Institution: Imperial College London

Unit of Assessment: 5 – Biological Sciences

Title of case study: A5-3 Environmental DNA technology for biodiversity and ecological monitoring

Period when the underpinning research was undertaken: 2012-2020

Details of staff conducting the underpinning research from the submitting unit:

Name(s):	**Role(s) (e.g. job title):**	**Period(s) employed by submitting HEI:**
i) Professor Vincent Savolainen	i) Professor of Organismic Biology	i) 2007-present
ii) Professor Alfried Vogler	ii) Professor of Molecular Systematics	ii) 1995-present
iii) Dr Carmelo Andujar	iii) Research Associate	iii) 2013-2016
iv) Dr Alba Herraiz	iv) Genomic Analyst and laboratory manager	iv) 2015-2017

Period when the claimed impact occurred: 2014-2020

Is this case study continued from a case study submitted in 2014? N

1. Summary of the impact (indicative maximum 100 words)

Environmental DNA (eDNA) can be used to characterise living organisms from any substrate (water, air, soil, sediment of water bodies etc.). eDNA is a very powerful tool to monitor biodiversity that outperforms traditional survey methods in terms of efficiency and sensitivity and it is non-invasive and easy to standardise. Research teams at Imperial College, led by Savolainen and Vogler, have used eDNA techniques to transform the way ecological monitoring is conducted. This has been achieved, firstly, via a successful start-up company, NatureMetrics Ltd, co-founded by Imperial College currently with 30 staff. Additionally, we have seen the wide uptake of eDNA by ecological consultancies in the UK; the development of efficient services for government mandated tests of endangered great crested newt; country-wide surveys of pollinator communities; novel methods for water filtration method and sensitive methods for individual-species or wider biodiversity audits brought to market internationally and nationally.

2. Underpinning research (indicative maximum 500 words)

The work led to a wider use of novel DNA-based approaches to taxonomy and biodiversity research in environmental monitoring, through applied research and direct involvement with the commercial sector. The use of eDNA involves two types of technology, the detection of single species using targeted high-sensitivity qPCR assays and the bulk analysis of mixed species communities using 'metabarcoding' and metagenomics. Research at Imperial contributed to both approaches **[1-5]**. Single-species detection for the endangered great crested newt was based on published protocols but its commercial availability in the UK was limited. Imperial researchers optimised the assay and rolled out widely to the ecological services industry, with funds from an Impact Acceleration Award (2014) and a Follow-on grant of NERC (2015). This work led to the formation of a start-up company, NatureMetrics, where these assays provided an early revenue stream. Related assays for invasive American signal crayfish were developed and eDNA quantification established that concentration levels correlate closely with egg-bearing females **[1]**; this has warned end users of eDNA technology that estimating densities of animals using eDNA is season specific. Research is also being done to use eDNA for monitoring shark populations by the Zoological Society of London. Furthermore, single-species sensitive detection methods were developed for conservation purposes, specifically to assess the presence of otters in Bornean rivers, and a technique to monitor hazel dormouse visits of nest tubes using new protocols for

DNA extracted from urine. The second approach for eDNA surveying based on bulk environmental samples originated from DNA taxonomy studies at Imperial and the Natural History Museum London, in part funded by their Biodiversity Initiative (2013-2016) led by Vogler (who holds a joint appointment). This approach using alternatively shotgun sequencing (metagenomic) and PCR amplification (metabarcoding), required extensive method development to optimise laboratory protocols and bioinformatics **[4]**. Combining both techniques provides reliable assessment of mixed species communities from the environment **[4]**. Proof-of-concept studies for eDNA analyses were carried out on mud samples from the shore in Harwich Estuary and identified all the marine invertebrate species present, which recovered almost double the number of animal phyla than the traditional morphological analysis **[2]**. A study of samples from an insecticide spill in 2016 at the river Kennet demonstrated the high resolution of entire invertebrate communities before and after the spill **[3]**. The EU COST Action DNAquaNet, a consortium of representatives of all EU countries, develops monitoring standards for European water bodies to complement existing morphotaxonomic approaches. Similar work was achieved to survey soil arthropods **[4]** and bees **[5]**. The latter has now become a central part of the UK Pollinator Monitoring Scheme (PoMS), conducted by UK-CEH and funded by Defra, and turned into a key revenue stream for NatureMetrics carrying out Environmental Impact Assessments (EIAs).

3. References to the research (indicative maximum of six references)

[1] Dunn, N., Priestley, V., Herraiz, A., Arnold, R., and V. Savolainen. 2017 Behavior and season affect crayfish detection and density inference using environmental DNA. Ecology and Evolution, 7:7777–7785. https://doi.org/10.1002/ece3.3316

[2] Steyaert, M., Priestley, V., Osborne, O., Herraiz, A., Arnold, R., and V. Savolainen. 2020. Metabarcoding and morphology-based methods recover contrasting patterns of animal diversity in intertidal marine benthos. Journal of Applied Ecology 57: 2234-2245 https://doi.org/10.1111/1365-2664.13729

[3] Andújar, C., P. Arribas, C. Gray, K. Bruce, G. Woodward, D. W. Yu, and A. P. Vogler. 2018. Metabarcoding of freshwater invertebrates to detect the effects of a pesticide spill. Molecular Ecology, 27: 146-166 https://doi.org/10.1111/mec.14410

[4] Arribas, P., Andújar, C., Hopkins, K., Shepherd, M., and A. P. Vogler. 2016. Metabarcoding and mitochondrial metagenomics of endogean arthropods to unveil the mesofauna of the soil. Methods in Ecology and Evolution, 7: 1071–1081 https://doi.org/10.1111/2041-210X.12557

[5] Creedy, T.J., H. Norman, C. Q. Tang, K. Qing Chin, C. Andujar, P. Arribas, R. S. O'Connor, C. Carvell, D. G. Notton, and A. P. Vogler. 2019. A validated workflow for rapid taxonomic assignment and monitoring of a national fauna of bees (Apiformes) using high throughput DNA barcoding, Molecular Ecology Resources, 20: 40-53 https://doi.org/10.1111/1755-0998.13056

4. Details of the impact (indicative maximum 750 words)

Impact on Commerce and the economy

The wider area of DNA taxonomy is the specialty of the Vogler & Savolainen labs. Method development for characterising eDNA either through direct sequencing from the water sample or complex specimen mixture was used for developing commercial products (eDNA tests), thanks to Research Council funding and the close collaboration with Thompson Ecology. Many laboratories have worked on these procedures, but Imperial has been particularly successful in transferring this expertise to the commercial sector.

NatureMetrics Ltd was co-founded by Imperial staff (Vogler, with postdoc Andujar, together with researchers at the University of East Anglia) in 2015, initially as a consultancy through Imperial Consultants, with two grants from NERC, in addition to funding from UEA and 'angel investors'. NatureMetrics currently employs ~30 members of staff (18 PhDs, Jan. 2021) and was established

to implement the techniques developed at Imperial by the Vogler lab and offer them to the private and public sector on commercial terms **[A]**. For example, in the spring of 2018 the Great Crested Newt test was conducted on ~2500 samples for ~60 different clients, from private individuals to small and large ecological consultancies **[A]**. The company has traded for four years, is profitable, and has doubled revenue each year with a current annual turnover of £800k **[A]**. NM is increasingly involved in Environmental Impact Assessments (EIAs) conducted by extractive industries and has set up regional offices and partner laboratories in South America, Southeast Asia and Africa. A second funding round concluded in 2019 raised £2.5M at a Pre-Money valuation of £5M **[A]**.

Thomson Unicomarine, the marine branch of the ecology consultancy company Thomson Ecology, worked with the Savolainen lab to assess the biodiversity in the Harwich Estuary. The work conducted in Harwich Estuary helped decipher the marine invertebrate community composition in more detail than the traditional morphology approach. eDNA extracted from mud samples from the shore in Harwich Estuary identified all the marine invertebrate species present. This analysis recovered almost double the number of animal phyla than the traditional morphological one; therefore monitoring programmes are being changed to include metabarcoding. Also commissioned by Thomson Ecology Ltd we use eDNA technology for monitoring the presence of Great Crested Newts, a protected species, which presence would preclude or delay large building works (ca. 350 samples) **[B]**.

Impacts on the environment

Covid19

Funded by College and an Urgency Grant from NERC, Savolainen and colleagues at Imperial are evaluating the potential for sewage (via effluent discharge, storm overflows, and other forms of run-off) to contribute to transmission to humans and wildlife by assessing RNA concentration and viral infectivity from environmental samples, from sewage outflows down to rivers, estuaries, and faeces from wildlife. Using data on concentrations of SARS-CoV-2 RNA fragments in raw sewage as well as in the environment, the team also provides models from population-level prevalence of SARS-CoV-2 to potential transmission routes. Savolainen has been joining fortnightly meetings chaired by Defra, and with representatives of UKRI, the Environmental Agency, the Government's Joint Biosecurity Centre and Office for National Statistics, etc, to advise SARS-Cov-2 risks to the environment. This being translated into policies for disease surveillance; e.g. after Savolainen was asked to inform Defra's Chief Scientist and Chief Vet about his findings of the presence of new, yet uncharacterised, coronaviruses in British bats **[C, D]**.

Surveys of targeted species, including invasive and protected species

The decline in pollinating insects across the UK is of particular concern due to their critical importance in the ecosystem. Since 2017 the UK Centre for Ecology & Hydrology (UKCEH) has led the UK Pollinator Monitoring Scheme (PoMS) which consists of 75 sites across Britain, using passive pan traps **[E]**. Each survey season this produces hundreds of thousands of specimens which need to be identified, a tedious and time-consuming task requiring specialist expertise **[E]**. This has led to two challenges (1) species identifications can be imprecise (2) expertise is expensive and in short supply meaning monitoring is difficult, or costly to implement at a sufficient scale **[E]**. The DNA-based methods described in [6] use deep sequencing of trap samples to identify bee species, which overcomes high error rates and inconsistencies in conventional taxonomical identifications and help solve the two above challenges of the monitoring scheme. Furthermore, these analyses allow for the study of associated pollen DNA to determine the plant visited by an insect **[E]**. In partnership with the UKCEH they have secured £50,000 from Defra to further develop the metabarcoding which will allow the DNA methods to be fully integrated in the PoMS as its main route for specimen identification, thereby increasing the capacity of the scheme for monitoring and as evidence base for pollinator declines across the UK **[E]**.

Our work with the American signal crayfish showed that eDNA levels correlate with crayfish numbers in egg-bearing females, meaning that population size can be estimated during the breeding season. This presents a great advantage in the management of this invasive species **[F]**.

With regard to protected species, both the European otter and the European water vole are protected under Schedule 5 of the Wildlife and Countryside Act 1981. This means that surveys are required before undertaking development projects that can alter their habitats. Currently traditional methods are used for such surveys based on looking for visual signs of their presence, including tracks, dens, dung, feeding remains, and live individuals. In collaboration with the National Trust, Mammal Society, Wildlife Trust, Somerset Otter Group, New Forest Wildlife Park, Cardiff University's Otter Project and the British Wildlife Centre, we developed eDNA tests for otters, mink (which threaten native water voles) and water voles that proved to be more efficient than traditional methods in detecting those species **[G, H]**.

As another example, the hazel dormouse, which is also protected, is traditionally surveyed using nest tubes. There is a risk of a false-negative result if dormice visit a survey nest tube but do not build a nest and the method therefore takes several months to complete, which can significantly delay building developments and infrastructure projects **[I]**. In collaboration with the British Wildlife Centre and London Zoo ZSL, we developed an eDNA test to detect hazel dormice visits to nest tubes by amplifying DNA from urine, to reduce the false-negative risk of the traditional survey method and hasten their detection. By having a robust test, it gives developers certainty and can mitigate possible delays **[I]**. Now, People's Trust for Endangered Species and Natural England have adopted this new method to measure abundance and/or offer population ecology of other species **[I, J]**.

5. Sources to corroborate the impact (indicative maximum of 10 references)

[A] Supporting letter from NatureMetrics
[B] Supporting letter from Thomson Ecology Ltd.
[C] Supporting letter from Public Health England
[D] Supporting letter from Defra
[E] Supporting letter from UKCEH
[F] News story from Environment Analyst on Invasive crayfish and the application of eDNA sampling https://environment-analyst.com/uk/58667/edna-sampling-during-breeding-could-control-invasive-crayfish (Archived here)
[G] Blogpost from the Somerset Otter Group regarding eDNA usage http://www.somersetottergroup.org.uk/archives/2288 (Archived here)
[H] News piece from Thomson Ecology on collaboration with Imperial College London with eDNA sampling https://www.thomsonec.com/news/working-with-imperial-college-london-to-expand-the-use-of-edna/ (Archived here)
[I] Supporting letter from an Independent mammal ecologist and ecological consultant
[J] Supporting letter from PTES