



**AVIT**  
AARUPADAI VEEDU INSTITUTE OF TECHNOLOGY



VINAYAKA MISSION'S  
RESEARCH FOUNDATION  
(Deemed to be University under section 3 of the UGC Act 1956)



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DEPARTMENT OF BIOTECHNOLOGY

**481171 L 1-ADVANCED BIOCHEMISTRY LABS**

## **STANDARD OPERATING PROCEDURE**

Name of the Lab./facility	<b>BIOCHEMISTRY AND BIO INSTRUMENTATION LAB</b>
Purpose	To describe the procedure for the operation and maintenance of the Soxhlet apparatus
Scope	This Standard Operating Procedure (SOP) applies to the staff and students using the Soxhlet apparatus in the Bio chemistry and Bioinstrumentation laboratory, Vinayaka missions' Research foundation
Responsibility	<p>It is the responsibility of Lab – In-charge of the lab to train Lab Assistant and students on this procedure and to ensure adherence to this procedure.</p> <p>It is the responsibility of the students/technicians to follow the SOP as described and to inform the Lab In-Charge about any deviations or problems that may occur while performing the procedure.</p>

### **STANDARD OPERATING PROCEDURE FOR SOXHLET APPARATUS**

- Assemble the apparatus.
- Fill the round bottom flask with solvent (n-Hexane).
- Put the thimble containing sample into extraction tube.
- Attach the extraction tube with flask containing solvent.
- Attach a condenser unit with the extraction tube and run the water.
- Fix the soxhlet apparatus on hot plate and heat the flask containing solvent.
- The solvent starts to evaporate and falls in the extraction tube after condensing.
- Continue this process till all the fat is extracted.
- Discontinue the process and take out the thimble.
- Again attach the extraction tube with flask containing solvent along with fat and

condenser unit.

- Again heat the flask to recover the solvent.
- Discontinue the process and clean the extraction tube and thimble.

### **PRECAUTIONS TO BE FOLLOWED**

- Always perform Soxhlet extractions in a fume hood (with sash down) to avoid inhalation of hazardous solvents and to contain potential fire
- Personal Protective Equipment – Eye protection: required – Hand protection: required
- Nitrile gloves are recommended
- Insulated gloves used for contact with hot glassware – Protective Clothing: lab coats, closed toe shoes and long pants should be worn

### **RECORD TO BE MAINTAINED**

- Laboratory Manual containing the experiments that can be performed with the equipment
- Maintenance Record

*A.H.*

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**481171L1-ADVANCED BIOCHEMISTRY LAB**

**STANDARD OPERATING PROCEDURE**

Name of the Lab./facility	<b>BIOCHEMISTRY AND BIO INSTRUMENTATION LAB</b>
Purpose	To describe the procedure for the operation and maintenance of the UV – Visible Spectrophotometer
Scope	This instrument is used to measure how much light of a given wavelength is absorbed by a liquid sample.
Responsibility	<p>It is the responsibility of Lab – In-charge of the lab to train Lab Assistant and students on this procedure and to ensure adherence to this procedure.</p> <p>It is the responsibility of the students/technicians to follow the SOP as described and to inform the Lab In-Charge about any deviations or problems that may occur while performing the procedure.</p>
<p align="center"><b>STANDARD OPERATING PROCEDURE FOR UV – VISIBLE SPECTROPHOTOMETER</b></p>	
<ul style="list-style-type: none"> <li>• Open panel door and make sure cuvette holders are empty, and then close the panel door.</li> <li>• Flip open the display unit and Turn spectrophotometer “ON” by flipping the yellow switch on the side of the machine. The machine will automatically initialize.</li> <li>• Hold the cuvette from the top to prevent tampering with the measurements, and wipe the sides with a lab tissue.</li> <li>• Open panel door and place the cuvettes with blank solution in the cuvette holders.</li> </ul>	

When the Auto Zero is complete, open the panel door and remove the front cuvette.

- Do not replace cuvette in rear holder.
- Using the same cuvette style, fill an empty cuvette with about 2-ml 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.
- Select spectrum from the drop down menu
- Press save as and give name and save.
- 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.
- Press RETURN to bring you back to step Note: This will erase your old data.
- Remove cuvettes remaining in holders.
- Then click on the Disconnect button near the bottom of the screen.
- Exit from the UV-Probe software.
- Push the mode button on the UV-VIS instrument keypad.
- Flip power switch located on the side, to turn off the machine.

**PRECAUTIONS TO BE FOLLOWED**

N/A

**RECORD TO BE MAINTAINED**

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**STANDARD OPERATING PROCEDURE**

Name of the Lab./facility	<b>BIOCHEMISTRY AND BIO INSTRUMENTATION LAB</b>
Purpose	To describe the procedure for the operation and maintenance of the Centrifuge
Scope	This Standard Operating Procedure (SOP) applies to the staff and students using the Centrifuge in the Biochemistry and Bioinstrumentation Lab, Vinayaka missions' Research foundation
Responsibility	<p>It is the responsibility of Lab – In-charge of the lab to train Lab Assistant and students on this procedure and to ensure adherence to this procedure.</p> <p>It is the responsibility of the students/technicians to follow the SOP as described and to inform the Lab In-Charge about any deviations or problems that may occur while performing the procedure.</p>
<b>STANDARD OPERATING PROCEDURE FOR CENTRIFUGE</b>	
<ul style="list-style-type: none"> <li>• Press the start/stop button and slowly increase the rpm to the desired speed using the dial</li> <li>• Once a run is complete, make sure the rotor has <b>COMPLETELY STOPPED</b> before opening the centrifuge lid by depressing the red stop/start button.</li> <li>• Remove sample vials.</li> <li>• Remember to return the rpm dial back to zero after finishing.</li> </ul>	

### **PRECAUTIONS TO BE FOLLOWED**

- Proper handling of the instrument
- Ensure level and stability
- Balance centrifuge tubes equally
- Ensure use of rubber cushion for glass tubes
- Bring speed Knob to off and increase the speed gradually.
- Do not open the lid in between the centrifugation cycle

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**STANDARD OPERATING PROCEDURE**

Name of the Lab./facility	<b>BIOCHEMISTRY AND BIO INSTRUMENTATION LAB</b>
Purpose	To describe the procedure for the operation and maintenance of the Conductivity Meter
Scope	This standard operating procedure provides the laboratory person with guidance on the procedure for determining conductivity in Liquid and water samples.
Responsibility	<p>It is the responsibility of Lab – In-charge of the lab to train Lab Assistant and students on this procedure and to ensure adherence to this procedure.</p> <p>It is the responsibility of the students/technicians to follow the SOP as described and to inform the Lab In-Charge about any deviations or problems that may occur while performing the procedure.</p>

**STANDARD OPERATING PROCEDURE FOR CONDUCTIVITY METER**

- Turn on the meter.
- Rinse the probe with DI water.
- Dip probe into sample making sure there are no air bubbles trapped on the slot of the probe.
- Stir the probe gently in the sample to create a homogenous sample, allowing a few seconds for the temperature reading to approach the solution temperature
- Take reading (when the reading is stable 'READY' will display on the left-middle

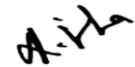
of the LCD.

### **PRECAUTIONS TO BE FOLLOWED**

Do not use or install the this equipment in any manner other than which is specified

### **RECORD TO BE MAINTAINED**

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A handwritten signature in black ink, appearing to be 'A. H.' or similar, written in a cursive style.

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### **STANDARD OPERATING PROCEDURE**

Name of the Lab./facility	<b>BIOCHEMISTRY AND BIO INSTRUMENTATION LAB</b>
Purpose	To describe the procedure for the operation and maintenance of the Magnetic Stirrer
Scope	This Standard Operating Procedure (SOP) applies to the staff and students using the Magnetic Stirrer in the Biochemistry and Bioinstrumentation laboratory, Vinayaka missions' Research foundation
Responsibility	<p>It is the responsibility of Lab – In-charge of the lab to train Lab Assistant and students on this procedure and to ensure adherence to this procedure.</p> <p>It is the responsibility of the students/technicians to follow the SOP as described and to inform the Lab In-Charge about any deviations or problems that may occur while performing the procedure.</p>
<b>STANDARD OPERATING PROCEDURE FOR MAGNETIC STIRRER</b>	
<ul style="list-style-type: none"> <li>• Place the magnetic stirrer on a stable well-levelled surface.</li> <li>• Place the stir bar at the bottom of a glass container.</li> <li>• Fill the glass container with the liquid to be stirred.</li> <li>• Plug the mains cable into a suitably earthed socket.</li> <li>• Check that the speed control knob is completely turned anticlockwise.</li> </ul>	

- Place the glass container on the centre of the magnetic stirrer.
- Press the On/Off switch to turn the magnetic stirrer On. The switch will light green.
- Adjust the speed control knob to a low stirring rate.
- Continue to adjust the speed control knob until the desired stirring speed is achieved.
- Wait until the liquid is properly mixed.
- Completely turn the speed control knob anticlockwise.
- Press the On/Off switch to turn the magnetic stirrer Off
- Manipulate another stir bar from the outside of the glass container to remove the immersed stir bar

#### **PRECAUTIONS TO BE FOLLOWED**

- Thoroughly wash the stir bar with distilled water after each application.
- Store stir bars in pairs to maintain their magnetic strength and increase their life span.

#### **RECORD TO BE MAINTAINED**

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### **STANDARD OPERATING PROCEDURE**

Name of the Lab./facility	<b>BIOCHEMISTRY AND BIO INSTRUMENTATION LAB</b>
Purpose	To describe the procedure for the operation and maintenance of the pH meter
Scope	This SOP describes how to use and maintain a pH meter. The pH meter is intended for accurate measurement of the pH of buffer solution used in the Biochemistry and Bioinstrumentation laboratory.
Responsibility	<p>It is the responsibility of Lab – In-charge of the lab to train Lab Assistant and students on this procedure and to ensure adherence to this procedure.</p> <p>It is the responsibility of the students/technicians to follow the SOP as described and to inform the Lab In-Charge about any deviations or problems that may occur while performing the procedure.</p>

### **STANDARD OPERATING PROCEDURE FOR pH METER**

- Before use, rinse the electrode with deionized water and blot dry with a soft, clean paper towel.
- Transfer the electrode to the test solution.
- Compensate for the temperature if necessary.
- Record the pH when the reading is stable (5–20 seconds after insertion of the electrode).

electrode into the solution)

- Rinse the electrode with deionized water and store according to the manufacturer instructions.

#### *Cleaning and maintenance*

- Clean the pH meter with a soft, clean, damp paper towel after use. No solvents should be used.
- Replace the electrode filling solution on a regular basis, according to the manufacturer's instructions. Record in the logbook (Adjust the speed control knob to a low stirring rate.

#### **PRECAUTIONS TO BE FOLLOWED**

- Thoroughly wash the stir bar with distilled water after each application.
- Store stir bars in pairs to maintain their magnetic strength and increase their life span.

#### **RECORD TO BE MAINTAINED**

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**STANDARD OPERATING PROCEDURE**

Name of the Lab./facility	<b>BIOCHEMISTRY AND BIO INSTRUMENTATION LAB</b>
Purpose	To describe the procedure for the operation and maintenance of the Colorimeter
Scope	This SOP describes how to use and maintain a Colorimeter. The Colorimeter is intended for accurate measurement of the concentration of the given sample used in the Biochemistry and Bioinstrumentation laboratory.
Responsibility	<p>It is the responsibility of Lab – In-charge of the lab to train Lab Assistant and students on this procedure and to ensure adherence to this procedure.</p> <p>It is the responsibility of the students/technicians to follow the SOP as described and to inform the Lab In-Charge about any deviations or problems that may occur while performing the procedure.</p>

**STANDARD OPERATING PROCEDURE FOR COLORIMETER**

- Switch on the instrument at least 5 minutes before use to allow it to stabilize.
- Select the most appropriate filter for the analysis and insert it in the light path or dial it in with the selector .
- Place the reagent blank solution (or water) in the cuvette and zero the instrument .

Make sure the clear faces of the cuvette are in the light path

- Place the sample in the colorimeter and read the absorbance of the solution. If the absorbance is "over range" (usually  $> 2.0$ ) then the sample must be diluted to yield a value within the limits of the instrument.
- At intervals, recheck the reagent blank to ensure that there is no drift in the zero value.

### **PRECAUTIONS TO BE FOLLOWED**

- The machine must be re-zeroed, if a new filter is chosen.
- The sample cuvettes must be at least two-thirds full.
- The outside of the cuvette must be dry and clean.

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**481171L1-ADVANCED BIOCHEMISTRY LAB**

**STANDARD OPERATING PROCEDURE**

Name of the Lab./facility	<b>BIOCHEMISTRY AND BIO INSTRUMENTATION LAB</b>
Purpose	To describe the procedure for the operation and maintenance of the Hematology Auto Analyzer
Scope	This SOP describes how to use and maintain a Hematology Auto Analyzer. This performs automated blood counts and requires no manual operations for aspirating blood, dilutions, measuring, calculations, print-outs and computer transfer used in the Biochemistry laboratory.
Responsibility	<p>It is the responsibility of Lab – In-charge of the lab to train Lab Assistant and students on this procedure and to ensure adherence to this procedure.</p> <p>It is the responsibility of the students/technicians to follow the SOP as described and to inform the Lab In-Charge about any deviations or problems that may occur while performing the procedure.</p>
<b>STANDARD OPERATING PROCEDURE FOR HEMATOLOGY AUTO ANALYZER</b>	
Specimen requirements:	
➤ About 2-3 ml of venous blood collected into EDTA tubes. Specimens should be	

transported at room temperature 18 - 26°C and can be store in the refrigerator of 2 - 8°C up to 6 hours. If stored in a refrigerator, samples should be returned to room temperature, for approximately 30 minutes, before analysis.

## **PROCEDURE:**

1. Check operation of the machine, ensuring it is clean and that all required supplies are present in sufficient quantities.
2. Switch the instrument on by pressing the ON/OFF switch, located on the back of the instrument.
3. The instrument performs an initialization phase for the internal electronics. Please wait.
4. Once the initialization phase is complete, the analyzer will automatically run a startup cycle.
5. If it does not automatically run a startup cycle after the initialization phase is completed, press "Startup" button in the "Status" area to initiate a startup cycle.
6. Then, the instrument will perform a blank cycle for a reference blank count (an analysis cycle based on reagents without any blood sample).
7. Check and verify that the reference blank counts do not exceed the following parameter limits: WBC < 0.3, RBC < 0.02, HGB < 0.3, PLT < 10 then: Press "OK" button to validate blank results.
8. Perform quality control analysis on 3 levels of control blood material (low, normal and high) to verify that the instrument is performing within the specified ranges of the quality control material.
9. Entering patient ID, sample ID, Patient name, etc
10. Follow the indications displayed in the "Sample analysis" dialog box to run the analysis.
  - a. Mix the sample gently and thoroughly.
  - b. Remove the cap from the sample tube.
  - c. Place the sample beneath the sampling needle.
  - d. Raise up the tube so that the sampling needle lowers into the blood and press the manual sample bar.
  - e. The analysis cycle will begin.

11. When the analysis is completed, the "Sample analysis" dialog box is closed and results are displayed in the "Result display" menu for print out.

### **PRECAUTIONS TO BE FOLLOWED**

Criteria for rejection haematology specimens 1. When the identification is missing /inadequate. 2. Insufficient quantity. 3. Inappropriate container 4. Inappropriate transport/storage. 5. Unknown duration of delay. 6. Clotted sample.

Equipment

### **RECORD TO BE MAINTAINED**

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### **STANDARD OPERATING PROCEDURE**

Name of the Lab./facility	<b>BIOCHEMISTRY AND BIO INSTRUMENTATION LAB</b>
Purpose	To describe the procedure for the operation and maintenance of the Biochemistry Analyzer
Scope	This SOP describes how to use and maintain a Biochemistry Analyzer. This performs automated blood counts and requires no manual operations for aspirating blood, dilutions, measuring, calculations, print-outs and computer transfer used in the Biochemistry laboratory.
Responsibility	<p>It is the responsibility of Lab – In-charge of the lab to train Lab Assistant and students on this procedure and to ensure adherence to this procedure.</p> <p>It is the responsibility of the students/technicians to follow the SOP as described and to inform the Lab In-Charge about any deviations or problems that may occur while performing the procedure.</p>

### **STANDARD OPERATING PROCEDURE FOR BIOCHEMISTRY ANALYZER**

#### **PROCEDURE:**

1. Check the Analyzer is attached with Electric supply.
2. Switch on the Analyzer

3. Press Aspiration key to take water, wash it thrice.
4. Keep the blank and press the aspiration button
5. Keep the standard solution and press the aspiration bottom.
6. Keep the test sample and press the aspiration button and record the reading
7. wash using water
8. Switch the Analyzer

#### **PRECAUTIONS TO BE FOLLOWED**

- **Keep away from direct Sunlight**
- **Keep in dry and cool space**

#### **RECORD TO BE MAINTAINED**

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**481171L1- ADVANCED BIOCHEMISTRY LAB**

**STANDARD OPERATING PROCEDURE**

Name of the Lab./facility	<b>BIOCHEMISTRY AND BIO INSTRUMENTATION LAB</b>
Purpose	To describe the procedure for the operation and maintenance of the Turbidity Meter.
Scope	<p>Clarity of water is extremely important in products destined for human consumption. Suspension free water is also considered necessary for industries manufacturing beverage, food products etc. Similarly, minimum limits of turbidity are required to be observed in drinking water &amp; sewage disposals.</p> <p>Digital Turbidity Meter and Nephelometer are ideal instruments for measurement of suspensions. They consist of a light source focused on a test tube containing the sample solution under test. The light, reflected at right angle to the focused light by the suspension in the solution, is detected by the photo electric detector, amplified and displayed on a 3½ digit LED display.</p>
Responsibility	<p>It is the responsibility of Lab – In-charge of the lab to train Lab Assistant and students on this procedure and to ensure adherence to this procedure.</p> <p>It is the responsibility of the students/technicians to follow the SOP as described and to inform the Lab In-Charge about any deviations or problems that may occur while performing the procedure.</p>
<b>STANDARD OPERATING PROCEDURE FOR TURBIDITY METER</b>	

## **PROCEDURE:**

1. Fill a clean container with at least 50 ml. of sample water and cover. Set sample aside to allow sample to equilibrate to air temperature and let gas escape. Avoid contaminants and analyze as soon as possible.
- 2) Rinse 2 empty turbidity tubes and caps with a portion of sample. Shake out excess water.
- 3) Fill both turbidity tubes to the neck by carefully pouring the sample down the side of the tube. This will prevent air bubbles from forming. (Similar to pouring soda or beer into a glass while trying to avoid foam on the top).
- 4) Cap the tubes and wipe dry with a clean lint free tissue.
- 5) Carefully invert the turbidity tube twice (do not create air bubbles) just before inserting the tube into the meter,
- 6) Open the meter lid. Align the indexing arrow on the tube with the indexing arrow on the meter. Insert the turbidity tube into the chamber.
- 7) Close the lid. Push the READ button. The turbidimeter in NTU units will be displayed within 5 seconds.
- 8) Record the results from each tube. Take a third reading from a new sample if the first two readings are significantly different.
- 9) The meter will turn off automatically one minute after the last button push. To turn the meter OFF manually, hold the READ button down for at least 2 seconds. Release the button when OFF is displayed.

## **PRECAUTIONS TO BE FOLLOWED**

- ER1 (ERROR 1) will be displayed if the battery is low.
- ER2 will be displayed when the measured turbidity is very low.
- ER3 will be displayed when the light source (bulb) has burned out or if the turbidity tube is misaligned.
- BAT will be displayed when the battery is getting low. The readings are reliable but the battery needs to be replaced as soon as possible. Carry extra batteries with the field kit.

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