

 <p>BLDEA'S Shri Sanganabasava Mahaswamiji College of Pharmacy & Research Centre Vijayapur 586103</p>	INSTRUMENT NAME – DOUBLE BEAM UV VISIBLE SPECTROPHOTOMETER	SOP NO.: BCP/CIL/SOP/ 004
	MODEL – ELICO	PAGE NO.: 1-3
	METHOD – SL 160	EFFECTIVE DATE: 01/01/2022
	SUBJECT: SOP UV VISIBLE SPECTROPHOTOMETER	REVIEW PERIOD: 31/12/2022

- ✓ **PURPOSE:** The purpose of this standard operating procedure (SOP) is to describe the operation of the uv visible spectrophotometer.
- ✓ **OBJECTIVES:** To describe the procedure for analysis on UV visible spectrophotometer.
- ✓ **SCOPE:** This machine is used to measure how much light of a given wavelength is absorbed by a liquid sample.
- ✓ **PRECAUTION:**
 - ✓ Limit access to areas where UV sources are used.
 - ✓ Post warning signs at the entrance to labs or other work areas using UV sources.
 - ✓ Wear protective eyewear and gloves.
 - ✓ Cover arms and neck and limit exposure time.
 - ✓ Never look directly at the beam
- ✓ **Standard Operating Procedure:**
 - Open panel door and make sure cuvette holder are empty, then close the panel door.
 - Turn spectrophotometer “ON” by flipping the yellow switch on the side of the machine.
 - The machine will automatically initialize and make a baseline correction.
 - Select “8” **CONDITION SET**, and then press **ENTER**.

- Select “5” for **LAMP SELECT** to turn off Uvlight bulb, then press **ENTER**.
- Press the **FILE**, Key.
- Select one of the following numbers appropriate for the sample being tested. Select “3” for **E.COLI**
 Select “7” for **PICHIA**
 Select “8” for **CHO**
 Select “10” for **PROTEIN**
- Once you have made your selection press **ENTER**.
 It will then ask you for a “**Parameter Change Y/N**”, Select **NO**, and press **ENTER**
- Fill 2 of the same cuvettes each with about 2mL of blank solution. Hold the cuvette from the top to prevent tampering with the measurements, and wipe the sides with lab tissue.
- Open panel door and place the cuvettes with blank solution in the cuvette holders. **Make sure to use the appropriate orientation for the cuvettes you’re using. Also make sure that the cuvettes used for the auto zeroing are the same cuvette you use for the sample reading.** If using a standard cuvette, see fig 9.3, any orientation of the cuvette in the holder is acceptable, just make sure you wipe the cuvette’s sides. If using a micro cuvette, see figure 9.3, the micro cuvette
- Press the **AUTO ZERO** key, and then press **ENTER**.
- When the Auto Zero is complete, open the panel door and remove the **front Cuvette. MUST** be oriented in the holder so the 1cm path length goes from left to right.
- Do not replace cuvette in rear holder.
- Using the same cuvette style, fill an empty cuvette with about 2ml of the sample.
- Clean the cuvette with a lab tissue.
- Place in front cuvette holder, using the appropriate orientation and close

the panel door.

- Press **START** to take a reading.
- Record the results or press **COPY** for a hardcopy print out.
Note: If the initial sample OD reading is greater than 1.0, the sample should be diluted until it reads below 1.0 and then multiply by the dilution factor to obtain the absorbance value.
- Open panel door and remove test sample from front cuvette holder.
- To test additional samples : Place cuvettes in front holder and press start for a reading.
- Record results, or press **COPY** for a hardcopy print out.
- Press **RETURN** to bring you back to step Note: This will erase your old data.
- Press **FILE** to return to the original screen.
- Remove cuvettes remaining in holders.
- Flip power switch located on the side, to turn off the machine.
- Ensure that instrument is clean and free from dust.
- Now fill the dried methanol up to the level to touch the sensors.
- Also, fill the methanol bottle and Karl Fischer reagent bottle.
- Switch "ON" the main and start the stirring.
- Now press the "START" button of the System to perform BLANK neutralization of Dried Methanol. After completion of Neutralization, a beep sound will appear.

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