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Investigating the Feasability of a Sum Frequency/Second Harmonic Generation Experiment to Examine the Behavior of RNA on Ice

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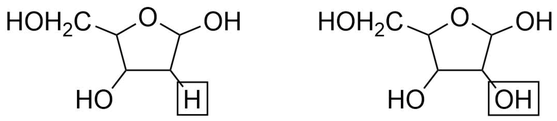
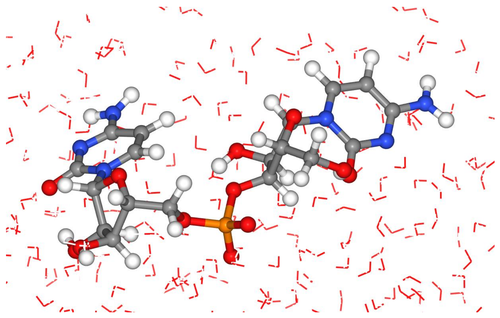
BACKGROUND

- The RNA world hypothesis asserts that life began with RNA, or ribonucleic acid. But a problem with RNA world theory is that RNA hydrolyses too quickly in water at room temperature.[1,2]



2-Deoxyribose Ribose

- This has led to a new hypothesis, the RNA world on ice hypothesis, that life began on the surface of ice.[2]



- Low temperatures are known to inhibit RNA hydrolysis.[2]
- A quasi-liquid layer (QLL) exists on the surface of ice at temperatures as low as -60 Celsius.
- **The guiding question of this work is: is it feasible to study experimentally the ability of RNA on ice to carry out essential life functions (self-replication and catalysis)?**

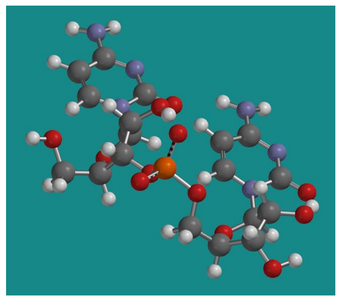
METHODS

- Sum frequency generation, or SFG, is a surface sensitive spectroscopic technique that observes the vibrational transitions of specific structures.[3]
- Second harmonic generation, or SHG, is also a surface sensitive spectroscopic technique that tells you about the electronic structure of a molecule.[4]
  - SFG and SHG are good candidates to further the study of RNA on ice because the techniques have already been used to study ice surfaces and the structures of surface bound proteins.
- Because these are surface sensitive techniques, they offer the possibility of investigating the **orientation of RNA on ice**, which is a first step towards addressing the larger guiding question.

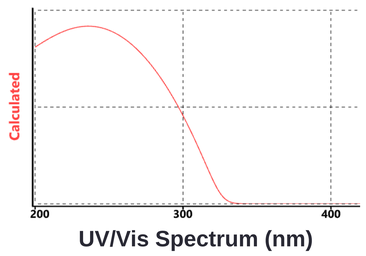
1. What can SHG and SFG tell us about the orientation of RNA on ice?
2. What are the obstacles to performing these experiments?

- I will attempt to answer these questions using software like Spartan (molecular modeling) and Gromacs (molecular dynamics).

SHG AS A POTENTIAL PROBE



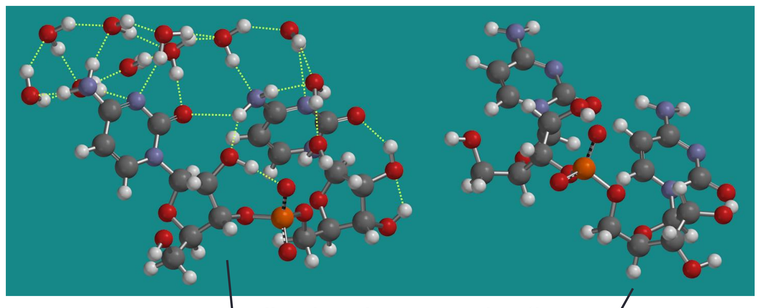
- This is a CC dinucleotide RNA strand that is the focus of this study. Below is a simulated UV/Vis spectrum generated in Spartan.

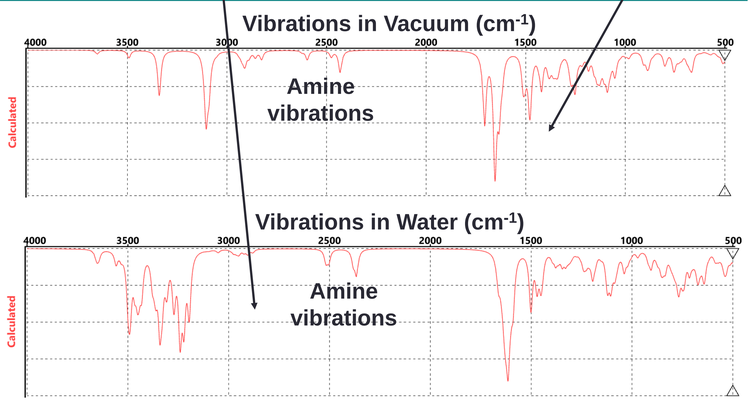


- Because RNA does not absorb at the right wavelength (400 nm) a chromophore must be attached to study with SHG.

SFG AS A POTENTIAL PROBE

- For SFG we compared the structure in water (left) to the strand in vacuum (right).

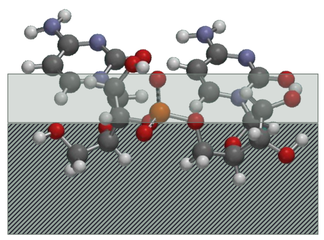


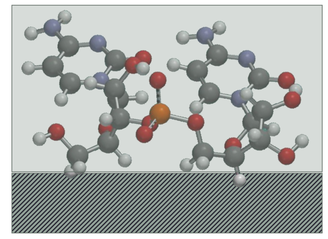


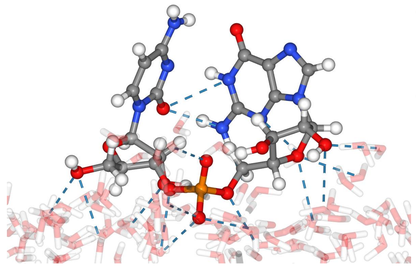
- Deuterating the amines moves the vibrational bands to an uncongested part of the spectrum that will be easy to detect with SFG.
- When the amines are in water, the vibrational bands are red-shifted by about 100 wavenumbers.
- This is a good probe to see if the amines are submerged in water or if they are in air
- Conclusion – SFG seems to be a more promising technique than SHG.

WHAT SFG & MD COULD TELL US

- Are parts of the RNA submerged in the quasi-liquid layer (QLL) or not?







- Which parts of the RNA are submerged in the QLL?
- We can use MD (molecular dynamics) to help us interpret the SFG.

FUTURE WORK

- Continue troubleshooting and work on furthering Gromacs calculations of an SFG spectrum
- Start compiling materials to hold ice for a cryo-SFG experiment, such as a temperature-controlled sample stage.
- Study the rate at which the deuterium will switch with the surrounding water.

REFERENCES

[1]Gilbert, W., Origin of life: The RNA world. *Nature* **1986**, *319*, 618.

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[3]Medders, G.; Paesani, F., Dissecting the Molecular Structure of the Air/Water Interface from Quantum Simulations of the Sum-Frequency Generation Spectrum. *J. Am. Chem. Soc.* **2016**, *138*, 3912-3919.

[4]Birman, Y.; et. al, Second-harmonic generation-based methods to detect and characterize ligand-induced RNA conformational changes. *Methods*, **2019**, *167*, 92-104.

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