ANALYSIS OF RIBOFLAVIN IN AQUEOUS SYSTEMS

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Abstract
We are developing an analytical method for determining the concentration of riboflavin (Vitamin B2) in aqueous samples based on fluorescence spectroscopy. This method will then be adapted to measure riboflavin in mock urine samples and used in undergraduate analytical chemistry courses as described by J.A. Henderleiter and R.M. Hyslop (J. Chem. Educ. 1996, 73, 563-564). The method can be used to track riboflavin uptake and clearance rates in the body. Since the method is being developed with a new spectrofluorimeter (obtained with funds from the National Science Foundation and Drake University), an examination of the capabilities and limitations of the instrument is also being conducted. Presented here are preliminary results for an analysis of riboflavin in water, upon which we can determine the concentration in a control solution to within one percent accuracy.

Why measure riboflavin levels?
Riboflavin deficiency can lead to
- Sore throats and redness and swelling of the lining of the mouth and throat,
- Vascularization of the cornea, and
- Decreased red blood cell count.
To link these symptoms to riboflavin deficiencies, methods for monitoring riboflavin are needed. We are using fluorescence spectroscopy to measure riboflavin concentrations in aqueous solutions and mock urine samples.

What is fluorescence?
- Molecules can absorb electromagnetic radiation (light).
- Different molecules absorb different wavelengths of light.
- After absorbing the light the molecules can re-emit it at different wavelengths.
- This process of absorption followed by emission of light is fluorescent and is depicted below.
- The amount of light emitted is proportional to the concentration of the fluorescent molecules in the sample.
In this way, measuring the intensity of the emitted light can be used to determine the concentration of riboflavin in solution.

Chemical structure of riboflavin

Measuring riboflavin in a control sample
- The calibration curve above and the resulting best straight line equation were applied to an independent control sample with a known riboflavin concentration of 58.75 ng/mL.
- The analysis yielded a measured concentration of 59.27 ± 0.26 ng/mL.
- The percent difference between the known and calculated values is 0.89%.

Conclusion
Using spectrofluorimetry we are able to measure the riboflavin concentration in an aqueous control solution to within one percent accuracy.

The next goal is to create mock urine samples and repeat the analysis mindful of potential interferences that may require the use of the method of standard additions.