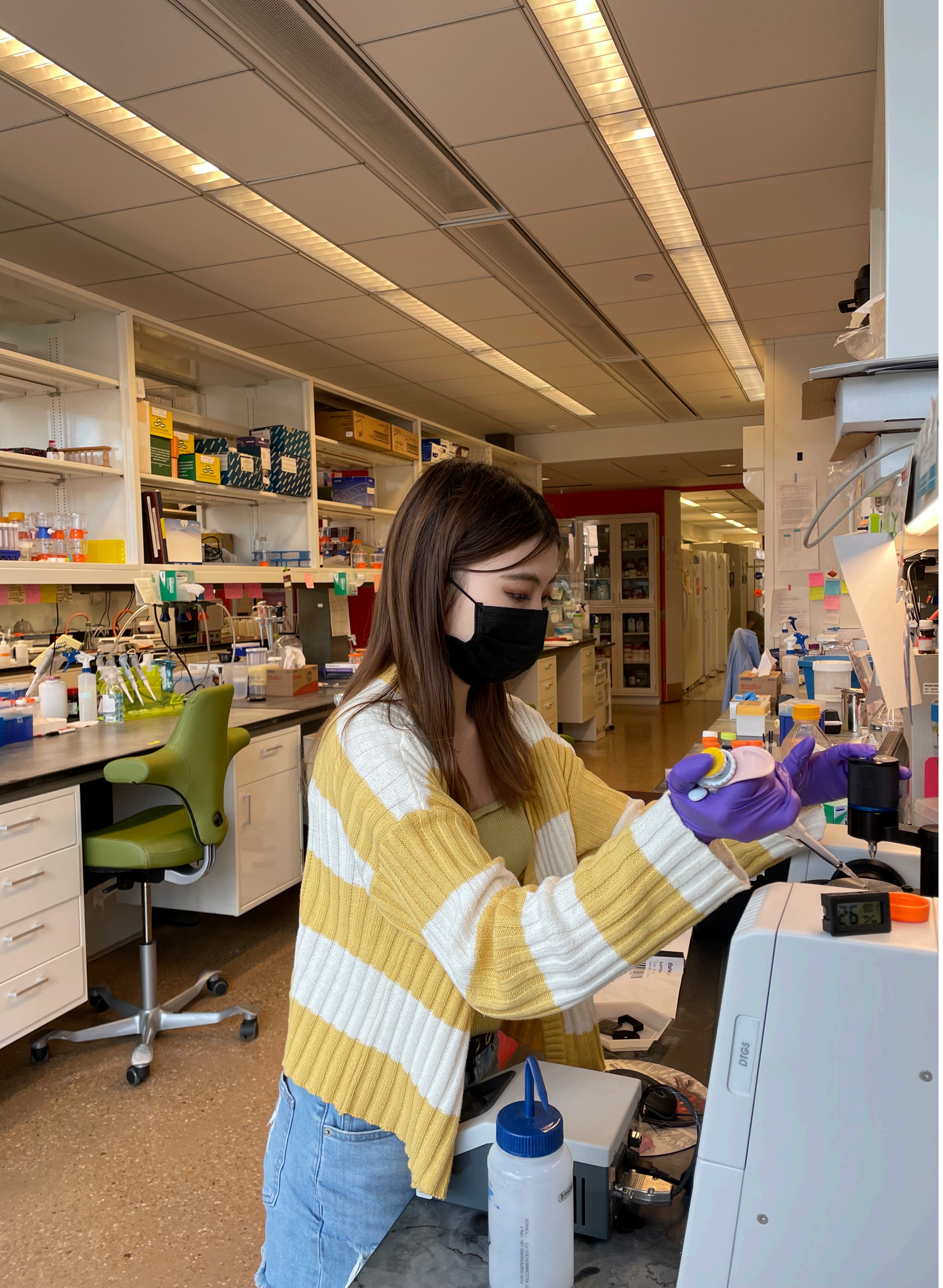

Effect of pH on CAHS D's Secondary Structure Using FTIR Spectroscopy

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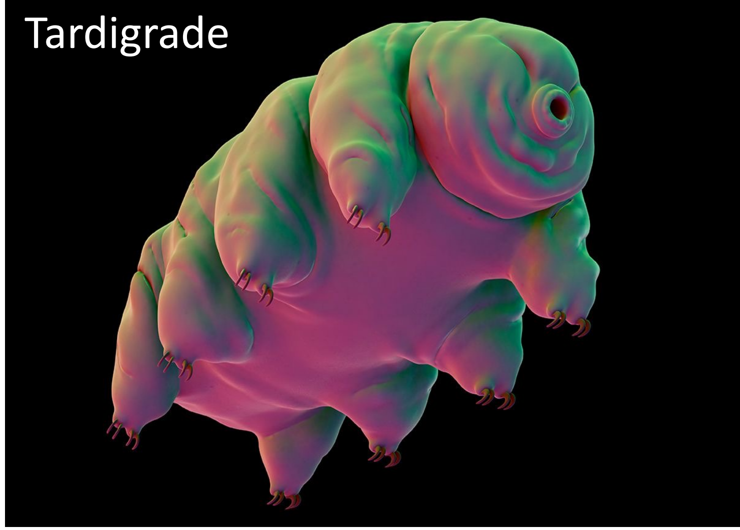
Research Question

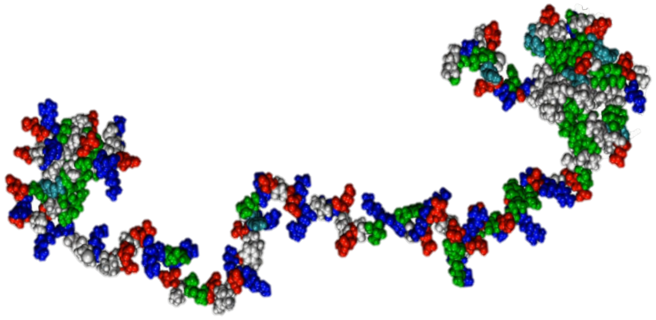


Tardigrades are famous for their ability to survive extreme conditions such as complete desiccation. Cytosolic abundant heat soluble proteins (CAHS), a type of tardigrade disordered protein, is essential for desiccation survival. Our Lab has found that purified CAHS D undergo gelation, which I hypothesize is stabilized by intramolecular interactions involving transient formation of secondary structure.

I investigated how pH impacts the secondary structure of CAHS D.

Low (10 g/L) and high (40 g/L) concentration samples of CAHS D were prepared in buffer at pH 5.5 and 8.0. Their secondary structures were measured and compared by using Attenuated Total Internal Reflectance Fourier Transform Infrared (ATR-FTIR) spectroscopy. CAHS D's N-terminal region contains histidine residues, whose pK_a is between 5.5 and 8.0. The repulsive and attractive interactions of histidine depend on protonation state. **Understanding the structure of CAHS D gels will aid in our understanding of its function.**

Molecular Dynamics simulation of CAHS D



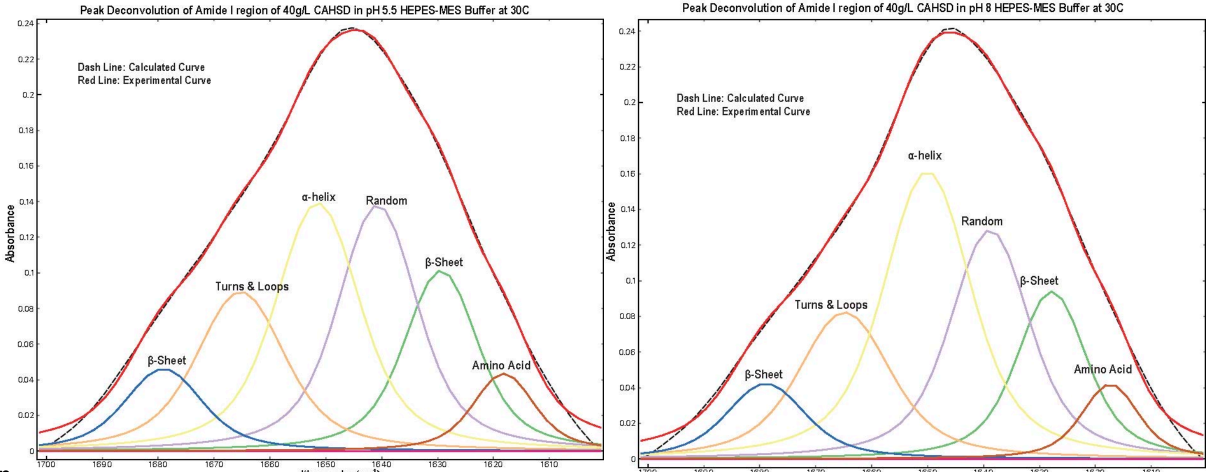
Results

- Experiments were performed in triplicate. Results are presented as averages. Uncertainties are presented as the standard deviation of the mean. At pH 8, 40 g/L CAHS D has 5±1 % more α-helix, 1±1% fewer turns and loops, 3.3±1.0% less random structures, and between 0.2±0.3% and 1.2±0.9% less β-sheet than at pH 5.5. I conclude that deprotonation of histidine increases the percentage of α-helix. (Spectra of the 10 g/L sample had a low signal to noise and was not analyzed).

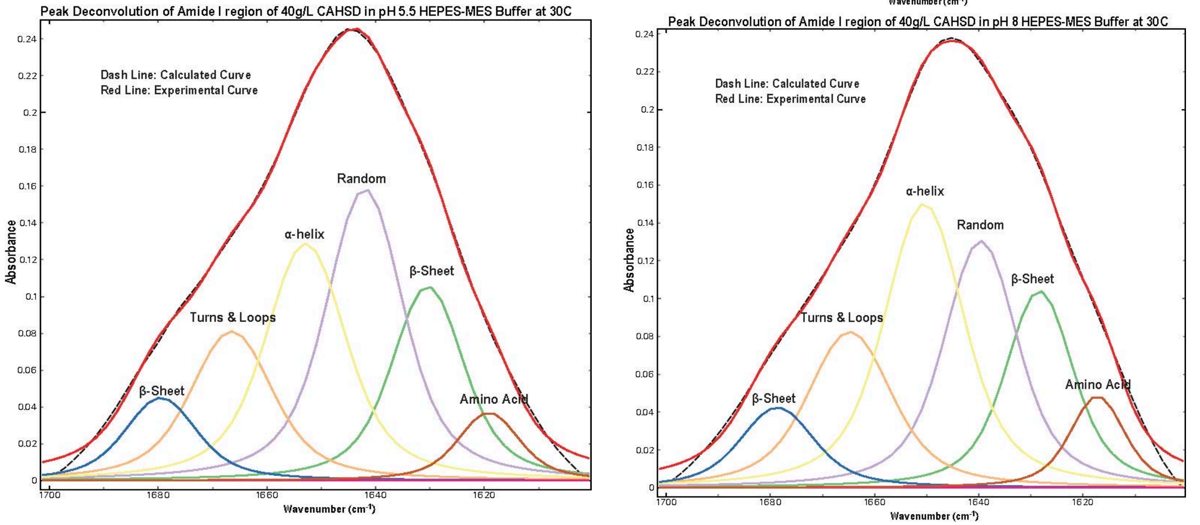
- The data shed light on the pH dependence of CAHS D's secondary structure. In the future research, I will expand the pH range. Investigating the structural rearrangement will aid in our understanding of CAHS D function in desiccation tolerance. Continued study of CAHS D is useful in real-world applications such as the manufacture of stress-tolerant crops and stabilizer for vaccines.

pH	Concentration (g/L)	Low frequency β -sheet (%)	Random coil (%)	Turn and Loops (%)	α-helix (%)	High Frequency β-sheet(%) (another)	Amino Acid (%)
5.5	40 ±2	17.8 ±0.5	27 ±1	16.6 ±0.6	25.2 ±0.7	7.5 ±0.3	5.9 ±0.3
8.0	40 ±2	16.6 ±0.8	23.7 ±0.4	16.0 ±0.8	30.3 ±0.9	7.3 ±0.1	6.1 ±0.3

First Run



Second Run



Third Run