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CHEMISTRY

INSTRUMENT: UV-VISIBLE SPECTROPHOTOMETER

MAKE: SHIMADZU MODEL: UV-1800

PROCURED ON: 18-06-2013

SUBJECT: SOP FOR

UV-VISIBLE

SPECTROPHOTOMETER

SOP NO.: BCP/PC/SOP/004

PAGE NO.: 01-02

EFFECTIVE DATE: 01/01/2022

REVIEW PERIOD: 31/12/2022

Objective:

The following document describes the standard operating procedure for UV-Visible spectrophotometer..

Scope:

UV-Visible Spectrophotometer is used to detect maximum absorbance for given sample, unknown concentration of sample, carry out calibration studies, etc.

Procedure:

- 1. Plug in to ensure the power supply.
- 2. Switch **ON** the main power supply and instrument mains.
- 3. The instrument will auto ensure that the program is functioning properly by displaying **OK** on the screen. This will take approximately 3-4 min.
- 4. From the Configure drop-down menu, select Parameters.
 - i. You may use the default parameters
 - ii. **OR** Adjust Wavelength Range before starting the test. Wavelength range is between 200-800 nm.
 - iii. Recording Range can be changed at any time. It is recommended to set Scan Speed to fast and Sample Interval to Auto.
- 5. For determining maximum wavelength for the given sample, go to **SPECTRUM** mode.
- 6. Select the wavelength scanning range between 800-200nm.

- 7. Fill the two cuvettes with blank solution and place both of them in two separate cuvette holder.
- 8. Click on **BASE** Correction to detect and nullify any background interference.
- 9. After the base correction is done the instrument will beep.
- 10. Replace one of the cuvettes with the sample under study.
- 11. Press the START button and wait till the scanning is completed.
- 12. A graph will be displayed on the screen with the highest peak as the maximum absorption for the given sample.
- 13. For obtaining linear curve/ calibration curve, go to **PHOPTOMETRIC** mode.
- 14. Clink on **GO TO WAVELENGTH** and set the maximum absorbed wavelength.
- 15. Fill the two cuvettes with blank solution and place both of them in two separate cuvette holder.
- 16. If some amount of the light will be absorbed by the blank, the reading will be displayed on the screen, then click on Auto Zero and wait until it reads 0.000A
- 17. Replace one of the cuvettes with the different serially diluted samples under study.
- 18. Note the absorbance values for each diluted sample and plot a graph of **ABSORBANCE** v/s **CONCENTRATION**.

Precautions:

- ➤ Before start of the instrument, ensure to remove silica bag from the cuvette holder chamber.
- Ensure that cuvettes are cleaned thoroughly after every using.
- > The concentrations of the solutions under study by dilute so as to obtain accurate results.

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