Letters

Suggestions To Expand the Laboratory Project 'The Sweetness of Aspartame'

In adapting Paul Stein's wonderful project "The Sweetness of Aspartame" *(1)* for use in my upper division biochemistry laboratory, I have expanded the project to include an aspartame standard curve, Beer's law calculations, and non-linear curve-fitting of a hyperbolic saturation curve. Initially, I discovered some problems with the lower concentrations used in the article. Stein recommends that students make up six masses of Equal sweetener dissolved in 50 mL of tap water: A, 0.031 g; B, 0.062 g; C, 0.125 g; D, 0.250 g; E, 0.500 g; and F, 1.00 g. Regarding the two most dilute solutions, A and B, the taste is so indistinct that it is difficult for students to estimate sweetness intensity. Also, using a Shimadzu spectrophotometer I found non-linearity in the aspartame standard curve below concentrations of 0.1 g Equal/50 mL. It is therefore best to avoid these dilute solutions. Accordingly, these masses of Equal sweetener per 50 mL should work well: A, 0.12 g; B, 0.20 g; C, 0.30 g; D, 0.40 g; E, 0.60 g; F, 0.80 g; and G, 1.20 g.

For upper-division laboratories, it is interesting to supply students with pure aspartame and ask them to determine its molar absorptivity at λ_{max}. The standard curve can be generated from a 5.0 mM aspartame stock solution (75 mg/50 mL), and dilutions of 1:2 down to 1:16. Students can then use Beer's law to convert the A_{257} measurements that they record for their Equal solutions (A–G) into aspartame concentrations in mol/L. Given the molecular weight of aspartame, they can calculate the mass of aspartame in each 50 mL solution (A–G), and finally, the mass of aspartame per gram of Equal sweetener in each solution. Students can compare their experimental value to the value given by the manufacturer (3.67 weight %) and cited in Stein's paper. Even more important, because aspartame is the active component of Equal sweetener, it makes sense to use concentration units of mol aspartame/L rather than g Equal/50 mL. This is especially significant for the hyperbolic saturation plot of sweetness intensity (S) versus concentration (ref *1,* Figure 2, p 1113).

For many years, the default plot used to characterize hyperbolic saturation data has been the Lineweaver–Burke double reciprocal plot (in this lab, 1/S versus 1/conc.). The statistical problem with the double reciprocal plot is well-known: experimental errors at low concentrations become magnified in reciprocals. The wide availability of non-linear data fitting in graphing software makes linearization of this type of data unnecessary nowadays. Delta Graph and Kaleidagraph can fit the hyperbolic plot of sweetness (S) versus [aspartame] to the appropriate equation,

$$S = S_{max}/(1 + K_d/[\text{aspartame}]) = S_{max} \times [\text{aspartame}]/([\text{aspartame}] + K_d)$$

using [aspartame] as the x values, S as the y values, and S_{max} and K_d as the fitted parameters. I recommend selecting the non-linear "user-defined" fitting for this project in an upper-division biochemistry laboratory. Knowing S_{max}, students can calculate how close to taste saturation their highest concentration of Equal sweetener (solution G) is. Also, the value of K_d expressed in units of mol/L describes not only the Equal sweetener solution with half-maximal sweetness intensity, but also the dissociation equilibrium constant for the complex between aspartame and the tongue's sweet receptor.

Literature Cited

1. Stein, Paul *J. Chem. Educ.* **1997**, *74*, 1112.

Todd Silverstein
Department of Chemistry
Willamette University
Salem, OR 97301
tsilvers@willamette.edu