

Letters to the Editor

The effect of vinegar on discharged nematocysts of *Chironex fleckeri*

We are writing because we have serious concerns about the statistical analyses and data interpretation reported by Welfare, Little, Pereira, and Seymour.¹ The authors state in the Abstract, “Part 1: There was a 69 +/- 32% ($F = 77, P < 0.001$) increase in venom discharge after vinegar was applied compared to post electrical stimulation.” The recovery of venom protein from a membrane after the application of vinegar subsequent to electrically stimulating tentacle cnidae to discharge, W4, was compared with protein recovered post stimulation in a saline wash, W3. Figure 2 shows W3 to be approximately 23 +/- 20%. While the authors imply the statistical difference between “venom discharge after vinegar was applied compared to post electrical stimulation”, or W4 vs W3, only the overall ANOVA significance comparing all four treatments was quoted ($F = 77, P < 0.001$) and no statistical significance was provided for this specific W4 vs W3 comparison. If we assume that standard errors of the means (SEMs) were used in Figure 2, a simple *t*-test will provide a *P*-value of only 0.11, comparing W4 and W3, an insignificant finding. The comparison of W4, post electrical stimulation to W1 the pre-stimulation control would yield a significant value of $P < 0.001$ but this is hardly surprising and intuitively obvious.

For this and the following reasons, it seems that the data may not have been properly analysed and not properly presented:

- The same three samples seem to have been used for W1–W4, resulting in internally matched samples, but the data were analysed using ANOVA, assuming samples in different treatments are all independent.
- It is not clear what the value after the “+/-” represents, CI, SEM, or SD, as this is not stated in the caption.
- It is not clear what the 3 x 82 subscript means for the reported $F = 77.12$ (page 32, right column, line 2 below Table 1).

We respectfully recommend that the editors engage a third-party statistician to run an independent analysis of the primary data. If these statistical errors exist, we suggest that the publication be retracted.

It is troubling that this small study reporting recovery of cytolytic activity from a placental membrane proxy of envenomation has been used to launch wildly extrapolative press releases despite over 40 years of using vinegar as a first-response treatment without a clear case of death resulting from its use. Statements such as “For decades experts have recommended vinegar to treat box jellyfish stings. Now, Queensland researchers have discovered the cure can kill”² are simply not true; there was no death or killing in the Welfare et al study.

Claims that “Vinegar may kill rather than cure victims of box jellyfish stings ... The remedy, used for decades to treat

stings, causes up to 60 per cent more venom from the lethal jellyfish to be discharged into the victim”³ are also not supported by these data. There were no ‘victims’ and the slight elevation in the amount of protein recovered in W4 vs W3 was not statistically significant. The authors also report that “vinegar promotes further discharge of venom from already discharged nematocysts” but data show only modest enhanced recovery of cytolytic activity from the membrane, not the degree of cnidae discharge. Finally, the authors do not consider alternative potential causes of enhanced cytolytic recovery, e.g., vinegar improves recovery of certain venom component activities. Thus, these findings may suggest the diametric opposite to the authors’ conclusion – that is, vinegar may enhance venom extraction from a sting site and thus increase survival. However, without validation of this membrane model in an authentic animal model, there is no clear way to interpret these data let alone extrapolate to make emergency medical care recommendations.

References

- 1 Welfare P, Little M, Pereira P, Seymour J. An in-vitro examination of the effect of vinegar on discharged nematocysts of *Chironex fleckeri*. *Diving Hyperb Med*. 2014;44:30-4.
- 2 Ross J. Vinegar doesn’t go with box jellyfish stings. *The Australian*. 2014 Apr 8, 2013. Higher Education. Available from: <http://www.theaustralian.com.au/higher-education/vinegar-doesnt-go-with-box-jellyfish-stings/story-e6frgcjx-1226877298034>.
- 3 AAP. Vinegar on jellyfish sting can be deadly. *The Sydney Morning Herald*. 2014 Apr 9, 2014. Available from: <http://www.smh.com.au/technology/sci-tech/vinegar-on-jellyfish-sting-can-be-deadly-researchers-20140408-zqs8b.html>.

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Reply:

We thank Yanagihara and Chen for their comments and for the opportunity to further the discussion.¹ Our statistician has re-examined (and reanalysed) these data, and we have supplied our data to an independent statistician (who supported our subsequent re-analysis) and are more than willing to supply these data to the journal editors should they feel this is necessary. Furthermore, the manuscript was independently reviewed by two reviewers who expressed no concern over our analysis. We are confident of our results.