Gilberto Gonzalez

Spectrophotometric Determination of pH 50

(Lab 6)

Purpose:

we will determine the pH of our UnKnown using spectrophotometric measurements.

Procedure:

Lab 6 - Spectrophotometric Determination of pH

Spectrophotometric Determination of pH 37 N4

Notes: Use the sample labeled "Spectro pH Unk." Measure the pH of 4 aliquots of the sample buffer solution spectrophotometrically, as described in your text. Using the diode-array spectrophotometer, measure and record the absorbance values at each of two wavelengths at, or near, the peaks for each of the two forms of the indicator. This may not be as clear as intended so be sure to ask for clarification.

Procedure Modification: After preparing your bromocresol green solutions in HCl and NaOH perform serial dilutions (1:2 volume dilutions each time) to generate three more BCG + HCl solutions and three more BCG + NaOH solutions. Measure the absorbance of these solutions at both lambda max values so you can generate two standard curves. Refer to the prelab serial dilution video regarding this. You need come up with a plan to do this with appropriate volumetric glassware. Discuss this with me prior for approval to proceed with your plan.

Use your standard absorbance curves to solve for the concentrations of protonated and deprotonated indicator in your unknown, which will allow you to determine the pH of your solution. Report the mean pH for your unknown buffer along with the absolute and relative (ppt) 95 % confidence intervals. Also report the propagated absolute and relative (ppt) uncertainty in the mean pH of you unknown buffer.

37N-4 The Spectrophotometric Determination of pH

Discussion

The pH of an unknown buffer is determined by addition of an acid/base indicator and spectrophotometric measurement of the absorbance of the resulting solution. Because there is overlap between the spectra for the acid and base forms of the indicator, it is necessary to evaluate individual molar absorptivities for each form at two wavelengths. See page 796 for further discussion.

The relationship between the two forms of bromocresol green in an aqueous solution is described by the equilibrium

$$HIn + H_2O \rightleftharpoons H_3O^+ + In^-$$

for which

$$K_a = \frac{[H_3O^+][In^-]}{[HIn]} = 1.6 \times 10^{-5}$$

The spectrophotometric evaluation of $[In^-]$ and $[HIn]$ permits the calculation of $[H_3O^+]$ and thus pH.

dilstock

PREPARATION OF SOLUTIONS

1. *Bromocresol green, 1.0 × 10⁻⁴ M* (sufficient for about five determinations). Dissolve 40.0 mg (to the nearest 0.1 mg) of the sodium salt of bromocresol green (720 g/mol) in water, and dilute to 500 mL in a volumetric flask.
2. *HCl, 0.5 M.* Dilute about 4 mL of concentrated HCl to approximately 100 mL with water.
3. *NaOH, 0.4 M.* Dilute about 7 mL of 6 M NaOH to about 100 mL with water.

PROCEDURE

Determination of Individual Absorption Spectra

Transfer 25.00-mL aliquots of the bromocresol green indicator solution to two 100-mL volumetric flasks. To one add 25 mL of 0.5 M HCl; to the other add 25 mL of 0.4 M NaOH. Dilute to the mark and mix well.

Obtain the absorption spectra for the acid and conjugate-base forms of the indicator between 400 and 600 nm, using water as a blank. Record absorbance values at 10-nm intervals routinely and at closer intervals as needed to define maxima and minima. Evaluate the molar absorptivities for HIn and In^- at wavelengths corresponding to their absorption maxima.

Determination of the pH of an Unknown Buffer

Transfer 25.00 mL of the stock bromocresol green indicator to a 100-mL volumetric flask. Add 50.0 mL of the unknown buffer, dilute to the mark, and mix well. Measure the absorbance of the diluted solution at the wavelengths for which absorptivity data were calculated.

Report the pH of the buffer.

Procedure Notes:

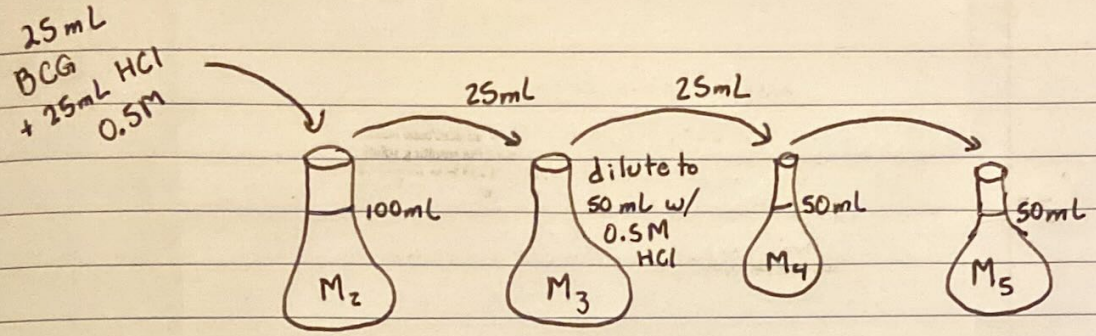
UnK: 3H-44

Solution 1: 40 mg of sodium salt in water, dilute in 500mL Volumetric flask

Solution 2: 4mL of HCl in 100mL DI H_2O

Solution 3: 7mL of 6M NaOH in 100mL of DI H_2O

Calculations:



BCG

$M_1 = 0.002$

$V_1 = ?$

$M_2 = 0.001$

$V_2 = 50mL$

$$V_1 = \frac{M_2 V_2}{M_1} = \frac{(0.001)(0.5)}{0.002} = 0.025L = 25mL$$

	HCl		NaOH			
Concentration	Absorbance (443nm)	[HIn]unk	Absorbance (615nm)	[In-]unk	[H+]	pH
0.000005	0.40925	3.56191E-06	0.97245	1.9792E-06	2.88E-05	4.540686472
0.0000025	0.19069	3.62687E-06	0.46914	1.99926E-06	2.90E-05	4.537216723
0.00000125	0.084579	3.60943E-06	0.2308	2.00708E-06	2.88E-05	4.541006727
0.000000625	0.026732	3.92278E-06	0.11219	2.10993E-06	2.97E-05	4.526554393
Mean:	0.17781275	3.68025E-06	0.446145	2.02387E-06	2.90853E-05	4.536366079
Std:	0.168569421	1.64005E-07	0.380974686	5.85665E-08	4.55786E-07	0.006762499

formula sheet is near the end of this lab report

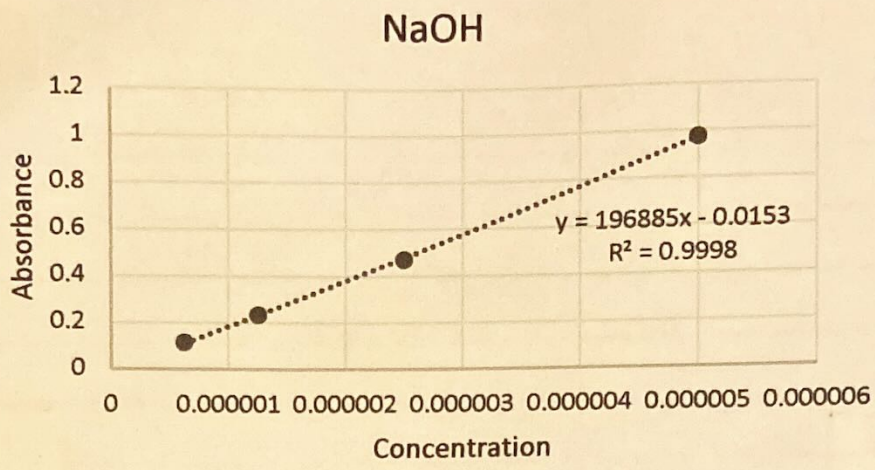
BCG W/ Unk		
	443nm	615nm
Trial1	0.28394	0.37437
Trial2	0.2896	0.37832
Trial3	0.28808	0.37986
Trial4	0.31538	0.40011
Mean:	0.29425	0.383165
Std:	0.01428827	0.01153086
G calc (Trial 4)	1.47883493	1.46953497
G crit (Trial 4)	1.463	1.463

Grubbs Test on Trial 4:
Gcalc > Gcrit so trial 4 should be thrown out.

Std for Trials 1-3.

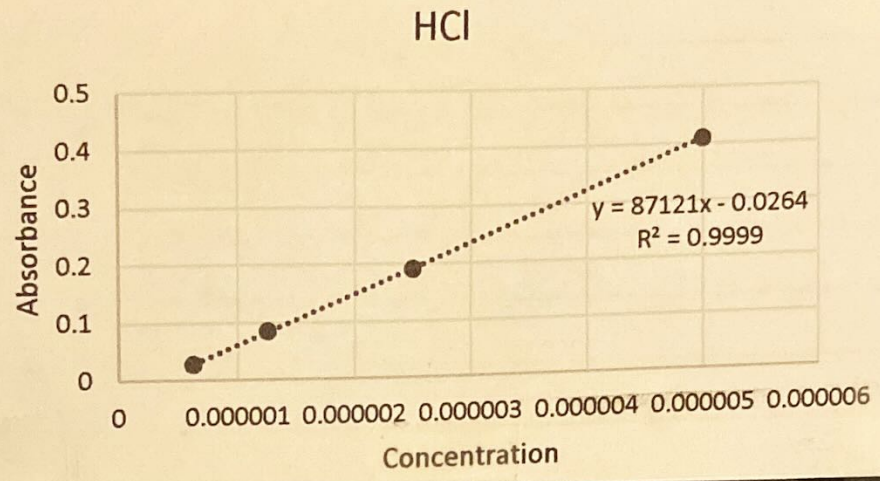
443	615
0.0029293	0.0028318

pH std for T1-T3
0.0021018



	A	B	C	D	E	F
2	Number of Stds (N)	Xi		Yi		
3	4	Molarity per Flask		Absorbance Signal of NaOH		
4	Number of Unknowns (M)	0		0		
5		0.000005		0.97245		
6		0.0000025		0.46914		
7		0.00000125		0.2308		
8		0.000000625		0.11219		
9	SUM(Xi)/N	2.34E-06	SUM(Yi)/N	0.446145		
10	SUM(Xi)	0.000009375				
11						
12						
13	Computing Regression Values					
14		Xi^2		Yi^2		XiYi
15		2.5E-11		0.945659003		4.86225E-06
16		6.25E-12		0.22009234		1.17285E-06
17		1.5625E-12		0.05326864		2.885E-07
18		3.90625E-13		0.012586596		7.01188E-08
19	SUM(Xi)^2	3.32031E-11	SUM(Yi)^2	1.231606578	SUM(XiYi)	6.39372E-06
20	(SUMXi)^2/N	2.19727E-11	(SUMyi)^2/N	0.796181444	(SUMXiSUMYi)/N	4.18261E-06
21	Sxx=	1.12305E-11	Syy=	0.435425134	Sxy=	2.21111E-06
22						
23	Defining The Trendline					
24	Slope(m)	196884.8696				
25	Intercept(b)	-1.53E-02				
26						
27	Evaluating The Fit to The Data					
28	Molarity Per Flask	Ycalc	(Y-Ycalc)^2			
29	0	-1.53E-02	2.34E-04			
30	0.000005	9.69E-01	1.11E-05			
31	0.0000025	4.77E-01	6.03E-05			
32	0.00000125	2.31E-01	4.73E-12			
33	0.000000625	1.08E-01	1.97E-05			

	A	B	C	D	E	F
1	Inputing Data					
2	Number of Stds (N)	Xi		Yi		
3	4	Molarity per Flask		Absorbance Signal of NaOH		
4	Number of Unknowns (M)	0		0		
5		0.000005		0.97245		
6		0.0000025		0.46914		
7		0.00000125		0.2308		
8		0.000000625		0.11219		
9	SUM(Xi)/N	=SUM(B4:B8)/4	SUM(Yi)/N	=SUM(D4:D8)/4		
10	SUM(Xi)	=SUM(B4:B8)				
11						
12						
13	Computing Regression Values					
14		Xi^2		Yi^2		XiYi
15		=B5^2		=D5^2		=B5*D5
16		=B6^2		=D6^2		=B6*D6
17		=B7^2		=D7^2		=B7*D7
18		=B8^2		=D8^2		=B8*D8
19	SUM(Xi)^2	=SUM(B15:B18)	SUM(Yi)^2	=SUM(D15:D18)	SUM(XiYi)	=SUM(F15:F18)
20	(SUMXi)^2/N	=((SUM(B5:B8)^2)/4)	(SUMyi)^2/N	=((SUM(D5:D8)^2)/4)	(SUMXiSUMYi)/N	=(SUM(B5:B8)*SUM(D5:D8)/4)
21	Sxx=	=B19-B20	Syy=	=D19-D20	Sxy=	=F19-F20
22						
23	Defining The Trendline					
24	Slope(m)	=F21/B21				
25	Intercept(b)	=D9-B24*B9				
26						
27	Evaluating The Fit to The Data					
28	Molarity Per Flask	Ycalc	(Y-Ycalc)^2			
29	0	=(B24*A29)+(B25)	=(D4-B29)^2			
30	0.000005	=$B$24*A30+$B$25	=(D5-B30)^2			
31	0.0000025	=$B$24*A31+$B$25	=(D6-B31)^2			
32	0.00000125	=$B$24*A32+$B$25	=(D7-B32)^2			
33	0.000000625	=$B$24*A33+$B$25	=(D8-B33)^2			



	A	B	C	D	E	F
37	Inputing Data					
38	Number of Stds (N)	Xi		Yi		
39	4	Molarity per Flask		Absorbance Signal of HCl		
40	Number of Unknowns (M)	0		0		
41		0.000005		0.40925		
42		0.0000025		0.19069		
43		0.00000125		0.084579		
44		0.000000625		0.026732		
45	SUM(Xi)/N	2.34E-06	SUM(Yi)/N	0.17781275		
46	SUM(Xi)	0.000009375				
47						
48						
49	Computing Regression Values					
50		Xi^2		Yi^2		XiYi
51		2.5E-11		0.167485563		2.04625E-06
52		6.25E-12		0.036362676		4.76725E-07
53		1.5625E-12		0.007153607		1.05724E-07
54		3.90625E-13		0.0007146		1.67075E-08
55	SUM(Xi)^2	3.32031E-11	SUM(Yi)^2	0.211716446	SUM(XiYi)	2.64541E-06
56	(SUMXi)^2/N	2.19727E-11	(SUMyi)^2/N	0.126469496	(SUMXi)(SUMYi)/N	1.66699E-06
57	Sxx=	1.12305E-11	Syy=	0.085246949	Sxy=	9.78412E-07
58						
59	Defining The Trendline					
60	Slope(m)	87121.18261				
61	intercept(b)	-2.64E-02				
62						
63	Evaluating The Fit to The Data					
64	Molarity Per Flask	Ycalc		(Y-Ycalc)^2		
65	0	-2.64E-02		6.96E-04		
66	0.000005	9.69E-01		3.13E-01		
67	0.0000025	4.77E-01		8.19E-02		
68	0.00000125	2.31E-01		2.14E-02		
69	0.000000625	1.08E-01		6.56E-03		

	A	B	C	D	E	F
38	Number of Stds (N)	Xi		Yi		
39	4	Molarity per Flask		Absorbance Signal of HCl		
40	Number of Unknowns (M)	0		0		
41		0.000005		0.40925		
42		0.0000025		0.19069		
43		0.00000125		0.084579		
44		0.000000625		0.026732		
45	SUM(Xi)/N	=SUM(B40:B44)/4	SUM(Yi)/N	=SUM(D40:D44)/4		
46	SUM(Xi)	=SUM(B40:B44)				
47						
48						
49	Computing Regression Values					
50		Xi^2		Yi^2		XiYi
51		=B41^2		=D41^2		=B41*D41
52		=B42^2		=D42^2		=B42*D42
53		=B43^2		=D43^2		=B43*D43
54		=B44^2		=D44^2		=B44*D44
55	SUM(Xi)^2	=SUM(B51:B54)	SUM(Yi)^2	=SUM(D51:D54)	SUM(XiYi)	=SUM(F51:F54)
56	(SUMXi)^2/N	=(SUM(B41:B44)^2)/4)	(SUMyi)^2/N	=([SUM(D41:D44)^2)/4)	(SUMXi)(SUMYi)/N	=(SUM(B41:B44)*SUM(D41:D44)/4)
57	Sxx=	=B55-B56	Syy=	=D55-D56	Sxy=	=F55-F56
58						
59	Defining The Trendline					
60	Slope(m)	=F57/B57				
61	intercept(b)	=D45-B60*B45				
62						
63	Evaluating The Fit to The Data					
64	Molarity Per Flask	Ycalc		(Y-Ycalc)^2		
65	0	=(B60*A65)+(B61)		=(D40-B65)^2		
66	0.000005	=$B$24*A66+$B$25		=(D41-B66)^2		
67	0.0000025	=$B$24*A67+$B$25		=(D42-B67)^2		
68	0.00000125	=$B$24*A68+$B$25		=(D43-B68)^2		
69	0.000000625	=$B$24*A69+$B$25		=(D44-B69)^2		

$$e_{num_1} = \sqrt{(0.0029293)^2 + (-0.026377522)^2}$$

$$e_{num_1} = 0.026539677$$

$$\frac{e_a}{a} = \sqrt{\left(\frac{0.026539677}{0.310317522}\right)^2 + \left(\frac{540.568164}{87121.1826}\right)^2}$$

$$\frac{e_a}{a} = 0.085749045$$

$$e_{num_2} = \sqrt{(0.0028318)^2 + (-0.015303913)^2}$$

$$e_{num_1} = 0.015563703$$

$$\frac{e_b}{b} = \sqrt{\left(\frac{0.015563703}{0.389673913}\right)^2 + \left(\frac{2014.52212}{196884.87}\right)^2}$$

$$\frac{e_b}{b} = 0.041230123$$

$$\frac{e_{[H^+]}}{[H^+]} = \sqrt{(0.085749045)^2 + (0.041230123)^2}$$

$$\frac{e_{[H^+]}}{[H^+]} = \pm 0.095146318 \qquad e_{[H^+]} = 0.095146318 \cdot 2.88 \times 10^{-5}$$

$$e_{[H^+]} = 0.00000274$$

$$e_{pH} = 0.095146318 \cdot \left(\frac{1}{\ln(10)}\right)$$

$$e_{pH} = 0.041321521$$

ppt: $0.041321521 / \text{Trial 1 pH} \cdot 1000 = 9.100280541$ ppt

95% CL

$$\mu = 4.54068647 \pm \frac{3.182 \cdot 0.0021018}{\sqrt{3}}$$

$$\mu = 4.54068647 \pm 0.003861277$$

	B	C	D	E	F	G	H
11		HCl		NaOH			
12	Concentration	Absorbance (443nm)	[Hln]unk	Absorbance (615nm)	[ln-]unk	[H+]	pH
13	0.000005	0.40925	=(O14+C24)/C23	0.97245	=(P14+C28)/C27	=((D13)*O2)/F13	=-LOG(G13)
14	0.0000025	0.19069	=(O15+C24)/C23	0.46914	=(P15+C28)/C27	=((D14)*O3)/F14	=-LOG(G14)
15	0.00000125	0.084579	=(O16+C24)/C23	0.2308	=(P16+C28)/C27	=((D15)*O4)/F15	=-LOG(G15)
16	0.000000625	0.026732	=(O17+C24)/C23	0.11219	=(P17+C28)/C27	=((D16)*O5)/F16	=-LOG(G16)
17							
18	Mean:	=AVERAGE(C13:C16)	=AVERAGE(D13:D16)	=AVERAGE(E13:E16)	=AVERAGE(F13:F16)	=AVERAGE(G13:G16)	=AVERAGE(H13:H16)
19	Std:	=STDEV(C13:C16)	=STDEV(D13:D16)	=STDEV(E13:E16)	=STDEV(F13:F16)	=STDEV(G13:G16)	=STDEV(H13:H16)

22	HCl	
23	m	87121.1826086957
24	b	0.0263775217391305

26	NaOH	
27	m	196884.869565217
28	b	0.0153039130434782

	N	O	P
2	Ka=	0.000016	
3		0.000016	
4		0.000016	
5		0.000016	

12	BCG W/ Unk		
13		443nm	615nm
14	Trial1	0.28394	0.37437
15	Trial2	0.2896	0.37832
16	Trial3	0.28808	0.37986
17	Trial4	0.31538	0.40011
18			
19	mean:	=AVERAGE(O14:O17)	=AVERAGE(P14:P17)
20	std:	=STDEV.S(O14:O17)	=STDEV.S(P14:P17)
21	G calc:	=(O17-O19)/O20	=(P17-P19)/P20
22	G crit:	1.463	1.463

Conclusion:

	T1	T2	T3
$[HIn]$	$3.56191 \times 10^{-6} \pm 0.09$	$3.62687 \times 10^{-6} \pm 0.09$	$3.60943 \times 10^{-6} \pm 0.09$
$[In^-]$	$1.9792 \times 10^{-6} \pm 0.04$	$1.99926 \times 10^{-6} \pm 0.04$	$2.00708 \times 10^{-6} \pm 0.04$
$[H^+]$	$2.88 \times 10^{-5} \pm 0.2 \times 10^{-5}$	$2.90 \times 10^{-5} \pm 0.2 \times 10^{-5}$	$2.88 \times 10^{-5} \pm 0.2 \times 10^{-5}$
pH	4.540686472 ± 0.04	4.537216723 ± 0.04	4.541006727 ± 0.04

error probs

$[HIn]$	± 0.085749045
$[In^-]$	± 0.041230123
$[H^+]$	± 0.00000274
pH	± 0.041321521
ppt	9.100280541 ppt

All done using Trial 1 data

UnKnown 3H-44, gave me an average pH of $4.539636641 \pm 0.041321521$.