eDNA for aquatic monitoring and field surveys in tropical waters

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Environmental DNA (eDNA) techniques have the potential to allow rapid and cost-effective surveys of remote aquatic ecosystems. This project is developing eDNA techniques for a selection of species of conservation or management concern and testing their usefulness under field conditions, as well as investigating ways to improve sample handling and laboratory analyses.

What's new?

- We developed an eDNA probe for the invasive aquatic plant cabomba (Cabomba caroliniana) and used it to test if cabomba had been eradicated from a section of the Darwin River. In November 2018, our project partners from the NT Dept of Environment and Natural Resources (DENR) collected water samples from the area where cabomba had been sprayed with herbicide 18 months earlier. We detected cabomba eDNA in some samples but further monitoring is required to assess whether we are detecting 'legacy' eDNA bound to bottom sediments.
- Our monitoring of Mozambique tilapia
 (Oreochromis mossambicus) in north
 Queensland's Walsh River with collaborators
 from the Biosecurity Queensland found no
 eDNA from this species, but suggested that
 another species of tilapia may be present. We
 modified our eDNA primer for Mozambique
 tilapia to include the spotted tilapia (Tilapia
 mariae) and rescreened the samples from the
 Walsh River. Spotted tilapia eDNA was detected.
 In July 2019 we sampled water from the Walsh,
 Wild and Mitchell Rivers that will be screened
 for the eDNA of both species.
- In April 2019 we collected water samples
 from creeks in the Carbine Tablelands of
 Queensland's Wet Tropics to screen for eDNA
 of three species of frog: Litoria dayi (listed as
 Vulnerable but thought to be extinct in the area),
 L. nannotis (Endangered) and L. lorica (Critically
 Endangered). We were able to detect the
 eDNA of L. nannotis and L. lorica up to 20 km

- downstream of a location where they are known to still occur.
- We conducted experiments to test the effectiveness of using eDNA to detect cane toads (*Rhinella marina*). We were able to detect cane toad eDNA in an 800-litre pond after a toad had been placed in the pond for five minutes, which simulates the minimum time that a toad might spend rehydrating. We were able to detect the cane toad's eDNA 2–3 days later, even at water temperatures as high as 35°C.
- We developed new protocols to collect and preserve water samples, rather than filtering them. This extends the length of time needed between the collection of samples and their delivery to the laboratory for analysis, and does not require their refrigeration.





eDNA assays have been developed for the threatened Australian lace-lid frog Litoria dayi (top) and the invasive cane toad Rhinella marina (bottom), photos Trent Townsend, NESP Northern Hub.

- A dedicated eDNA lab has been fitted out with new equipment at JCU Townsville. eDNA analysis works with traces of DNA, and strict laboratory protocols are necessary to reduce the risks of sample contamination.
- We completed four technical reports related to the eDNA surveys and assays for cabomba, tilapia, rainforest frogs and cane toad.
- We resampled the Carbine Tablelands and several creeks in the Daintree and Cape Tribulation, where we found presence of *L. nannotis* and *L. dayi*.
- We were grateful to receive samples collected by the Anindilyakwa Land and Sea Rangers from a saltwater lake on NT's Groote Eylandt to be screened for presence of largetooth sawfish, Pristis pristis.

What's next?

- We will continue to work with NT DENR on the cabomba eradication program and test water samples collected before and after the next application of herbicide in September 2020.
- We will continue to test the effectiveness of using eDNA to monitor for tilapia in the river systems of far north Queensland.
- We're preparing a paper on our experiments on the detection and decay rates of cane toad eDNA, as well as a paper on the results of the *L. lorica* and *L. nannotis* surveys.
- We'll continue to refine our protocols for field sampling and laboratory analysis, including further testing of the efficiency of precipitating rather than filtering eDNA from water samples.

Project summary

The pressure to develop the water resources of Northern Australia is increasing. In order to make appropriate management decisions, water resource managers and policy makers need a good understanding of the distribution and abundance of aquatic species, as well as their presence or absence at individual sites. This is particularly so for species of conservation concern, for invasive species, and for cryptic species that are difficult to detect. Aquatic field surveys generally require specialised equipment and expertly trained staff. In remote northern Australia, they are expensive, difficult and often dangerous. Yet all organisms constantly shed DNA into their environment as they release faeces, urine and mucus or shed their skin cells. Modern genetic sequencing techniques enable scientists to identify a species from these tiny traces of its DNA. Aquatic environments are especially suitable for eDNA approaches, as a sample can be obtained by simply collecting water in a bottle.

Further information

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The project page can be found on the Hub website, along with the project start-up factsheet.

This project is due for completion in December 2020.















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