Year 2 - Practical Notes

Biochemistry and Clinical Pathology

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Molisch Test for Carbohydrates

Experiment 1 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To perform or observe the assigned qualitative test and interpret its controls.

Principle

A general carbohydrate reaction forms furfural-type products that react with the Molisch reagent at an acid interface.

Expected Teaching Response

Violet or purple interface ring supports a carbohydrate response.

Requirements

- Instructor-prepared carbohydrate solutions, Molisch reagent and assigned acid system
- Clean labelled tubes or product-specific test equipment; PPE

Procedure

1. Label

Prepare separate test, positive control and reagent blank/negative control entries.

2. Perform

Use the assigned small-scale layering demonstration under supervision; observe the interface without vigorous mixing.

3. Read

Observe at the method-defined time and compare with controls.

4. Record

Write the actual colour, precipitate or category, then the supported inference.

5. Interpret

This is a general class test, not glucose-specific.

Observations and Record

Material	Observed response	Inference
Test material	_____	_____
Positive control	_____	_____
Blank / negative control	_____	_____

Result

The observed response was _____. Controls were _____. The result _____ supports _____ under the stated method.

Precautions

- Use simulated biological material or authorised supervised demonstrations.
- Follow reagent, solvent, heating and waste controls; corrosive or metal reagents are instructor-managed.
- Do not copy an expected response into the actual observation field.

Viva Questions and Answers

1. What response is expected in the teaching positive?

Violet or purple interface ring supports a carbohydrate response.

2. Why use controls?

They show whether the reagents and conditions produced a valid comparison.

3. What limits the conclusion?

This is a general class test, not glucose-specific.

Benedict Test for Reducing Sugars

Experiment 2 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To perform or observe the assigned qualitative test and interpret its controls.

Principle

Reducing sugars reduce alkaline copper(II) to copper(I) oxide under the test conditions.

Expected Teaching Response

Green-to-yellow/orange/red precipitate may appear depending on response and concentration.

Requirements

- Assigned sugar solutions, Benedict reagent and controlled heating bath
- Clean labelled tubes or product-specific test equipment; PPE

Procedure

1. Label

Prepare separate test, positive control and reagent blank/negative control entries.

2. Perform

Combine the prescribed portions and heat using the assigned time and conditions; compare the cooled response with controls.

3. Read

Observe at the method-defined time and compare with controls.

4. Record

Write the actual colour, precipitate or category, then the supported inference.

5. Interpret

Sucrose is nonreducing before hydrolysis; other reducing substances can respond.

Observations and Record

Material	Observed response	Inference
Test material	_____	_____
Positive control	_____	_____
Blank / negative control	_____	_____

Result

The observed response was _____. Controls were _____. The result _____ supports _____ under the stated method.

Precautions

- Use simulated biological material or authorised supervised demonstrations.
- Follow reagent, solvent, heating and waste controls; corrosive or metal reagents are instructor-managed.
- Do not copy an expected response into the actual observation field.

Viva Questions and Answers

1. What response is expected in the teaching positive?

Green-to-yellow/orange/red precipitate may appear depending on response and concentration.

2. Why use controls?

They show whether the reagents and conditions produced a valid comparison.

3. What limits the conclusion?

Sucrose is nonreducing before hydrolysis; other reducing substances can respond.

Barfoed Test for Monosaccharides

Experiment 3 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To perform or observe the assigned qualitative test and interpret its controls.

Principle

In the prescribed acidic copper-reagent test, monosaccharides commonly reduce the reagent faster than many disaccharides.

Expected Teaching Response

A red copper(I) oxide response within the defined time supports the assigned monosaccharide comparison.

Requirements

- Assigned mono/disaccharide solutions, Barfoed reagent and timed water bath
- Clean labelled tubes or product-specific test equipment; PPE

Procedure

1. Label

Prepare separate test, positive control and reagent blank/negative control entries.

2. Perform

Use matched portions, heat for the prescribed brief interval and observe promptly; record the elapsed time.

3. Read

Observe at the method-defined time and compare with controls.

4. Record

Write the actual colour, precipitate or category, then the supported inference.

5. Interpret

Excess heating can cause misleading delayed disaccharide responses.

Observations and Record

Material	Observed response	Inference
Test material	_____	_____
Positive control	_____	_____
Blank / negative control	_____	_____

Result

The observed response was _____. Controls were _____. The result _____ supports _____ under the stated method.

Precautions

- Use simulated biological material or authorised supervised demonstrations.
- Follow reagent, solvent, heating and waste controls; corrosive or metal reagents are instructor-managed.
- Do not copy an expected response into the actual observation field.

Viva Questions and Answers

1. What response is expected in the teaching positive?

A red copper(I) oxide response within the defined time supports the assigned monosaccharide comparison.

2. Why use controls?

They show whether the reagents and conditions produced a valid comparison.

3. What limits the conclusion?

Excess heating can cause misleading delayed disaccharide responses.

Iodine Test for Starch

Experiment 4 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To perform or observe the assigned qualitative test and interpret its controls.

Principle

Iodine interacts with suitable polysaccharide structures, producing a characteristic starch colour.

Expected Teaching Response

Blue-black colour supports the starch response; different polysaccharides can give other colours.

Requirements

- Assigned starch and sugar solutions, iodine test reagent
- Clean labelled tubes or product-specific test equipment; PPE

Procedure

1. Label

Prepare separate test, positive control and reagent blank/negative control entries.

2. Perform

Add the prescribed iodine reagent to each labelled teaching solution and compare with blank and starch control.

3. Read

Observe at the method-defined time and compare with controls.

4. Record

Write the actual colour, precipitate or category, then the supported inference.

5. Interpret

Colour changes with temperature and starch structure; it is not a reducing-sugar test.

Observations and Record

Material	Observed response	Inference
Test material	_____	_____
Positive control	_____	_____
Blank / negative control	_____	_____

Result

The observed response was _____. Controls were _____. The result _____ supports _____ under the stated method.

Precautions

- Use simulated biological material or authorised supervised demonstrations.
- Follow reagent, solvent, heating and waste controls; corrosive or metal reagents are instructor-managed.
- Do not copy an expected response into the actual observation field.

Viva Questions and Answers

1. What response is expected in the teaching positive?

Blue-black colour supports the starch response; different polysaccharides can give other colours.

2. Why use controls?

They show whether the reagents and conditions produced a valid comparison.

3. What limits the conclusion?

Colour changes with temperature and starch structure; it is not a reducing-sugar test.

Biuret Test for Proteins

Experiment 5 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To perform or observe the assigned qualitative test and interpret its controls.

Principle

Copper(II) in an alkaline medium forms a coloured complex with suitable peptide-bond arrangements.

Expected Teaching Response

Violet colour supports peptide-containing material under the method conditions.

Requirements

- Assigned protein, amino-acid and blank solutions; Biuret reagent
- Clean labelled tubes or product-specific test equipment; PPE

Procedure

1. Label

Prepare separate test, positive control and reagent blank/negative control entries.

2. Perform

Add the prescribed reagent to matched portions and read after the assigned time.

3. Read

Observe at the method-defined time and compare with controls.

4. Record

Write the actual colour, precipitate or category, then the supported inference.

5. Interpret

A free amino acid is not equivalent to a protein in this test.

Observations and Record

Material	Observed response	Inference
Test material	_____	_____
Positive control	_____	_____
Blank / negative control	_____	_____

Result

The observed response was _____. Controls were _____. The result _____ supports _____ under the stated method.

Precautions

- Use simulated biological material or authorised supervised demonstrations.
- Follow reagent, solvent, heating and waste controls; corrosive or metal reagents are instructor-managed.
- Do not copy an expected response into the actual observation field.

Viva Questions and Answers

1. What response is expected in the teaching positive?

Violet colour supports peptide-containing material under the method conditions.

2. Why use controls?

They show whether the reagents and conditions produced a valid comparison.

3. What limits the conclusion?

A free amino acid is not equivalent to a protein in this test.

Xanthoproteic Test for Aromatic Amino Acids

Experiment 6 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To perform or observe the assigned qualitative test and interpret its controls.

Principle

The assigned reaction produces yellow nitro derivatives from suitable aromatic residues; the controlled alkaline stage can deepen the colour.

Expected Teaching Response

Yellow, with orange after the assigned alkaline stage, supports a responsive aromatic group.

Requirements

- Instructor-prepared protein/amino-acid solutions and approved reagent system
- Clean labelled tubes or product-specific test equipment; PPE

Procedure

1. Label

Prepare separate test, positive control and reagent blank/negative control entries.

2. Perform

Observe the instructor-controlled reaction and subsequent prescribed colour-development stage; record the response without preparing reagents.

3. Read

Observe at the method-defined time and compare with controls.

4. Record

Write the actual colour, precipitate or category, then the supported inference.

5. Interpret

Not every amino acid gives the same reaction; the test is not a complete protein assay.

Observations and Record

Material	Observed response	Inference
Test material	_____	_____
Positive control	_____	_____
Blank / negative control	_____	_____

Result

The observed response was _____. Controls were _____. The result _____ supports _____ under the stated method.

Precautions

- Use simulated biological material or authorised supervised demonstrations.
- Follow reagent, solvent, heating and waste controls; corrosive or metal reagents are instructor-managed.
- Do not copy an expected response into the actual observation field.

Viva Questions and Answers

1. What response is expected in the teaching positive?

Yellow, with orange after the assigned alkaline stage, supports a responsive aromatic group.

2. Why use controls?

They show whether the reagents and conditions produced a valid comparison.

3. What limits the conclusion?

Not every amino acid gives the same reaction; the test is not a complete protein assay.

Ninhydrin Test for Amino Acids

Experiment 7 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To perform or observe the assigned qualitative test and interpret its controls.

Principle

Ninhydrin reacts with many free amino groups to form a characteristic purple product.

Expected Teaching Response

Many amino acids give blue-purple colour; proline-type responses may be yellow.

Requirements

- Assigned amino-acid solutions, ninhydrin reagent and controlled heating equipment
- Clean labelled tubes or product-specific test equipment; PPE

Procedure

1. Label

Prepare separate test, positive control and reagent blank/negative control entries.

2. Perform

Use matched test and control portions and the prescribed reaction conditions; read the developed colour.

3. Read

Observe at the method-defined time and compare with controls.

4. Record

Write the actual colour, precipitate or category, then the supported inference.

5. Interpret

The response depends on amino-group chemistry and is not identical for all amino acids.

Observations and Record

Material	Observed response	Inference
Test material	_____	_____
Positive control	_____	_____
Blank / negative control	_____	_____

Result

The observed response was _____. Controls were _____. The result _____ supports _____ under the stated method.

Precautions

- Use simulated biological material or authorised supervised demonstrations.
- Follow reagent, solvent, heating and waste controls; corrosive or metal reagents are instructor-managed.
- Do not copy an expected response into the actual observation field.

Viva Questions and Answers

1. What response is expected in the teaching positive?

Many amino acids give blue-purple colour; proline-type responses may be yellow.

2. Why use controls?

They show whether the reagents and conditions produced a valid comparison.

3. What limits the conclusion?

The response depends on amino-group chemistry and is not identical for all amino acids.

Sulphur Test for Selected Amino Acids

Experiment 8 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To perform or observe the assigned qualitative test and interpret its controls.

Principle

The prescribed alkaline test can reveal sulphur released from suitable sulphur-containing amino acids and produce a dark metal-sulphide response.

Expected Teaching Response

A dark sulphide response supports the assigned cysteine/cystine test.

Requirements

- Instructor-prepared cysteine/cystine comparison, negative control and approved reagents
- Clean labelled tubes or product-specific test equipment; PPE

Procedure

1. Label

Prepare separate test, positive control and reagent blank/negative control entries.

2. Perform

Observe the supervised prescribed test with designated waste collection; compare positive and negative controls.

3. Read

Observe at the method-defined time and compare with controls.

4. Record

Write the actual colour, precipitate or category, then the supported inference.

5. Interpret

Methionine does not necessarily give the same positive response in this classical test.

Observations and Record

Material	Observed response	Inference
Test material	_____	_____
Positive control	_____	_____
Blank / negative control	_____	_____

Result

The observed response was _____. Controls were _____. The result _____ supports _____ under the stated method.

Precautions

- Use simulated biological material or authorised supervised demonstrations.
- Follow reagent, solvent, heating and waste controls; corrosive or metal reagents are instructor-managed.
- Do not copy an expected response into the actual observation field.

Viva Questions and Answers

1. What response is expected in the teaching positive?

A dark sulphide response supports the assigned cysteine/cystine test.

2. Why use controls?

They show whether the reagents and conditions produced a valid comparison.

3. What limits the conclusion?

Methionine does not necessarily give the same positive response in this classical test.

Sudan Test for Lipids

Experiment 9 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To perform or observe the assigned qualitative test and interpret its controls.

Principle

A suitable Sudan dye preferentially colours a lipid phase, supporting recognition of lipid material.

Expected Teaching Response

An appropriately coloured lipid phase supports the test.

Requirements

- Teaching oil, aqueous blank and the assigned Sudan reagent
- Clean labelled tubes or product-specific test equipment; PPE

Procedure

1. Label

Prepare separate test, positive control and reagent blank/negative control entries.

2. Perform

Mix the prescribed portions gently and observe where the dye partitions; compare with controls.

3. Read

Observe at the method-defined time and compare with controls.

4. Record

Write the actual colour, precipitate or category, then the supported inference.

5. Interpret

Dye partition is a class observation, not an identification of a particular fatty acid.

Observations and Record

Material	Observed response	Inference
Test material	_____	_____
Positive control	_____	_____
Blank / negative control	_____	_____

Result

The observed response was _____. Controls were _____. The result _____ supports _____ under the stated method.

Precautions

- Use simulated biological material or authorised supervised demonstrations.
- Follow reagent, solvent, heating and waste controls; corrosive or metal reagents are instructor-managed.
- Do not copy an expected response into the actual observation field.

Viva Questions and Answers

1. What response is expected in the teaching positive?

An appropriately coloured lipid phase supports the test.

2. Why use controls?

They show whether the reagents and conditions produced a valid comparison.

3. What limits the conclusion?

Dye partition is a class observation, not an identification of a particular fatty acid.

Emulsion Test for Lipids

Experiment 10 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To perform or observe the assigned qualitative test and interpret its controls.

Principle

Lipids dissolved in the assigned alcohol can form a dispersed milky phase on dilution into water.

Expected Teaching Response

A milky dispersion supports the presence of suitable lipid material.

Requirements

- Teaching oil, assigned alcohol, water and blank
- Clean labelled tubes or product-specific test equipment; PPE

Procedure

1. Label

Prepare separate test, positive control and reagent blank/negative control entries.

2. Perform

Use the prescribed small-scale alcohol extraction and water-dilution test; compare the resulting appearance.

3. Read

Observe at the method-defined time and compare with controls.

4. Record

Write the actual colour, precipitate or category, then the supported inference.

5. Interpret

A cloudy unrelated specimen or insoluble solid can interfere.

Observations and Record

Material	Observed response	Inference
Test material	_____	_____
Positive control	_____	_____
Blank / negative control	_____	_____

Result

The observed response was _____. Controls were _____. The result _____ supports _____ under the stated method.

Precautions

- Use simulated biological material or authorised supervised demonstrations.
- Follow reagent, solvent, heating and waste controls; corrosive or metal reagents are instructor-managed.
- Do not copy an expected response into the actual observation field.

Viva Questions and Answers

1. What response is expected in the teaching positive?

A milky dispersion supports the presence of suitable lipid material.

2. Why use controls?

They show whether the reagents and conditions produced a valid comparison.

3. What limits the conclusion?

A cloudy unrelated specimen or insoluble solid can interfere.

Qualitative Detection of Urinary Glucose

Experiment 11 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To perform or observe the assigned qualitative test and interpret its controls.

Principle

A validated glucose-oxidase strip or enzymatic method detects glucose under product-specific conditions.

Expected Teaching Response

Read the glucose pad against its supplied chart; enter the actual category and units if provided.

Requirements

- Simulated urine, positive/negative controls and approved glucose strips
- Clean labelled tubes or product-specific test equipment; PPE

Procedure

1. Label

Prepare separate test, positive control and reagent blank/negative control entries.

2. Perform

Use the correct specimen contact and timed colour reading specified by the product.

3. Read

Observe at the method-defined time and compare with controls.

4. Record

Write the actual colour, precipitate or category, then the supported inference.

5. Interpret

A general reducing-sugar test is less specific; a urine result alone does not diagnose diabetes.

Observations and Record

Material	Observed response	Inference
Test material	_____	_____
Positive control	_____	_____
Blank / negative control	_____	_____

Result

The observed response was _____. Controls were _____. The result _____ supports _____ under the stated method.

Precautions

- Use simulated biological material or authorised supervised demonstrations.
- Follow reagent, solvent, heating and waste controls; corrosive or metal reagents are instructor-managed.
- Do not copy an expected response into the actual observation field.

Viva Questions and Answers

1. What response is expected in the teaching positive?

Read the glucose pad against its supplied chart; enter the actual category and units if provided.

2. Why use controls?

They show whether the reagents and conditions produced a valid comparison.

3. What limits the conclusion?

A general reducing-sugar test is less specific; a urine result alone does not diagnose diabetes.

Qualitative Detection of Urinary Protein

Experiment 12 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To perform or observe the assigned qualitative test and interpret its controls.

Principle

A validated reagent-strip protein response is commonly most sensitive to albumin; other assigned methods detect proteins differently.

Expected Teaching Response

A positive response supports protein detectable by that method.

Requirements

- Simulated urine, controls and approved protein-testing system
- Clean labelled tubes or product-specific test equipment; PPE

Procedure

1. Label

Prepare separate test, positive control and reagent blank/negative control entries.

2. Perform

Use the assigned strip or instructor-demonstrated method and interpret its method-specific response.

3. Read

Observe at the method-defined time and compare with controls.

4. Record

Write the actual colour, precipitate or category, then the supported inference.

5. Interpret

pH, contamination and protein type can affect results; a negative strip does not exclude every protein.

Observations and Record

Material	Observed response	Inference
Test material	_____	_____
Positive control	_____	_____
Blank / negative control	_____	_____

Result

The observed response was _____. Controls were _____. The result _____ supports _____ under the stated method.

Precautions

- Use simulated biological material or authorised supervised demonstrations.
- Follow reagent, solvent, heating and waste controls; corrosive or metal reagents are instructor-managed.
- Do not copy an expected response into the actual observation field.

Viva Questions and Answers

1. What response is expected in the teaching positive?

A positive response supports protein detectable by that method.

2. Why use controls?

They show whether the reagents and conditions produced a valid comparison.

3. What limits the conclusion?

pH, contamination and protein type can affect results; a negative strip does not exclude every protein.

Qualitative Detection of Urinary Ketones

Experiment 13 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To perform or observe the assigned qualitative test and interpret its controls.

Principle

Common nitroprusside urine tests detect mainly acetoacetate; they do not directly quantify beta-hydroxybutyrate.

Expected Teaching Response

A purple-type positive response supports the measured ketone component.

Requirements

- Simulated urine, ketone controls and approved testing product
- Clean labelled tubes or product-specific test equipment; PPE

Procedure

1. Label

Prepare separate test, positive control and reagent blank/negative control entries.

2. Perform

Follow the product contact and timed-reading instructions and record the category.

3. Read

Observe at the method-defined time and compare with controls.

4. Record

Write the actual colour, precipitate or category, then the supported inference.

5. Interpret

Do not use a negative urine result to exclude an acutely unwell person's ketoacidosis.

Observations and Record

Material	Observed response	Inference
Test material	_____	_____
Positive control	_____	_____
Blank / negative control	_____	_____

Result

The observed response was _____. Controls were _____. The result _____ supports _____ under the stated method.

Precautions

- Use simulated biological material or authorised supervised demonstrations.
- Follow reagent, solvent, heating and waste controls; corrosive or metal reagents are instructor-managed.
- Do not copy an expected response into the actual observation field.

Viva Questions and Answers

1. What response is expected in the teaching positive?

A purple-type positive response supports the measured ketone component.

2. Why use controls?

They show whether the reagents and conditions produced a valid comparison.

3. What limits the conclusion?

Do not use a negative urine result to exclude an acutely unwell person's ketoacidosis.

Qualitative Detection of Urinary Bilirubin

Experiment 14 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To perform or observe the assigned qualitative test and interpret its controls.

Principle

Validated urinary bilirubin tests use a product-specific colour reaction with detectable conjugated bilirubin.

Expected Teaching Response

Read the supplied method colour chart and report positive/negative or the stated category.

Requirements

- Simulated urine, bilirubin controls and the prescribed kit
- Clean labelled tubes or product-specific test equipment; PPE

Procedure

1. Label

Prepare separate test, positive control and reagent blank/negative control entries.

2. Perform

Protect the material as instructed and perform a timed test with controls.

3. Read

Observe at the method-defined time and compare with controls.

4. Record

Write the actual colour, precipitate or category, then the supported inference.

5. Interpret

Light exposure and interfering substances may alter the response; interpret with the clinical/laboratory context.

Observations and Record

Material	Observed response	Inference
Test material	_____	_____
Positive control	_____	_____
Blank / negative control	_____	_____

Result

The observed response was _____. Controls were _____. The result _____ supports _____ under the stated method.

Precautions

- Use simulated biological material or authorised supervised demonstrations.
- Follow reagent, solvent, heating and waste controls; corrosive or metal reagents are instructor-managed.
- Do not copy an expected response into the actual observation field.

Viva Questions and Answers

1. What response is expected in the teaching positive?

Read the supplied method colour chart and report positive/negative or the stated category.

2. Why use controls?

They show whether the reagents and conditions produced a valid comparison.

3. What limits the conclusion?

Light exposure and interfering substances may alter the response; interpret with the clinical/laboratory context.

Quantitative Estimation of Urinary Glucose

Experiment 15 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To estimate glucose in a urine teaching specimen using the assigned validated method.

Principle

A glucose-oxidase/peroxidase method converts glucose-related reaction products into a measurable colour. The blank-corrected test response is compared with calibration under the method's linearity and quality-control requirements.

Method and Interpretation

Urinary glucose reflects renal handling as well as blood glucose and must be interpreted in context.

Requirements

- Simulated urine, 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

Carry out the kit-defined reaction and photometric reading with matched blank, standard and controls.

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 0.400, standard 0.500; standard 100 mg/dL; dilution factor 2.
- Calculated teaching result = 160 mg/dL.

Result

Concentration of glucose: _____ 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?

Urinary glucose reflects renal handling as well as blood glucose and must be interpreted in context.

Quantitative Estimation of Urinary Creatinine

Experiment 16 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To estimate creatinine in a urine teaching specimen using the assigned validated method.

Principle

An assigned kinetic Jaffe method measures the prescribed creatinine-related colour change with a prepared alkaline-picrate reagent; an enzymatic alternative uses its own reaction scheme. The blank-corrected test response is compared with calibration under the method's linearity and quality-control requirements.

Method and Interpretation

Creatinine supports assessment of urine concentration or timed excretion; nonspecific Jaffe interferences and collection quality matter.

Requirements

- Simulated urine, 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

Use the kit's timed response difference or enzymatic reading; match the calculation to the selected method.

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 0.300, standard 0.500; standard 100 mg/dL; dilution factor 2.
- Calculated teaching result = 120 mg/dL.

Result

Concentration of creatinine: _____ 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?

Creatinine supports assessment of urine concentration or timed excretion; nonspecific Jaffe interferences and collection quality matter.

Quantitative Estimation of Urinary Chlorides

Experiment 17 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To estimate chloride in a simulated urine specimen by the assigned validated method.

Principle

An instructor-assigned argentometric method measures chloride through silver-chloride precipitation. Specimen pretreatment, pH and interfering ions must follow the validated urine procedure; an approved ion-selective method is a different option.

Requirements

- Simulated urine, chloride standard and controls
- Standardised silver titrant and prescribed endpoint system or approved analyser
- Assigned urine-method sheet and designated silver waste

Procedure

1. Review

Record pretreatment, tested volume, endpoint system, blank and titrant strength.

2. Prepare

Use the method-approved specimen preparation and dilution.

3. Measure

Perform the supervised titration or approved analyser method and record the actual response.

4. Check

Assess blank, controls and repeatability.

5. Calculate

Use the method stoichiometry and dilution; report mmol/L or the specified units.

Observations and Record

Field	Record
Test volume / dilution	_____
Test / blank titre	_____
Titrant molarity	_____
Control validity	_____
Chloride concentration / units	_____

Worked Example

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

- For a valid 1:1 silver-to-chloride titration: chloride mmol/L = corrected titre (mL) × titrant M × 1,000 / aliquot (mL) × dilution factor.
- Illustrative values: 5.00 mL of 0.0100 M silver titrant for a 1.00 mL aliquot at dilution factor 1 gives 50.0 mmol/L.

Result

Record the observed outcome and calculated value, with units. State whether the stated laboratory specification was met; do not copy the worked example as your result.

Precautions

- Use the validated urine method; a water-only chloride test is not automatically suitable for urine.
- Handle silver reagents and waste as instructed.
- Do not infer daily excretion without a valid timed collection and total volume.

Viva Questions and Answers

1. Why is the silver-chloride ratio 1:1?

One silver ion reacts with one chloride ion.

2. Why does the matrix matter?

Other components can affect precipitation or the endpoint.

3. How is timed excretion calculated?

Concentration multiplied by correctly collected urine volume, with consistent units.

Estimation of Serum Creatine

Experiment 18 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To estimate creatine using an assigned enzymatic calibration exercise.

Principle

A validated creatine assay can produce a proportional colour through coupled enzyme reactions. Creatine is chemically distinct from creatinine, so the correct assay and analyte name are essential.

Requirements

- Simulated serum and creatine calibrators
- Assigned creatine assay kit and photometer; a selected product may use a 570 nm colour reading
- Product method sheet and controls

Procedure

1. Read method

Confirm the assay measures creatine, not creatinine; record specimen and interference requirements.

2. Calibrate

Prepare the product-specified blank and calibration series.

3. React

Use the prescribed kit volumes, timing and temperature.

4. Read

Measure the kit-defined response and verify control validity.

5. Calculate

Read concentration from the validated calibration curve and apply any dilution.

6. Report

State creatine units and keep the result separate from a creatinine result.

Observations and Record

Field	Actual record
Kit / analyte / wavelength	_____
Calibrator responses	_____
Test response / controls	_____
Curve-derived concentration	_____
Dilution / final units	_____

Worked Example

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

- Original linear teaching curve: 0 micromol/L gives response 0.000; 50 gives 0.250; 100 gives 0.500.
- A corrected response 0.300 corresponds to 60 micromol/L on this illustrative curve before any dilution factor.

Result

Record the observed outcome and calculated value, with units. State whether the stated laboratory specification was met; do not copy the worked example as your result.

Precautions

- Use the actual product calibration, which may not follow the illustrative line.
- Do not substitute a creatinine assay or its clinical interpretation.
- Use simulated biological material in the classroom.

Viva Questions and Answers

1. Are creatine and creatinine the same?

No; they are different analytes with different assay schemes.

2. Why use a calibration curve?

It links the measured response to known concentrations over the validated range.

3. Why verify the kit name?

Similar analyte names can lead to the wrong assay and interpretation.

Estimation of Serum Creatinine

Experiment 19 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

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

Principle

A validated enzymatic or kinetic Jaffe assay estimates creatinine; the selected method determines interference control and calibration. The blank-corrected test response is compared with calibration under the method's linearity and quality-control requirements.

Method and Interpretation

Renal interpretation uses clinical context and appropriate kidney-function assessment; creatinine concentration alone is not a complete diagnosis.

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

Use the kit-defined kinetic response difference or enzymatic endpoint and its corresponding calibration.

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 0.250, standard 0.500; standard 2 mg/dL; dilution factor 1.
- Calculated teaching result = 1 mg/dL.

Result

Concentration of creatinine: _____ 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?

Renal interpretation uses clinical context and appropriate kidney-function assessment; creatinine concentration alone is not a complete diagnosis.

Estimation of Serum Glucose

Experiment 20 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

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

Principle

A glucose-oxidase/peroxidase endpoint or validated hexokinase method links glucose concentration to a measurable reaction. The blank-corrected test response is compared with calibration under the method's linearity and quality-control requirements.

Method and Interpretation

Meal timing, collection/processing and clinical context are needed; diagnostic decisions use the required validated assessment.

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

Use the selected product's endpoint, wavelength and calibration with suitable controls.

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 0.600, standard 0.500; standard 100 mg/dL; dilution factor 1.
- Calculated teaching result = 120 mg/dL.

Result

Concentration of glucose: _____ 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?

Meal timing, collection/processing and clinical context are needed; diagnostic decisions use the required validated assessment.

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.

Estimation of Serum Calcium

Experiment 22 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

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

Principle

An assigned o-cresolphthalein-complexone method forms a calcium-related colour; alternative validated methods use their own chemistry. The blank-corrected test response is compared with calibration under the method's linearity and quality-control requirements.

Method and Interpretation

Total calcium is affected by protein binding and is different from ionised calcium; interpretation requires the appropriate context.

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

Use the prescribed reagent, blank, calibration and photometric conditions.

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 0.460, standard 0.500; standard 10 mg/dL; dilution factor 1.
- Calculated teaching result = 9.2 mg/dL.

Result

Concentration of total calcium: _____ 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 calcium is affected by protein binding and is different from ionised calcium; interpretation requires the appropriate context.

Estimation of Serum Urea

Experiment 23 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

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

Principle

A urease-based method converts urea to products measured by a coupled system; a GLDH/NADH method uses the specified absorbance change. The blank-corrected test response is compared with calibration under the method's linearity and quality-control requirements.

Method and Interpretation

Urea and blood urea nitrogen have different mass reporting conventions; hydration, protein metabolism and renal context affect interpretation.

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

Read the method-defined reaction difference or endpoint and calibrate accordingly.

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 0.350, standard 0.500; standard 50 mg/dL; dilution factor 1.
- Calculated teaching result = 35 mg/dL.

Result

Concentration of urea: _____ 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?

Urea and blood urea nitrogen have different mass reporting conventions; hydration, protein metabolism and renal context affect interpretation.

Estimation of SGOT and SGPT Enzyme Activity

Experiment 24 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To calculate AST and ALT activity from supplied kinetic teaching records.

Principle

SGOT is the older name for AST; SGPT is the older name for ALT. Coupled kinetic methods commonly follow NADH absorbance change at 340 nm. The actual method supplies the activity factor and conditions.

Reaction Overview

- AST transfers an amino group from aspartate to alpha-ketoglutarate, forming oxaloacetate and glutamate.
- ALT transfers an amino group from alanine to alpha-ketoglutarate, forming pyruvate and glutamate.
- The coupled detection system links these products to the measured kinetic signal.

Requirements

- Simulated serum or supplied kinetic records
- Assigned AST and ALT kits, controls and photometer
- Product-specified temperature, timing and factor

Procedure

1. Review

Record enzyme name, reaction conditions, interval and activity factor.

2. Run

Use the product's preparation and timed readings; assess controls.

3. Calculate slope

Determine the method-approved absorbance change per minute.

4. Calculate activity

Multiply by the kit's factor and any required dilution.

5. Interpret

Report U/L and distinguish enzyme activity from enzyme mass or a stand-alone diagnosis.

Observations and Record

Time / activity	AST	ALT
Initial absorbance	_____	_____
After 1 minute	_____	_____
After 2 minutes	_____	_____
Change per minute	_____	_____
Factor / activity, U/L	_____	_____

Worked Example

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

- Illustrative kinetic record: absorbance 0.800, 0.780 and 0.760 at 0, 1 and 2 minutes gives a decrease of 0.020/min.
- With an instructor-supplied factor 1,746, activity = 0.020 × 1,746 = 34.92 U/L. This factor is not universal.

Result

Record the observed outcome and calculated value, with units. State whether the stated laboratory specification was met; do not copy the worked example as your result.

Precautions

- Match the defined temperature and timing.
- Do not use one kit's factor with another method.
- Haemolysis, matrix interference and nonlinearity may invalidate interpretation.

Viva Questions and Answers

1. What are AST and ALT?

Aminotransferase enzymes measured through an activity assay.

2. What does U/L express?

Enzyme activity per litre under the defined assay conditions.

3. Can an isolated rise diagnose a cause?

No; clinical context and other findings are needed.

Comparison of Acid and Amylase Hydrolysis of Starch

Experiment 25 - Practical Notes

Name: _____ Roll no.: _____ Date: _____

Aim

To compare starch hydrolysis in two instructor-assigned teaching systems.

Principle

Both acid treatment and amylase can break starch into smaller carbohydrate products, but their mechanisms and suitable conditions differ. Iodine and reducing-sugar tests monitor different aspects of the change.

Requirements

- Assigned starch preparation, instructor-prepared acid and amylase systems
- Timed water baths, labelled aliquots, iodine and reducing-sugar test reagents

Procedure

1. Plan

Label starch blank, acid-treated and enzyme-treated systems with the assigned conditions.

2. Begin

Start the supervised systems and record time and conditions.

3. Take teaching aliquots

At the assigned times, perform the prescribed stopping/neutralisation where needed before tests.

4. Compare

Test appropriate portions for residual starch and reducing products using controls.

5. Interpret

Explain why disappearance of iodine colour and appearance of reducing response are complementary findings.

Observations and Record

System / time	Iodine response	Reducing-sugar response
Untreated control	_____	_____
Acid system - assigned time 1	_____	_____
Acid system - assigned time 2	_____	_____
Amylase system - assigned time 1	_____	_____
Amylase system - assigned time 2	_____	_____

Result

Under the assigned conditions, starch response changed as _____ and reducing-product response as _____. Comparison and limitations: _____.

Precautions

- Use assigned controlled conditions; enzyme activity depends on temperature and pH.
- Acid-treated aliquots require the prescribed preparation before alkaline tests.
- Do not infer that every hydrolysis product is glucose.

Viva Questions and Answers

1. What does iodine indicate?

Suitable residual starch structure.

2. What does a reducing test indicate?

Reducing carbohydrate products under the test conditions.

3. Why can excessive heat stop amylase activity?

It can denature the enzyme.

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