Estimation of Serum Cholesterol

Experiment 21 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To estimate total cholesterol in a serum teaching specimen using the assigned validated method.

Principle

A cholesterol-esterase/oxidase/peroxidase method uses enzymatic reactions to generate a colour related to total cholesterol. The blank-corrected test response is compared with calibration under the method's linearity and quality-control requirements.

Method and Interpretation

Total cholesterol is one lipid measurement; broader cardiovascular-risk assessment also needs other clinical information.

Requirements

- Simulated serum, calibrator and low/high quality controls
- Assigned validated reagent kit, pipettes and photometer or analyser
- Current product method sheet

Procedure

1. Read method

Record specimen suitability, reagent volumes, wavelength, incubation/read times, calibration, linearity and interference information.

2. Prepare

Label blank, calibrator, controls and test; dispense the kit-specified quantities.

3. Develop and read

Perform the kit-defined enzymatic reaction and endpoint reading.

4. Check validity

Confirm controls meet their assigned limits and the response is within the method range.

5. Calculate

Use the validated calibration procedure and any documented specimen dilution.

6. Report

State result, units, method, dilution and whether the controls support a valid measurement.

Observations and Record

Field	Student record
Method / kit / wavelength	_____
Blank / standard / test response	_____
Control results / limits	_____
Standard concentration / units	_____
Dilution factor	_____
Calculated concentration	_____

Worked Example

The numbers below are illustrative; enter your own observations in the record.

- One-point teaching calculation, only when permitted by the method: concentration = corrected test response / corrected standard response × standard concentration × dilution factor.
- Illustrative responses: test 0.450, standard 0.500; standard 200 mg/dL; dilution factor 1.
- Calculated teaching result = 180 mg/dL.

Result

Concentration of total cholesterol: _____ mg/dL. Controls: _____. Dilution and limitations: _____.

Precautions

- A multi-point calibrated or kinetic method may require its own curve/equation rather than the one-point illustration.
- Do not treat a failed control or out-of-range response as reportable.
- Biological specimens require authorised collection and handling; the classroom exercise uses simulated material.

Viva Questions and Answers

1. Why subtract the blank?

It removes the assigned reagent/background contribution.

2. Why include dilution?

The tested solution may be less concentrated than the original material.

3. Can a concentration alone diagnose disease?

Total cholesterol is one lipid measurement; broader cardiovascular-risk assessment also needs other clinical information.

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