Shelf Life of Lidocaine

Rural Scholars 2017/2018
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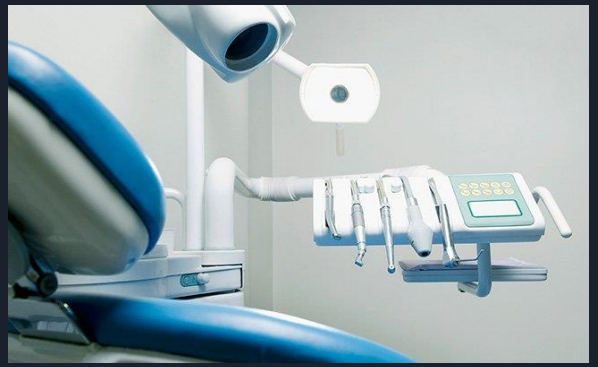
"two-thirds of the time you'll probably fail" - scott shaw

Lidocaine Background

- Lidocaine is a local anesthetic used to numb tissue
  - Ex) used to numb gums at the dentist office
- A buffer is added to resist major changes in the pH
  - The buffer used is $NaHCO_3$
  - Makes the solution less acidic, which is less painful for patient
- Why:
  - We chose this because all of us are going into dentistry or medical fields and lidocaine related to both of these



Societal Impacts

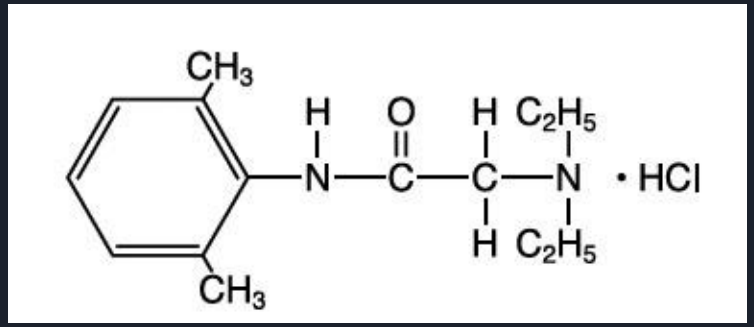
Cost efficiency for clinics that use lidocaine

- Know more about the shelf-life of normal + buffered lidocaine
  - Minimize waste
- Know which conditions maximize the shelf life of each solution
- Gain insight on why the efficiency of lidocaine injections changes between patients

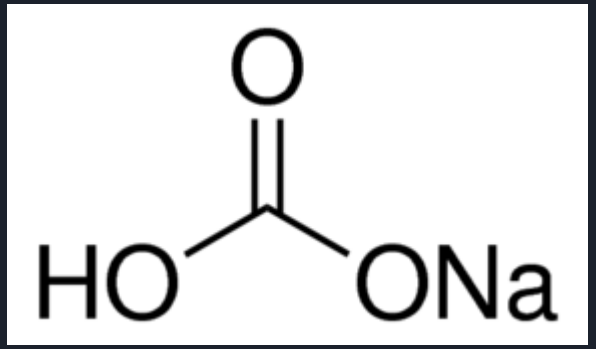
Experimental Question

What factors affect the degradation of lidocaine?

- How fast does degradation occur under each factor?
- Which environment is most suitable to maximize the shelf life of lidocaine?



Lidocaine HCl



$NaHCO_3$ Buffer

Experimental Plan

- Investigating degradation of buffered and unbuffered lidocaine
  - Concentration of lidocaine and epinephrine in solution have been shown to decrease significantly after one week
- Testing in presence of different factors
  - Light
  - Dark
  - Cold
  - Heat
  - Control groups
- Determine what environments affect how long buffered and unbuffered lidocaine lasts

Controls

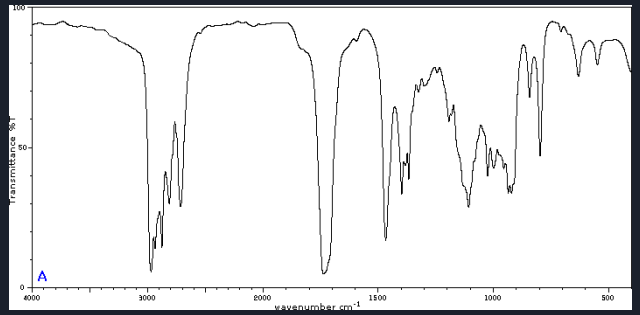
Cold vs Heat

Dark vs Light

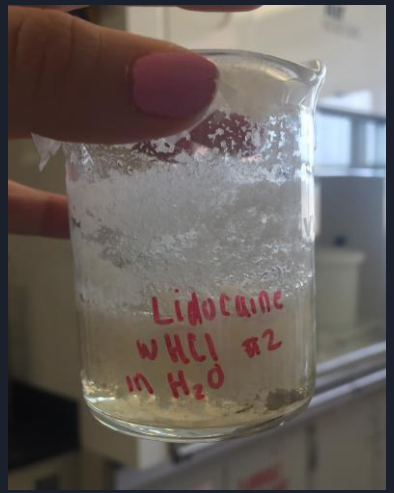
Expected Outcomes

- We predict that the solutions placed in the dark and cold will have the longest shelf life, as compared to those placed in heat or light, which should degrade faster.

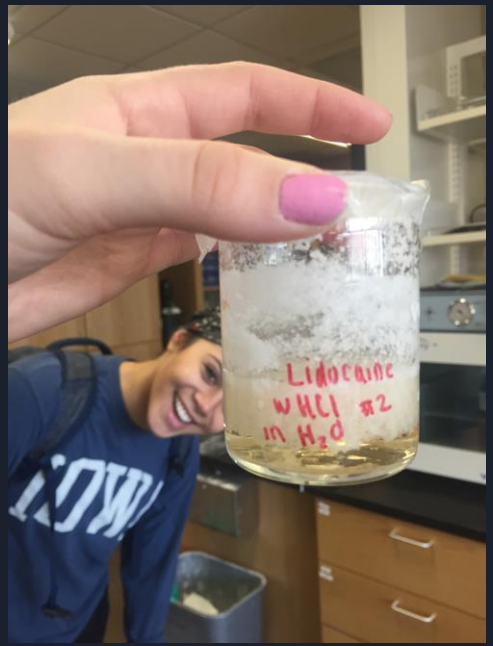
Challenges



- Unfamiliar unit conversions
  - Medical units (mEq - milliequivalent units)
- Inexperience in lab devices
  - Infrared
  - NMR
  - Mass Spectrometry
- Time constraints
- Insolubility of lidocaine and buffer
  - $NaHCO_3$ needed more water to dissolve fully
  - Need to recheck calculations
  - 3 different solutions were made-all were insoluble



Solubility of Sodium Bicarbonate



- Sodium bicarbonate is a salt that is normally very dissolvable in water
- We used 4.2 grams NaHCO3 (8.4%)
- Prediction:
  - First we must dilute the Sodium Bicarbonate so it is 8.4% when mixed with water
  - This diluted version must then be added to the Lidocaine Epinephrine solution in a 1:10 ratio
    - 5 mL diluted Sodium Bicarbonate with 45 mL Lidocaine Epinephrine solution
- Changed experiment plan - did not work for us

Mechanisms

NMR - Nuclear Magnetic Resonance

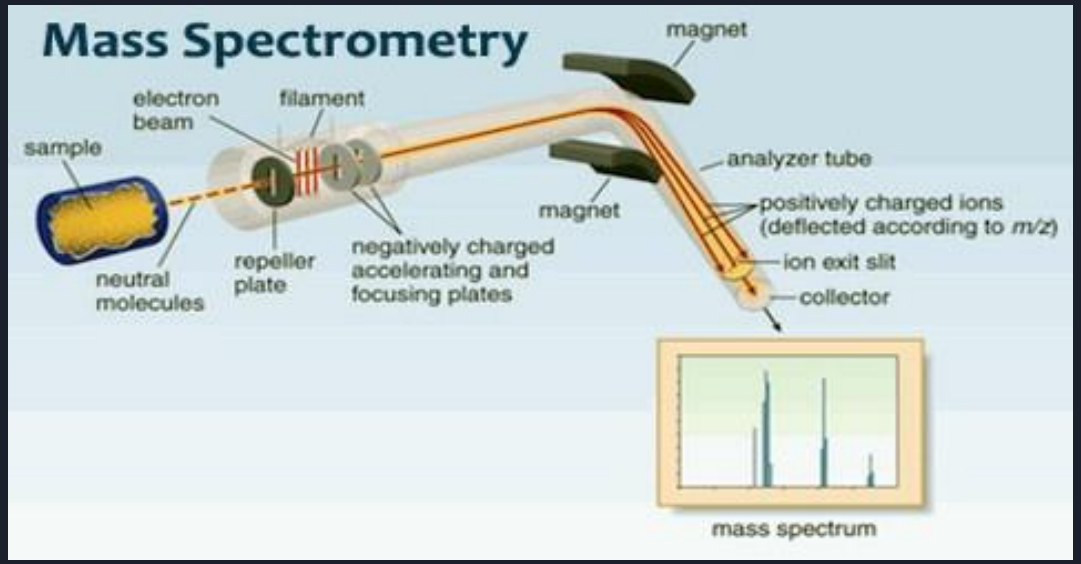
- NMR is used to determine physical and chemical properties of molecules
  - Can show us where hydrogen groups or carbon groups are located in relation to each other
  - Shows the proton environment

IR Spectroscopy - Infrared Spectroscopy

- IR uses infrared light to determine the functional groups present in a molecule
  - Measured onto a graph of absorbance vs wavenumber
  - Each peak at a wavenumber range corresponds to different functional groups
- Helps us see structure of samples

Together we can use NMR and IR Spectroscopy to determine the structure of the lidocaine samples

Mass Spectrometry (MS)



- Sorts ions by mass-to-charge ratio
- Uses electromagnetic fields
- We use it to compare the weight of our standard solution of lidocaine to the samples of lidocaine that were sitting for 2 months
- We didn’t actually do this

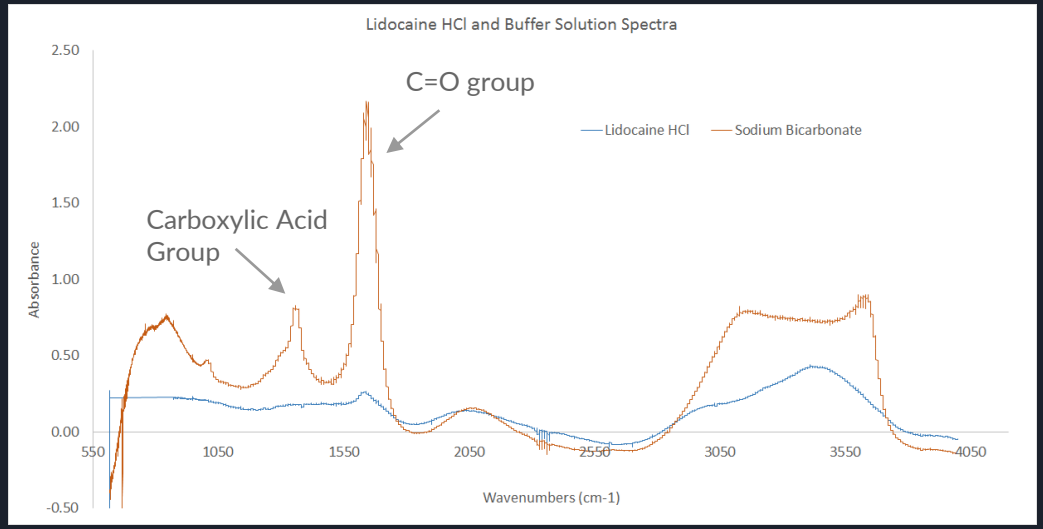
Data

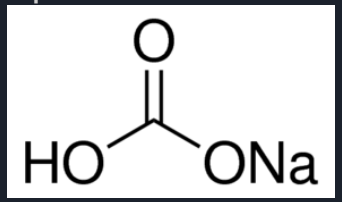
Obtained using IR, MS, and NMR

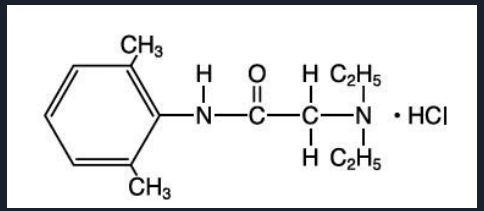
Spectra for IR Spectroscopy

IR Spectroscopy of Sodium Bicarbonate (buffer) and Lidocaine HCl w/ Epinephrine.

- IR spectrum has peaks representing amount of light transmitted & is used to determine functional groups in molecules
- Carbon Dioxide and C=O bonds are the cause of some of the peaks



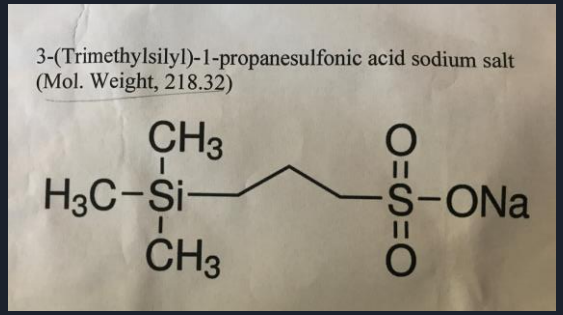


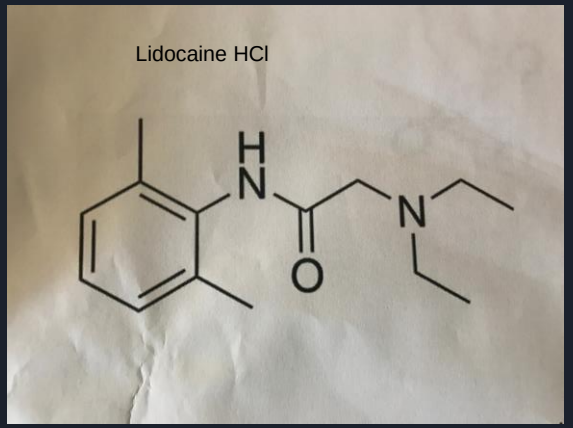


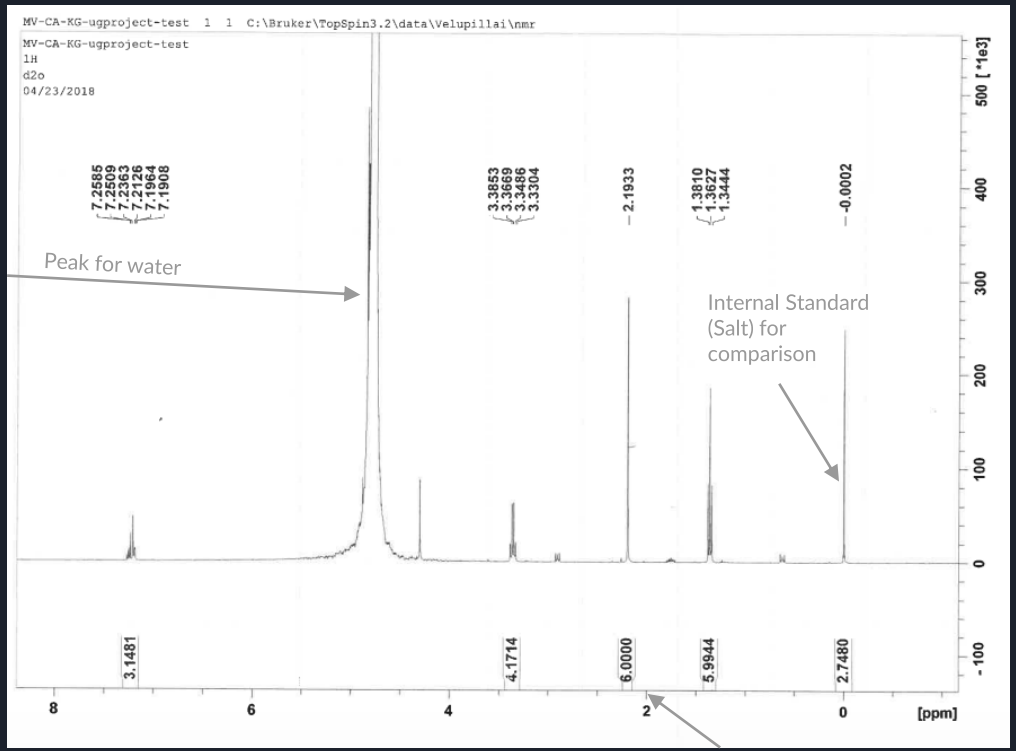
1870-1540 cm^{-1}					
1818 1750	strong	C=O	stretching	anhydride	
1740-1720	strong	C=O	stretching	aldehyde	
1730-1715	strong	C=O	stretching	α,β-unsaturated ester	or formates
1725-1705	strong	C=O	stretching	aliphatic ketone	or cyclohexanone or cyclopentenone
1720-1706	strong	C=O	stretching	carboxylic acid	dimer
1710-1680	strong	C=O	stretching	conjugated acid	dimer
1710-1685	strong	C=O	stretching	conjugated aldehyde	
1690	strong	C=O	stretching	primary amide	free (associated: 1650)
1400-1000 cm^{-1}					
1440-1395	medium	O-H	bending	carboxylic acid	
1420-1330	medium	O-H	bending	alcohol	

Libretexts. “Infrared Spectroscopy Absorption Table.” *Chemistry LibreTexts*, Libretexts, 21 July 2016. Accessed on 4/5/18.

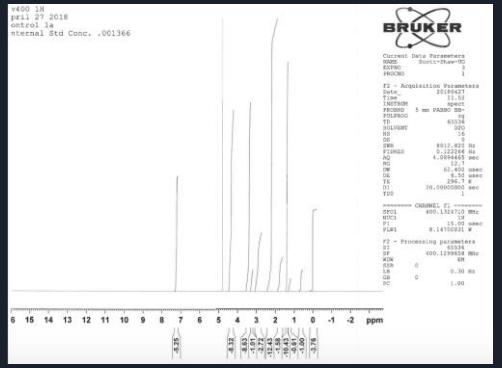
NMR Spectra for Standard Solution



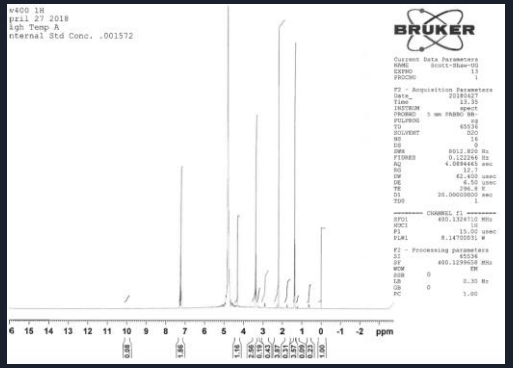




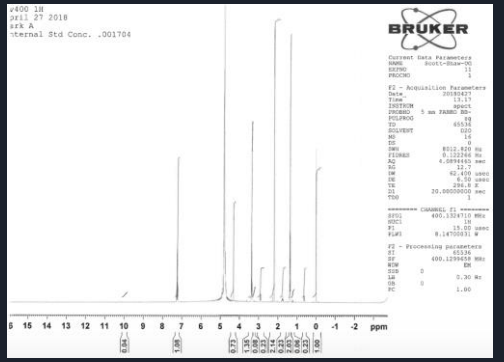
Integral Intensity
Gives us the number of hydrogen atoms in each region of the molecule



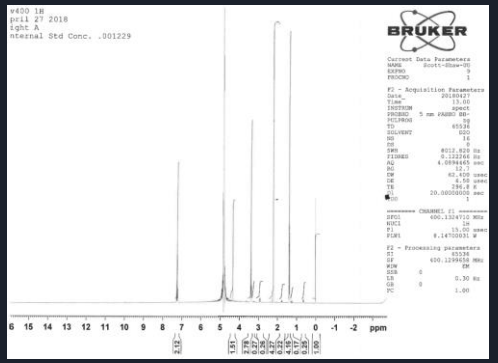
Control 1A



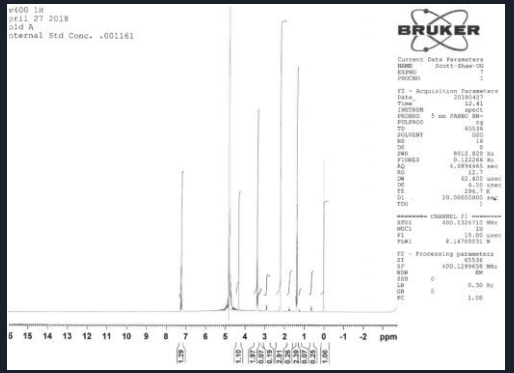
High Temp A



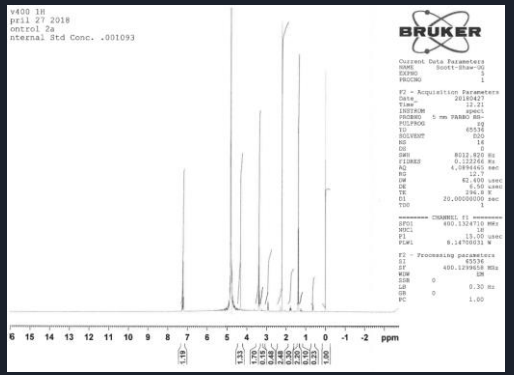
Dark A



Light A



Cold A



Control 2A

We conducted ran NMR tests on two tubes of each of the different lidocaine sample in the different environments.

Quantitation: Internal Standard Method

$$I.I_{ox} \alpha n_{ox}$$

$$I.I_{ox} \alpha M_{ox}$$

$$I.I_{ox} = K_s M_{ox} n_{ox} \frac{V_{gas}}{V_{tot}} \quad \text{.....[1]}$$

$I.I_{ox}$ –Integrated intensity of a group of resonances due to the oxygenate

K_s -Spectrometer constant

n_{ox}-Number of protons generating the signal

V_{gas}= 100ul

V_{tot} = 600ul

M_{ox}- Molar concentration of oxygenate

The integrated intensity ratio of the oxygenate to the internal standard is then:

$$\frac{I.I_{ox}}{I.I_{DMO}} = \frac{\cancel{K_s} n_{ox} M_{ox} (V_{gas}/ V_{tot})}{\cancel{K_s} n_{DMO} M_{DMO}} \text{.....[2]}$$

M_{DMO}- Molar concentration of DMO in the NMR sample

Rearranging equation [2]

$$M_{ox} = \frac{I.I_{ox}}{I.I_{DMO}} \frac{n_{DMO}}{n_{ox}} M_{DMO} \frac{V_{tot}}{V_{gas}} \quad \text{.....[3]}$$

Nuclear Magnetic Resonance (NMR)

	I.I DMO	I.I ox	n DMO	n ox	M DMO	V total	V gas	M ox (final concentration)	average M ox
3 - Control 1A	1	-12.43	9	6	0.00137	670	70	-0.24378	
4 - Control 1B	1	2.610	9	6	0.00130	670	70	0.04871	0.04871
5 - Control 2A	1	2.480	9	6	0.00109	670	70	0.03892	
6 - Control 2B	1	1.150	9	6	0.00287	670	70	0.04739	0.04315
7 - Cold A	1	2.910	9	6	0.00116	670	70	0.04851	
8- Cold B	1	2.000	9	6	0.00130	670	70	0.03733	0.04292
9 - Light A	1	4.270	9	6	0.00123	670	70	0.07534	
10- Light B	1	3.930	9	6	0.00157	670	70	0.08870	0.08202
11-Dark A	1	2.140	9	6	0.00170	670	70	0.05235	
12-Dark B	1	1.910	9	6	0.00226	670	70	0.06186	0.05711
13-High Temp A	1	3.870	9	6	0.00157	670	70	0.08734	
14-High Temp B	1	3.150	9	6	0.00171	670	70	0.07729	0.08232

- Original concentration of Lidocaine was 85 mM = 0.085 M

Mass Spectrometry Data

Compound 1: Lidocaine											
											start at 0.085 M
	Name	Sample Text	Type	Std. Conc	RT	Area	Response	ng/ul	%Dev	S/N	Conc Before Dilutions (ng/uL)	Conc in mol/L
1	T04261812	0.147 ng/ul	Standard	0.147	2.02	15651	15651.02	0.14	-7.3	7161.039		
2	T04261813	0.293 ng/ul	Standard	0.293	2.02	21433	21433.43	0.31	4.8	10546.983		
3	T04261814	0.588 ng/ul	Standard	0.588	1.99	32058	32057.654	0.62	5.5	17234.482		
4	T04261815	1.175 ng/ul	Standard	1.175	1.99	49626	49626.387	1.14	-3.1	23387.758		
5	T04261818	water	Blank									
6	T04261819	Control	Analyte	0.2772286614	2.02	20425	20424.912	0.28		9118.418	2772.286614	0.01023735737
7	T04261820	Low temp	Analyte	0.253367246	2.02	19616	19616.461	0.25		8097.082	2533.67246	0.009356215304
8	T04261821	Hight temp	Analyte	0.2800052537	1.99	20519	20518.986	0.28		10131.689	2800.052537	0.01033988994
9	T04261822	Light	Analyte	0.2273254115	2.02	18734	18734.135	0.23		9809.8	2273.254115	0.008394555836
10	T04261823	Dark	Analyte	0.2401462762	2.02	19169	19168.52	0.24		8082.41	2401.462762	0.008867998133
11	T04261824	Control 2	Analyte	0.1723776678	2.02	16872	16872.445	0.17		9081.951	1723.776678	0.006365473827
12	T04261825	water	Blank									

- Original concentration of Lidocaine was 85 mM = 0.085 M

Results

- Never finished buffered solution
- Cold and dark samples degraded the most
  - Based on the data from NMR and MS
  - Final concentrations were the lowest
  - We don’t know why
- Coming back in the fall for more research