

Name(s) _____

DATA SHEET
SPECTROPHOTOMETRY

I. Establishing a calibration curve

Solution	Concentration (g Hb/100 mL)	Absorbance
1	4.0	
2	8.0	
3	12.0	
4	16.0	
5	20.0	

Record the equation for your calibration line below:

$$\text{Absorbance} = \frac{\quad}{\text{slope}} \times \text{Concentration} + \frac{\quad}{\text{intercept}}$$

II. Determination of unknown concentration of blood

Record the designation of your unknown. _____

Trial #	Absorbance	Hb Concentration in "Diluted Blood" sample	Actual Hb Concentration in "Blood"
1			
2			
Average	-----	-----	

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REVIEW QUESTIONS
SPECTROPHOTOMETRY

1. What is a spectrophotometer? What does it do?
2. What wavelengths comprise white light?
3. Why does an object or a solution appear to have color?
4. What is a calibration curve, in general, and how is it prepared?
5. How can your calibration curve be used to determine the status of someone's health?
6. If the absorbance of a sample falls outside, or off the graph, what can be done to make the absorbance fit onto the curve? Hint: What did you do to your unknown?
7. Is your "patient" healthy? Why or why not?
8. How can you report g Hb/100 mL when you only analyzed 10 mL?
9. Comment on the reproducibility of this method of analysis. Were your two values similar?
10. The iron-thiocyanate complex $[\text{Fe}(\text{SCN})]^{2+}$ is considered an unstable complex. It decomposes over time. What constraints might this impose on your experimental measurement?