

# Fluorescence Lifetimes of Retinol in Different Lipid Phases

Laura Smith

Dr. Jay Nadeau

Dr. Drake Mitchell

Louis Sumrall

2022 REU Portland State University Symposium, August 12, 2022



# Overview

- Introduction
  - Retinol's Properties
  - Liposomes and Retinol Encapsulation
  - FLIM and FLS
- Methods
- Results
- Discussion
- Conclusions

# Retinol's Properties

- Retinol, or Vitamin A, has important pharmaceutical and cosmetic applications
- It degrades readily when exposed to light or oxygen
- It's fluorescent

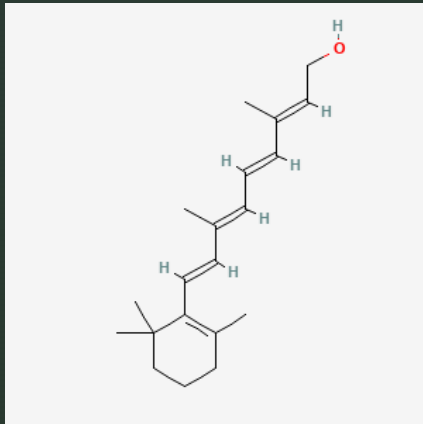


Image credit from PubChem



Image credit from Neutrogena

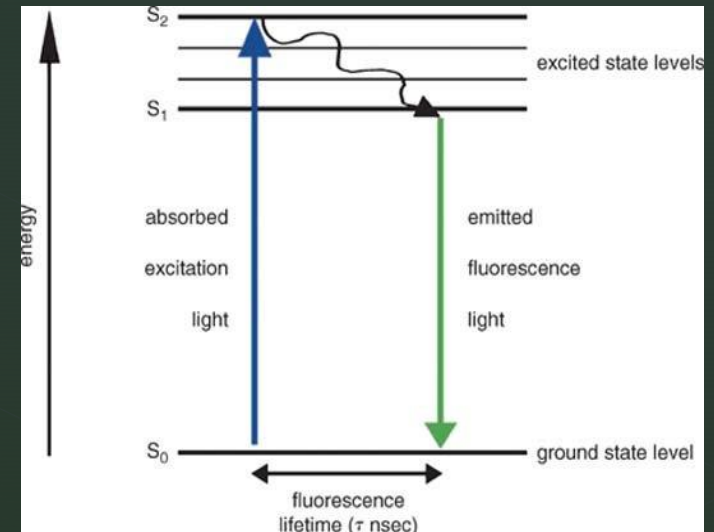


Image credit from Research Gate

# Liposomes and Retinol Encapsulation

- Liposomes are vesicles that consist of lipid bilayers
- Encapsulation minimizes oxygen and light exposure preventing degradation

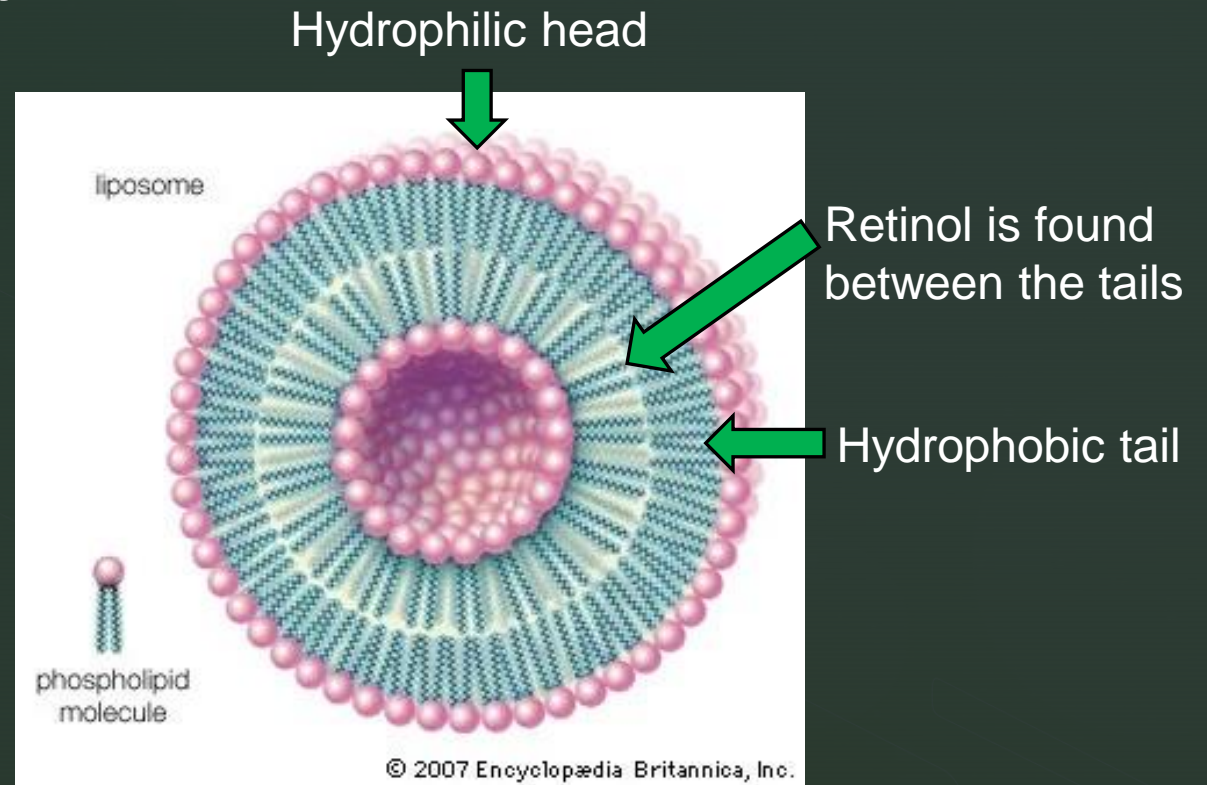


Image credit from Britannica

# FLIM and FLS

- FLIM images on the microscopic scale to determine the fluorescence lifetime of specific regions of the liposome
- FLS determines bulk time resolved measurements of liposomes in an aqueous solution

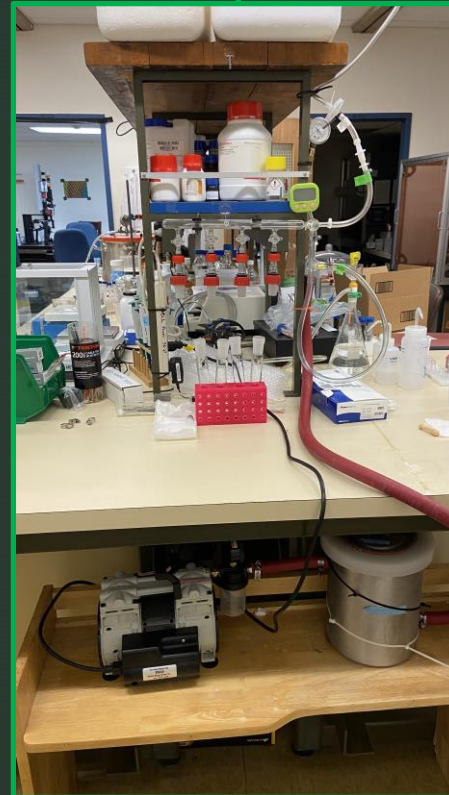
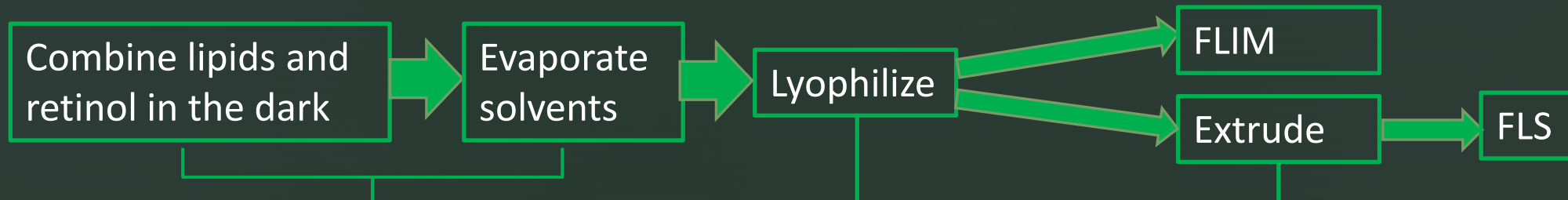


Image credit from OHSU

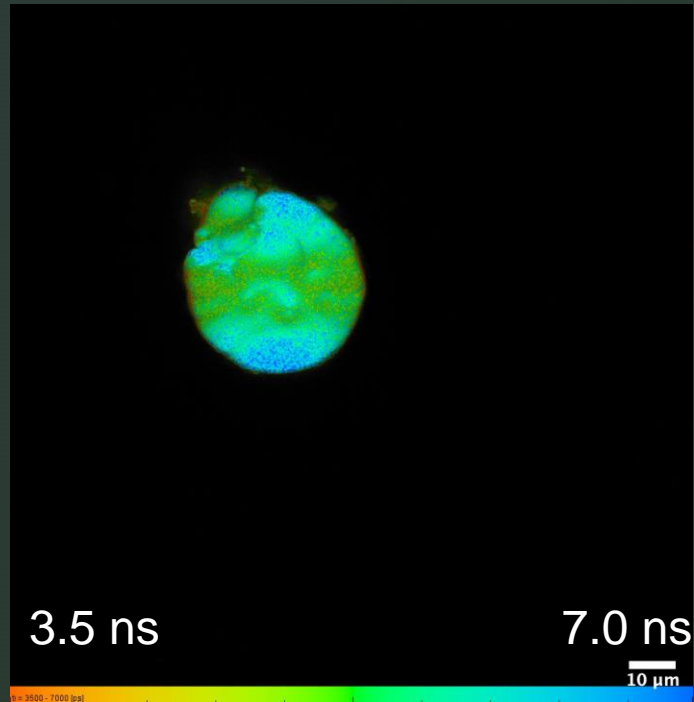


Image credit from ISS

# Methods

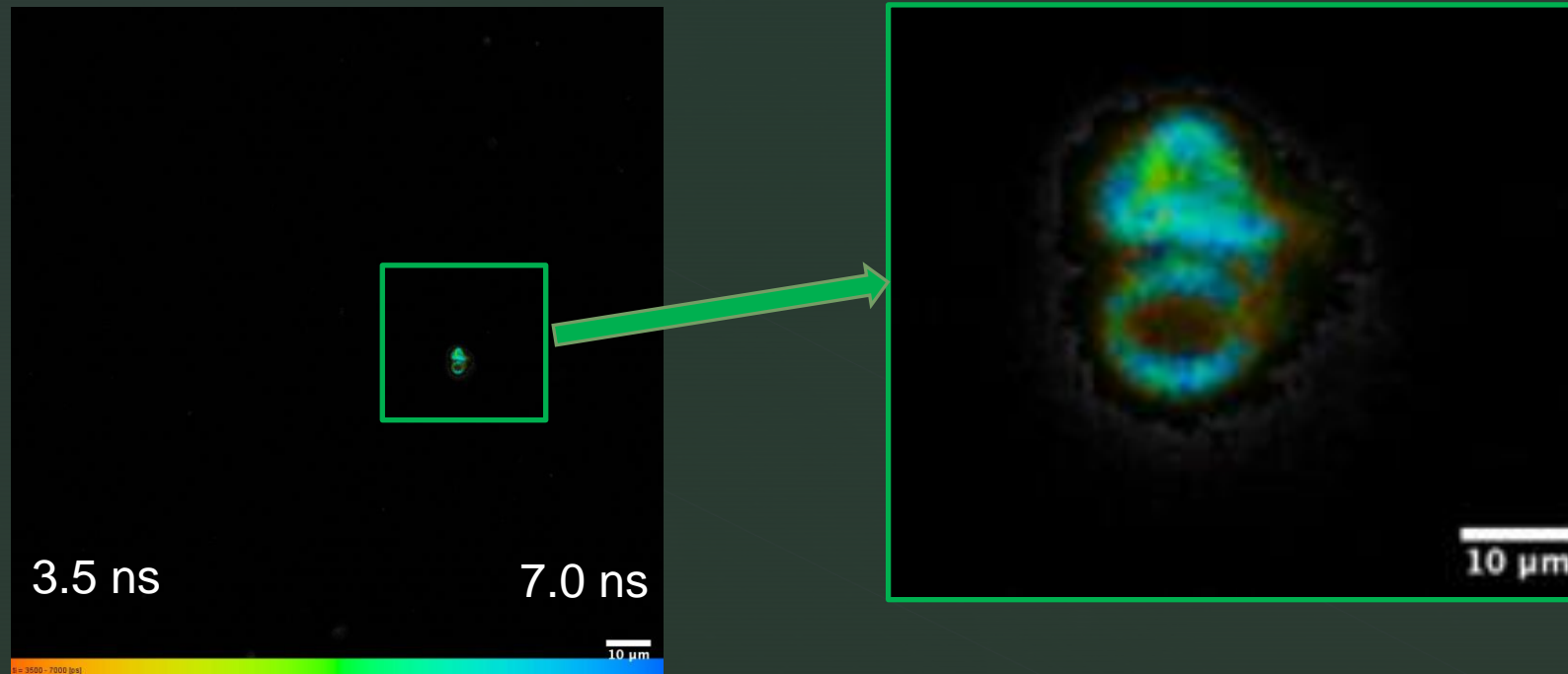


# FLIM Measurement of POPC Liposome #1



- Lifetimes ranged from 6.3 to 8.0 ns based on ROIs
- There were small variations in lifetime amongst the regions
- Consistent lamellarity

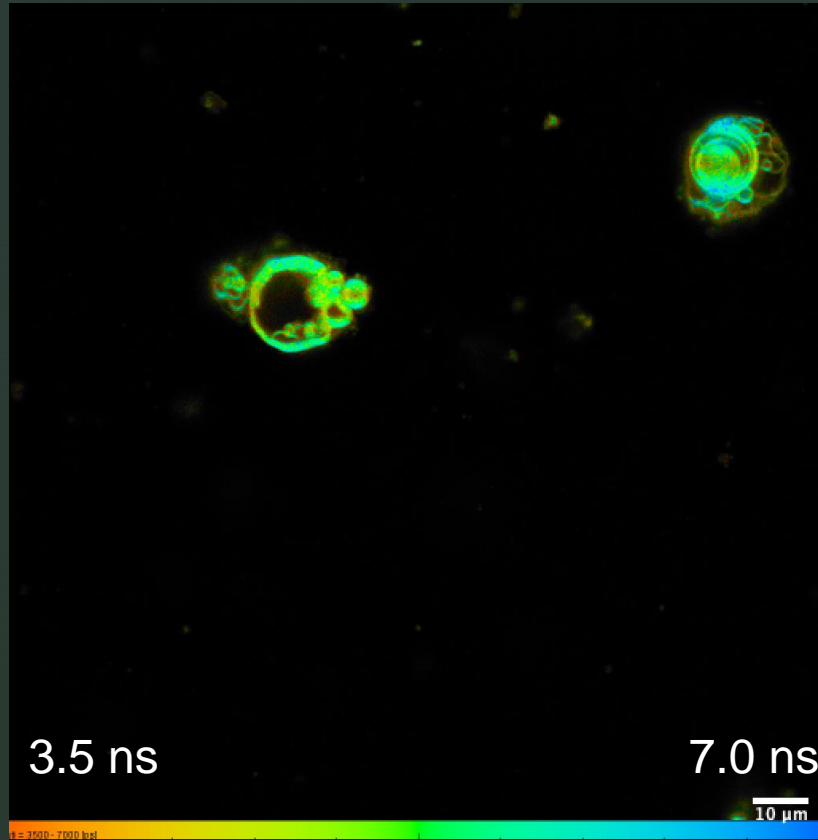
# FLIM Measurement of POPC Liposomes #2



- Lifetimes ranged from 6.8 to 7.2 ns
- There were minimal variations in lifetime amongst the regions
- Inconsistent lamellarity

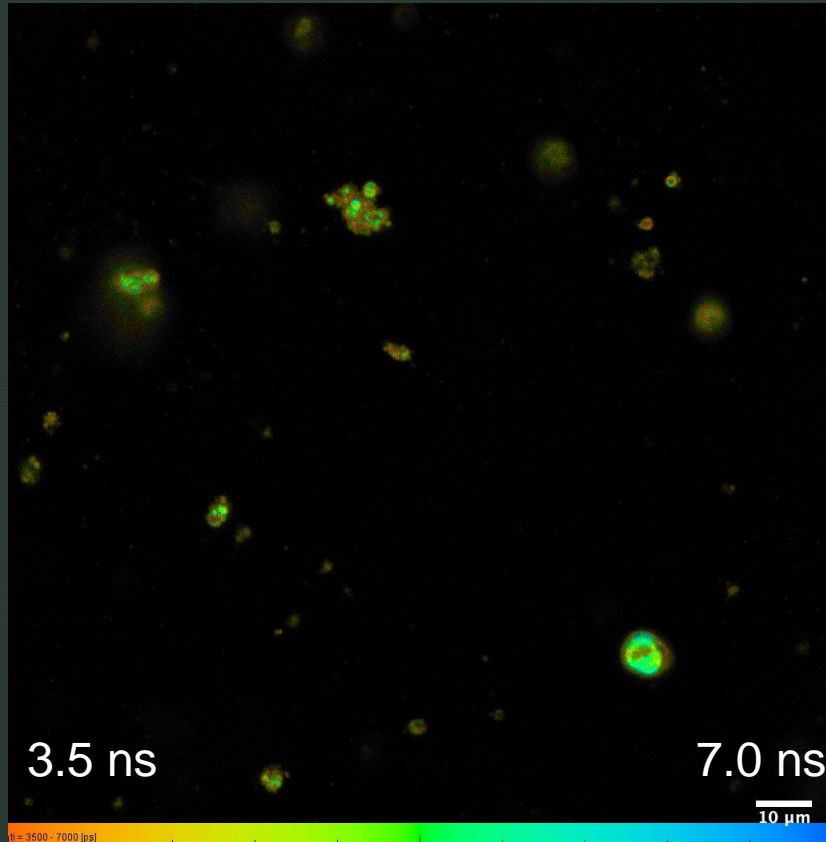


# FLIM Measurement of POPC Liposomes with 10 molar percent cholesterol #1



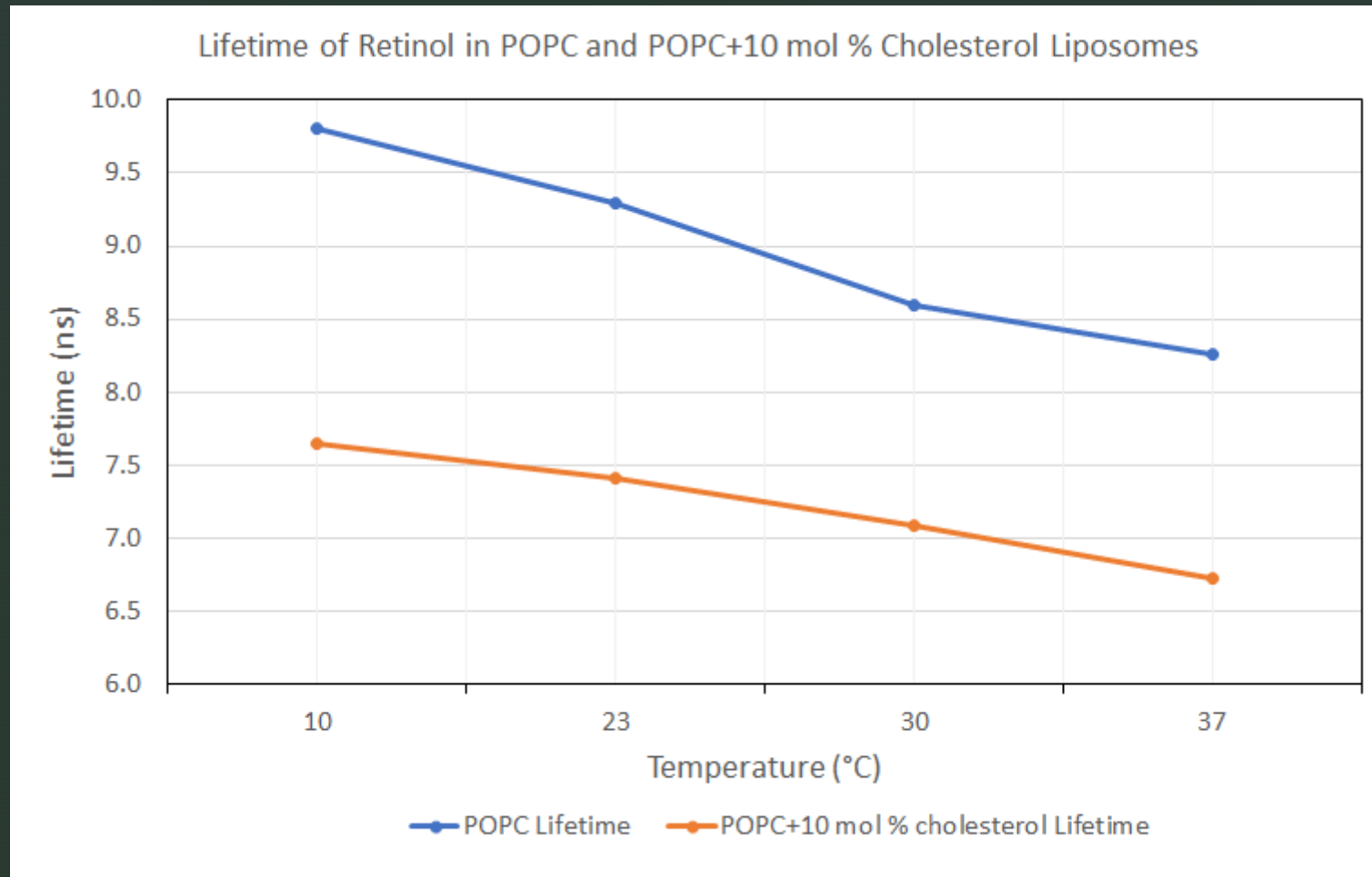
- Lifetimes ranged from 6.8 to 7.1 ns based on ROIs
- Lamellarity was inconsistent in the liposome to the left

# FLIM Measurement of POPC Liposomes with 10 molar percent cholesterol #2



- Lifetimes ranged from 6.2 to 6.8 ns based on ROIs
- Consistent lamellarity
- Size of the liposomes varied

# FLS Measurement



- As temperature increased, lifetime decreased

# Discussion

- Fluorescence lifetimes were greater for POPC liposomes from both FLIM and FLS measurements
- As temperature increased, lifetimes decreased
- Careful handling of retinol contributed to improved results

# Conclusion

- Stability of retinol was improved in the POPC liposomes more than in the POPC+10 mol % cholesterol liposomes
- As disorder in the lipid bilayer increases, stability decreases based off these preliminary results
- Further work with different amounts of cholesterol and lipids with saturated fatty acid tails are recommended

# References

1. Kafi, R. et al. Improvement of naturally aged skin with vitamin A (retinol). *Archives of dermatology* 143, 606-612 (2007). <http://www.ncbi.nlm.nih.gov/pubmed/17515510>
2. Varani, J. et al. Vitamin A antagonizes decreased cell growth and elevated collagen-degrading matrix metalloproteinases and stimulates collagen accumulation in naturally aged human skin. *The Journal of investigative dermatology* 114, 480-486 (2000). <http://www.ncbi.nlm.nih.gov/pubmed/10692106>
3. Fu, P.P. et al. Photoreaction, phototoxicity, and photocarcinogenicity of retinoids. *Journal of environmental science and health. Part C, Environmental carcinogenesis & ecotoxicology reviews* 21, 165-197 (2003). <http://www.ncbi.nlm.nih.gov/pubmed/15845224>
4. Cho, H. S. et al. Lipid Domains in Bicelles Containing Unsaturated Lipids and Cholesterol. *The Journal of Physical Chemistry B* 114, 9238–9245 (2010).
5. Lee, S.C. et al. The effect of cholesterol in the liposome bilayer on the stabilization of incorporated retinol. *Journal of Liposome Research* 15, 157-166 (2005). <https://doi.org/10.1080/08982100500364131>
6. Chen, W. et al. Determination of the Main Phase Transition Temperature of Phospholipids by Nanoplasmonic Sensing. *Sci Rep* 8, 1-11 (2018). <https://doi.org/10.1038/s41598-018-33107-5>
7. Raghunathan, V. A., and John Katsaras. "L  $\beta$  '  $\rightarrow$  L C ' Phase Transition in Phosphatidylcholine Lipid Bilayers: A Disorder-order Transition in Two Dimensions." *Physical Review E Phys. Rev. E*: 4446-449. (2015). <https://journals.aps.org/pre/abstract/10.1103/PhysRevE.54.4446>
8. Radda, G.K. & Smith, D.S. Retinol: A fluorescent probe for membrane lipids. *FEBS Lett* 9, 287-289 (1970). <https://www.ncbi.nlm.nih.gov/pubmed/11947694>
9. Imanishi, Y. & Palczewski, K. Visualization of Retinoid Storage and Trafficking by Two-Photon Microscopy. *Methods in Molecular Biology* 652. 247-261 (2010). <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4607074/>

# Acknowledgements

- The Author's participation in this project was supported by the National Science Foundation's *Application of Microscopy and Microanalysis in Multidisciplinary Research* REU program at Portland State University (#1851851). It was also supported through guidance from Dr. Jay Nadeau, Louis Sumrall, Dr. Drake Mitchell, Dr. Elmukhtar Alhatmi, Dr. Jun Jiao, and Dr. Erik Sánchez.

