Purpose: to investigate possible interactions between H$_2$O$_2$, Aquaporins, and Ca$^{2+}$ channels.

Background: Honey (largely through osmotic stress) is antimicrobial- no discussion why it does not have the same effect on host cells. Honey also produces H$_2$O$_2$ which, may be antimicrobial or have an effect on gene expression. Aquaporins are known to facilitate passive diffusion of H$_2$O$_2$. Aquaporins can also influence Ca$^{2+}$ levels. Ca$^{2+}$ to regulate several processes involved in wound healing.

Results:

- Showed that honey could trigger Ca changes. The levels and profile was dependent on the source of honey and the source of the calcium is extracellular.
- Honey induced micromolar levels of H$_2$O$_2$. Catalase blocks the Ca$^{2+}$ response indicating that it is a function of H$_2$O$_2$.
- Pharmacetical and genetic (siRNA) inhibition of TRPM2 also inhibited the Ca response to honey.
- Pharmaceutical inhibition of Orail also diminished the Ca response to honey.
- Inhibition of both types of channels completely abrogated the Ca response.
- Examination of aquaporin expression found that AQP3 was upregulated by honey. siRNA for AQP3 could block the Ca increase elicited by honey.

Comments

- This study would be strengthened by the authors directly demonstrating H$_2$O$_2$ production by honey rather than relying on catalase. It is possible that H$_2$O$_2$ may be indirectly acting. Also, since the observed effects were dependent on the type of honey- does this correlate with H$_2$O$_2$ production?
- Fig 5a: Individual lanes are not identified in the immunoblot (C is probably control and M is probably honey).
- The authors should note that the keratinocyte scratch assay may have little or nothing to do with in vivo wound healing. Was “wound closure” a function of increased proliferation or migration?

Conclusion: This is a nice study with the results largely supporting the questions asked. This reviewer is a little troubled by the equating of the scratch assay with wound healing and it would be nice to see direct measurement of extracellular H$_2$O$_2$. The English writing could also be improved but at present does not significantly detract from the study.