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Reply to the 'Comment on "Why there is no evidence that pyridine killed the English crabs"' by A. Peters, *Environ. Sci.: Adv.*, 2025, 4, DOI: 10.1039/D4VA00420E

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We highlight that the commentary by Peters which uses a CREED/CRED risk assessment framework supports the previous government reports that rejected the pyridine hypothesis and considers the previous evidence as unreliable. This reaffirms our conclusions that pyridine didn't kill the Teesside crabs.

The Comment by Peters, in response to our paper,¹ raises the question as to whether we could, or should, have used an environmental or chemical risk assessment framework such as CREED² or CRED³ when making a judgement as to whether pyridine caused the crabs mortality event off Teesside. We agree that these frameworks are a useful and transparent approach. However, we must highlight that our paper was a perspective piece, based on an independent panel report,⁴ about why pyridine was an unlikely culprit in the mass mortality event. It was not intended to be a full environmental risk assessment. Our perspective was drafted in Autumn 2023 in response to ongoing speculation that pyridine was a causal factor despite the published findings of the independent panel, and a subsequent study.^{5,6} We also wanted to contest unfounded allegations by some politicians and journalists that the independent panel of university scientists had falsified elements of their report on behalf of the UK Government. These concerns were also published to a more general public audience shortly after our perspective was published.⁷ Hence our paper aim states '*Unfortunately, the debate became highly politicised, with misleading information aired by the two largest political parties. Here, several members of that independent review panel refute the pyridine link to the mass mortality and highlight where the science has been misrepresented by the media. In doing so, we explain why pyridine did not kill the Teesside crabs. We do this by answering the following 5 questions.*' It is also worth noting that all the CREED guidelines and associated literature cited by Peters were published after our manuscript was submitted for publication (9th January 2024).

For a risk assessment to have validity, the best possible data (empirical and modelled) should be used and the premise

behind those data ascertained. The pyridine hypothesis was predicated on two main pieces of evidence, which had not been peer reviewed. Firstly, concentrations of pyridine were higher in crab tissues from areas associated with the mortality event compared to reference areas. Not only did this turn out to be false (*i.e.* there was no significant difference between locations), but the Environment Agency (EA) considered their own data to be unreliable, due to method limitations. Unfortunately, some elements of the media presented the pyridine hypothesis as highly credible and promoted the idea of a cover-up. The EA's concern that their pyridine data was unreliable was later justified when CEFAS reanalysed archived and new samples using an optimised and validated methodology published as a DEFRA/CEFAS report⁵ and later as a peer reviewed paper.⁶ They not only found that the original concentrations of pyridine in crab tissues were three orders of magnitude too high, but that there were also no differences between impacted and control locations. Therefore, as we state in our manuscript, the rationale for considering the pyridine hypothesis in the first place is substantially diminished (or unreliable). Peters judges the DEFRA/CEFAS report⁵ to be '*reliable with restrictions and relevant without restrictions at the silver assessment level, with the limitation on the reliability that only sampling dates were reported, but the sampling times were not reported*'. We are unclear why Peters makes a judgement on the reliability of the CEFAS analysis but not the reliability of the former EA data which predicated the pyridine hypothesis. Either way it was judged that the new data were reliable in confirming that the old data were not so.

The second piece of important evidence came from an unpublished piece of research which was presented to the EFRA committee and became key to the launch of an independent panel inquiry. We believe that using grey literature (*e.g.* Eastbrook *et al.* *BioRxiv* preprint⁸) as the core tenet of a risk assessment providing both hazard data (LC₅₀) and exposure assessment (*e.g.* 'dose to crabs') provides huge uncertainty and the 'ecotox' data is uncertain and unreliable – for example, it

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would score 3 ('unreliable') on the Klimisch scale.⁹ The CRED framework was designed to determine the suitability of peer reviewed literature for regulatory purposes. Eastabrook *et al.* was uploaded as a *BioRxiv* preprint in November 2022 and to date has not appeared in the peer reviewed literature so is unlikely to be used in an environmental risk assessment. Peters, using the CRED framework, considers the study by Eastabrook *et al.*⁸ to be unreliable which reaffirms our conclusions. However, Peters' comment on our paper does pick out a nematode study and appears to suggest that we aren't comparing like for like when assessing the statement that pyridine is 'exceptionally toxic to crustaceans'. We think Peters has misinterpreted our figure which presented all the available LC₅₀ data in the US EPA ecotox database. The opinion that pyridine was exceptionally toxic to crustaceans was presented to the EFRA committee.¹⁰ It was based on data contained in Eastabrook *et al.*⁸ and suggested as a potential reason for why only crabs and lobsters were washing up dead and not other marine life. When we presented data we were highlighting that pyridine was not exceptionally toxic to crustaceans comparative to other phyla, and not exceptionally toxic compared with other common contaminants found in industrialised sediments. The nematode study which Peters scrutinised did not factor into our conclusions in addressing this question and hence we don't mention it in our paper. Our figure details 81 LC₅₀ values across many vertebrate and invertebrate phyla including those of marine and freshwater crustaceans.¹

Peters when addressing our question 'has pyridine ever been recorded at concentrations likely to cause acute toxicity' reports that '*the most relevant information for addressing this question is aquatic monitoring data for pyridine, from the time and location of the incident.*' Pyridine was monitored around the time of the incident by the EA and was below the levels of detection in water.⁴ Unpublished data were also provided to the independent panel generated from York University on pyridine in sediments.⁴ These data were not scrutinised under the CREED criteria by Peters. Peters does make a judgement on the pyridine found in water and sediments published in November 2023 by DEFRA⁵ but not the data presented previously by the EA, CEFAS and independent research presented to the independent panel.¹¹ Peters considers the data in the DEFRA report '*as reliable, although an additional relevance assessment is required for the new purpose of the assessment. In this case water samples are identified as the appropriate medium for sampling, but samples from other media are acceptable if no water samples are available, but the data from them would have significant limitations associated with it.*'

Overall we think this approach by Peters is a valuable addition. However, as it makes judgements on some, but not all, of the available data, it could potentially bias the overall conclusions, especially when some data might support or conflict with existing hypotheses. The independent panel was reviewing large numbers of unpublished datasets from multiple agencies and universities as well as published literature covering algal blooms, deoxygenation events, disease/parasite outbreaks, dredging, environmental chemistry and toxicology. Peters does not come to any conclusions from the CREED/CRED approach

although does appear to support the DEFRA report⁵ that rejected the pyridine hypothesis and considers the data presented by Eastabrook *et al.*⁸ as unreliable. This reaffirms our conclusions that pyridine didn't kill the Teesside crabs.

Conflicts of interest

There are no conflicts of interest to declare.

Data availability

This is a rebuttal and contains no data.

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