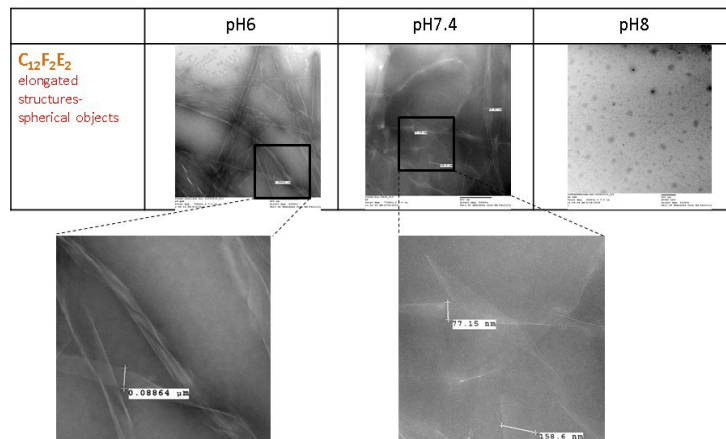


**PRF#: 57434-DNI7. Project Title:** The Evolution of Self-Assembled Organic Materials

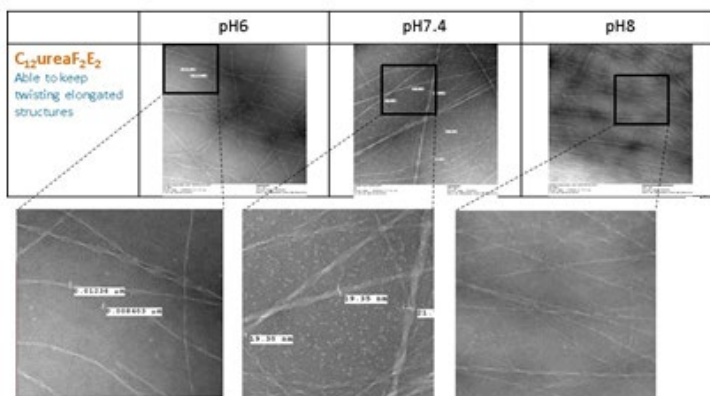
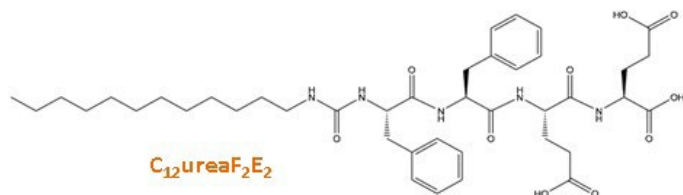
**PI:** Martin Conda-Sheridan, University of Nebraska Medical Center

The goal of this project is to synthesize petroleum –based molecules that mimic traditional peptide amphiphiles (PAs). We hypothesize these compounds will still self-assemble into supramolecular structures. Although the (final) proposed compounds were not prepared due to setbacks with the chemistry, we made tremendous progress in our understanding of the synthesized molecules and the key parameters that dictate the shape of the nanostructures they form. We are currently writing a paper and are expecting to soon start writing two additional manuscripts, all focused on the material behavior of these new nanostructures. Because of these future papers, the ACS-PRF funding had and will have tremendous impact on my career. Further, securing funding from an external source is seen favorably for my P&T package. In addition, some of the controls we made during the funding period were the basis of a theoretical paper published by our collaborators at the University of Buenos Aires-Argentina showing the international dimension this grant gave to my research group (<https://doi.org/10.1021/acs.jpcc.9b04280>).

Regarding the project, we found the inclusion of an urea motif near the lipid region of the molecules leads to more stable nanostructures. We believe this is related to the presence of this divalent hydrogen-bonding group. For example, the molecules  $C_{12}F_2E_2$  (**Figure 1**) and  $C_{12}ureaF_2E_2$  formed nanostructures as seen by transmission electron microscopy (TEM) (**Figure 2**) and atomic force microscopy (AFM, not shown) at pHs 6 and 7.4 but only the urea-containing one showed stability at higher pHs.

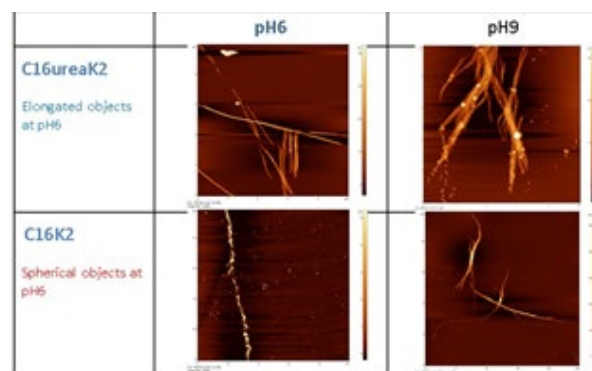


**Figure 1.** TEM micrographs of the nanostructures formed by  $C_{12}F_2E_2$  at different pHs. High pHs promote formation of spheres presumably due to coulombic repulsions.



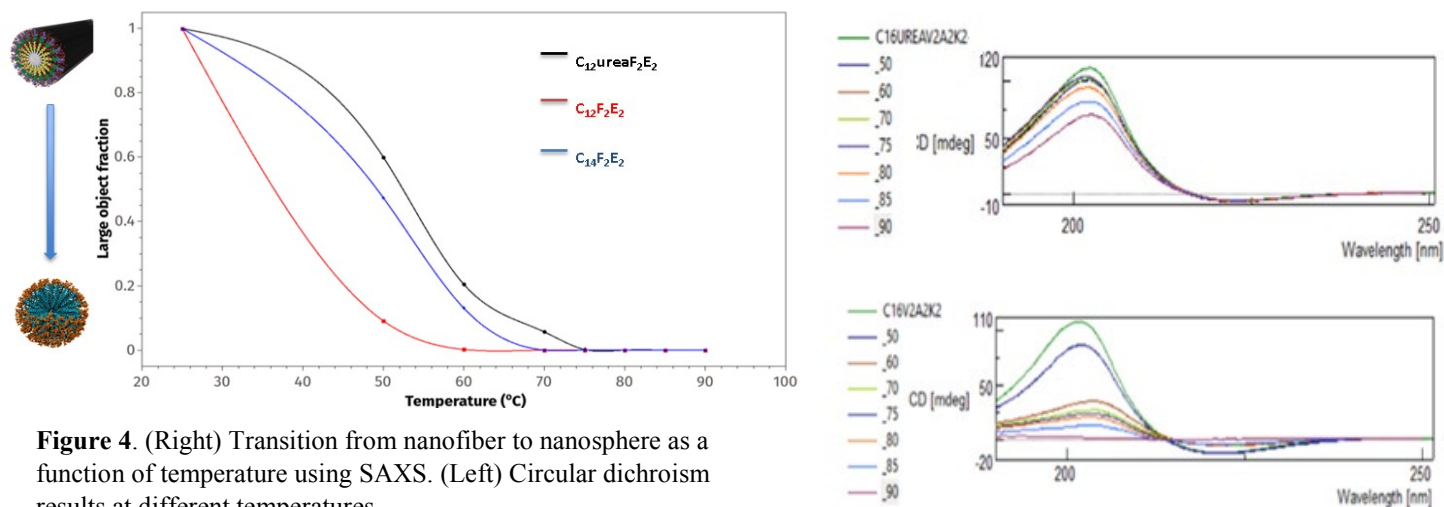
**Figure 2.** Top; structure of small molecule containing an urea functionality. Right; TEM micrograph of the nanostructures formed by this molecule at different pHs. The nanofiber morphology was maintained at the higher pH.

This trend was also observed in “simpler” self-assembling molecules as seen by the AFM micrograph on **Figure 3**, which indicates only urea-containing molecules form nanofiber at pH 6 (the results were confirmed by TEM). The same phenomena was perceived at different temperatures. As can be observed in the small angle X-ray scattering (SAXS) experiment shown in **Figure 4, Right**; compounds with urea were more resistant to transitions from fibers to spherical micelles at higher temperatures. Further, studies using variable temperature circular dichroism (VT-CD) showed that  $\beta$ -sheet signature peaks disappeared first (at lower temperatures) in nanostructures lacking the urea group (**Figure 4, Left**).



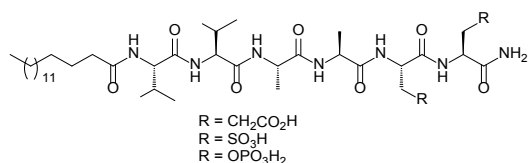
**Figure 3.** Effect of the pH in simple self-assembling

Altogether, we prepared and characterized 5 series of molecules (each containing 3 compounds) and the same trend was observed in all the nanostructures.



**Figure 4.** (Right) Transition from nanofiber to nanosphere as a function of temperature using SAXS. (Left) Circular dichroism results at different temperatures.

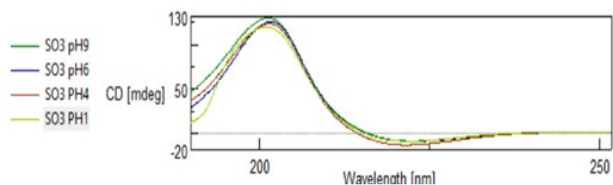
In addition, we studied the effect that groups with diverse pKas have on the self-assembling characteristics of designed nanostructures contain charged groups such as carboxylic acid (pKa ~ 4.7), phosphoric acid (pKa1 ~ 2.1, pKa2 ~ 7.0) and sulfonic acid (pKa ~ -3.0)



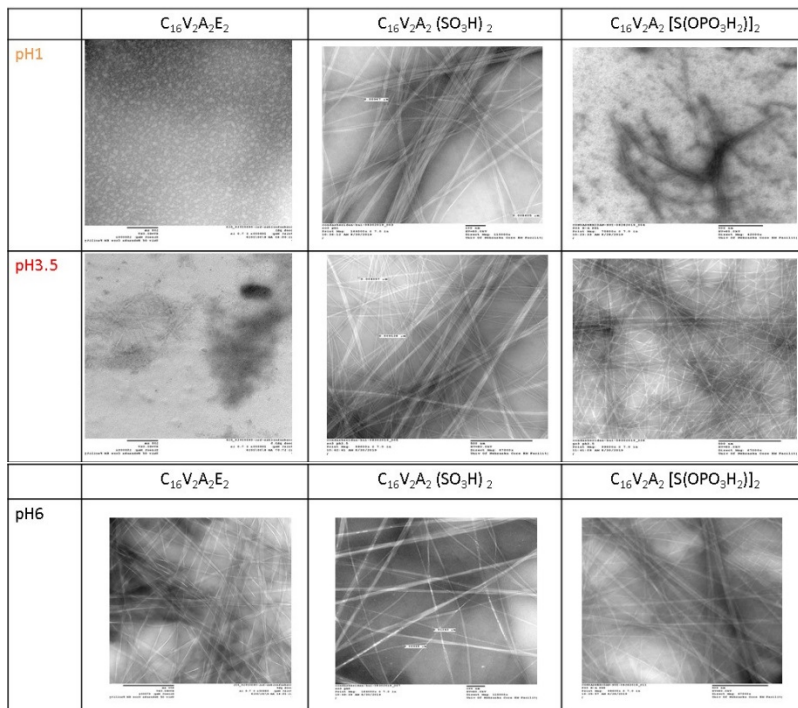
**Figure 5.** Formulas of self-assembling molecules with negatively charged ionizable groups.

(Figure 5).

As can be seen on the TEM micrographs in Figure 6, the supramolecular morphology is interconnected to the pKa of the ionizable group. If the solution pH < pKa, no fiber-like nanostructure is formed, we presume this is related to molecular aggregation. CD studies at various pHs (Figure 7) indicate the presence of a  $\beta$ -sheet region (which can be linked to fiber-like morphologies) is also related to the pKa of the nanostructure. We prepared 6 additional nanostructures and are currently investigating the effect of the pKa on their behavior.



**Figure 7.** CD data indicating  $\beta$ -sheet signature peaks, at diverse pHs, of the sulfone-substituted nanostructure.



**Figure 6.** TEM micrographs of three nanostructures with negatively charged groups of different pKas at diverse temperatures.

In conclusion, the funds of the ACS-PRF helped us to unravel the characteristics and properties of self-assembling systems. Future efforts will focus in removing the peptide bonds and amino acid segments for other chemical functionalities in order to create new self-assembling systems different from the ones derived from nature.

Regarding personnel, a first year graduate student, Huihua Xing, participated in the project. She learned several techniques that are essential for material sciences research including: transmission electron microscopy (TEM), atomic force microscopy (AFM), dynamic light scattering (DLS), and circular dichroism (CD).