

Analytical Estimation of Riboflavin Using Different Buffer Solution by Using UV Spectroscopy

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ABSTRACT: The detection and assay of vitamin B-2 (Riboflavin) was accomplished under aqueous conditions using different buffer solution such as citrate buffer, sodium Borate buffer at different Ph. The absorbance spectrum of Riboflavin was determined at different pH values by using several buffers. The accurate and sensitive assay of Riboflavin is done by spectrophotometer at 440 nm wavelength. Where indicated an origin solution was employed by dissolving sufficient vitamin to make a stock solution of 1.403×10^{-4} molar concentration. Measurement of various aqueous solutions containing Riboflavin were accomplished included aqueous test samples, vitamin capsule/tablet, and water vitamin mixtures. The B vitamin Riboflavin can be assayed by using UV/VIS spectrophotometry at 440 nm in aqueous media by using different buffer solution.

KEYWORDS: Riboflavin , citric acid buffer, sodium borate buffer , phosphate buffer

I. INTRODUCTION:

The name vitamin is derived from a Latin word "vita" which means life. Riboflavin (vitaminB2) is a yellow- green fluorescent water soluble vitamin an irresponsible for imparting yellow colouration to B vitamin preparations. Vitamins usually act as catalysts or co-enzymes or Often are essential parts of coenzymes. The sources of vitamins include Dairy products, fishes, rice, wheat, egg yolk ,vegetables, fortified cereals, chicken and various meat. Ascorbic acid protect Riboflavin from degradation. There are different diseases that are associated with the deficiency of each vitamin.[1]

Vitamins are divided into water-soluble and fat-soluble vitamins on the basis of their solubility and chemical nature. Some analytical methods based on UV-V is spectrophotometry and

spectrofluorimetry have also been optimized for the simultaneous quantification of vitamins. However, these methods usually require some chemical reaction or separation steps, 7-9 where sample manipulation can increase the risk of human error in the results and the cost of the analysis. When consumed this vitamin is not considered to be toxic partly due to its lower aqueous solubility [2] and because excess amounts of the vitamin is readily excreted into the urine [3].

Riboflavin has been popular as a supplement in the form of tablets or capsules and is often utilized in energy drinks or similar health-related beverages. There have been few studies conducted for applying Riboflavin in clinical and therapeutic scenarios. For example, Riboflavin in higher dose has been found to help and prevent migraine headaches [4,5]. Simultaneous dose of Riboflavin and uv light with blood products has been found to reduce harmful activity of pathogens through reduction of replication [6]. Riboflavin is considered effective against nuclear cataracts [7]. Various methodologies were investigated for the determination of Riboflavin.

These include isocratic reversed-phase column high-performance liquid chromatography separation followed by fluorometry detection of the analyze [8]. Fluorescence detection following high-performance liquid chromatography [9,10]. Other work showed an assay with straight fluorescence following pre-treatment of sample [11]. Simultaneous detection of various vitamins, including Riboflavin were found to be feasible utilizing planar chromatography followed by an application of fluorescence, ultraviolet-visible detection and confirmation with electro spray ionization mass spectrometry Riboflavin vitamin.

II. MATERIAL AND PROCEDURE:

2.1 Materials:

2.1.1 Riboflavin Drug Standard was a generous donation from Popular Pharmaceuticals Ltd. in India.

2.1.2 Dosage method: Riboflavin tablets (10mg) were obtained from a local drug store in Dharmapuri. The samples manufacturing license numbers, batch numbers, production, and expiry dates were all double-checked. They were coded A, B, and C at random and stored properly.

2.1.3 Reagents : Buffer solution (citric acid buffer , sodium borate buffer ,Phosphate buffer) these buffers are employed in this study and distilled water are also employed in this study.

2.1.4 Instruments :

Single pan balance, UV/visible spectrophotometer.

2.2 Procedure :

2.2.1. By using citric acid buffer :

Preparation of standard and test solution:

Riboflavin solid consists of orange crystals and produces a yellow aqueous solution. A stock solution of Riboflavin were prepared by dissolving 0.0526 g into one liter of distilled water making a conc. of 1.403×10^{-4} molar. This container was wrapped in aluminum foil to protect Riboflavin stock solution from light exposure. In the case of Tablet or capsule:

- 1) The tablet is weighed
- 2) Then ground in mortar and pestle
- 3) The dry amount of solid should be dissolved in volumetric flask.
- 4) The desired amount of solid is carefully placed in volumetric flask and dissolved in distilled water
- 5) Then filter in gout insoluble solid through Whatman #1 filter paper
- 6) The filtered liquid is ready for assay.

2.2.2. By using sodium borate buffer:

Preparation of standard and test Solution:

Riboflavin solid consist of orange crystals and produce yellow aqueous solution. A stock solution of Riboflavin was prepared by dissolving 0.0526 g in one liter of distilled water making a conc. of 1.403×10^{-4} molar. This container was wrapped in aluminum foil to protect Riboflavin stock solution from light exposure. Stock solution

of sodium borate buffer of 0.110 molar were prepared in distilled water at pH value of 7.54

To as certain feasibility for measuring industrial or beverage aqueous vitamin mixtures, as In the case of tablet or capsule,

- (1) The tablet gets weighed
- (2) Then ground in mortar and pestle
- (3) The dry amount of solid to be dissolved in volumetric flask
- (4) The desired amount of solid is carefully placed in volumetric flask in distilled water
- (5) Filter out of insoluble solids through Whatman#1 filter paper
- (6)The Filtered liquid is ready for assay

2.2.3. By using phosphate buffer:

Preparation of standard and test solution:

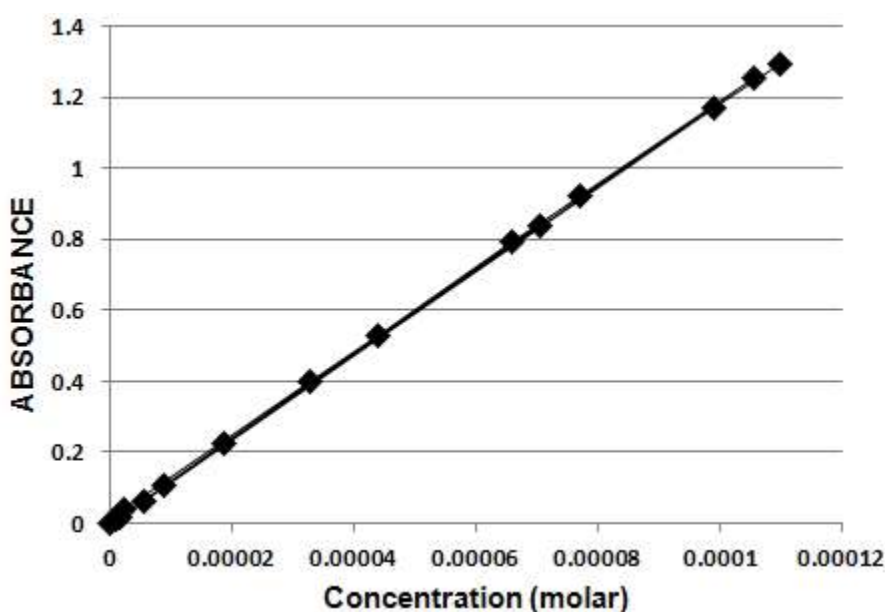
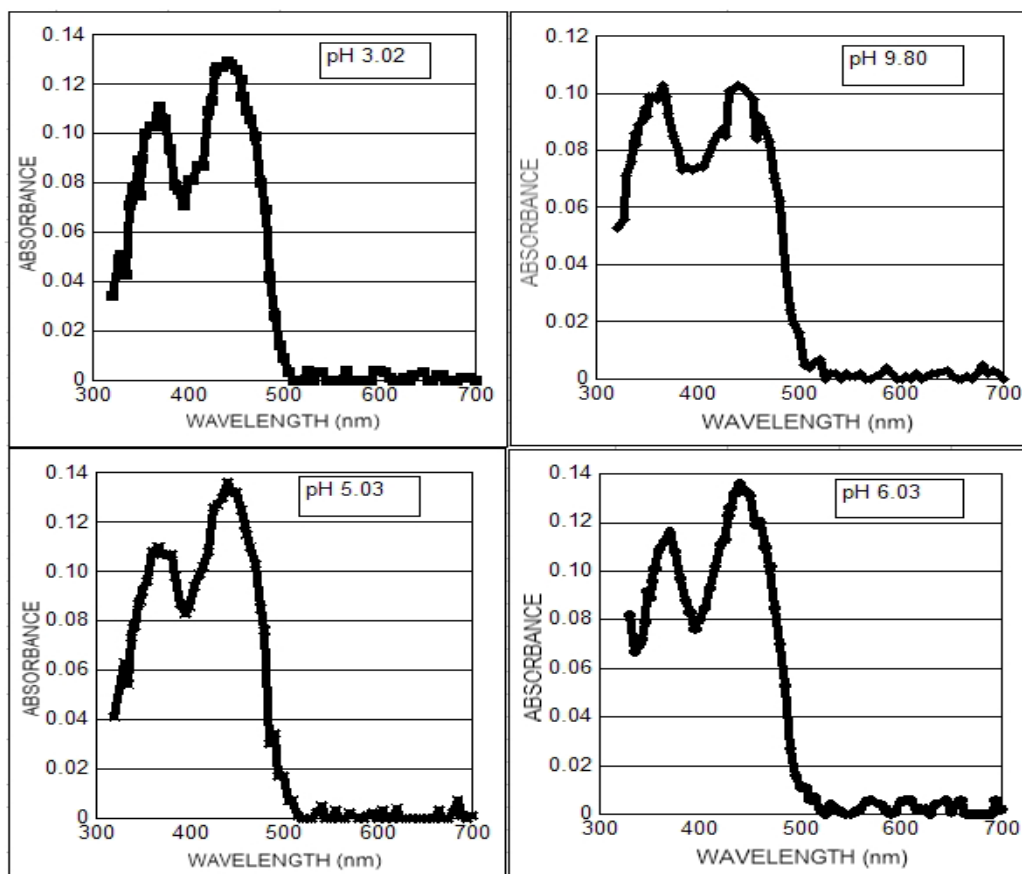
Riboflavin solid consists of orange crystal and produce yellow aqueous solution. A stock solution of Riboflavin were prepared by dissolving 0.0528 g in one liter of distilled water making a conc. Of 1.403×10^{-4} molar. This container was wrapped in aluminum foil to protectsssss Riboflavin stock solution from light exposure. Stock solution of Phosphate buffer of 0.010 molar was prepared in distilled water at pH value of 6.8. In the case of tablet or capsule:

- 1) The tablet were weighed
- 2) Then ground in mortar and pestle
- 3) The dry amount of solid to be dissolved in volumetric flask
- 4) The desired amount of solid is carefully placed in volumetric flask and dissolve in distilled water
- 5) Filter out insoluble solid through Whatman#1 filter paper
- 6) The filtered liquid is ready for assay

III. RESULT

3.1 By citric acid buffer:

Riboflavin consists of a yellow to orange yellow crystal like powder and has slight odor. The solid is not significantly affected by light but when in solution, the vitamin quickly degrade under light. Percent recovery of Riboflavin was on average of 101.9%, having standard deviation of 1.1%.The min percent recovery was 100.25 and max of 103.7% . Median percent recovery was 102.8% and values are approximately symmetric with scenes of 0.152.Coefficient of variation for percent recovery is 1.1%.



Standard curve showing line equation $y = 11872x$ with $R^2 = 0.9988$ and Pearson correlation $r = 0.9998$.

3.2 By using sodium borate buffer:

Riboflavin has a variable but low solubility in water approximately 1 mg in 20 ml

with the variation due to differences in the internal crystalline structure of Riboflavin [2]. The equation of the line was $y = 12546x$ (including origin) and with a correlation coefficient of essential $r = 1.100$ with a coefficient of determination $R^2 = 1.100$. The standard deviation of slope is 1.66 and the 95% confidence interval for the slope is from 12533 to 12565.

3. 3 By using phosphate buffer:

Riboflavin consist of a yellow to orange yellow crystal like powder. The solid is not significantly affected by light but when in solution, the vitamin quickly degrade under light. Percent recovery of Riboflavin was on the average of 105.9% ,having standard deviation of 1.2%. The min percent recovery was 103.25 and max of 108.7%. Median percent recovery was 107.8% and value is approximately symmetric with skew of 0.151. Coefficient of variation for percent recovery is 1.1%.

IV. CONCLUSION :

The measurement of various types of aqueous mixture which contain Riboflavin were accomplished. These include aqueous test samples, vitamin capsule/tablet and water vitamin mixtures. This approach is fast, broadly applicable due to the aqueous solubility of Riboflavin, easily applied, facile for use in quality control and efficient for manufacturing. I conclude that vitamin B-2 (Riboflavin) can be assayed by using UV/VIS spectrophotometry at max 310 nm - 440nm in aqueous media by using different buffer solutions at different Ph values.

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