

## 2018/05/22 Experiment 235.1 – CaPS\_RACE NMR of Precipitated Human Sepsis Serum

### Supplies

- 5mm 7-inch long NMR sample tubes (Wilmad LabGlass Cat# 528-PP-7)
- 13.5 inch Glass Pasteur Pipets (Wilmad LabGlass Cat# 803A)
- Cryotubes (Fisher 02-912-729 or similar)
- Internal standard: 9.636 mM formate **Check concentration!**

### Procedure:

1. Thaw samples at room temperature, or if samples have been stored at 4°C, bring samples up to room temperature.
2. Do 3-point pH calibration on electrode prior to measuring pH
3. pH samples, **record the initial pH**. Since samples are resuspended in phosphate buffer, the concentration will very rarely need to be adjusted; if the pH is not between 6.5 and 7.5 ( $\pm 0.1$ ) use DCl and/or NaOD (20uL drops with a micropipettor), **record the final pH**.
4. Transfer samples to labeled NMR tubes.
5. Acquire spectra on Varian 500 MHz NMR using “metnoesy temp”  
Array for 90° pulse width, 12ppm spectral width, nt=32, 1 sec relaxation delay, tmix=100ms, select satfrq manually, satpwr=80Hz, satmode=ynyn, receiver gain=6db  
Shims: most current d2o\_shims

Sample	Initial vol (uL)	Initial pH	Acid/base added	Final pH	Tube label	rms error	satfrq	PW 360	PW 90	Formate LW
R_001										
R_002										
R_003										
R_004										
R_005										
R_006										
R_007										
R_008										
R_009										
R_010										

**Internal Standard: 9.636 mM Ca Formate – 50uL**