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Assay of paracetamol in tablets

Introduction

A method for quantification of paracetamol in a tablet formulation by Ultra-Violet-Visible Spectrophotometry (UV-Vis) using a UV-1900i and an external standard is described. **UV-Visible spectrophotometry** is one of the most frequently employed technique in pharmaceutical analysis. It involves measuring the amount of ultraviolet or visible radiation absorbed by a substance in solution.

A series of standards was prepared by making a 100ppm stock solution and preparing a series of working standards to generate a 6-point calibration curve. The paracetamol is detected using UV detection at a wavelength of approximately 243nm. Quantification is achieved by scanning each sample against the calibration curve.

Reagents

Paracetamol	-	Harmful if ingested in quantity; irritant; reproductive effects
Methanol	-	Toxic if swallowed, harmful to organs,
Water	-	Wear PPE like lab coats, gloves and safety specs when handling

Safety

Avoid skin and eye contact with reagents by wearing a lab coat, gloves and safety glasses. Do not expose acetonitrile to a source of ignition. Avoid inhalation of methanol vapour. Provided the recommended precautions are adopted, the risk to operators during this procedure is minimal.

Apparatus

Balance capable of weighing 0.0001 g
Double beam UV-Vis Spectrophotometer
Mortar and Pestle
Weighing boats
Methanol
Distilled Water
Volumetric Flasks (100mL and 50mL)
Ultrasonic bath

Table 1: UV-Visible Spectrophotometer Parameters: [Spectrum]

Wavelength Range:	400-200 nm
Data Interval:	1 nm
Scan Speed:	High Speed
Measurement Type:	Absorbance

Table 2: UV-Visible Spectrophotometer Parameters: [Quantitation]

Wavelength Range:	λ_{max} [Spectrum]
Accumulation Time:	0.1 seconds
Calibration Equation:	$Calculated\ Value = K_1 \times Concentration + K_0$
Measurement Type:	Absorbance

Preparation of Diluent:

Prepare 500 mL of 15% methanol diluent. Once made up, use an ultrasonic bath to degas the diluent for 10 minutes. This will be used to prepare the stock and working standards as well as the samples.

Preparation of standard solutions[1]

Use a calibrated balance and one-mark pipettes to prepare standard stock solution and dilutions. All standards should be prepared in 15% methanol.

To prepare the solution, Dissolve 10 mg of a paracetamol reference standard in a minimum amount of the previously prepared diluent and ensure the standard is completely dissolved. Once fully dissolved, make up to the mark to 100 ml, establishing a concentration of 100 parts per million (ppm). From this prepared solution, prepare a series of working standards for a 6-point calibration. (0, 20, 40, 60, 80, 100 ppm)

Preparation of sample (tablet) solution[1]

Use a calibrated balance and one-mark pipettes to prepare sample solution and dilutions. All solutions should be prepared in 15% methanol.

Weigh 20 tablets (each tablet contains 500 mg of paracetamol). Powder one tablet and weigh a quantity equivalent to 100 ±10 mg of paracetamol (weigh by difference). This should be done in triplicate. Transfer the weighed samples into 100 ml volumetric flasks and make up to the mark with the diluent. Shake and degas each 100 mL flask for 10 minutes. From each flask, extract 1 mL and transfer to a 100 mL volumetric flask. Make up to the mark with the diluent.

Analysis of samples and standards

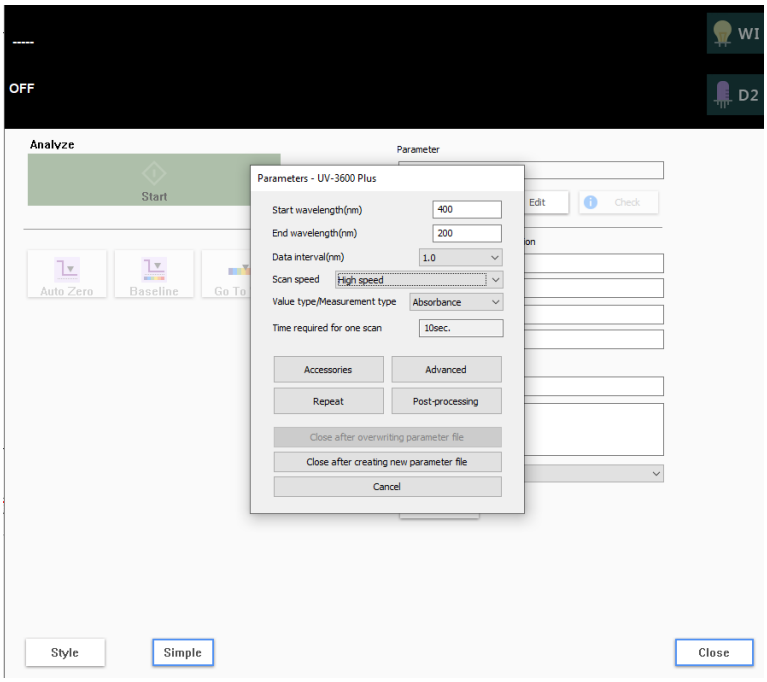
Scan each of the calibration standards and sample dilutions and analyze using the conditions described above – *see appendix 1*.

Construct a calibration curve and determine the amount of paracetamol in the tablet. – *see appendix 2*.

Calculate the % of stated amount present in the tablet.

Appendix 1 – LabSolutions UV-Vis: Spectrum

1) Load the **[Spectrum]** module and connect the instrument by clicking **[Connect]**
2) When the Instrument control panel launches, press **[Edit]**
3) Set parameters in accordance to the conditions in Table 1



4) Press [Close after creating new parameter file] and name the parameters
5) Fill in the file name and sample information
6) Fill 2 cuvettes with 15% MeOH and place in both cell holder
7) Baseline the UV-Vis Spectrophotometer
8) Remove the cuvette from the front cell holder, leaving the rear cuvette in place
9) Place your sample in a cuvette and place in the front cell holder
10) Close the lid and press sample scan
11) Once the sample scan is complete, click on the **[Active]** tab
12) Click on **[Point Pick]**
13) Move the black bar to λ_{max} at approximately 243nm and click **[Add]** to report the Abs value at λ_{max}.
14) Repeat this for the rest of your calibration standards.

Appendix 2: LabSolutions UV-Vis: Quantitation Calibration

Configuration of the measurement parameters may be performed by a laboratory technician in advance of students commencing the experiments. The method is as follows:

1) Launch *LabSolutions UV-Vis,* then select **[Quantitation]**
2) Select the **[Inst. Control]** icon from the toolbar to display the instrument control window
3) Click **[Parameter] – [Edit]** to display the **[Set Parameters]** window.
4) Click **[Registration of Wavelength]** to display the [Register Wavelengths] window, then select '*Point'* for **[Measuring Method]** and type in the λ_{max} that you found by scanning your calibration standards in spectrum. Finally select [Add], then click [OK].
5) Click **[Calibration Curve]** to display the [Calibration Curve] window and select: '*Measure a standard sample',* '*Fixed Wavelength'* for **[Calculation Method]** and '*WL* λ_{max}*'* for **[Wavelength 1]**. Then, click **[OK]**.
6) Click [Standard Sample], then select '*Acquire a measuring mode by measurement'.* Then, click [Ok].
7) Click **[Unknown Sample],** then select '*Acquire a measuring mode by measurement'.* Then, click **[OK]**.
8) Click **[Close after creating new parameter file]** in the **[Set Parameters]** window. Enter a file name and click **[Save]**.

For Students:

1. Quantitation Procedure

Specifying Parameter File

1) Launch *LabSolutions UV-Vis,* then select **[Quantitation]**
2) Click the **[Connect]** icon from the toolbar to connect the spectrometer, this will automatically launch the instrument control panel upon successful connection
3) Click **[Parameter] – [Read]** and select the file generated in *Appendix 2.*

Specifying File Names

4) In the instrument control window, click **[File name] – [Edit]**
5) Enter the details for the **[Filename]** *(where the data is to be stored)*, **[Analyst]** and **[Comment]** and click **[OK]**. Leave the instrument control window open.

Measuring Standard Samples

6) In the Standard Table enter the calibration standard solutions and their concentrations.
7) Click in the standard sample table to activate it *(table becomes enclosed in a red frame)*
8) Set the blank sample (15% MeOH) in the instrument's sample compartment.

9) In the instrument control window, click  **[Baseline]**.
10) Place the first standard sample in the instrument's sample compartment and close the lid.
11) In the instrument control window, click  **[Standard]**. The measurement value is displayed in the wavelength column.
12) Repeat the same operations for the remaining prepared standard samples.
13) Once measurement of all standard samples is complete, click **[Save]** on the toolbar. With all standards scanned, a calibration curve will be generated on the right of the screen.

Measuring Unknown Samples

14) Click in the unknown sample table to activate it *(table becomes enclosed in a red frame)*
15) Enter the sample information.
16) Place the first sample in the instrument's sample compartment and close the lid.
17) In the instrument control window, click  **[Unknown Sample]**.
18) Repeat the same operations for the remaining prepared samples.
19) The concentration calculated using the calibration curve is displayed in the concentration column and will be shown after the scan.

References:

1. **https://www.omicsonline.org/uv-visible-spectrophotometric-method-development-and-validation-of-assay-of-paracetamol-tablet-formulation-2155-9872.1000151.php?aid=9714**