Procedure

Materials

Chemicals

- ❑ Stock bromothymol blue solution (2.5×10^{-4} M)
- ❑ Potassium dihydrogen phosphate, anhydrous, KH_2PO_4
- ❑ Potassium hydrogen phosphate, monohydrate, $K_2HPO_4 \cdot H_2O$ pH 4 buffer solution
- ❑ pH 10 buffer solution

Equipment

- ❑ Labeling tape
- ❑ Sharpie marker or wax pencil
- ❑ Large test tubes (5)
- ❑ Test tube rack
- ❑ Cuvettes with caps (6)
- ❑ Cuvette rack
- ❑ 100 mL graduated cylinder
- ❑ 100 mL beakers (3)
- ❑ 250 mL beaker
- ❑ Volumetric pipettes: 5 mL, 10 mL, 25 mL
- ❑ Pipette bulb
- ❑ Glass stir rod
- ❑ Spatula
- ❑ Deionized water wash bottle

Instrumentation

- ❑ pH sensor
- ❑ Visible spectrometer
- ❑ Digital device for collecting spectroscopic data

PART A Getting Ready

1 Prepare a waste container.

2 Label your 100 mL beakers KH_2PO_4, K_2HPO_4, and Stock BTB.

3 Label your 250 mL beaker DI H_2O.

4 Label your large test tubes 1, 2, 3, 4, and 5, and then arrange them in numerical order in your test tube rack.

5 Label one of your cuvette caps Blank, and the remaining caps 1, 2, 3, 4, and 5.

6 Review General Protocol 1: Measuring Mass with an Electronic Balance, page 13.

7 Prepare the KH_2PO_4 solution using the following steps:

- **a** Weigh out enough KH_2PO_4 to make 50 mL of 0.25 M solution. Consult Table 9.2 for the theoretical value you calculated in pre-lab question 1, and record the actual value you measured.
- **b** Transfer the solid to your beaker labeled KH_2PO_4.
- **c** Measure 50.0 mL of deionized water with your graduated cylinder and add it to the KH_2PO_4 beaker.
- **d** Stir the solution with your glass stir rod until the solid has completely dissolved.
- **e** Rinse the stir rod with deionized water, catching the rinse water in your waste container.

8 Prepare the K_2HPO_4 solution as described in step 7. Consult Table 9.2 for the theoretical value you calculated in pre-lab question 2, and record the actual value you measured.

PART B Making Solutions of BTB at Five Different pHs

Making the following solutions may take about an hour, but the measurements you make once the solutions are created will go very quickly. Refer to Table 9.1 for the volumes of stock BTB solution, KH_2PO_4 solution, K_2HPO_4 solution, and water required to make the solutions described in the following steps.

TABLE **9.1** Volumes of Various Compounds for the BTB/Buffer solutions

Solution	BTB (aq) (mL)	KH_2PO_4 (aq) (mL)	K_2HPO_4 (aq) (mL)	Water (mL)
1	5.00	0.00	10.00	35.00
2	5.00	5.00	10.00	30.00
3	5.00	10.00	10.00	25.00
4	5.00	15.00	10.00	20.00
5	5.00	10.00	0.00	35.00

1 Obtain about 200 mL of deionized water in your beaker labeled DI H_2O.

2 Obtain about 30 mL of stock BTB solution in your beaker labeled Stock BTB.

3 Review General Protocol 5: Conditioning a Volumetric Pipette (p. 16), and 6: Transferring a Volume with a Volumetric Pipette (p. 16).

4 Condition your 5.00 mL volumetric pipette with ~2 mL of the BTB solution. Collect the rinses in your waste container.

5 Using the conditioned 5.00 mL pipette, transfer 5.00 mL of the BTB solution to each test tube.

6 Rinse the 5.00 mL pipette twice with tap water and once with deionized water. Collect the rinse water in your waste container.

7 Condition your 10.00 mL and 5.00 mL volumetric pipettes with the KH_2PO_4 solution. Collect the rinses in your waste container.

8 Use the conditioned 10.00 mL and 5.00 mL pipettes as needed to deliver the scheduled volumes of KH_2PO_4 solution to Test Tubes 2–5.

9 Rinse the 10.00 mL and 5.0 mL pipettes twice with tap water and once with deionized water. Collect the rinse water in your waste container.

10 Condition your 10.00 mL volumetric pipette with the K_2HPO_4 solution. Collect the rinses in your waste container.

11 Using the conditioned 10.00 mL pipette, transfer 10.00 mL of the K_2HPO_4 solution to Test Tubes 1–4.

12 Rinse the 10.00 mL pipette twice with tap water and once with deionized water. Collect the rinse water in your waste container.

13 Use the 25.00 mL, 10.00 mL, and 5.00 mL volumetric pipettes to add the appropriate volume of water to each test tube.

14 Record the resulting colors of the BTB solutions in Table 9.3 on the data sheet, page 293.

PART C Measuring pH of the BTB Solutions

1 Calibrate your pH meter as described by your instructor.

2 Measure the pH of the five solutions prepared in Part B, making sure to thoroughly rinse the pH sensor with deionized water before each measurement.

3 Record the pHs in Table 9.3 on the data sheet, page 293.

4 **Reflect:** Compare your predictions of solution color vs. pH from the pre-lab questions to the actual pH measurements you just made. Were your predictions correct? Why or why not?

PART D Measuring Absorbance of the BTB Solutions

1 Review General Protocol 9: Holding and Cleaning a Cuvette, page 20.

2 Calibrate your spectrometer using the following steps:

  a Fill one of your cuvettes at least 3/4 full with deionized water and cap it with the cap labeled Blank.

  b Use a Kimwipe to clean the clear windows of the cuvette.

  c Place the cuvette in your spectrometer and run the calibration procedure as described by your instructor.

3 Fill one of your cuvettes at least 3/4 full with your Solution 1, and cap it with the 1 cap. Wipe the clear windows of the cuvette carefully with a Kimwipe.

4 Repeat step 3 for your remaining BTB solutions.

5 Collect the visible absorbance spectrum for Solution 1, and determine its λ_{max} using the following steps:

  a Place Cuvette 1 into the spectrometer and collect the full visible absorption spectrum.

  b Record λ_{max} for Solution 1 and the absorbance at λ_{max} in Table 9.3 on the data sheet, page 293.

  c Save the spectrum with an appropriate file name on the digital data collector.

6 **Reflect:** Do you need to determine λ_{max} and ϵ for both the acidic and basic forms of BTB in this experiment? Why or why not?

7 Collect the absorbance of Solution 5 at the λ_{max} for Solution 1 using the following steps:

  a Place Cuvette 5 into the spectrometer and collect the full visible absorption spectrum.

  b Record the absorbance of the solution at the λ_{max} for Solution 1 in Table 9.3.

  c Check with your instructor to make sure that the absorbance of this solution at the λ_{max} for Solution 1 is close enough to zero to proceed.

  d Save the spectrum with an appropriate file name on the digital data collector.

8 Collect the visible absorbance spectrum of Solution 2 at the λ_{max} for Solution 1 using the following steps:

a Place Cuvette 2 in the spectrometer and collect the full visible absorption spectrum.

b Record the absorbance of the solution at the λ_{max} for Solution 2 in Table 9.3.

c Save the spectrum with an appropriate file name on the digital data collector.

9 Repeat step 8 for Cuvettes 3 and 4.

10 **Reflect:** Why is it important that the absorbance of Solution 5 at the λ_{max} for Solution 1 is close to zero?

11 **Reflect:** Should the *K* of Reaction 5 depend on the pH of the solution? Why or why not? Explain your reasoning.

12 Discard any remaining solutions in your waste container, and dispose of the waste as instructed.

13 Rinse all glassware and cuvettes twice with tap water and once with deionized water.

DATA SHEET 9.1

Name

Section Date

Data Analysis and Calculations

TABLE **9.2** Masses of KH_2PO_4 and K_2HPO_4 Required to Make 50 mL of 0.25 M Solutions

	Theoretical	Actual
Mass KH_2PO_4 (g)	1.7 g	1.8g
Mass $K_2HPO_4 \cdot H_2O$	2.4g	2.868 g

TABLE **9.3** Data for Color, pH, and Absorbance of Solutions 1–5

λ_{max} for Solution 1: ______ nm

Solution	Color	pH	Absorbance at λ_{max} for Solution 1
1	Blue		-0.076
2	Bluish green		-0.077
3	Green		-0.079
4	Greenish Blue		-0.0880
5	Yellow	6.80	-0.077

1 Determine the molar absorptivity of B^{2-} (ϵ_b) from your absorbance data for Solution 1 using Equation 4. Record your result with proper units in the space provided.

ϵ_b:

Calculation:

7:40 AM Sat Jul 20

learn-us-east-1-prod-fleet01-xythos.s3.us-east-1.amazonaws.com

61%

2 Determine the following for Solutions 2 through 4, and record your results in Table 9.4.

a $[B^{2-}]$ using Equation 5.

Sample Calculation with Solution 2:

b $[HB^{-}]$ using Equation 2.

Sample Calculation with Solution 2:

c $[H_3O^+]$ from the pH using $[H_3O^+] = 10^{-pH}$.

Sample Calculation with Solution 2:

d K using Equation 1.

Sample Calculation with Solution 2:

DATA SHEET 9.1
(continued)

Name ______________________

Section ______________ Date ______________

3 Compare your *K*s to the literature value of 7.9×10^{-8}, and calculate the percent error in each case. Record your results in Table 9.4.

Sample Calculation with Solution 2:

TABLE **9.4** Data for Determination of the *K* for BTB

Concentration of BTB in each Solution (from pre-lab question 6): ______________ M

Solution	$[B^{2-}]$ (M)	$[HB^-]$ (M)	$[H_3O^+]$ (M)	*K*, Experimental	Percent Error
2					
3					
4					

4 If your spectrometer allowed you to collect the full spectrum for each solution, use Excel to construct a plot with the full spectra you took for all five solutions overlaid.

Post-Lab Questions

1 Compare the *K*s you calculated for each solution. Are they similar or different? How does the similarity of your results compare to the prediction you made in the pre-lab questions?

2 Use Le Chatelier's principle to describe how the concentrations of HB^- and B^{2-} change in response to increasing pH in Solutions 1–5. (***Hint:*** Consider the colors you observed for Solutions 1–5.)

3 Assume that when you were making Solution 1, you accidentally added 1 mL more water than called for without realizing it. (Assume that all other solutions were made perfectly.)

a How would this error affect the actual concentration of B^{2-} in Solution 1 (as opposed to the concentration you thought it was)?

b How would the incorrect concentration of B^{2-} affect the value of ϵ_b you calculated for Solution 1?

c How would the incorrect value of ϵ_b affect the value of $[B^{2-}]$ you calculated for Solutions 2 through 4?

9

d How would the incorrect values of $[B^{2-}]$ for Solutions 2 through 4 affect the value of $[HB^-]$ you calculated for these solutions?

e How would the incorrect values of $[B^{2-}]$ and $[HB^-]$ affect the values of *K* you calculated for Solutions 2 through 4?

4 Describe how a minor error at the beginning of an experiment can affect your final results.

a Consider the procedure you performed in this activity. What is one other possible measurement error a student could make?

Name

Section Date

b Following the framework in question 3, describe how this error would propagate through the subsequent measurements and data analysis in the experiment.

5 If your spectrometer allowed you to collect the full spectrum for each solution, consider your plot of the overlaid spectra. What trend do you notice as the pH of the solutions increases? How does this trend relate to the observed color of the solution?