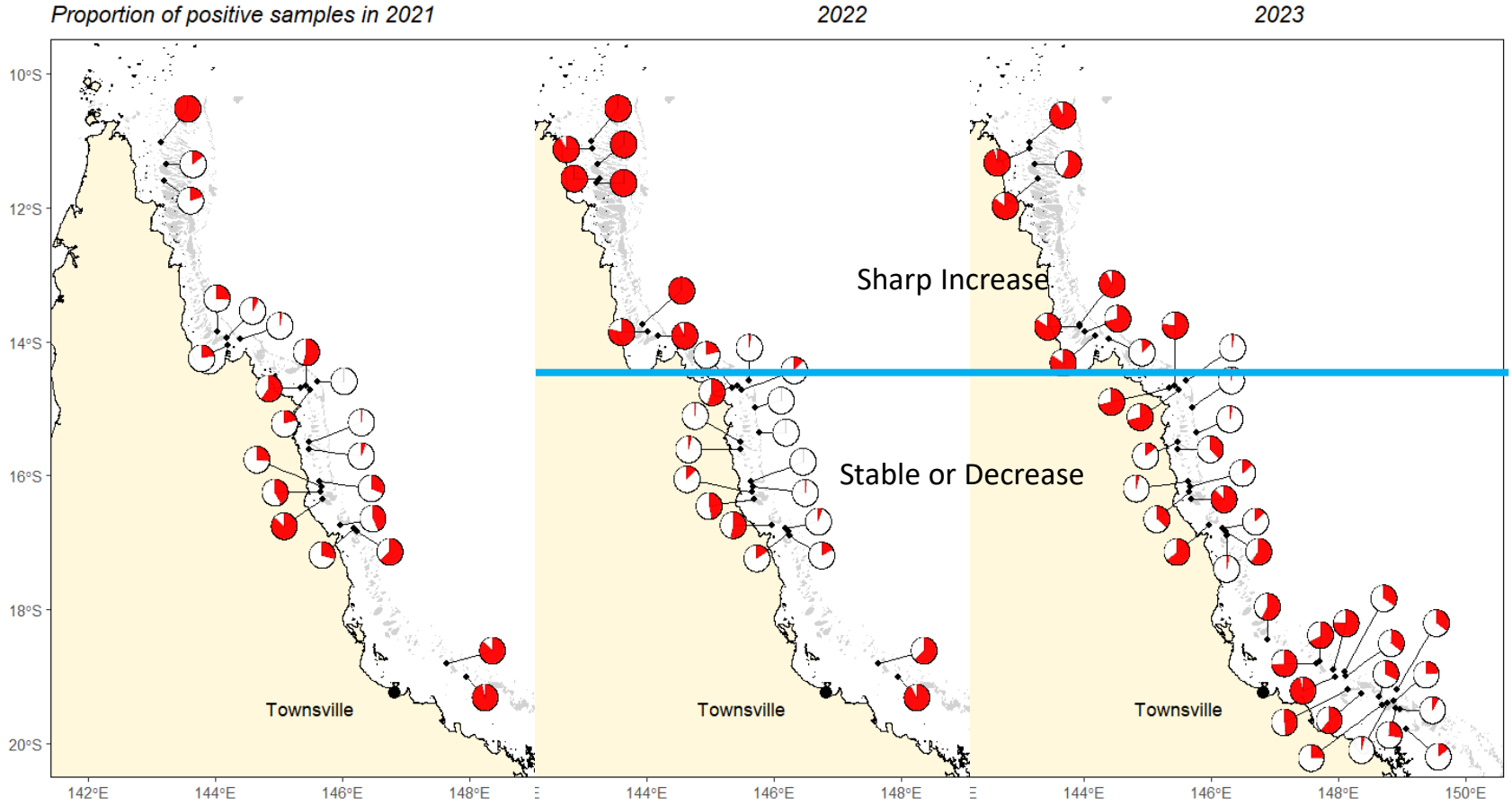
Chandler, J. F., D. Burn, C. F. Caballes, P. C. Doll, S. L. Kwong, B. J. Lang, K. I. Pacey and M. S. Pratchett (2023). Scientific Reports

Results: Good correlation between scooter surveys and eDNA

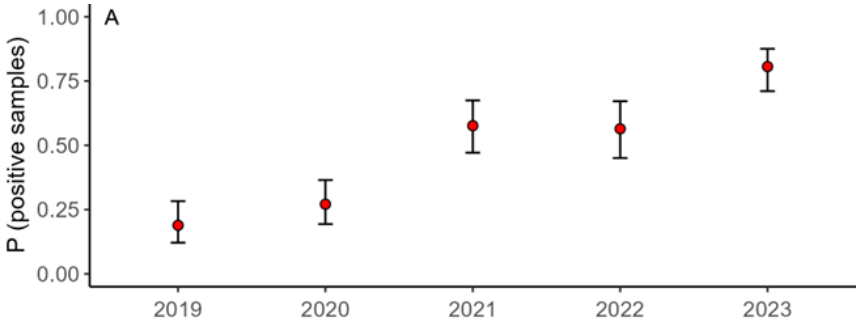


- On the reef level, 40 comparisons available
- 3rd independent dataset illustrating eDNA is quantitative
- Reliable detection at low densities confirmed

Large scale patterns over 3 years

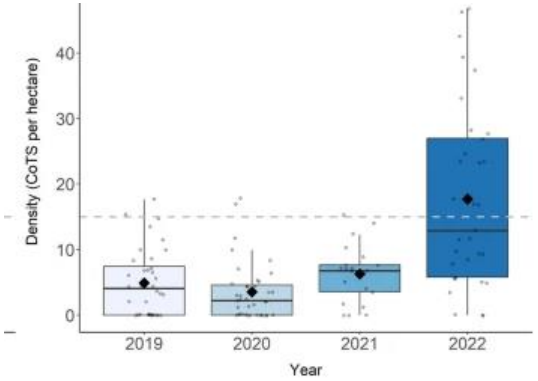


Small scale surveys, Lizard Island



Uthicke, S., Doyle, J. R., Gomez Cabrera, M., Patel, F., McLatchie, M. J., Doll, P. C., Chandler, J. F., & Pratchett, M. S. (2024). *Coral Reefs 43: 857-866*

Corresponds well to scooter surveys

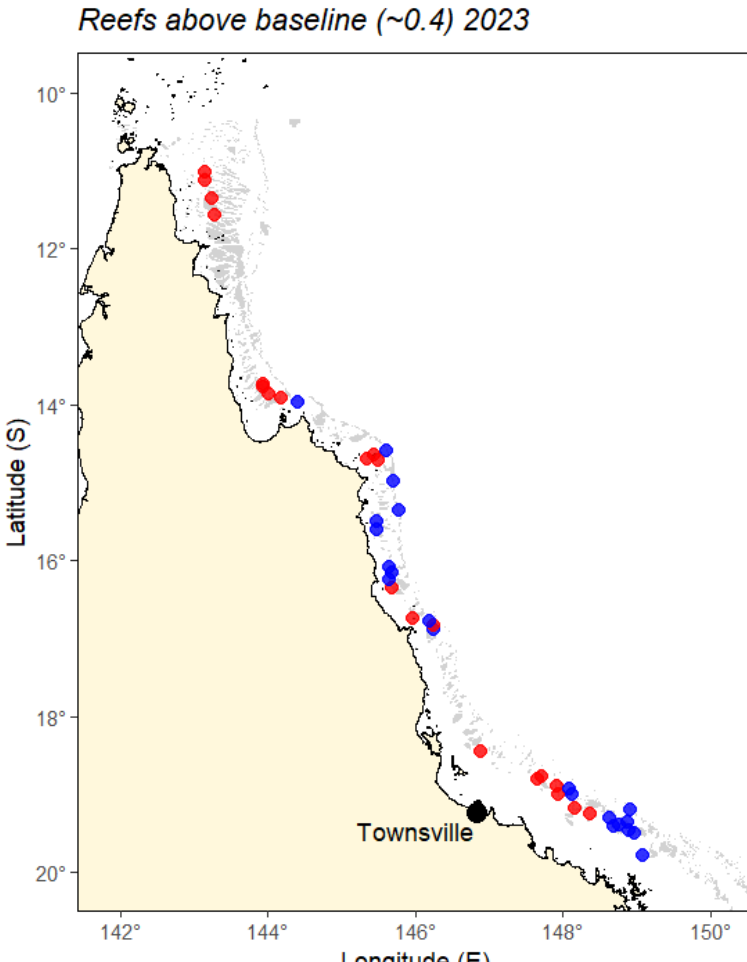


Chandler, J. F., D. Burn, C. F. Caballes, P. C. Doll, S. L. Kwong, B. J. Lang, K. I. Pacey and M. S. Pratchett (2023). *Scientific Reports 13(1): 1930*

Conclusions

- 5th outbreak detected on Lizard Island and Large Scale
  → Congruent patterns, agree with scooter observations
  → (NOT visible with standard surveys)
- Early detection and quantification work well
- .

→ Continue monitoring on ~ 20 reefs, add others through ships of opportunity
→ Continue Lizard Island
→ Modelling suggest sample size can be reduced (e.g., 4 x 6 samples per reef)
→ Continue methods development for fast in-field measurements (e.g. 'Biosensors')



Acknowledgements

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Sarah Kwong
Sam Jaworski
Ash Bean