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SOLUTIONS FOR PEOPLE, ANIMALS AND ENVIRONMENT

Assessing negative and positive evidence for animal pain

Commentary on **<u>Birch</u>** on *Precautionary Principle*

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Abstract: Jonathan Birch suggests that we should take one well-conducted study that produces results consistent with the idea of pain as being sufficient to invoke the animal sentience precautionary principle. Here, I consider how to balance negative and positive results from such studies using examples from my own work. I also consider which criteria of pain might provide strong inference about pain and which may prove to be weaker.

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Birch (2017a) considers the "Animal Sentience Precautionary Principle" and argues that one wellconducted study that is consistent with the idea of a taxon being able to feel pain should be sufficient to grant protection for the order within which that species is placed. He refers to work by me on decapods to illustrate his thoughts. I focus here on what to make of apparently negative findings and apparently positive findings. Some of the points I make here are touched upon in an excellent response to commentaries by Birch (2017b). In some cases, similar conclusions are reached; however, I go into more depth on specific issues, and I wrote before Birch (2017b) was made available.

Are negative findings a problem?

Mallatt (2017) and Woodruff (2017) are concerned by studies that fail to support the idea of pain in crustaceans and appear to believe that a study with negative findings should cancel one (or more) that has positive results. Lack of support is typically due to statistically non-significant results, whereas support is typically based on significant results. These outcomes, however, do not provide equal support for inference. A significant difference between experimental groups tells us that that under the conditions of the experiment, the animal is affected by the different conditions imposed by the experimenter. It provides strong inference. By contrast, a nonsignificant result does not allow the inference that the species in question cannot be affected by those conditions — just that the sample population failed to respond at that time. Thus, as a matter of logic, one negative result should not cancel a positive result. Absence of evidence is not evidence of absence (Sherwin 2001).

Woodruff notes that in my own work, two studies failed to support the expectations of pain. Magee and Elwood (2013) reported swift discrimination by shore crabs between a safe dark shelter and one that resulted in electric shocks, whereas another study (Magee and Elwood 2016), described by Woodruff as being 'slightly different', did not result in discrimination. Whilst the apparatus was broadly similar in the two experiments, the slight difference resulted in a very different paradigm. In the first, the animal was repeatedly placed in an enclosure containing the two shelters and had access to both during each trial. By contrast, in the second experiment, an opaque partition was placed between the shelters during each training trial so that the animal could only access one per training trial. In the first experiment, we had simultaneous presentation of the stimuli, whereas in the second, we had sequential presentation (eventually followed by a test when the crab could choose). Sequential presentation of stimuli in a discrimination task has been shown to be a more demanding task cognitively than simultaneous presentation in a variety of contexts (Dyer and Neumeyer 2005; Dougherty and Shuker 2015). The negative finding with respect to discrimination thus tells us more about the cognitive abilities of the animals than their ability to experience pain. However, even though there was no discrimination learning in Magee and Elwood (2016), the animals used different strategies to reduce their exposure to shock. An increasing number exited the shock shelter with successive trials; a comparison of those that exited on the first and last trials showed they exited more quickly in the later trial, thus markedly reducing the number of shocks. That is, they largely solved the problem of shock avoidance by a different means.

The second, apparently negative finding of concern to Woodruff was about the expectation that animals should show vertebrate-like responses to morphine in inducing analgesia (Bateson 1991). Two studies had shown that crabs given morphine become unresponsive to shock, because they did not move away (Maldonado and Miralto 1982; Lozada et al. 1988), and this was interpreted as crustaceans showing morphine-induced analgesia. However, crustaceans given morphine also fail to respond to visual stimuli (Tomsic and Maldonado 1990). This opens the possibility that the initial conclusion about a specific analgesia was wrong. It could be that morphine induces unresponsiveness to all stimuli (Barr and Elwood 2011). To test this, an experiment was devised in which a crab was offered the chance to enter a dark shelter in which they would receive a shock. If morphine acts as an analgesic, then those given morphine should be more likely to move into the shelter to escape the light because the effect of the shock would be reduced by the analgesia. However, crabs given morphine were less likely to enter the shelter than were control, saline-injected crabs. All crabs given morphine were unresponsive for about 30 minutes after injection and tended not to enter the shelter even if no shock was delivered.

Thus, it was concluded that morphine did not have a specific analgesic effect. The key question is, should we expect all taxa to have the same physiological response to morphine? We know that different taxa have very different physiology. For example, nociceptors in some taxa respond to capsaicin whereas others do not (Elwood et al. 2016). Thus, it is not reasonable to expect the physiological responses of all invertebrates to be the same as those of mammals, and this applies to the way responses to noxious stimuli might be modulated. A similar argument can be used to counter the suggestion by some (e.g., Key 2016) that only animals with brain structures identical to those of humans can experience pain. We know that similar functions — for example, the ability to use visual information — can occur in different taxa with very different brain structures (Elwood 2011). The same principle may apply to emotional responses to stimuli. Woodruff's

concerns on this point are hence unfounded: The results of the various studies on morphine in crustaceans are not fundamentally different because they all indicate a general lack of responsiveness. The results did not differ, although they were interpreted differently.

Which criteria should be regarded as important?

The second point I wish to consider is Birch's suggestion that only one study consistent with the idea of pain should be required to trigger protection under the precautionary principle. If it is to be one study, then it should investigate a criterion of pain that is very convincing (Sneddon et al. 2014).

Birch picks motivational trade-offs as a good criterion of pain. The original aim of examining trade-offs was to establish whether responses were reflexes or more than reflexes. If they are reflexes, then responses should be the same regardless of other motivational conditions. However, the finding that hermit crabs are less likely to evacuate from a shell in which they receive a shock when the shell is of good quality (Elwood & Appel 2009), or when there are clues to the presence of predators (Magee & Elwood 2016), demonstrates that the responses are influenced by other motivational requirements and are hence not purely reflexive in nature. These experiments indicate that the noxious stimulus of shock produces a motivation to avoid the shock that is traded-off against other requirements, such as the requirement of a good quality shell or the need to avoid predators. Importantly, these and other experiments also indicate that the shock produces a motivation that appears to be strong because highly valuable resources may be abandoned to escape the shock. For example, hermit crabs that leave their shells are at risk of predation (Appel & Elwood 2009a), as are shore crabs that abandon dark shelters and move into a light environment (Magee & Elwood 2013). Trade-offs, together with giving up valued resources, are consistent with expectations of pain. This approach enables us to judge the price the animal will pay to avoid the noxious stimulus.

Self-administration of analgesics provides a compelling case for pain experience (Colpaert et al. 1980), but there are problems with this approach in crustaceans, not least because it is not clear what might be an effective analgesic should the animals experience pain. I am not aware of any attempts with crustaceans, but one attempt with honey bees given the opportunity to ingest morphine did not find that their preference for morphine increased when they were injured (Groening et al. 2017). This might be because a) bees do not experience pain; b) they do feel pain but ingested morphine does not act as an analgesic; or c) they do feel pain, and morphine acts as an analgesic but the association was not formed because the bees could sample morphine and non-morphine solutions within the time that it took for morphine to act.

It is reasonable to assume that the ability to experience pain evolved because it enhances long-term fitness. One way pain might be adaptive is that it induces or facilitates avoidance learning of the noxious, potentially damaging stimulus. However, mere demonstration of avoidance learning is weak evidence of pain. Decerebrate shore crabs, for example, appear to learn to keep a leg out of shallow water to avoid shock but only after receiving over 1,500 shocks in the first minute, and continue to receive 400 per minute after 10 minutes of training (Dunn & Barnes 1981). Clearly the apparent learning is not effective in avoiding the stimulus. Contrast this with shore crabs noted above selecting between two dark shelters, one of which results in shock. In only the third trial, significantly more crabs switched their choice from the shock to the non-

shock shelter than vice versa (Magee and Elwood 2013). This demonstrates that crabs can make swift adjustments in their behaviour to avoid noxious stimuli. Thus, the key to evaluating learning is to consider whether the learning is swift. Unless it is swift, it cannot be expected to improve fitness.

Another response that might have positive fitness consequences is rubbing and attending to a wound. This has been seen in wounded brown crabs (McCambridge et al. 2016), hermit crabs that have been shocked on the abdomen (Appel & Elwood 2009b), and prawns (Barr et al. 2008) and shore crabs (Elwood et al. 2017) treated with acetic acid. These experiments show an awareness of the site of the wound and observations of wound guarding (McCambridge et al. 2016), which may reduce further wounding.

Another criterion concerns long-term modification of behaviour that is not easily explained by associative learning. For example, hermit crabs shocked within their shells show a much higher motivation to change to an alternative shell up to 24 hours later (Appel & Elwood 2009b). Also, crayfish (Fossat et al. 2014) become much more risk averse after electric shock, showing evidence of being anxious. Curiously, the anxiety is reduced if the animals are treated with an anti-anxiolytic used to treat anxiety in humans (Fossat et al. 2015). Amphipods show similar risk aversion after shock, which results in greater survival in the presence of a fish predator (Perrot-Minnot et al. 2017). Thus, expectations about pain enhancing fitness are upheld.

Finally, we see a range of physiological changes after wounding/electric shock (Fossat et al. 2014; Patterson et al. 2007) that would be expected if they mediate long-term protection. As noted above, however, we cannot expect that all physiological mechanisms are identical in all taxa. Moreover, physiological changes are difficult to attribute specifically to the shock rather than the immediate vigorous escape response that often occurs following shock, yet this too has been achieved (Elwood & Adams 2015).

Conclusions

We have a range of criteria that suggest pain, some being more persuasive than others. I have noted that slow avoidance learning is not as persuasive as fast avoidance learning. The presence or absence of physiological changes similar to those of mammals may not be particularly persuasive because there is large variation between taxa in physiological mechanisms. For the same reason, having a central nervous system that is morphologically homologous to that of mammals cannot be a criterion. By contrast, motivational trade-offs, coupled with giving up valuable resources, long-term attention to a wound, and guarding of wounds are also suggestive of experiencing pain. Long-term behavioural changes and evidence of anxiety are also consistent with the idea of pain. However, just one criterion does not prove pain and, indeed, all of them combined do not prove pain in any taxon. It remains to be seen whether Birch's precautionary principle based on just one criterion will be adopted. But if protection is to be given, it should be aimed to provide maximum effect. Hence, surely it should be applied first and foremost to the commercial sector that processes billions of decapods and other possibly sentient invertebrates each year rather than science, which uses relatively trivial numbers and provides the evidence to judge if these species are sentient (Elwood 2011, 2012).

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