Review of: "Behavioral effects of ethanol in the Red Swamp Crayfish (Procambarus clarkii)"

Enrique M Rodríguez[1]

1 University of Buenos Aires

Potential competing interests: The author(s) declared that no potential competing interests exist.

This is a well-developed study; quite clear results are shown, which reveals that the selected species, the crayfish *Procambarus clarkii*, is a sensitive and responsive model to be used for testing the effects of ethanol, and likely of similar substances, on its locomotor behavior. For further experiments, though, I recommend a better standardization of the biological material to be used, i.e., according to that mentioned in the Discussion (item 4.5, 2nd paragraph), the first experiment seems to be run with late juveniles, while early adults were used for the second experiment. The same stage of the life cycle would warrant a better comparison of results among different experiments, and therefore more robust conclusions.

My main concern has to do with the possible extrapolation of the results obtained from an invertebrate species, like the crayfish, to vertebrate species, particularly humans. Experiments made with rats or monkeys (mentioned several times in the manuscript) have indeed more direct implications for humans, since the structure of their nervous system has a high degree of similarity with that of humans. However, the nervous system of a crayfish is very far from that of vertebrates, in terms of its structure, neurotransmitters, and other neurophysiological aspects. For instance, the muscle innervation of crustaceans is dual, given by both excitatory neurons, secreting glutamate as neurotransmitters, and inhibitory neurons, which release GABA. This innervation pattern is quite different from that of vertebrates, which also have a cholinergic, excitatory innervation to skeletal muscles. Therefore, the imbalances caused by a substance like ethanol on the nervous control of locomotion should not be *a priori* expected to be the same between invertebrate and vertebrate species. I think that this whole picture should be taken into account by the authors, who should also discuss the last findings of the effect of ethanol on neurotransmission, for a better interpretation and extrapolation of the results shown in crayfish.