ACID DISSOCIATION CONSTANT OF METHYL RED

The spectrophotometric determination of the acid dissociation constant of a dye is illustrated.

<u>Apparatus</u>
Bausch and Lomb Spectronic 20 (L); Set of four matched cells (13x100 mm test tubes)(S); pH meter (L); Standard buffer of pH 6.00 (S); six 100 ml volumetric flasks (D,S); 5 ml, 10 ml, and 25 ml pipettes (D).

Chemicals
Stock solution of Methyl Red (L); 60 ml 95% ethanol (L); distilled H_2O (L); 250 ml 0.04 M sodium acetate (P); 100 ml 0.01 M sodium acetate (P); 100 ml 0.05 M acetic acid (P); 25 ml 0.1 M HCl (P); 100 ml 0.01 M HCl (P); stock $CoCl_2$ solution [2g $CoCl_2 \bullet 6H_2O$ per 100 ml 0.3 M HCl](L); glacial acetic acid (L).

THEORY. Calculations involving the dissociation constants of weak acids and bases are fundamental to any discussion of homogenous equilibrium. Equilibrium is defined at the point when the change in Gibbs Free Energy for the process (or reaction) is equal to zero. This equilibrium point will depend on the concentration of the various reactants. For an ideal solution (and only for an ideal solution) the Gibbs Free Energy for a compound can be written as a function of the concentration, C, of the compound and the standard Free energy of the compound in some standard state ΔG^o (usually when the concentration equals one molar).

$$\Delta G = \Delta G^o + RT\ln([C])$$

For acid base equilibrium some acid HA will dissolve in water with a concentration we will label [HA]. Part of the dissolved acid will dissociates into a hydrogen ion H^+ and a conjugate acid A^- by the following reaction.

The Free Energy change of this reaction can be expressed as follows:

$$\Delta G_R = \Delta G_{H^+} + \Delta G_{A^-} - \Delta G_{HA}$$

$$= \Delta G^o_{HA} + RT\ln([HA]) - (\Delta G^o_{H^+} + RT\ln([H^+]) + \Delta G^o_{A^-} + RT\ln([A^-]))$$

$$= \Delta G^o_R + RT\ln\left(\frac{[H^+][A^-]}{[HA]}\right) = 0 \ \textit{At equilibrium}$$

Therefore at equilibrium, the quantity $[HA]/[H^+][A^-]$ must be a constant since ΔG^o is not a function of concentration. We will call this quantity K. For methyl red, the ion A^-, is given in Figure 1.

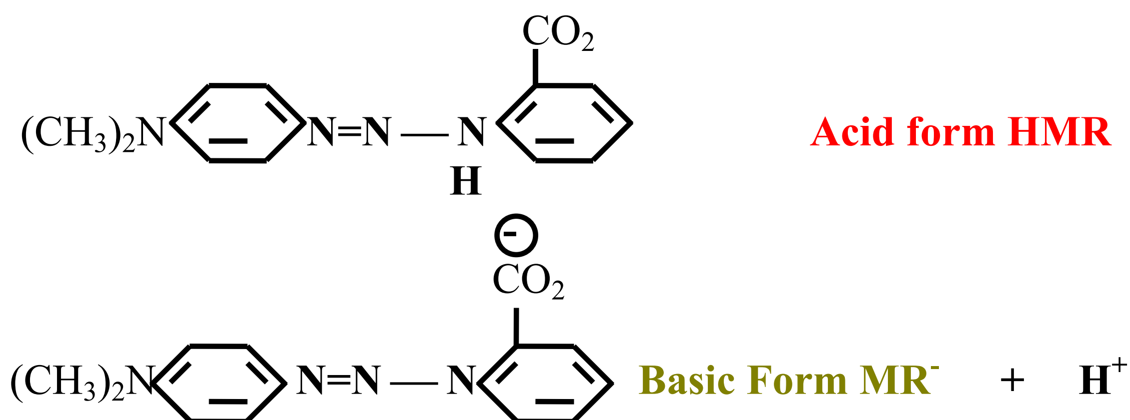


Figure 1. Acid (HMR) and basic (MR-) form of methyl red.

Methyl red provides a particularly good system with which to study acid-base equilibrium since both HMR and MR^- have strong absorption peaks in the visible portion of the spectrum.

In aqueous solution methyl red is a zwitterion and has a resonance structure somewhere between the two extreme forms. This is the red form HMR in which methyl red exists in acid solutions. When base is added, a proton is lost and the yellow anion MR^- of methyl red has the structure shown at the bottom of the figure. The basic form is yellow because it absorbs blue and violet light. The equilibrium Constant for the ionization of methyl red is

$$K_c = \frac{(H^+)(MR^-)}{(HMR)} \qquad (1)$$

It is convenient to use this equation in the form

$$pK_c = pH - \log\frac{(MR^-)}{(HMR)} \qquad (2)$$

The ionization constant may be calculated from measurements of the ratio $(MR^-)/(HMR)$ at known pH values.

Since the two forms of methyl red absorb strongly in the visible range, the ratio $(MR^-)/(HMR)$ may be determined spectrophotometrically. The absorption spectra of methyl red in acidic and basic solutions are determined, and two wavelengths are selected for analyzing mixtures of the two forms. These two wavelengths, λ_1 and λ_2, are chosen so that at one, the acidic form has a very large absorbancy

index compared with the basic form, and at the other, the situation is reversed. The absorbancy indices of HMR and MR^- are determined at both of these wavelengths, using several concentrations to determine whether Beer's law is obeyed.

The composition of a mixture of HMR and MR^- may be calculated from absorbancies A_1 and A_2 at wavelengths λ_1 and λ_2 using, at unit cell thickness,

$$A_1 = a_{1,HMR}(HMR) + a_{1,MR^-}(MR^-) \quad (3)$$
$$A_2 = a_{2,HMR}(HMR) + a_{2,MR^-}(MR^-) \quad (4)$$

PROCEDURE. The procedure for this experiment has been described by Tobey[1]. The methyl red is conveniently supplied as a stock solution made by dissolving crystalline methyl red in 300 ml of 95 percent ethanol and diluting to 500 ml with distilled water. The standard solution of methyl red for use in this experiment; is made by adding 5 ml of the stock solution to 50 ml of 95 percent ethanol and diluting to 100 ml with water.

Preparation of Standard Solution

The absorption spectrum of methyl red is determined in hydrochloric acid solution as solvent to obtain the spectrum of HMR and in sodium acetate solution as solvent to obtain the spectrum of MR^-. Distilled water is used in the reference cell. Since the equilibrium to be studied is affected by temperature, it is important that all the spectrophotometric and pH measurements be made at the same temperature. If the cell compartment of the spectrophotometer is slightly above room temperature, the filled cells should be placed in the spectrophotometer, pictured in Figure 2, just before making the measurements. In order to obtain the best results, the cell compartment should be thermostated. Distilled water should be used to set the 0% and 100% transmission for each sample and each wavelength.

The acidic solution is conveniently prepared by diluting a mixture of 10 ml of the standard methyl red solution and 10 ml of 0.1 M hydrochloric acid to 100 ml. The pH of this solution should be around 2.

The basic solution is conveniently prepared by diluting a mixture of 10 ml of the standard methyl red solution and 25 ml of 0.04 M sodium acetate to 100 ml. The pH of this solution should be around 8.

Figure 2. Apparatus for the determination of the transmission and absorbance of a sample, a spectrophotometer.

You must prepare your own 0.04 M sodium acetate; weigh out the appropriate amount of $NaC_2H_3O_2$, place in a 250 ml volumetric flask and add distilled H_2O up to the fiducial line. Dilute 25 ml of this 0.04M solution to 100 ml to prepare 100 ml of 0.01 M sodium acetate. Dilute 10 ml 0.1 M HCl to 100 ml to prepare 0.01 M HCl. To prepare the acetic acid solution: weigh 100 ml volumetric flask; add 3 ml glacial acetic acid, reweigh, dilute to 100 ml mark giving 0.5 M solution; now dilute 10 ml of this to 100 ml to get 100 ml 0.05 M acetic acid.

Using these standard solutions, the appropriate solution(s) for the measurement of the acid's equilibrium constant will be prepared.   Check to see that the spectrophotometer cells are matched using the following procedure: (1) half-fill a tube with the stock $CoCl_2$ solution; (2) set λ = 510 nm, place tube in compartment and adjust light control so that meter reads 90% transmission; (3) find three other tubes that give 90% transmission (or less than 1% variation from the reference tube). Operating instructions for the Spectronic 20 are given on the instrument case.

Measure the absorption spectrum of methyl red in the HCl solution and in the sodium acetate solution between 350 and 600 nm using H_2O in the reference

cell. This is done to find the absorption maxima for each species. Sample results are presented in Figure 3. From the plots of absorbancy versus wavelength which just obtained, two wavelengths are selected for analyzing mixtures of the acidic and basic forms of methyl red. These wavelength need not be at the maximums, but should have the greatest difference between the absorbancy of the acid and base form.

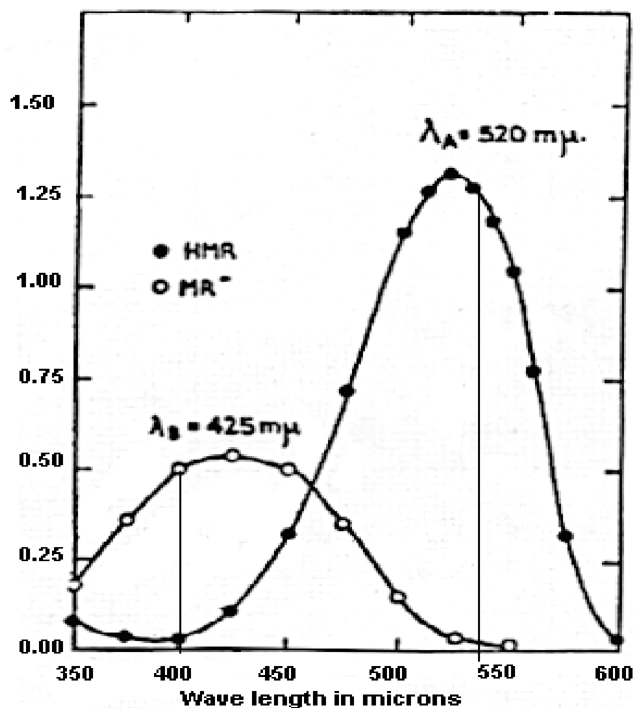


Figure 3. Absorbancy of HMR and MR- as a function of wavelength λ.

Further spectrophotometric measurements over a range of concentration are made at these two wavelengths with both acidic and basic solutions to check whether Beer's law is obeyed. Portions of the acid solutions (A) and basic solution (B) are diluted to 0.75, 0.50, and 0.25 times their initial concentration using 0.01 M HCl and 0.01 M NaAc respectively. Sample results are presented in Figure 4. This plot is used to calculate the four constants in equations 3 and 4.

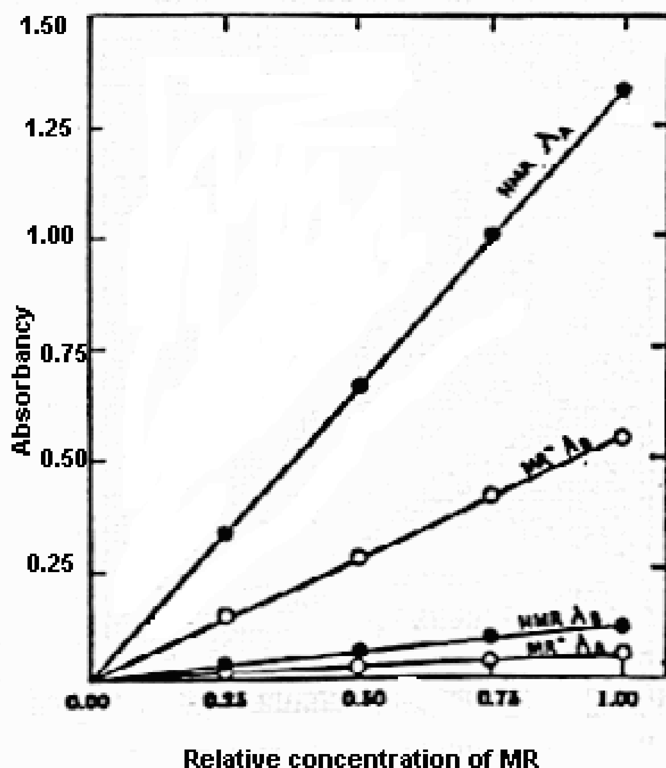


Figure 4. Absorbance of acid and basic form of methyl red at two wavelengths.

To determine the ionization constant of methyl red, the relative amounts of HMR and MR^- present in solution must be obtained as a function of pH. Spectrophotometic analyses are carried out on solutions containing 0.01 N sodium acetate, a constant total concentration of indicator, and various concentrations of acetic acid. The pH values of these solutions are measured, using a pH meter as pictured in Figure 5, at the same temperature as the spectrophotometric measurements. A standard buffer solution can be used to calibrate the pH meter using the pH set knob, and an additional buffer solution with a different pH should be used to confirm that the meter is calibrated.

For methyl red it is convenient to use acetic acid concentrations ranging from 0.001 to 0.05 N. The color of these methyl red solutions should vary from the acidic color to the basic color as seen in Figure 6.

Figure 5. pH meter

Figure 6. Methyl red solution with varying pH.

CALCULATIONS. Plots are prepared of absorbancy versus wavelength and absorbancy versus concentration of dye in acidic and basic solutions at λ_1 and λ_2. The values of the various absorbancy indices are calculated.

The concentrations of the acidic and basic forms of the dye in the various buffer solutions are calculated by using Eqs. (3) and (4).

Equation (2) is used to calculate the pK value for the dye. As a means of testing and averaging the data, log $[(MR^-)/(HMR)]$ may be plotted versus the pH. An average value from the literature[2] is 5.05 $\pm$ 0.05 for the 25 to 30° temperature range.

Practical applications. This method is useful for studying dyes for use as indicators in acid- base titrations, or by an analogous procedure for indicators for oxidation-reduction titrations.

Suggestions for further work. The pK values for other common dyes may be determined. General references are cited.[3]

References

1. S. W. Tobey, J. Chem. Educ., 35: 514 (1958).
2. 1. M. Kolthoff, "Acid-Base Indicators," The Macmillan Company, New York, 1953.
3. R. P. Bell, "Acids and Bases: Their Quantitative Behavior," Methuen & Co., Ltd., London, 1952; "The Proton in Chemistry," Cornell University Press, Ithaca, N.Y., 1959.

SPECTROPHOTOMETRIC DETERMINATION OF AN EQUILIBRIUM CONSTANT

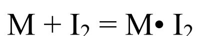
The equilibrium constant for a reaction in solution is determined from a study of the concentration dependence on the intensity of an absorption band in the spectrum of the solution.

THEORY. The present experiment illustrates an important method for the study of chemical equilibrium in solution. The method utilizes differences in the light absorbing properties of reactants and products and is particularly appropriate for systems for which classical methods of chemical analysis cannot be used to find the concentrations of the various species present.

The optical absorption spectrum, i.e., the percentage transmission of light as a function of wavelength, has been investigated for iodine in a variety of solvents. Associated with the color of the solutions is a strong absorption in the neighborhood of 500 mμ. In certain solvents, especially aromatic compounds, a new absorption band appears in the violet or ultraviolet region. The new band has been attributed to a complex (a molecular combination of iodine with solvent) existing in equilibrium with uncomplexed iodine and solvent. By means of a quantitative study of the intensity of absorption at the peak of this band, as a function of concentration, it is possible to test this interpretation and to obtain a value for the equilibrium constant for the formation of the complex.[1,2]

The study is best carried out with iodine and the complexing organic substance, mesitylene in this experiment, both present as solutes in dilute solution in an inert solvent such as CCl_4. (The inertness of CCl_4, expected on chemical grounds, is verified by the absence of new absorption bands in solutions of I_2 in CCl_4.) The initial step involves the measurement of the percentage transmission over the appropriate wavelength range, of three solutions: one containing solute only, one containing mesitylene solute only, the third containing both I_2 and mesitylene as solutes. An absorption band present only in the spectrum of the third solution is attributed to a 1:1 complex, $M \bullet I_2$, existing in equilibrium with free mesitylene

(M) and I_2,



$$M + I_2 = M \bullet I_2$$

$$K = X/(c_1 - X)(c_2 - X) \qquad (1)$$

where c_1 = total concentration of mesitylene
c_2 = total concentration of I_2
X = concentration of complex at equilibrium
K = equilibrium constant

It remains to verify that an equilibrium condition of the form of Eq. (1) is consistent with the concentration dependence of the absorption intensity and to evaluate K The new

absorption band occurs in a wavelength region in which absorption by uncomplexed iodine and mesitylene is very slight; the investigation can therefore proceed without serious interference from absorption due to uncomplexed solutes.

Let I and I_o be intensities of light of a specified wavelength transmitted, respectively, by solution and by pure solvent. Then the optical absorbancy A, defined by

$$A = -\log I/I_o \qquad (2)$$

is given by the Beer-Lambert law, for the case in which only one solute absorbs at the given wavelength, by

$$A = abc$$

where a = molar absorbancy index of absorbing solute
b = length of light path in cell
c = concentration of absorbing solute

The molar absorbaney index depends on the wavelength, temperature, and solvent.

If several solutes absorb independently, the absorbances are additive. Thus

$$A = A_l + A_2 + A_3 \qquad (3)$$
$$A_l = a_l b(c_l - x) \qquad (4)$$
$$A_2 = a_2 b(c_2 - x) \qquad (5)$$
$$A_3 = a_3 bx \qquad (6)$$

where a_l = molar absorbancy index of mesitylene
a_2 = molar absorbancy index of I_2
a_3 = molar absorbancy index of complex

Hence, in the present case, measurement of absorbancy does not lead directly to values for the concentration of complex. A_l and A_2, though smaller than A_3 at the wavelength of the peak of the new complex band, may not always be negligible; also, a_3 is initially unknown. However, the following indirect procedure yields values for a_3 as well as for K.

A considerable simplification in the work results from restricting the investigation to solutions in which mesitylene is present in large excess. Thus

$$c_1 \gg c_2 > X$$

Accordingly, for the calculation of A_3 from A, the approximations

$$A_l = a_l bc_l$$
$$A_2 = 0$$

may be used, with a_l calculated from the absorbancy measured at the given wavelength for a solution containing only mesitylene as solute at a known concentration. The equilibrium condition (1), with the approximation (7), simplifies to

$$K = X/c_1(c_2-x)$$

Upon replacement of x by A_3/a_3b and rearrangement, this becomes

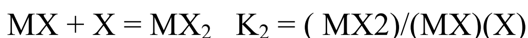
$$A_3/c_1c_2 = ba_3K - (A_3/c_2)K \qquad (9)$$

Absorbancies are measured for a series of solutions made up with various known values of c_l and c_2, and values of A_3/c_lc_2 plotted against A_3/c_2. If the data are well represented by a straight line, the slope and intercept lead to values for a_3 and K.

The inclusion of a correction for absorption due to uncomplexed I_2 may be warranted if the data are of sufficient accuracy. A procedure for carrying out this refinement is outlined under Suggestions for Further Work.

A word of caution is in order with regard to the interpretation of the results. While it is possible by the procedure outlined here to learn whether or not the absorbancies are consistent with the postulated reaction equilibrium, linearity of the graph does not of itself constitute proof that Eq. (1) represents the true state of affairs. It has in fact been shown[3,4] that a functional relationship among c_1, c_2, and A_3 identical with that of Eq. (9) will exist for a system described by two stages of complex formation,

$$M + X = MX \quad K_1 = (MX)/(M)(X)$$



$$MX + X = MX_2 \quad K_2 = (MX2)/(MX)(X)$$

if the ratio of the molar absorbancies of MX and MX_2 happens to satisfy a certain condition. (In this case A_3, found experimentally as before, stands for the total absorbancy due to complexed M.) The best procedure, therefore, when it can be used, is to make measurements on more than one of the absorption bands due to the complex. The necessary condition on the ratio of the absorbancies is likely not to be satisfied at each of several widely different wavelengths, so that it will then become obvious if the system is not obeying the equilibrium equations corresponding to Eq. (1).

Finally, a few comments will be made about the principle and operation of the spectrophotometer.[5] The function of a spectrophotometer is to produce monochromatic light of a selected wavelength and to measure the intensity of light transmitted by a solution relative to that transmitted by a sample of the pure solvent. The source is a tungsten lamp. The wavelength to be used is selected by turning a knob which moves the prism until light of the desired wavelength is directed toward the slit. The wavelength scale is mechanically coupled to the same shaft. The slit, an aperture of adjustable width, serves to admit only light of a narrow band of wavelengths; at the same time, it

necessarily determines the intensity of light passed. As the slit is widened to provide greater intensity of light for the measurement, a wider range of wavelengths is permitted to pass through. At wavelengths where the transmission of a sample varies rapidly with wavelength, the slit width should be kept as narrow as possible, consistent with the need for adequate intensity.

With the shutter closed, the so-called dark current of the phototube is balanced out so as to give a meter deflection corresponding to zero percent transmission. With the shutter open, and the pure solvent sample in the light path, the sensitivity and slit-width controls are adjusted to give a deflection corresponding to 100 percent transmission. Finally, with the solution sample in the light path, the deflection indicates directly the percent transmission. The output meter scale is also calibrated directly in absorbance.

If the solution transmission is low, better accuracy may be attained by increasing the sensitivity control by one or more steps; if this is done, the scale is altered: for the Beckman model B spectrophotometer, 0.5 must be added to the absorbancy scale reading for each step by which the sensitivity control has been advanced above that at which the reference setting was made with the pure solvent.

For accurate work it is necessary to take into account differences among spectrophotometer sample cells. The first to be considered is that resulting from imperfections in the cell windows. To determine the correction for this effect, the cells are cleaned and filled with nonabsorbing solvent. It is convenient to select the cell with the highest transmission as the reference cell. The slit width is adjusted to give a deflection corresponding to 100 percent transmission with this cell in the light path. The other cells are then placed in turn in the light path, and the absorbancies, designated A_c noted. The value of A_c for a particular cell is called the cell correction; A_c may vary with wavelength. In subsequent measurements with solutions, the true sample absorbancy is then found by subtracting the value of A_c for the cell used from the measured absorbancy of the cell filled with the solution sample.

Second, the optical path lengths may be different for two cells of nominally the same thickness. The path lengths for two cells can most easily be compared by measuring the transmission for both filled with a series of absorbing solutions. The manufacturing tolerances on cell thickness are close enough to permit omission of the path-length correction in most student work.

Suggestions for further work. Combination of Eq. (8) and Eq. (9) give an expression which may be used to calculate values of A2 for each solution after the first approximation to K has been obtained by the procedure given above. Improved values of A_3 may then be calculated, and a new series of points plotted on the original graph. The cycle may be repeated if the new value for K leads to appreciably different estimates for values of A_2. A value for a_2 at the appropriate wavelength may be obtained from measurements on the stock I_2 solution.

References

1. H. A. Benesi and J. H. Hildebrand, J. Am. Chem. Soc. 71:2703 (1949).
2. L. J. Andrews and R. M. Keefer, J. Am. Chem. Soc., 74: 4500 (1952).
3. R. Kruh, J. Am. Chem. Soc., 76: 4865 (1954).
4. T. W. Newton and F. B. Baker, J. Phys. Chem., 61: 934 (1957).
5. W. West in A. Weissberger (ed.), “Technique of Organic Chemistry,” vol. 1, “Physical Methods of Organic Chemistry,” 3d ed., pt. 3, chap. 28, Interscience Publishers, Inc., New York, 1960.
6. O. J. Walker, Trans. Faraday Soc., 31: 1432 (1935).