

Below we outline our ongoing and near-term research plans:

- 1) Ongoing: A repeated and unevicenced line of defence from Cefas and the Environment Agency is that the pyridine measured in the dead crabs from the impacted zone was produced post-mortem. This is despite there being no literature support for this, plus Cefas renegeed on a prior commitment to test this hypothesis. We have therefore begun an experiment to test this hypothesis ourselves. This experiment is ongoing and involves the controlled decomposition of *Cancer pagurus* in air, seawater, and sediment (see photos below). Carcasses are sampled at fixed time intervals with tissue collected for pyridine analysis. Samples are also taken for bacterial community composition using Illumina MiSeq. We intend to submit this work to a peer reviewed journal. Funded by Newcastle HEIF money.



- 2) Ongoing: continued reanalysis of the MMO landings data. We intend to submit this work to a peer reviewed journal. Unfunded.
- 3) Ongoing: awaiting return of pyridine data from the *Cancer pagurus* exposure trials (exposure water, gills, hepatopancreas, muscle), which will allow us to determine the tissue equilibrium concentration. Once these data are available, they will be integrated into our existing preprint and will be submitted for peer review. Funded by Fishmongers' Company.
- 4) Near-term (post-Christmas). Water samples collected and pending analysis for pyridine loss from our toxicity exposure tanks. These data will allow us to recalibrate our LD data rather than assuming a constant pyridine concentration through the 72h exposures. Funded by Fishmongers' Company.
- 5) Near-term (pre-Christmas). We will repeat the *Cancer pagurus* toxicity trials with using *Carcinus maenas* to determine if there are any species-specific responses. These experiments will be further expanded to test whether the presence of sediment affects pyridine toxicity. Funded by Fishmongers' Company.
- 6) Near-term (post-Christmas). Further optimisation of our pyridine transport model as outlined in the manuscript Discussion. Unfunded.
- 7) Near-term (post-Christmas). Experimental validation of the pyridine adduct formation secondary toxicity mechanism (using ¹⁵N labelled pyridine) through substitution of nicotinamide adenine dinucleotide (NAD). Presently, we have run a comprehensive set of experiments on NAD and we have assigned the proton and carbon peaks (with the aid of some 2D spectra) that has allowed us to assign the ¹⁵N peaks ([NAD assigned NMR spectra.pdf](#)). We have identified the nitrogen peak coming from the nicotinamide part of the molecule, and we expect the chemical shift for pyridine will probably be similar. If we do get substitution of the ¹⁵N labelled pyridine then the intensity of this ¹⁵N signal should allow us to detect it directly. If we can't detect any ¹⁵N directly then that may be evidence that the pyridine hasn't replaced the nicotinamide. Funded by Newcastle HEIF money.
- 8) Near-term (February-March) Implementation of a sophisticated mixture-based risk assessment tool (Knowledge Graphs) to assess the risk of dredging to the marine environment. Funded by Sea-Changers.
- 9) Aspirational – we have repeatedly sought permission to take vibro-corer sample in the Tees estuary to determine total pyridine concentrations, in addition to other contaminants of concern. Our requests have been resolutely ignored, despite offering to cover the costs of the work. This is a vital part of missing evidence and was highlighted as such during the EFRA Select Committee session. We would fund this through a combination of contributions from the Fishmonger's Company, an existing gofundme pot and through various internal university funding pots.