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URINALYSIS.....

ROUTINE & MICROSCOPIC
TECHNICAL PROCEDURE MANUAL

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URINALYSIS PRINCIPLE

Urinalysis is a routine laboratory test that analyzes the physical, chemical, and microscopic properties of urine. It provides valuable information about the health of the kidneys and urinary tract, as well as other organ systems. This manual outlines the procedures for performing a complete urinalysis, including routine analysis (physical and chemical properties) and microscopic examination.

This principle is further strengthened by the dynamic nature of urine, which reflects the body's internal milieu. Variations in dietary intake, hydration status, medication use, and even stress can influence the composition of urine. By meticulously analyzing these variations, urinalysis provides a snapshot of the body's current state, potentially revealing early signs of disease or dysfunction. This makes it a valuable tool for both routine health screenings and the diagnosis and management of a wide range of medical conditions.



URINALYSIS

SPECIMEN REQUIREMENTS

Criteria for Specimen Collection, Labeling, Rejection, Storage, and Transport

Specimen Collection

A. Specimen Types

- **Random Specimen:** Collected at any time of day, suitable for routine urinalysis (physical and chemical analysis) and microscopic examination.
- **First Morning Specimen (8-hour Specimen):** Collected immediately upon waking, preferred for routine and microscopic urinalysis as it is more concentrated.
- **Midstream “Clean Catch” Specimen:** Collected after cleansing the genital area, provides a less contaminated specimen for microscopic analysis and bacterial culture (if indicated).

B. Patient Instructions:

- Provide clear and concise instructions to patients on how to collect the appropriate specimen type.
- Explain the importance of proper hygiene and collection techniques to minimize contamination.
- Include information on the required volume of urine for analysis (typically 12 ml).

Specimen Labeling

- **Patient Identification:** Each specimen container must be labeled with the patient's full name, date of birth, unique identification number (e.g., medical record number), and date and time of collection.
- **Specimen Type:** Clearly indicate the type of specimen collected (e.g., random, first morning, clean catch).
- **Labelling Materials:** Use waterproof, permanent labels that will not smudge or fade.
- **Legibility:** Ensure all labels are legible and complete.

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SPECIMEN REQUIREMENTS

Specimen Rejection Criteria

- Improperly Labeled Specimens: Specimens lacking complete and accurate labeling must be rejected and recollected.
- Contaminated Specimens: Specimens showing evidence of contamination (e.g., fecal material, vaginal discharge) must be rejected.
- Insufficient Volume: Specimens with less than the minimum required volume (usually 12 ml) may be rejected or analyzed with a note indicating the reduced volume.
- Inappropriate Containers: Specimens collected in inappropriate containers (e.g., syringes) must be rejected.
- Leaking Specimens: Specimens leaking from the collection container are unacceptable.

Storage Guidelines

- Refrigeration: Urine specimens should be refrigerated at 2-8°C (36-46°F) as soon as possible after collection.
- Storage Time: Refrigerated specimens should be transported to the laboratory within 2 hours of collection.



URINALYSIS

SPECIMEN REQUIREMENTS

Transport Recommendations

- Transport Containers: Use leak-proof, sterile containers for transport.
- Transport Time: Transport specimens to the laboratory as soon as possible after collection.
- Temperature Control: Maintain the appropriate temperature during transport (refrigerated).



URINALYSIS

REAGENTS, MEDIA, SUPPLIES, AND EQUIPMENTS

Reagents and Media

- **Reagent Strips:** Commercially available strips containing multiple reagent pads for testing various chemical parameters in urine (e.g., pH, glucose, protein, ketones, blood, nitrites, leukocyte esterase, bilirubin, urobilinogen, specific gravity).

- **Staining Solutions:** For microscopic examination, staining solutions are used to enhance the visibility of cellular elements and casts in urine sediment. Commonly used stains include:

Gram Stain: Differentiates bacteria based on their cell wall structure.

Methylene Blue: Stains nuclear material and bacteria.

Sudan III: Stains fat droplets.

Acetic Acid: Used to enhance the visibility of red blood cells and casts.

- **Microscopic Mounting Medium:** A transparent medium used to mount urine sediment on a microscope slide for examination.



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REAGENTS, MEDIA, SUPPLIES, AND EQUIPMENTS

Supplies

- Urine Collection Containers: Sterile, leak-proof containers with a capacity of at least 12 ml.
- Microscope Slides: Clean, standard microscope slides for mounting urine sediment.
- Coverslips: Clean, standard coverslips to cover the urine sediment on the microscope slides.
- Pipettes: Disposable pipettes for transferring urine to reagent strips and for preparing urine sediment.
- Test Tubes: Small test tubes for holding urine samples.
- Centrifuge Tubes: Graduated centrifuge tubes for concentrating urine sediment.
- Centrifuge: A laboratory centrifuge to spin urine samples to concentrate the sediment.
- Gloves: Disposable gloves for handling urine specimens and reagents.
- Lab Coats: Protective lab coats for handling specimens and reagents.
- Safety Glasses: Protective safety glasses for handling reagents and specimens.



URINALYSIS

REAGENTS, MEDIA, SUPPLIES, AND EQUIPMENTS

Equipment

- **Microscope:** A compound light microscope with objective lenses of 10x, 40x, and 100x magnification for examining urine sediment.
- **Reagent Strip Reader:** A device used to interpret the results of reagent strip tests.
- **Centrifuge:** A laboratory centrifuge for spinning urine samples to concentrate sediment.
- **Incubator:** For culturing bacteria (if performing a urine culture).
- **Water Bath:** For warming reagents or solutions.
- **Biohazard Waste Containers:** For proper disposal of biohazardous materials, including urine specimens and used reagents.



URINALYSIS CALIBRATION

I. Reagent Strips

- **Manufacturer's Instructions:** Follow the manufacturer's instructions meticulously for calibrating reagent strips. These instructions will typically include:
- **Storage:** Proper storage is crucial for maintaining reagent strip accuracy. Follow the manufacturer's recommendations for storage temperature and humidity. Often, this means storing strips in a cool, dry, dark place.
- **Expiration Dates:** Use reagent strips within their expiration dates. Expired strips may not provide accurate results due to the potential for degradation of the chemical reagents on the strips.
- **Quality Control:** Use manufacturer-supplied quality control materials (e.g., positive and negative controls) to verify the accuracy of the reagent strips before use. This helps to ensure that the strips are functioning correctly and can detect the target analytes.



URINALYSIS CALIBRATION

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II. Microscope

- **Stage Micrometer:** Calibrate the microscope using a stage micrometer, a slide with a precisely etched scale, to ensure accurate measurements of microscopic elements in urine sediment.

Procedure:

1. Place the stage micrometer on the microscope stage and focus on the scale under the desired objective lens (e.g., 40x).
2. Align the stage micrometer scale with the eyepiece graticule (a reticle with a scale in the eyepiece).
3. Adjust the eyepiece graticule until a known number of divisions on the stage micrometer scale correspond to a specific number of divisions on the eyepiece graticule. This establishes a conversion factor for measuring microscopic elements.
4. Record the conversion factor for each objective lens used.
5. Repeat the calibration procedure regularly to ensure accuracy.

III. Refractometer (for specific gravity)

- **Distilled Water:** Calibrate the refractometer using distilled water, which has a specific gravity of 1.000.

Procedure:

1. Place a few drops of distilled water on the prism of the refractometer.
2. Close the lid and look through the eyepiece.
3. Adjust the calibration screw until the reading on the scale aligns with the specific gravity of 1.000.
4. Clean the prism with a soft cloth and distilled water after calibration.
5. Repeat the calibration procedure regularly to ensure accuracy.

URINALYSIS CALIBRATION

IV. pH Meter

- **Standard Buffer Solutions:** Calibrate the pH meter using standard buffer solutions with known pH values.

Procedure:

1. Immerse the pH meter probe in a buffer solution with a pH of 7.00 (neutral).
2. Allow the meter to stabilize and adjust the calibration knob until the reading matches the known pH of the buffer solution.
3. Repeat the calibration process with a second buffer solution with a different pH value (e.g., 4.01 or 10.01).
4. Clean the pH meter probe thoroughly with distilled water after calibration.
5. Repeat the calibration procedure regularly to ensure accuracy.

V. Documentation and Quality Control

- **Calibration Records:** Maintain accurate records of all calibration procedures, including:
 - **Date and Time of Calibration:** Record the date and time of each calibration event.
 - **Calibration Materials Used:** Document the specific calibration materials used (e.g., lot numbers, expiration dates).
 - **Calibration Results:** Record the calibration results, including any deviations from expected values.
 - **Corrective Actions:** Document any corrective actions taken if calibration results indicate a deviation from expected values.
 - **Regular Monitoring:** Perform regular quality control checks using positive and negative controls to monitor the accuracy and reliability of the reagent strips, reader (if applicable), and other equipment.
 - **Proficiency Testing:** Participate in proficiency testing programs to evaluate the accuracy and reliability of the urinalysis procedures, including the calibration of equipment and reagents.

URINALYSIS QUALITY CONTROL

Specimen Collection and Handling

Proper Collection:

- Ensure clear instructions are provided for appropriate specimen collection, including type of container, volume required, and specific instructions for midstream clean-catch urine collection.

- Emphasize the importance of labeling specimens correctly with patient information and the date and time of collection.

Storage and Transportation:

- Define appropriate storage conditions for urine specimens, including temperature and time limits for analysis.

- Establish protocols for safe and timely transportation of specimens to the laboratory.

Specimen Integrity:

- Implement procedures to assess specimen integrity, including visual inspection for contamination, turbidity, or unusual color.

- Document any observations and take appropriate action if necessary (e.g., reject specimen, request a new collection).

Routine Urinalysis

Dipstick Testing:

- Utilize commercially available dipstick reagents with established quality control procedures.

- Run control materials (positive and negative) with each batch of dipsticks to ensure their accuracy and reliability.



URINALYSIS

QUALITY CONTROL

- Document control results and investigate any discrepancies promptly.
- Establish clear interpretation guidelines for dipstick results, including reference ranges and potential clinical significance.

Specific Gravity:

- Calibrate refractometers or other specific gravity measurement devices regularly.
- Use control solutions with known specific gravity values to verify instrument accuracy.
- Document calibration and control results.

Quality Control Records:

- Maintain detailed records of all quality control activities, including dates, control materials used, results obtained, and any corrective actions taken.

Microscopic Urinalysis

Microscope Calibration:

- Ensure the microscope is properly calibrated and maintained.
- Use a stage micrometer to verify the accuracy of the objective lenses.
- Document calibration results and perform regular checks.



URINALYSIS QUALITY CONTROL

Staining Procedures:

- Use standardized staining procedures for microscopic examination (e.g., Sternheimer-Malbin stain, Gram stain).
- Utilize control slides with known cellular elements to verify staining quality.
- Document control results and troubleshoot any staining issues.

Microscopic Examination:

- Establish clear guidelines for microscopic examination, including the number of fields to be examined, the criteria for identifying different cellular elements, and the reporting of findings.
- Use reference materials (e.g., atlases, images) to ensure consistent identification and interpretation.

Reporting and Interpretation:

- Develop standardized reporting formats for microscopic findings, including the number and types of cells, casts, crystals, and other elements observed.
- Provide clear interpretations of microscopic findings, linking them to potential clinical conditions.



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STEP-BY-STEP TESTING PROCEDURES

Sample Collection and Handling

- Collect a fresh urine sample: Ideally, collect the sample first thing in the morning, as the urine is more concentrated at this time .
- Use a clean-catch method: This involves cleansing the urinary opening to minimize contamination with bacteria from the surrounding skin .
- Store the sample properly: If the sample cannot be analyzed within 30 minutes, refrigerate it to prevent bacterial growth and changes in the chemical composition of the urine .
- Warm the sample to room temperature before analysis: If the sample was refrigerated, allow it to warm up to room temperature before proceeding with the analysis.

Routine Urinalysis

2.1. Visual Examination

- Color: Normal urine ranges from pale yellow to amber, depending on concentration. Dark yellow to brown urine may indicate bilirubin presence. Red urine can be caused by hematuria (blood in the urine), hemoglobin, or myoglobin .
- Clarity: Healthy urine is typically clear. Cloudiness indicates the presence of formed elements like cells, crystals, mucus, casts, bacteria, or spermatozoa .
- Odor: While not routinely assessed, a strong or unusual odor might suggest an infection .



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STEP-BY-STEP TESTING PROCEDURES

2.2. Dipstick Test

- **Prepare the dipstick:** Ensure the dipstick is within its expiration date. Remove a dipstick from the container, avoiding contact with the reagent squares.
- **Immerse the dipstick:** Dip the dipstick into the well-mixed urine sample, ensuring all reagent squares are fully immersed.
- **Remove and blot excess urine:** Remove the dipstick immediately and gently tap off excess urine onto a paper towel, holding it horizontally to prevent cross-contamination of the reagent squares.
- **Interpret the results:** Use the urinalysis guide on the dipstick container to interpret the findings. Different reagent squares require different interpretation times, so ensure you interpret the correct test at the appropriate time interval.

Microscopic Examination

- **Centrifuge the urine sample:** Transfer about 10 ml of urine to a centrifuge tube and spin it at 1,500 rpm for 5 minutes. This will concentrate the formed elements at the bottom of the tube.
- **Prepare a slide:** Discard the supernatant and resuspend the sediment by gently tapping the tube. Place a drop of the sediment onto a microscope slide and cover it with a coverslip.
- **Examine under the microscope:** Start by scanning the slide under low magnification (100x) and then increase the magnification (400x) for a closer look.



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STEP-BY-STEP TESTING PROCEDURES

Identify and quantify formed elements:

Cells:

- Red blood cells (RBCs): High numbers (more than 5 per high-power field) indicate bleeding along the urinary tract .
- White blood cells (WBCs): High numbers (more than 5 per high-power field) suggest a urinary tract infection or inflammation .
- Squamous epithelial cells: These are normal in small numbers but high numbers indicate contamination .
- Renal tubular epithelial cells (RTEs): These cells originate from the kidney tubules and their presence in high numbers indicates kidney damage .
- Transitional epithelial cells: These cells line the ureters, bladder, and urethra. High numbers can indicate a urinary tract infection .
- Crystals: These can form due to dehydration or other factors and may indicate kidney stones or other conditions .
- Casts: These are cylindrical structures formed in the kidney tubules and can indicate kidney disease or infection .
- Microorganisms: The presence of bacteria, fungi, or parasites indicates an infection .



URINALYSIS CALCULATIONS

Routine Urinalysis Calculations

- **Specific Gravity:** This is a measure of the concentration of solutes in the urine. It is calculated by comparing the density of urine to the density of water.
- **Creatinine Clearance:** This test measures how well your kidneys are filtering waste products from your blood. It is calculated using a urine sample and a blood sample.

Microscopic Urinalysis Calculations

- **Red Blood Cell Count:** This is typically reported as the number of red blood cells per high-power field (HPF) in a microscopic examination of urine.
- **White Blood Cell Count:** Similar to red blood cell count, this is reported as the number of white blood cells per HPF.
- **Casts:** The type and number of casts are typically reported, rather than calculated.



URINALYSIS REPORTING RESULTS

Reference Intervals for Urinalysis Parameters

Reference ranges may vary slightly based on the laboratory and individual health conditions. The general adult reference values are as follows in the given table below:

SPECIMEN VALIDITY TESTING

| REPORTED PRESCRIPTION | TEST OUTCOME | MEASURED RESULT | UNITS | REFERENCE RANGE |
|-----------------------|--------------|-----------------|---------|--|
| pH | Normal | 6 | pH | 5 - 9 |
| Specific Gravity | Normal | 1.015 | | 1.000 - 1.060 |
| Glucose | Normal | 0 | mg/dL | <20 (Max > 500) |
| Protein | Normal | 0 | mg/dL | <20 (Max > 500) |
| Bilirubin | Normal | 0 | mg/dL | <1.80 (Max > 4.00) |
| Urobilinogen | Normal | 0 | mg/dL | <1.60 (Max > 4.00) |
| Blood | Normal | 0 | mg/dL | <0.02 (Max > 1.00) |
| Ketone | Normal | 0 | mg/dL | <3 (Max > 80) |
| Nitrite | Normal | Negative | mg/dL | 0 - 1 (Max > 1) |
| Leukocytes | Normal | 0 | WBCs/uL | 15 - 40 (Max > 500) |
| Ascorbic Acid | Abnormal | 0 | mg/dL | <17 (Max > 40) |
| Clarity | Normal | Clear | | Clear/ Slightly-Cloudy/ Cloudy/ Turbid |
| Color | Normal | Yellow | | Yellow/ Amber/ Red/ Blue/ Colorless/ Straw |

URINALYSIS REPORTING RESULTS

Reference for Dipstick Test

| TESTS AND READING TIME | | | | | | | | |
|------------------------|------------------|----------|---------------------|--------------------|-----------------|-------------|-------------|---|
| LEU | LEUKOCYTES | Negative | | Trace | Small + | Moderate ++ | Large +++ | |
| | 2 minutes | | | | | | | |
| NIT | NITRITE | Negative | | | | | | |
| | 60 seconds | | | | | | | Positive (any degree of uniform pink color) |
| URO | UROBILINOGEN | 0.2 | 1 | 2 | 4 | 8 | | |
| | 60 seconds | | | | | | | |
| PRO | PROTEIN | Negative | Trace | mg/dL | 30 + | 100 ++ | 300 +++ | 2000 or more ++++ |
| | 60 seconds | | | | | | | |
| pH | pH | 5.0 | 6.0 | 6.5 | 7.0 | 7.5 | 8.0 | 8.5 |
| | 60 seconds | | | | | | | |
| BLO | BLOOD | Negative | Non-hemolyzed Trace | Hemolyzed Moderate | Hemolyzed Trace | Small + | Moderate ++ | Large +++ |
| | 60 seconds | | | | | | | |
| SG | SPECIFIC GRAVITY | 1.000 | 1.005 | 1.010 | 1.015 | 1.020 | 1.025 | 1.030 |
| | 45 seconds | | | | | | | |
| KET | KETONE | Negative | mg/dL | Trace 5 | Small 15 | Moderate 40 | Large 80 | Large 160 |
| | 40 seconds | | | | | | | |
| BIL | BILIRUBIN | Negative | | | Small + | Moderate ++ | Large +++ | |
| | 30 seconds | | | | | | | |
| GLU | GLUCOSE | Negative | g/dL (%) | 1/10 (tr.) | 1/4 | 1/2 | 1 | 2 or more |
| | 30 seconds | | mg/dL | 100 | 250 | 500 | 1000 | 2000 or more |

URINALYSIS REPORTING RESULTS

Reporting Format

The proper reporting format for Urinalysis is as follows:

| Test Report | | | |
|--|-----------------------|-------|--------------------|
| Test Name | Results | Units | Bio. Ref. Interval |
| URINE EXAMINATION, ROUTINE; URINE R/E, AUTOMATED | | | |
| Physical | | | |
| Colour | Pale Yellow | | Pale Yellow |
| Specific gravity | 1.025 | | 1.000 -1.030 |
| Ph | 6.0 | | 5.0 - 8.0 |
| Chemical | | | |
| Proteins | Trace | | Negative |
| Glucose | Negative | | Negative |
| Ketones | Negative | | Negative |
| Bilirubin | Negative | | Negative |
| Urobilinogen | Negative | | Negative |
| Blood | Negative | | Negative |
| Leukocyte esterase | Present 1+(25 WBC/uL) | | Negative |
| Nitrite | Negative | | Negative |
| Ascorbic Acid | Negative | | Negative |
| Microscopy | | | |
| RBC | 1.80 | /hpf | 0.0 - 2.0 |
| Pus cells | 5.70 | /hpf | 0.0 - 5.0 |
| Epithelial cells | 13.60 | /hpf | 0.0 - 5.0 |
| Calcium oxalate monohydrate crystals | 0.00 | /hpf | 0.0 - 0.99 |
| Calcium oxalate dihydrate crystals | 21.50 | /hpf | 0.0 - 0.99 |
| Triple Phosphate crystals | 0.00 | /hpf | 0.0 - 0.99 |
| Uric acid crystals | 0.00 | /hpf | 0.0 - 0.99 |
| Calcium Phosphate | 0.00 | /hpf | 0.0 - 0.99 |
| Cystine crystals | 0.00 | /hpf | 0.0 - 0.99 |
| Leucine crystals | 0.00 | /hpf | 0.0 - 0.99 |
| Tyrosine crystals | 0.00 | /hpf | 0.0 - 0.99 |
| Amorphous urates crystals | 0.00 | /hpf | 0.0 - 0.99 |
| Amorphous phosphate crystals | 0.00 | /hpf | 0.0 - 0.99 |



URINALYSIS PROCEDURE NOTES

Specimen Integrity: Handle specimens carefully to avoid contamination and maintain their integrity.

Reagent Strip Handling: Use reagent strips according to the manufacturer's instructions.

Microscope Use: Ensure proper lighting and focus when using the microscope.

Reporting: Report results accurately and completely.

Safety: Wear appropriate personal protective equipment (PPE) when handling specimens and reagents.



URINALYSIS

METHOD LIMITATIONS

Reagent Strips: Reagent strips may have limitations in sensitivity and specificity, leading to false-positive or false-negative results.

Microscopic Examination: Microscopic examination is subjective and may be influenced by the experience of the examiner.

Specimen Quality: Specimen quality can affect the accuracy of the results.



URINALYSIS

TROUBLESHOOTING OR BACKUP PLANS

Reagent Strip Issues: If reagent strips are not functioning correctly, use a different lot number or brand of strips.

Microscope Problems: If the microscope is not working properly, use a different microscope or contact the manufacturer for repair.

Specimen Contamination: If the specimen is contaminated, collect a new specimen.

Unexpected Results: If unexpected results are obtained, repeat the test or consult with a senior laboratory professional.



REFERENCES

Ag. (2023, March 6). *Urine test report* [Online forum post]. BabyCenter.

<https://community.babycenter.com/post/a78054082/urine-test-report>

Alcala Testing and Analysis Services. (2019, September 26). *Urine Toxicology*. <http://www.alcalalabs.com/urine-toxicology/>

DJMed Pty Ltd. (2024, October 23). *MultiStix 10SG Urinalysis Test Strips*. DearJane Medical.

<https://dearjane.com.au/product/multistix-10sg-urinalysis-reagent-test-strips/>

Krans, B. (2018, September 20). *Urinalysis*. Healthline. <https://www.healthline.com/health/urinalysis#test-process>

Urinalysis - mayo clinic. (n.d.). <https://www.mayoclinic.org/tests-procedures/urinalysis/about/pac-20384907>

Urinalysis (urine test). (n.d.). National Kidney Foundation. <https://www.kidney.org/kidney-topics/urinalysis-urine-test>

Urinalysis (Urine test). (2024, September 5). WebMD. <https://www.webmd.com/a-to-z-guides/what-is-urinalysis>



EFFECTIVE DATE
EFFECTIVE UPON APPROVAL

**SIGNATURE OF THE
LABORATORY DIRECTOR**



KIARRA LEE HYACINTH P. CARREON