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- Subject: Advance Instrumentation Techniques (BP-803TE)
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PROCEDURE OF CALIBRATION

1. Control of wavelength
2. Control of absorbance
   - For ultraviolet region
   - For visible region
   - Limit of stray light
   - resolution power
3. Maintenance
4. Precautions
5. Abbrevations

CONTROL OF WAVELNGTH

CHEMICALS: 1.4M perchloric acid,

holmium perchlorate solution

STEP 1: Dissolve 1 gram of holmium oxide in 1.4M perchloric acid with the aid of heating on water bath, cool and dilute to 25ml with same solvent.

STEP 2: Record the spectrum holmium perchlorate solution from 200nmto 600nm using 1.4M perchloric acid as reference solution

CONTROL OF ABSORBANCE

STEP 1:Weigh 57 to 63mg of potassium dichromate primary standard and transfer to 1000ml volumetric flask. Dissolve in 0.005m sulphuric acid and make up to the mark with the same acid.

Step 2: Measure the absorbance at 235 nm, 257nm, 313nm and 350nm using 0.005M sulphuric acid as reference

- **FOR VISIBLE REGION:** Whole procedure is same as UV region but at the end measure the absorbance at 430nm.
- **CALCULATION:** Value of A(1%,1cm)

$$A(1\%,1cm) = \frac{\text{absorbance} \times 100}{\text{weight in gm} \times 100}$$

LIMIT OF STRAY LIGHT

STEP 1: Prepare the solution of 1.2%v/v potassium chloride and dissolve with 50ml distilled water.

STEP 2:Determine the absorbance using path length of 1cm at 200nm against purified water as blank

Acceptance Criteria: Absorbance at 200nm should be Greater then 2.0

RESOLUTION POWER

STEP 1: Prepare a solution 0.02%v/v toluene in hexane

STEP 2: Record the spectrum of 0.02%v/v toluene in hexane from 250nm – 300nm using hexane as reference

STEP 3: record the absorbance at 269nm (max) and 266nm (min)n

STEP 4: Calculate the ration of absorbance by dividing the absorbance at maxima and minima

ACCEPTANCE CRITERIA: Absorbance ratio at 269nm to 266nm is not less then 1.5

WAVELENGTH ACCURACY

- Deviation of wavelength reading at an absorbance band

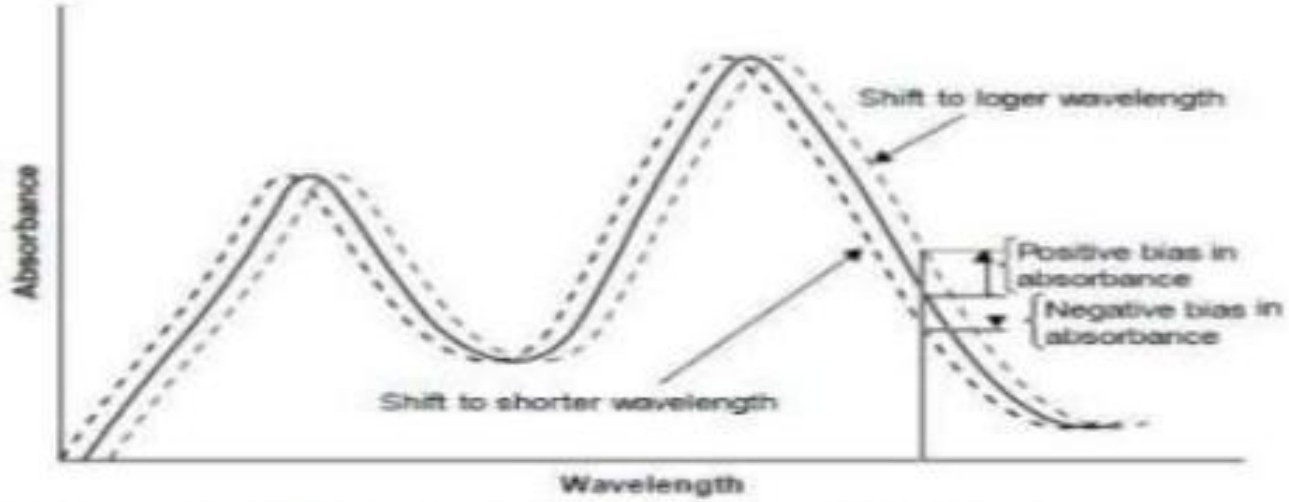

Figure 10.2. Effect of wavelength accuracy on UV–Vis measurements.

Table I

S.No.	Wavelength (nm)	Absorbance E (1%1cm)	Maximum tolerance
1.	235	124.5	122.9 to 126.2
2.	257	144.0	142.8 to 145.7
3.	313	48.6	47.0 to 50.3
4.	350	106.6	104.9 to 108.2
5.	430	15.9	15.7 to 16.1

UV APPARATUS

SHIMADZU UV 1601

SHIMADZU UV 2600

- Note down the maxima observed at wavelength against the acceptance criteria given below:

S.No.	Maxima Wavelength (nm)	Tolerance (nm)
1.	241.15nm	240.15nm to 242.15nm
2.	287.15nm	286.15nm to 288.15nm
3.	361.5nm	360.50nm to 362.50nm
4.	536.3nm	533.30nm to 539.30nm

UNIT& TOPIC-III/01

Content

SOP-FTIR

- **1.0 OBJECTIVE**

To describe the procedure for operation and calibration of FT-IR Spectrometer.

- **2.0 SCOPE**

This SOP is applicable for the operation and calibration of FT-IR Spectrometer.

- **3.0 RESPONSIBILITY**

Officer/ Executive - Quality Control.

- **4.0 ACCOUNTABILITY**

Manager - Quality Control

5.0 PROCEDURE

- **5.1 Procedure for General Cleaning**

5.1.1 Ensure that the power to the instrument is switched OFF before cleaning Clean the instrument with clean dry cloth daily.

5.2-Operation of Instruments

- 5.2.1 Ensure that the instrument is properly connected to the power supply.
- 5.2.2 Switch ON the main switch and power switch of the instrument situated at backside panel.
- 5.2.3 Switch the computer ON by the switches situated on right of the monitor and PC respectively.
- 5.2.4 Switch on the instrument, kept it for 10-15 minutes to get warm.
- 5.2.5 Click on software & select collect mode
- 5.2.6 In collect mode you select background & put file name or as per your requirement (in case of DRA assembly put KBr powder).
- 5.2.7 Now click on 'ok collect' so that it will scan background spectra.
- 5.2.8 After completion of background spectra, put sample pellet (in case of DRA put mixture of KBr & sample).

5.3 For Solids

- 5.3.1 Preparation of potassium bromide disc: Take about 200 mg of potassium bromide in pestle and mortar and grind it to a fine powder. Add about 2-3 mg of substance being examined and again grind to fine powder, Mix well the contents
- 5.3.2 Clean the Die and it components with soft cloths or tissue paper. Do not use a cloth having hard abrasive texture to wipe to wipe the polished surface.
- 5.3.3 Ensure that the rubber seal is properly fixed in the groove of the die.
- 5.3.4 Carefully place on the pellets (polished face in up side position) into the bore of the cylinder .
- 5.3.5 Using a clean thin stainless steel spatula, transfer the fine powder sample + Kbr preparation in the bore of the cylinder. Tape the side of die gently so that the powder is evenly distributed across the face of the police face. Insert the second pellet with police facing down.
- 5.3.6 Place the die assembly under “hydraulic press “ Evacuate die assembly. Then apply pressure of 10 tons to the plunger to produce transparent disc.
- 5.3.7 Remove die base from cylinder leaving plunger in released position.
- 5.3.8 Put cylinder bore below the cylinder and place them “hydraulic press” Apply pressure across the plunger until the pellet immersed from the cylinder followed by disc .
- 5.3.9 Remove the disc carefully and put into plastic tweezer switch is further fixed into the sample compartment of the instrument

5.4 For Liquids

- 5.4.1 Prepare the blank and sample solution as directed in the STP.
- 5.4.2 Fill the blank / test solution into the cell, preferably with a syringe.
- 5.4.3 Rinse the cell 2 times with the solution and finally fill the cell taking care that no air bubble is trapped inside.

5.5 For Sample in Mineral Oil Dispersion

- 5.5.1 Take about 200 to 300 mg of the sample in the clean mortar and add about 3 to 5 drops of liquid paraffin (AR grade) and grind it to a fine paste. Mix the contents properly.
- 5.5.2 Clean the sodium chloride cell with a tissue paper mildly so that no fiber /small piece of tissue paper stick to it.
- 5.5.3 Hold a cell horizontally and pour 2 to 3 drops of the sample preparation on it and spread it evenly slightly tilting.
- 5.5.4 Place the second cell horizontally on the first and press it mildly so that it stick together.
- 5.5.5 Now place these cell horizontally, inside the window and tighten the screws

5.6 Scanning

- 5.6.1 Open the sample compartment by raising the left hand side cover of spectrum one FT-IR Spectrometer and insert the sample holder into sample the compartment.
- 5.6.2 Close the sample compartment
- 5.6.3 Click the scan display on right sight of the screen.
- 5.6.4 The spectrum appears on the screen.
- 5.6.5 Compare the spectrum obtained with standard spectra that is stored in the computer, by proceeding as follows.
- 5.6.6 Select file on the top left hand side then open.
- 5.6.7 In open choose the relevant spectra and clicking the one that is needed . If relevant standard spectra not available in computer scan standard also with same operation parameter.
- 5.6.8 Two spectra are displayed on the screen, the test in black colour and the standard over laying it in blue colour.
- 5.6.9 After comparison take the spectra by print command.
- 5.6.10 The spectrum is printed by the printer.
- 5.6.11 Current official reference standard shall be chronogram and the spectra shall be kept in the file.
- 5.6.12 These spectra shall be updated once every year /or whenever there change in lot no. of the official reference.

5.7 Calibration

- 5.7.1 Switch on the system.
- 5.7.2 Set instrument parameter as follows.
- RESOLUTION 2.0
- APODIZATION STRONG
- RANGE 4000-400 cm-1
- MODE RATIO
- NUMBER OF SCAN 16
- 5.7.3 Allow the instrument to warm for 30 minutes.
- 5.7.4 Operate the instrument as per operation SOP.
- 5.7.5 Open the sample compartment cover of FTIR 1600 and place the polystyrene film in the sample holder and the close the cover.
- 5.7.6 Click OK in the SCAN MODE.
- 5.7.7 The spectrum of the polystyrene film is displays on the screen.
- 5.7.8 PRINT the spectrum by going to print function.
- 5.7.9 When the printer of the spectra is completely go to DATA and then to PEAK.
- 5.7.10 The peak data's of the spectra of the polystyrene film is displays on the screen.
- 5.7.11 PRINT out these data's by going to print function.

5.8 Frequency of Calibration

- 5.6.1 Once in three month and after each maintenance job

6.0 ABBREVIATIONS

- 6.1 SOP - Standard Operating Procedure
- 6.2 Kbr - Potassium Bromide
- 6.3 FTIR - Fourier Transforms Infra Red
- 6.4 AR - Analytical Reagent Grade

ANNEXURE-I

ANNEXURE-I

FT-IR USAGE LOG BOOK

Sr.No	Date	Product/ Material Name	Batch No./ Lot No.	Time		Analyzed by	Remarks
				Started at	Completed On		

ANNEXURE-II

ANNEXURE-II
CALIBRATION REPORT OF FTIR

Asset ID No.	:	**Ref. SOP No.**	:
Make	:	**Model No.**	:

Limit of Wave Number Accuracy :
By Polystyrene Film
OBSERVATION :

Sr. No.	Wave Number	Observation	Limit	Remark
1	3060.0 (±1.5)		(±1.5)	
2	2849.5 (±1.5)		(±1.5)	
3	1942.9 (±1.5)		(±1.5)	
4	1601.2 (±1.0)		(±1.0)	
5	1583.0 (±1.0)		(±1.0)	
6	1028.3 (±1.0)		(±1.0)	

Resolution Performance
By Polystyrene Film
OBSERVATION :

Sr. No.	Wave Number	Absorbance	Transmittance Deference Between X and Y	Limit Greater Than in %	Remark
	2870.0			18	
	2849.5				
	1589.0			12	
	1583.0				

Remarks : The instrument is working satisfactorily/not satisfactorily as per SOP.
Next calibration due on :

Date of calibration	Calibrated by (name/sign)	Checked by (name/sign)/on

REFERENCE

- http://www.pharmaguideline.com/2011/02/sop-for-operation-and-calibration-of-ft.html