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Author for correspondence: E. D. Stevens e-mail: dstevens@uoquelph.ca

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Neurobiology

Stress is not pain. Comment on Elwood and Adams (2015) 'Electric shock causes physiological stress responses in shore crabs, consistent with prediction of pain'

- E. D. Stevens¹, R. Arlinghaus², H. I. Browman³, S. J. Cooke⁴, I. G. Cowx⁵,
- B. K. Diggles⁶, B. Key⁷, J. D. Rose⁸, W. Sawynok⁹, A. Schwab¹⁰,
- A. B. Skiftesvik³, C. A. Watson¹¹ and C. D. L. Wynne¹²

¹Department of Biomedical Science, Atlantic Veterinary College, Charlottetown, Prince Edward Island, Canada ²Leibniz-Institute of Freshwater Ecology and Inland Fisheries, Humboldt-Universität zu Berlin, Berlin, Germany ³Institute of Marine Research, Storebø, Norway

⁴Department of Biology, Carleton University, Ottawa, Ontario, Canada

- ⁵Hull International Fisheries Institute, University of Hull, Hull, Humberside, UK
- ⁶DigsFish Services, Banksia Beach, Australia
- ⁷Department of Biomedical Sciences, University of Queensland, St Lucia, Queensland, Australia

 8 Department of Zoology and Physiology, University of Wyoming Laramie, WY, USA

⁹Infofish Australia, Frenchville, Australia

¹⁰Biglen, Switzerland

¹¹Tropical Aquaculture Laboratory, University of Florida, Gainesville, FL, USA

¹²Department Psychology, Arizona State University, Tempe, AZ 85287, USA

(D) EDS, 0000-0002-3113-1543

Elwood & Adams [1] exposed *Carcinus maenas* to electric shocks. They reported that 6/20 unshocked crabs did not move but all (20/20) shocked crabs did move. No control crabs showed 'extreme responses' [1, p. 2] but 4/20 shocked crabs did. Most crabs walked (14 control and 16 shocked). When comparing only walking crabs, haemolymph lactate was higher in shocked crabs. At the end of the discussion, they state 'Although these physiological responses are expected should an animal experience pain, they do not prove the feeling of pain in decapods...' [1, p. 3]. They nevertheless conclude that their results, interpreted in the context of previous research, '...fulfils the criteria expected of a pain experience' [1, p. 1]. We contend that their conclusion that crabs experienced pain (as summarized in the title and abstract) is unfounded, because neither is stress a necessary prerequisite for pain nor is it always coupled with pain.

We are concerned by the authors relating stress to pain. Lactate may indicate physiological stress in some crustaceans, but physiological stress is not pain. Many stimuli used to study stress, confinement of fish, for example, are not associated with stimulation of nociceptors. There is a wealth of literature demonstrating that many different stimuli elicit physiological stress, including benign experimenter interventions [2]. Physiological stress is neither specifically nor selectively related to pain and cannot be inferred to be a useful index of pain. Stress is a confounding factor in 'pain' experiments [3] and efforts often are made to show stress does not occur (e.g. [4]). Hence, elevated lactate in shocked crabs does not necessarily depend on pain.

Similarly, pain is not necessarily stress, as the former, at the very minimum, requires activation of nociceptive pathways, followed by conscious higher level neural processing [5], neither of which was demonstrated.

In discussing behaviours that they argue reflect pain, the authors cite learning to avoid aversive stimuli and 'activities that go beyond mere reflex response' [1, p. 1]; neither are adequate criteria for pain [5]. Likewise, behaviours like shell

2

abandonment or grooming described in prior publications by the same laboratory are insufficient to demonstrate pain. The authors do not provide a persuasive argument for the continued use of these criteria for pain.

There also are methodological shortcomings that confound their interpretations. Walking was not quantified; animals were subjectively partitioned into subgroups; the tissue producing lactate was not determined and muscle activity was not quantified.

The authors state that shocks 'did not appear to cause substantial muscle contraction, which might have accounted for high lactate, because the crabs walked normally' [1, p. 3]. Crustacean muscle is characterized by large motor units, the majority innervated with only two large axons with low thresholds that are much more easily stimulated with external electrodes than small sensory axons [6]. Thus, an alternative and more parsimonious explanation for their lactate results is that the shocks induced some muscle activity.

A variety of stressors can elevate lactate in crabs; the most potent is activity, which can elevate lactate an order of magnitude above the values the authors reported for shocked crabs [7]. Moreover, the 'elevated' [1, p. 2] lactate levels reported are within the normal range for this species, suggesting that they were not unduly stressed [8]. The authors did not report heart or breathing rates, making it difficult to assess if their results were associated with shocks, or indirect physiological effects. Another alternative explanation of their results is that their shocks stopped the heart and respiratory movements [9], thus increasing anaerobic metabolites (lactate) before sampling.

In conclusion, we argue that the reported data fail to show that haemolymph lactate is an appropriate measure of 'pain-like states' [1, p. 2] in crabs or provide evidence to support the conclusion that crabs exhibit 'pain-like states'. We agree with Dawkins that one should address animal welfare pragmatically using stress-related indicators without reference to conscious experiences [10]. However, legally, the ability to feel pain often is used as a criterion to distinguish among species that deserve moral attention from those that do not [11]. Thus, we are concerned that policy-makers may make inappropriate decisions about crab welfare based on this over-interpreted study.

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