Urine Tests

A Case-Based Guide to Clinical Evaluation and Application

Victoria J. A. Sharp
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Preface

Consider a medical environment in which urine tests are not readily available for interpretation. Urinalysis for identification of blood and glucose, urine culture for diagnosis of urinary tract infection, and urine pregnancy and drug screening are just a few of the urine tests that have become mainstays in modern medical practice. More recent technological advances in laboratory testing have allowed for expansion of urine-based testing; they are now considered accurate, noninvasive, and cost-effective options for diagnosis of sexually transmitted infections, renal insufficiency, and even cancer screening.

Urine Tests: A Case-Based Guide to Clinical Evaluation and Application was conceived in response to the recognition that urine tests are essential in both inpatient and outpatient care settings, yet no clinically based resource exists devoted to understanding their nuances. Appropriate collection, analysis, and interpretation of urine for testing are essential for the diagnosis and management of both common and rare patient concerns. Thus, a handbook describing urine collection technique, methods of analyzing urine, specific tests available, and interpretation for simple and complex diagnoses is an essential tool.

This book is a collaborative effort between specialists, primary care providers, administrators, and researchers who have expert knowledge of urine testing. The authors of this book perform urine testing on patients daily and fully understand the advantages and pitfalls of urine-based tests. The

focus of this book is to provide practical knowledge and clinically relevant scenarios for urine-based tests in order to improve overall patient care.

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Abbreviations

AAP	American Academy of Pediatrics
ACEi	Angiotensin-converting enzyme inhibitor
ACR	Albumin creatinine ratio
ADH	Antidiuretic hormone
AFB	Acid-fast bacillus (bacilli)
AG	Anion gap
AIN	Acute interstitial nephritis
AKI	Acute kidney injury
ANA	Antinuclear antibody
ANC	Absolute neutrophil count
ANCA	Antineutrophil cytoplasmic antibodies
Anti-dsDNA	Anti-double stranded DNA antibodies
APOL1	Apolipoprotein L1
ARB	Angiotensin II receptor blocker
ARR	Aldosterone:renin ratio
ASO	Anti-streptolysin O
ATN	Acute tubular necrosis
AUA	American Urological Association
BCG	*Bacille Calmette-Guérin*
β-hCG	beta-human chorionic gonadotropin
BMP	Basic metabolic panel
BP	Blood pressure
BPH	Benign prostatic hyperplasia
BUN	Blood urea nitrogen
C	Celsius
C3	Complement C3
C3GN	Complement C3 glomerulonephritis

C4	Complement C4
Ca	Calcium
c-ANCA/PR3	cytoplasmic antineutrophil cytoplasmic antibodies/proteinase 3
CBC	Complete blood count
CBC w/diff	Complete blood count with differential
CDC	Centers for Disease Control and Prevention
CDI	*Clostridium difficile* infection
CFU	Colony-forming unit
CK	Creatinine kinase
CKD	Chronic kidney disease
Cl	Chloride
CLIA	Clinical Laboratory Improvement Amendments
CMS	Centers for Medicare and Medicaid Services
CNS	Central nervous system
CPT	Current Procedural Terminology
Cr	Creatinine
CT	Computed tomography
CTU	Computed tomography urography
CVD	Cardiovascular disease
DDAVP	Desmopressin
DDD	Dense deposit disease
DI	Diabetes insipidus
DKA	Diabetic ketoacidosis
dL	deciliter
DM	Diabetes mellitus
DNA	Deoxyribonucleic acid
DRE	Digital rectal exam
EAU	European Association of Urology
ED	Emergency department
EGPA	Eosinophilic granulomatosis with polyangiitis
ELISA	Enzyme-linked immunosorbent assay
ENaC	Epithelial sodium channel
ESRD	End-stage renal disease
F	Fahrenheit or French size
FDA	Food and Drug Administration
FeNa	Fractional excretion of sodium
FeUrea	Fractional excretion of urea

FISH	Fluorescence in situ hybridization
FSGS	Focal segmental glomerulosclerosis
g	gram
GBM	Glomerular basement membrane
GC-MS	Gas chromatography-mass spectrometry
GFR	Glomerular filtration rate
GI	Gastrointestinal
GPA	Granulomatosis with polyangiitis
GU	Genitourinary
H	Hydrogen (hydronium)
H_2O	Water
H_2O_2	Hydrogen peroxide
HAGMA	High anion gap metabolic acidosis
Hb, Hgb	Hemoglobin
HBV	Hepatitis B virus
HCO_3	Bicarbonate
HCPCS	Healthcare Common Procedure Coding System
HCV	Hepatitis C virus
HIV	Human immunodeficiency virus
hpf	high power field
HPLC/MS	High-performance liquid chromatography mass spectrometry
HPV	Human papilloma virus
HTN	Hypertension
IDSA	Infectious Diseases Society of America
IgA	Immunoglobulin A
IgAN	Immunoglobulin A nephropathy
IgM	Immunoglobulin M
INR	International normalized ratio
IV	Intravenous
K	Potassium
KDIGO	Kidney Disease Improving Global Outcomes
kg	kilogram
KOH	Potassium hydroxide
KUB	Kidney, ureter, and bladder x-ray
L	Liter
LC/MS	Liquid chromatography mass spectrometry
LE	Leukocyte esterase

LMW	Low molecular weight
LN	Lupus nephritis
lpf	low power field
LSD	Lysergic acid diethylamide
MAR	Medication Administration Record
Mb	Myoglobin
MCD	Minimal change disease
mEq	milliequivalent
mg	milligram
Mg	Magnesium
MGN	Membranous nephropathy
mL	milliliter
mm	millimeter
mmol	millimole
mOsm	milliosmole
MPA	Microscopic polyangiitis
MPGN	Membranoproliferative glomerulonephritis
MRI	Magnetic resonance imaging
MRU	Magnetic resonance urography
Na	Sodium
NAAT	Nucleic acid amplification test
NAGMA	Non-anion gap metabolic acidosis
NH_3	Ammonia
NH_4	Smmonium
nm	nanometer
NPV	Negative predictive value
NSAID	Nonsteroidal anti-inflammatory drug
PAC	Plasma aldosterone concentration
p-ANCA/ MPO-ANCA	perinuclear antineutrophil cytoplasmic antibodies/myeloperoxidase antineutrophil cystoplasmic antibodies
Pap	Papanicolaou
PCA3	Prostate cancer antigen 3
PCH	Paroxysmal cold hemoglobinuria
PCN	Percutaneous nephrostomy
PCNL	Percutaneous nephrolithotomy
PCP	Phencyclidine
PCR	Protein creatinine ratio OR polymerase chain reaction

pH	potential of hydrogen
PID	Pelvic inflammatory disease
PIGN	Postinfectious glomerulonephritis
PLA2R	Phospholipase A2 receptor
PNH	Paroxysmal nocturnal hemoglobinuria
PPI	Proton pump inhibitor
PPV	Positive predictive value
PPM	Provider-performed microscopy
PRA	Plasma renin activity
PSA	Prostate-specific antigen
RAS	Renal artery stenosis
RAAS	Renin-angiotensin-aldosterone system
RBC	Red blood cell
RNA	Ribonucleic acid
RTA	Renal tubular acidosis
rTEC	renal tubular epithelial cell
RVU	Relative value unit
SG	Specific gravity
SGLT2	Sodium glucose co-transport protein 2
SIADH	Syndrome of inappropriate antidiuretic hormone secretion
SLE	Systemic lupus erythematosus
SPEP/IFE	Serum protein electrophoresis/ immunofixation
STI	Sexually transmitted infection
SWL	Shock wave lithotripsy
TB	Tuberculosis
TCA	Tricyclic antidepressant
THC	Tetrahydrocannabinol
ttg Ab	tissue transglutaminase antibody
TTP	Thrombotic thrombocytopenic purpura
UA	Urinalysis
UPEP/IFE	Urine protein electrophoresis/ immunofixation
US	Ultrasound
USPSTF	United States Preventative Services Task Force
UTI	Urinary tract infection
WBC	White blood cell

Contents

Editor Bios

Sharp obtained her MD degree from the University of Arizona following a BS in Nursing from George Mason University and later an MBA from the University of Iowa. She did residency training in Urology and Family Medicine at the University of Iowa Hospitals and Clinics and is Board Certified in Family Medicine. As Clinical Professor of Urology and Family Medicine at the University of Iowa Carver College of Medicine, she practices and teaches medical urology at the Iowa City VA Medical Center on a limited basis. Previously she was Chief Medical Officer for AmeriHealth Caritas Iowa, a Medicaid managed care organization, and is currently a Clinical Transformation Physician Consultant for 3M Health Information Systems. She is interested in quality improvement, performance optimization, and change management.

Sanders obtained his MD degree from the University of Tennessee Health Science Center after completion of a PhD in Pharmaceutical Sciences. He completed internal medicine residency and chief residency at the University of Iowa Hospitals and Clinics followed by completion of both a general nephrology and transplant nephrology fellowship at Vanderbilt University Medical Center. He is Board Certified in both Internal Medicine and Nephrology, and his current medical practice incorporates internal medicine, general nephrology, and transplant nephrology. His academic interests include medical education and kidney transplant outcomes.

Lockwood obtained her MD degree from Southern Illinois University School of Medicine. She completed urology residency at the Medical College of Wisconsin. She completed her pediatric urology fellowship at Connecticut Children's Medical Center and obtained her Master of Science degree in Clinical and Translational Research at University of Connecticut. She is Board certified in Urology. She currently practices pediatric urology and has research interests in patient and parent education and quality improvement.

Antes obtained her MD degree from UMDNJ Robert Wood Johnson Medical School. She completed internal medicine residency and chief residency at Montefiore Medical Center and her nephrology fellowship at the University of Pennsylvania. She was in private practice for 2 years prior to joining the nephrology division at the University of Iowa Hospitals and Clinics in 2000. She has been the Nephrology Fellowship Program Director since 2015. Her current medical practice incorporates patient care in internal medicine and nephrology and education of students, residents, and fellows.

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Chapter 1
Urine: The Golden Elixir of Life

M. Lee Sanders and Lisa M. Antes

Objectives

- Understand the basic anatomy of the urinary system
- Discuss glomerular filtration as the process of urine formation
- Describe how urine composition and amount are determined by a series of specialized tubules

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Overview

Many scientists as well as philosophers have recognized the importance of urination and how this basic physiologic function is vital to sustain life. The urinary system and tract are responsible for the production, refinement and elimination of urine from the body. A thorough examination of urine provides valuable information that assists in patient care. This book is dedicated to the clinical application of urine tests; however before test application, one should understand some basic principles of urine production, refinement and elimination.

The kidney presents in the highest degree the phenomenon of sensibility, the power of reacting to various stimuli in a direction which is appropriate for the survival of the organism; a power of adaptation which almost gives one the idea that its component parts must be endowed with intelligence. (Frank Starling, 1909)

Urine Production

The kidney is the organ responsible for urine production. Humans normally have two separate "bean shaped" kidney organs located to the left and right of the spine in the retroperitoneal space. The kidneys are highly vascular organs receiving an average 20–25% of the cardiac output, which is remarkable since the kidneys only comprise 0.5% of total body weight [1, 2].

The basic filtering unit of the kidney is the nephron. The nephron is composed of a glomerulus, a series of tubules and a collecting duct. Exact nephron number across individuals is variable but total number is determined/finalized at birth. After birth, new nephrons cannot be developed and lost nephrons cannot be replaced. Each kidney on average contains approximately one million nephrons [1, 2].

Blood is supplied to the nephron through a series of arteries finally reaching the glomerular capillaries via the afferent arteriole and leaving the capillary bed via the efferent arteriole (Fig. 1.1). The hydrostatic pressure in the capillary bed forces

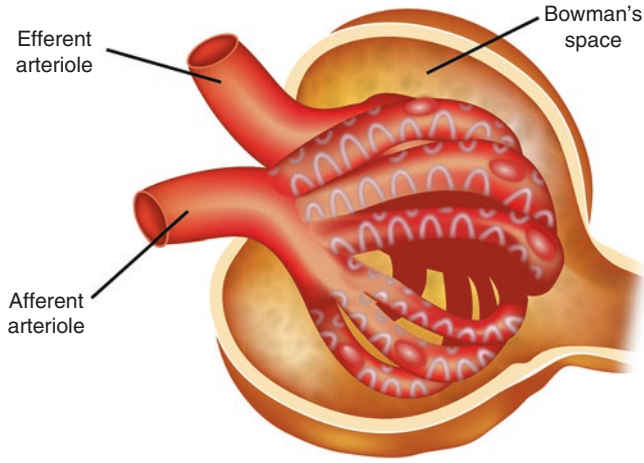

FIGURE 1.1 Capillary bed of the glomerulus. (Courtesy of Teresa Ruggle, University of Iowa.)

fluid to move from the blood compartment across the semi-permeable glomerular membrane into the urinary space (Bowman's space). This ultrafiltrate is essentially the same osmolality as plasma and includes water, small molecules, and ions that easily pass through the filtration membrane [1, 2].

Larger molecules such as proteins and red blood cells are normally prevented from passing through the filtration membrane. This filtration membrane or barrier is comprised of three components: (a) the endothelial cells of the renal capillaries, (b) the basement membrane and (c) the epithelial cells lining the urinary space. Evidence of protein (see Chap. 5) or red blood cells (see Chap. 9) in the urine could therefore be a sign that this barrier is compromised [3–5].

Urine Refinement

The ultrafiltrate in Bowman's space will then pass through a series of tubules and a collecting duct. This refinement process allows for the secretion of additional waste products in

addition to reabsorption of water and solutes from the ultrafiltrate. This refinement process is also required to sustain life. In a typical 70 kilogram individual, the kidney filters approximately 180 liters of fluid daily. Life would cease to exist without the ability to reabsorb solutes and water from the filtrate. The kidneys are very efficient at this process as only 1–2 liters of urine on average are excreted daily while maintaining electrolyte, mineral and pH balance in the blood.

Four major tubular segments of the nephron (Fig. 1.2) determine the final composition and volume of the urine: (a) proximal convoluted tubule, (b) loop of Henle, (c) distal convoluted tubule and (d) collecting duct. Each individual segment of the tubule possesses a unique set of channels and transporters that allow reabsorption and excretion to occur. The main ion that is reabsorbed through these segments is sodium. Filtering 180 liters a day would lead to the theoretical excretion of approximately 25,500 mmol per day of sodium, a

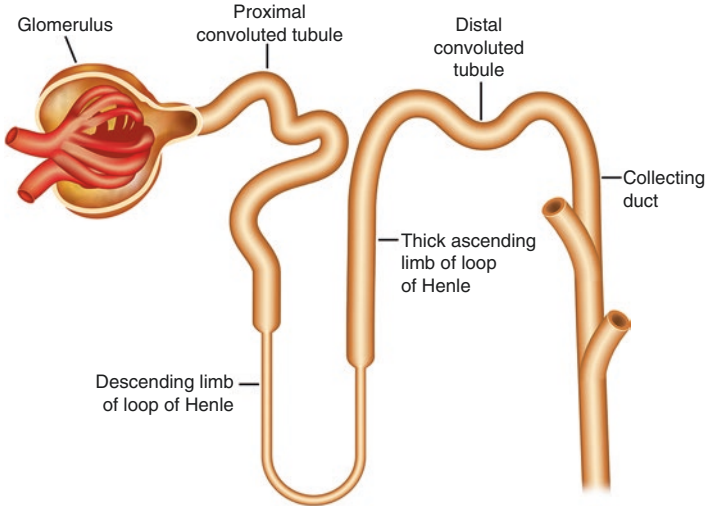

FIGURE 1.2 The nephron of the kidney. (Courtesy of Teresa Ruggle, University of Iowa.)

loss that would be incompatible with life. The efficient reabsorption mechanism of the renal tubules allows for over 99% of this sodium to be reabsorbed, leading to only about 100 mmol per day sodium excretion [1, 2]. Additional examples of the reabsorption efficiency of the kidney are listed in Table 1.1.

The proximal convoluted tubule is the workhorse of the kidney and reabsorbs more solute and water than any other segment of the nephron. Approximately 55–65% of the total ultrafiltrate is reabsorbed in this segment. Almost all of the filtered glucose and amino acids are reabsorbed in this segment along with 90% of the bicarbonate, 65% of the sodium and 55% of the chloride. Since both solutes/ions and water are reabsorbed in the proximal convoluted tubule, the ultrafiltrate leaving the proximal tubule is essentially the same osmolality as the ultrafiltrate that entered. Stated another way, urine is neither concentrated nor diluted in this segment [1, 2].

The loop of Henle is composed of a descending limb and an ascending limb. The descending limb is relatively impermeable to solutes but freely permeable to water. The ascending limb is water impermeable; thus here begins the diluting

Table 1.1 Filtration, excretion, and reabsorption of water, electrolytes, and solutes by the kidney in a normal adult

Substance	Amount	Filtered	Excreted	Reabsorbed	% Filtered amount reabsorbed
H_2O	L/day	180	1.5	178.5	99.2
Na^+	mEq/day	25,200	150	25,050	99.4
K^+	mEq/day	720	100	620	86.1
Ca^{++}	mEq/day	540	10	530	98.2
HCO_3^-	mEq/day	4320	2	4,318	99.9+
Cl^-	mEq/day	18,000	150	17,850	99.2
Glucose	mmol/day	800	0	800	100.0
Urea	g/day	56	28	25	50.0

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segment of the nephron because removal of solutes and not water dilutes the ultrafiltrate concentration. The $Na^+K^+2Cl^-$ symporter on the apical membrane of the thick ascending limb allows for 25% of the filtered sodium and chloride to be reabsorbed in this segment of the nephron [1, 2]. This transporter is the site of action for the class of diuretics known as loop diuretics. Hereditary or acquired dysfunction of this transporter results in Bartter syndrome. This disorder has clinical features (hypokalemia, metabolic alkalosis, hypercalciuria) similar to those seen in patients given a loop diuretic [6–8].

The distal tubule is relatively impermeable to water, continuing the diluting segment of the nephron. The Na^+Cl^- symporter on the apical membrane of the distal convoluted tubule allows for an additional 5–10% of filtered sodium and chloride to be reabsorbed in this segment of the nephron [1, 2]. This transporter is the site of action for the class of diuretics known as thiazide diuretics. Hereditary or acquired dysfunction of this transporter results in Gitelman syndrome. This disorder has clinical features (hypokalemia, metabolic alkalosis, hypocalciuria) similar to those seen in patients given a thiazide diuretic [6–8].

The collecting duct fine tunes sodium reabsorption as well as potassium and acid excretion. The epithelial sodium channel (ENaC) on the apical membrane of the principal cell in the collecting duct allows for an additional 1–3% of the filtered sodium load to be reabsorbed. This channel is upregulated by the mineralocorticoid aldosterone [1, 2]. Mineralocorticoid receptor antagonists, such as spironolactone and eplerenone, cause a downregulation of ENaC resulting in sodium diuresis. A hereditary or acquired activating mutation of ENaC results in Liddle syndrome, which is a disorder with clinical characteristics similar to those of a high aldosterone state (excessive sodium reabsorption, hypervolemia, hypertension, hypokalemia) [8–10]. An inactivating mutation has also been described that results in clinical char-

acteristics similar to those of a low aldosterone state (sodium wasting, hypovolemia, hyperkalemia) [11].

The collecting duct is impermeable to water in the absence of antidiuretic hormone (ADH). When ADH is present, the collecting duct becomes permeable to water through the use of aquaporin channels. It is here in the collecting duct that the urine can be further diluted or concentrated depending on the needs of an individual. The major stimuli for ADH secretion are hyperosmolality and effective circulatory volume depletion [1, 2].

Urine Elimination

Urine flows from the collecting duct of the nephron to join a converging system of tubules with other collecting ducts. These ducts then join together to form the minor calyces followed by the major calyces that ultimately converge in the renal pelvis (Fig. 1.3). Urine continues to flow from the renal pelvis into the ureter, transporting urine into the urinary bladder. Urine from both kidneys is stored in the bladder until the process of micturition (urination) occurs.

The first urge to void is felt at a bladder volume of approximately 150 milliliters (mL). A marked sense of fullness is felt at about 400 mL and bladder capacity is approximately 500 mL [12]. Urine is normally excreted voluntarily from the body by flowing through the urethra away from the bladder.

Average daily urine production for adults is usually between 1–2 liters (L) depending on the state of hydration, activity level, and health of the individual. Producing too much or too little urine requires medical attention. Polyuria (see Chap. 15) is a condition of excessive urine production (more than 3 L/day). Oliguria is the production of less than 400–500 mL/day and anuria is the production of less than 50–100 mL/day.

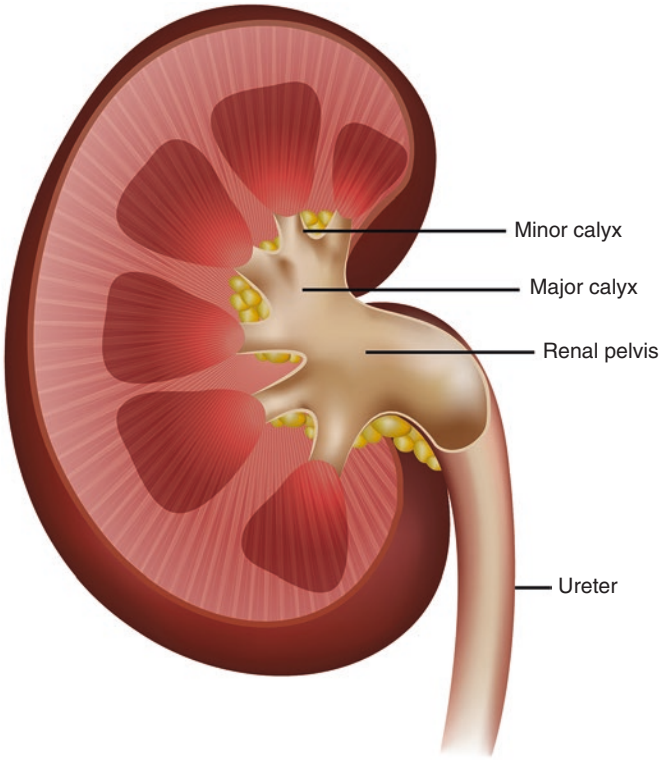

FIGURE 1.3 Anatomy for urine flow through the kidney. (Courtesy of Teresa Ruggle, University of Iowa.)

Summary

Normal urinary system anatomy consists of two kidneys, two ureters, one urinary bladder and one urethra. The urinary system is responsible for urine production, refinement and elimination. The final composition and amount of urine is determined by the specialized actions of glomerular filtration followed by tubular reabsorption and secretion. Urine can be collected from a patient and its contents provide additional clinical information to the provider. This information may be obtained by a urinalysis using a dipstick to determine chemical composition; microscopic analysis to look for cells, casts or

crystals; cultures to assist with infection diagnosis; or specialized tests to assess for cancer, electrolyte abnormalities or substance abuse. Whatever the test, analyzing this golden elixir of life provides a valuable, noninvasive means to assist with patient care.

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Chapter 2
Follow the Money: Costs, Reimbursement and Regulations of Urine Based Testing

Matthew A. Uhlman, Victoria J. A. Sharp, Nora Kopping, and Mark S. Uhlman

Objectives

- Gain an understanding of costs and reimbursement issues related to urine studies
- Recognize regulatory requirements related to obtaining and maintaining accreditation to perform urine-based testing
- Differentiate which urine-based tests are preferable to other testing modalities from financial and quality perspectives

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Overview

Urine-based tests are a critical and increasingly utilized part of the investigative workup for many diseases. As such tests continue to evolve, urine-based diagnostics will increasingly represent a relatively painless and easy-to-collect tool for healthcare providers. While many urine-based tests are available, reimbursement for them may not be, and for others, cost benefit ratios are unacceptable. An understanding of the associated costs, reimbursement and regulatory aspects of such tests is paramount in their successful deployment and utilization. In this chapter we will highlight key financial and regulatory considerations for the clinician considering the use of the urine-based tests currently available.

Costs

Urine-based testing has been a part of medical care since the time of the Sumerian and Babylonian empires [1] and while similarities still exist in urine-based diagnostics, financial considerations are (assumedly) far more complex today. Most clinicians and providers are familiar with urine-based testing in some respect, but far fewer are versed in the financial and regulatory aspects of such tests. Whether the testing is done under a microscope at a provider's office or delivered overnight for analysis at a reference lab, an understanding of the many different considerations surrounding such tests is increasingly important.

In the United States each year, nearly 1 billion physician office visits occur, with over 500 million visits to primary care offices [2, 3]. While only a fraction of patients undergo a urine-based diagnostic test, the number of studies and accrued costs quickly become staggering. In urine-centric specialties such as urology or nephrology, the percentage of patients requiring testing and the number of potential urine-based tests increases drastically.

In 2015, urinary tract infections accounted for over 10.5 million office and 2–3 million ER visits [4, 5] at an estimated cost of at least $3.5 billion [4], and in 2014, urine-based screening tests (drugs of abuse) were estimated to cost in excess of $8.5 billion [6]. Such numbers increase each year, and while a myriad of urine-based tests exist currently, increasing numbers are coming onto the market. While such tests are minimally-invasive and more convenient than many current options, many are expensive. Being aware of the financial implications of both routine and specialized urine-based testing will continue to be important going forward, especially as patients are responsible for an increasing percentage of their overall healthcare spending.

Laboratory testing in the United States is a market and understanding the financial implications of urine testing requires knowledge of the market landscape. In general, three types of laboratories exist: laboratories owned by hospitals or practices, hospital outreach laboratories, and national commercial laboratories. Traditionally, laboratories were owned and operated by physician practices and hospitals. The costs of operating these types of laboratories tend to be higher because of lower volumes of testing. To increase volumes, many such laboratories do outreach testing for other providers, better leveraging economies of scale. There is also an independent national commercial laboratory market, half of which is dominated by two laboratories: LabCorp and Quest Diagnostics. Laboratories operating on a national scale receive the highest test volumes and therefore are able to offer competitive pricing. While Medicare has shifted toward a single national fee schedule for reimbursement in recent years, private payers tend to reimburse for independent laboratory testing at lower rates than hospital outreach laboratories, which in turn are reimbursed at lower rates than physician office laboratories [7].

Urine-based tests run the gamut from budget- friendly to incredibly expensive, depending on the type of test, the setting in which the test is performed, and the reason the test is

Table 2.1 Cost of urine-based tests

Test	Cost
Urine pregnancy test (store bought)	$
Urinalysis – dipstick	$
Urinalysis – complete microscopy	$$
Urine sodium, creatinine, urea nitrogen	$$
Urine pregnancy test (lab)	$$
NMP22 bladderChek – urine bladder cancer test	$$
24-hour urine – protein	$$
Urine culture	$$
Urine drug screen	$$$
Urine catecholamines	$$$
Urine cytology	$$$$
CxBladder – urine bladder cancer test	$$$$
24-hour urine – Litholink	$$$$
SelectMDX – urine prostate cancer test	$$$$$
Urovysion – urine bladder cancer test	$$$$$

Key: $ <$10, $$ $10–50, $$$ $50–100, $$$$ $100–500, $$$$$ >$500.

ordered (Table 2.1). Laboratory testing ordered as part of preventive care visits is generally covered in full by insurance, but insurance coverage for diagnostic tests varies depending on a patient's individual insurance plan. With the patient sharing an increasing portion of the cost of care, knowing not only the cost of tests, but also if there are related diagnostic fees is important; such fees may also vary, depending on how a test is performed. Examples include urine microscopy which may be performed during an office visit by the clinician, or sent to an outside lab or done at a hospital where a

microbiologist performs the analysis. Urine cytology is another highly variable test; physicians may work at a facility with or without urologic pathologists who read such tests. In the case where one is not present, they may be sent out to a regional center. Any situation that involves sending the test to an outside lab, physician, or medical center can result in additional fees. All such aspects of testing can have serious financial implications for patients and potentially for providers.

Regardless of where testing is performed, there are circumstances in which providers and patients alike may be surprised to find that testing is not reimbursable or is reimbursed at low rates. The form of payment varies by the setting of testing and patient diagnosis. Inpatient care is reimbursed with a bundled payment and depending on diagnosis, some testing is included with these bundles. In cases where testing is billed separately, payers may apply different reimbursement arrangements. Similarly, when test panels are ordered, payers may elect not to reimburse for individual tests on the panel. Both situations result in higher total cost of care for the patient, and may result in additional costs to the patient. It is also important to note that not all insurance plans cover all diagnostic tests. While some tests are covered by large plans (Medicare, Medicaid), other insurances still consider them "investigational," leaving the patient to cover the entire cost. Some companies have become more patient/customer savvy by offering significant discounts for early payment of diagnostic tests. Having a working knowledge of such programs can ultimately save patients significant money as well as give them a realistic expectation of their possible financial responsibilities. To avoid patient frustration and confusion, prior to undergoing such tests many clinics require patients to read and sign a form outlining the potential associated patient obligation. An example exists with the urine-based SelectMDX (MDxHealth, Irvine, CA) test, a prostate cancer screening diagnostic (see Chap. 14); patients may be respon-

sible for up to $350, with a reduced cost of $200 if the balance is paid within 30 days. Regardless of the circumstance, it behooves the provider to be aware of all costs associated with urine testing.

As payment models continue to evolve (i.e., bundled payments), it will become all the more important that providers understand the costs of tests. The Centers for Medicare and Medicaid Services (CMS) has developed payment models in which the provider assumes financial risk for patient outcomes, including accountable care organizations, bundled payments for episodes of care and capitated payments for primary care, and has hinted that some of these models may become mandatory. To practice successfully in this new payment environment, providers will need to consider lower cost alternatives in urine testing [8].

In many instances, urine-based tests provide the most cost-effective option for patients and clinics alike, and may be available for use at home. Tests such as home use urinalysis (Dipstick) which can allow for early detection of urinary tract infections (see Chap. 6), are no different than those used in a clinic and cost significantly less to the patient (<$1 at home vs. $30+ in a walk-in clinic). Urine-based pregnancy tests can cost less than $1, while blood-based tests are significantly more expensive, are invasive, and may not increase detection rates except during very early stages of pregnancy (see Chap. 3). Conversely, a urine-based HIV test is available that offers consumers a convenient, non-invasive testing option, but comes with an unacceptable tradeoff of decreased accuracy. Considerations of tradeoffs in accuracy, precision, sensitivity and specificity must be made when considering a urine-based testing modality and such knowledge is paramount to ensure adequate patient care. See Table 2.2 for a list of urine-based tests with serum or urethral swab-based options. Vendors for various testing modalities can provide this information, but providers should corroborate these claims by review of relevant literature.

Table 2.2 Tests that have urine-based options available

Blood tests
Human papilloma virus (HPV)
Human immunodeficiency virus (HIV)
Tuberculosis
Pregnancy testing
Down syndrome screening
Drug screening
Catecholamines/metanephrines
H. pylori
Glucose
Phenylalanine
Urethral swab tests
Chlamydia
Gonorrhea

A growing trend in health care is provider alignment with large organizations; independent practices are becoming increasingly rare. Providers aligned with large organizations can make use of administrative resources to understand their own practice patterns across their entire panel of patients. Data analytics teams can create reports that show how often specific tests are ordered or review claims data to identify frequent ordering of costly tests. Supply chain teams can conduct value analysis assessments to compare collection methods on cost and quality and can assist with piloting new collection supplies. Some providers may be surprised to learn that they order expensive tests at higher rates than their peers with like practices, or that their go-to collection method can be done with clinically equivalent but lower cost supplies.

Reimbursement

Providers must not only be familiar with the cost of tests, but also the reimbursement of such tests. The vast majority of urine-based tests are considered diagnostics and as such, are generally not billable by providers. A common example is the urinalysis which is often billed under the Current Procedural Terminology (CPT) code 81000 but does not result in any work relative value unit (RVU) compensation for the provider interpreting it. The test itself is billed and ultimately a clinic or hospital is reimbursed for it, meaning it can be a source of income (albeit modest at best).

The level of reimbursement can also vary significantly based on where it is performed (hospital, doctor's office, walk in clinic, etc.). Walk in clinics routinely charge $20–40 for a urinalysis (UA), while hospitals may charge in excess of $100, both of which may be billed to the patient or insurance. Meanwhile the average reimbursement is often much lower (Washington (WA) Medicare – $4.02, WA Medicaid – $3.62, Iowa (IA) Medicaid – $3.82). Other common tests (coding information seen in Table 2.3), such as a urine culture, urine pregnancy test and drug screens (see Chap. 16), generally result in no payment to the provider, but rather for the clinic or hospital. Reimbursement for such tests is determined on a contractual basis and while average reimbursement varies with where a patient is seen, the majority of urine-based tests are modest in price, generally costing between $10 and $100. However, specialty tests may cost over $1000 (Table 2.1). The Medicare fee schedule, which lists Medicare reimbursement rates for clinical laboratory tests identifiable by Healthcare Common Procedure Coding System (HCPCS) codes is available for download from CMS.gov [9]. Medicaid reimbursement varies by state. For example, a urine pregnancy test (procedure code 81025) performed at an independent lab is reimbursed by Iowa Medicaid at $8.18 (Table 2.3), Ohio Medicaid at $6.46 and Wisconsin Medicaid at $8.61. When performed at a outpatient clinic the same test is reimbursed by Iowa Medicaid at $8.97, Ohio Medicaid at $8.77 and by

Table 2.3 Common urine-based testing CPT codes

CPT code	Explanation	Medicare fee schedule [9]	IOWA Medicaid fee schedule (independent lab) [13]
81000	UA, with dipstick, non-automated, with microscopy	$4.02	$3.82
87077	Culture, bacterial; aerobic isolate, additional methods required for definitive identification, each isolate	$8.97	$9.47
87086	Culture, bacterial; quantitative colony count, urine	$8.97	$9.46
87088	Culture, bacterial; with isolation and presumptive identification of each isolate, urine	$8.99	$9.49
81025	Urine pregnancy test, by visual color comparison methods	$8.61	$8.18
80305	Drug Tests(s), presumptive, any number of drug classes; any number of devices or procedures, capable of being read by direct optical observation	$12.60	$12.79
80306	Drug Tests(s), presumptive, any number of drug classes; any number of devices or procedures, read by instrument-assisted direct optical observation	$17.14	$17.06

Wisconsin Medicaid at $8.90. California Medicaid reimbursement rate is $2.80 across the board with no adjustment in rate for differences in testing location [10–14].

Given that many urine tests are relatively inexpensive and covered in full by payers, providers may not be aware of cost-effectiveness considerations. New payment models require providers to think in terms of the health of their panel population; when extrapolated out to one's full panel, the cost of urine testing can add up.

Regulations

As with many aspects of medicine, many clinicians are blissfully unaware of the intricacies and requirements for processing something as simple as a urine culture. The Clinical Laboratory Improvement Amendments (CLIA) [15] are a set of standards that regulate laboratory testing and require such facilities to be certified by their state and CMS. CLIA ultimately dictates what tests can be performed in a given laboratory, though many tests are considered exempt (the urinalysis being one of them). Many larger organizations maintain broad CLIA certifications, including some that serve as regional centers, while smaller clinics understandably limit the breadth of their services to control costs.

Many urine-based analyses are performed by outside companies, either due to proprietary technology, specialist availability, or due to the small number that would be performed at an institution. From the standpoints of provider planning, patient satisfaction, and simple logistics (i.e. can a lab be done on a Friday?), knowing what happens after a test is ordered is fundamental to proper utilization.

Additionally, as the medical community continues to seek less invasive and more accurate testing options, urine-based testing will undoubtedly increase in popularity. Over the past decade, molecular tests have become more widespread and are now being used for cancer screening, diagnostic stratification and treatment follow up (see Chap. 14). Numerous other

urine tests for communicable diseases such as chlamydia, gonorrhea, human immunodeficiency virus (HIV), human papilloma virus (HPV) and tuberculosis (TB) are also available.

Given the rapid expansion of urine-based diagnostics, it behooves all healthcare providers to have an evolving knowledge of available and cost-effective tests. A familiarity of the advantages and disadvantages of such tests is equally important. As an example, in the early 2000's, a urine-based test for H. pylori showed promise; however, a breath test proved to be more accurate and easier to administer [16].

Additional Considerations

The costs associated with performing urine-based testing are important to consider when determining if offering such tests is worthwhile within a facility. At first glance, the costs related to microscopy seem limited to the purchase of the unit plus supplies, however it quickly becomes *much* more complex. Costs include the initial microscope purchase, replacement bulbs, slides/covers, gloves, a centrifuge, disposal containers (and the associated disposal costs), periodic cleaning, and lens replacement and calibration every 6 months (CLIA requirement). Furthermore, clinics must apply for and obtain a provider performed microscopy (PPM) certificate, renew the certificate every 2 years, appoint a qualified lab director, provide and maintain procedural manuals, retain the last 2 years of testing and maintenance records, and administer proficiency testing at least twice per year to all providers [17]. For labs offering testing more complex than a simple urinalysis, the requirements grow in both quantity and complexity.

While simple at the core, offering urine-based testing within a clinic or hospital setting should be evaluated with a validated business plan prior to implementation. Providers should also understand issues related to contracting with outside labs. Contracts can be advantageous if they lock in lower costs for testing, but there may be quality or timeliness con-

cerns with outside laboratories. Laboratory contracts should be reviewed carefully to ensure they achieve access to the highest quality, lowest cost tests possible. Another consideration is having to send specimens to certain designated laboratories based on contracts by insurers. If as a provider you want to send to a different laboratory based on experience, this would require an appeal with the insurer which may take a lot of time and may still get denied.

Future Directions

With the rising popularity of telehealth and remote health monitoring, urine-based testing will remain an important diagnostic tool for all healthcare providers. In rural settings, and for patients in isolated locations, remote evaluation and monitoring gives them access to healthcare that would otherwise be inaccessible. Examining a urine sample via a smart phone over a secure video connection gives the provider important information with the same accuracy as if the test were performed during a face to face visit.

In addition, with continued improvements and increasing speed in point-of-care diagnostics, and with the availability of such technology to the general public, patients may soon be able to perform some tests at home, followed by interpretation by a physician via telehealth applications. The ability to accurately and objectively diagnose ailments via telehealth applications has the potential to increase efficiency and satisfaction for providers and patients alike.

Summary

Urine-based tests are an integral part of the diagnostic armamentarium of healthcare providers and we hope that this chapter has helped inform you of some of the financial aspects of the subject. With the number of available tests continuing to grow, it is imperative that providers understand

the potential benefits and limitations of such tests, both in terms of disease diagnosis and finances. As the focus on the cost and value of healthcare continues to intensify, it will become increasingly important for providers to understand the financial implications of ordering (and processing) urine-based tests.

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Chapter 3
Going with the Flow: Proper Urine Testing Methods for Clinical Practice

Gina M. Lockwood and Victoria J. A. Sharp

Objectives

- Discuss proper methods of collecting urine for different urine tests
- Understand how both urine dipstick and urine microscopy are performed in the office or laboratory
- Incorporate best practice guidelines for appropriate urine testing in clinical practice
- Identify special urine testing available to clinicians, including pregnancy/ovulation testing, testing for urinary calculi, and cytopathologic examination

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Overview

This book details specific urine tests obtained in clinical practice, their indications and clinical implications. However, before a provider orders urine testing, he or she must understand appropriate methods of urine collection for different situations, as well as how urine testing is performed. With this knowledge, false negative and false positive results can be more easily identified, and testing can be optimized for efficiency and cost to the patient and provider. In addition to basic urine dipstick and urine microscopy testing, multiple other uses for urine testing will be discussed throughout this book. In this chapter pregnancy and ovulation testing, urinary calculi, metabolic testing, and cytopathologic examination of cells in the urine will be discussed.

Case 1: Urine Specimen Collection

A 40 year old diabetic female complains of 48 hours of painful urination as well as blood in the urine. She has associated intermittent suprapubic pain and right flank pain. She has a history of recurrent urinary tract infections as well as a history of nephrolithiasis. On physical examination, she is obese. She has mild suprapubic tenderness to palpation and mild right costovertebral angle tenderness. What logistical considerations should be made when obtaining a urine sample from this patient?

All urine specimens must be collected in clean, dry, leakproof containers. Disposable containers are preferred to eliminate the chance of contamination due to improper washing. Individually packaged sterile containers with secure closures should be used for microbiological studies [1]. Routine urinalysis protocols usually require 10–25 mL of urine. Volumes of <12 mL can sometimes hinder the performance of microscopic examination [2]. As a general rule, urine specimens should be examined within 2 hours of collec-

tion. If transportation to the laboratory or office will be longer than 2 hours, precautions should be taken to preserve specimen integrity. Unpreserved urine can cause both false positive and false negative chemical and microscopic results. No single preservative is suitable for every urine test, thus the most easy and common preservative method is refrigeration at 4–6 °C. Refrigeration prevents bacterial proliferation and yields a specimen that is suitable for culture for up to 24 hours [2].

Methods of Urine Collection

The method of urine collection depends on the parameters being tested. For example, if urine is being tested only for protein, sterile technique is not as vital as with urine being collected to diagnose urinary tract infection. Special considerations for urine collection in pediatric patients will be discussed in Chap. 13.

Random (Routine) Specimen

The most common type of specimen for routine testing or screening because of ease of collection and convenience is a one-time "spot" urine collection. Care should be taken to avoid inaccurate results because of recent dietary intake (beets causing red urine), physical activity (exercise-induced hematuria or proteinuria), or hydration causing urinary dilution. A routine void requires no patient preparation, and it is simply collected by having the patient urinate into an appropriate container.

First Morning Specimen

This one-time urine collection is ideal for urine screening, especially in the prevention of false-negative pregnancy tests and for evaluation of proteinuria that is suspected to be orthostatic in nature. This specimen is concentrated and thus

ensures detection of chemicals or formed elements that may not be recognized in more dilute urine [1]. It is also ideal for urine parameters affected by increased physical activity. However, it can be inconvenient because it may require the patient to pick up and drop off the container at separate visits.

Timed Collection

A 12 or 24-hour urine collection is obtained when a quantitative measurement of a urine substance is desired. Some solutes exhibit diurnal variation in urine excretion, like urine catecholamines and 17-hydroxysteroids. When the concentration of the substance to be measured changes diurnally or with daily activities like activity, food intake, and body metabolism, a 24-hour urine collection may be necessary, especially if exact quantification is needed. Some examples of urine testing that can be obtained by 24-hour collection include urine creatinine, protein, cortisol, aldosterone, metanephrines, urine electrolytes, and heavy metals. Quantitative measurement of some substances secreted in the urine can rule in or out a diagnosis (like metanephrines for pheochromocytoma).

Collection of urine over a 24-hour period has been considered the "gold standard" for measurement of creatinine clearance and urine protein excretion. Unfortunately, this collection is time consuming and inconvenient for the patient. More importantly, it can be inaccurate, both because of over- and under-collection. One large retrospective chart review of pregnant women with a hypertensive disorder found 24-hour urine collection to be inaccurate for measurement of proteinuria and creatinine clearance [3]. In non-pregnant patients, it is generally accepted that prediction equations are generally better predictors of glomerular filtration rate than 24-hour creatinine clearance due to suboptimal collection techniques [4]. The same holds true for spot urine protein/creatinine compared to 24-hour urine protein excretion [5].

However, 24-hour measurement may be needed in special situations, as in patients with significant abnormalities in muscle mass.

If 24-hour urine collection is to be obtained, the patient must be provided detailed instructions as to the procedure of collection, and the patient should be provided a urine collection container. Depending on the urine tests obtained, chemical preservatives may be added to the urine container, and patients should be notified not to empty this additive. If an acid preservative is used, a warning label should be placed on the container, as the acid can cause chemical burns. In some cases, simple refrigeration may be the only preservation method necessary. The collection should begin and end with an empty bladder. For example, at 7 AM on the morning of day one, the patient voids and discards the urine specimen, and records the time. All voids for the next 24 hours are collected including a final void at the same time of the void in which urine was discarded the day prior (7 AM, in this situation). Addition of urine before the start of collection will falsely elevate levels, and failure to include the end void will falsely decrease levels.

Midstream "Clean-Catch" Specimen

When obtaining urine for routine urinalysis and/or urine culture, this type of collection is used in situations in which contamination (e.g., from vaginal discharge or skin flora) could alter results or in which culture is desired. Contamination of a clean-catch specimen is common, especially in the obese, children, and those with limited manual dexterity. Strong antibacterial agents are not recommended for pre-collection cleansing, but gentle cleansing agents are sometimes recommended, although the effect of use of antibacterial cleansing wipes on contamination rates is debated. Figure 3.1 shows patient instructions for proper collection of midstream urine [1, 15].

Female	Male
1. Wash hands with soap and water. 2. Remove lid from sterile container without touching the inside of the lid or container. 3. Separate the genital skin folds (labia). 4. Use one side of the cleaning towelette to wipe the inner folds of the labia from front to back.* 5. Use a second towelette or other side of the towelette to wipe the the urethra where the urine comes out, just above the opening to the vagina.* 6. Begin to urinate into the toilet. 7. Once you have begun to urinate, bring the container into the stream to collect an adequate amount of urine (about 1/2 of the container). Do not touch the inside of the container with your hands or allow the container to touch the genital area. 8. Finish urinating into the toilet. 9. Place the lid tightly over the specimen container touching only the outside of the lid.	1. Wash hands with soap and water. 2. Remove lid from sterile container without touching the inside of the lid or container. 3. Cleanse the head of the penis with a cleaning towelette.* If you are uncircumcised, pull back the foreskin first and keep it pulled back (retracted). 4. Begin to urinate into the toilet. 5. Once you have begun to urinate, bring the container into the stream to collect an adequate amount of urine (about 1/2 of the container). Do not touch the inside of the container with your hands or allow the container to touch the genital area. 6. Finish urinating into the toilet. If you are uncircumcised, replace the foreskin back over the head of your penis. 7. Place the lid tightly over the specimen container touching only the outside of the lid.

*The effects of using cleansing towelettes on rates of urine specimen contamination are debated in adults and children. Some providers do not recommend their use.

FIGURE 3.1 Patient instructions for obtaining midstream clean-catch urine specimens in adults

Catheterized Specimen

In patients in whom an indwelling catheter is present for the short-term, urine should ideally be collected with initial catheter insertion. If a catheter is already in place, it can be obtained from the catheter tubing port with a luer lock syringe using aseptic technique (Fig. 3.2) [6]. In the case of a long-term indwelling catheter, a new catheter should be placed prior to obtaining urine for culture, if possible [7]. It must be remembered that any patient with an indwelling

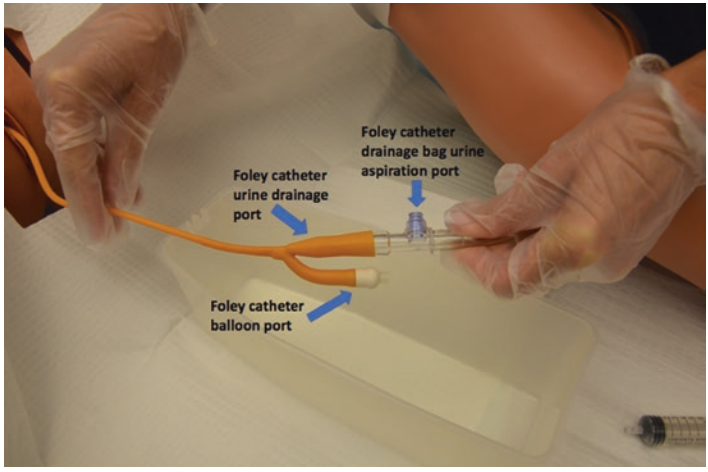


FIGURE 3.2 Foley catheter and drainage bag tubing. Aspiration port allows for luer lock syringe drainage of urine without disruption of closed system. Figure included with adaptation from [17], under the Creative Commons Attribution 4.0 International Public License (https://creativecommons.org/licenses/by/4.0/legalcode)

catheter may have bacterial colonization. Culture specimens should never be obtained from urinary drainage bags. Straight catheter urine specimens may also be required in those who do not void volitionally (infants, elderly, and patients with neurogenic bladder).

Suprapubic Aspirate

This type of collection is sometimes required to obtain a sterile specimen. Because of invasiveness, it is not commonly used, but the most common use is in infants.

Ordering of Urine Tests

Providers may order urine tests individually or as a panel. Tests may be ordered and performed during primary or specialty clinic visits or for patients during an inpatient hospital stay. Some

patients require frequent repeat testing (e.g. every day during a hospital stay) or on a less frequent basis for disease monitoring (e.g. twice per year). Various non-clinical parameters can influence what types of tests are ordered, and how frequently. Providers may feel pressure to order tests that would not impact diagnosis as a defensive mechanism or to show patients they are receiving every possible health care service. Others may order tests with equivalent diagnostic values because they are new and state-of-the-art. When tests are clinically equivalent, some providers prefer to order the test that requires less provider time, or a test that avoids a lengthy prior authorization process. The financial implications of such decisions for patients and the health system are discussed in greater detail in Chap. 2.

Returning to case 1, this patient should be instructed on proper clean-catch collection technique, given risk factors for urine contamination (obesity). Alternatively, a catheterized urine specimen could be obtained.

Case 2: Urine Dipstick Versus Urine Microscopy

An 85 year old female who is incontinent of urine has had pink-tinged urine for 1 week. She has no burning with urination, urinary urgency, or fevers. She has multiple medical comorbidities and takes 13 medications daily. She has a 45 pack-year smoking history. Her abdominal, back and genitourinary exams are all unremarkable. What urine testing would be more appropriate as a next step: urine dipstick or microscopic urinalysis?

Methods of Urine Testing

The two most common types of urine testing performed in inpatient and outpatient settings are chemical examination by reagent strip (dipstick) and sediment examination by microscopy. The latter simply adds a microscopic examination of urine sediment to the dipstick test.

Table 3.1 Components of urine reagent strip and microscopic urinalysis with examples of normal values

Gross/physical examination	**Normal values**
Color	Pale/dark yellow
Appearance (Clarity)	Clear
Odor	Faint aromatic
Chemical examination/reagent strip	
pH	4.5–8
Specific gravity	1.007–1.030
Blood (Hemoglobin/myoglobin)	Negative
Glucose	Negative
Ketone	Negative
Protein	Negative
Bilirubin	Negative
Urobilinogen	0.2–1.0 mg/dL
Leukocyte esterase	Negative
Nitrite	Negative
Sediment/microscopic examination	
White blood cells (WBC)	0–5/hpf
Red blood cells (RBC)	0–2/hpf
Epithelial (squamous) cells	≤10/lpf
Casts	0/hpf
Bacteria	0/hpf
Crystals	<5/hpf
Yeast	0/hpf
Parasites	0/hpf
Mucus	None, rare
Spermatozoa	0/hpf

(Normal values obtained from multiple sources)
Note individual laboratories will have distinct normal values.
hpf high power field, *lpf* low power field

Methods for Chemical Screening

Reagent strips, or dipsticks, are the primary tools used for chemical examination of urine. Generally they prove to act as good screening tests, with high sensitivity. They allow for quick, easy, and inexpensive processing by personnel in whom minimal training is required; some are even designed to be performed at home. Despite their ease of use, multiple complex chemical reactions are facilitated by this test. Dipsticks consist of chemical-impregnated absorbent pads adherent to a plastic strip. When this pad comes into contact with urine, a color-producing chemical reaction occurs. The color produced on the pad is compared to a manufacturer's chart in a specified time frame (Figs. 3.3 and 3.4). A semi-quantitative value of trace, 1+, 2+, 3+ or 4+ can be reported, and an estimate of mg/dL is available for calculation for some parameters [1]. Table 3.2 shows practical recommendations for care and use of reagent strips in the office or laboratory, given that untrained office personnel will need specific instruction in their use [8]. Although traditionally dipstick evaluation is performed by manual visualization, semi-automated and fully-automated instruments are available to read the chemical reactions by reflectance. Automated

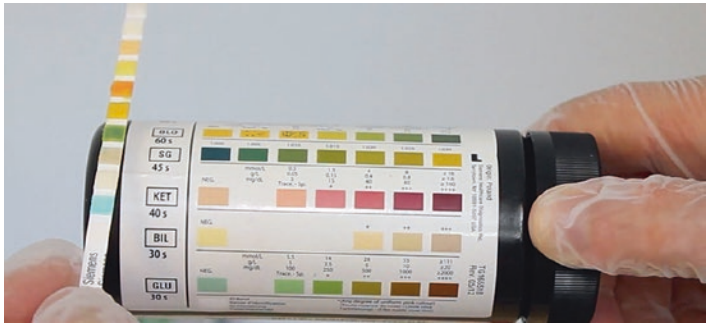


Figure 3.3 Urine reagent strip and manufacturer's chart. Used with permission from Siemens Healthcare Diagnostics Inc.

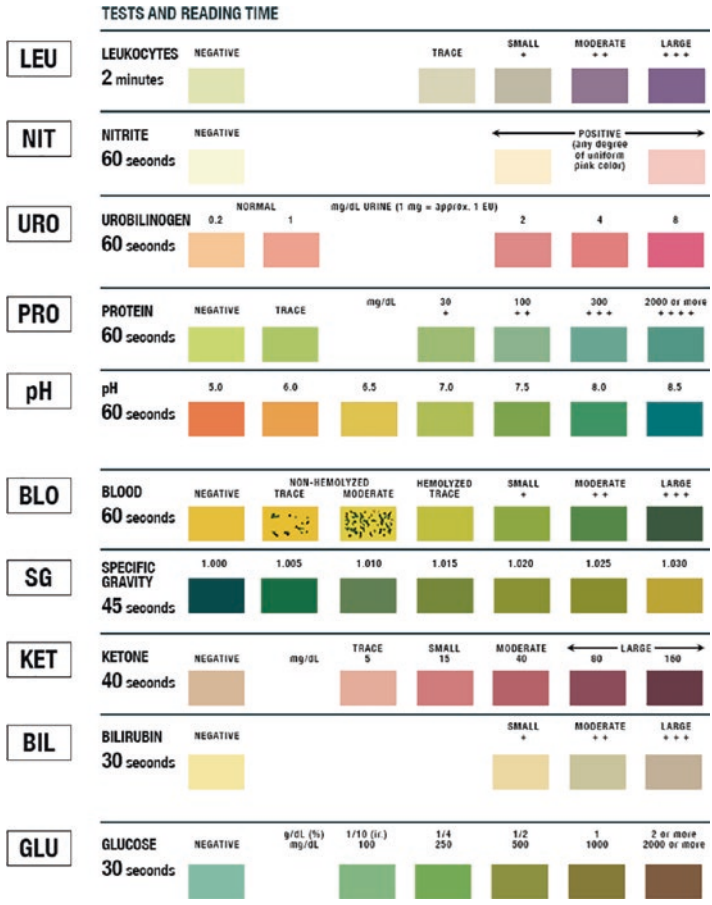


FIGURE 3.4 Sample color chart for urine dipstick testing. Used with permission from Siemens Healthcare Diagnostics Inc. This is for educational purposes only and not intended for use to interpret test results. The colors as they appear may not be the exact color on the official product labeling

instrumentation is discussed in detail later in this chapter. Chapters 4, 5, 6, and 7 address specific aspects of urine dipstick testing and their clinical applications.

Table 3.2 Recommendations for handling and use of urine reagent strips

Storage	Testing/use
• Protect from excessive moisture and heat. • Store in a cool, dry area (not refrigerator). • Check for discoloration that may indicate loss of reactivity with each use. • Do not use discolored strips. • Keep container tightly sealed. • Check manufacturer's directions with each new lot number for changes in procedure.	• Test urine as soon as possible after receiving specimen. • Remove enough strips only for immediate use. • Test a well-mixed, unspun urine sample only at room temperature. • Do not touch test area with fingers. • Do not use reagent strips in presence of volatile acids or alkaline fumes. • Dip strip into urine briefly, no longer than 1 second. • Drain excess urine; run edges of strip along rim of tube, or blot edge on absorbent paper. • Do not allow reagents to run together. • Do not lay strip directly on workbench surface. • Follow timing recommendations from manufacturer for each chemical test. • Hold strip close to color chart and read under good lighting. • Know sources of error, sensitivity and specificity for each test.

Methods for Examination of Urine Sediment (Microscopy)

Cellular and noncellular elements of the urine that do not yield distinct chemical reactions can be detected by urine microscopy. Microscopy can serve as a confirmatory test when urine dipstick is abnormal [8, 9], but it also recognizes solutes in the urine not seen on dipstick in up to 66% of tests [10]. Chapters 8, 9, 10, 11, and 12 address specific microscopic urine tests and their clinical applications. Centrifuged urine sediment can be examined for these elements using different types of urine microscopy (Table 3.3) [1]. Accurate urine microscopy requires the expertise of trained personnel with the ability to identify and differentiate cells found in urine sediment. It is also helpful if the microscopist understands the clinical relevance of findings and possible inconsistencies with chemical examination. Table 3.4 gives an example of laboratory preparation of a urine sample for microscopy. In general, random urine collection is adequate for microscopy, but examination is recommended when the sample is fresh; cells and casts begin to lyse within 2 hours of collection.

Bright-field microscopy is the most common type performed for urinalysis, but other types of microscopy may be useful in examining specific aspects of urine sediment (Table 3.3). The type of microscopy used depends on the specimen type, refractive index of the object, and ability to image unstained living cells (Figs. 3.5 and 3.6). Microscopic examination should be performed in a consistent way, but techniques vary from laboratory to laboratory.

Table 3.3 Types of microscopy used in urine testing

Technique	Functions
Bright-field microscopy	Routine urine analysis
Phase-contrast microscopy	Enhances visualization of elements with low refractive indices (hyaline casts, mixed cellular casts, mucous threads, *Trichomonas,* dysmorphic red blood cells)
Polarizing microscopy	Aids in identification of cholesterol in oval fat bodies, fatty casts and various crystals
Fluorescence microscopy	Allows visualization of naturally fluorescent micro-organisms or those stained by fluorescent dye
Interference contrast microscopy	Produces three-dimensional microscopy image and layer-by-layer imaging of cellular elements

Automated Urinalysis

Several automated instruments are available that standardize sample processing, analyze reagent strips, analyze sediment, and report results with consistent quality. In addition to reducing subjectivity, human error, and time, automation can interface with computers for results, flag abnormal results, store patient and control results, and minimize calibration, cleaning and maintenance [1]. Semi-automated machines interpret reagent strips but require an operator to dip the strip into urine, place the strip on a platform, and press a button to analyze (Fig. 3.7). The machine prints out the results. These machines are satisfactory for small volume laboratories and physicians' offices. Fully automated chemistry analyzers are also available in which the machine aspirates and analyzes a urine sample. Automated microscopy is especially useful and cost-effective, as interrater reliability of manual microscopy is poor. Combining automated urine chemistry

Table 3.4 Example of standardization for preparation of samples for urine microscopy

Written instructions to patients for urine collection
Collection in disposable containers of the second urine of the morning after discarding the first few milliliters of urine (midstream technique)
Sample handling and analysis within 2–3 hours from collection
Removal by suction of 9.5 mL of supernatant urine
Gentle, but thorough, resuspension with a pipette of the sediment in the remaining 0.5 mL of urine
Transfer by pipette of 50 μL of resuspended urine to a slide
Covering sample with a 24 × 32-mm coverslip
Examination of all samples by a phase contrast microscope at original magnifications ×160 and ×400
Use of polarized light to identify doubtful lipids and crystals
Match of microscopic findings with dipstick for pH, specific gravity, hemoglobin, leukocyte esterase, and albumin
For routine work, cells expressed as lowest to highest number seen/high-power field, casts as number/low-power field, all the other elements on a scale from 0 to ++++
For scientific work, cells expressed as total number counted on 20 high-power fields

Note: This method is used in the authors' laboratory
Reprinted from [19] with permission from Elsevier

analysis and cell analysis has further increased efficiency in large laboratories. See Chap. 2 for further discussion of cost implications of different types of laboratory analysis.

Returning to case 2, urine dipstick would be an appropriate screening for blood in the urine given its pink-tinge. However, if there are any abnormalities, these need to be confirmed on urine microscopy, with standard bright-field microscopy. This is especially true given the woman's significant smoking history and thus increased risk for genitourinary malignancy.

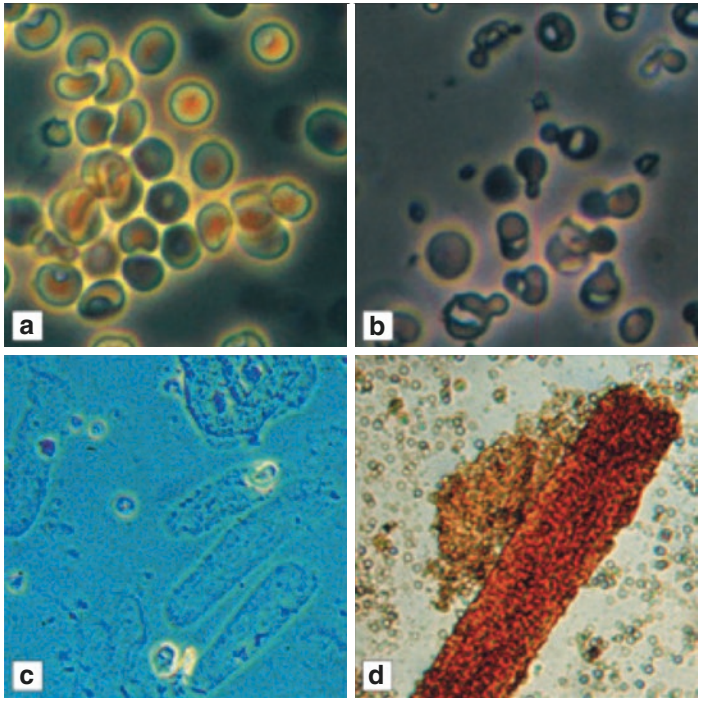


FIGURE 3.5 Urine microscopy. (**a**) Phase-contrast image of erythrocytes from lower urinary tract bleeding (×400). (**b**) Phase-contrast image of dysmorphic erythrocytes from glomerular inflammation (×400), as seen in glomerulonephritis. (**c**) Hyaline casts on phase-contrast microscopy as seen with solidification of Tamm-Horsfall mucoprotein. (**d**) Erythrocytes and a red cell cast in glomerulonephritis (×100) on bright field microscopy. Reprinted from [17] with permission by Elsevier. (A, B) Courtesy of Dr G.M. Iadorola and Dr F. Quarello, B. Bosco Hospital, Turin (from www.sin-italia.org/imago/sediment/sed.htm)

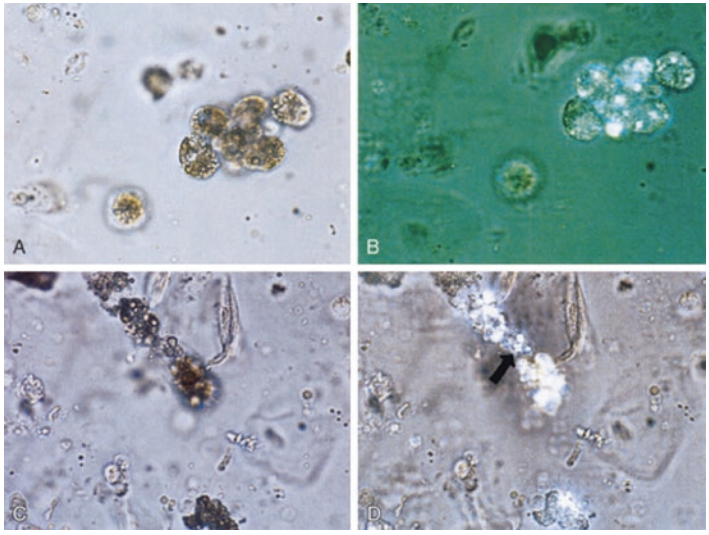


FIGURE 3.6 Lipids on urine microscopy. (**a**, **b**) Oval fat bodies seen on bright field microscopy and polarized microscopy, respectively (can be associated with nephrotic syndrome or diabetic nephropathy). (**c**, **d**) Lipid-laden cast on bright field microscopy and polarized microscopy, respectively, with the same clinical implications as (**a**). Reprinted from [18] with permission from Elsevier

CLINITEK advantus urine chemistry analyzer

FIGURE 3.7 Semi-automated urine chemistry analyzer. Reprinted with permission from Siemens Healthcare Diagnostics Inc.

Case 3: Special Urine Testing

A 34 year old female and her significant other have been trying to conceive for 8 months. They had intercourse regularly during the time of her ovulation in the month of June, and she is eager to find out if she is pregnant. Menstrual cycles are regular, and she states that according to her calendar, she will start her menstrual cycle in 3 days. In your office, she inquires when is the earliest she can take a urine pregnancy test to achieve accurate results? Can she find out any sooner with a blood test?

As this book will illustrate, in addition to commonly-used urine dipstick and urinalysis testing, many other urine tests and assays are used for diagnosis and evaluation of various patient conditions. Many of these tests will be discussed in the upcoming chapters, but the following section addresses three commonly-used tests that do not utilize routine urinalysis.

Pregnancy and Ovulation Testing

Pregnancy tests are based on the detection of β-human chorionic gonadotropin (β-hCG), secreted by trophoblastic cells of the placenta from the time of implantation, in the serum or urine. Qualitative urine testing can detect levels as low as 15 and 25 mIU/mL. The concentration is slightly lower in the urine than in the serum, so this is not quite as sensitive in detecting early pregnancy [11].

Most urine pregnancy tests are based on Enzyme Linked Immunoassay (ELISA) methodology and use monoclonal antibodies selective for the beta portion of the hCG molecule. For best results the test should not be performed before the first day of missed menses, or approximately 15 days after conception, although some claim accuracy earlier [12]. False negative results can occur secondary to early pregnancy or dilute urine. Optimal results are obtained with concentrated, first-morning specimens. Many package inserts claim >99% accuracy; however, the actual accuracy may differ depending

on who is performing the test. Office based personnel routinely perform the test, and home testing is also often used.

Results on test strips are shown by a color change, a plus or minus sign, one or two lines and on digital brands, the words "pregnant" or "not pregnant." Most test strips also have a control indicator, a line or symbol, which will appear near the test result in the result window to ensure test accuracy. Detectable levels of hCG (>5 IU/L) can be present in the serum from 8 to 11 days following conception [12]. Factors to consider when deciding between an in-office serum or urine test versus a home urine pregnancy test are length of time since missed menses, accuracy, cost and convenience.

Home ovulation predictors have become frequently-used tools for those trying to conceive in recent years. Luteinizing hormone (LH) can be detected in the urine after a monthly serum LH surge and 24–36 hours before ovulation occurs. Measurement of LH at this time does not ensure ovulation will occur, but rather when it should occur, guiding time of intercourse for couples trying to conceive. "Home LH kits" have become a relatively inexpensive option for patients wishing to track ovulation schedule. Most home ovulation kits use ELISA "dipsticks." The color change seen is proportional to the amount of LH present, and a reference range is provided. These tests have been found to accurately predict ovulation in about 70% of women [13].

Returning to case 3, a serum pregnancy test could be obtained at this time and may be able to detect the presence of β-hCG. Urine pregnancy testing, whether in the office or at-home, will not likely be accurate for another 3 days, until the day of her missed menses. If urine testing is performed, it should be a first morning specimen.

Urinary Calculi

The use of one-time routine urinalysis in diagnosis of nephrolithiasis is limited in determination of stone composition, but it can be useful in the acute diagnosis of the presence of uro-

lithiasis. Hematuria is a consistent finding in patients with urinary tract stones, even when asymptomatic. Additionally, urine pH can assist in determining type of crystal likely to form a precipitate. For example, uric acid stones are likely to form in acidic urine with pH <6.

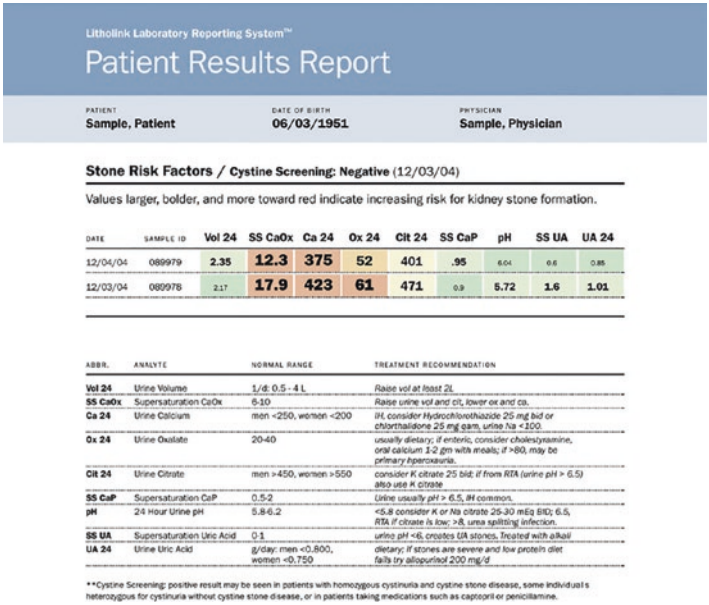
Urine collection over 24 hours is more useful in the metabolic evaluation of stone-formers. The American Urological Association (AUA) Guidelines for the Medical Management of Kidney Stones recommend metabolic testing in high risk first-time stone-formers or recurrent stone-formers. This should consist of one or two 24-hour urine collections obtained on a random diet and analyzed at a minimum for total volume, pH, calcium, oxalate, uric acid, citrate, sodium, potassium, and creatinine [14].

Multiple laboratories offer services focusing on simple, accurate 24-hour urine assessment for stone-formers. These labs provide patients with collection containers and chemical preservatives that obviate the need for refrigerated storage and transport. Cumulative data is extrapolated from a small aliquot of the entire collection. After values of urinary elements and saturations have been calculated, the physician receives a numeric interpretation of results (Fig. 3.8).

Analysis of stone composition itself is performed by various methods including optical crystallography, radiograph diffraction, and infrared spectroscopy [8]. Most laboratories refer calculi specimens to specialized laboratories where chemical and specialized testing are used. See Chap. 12 for further details on urine testing for crystals and stones.

Cytopathologic Examination

The preparation of permanent slides using cytocentrifugation followed by Papanicolaou staining is frequently performed for detection of malignancies of the urinary tract. A first-morning voided specimen is recommended. This testing can also assist with diagnosis of transplant rejection, uncom-



Litholink Laboratory Reporting System™

Patient Results Report

PATIENT	DATE OF BIRTH	PHYSICIAN
Sample, Patient	06/03/1951	Sample, Physician

Stone Risk Factors / Cystine Screening: Negative (12/03/04)

Values larger, bolder, and more toward red indicate increasing risk for kidney stone formation.

DATE	SAMPLE ID	Vol 24	SS CaOx	Ca 24	Ox 24	Cit 24	SS CaP	pH	SS UA	UA 24
12/04/04	089979	2.35	12.3	375	52	401	.95	6.04	0.6	0.85
12/03/04	089978	2.17	17.9	423	61	471	0.9	5.72	1.6	1.01

ABBR.	ANALYTE	NORMAL RANGE	TREATMENT RECOMMENDATION
Vol 24	Urine Volume	1/d: 0.5 - 4 L	*Raise vol at least 2L*
SS CaOx	Supersaturation CaOx	6-10	*Raise urine vol and cit, lower ox and ca.*
Ca 24	Urine Calcium	men <250, women <200	*IH, consider Hydrochlorothiazide 25 mg bid or chlorthalidone 25 mg qam, urine Na <100.*
Ox 24	Urine Oxalate	20-40	*usually dietary; if enteric, consider cholestyramine, oral calcium 1-2 gm with meals; if >80, may be primary hyperoxaluria.*
Cit 24	Urine Citrate	men >450, women >550	*consider K citrate 25 bid; if from RTA (urine pH > 6.5) also use K citrate*
SS CaP	Supersaturation CaP	0.5-2	*Urine usually pH > 6.5, IH common.*
pH	24 Hour Urine pH	5.8-6.2	*<5.8 consider K or Na citrate 25-30 mEq BID; 6.5, RTA if citrate is low; >8, urea splitting infection.*
SS UA	Supersaturation Uric Acid	0-1	*urine pH <6, creates UA stones. Treated with alkali*
UA 24	Urine Uric Acid	g/day: men <0.800, women <0.750	*dietary; if stones are severe and low protein diet fails try allopurinol 200 mg/d*

**Cystine Screening: positive result may be seen in patients with homozygous cystinuria and cystine stone disease, some individuals heterozygous for cystinuria without cystine stone disease, or in patients taking medications such as captopril or penicillamine.

FIGURE 3.8 Quantitative results of urine metabolic evaluation from 24-hour urine collection, reprinted with permission from Litholink Corporation (Itasca, IL)

mon infections, cellular inclusions, pathologic casts, and inflammatory conditions [1]. Examination by a pathologist is usually required to confirm suspicious cells. See Chap. 14 for further details on cytologic examination.

Summary

When obtaining urine tests, collection and analysis methods should be fully understood by the provider. This knowledge not only allows accurate interpretation of testing, but provision of adequate patient and personnel training, and determination of the most appropriate test for a particular diagnosis.

Test analysis methods of varying scales can achieve accurate results, from visual dipstick analysis to completely automated analysis. Urine dipstick, microscopic urine analysis, and a multitude of other urine tests can serve as minimally-invasive, inexpensive first-line testing for both urinary tract and systemic disease.

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Chapter 4
Urine Dipstick: Blood – The Spectrum of Red

Alexandra J. Sharp and Victoria J. A. Sharp

Objectives

- Understand recommendations and clinical indications for performing a urine dipstick test for blood
- Understand causes of false negative and false positive results of blood on urine dipstick
- Correlate positive urine dipstick for blood with urine microscopic exam for the presence or absence of red blood cells and discuss differential diagnoses of each
- Discuss appropriate referrals to urology and/or nephrology based on results of urine dipstick for blood

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Overview

Dipstick urinalysis for blood is a quick, easy, noninvasive and relatively inexpensive test to perform. Of note, this test should only be performed in the appropriate clinical setting due to potential risks to the patient and unnecessary costs if an evaluation is undertaken for false positive results [1]. It is a screening test and if positive, should always be followed up with urine microscopy. The focus of this chapter is the urine dipstick test for blood: the test itself, sensitivity and specificity, factors associated with false positive and false negative results, indications/recommendations, risk factors, differential diagnoses and appropriate referrals.

The Urine Dipstick Test for Blood

A urine reagent dipstick is a plastic strip that has reagent pads bonded to it. Most testing strip pads use an oxidative chemical reaction that changes the color of the pad depending on what is present in the urine [2]. For urine dipstick testing for blood, hemoglobin in the blood acts as a peroxidase that releases an oxidizing agent that causes the color of the testing pad to change (Fig. 4.1) [3].

The color to which the pad changes varies depending on the brand of testing strip that is being used (Table 4.1). After 60 seconds the color can be compared to a standardized color chart for the brand of testing strip. Accutest, Multistix and Chemstrip turn green in the presence of heme [3–5], while Atlas Medical strips turn blue in the presence of heme [6]. If the testing pad on the reagent strip is uniform in color, there

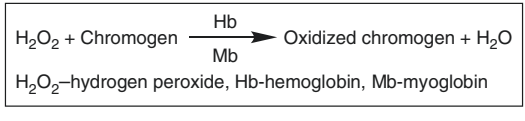


Figure 4.1 Chemical peroxidase reaction that causes a color change on the testing reagent pad for blood positivity [2]

Table 4.1 Urine reagent color characteristics of various brands of test strips

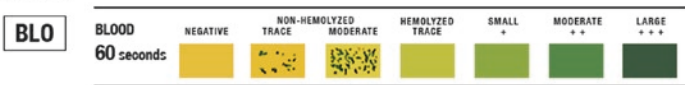
Mulitstix[a]	**Color characteristics** Equally sensitive to myoglobin and hemoglobin Color ranges from orange to green to blue Most commonly used **Color Chart** 
Atlas Medical	**Color characteristics** Separate color scale for hemoglobin and erythrocytes Color ranges from orange to green to dark blue **Color chart** 

Table 4.1 (continued)

ChemstSrip	**Color characteristics**
	Separate color blocks for erythrocytes and hemoglobin
Accutest	**Color characteristics**
	Tests for hemoglobinuria and myoglobinuria equally

Color charts reprinted with permission from Atlas Medical and Siemens Healthcare Diagnostics Inc.
Refs. [3–6]
[a]This is for educational purposes only and not intended for use to interpret test results. The colors as they appear may not be the exact color on the official product label

may be free hemoglobin or myoglobin present from lysed cells, as the test pad is sensitive to both. A testing pad that is not uniform in color and appears blotchy or spotty is a good indication of intact red blood cells (RBC) [3–5]. There may be a combination of the two caused by hemoglobin being released from lysed red blood cells.

Urine maintained at room temperature should be tested within two hours to avoid changes that can occur in unpreserved urine leading to false positive or negative results [2]. If the urine was maintained in a refrigerator, it should be warmed to room temperature and stirred so if there are blood cells present they are resuspended in order to interact with the testing pad of the strip. Unmixed urine can be the cause of a false negative result [7].

The mode of sample collection must also be considered. Menstrual blood can cause contamination. For women with atrophic vaginitis, red blood cells from the dry and easily abrased tissue can also contaminate the sample [6]. A urinary catheterized sample can decrease contamination but is more invasive. Proper collection and testing techniques are further discussed in Chap. 3. The dipstick should be dipped very briefly, as extended exposure can cause incorrect readings [2]. Testing strips that are expired or stored incorrectly can also provide inaccurate results [8].

The urine dipstick for blood measures if there are intact red blood cells, hemoglobin, or myoglobin present in the urine. Test results are reported in different ways: some manufacturers report their results in concentrations (mg/dL) corresponding to different colors on the strip; some report as trace, small, moderate, large; some report in a plus system (1+, 2+, 3+, 4+); and some report as positive, negative, or normal [2]. Multistix, one of the more commonly used brands, reports results as non-hemolyzed negative, trace, or moderate, and hemolyzed trace, small +, moderate ++, or large +++ [4].

Sensitivity/Specificity

The sensitivity for multiple reagent strips assessing for blood ranges from 91% to 100% and the specificity ranges from 65% to 99% [9]. Reagent strips can detect free hemoglobin as low as 0.015–0.062 mg/dL and 5–10 Ery/μL (in urine specimens with vitamin C (ascorbic acid) content of <50 mg/dL) [4–6]. High urine pH can decrease the sensitivity of the reagent strips and high vitamin C content >50 mg/dL can inhibit color formation on some test strips [6]. Sensitivity can also vary depending on the variability of color perception and on the lighting conditions when strips are read manually [3].

Both patient and laboratory factors that can cause false positive or negative results are listed in Table 4.2. Due to possible false positive results, a positive dipstick should always be followed by microscopy [2]. If a patient is taking high supplemental daily doses of vitamin C, which can cause a false negative result, the dose of vitamin C should be limited for 1–2 days before testing to get an accurate result [10].

The next step in testing following a positive dipstick for blood is microscopy [11]. Microscopy has the ability to differentiate true hematuria from hemoglobinuria and myoglobinuria by detecting the presence of red blood cells. There may not be visual color clues in the urine for blood [2]. Urine dipstick with microscopy is often the most efficient way to order these tests. Most laboratories will only perform the microscopy if the urine dipstick is positive. If the provider (usually urologist or nephrologist) performs these tests in their own Clinical Laboratory Improvement Amendments (CLIA) approved laboratory, they may routinely perform both components.

TABLE 4.2 Possible causes for false dipstick results for blood in urine [2, 8, 9, 10, 12, 13]

False positive causes
Peroxidases (microbial peroxidase) - associated with urinary tract infection
Myoglobin
Oxidizing agents (hypochlorite)-cleaning agents in collection cup
Menstrual contamination
Atrophic vaginitis
Dehydration
False negative causes
Medications Acetylcysteine Quinidine Cefoxitin Levodopa Mesna Keflin Lodine Hydrochlorothiazide Metformin *Curcuma* (turmeric) Chloroquine Captopril Vitamin C (ascorbic acid)
Chlorhexidine staining
High specific gravity
pH < 5.1
Proteinuria
High nitrite (>10mg/dL)
Formalin-used to preserve urine
Unmixed specimens

When Is it Appropriate to Check Urine for Blood?

Based on current consensus, a urine dipstick test for blood should not be ordered for screening in the absence of symptoms [8]. Testing would be appropriate in patients with gross hematuria, lower urinary tract symptoms (urinary frequency, hesitancy, urgency, dysuria or nocturia), renal failure, systemic signs of disease (rash, joint pain), trauma, vascular catastrophe, renal infarct, suspicion of stones, vasculitis, glomerulonephritis, or hereditary renal diseases such as thin basement membrane disease. The US Preventive Services Task Force (USPSTF) recommendation states that the current evidence is insufficient to routinely screen for bladder cancer in asymptomatic adults. This recommendation is supported by The American Academy of Family Physicians. The American Cancer Society recommends prompt evaluation if there are symptoms present (such as gross hematuria). No major organization currently recommends asymptomatic screening [14].

In addition to the urine dipstick, a thorough history and physical exam should be performed for evaluation of hematuria. Part of the history should include any possible medications and foods that could discolor the urine to appear red. Possibilities include beets, blackberries, rhubarb, phenolphthalein and rifampin [9]. It is still important to perform a urine dipstick to check for blood in patients who have discolored urine and have ingested any of these medications/foods. Urine can look red visually but be dipstick negative. If the dipstick is positive, follow up microscopy is needed to assess for pathology.

Case 1: Positive Dipstick for Blood, Negative Microscopy for Red Blood Cells

A 20 year old college football player has been training more than usual with a new conditioning coach previously employed by the NFL. He presents to student health with fever, nausea, myalgia, muscular weakness and dark red urine.

Urine dipstick and urine microscopy	
Component	**Result**
Color	Red
Appearance	Cloudy
pH	6.0
Specific gravity	1.010
Blood	3+
Glucose	Negative
Ketone	Negative
Protein	Negative
Bilirubin	Negative
Urobilinogen	Negative
Leukocyte esterase	Negative
Nitrite	Negative
White blood cells (WBC)	0–5/hpf
Red blood cells (RBC)	0–2/hpf
Squamous epithelial cells	0/lpf
Casts	2/hpf, Granular
Bacteria	0/hpf

In this case the dipstick for blood was positive, the supernatant was red (Fig. 4.2), and a follow up microscopic urine examination was performed showing no red blood cells (dipstick +/microscopic exam−).

The differential diagnoses for this scenario include myoglobinuria and hemoglobinuria. It is very important to determine the diagnosis because myoglobinuria can cause acute kidney injury quickly [15]. Myoglobin is an intracellular protein involved in the transport of oxygen in the muscles and increases in the bloodstream when muscle tissue is damaged [2]. Myoglobinuria is usually caused by rhabdomyolysis/myositis or muscle destruction [10, 15]. Table 4.3 lists possible etiologies of myoglobinuria.

FIGURE 4.2 Urine of patient with rhabdomyolysis (Courtesy of University of Iowa, Division of Nephrology)

A positive reagent strip detects the presence of heme pigment, but cannot distinguish whether this is due to hemoglobin, myoglobin or intact red blood cells. A rapid ammonium sulfate precipitation test, not routinely performed, can differentiate between hemoglobin and myoglobin because it precipitates hemoglobin. For this test ammonium sulfate (2.8 g) is slowly added and thoroughly mixed in a 5 ml urine sample and then centrifuged. The test is based on the different solubilities of hemoglobin and myoglobin when saturated with 80% ammonium sulfate. At this concentration, hemoglobin precipitates out and myoglobin remains soluble in the supernatant. If the supernatant is clear, hemoglobin was precipitated by ammonium sulfate. If myoglobin is present, the supernatant is red [10, 16].

Table 4.3 Possible causes of myoglobinuria [2, 10, 16]

Muscle trauma
Crushing injuries
Surgery
Contact sports
Vigorous muscle exercises
Compartment syndrome
Polymyositis
Muscle ischemia
Carbon monoxide poisoning
Alcohol use
Illicit drug use
Muscle infections (myositis)
Viral (Coxsackie, influenza, or myxoviruses)
Bacterial (trichinosis)
Myopathy due to medications
HMG CoA reductase inhibitors (statins)
Cocaine
Salicylates
Succinylcholine
Anti-psychotics
Anti-depressants
Sedative-hypnotics
Antihistamines
Seizures/convulsions
Toxins
Snake venoms
Spider bites
Hyperthermia, heat stroke
Electric shock
Haff disease (after ingestion of fish—cause unknown)

Symptoms of myoglobinuria manifest as the classic triad of myalgia, muscle weakness, and dark urine. Other symptoms include fatigue, fever, tachycardia, nausea and vomiting [15]. Myoglobin is freely filtered through the glomerulus and is rapidly excreted in urine. When it reaches the distal convoluted tubules, it precipitates in acidic urine causing acute renal injury [15]. Figure 4.3 shows urine microscopy of patient with acute tubular necrosis caused by myoglobinuria. Evaluation should include a serum creatine kinase (CK), an enzyme released by damaged muscle. A level above 1000 U/L (5 times the upper limit of normal) is consistent with a diagnosis of rhabdomyolysis. Intravenous hydration helps protect the kidneys by increasing excretion of myoglobin [15].

There are several different causes of hemoglobinuria (Table 4.4), including paroxysmal nocturnal hemoglobinuria (PNH), March hemoglobinuria, and paroxysmal cold hemoglobinuria (PCH). PNH usually occurs at night or in the early morning as a result of a breakdown of red blood cells. Symptoms include dark/red urine, weakness, shortness of breath, headache, irregular heartbeat, chest pain, abdominal pain, ulcers, pallor, jaundice, easy bruising, hemoptysis, impo-

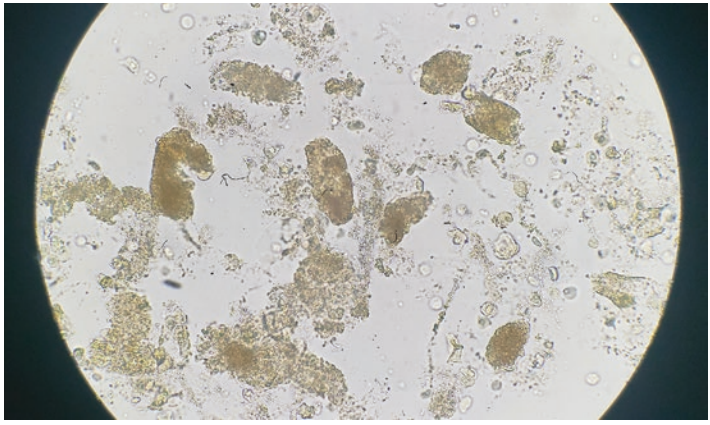


Figure 4.3 Urine of patient with rhabdomyolysis and acute tubular necrosis (Courtesy of University of Iowa, Division of Nephrology)

Table 4.4 Possible causes of hemoglobinuria [2, 10]

Intravascular hemolysis
Transfusion reactions
Autoimmune and microangiopathic hemolytic anemia
Paroxysmal nocturnal hemoglobinuria (PNH)
Thrombotic thrombocytopenic purpura (TTP)
Paroxysmal cold hemoglobinuria (PCH)
Extensive burns
Infections
Falciparum malaria
Clostridium perfringens
Syphilis
Mycoplasma spp.
Chemical toxicity
Copper
Nitrites
Nitrates
Drug induced
Strenuous exercise
Marching
Karate
Long-distance running
Sudden cold
Eclampsia
Sickle cell crisis
Multiple myeloma
Alkaloids
Mushrooms

tence, seizures and blood clots [17]. Treatment options for hemolytic (classical) PNH include allogeneic hematopoietic cell transplantation (HCT) and complement inhibition with eculizumab [18].

March hemoglobinuria is a result of strenuous exercise. It causes red urine and occasionally cough, fatigue and myalgias [19]. Symptoms usually resolve with rest.

Paroxysmal cold hemoglobinuria is rare and occurs when an individual is exposed to the cold. The acute form is seen in young children after a viral illness and the chronic form is seen with some hematological malignancies and tertiary syphilis [20]. Symptoms of PCH include chills, fever, back pain, leg pain, abdominal pain, headache, blood in the urine/red urine, and general discomfort [21]. Treatment for an acute episode is usually supportive consisting of cold avoidance, rest, pain medication, blood transfusion and immunosuppressive therapy in severe cases [20]. It is important to distinguish if hemolytic anemia is also present with the hemoglobinuria because if present it can help distinguish PCH from other forms of hemolytic anemia [15].

Returning to case 1, based on the patient's symptoms, clinical history and urine test results (dipstick 3+ blood/micro-), his most likely diagnosis is myoglobinuria and possibly rhabdomyolysis if his CK is above 1000 U/L. Severity of his illness would determine if inpatient admission is required.

Case 2: Positive Dipstick for Blood, Positive Microscopy for Normal Shaped Red Blood Cells

A 70 year old male with a 35 pack-year smoking history, who has worked in his family's dry cleaning business for about 50 years, presents to his physician's office complaining of a single episode of gross hematuria.

Urine dipstick and urine microscopy	
Component	**Result**
Color	Red
Appearance	Cloudy

Urine dipstick and urine microscopy	
Component	**Result**
pH	6.0
Specific gravity	1.015
Blood	3+
Glucose	Negative
Ketone	Negative
Protein	Negative
Bilirubin	Negative
Urobilinogen	Negative
Leukocyte esterase	Negative
Nitrite	Negative
White blood cells (WBC)	0–5/hpf
Red blood cells (RBC)	>50 isomorphic/hpf
Squamous epithelial cells	0/lpf
Casts	0/hpf
Bacteria	0/hpf

In this case the patient has a positive urine dipstick for blood (in the absence of other positive dipstick results), followed by a positive urine microscopy showing a large amount of isomorphic red blood cells (in the absence of other cells such as white blood cells and casts).

The differential diagnosis for this scenario includes urothelial cancers. A focused history can help with risk stratification. See Table 4.5 for risk factors for urinary tract malignancies. Table 4.6 shows possible etiologies of nonglomerular hematuria, which are more likely in the case of isomorphic RBCs.

Table 4.5 Common risk factors for urinary tract malignancies in patients with hematuria [22, 23]

Smoking
>35 years of age
Male
Exposure to chemicals or dyes (benzenes or aromatic amines)
Family history of urothelial cancer
Dry cleaners
Hairdressers
Painters
Truck drivers
Petroleum, chemical, leather, textile, tire, rubber workers
Pelvic irradiation
Drugs
Cyclophosphamide
Pioglitazone
Chronic cystitis
Chronic urinary tract infection
Indwelling bladder catheters
Chronic bladder stones
Schistosoma haematobium infection

Table 4.6 Non-glomerular causes of hematuria [2, 9, 10, 24, 25]

Infection/inflammation
Cystitis, pyelonephritis, urethritis
Sexually transmitted infection
Atypical urinary tract infection (e.g., schistosomiasis)
Radiation cystitis
Interstitial cystitis
Drugs
Cyclophosphamide (hemorrhagic cystitis)
Anticoagulants (e.g., warfarin)

Table 4.6 (continued)

Calculi
Renal, ureteral, bladder
Asymptomatic crystalluria (hypercalciuria, hyperuricosuria)
Benign prostatic hyperplasia
Obstruction
Urethral/ureteral stricture
Ureteropelvic junction obstruction
Posterior urethral valves
Tumor/neoplasm
Kidney, ureter, bladder, prostate, urethra
Gynecologic
Endometriosis
Menstrual contamination
Atrophic vaginitis
Other anatomic/structural causes
Cystic renal disease
Urethral diverticulum
Urogynecologic or uroenteric fistula
Arteriovenous malformation
Renal artery/vein thrombosis
Miscellaneous causes
Blunt trauma
Recent urinary tract instrumentation (catheterization, cystoscopy)
Sexual trauma
Vigorous exercise
Sickle cell disease
Loin pain hematuria syndrome

Returning to case 2, based on the patient's true hematuria (dipstick+/micro+), isomorphic RBCs, and high risk for a urinary tract malignancy, further evaluation would be required, and he should be referred to a urologist (see Chap. 9).

Case 3: Positive Dipstick for Blood, Positive Microscopy for Dysmorphic RBCs

A 22 year old black female presents to her primary care physician with new onset rust colored urine and swelling in legs, ankles and feet. Her blood pressure is 165/100.

Urine dipstick and urine microscopy	
Component	**Result**
Color	Red
Appearance	Cloudy
PH	6.0
Specific gravity	1.015
Blood	2+
Glucose	Negative
Ketone	Negative
Protein	2+
Bilirubin	Negative
Urobilinogen	Negative
Leukocyte esterase	Negative
Nitrite	Negative
White blood cells (WBC)	3–5/hpf
Red blood cells (RBC)	15–20 dysmorphic/hpf
Squamous epithelial cells	0/lpf
Casts	2/hpf, Red blood cell
Bacteria	0/hpf

Table 4.7 Glomerular causes of hematuria

Immune complex diseases
IgA nephropathy
Membranoproliferative disease
Post-infectious glomerulonephritis
Lupus nephritis
Basement membrane diseases
Alport syndrome
Anti-glomerular basement membrane (GBM) disease
Thin basement membrane disease
Pauci-immune diseases
Granulomatosis with polyangiitis
Eosinophilic granulomatosis with polyangiitis
Microscopic polyangiitis

Due to hematuria in the presence of proteinuria and casts, as well as her clinical symptoms and elevated blood pressure, the differential diagnosis needs to include glomerular etiologies (Table 4.7) and she needs further evaluation with referral to a nephrologist (see Chap. 5).

Summary

A urine dipstick assessing for blood should only be performed in patients based on clinical indications. There are no guidelines to test an asymptomatic individual. A positive dipstick for blood should always be confirmed with a microscopic assessment looking for a positive result of ≥3 RBCs/hpf. If the dipstick is positive and the microscopic exam is negative, other causes should be explored, such as a negative test, hemoglobinuria or myoglobinuria (Fig. 4.4) [10]. Depending on the clinical scenario, referral to a specialist (urologist and/or nephrologist) may be required (Fig. 4.5) [26].

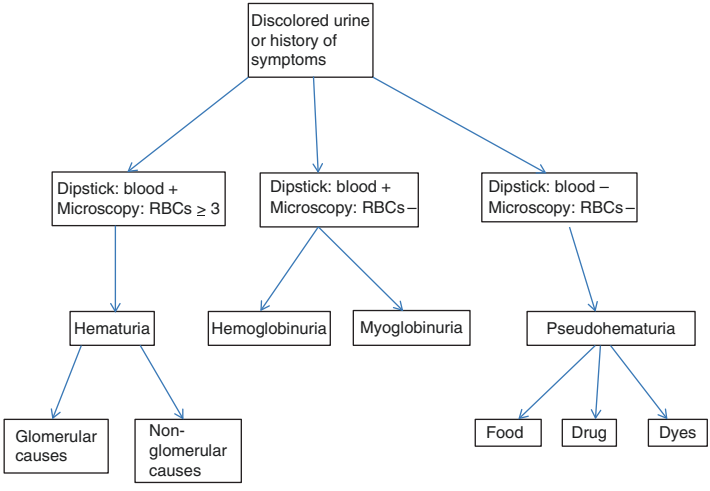


FIGURE 4.4 Algorithm for the differential diagnosis of hematuria

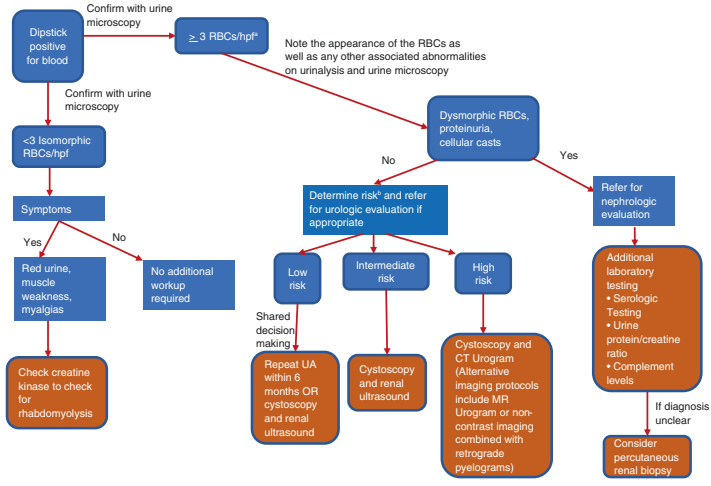


FIGURE 4.5 Workup for positive urine dipstick for blood
[a]If urinary tract infection or gynecologic source, treat and recheck UA to confirm
[b]**Low risk** (all of the following): women age < 50 yrs, men age < 40 yrs, never smoker or < 10 pack-years, 3–10 RBC/hpf on 1 UA, no other risk factors for urothelial cancer and no prior episode of microscopic hematuria
Intermediate risk (any of the following): women age 50–59 yrs, men age 40–59 yrs, 10–30 pack-years smoking, 11–25 RBC/hpf on 1 UA, > 1 additional risk factor for urothelial cancer, and previously low risk, no prior evaluation and 3–25 RBC/hpf on repeat UA
High risk (any of the following): age > 60 yrs, > 30 pack-years smoking, > 25 RBC/hpf on 1 UA, history of gross hematuria, and previously low-risk, no prior evaluation and > 25 RBC/hpf on repeat UA

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Chapter 5
Urine Dipstick: Proteinuria – Causes, Consequences and Diagnostic Approach

Lewis Mann, Lisa M. Antes, and M. Lee Sanders

Objectives

- Recognize the pathophysiology and significance of proteinuria
- Understand the different methods to test for urine protein as well as the strengths and weaknesses of each method
- Differentiate between nephrotic and nephritic causes of proteinuria and discuss initial testing considerations for each cause
- Identify which patients should be screened for proteinuria
- Discuss the medical management of proteinuria

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Overview

The urine is typically void of protein due to a complex filtration barrier in the glomerulus. Urine dipstick detection of protein is usually an indication that this glomerular barrier is compromised. Quantifying the amount of urine protein is a vital part of the work-up and management of proteinuria. Both nephrotic and nephritic glomerular diseases can cause proteinuria. A nephrologist should be consulted when an intrinsic kidney pathology is suspected as the cause of proteinuria as a kidney biopsy may be indicated. Certain patient populations should be routinely screened for proteinuria as early detection and treatment may mitigate kidney disease progression.

The Pathophysiology and Significance of Proteinuria

Urine starts as ultrafiltrate that is formed by the constant filtering of blood through the glomerulus (see Chap. 1). Electrolytes and glucose are small enough to pass freely through the filter, while large molecules such as most proteins are almost entirely excluded from the ultrafiltrate. The ultrafiltrate flows through the tubules where useful molecules are reabsorbed and waste products are secreted. Any proteins which were able to pass through the filter are reabsorbed primarily in the proximal tubule. Therefore, in a healthy individual, urine should be void of protein [1–2].

Three layers combine to create the filtration barrier that excludes protein from entering the ultrafiltrate: the endothelial cells of the renal capillaries, the glomerular basement membrane, and the epithelial cells lining the urinary space (Fig. 5.1). Gaps called fenestrations exist between the endothelial cells measuring 50–100 nm. These fenestrations which comprise 20–50% of the endothelial surface in theory should allow molecules like albumin with a diameter of approximately 3.6 nm to pass between the cells during ultrafiltation;

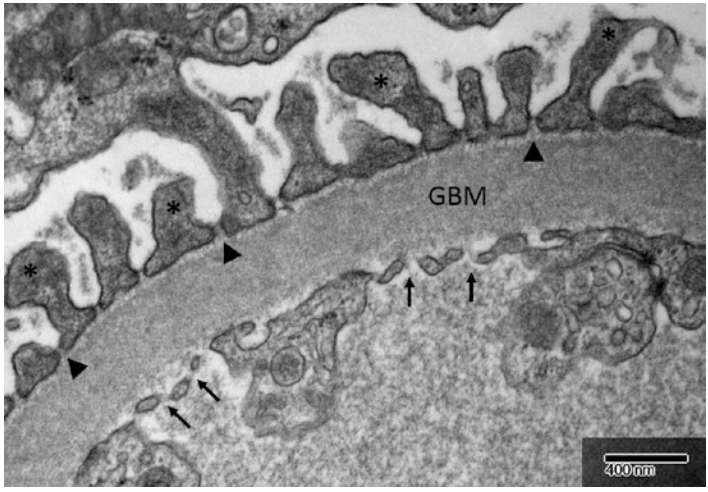

FIGURE 5.1 Transmission electron micrograph of a normal intact human glomerular filtration barrier. Arrows indicate the endothelial fenestrations of the renal capillaries, GBM is the glomerular basement membrane, asterisks indicate podocyte foot processes, and arrowheads indicate slits between podocyte foot processes (×40,000). (Courtesy of Danni Holanda MD, University of Iowa, Department of Pathology)

however, in actuality, approximately 90% of albumin does not pass through the fenestrations. The reason is both albumin and the endothelial surface glycoproteins have a net negative charge which results in charge repulsion and decreased filtration of albumin [3].

The second barrier is the glomerular basement membrane (GBM) which is comprised of type IV collagen. The collagen forms a meshwork of fibers with gaps that are too small for most remaining proteins to fit through.

The third barrier is specialized epithelial cells called podocytes that line the urinary space. Podocytes form a complex of foot processes that interdigitate with the foot processes of neighboring podocytes. The space between them measures only 2.0 nm [3]. A network of adhesion molecules forms a diaphragm between the neighboring foot processes that also

contributes to charge and size selectivity that completes the filter. Its most important function however, is to resist the outward pressure of fluid within the capillaries. Without the podocytes, the collagen meshwork of the GBM would stretch and thin, widening the spaces between the collagen fibers, allowing more protein to pass through.

Overall, this combined filtration barrier uses both charge and size selectivity to almost entirely exclude protein from the ultrafiltrate. What protein does pass through is almost entirely reabsorbed in the proximal tubule. The end result is urine that is void of protein [1, 4].

Proteinuria is a marker for renal dysfunction, reflecting a disruption of the filtration barrier. The primary protein detected in the urine in patients with renal dysfunction is albumin, which is the most common protein in the serum. The Kidney Disease: Improving Global Outcomes (KDIGO) classification system of chronic kidney disease in addition to glomerular filtration rate also includes albuminuria detection and quantification as staging criteria (Fig. 5.2).

The amount of proteinuria has been correlated to the risk of kidney disease progression as well as an increased risk for myocardial infarction, stroke, and heart failure. Even low levels of albuminuria are associated with an increase in cardiovascular events that is independent of the presence of diabetes, hypertension, or chronic kidney disease. There is also evidence that proteinuria reduction using medications that target the renin-angiotensin system can reduce these cardiovascular risks [5].

Case 1: Dipstick Positive Proteinuria in a Healthy/Low Risk Patient

A 35 year old woman with no medical problems and no complaints presents for an annual physical exam. On review of her medical record, you note that a urinalysis from 2 months ago showed 2+ proteinuria with no other abnormalities. What is the significance? What further workup is indicated?

CKD is classified based on: · Cause (C) · GFR (G) · Albuminuria (A)				Albuminuria categories Description and range		
				A1	A2	A3
				Normal to mildly increased	Moderately increased	Severely increased
				<30 mg/g <3 mg/mmol	30-299 mg/g 3-29 mg/mmol	≥300 mg/g ≥30 mg/mmol
GFR categories (ml/min/1.73m^2) Description and range	G1	Normal or high	≥90	1 if CKD	Treat 1	Refer* 2
	G2	Mildly decreased	60-89	1 if CKD	Treat 1	Refer* 2
	G3a	Mildly to moderately decreased	45-59	Treat 1	Treat 2	Refer 3
	G3b	Moderately to severely decreased	30-44	Treat 2	Treat 3	Refer 3
	G4	Severely decreased	15-29	Refer* 3	Refer* 3	Refer 4+
	G5	Kidney failure	<15	Refer 4+	Refer 4+	Refer 4+

FIGURE 5.2 The Kidney Disease: Improving Global Outcomes (KDIGO) classification system of chronic kidney disease (CKD) is based on the glomerular filtration rate (GFR) and amount of albuminuria. Colors represent the risk for CKD progression: green (low risk), yellow (moderately increased risk), orange (high risk), red (very high risk). Numbers in each of the colored boxes represent a recommendation for the number of times per year the patient should be monitored. "Treat" corresponds to medical management which can be done by a primary care provider whereas "Refer" indicates that nephrology referral and services are recommended. In those "Refer" recommendations containing an asterisk (*), referring clinicians may wish to discuss with their nephrology colleagues depending on local arrangements regarding monitoring or referral. (Reprinted from The American Journal of Medicine, Volume 129/ Issue 2, Vassalotti JA, Centor R, Turner BJ, Greer RC, Choi M, Sequist TD, Practical Approach to Detection and Management of Chronic Kidney Disease for the Primary Care Clinician, p153-162e7, Copyright (2016), with permission from Elsevier)

The Dipstick Analysis

The urine dipstick should be negative for protein in an otherwise healthy individual whereas a positive urine dipstick for protein raises the possibility for renal pathology. A urine

dipstick reports the presence of protein (albumin) in a range of negative to 4+. This range can be roughly corresponded to a daily excretion of protein shown in Table 5.1.

There are several problems inherent in the measurement of proteinuria via dipstick. First, the analysis is done by a human observer so there will always be the possibility of error comparing the color of the dipstick indicator to the guide on the dipstick bottle. Second, a measurement of trace protein on a dipstick is only 69% sensitive for albuminuria in the range of 30–300 mg/g which was previously called "microalbuminuria" but now called "moderately increased" albuminuria [6]. Due to its low sensitivity, the urine dipstick should not be used to screen for proteinuria in high risk populations. Instead, screen for microalbuminuria using special albumin-specific urine dipsticks which have a lower detection threshold compared to standard urine dipsticks. Third, the dipstick protein measurement can be altered by the overall urine concentration. If the urine is more dilute, the measured dipstick concentration of protein can be falsely low while concentrated urine can overestimate the amount of proteinuria.

Another inherent problem with the routine protein dipstick analysis is related to the mechanism of the test itself. The indicator paper of the dipstick contains one of several chemicals

Table 5.1 Urine dipstick result rough correlation with daily protein excretion

Urine dipstick result	Urine sample estimated protein concentration (mg/dL)	Daily estimated protein excretion (mg/day)
Negative	<10	n/a
Trace	10–30	<500
1+	30–100	<500
2+	100–300	500–1000
3+	300–1000	1000–2000
4+	>1000	>2000

(manufacturer dependent) which undergo a color change when bound to protein resulting in a "positive" test [1, 7]. This color change detection is the result of the urine protein being able to accept protons. Albumin is the most abundant serum protein so a defect in the filtration barrier usually results in albumin being the most significant contributor to proteinuria. Albumin is a large negatively charged protein with many sites that can be protonated at different pKas. The majority of these pKas are 5–7 [8] which is also the normal pH of urine on a typical high-protein Western diet. Most other proteins in contrast have fewer sites that can be protonated and have lower pKas, meaning these proteins will not be well detected except in very acidic urine. Immunoglobulins have no sites that can be protonated and therefore will not be identified on a dipstick analysis [1, 4].

Urine Protein Quantification

The next step after a positive dipstick for protein is to quantify the amount of protein present in the urine. The gold standard for quantifying urine protein amount is a 24-hour urine collection. Collecting a 24-hour urine sample for protein quantification can be readily completed on inpatients but is cumbersome and error prone for outpatients (see Chap. 3).

Much more common is to collect a spot urine protein to creatinine ratio (PCR). The spot ratio theoretically works if you presume that protein and creatinine excretion rates remain constant during the day, i.e., that the glomerular filtration rate remains constant. There are two problems with this presumption: first, it assumes that the patient has excreted the normal amount of creatinine per 24 hours (approximately 1 g). This assumption is not quite true as the amount of daily creatinine produced and excreted varies based on an individual's muscle mass, metabolism, and kidney function. Second, it also assumes that the rate of creatinine filtration is constant but the rate actually varies day to day and hour to hour in the same individual [9].

Despite these two problems, the protein to creatinine ratio has become the standard metric for quantifying and trending

urinary protein. The reason is two-fold: first, PCR is easy to obtain in a clinic setting as it requires a single urine specimen. Second, this single PCR measurement correlates well with a 24-hour protein measurement; however, the correlation is less reliable as the amount of proteinuria increases [10] (Fig. 5.3). If there is doubt as to the reliability of a PCR measurement, it should be verified by obtaining a 24-hour urine collection. The reliability of a 24-hour urine protein collection can also be verified by measuring the 24-hour urine creatinine. Depending on amount of lean body mass, 24-hour total creatinine excretion should be 10–15 mg/kg for women and 15–20 mg/kg for men [4]. If the measured 24-hour creatinine in the patient-provided sample is outside this range, the most likely explanation is that the 24-hour collection was not done properly [5].

Clinical Pearl
If a urine dipstick is positive for protein, the next step is to quantify the amount of protein.

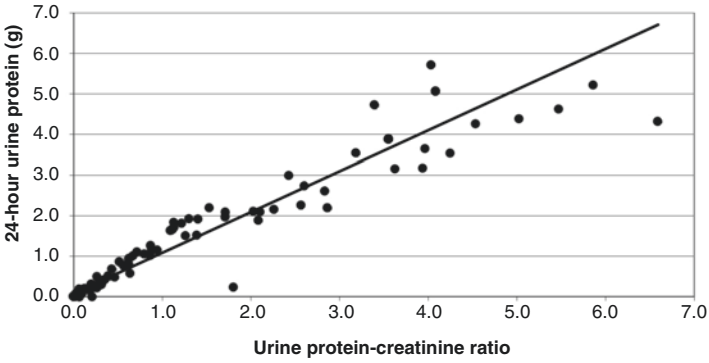

Figure 5.3 Correlation between a single void urinary protein to creatinine ratio (PCR) and a 24-hour urine collection for urine protein quantification. The PCR is easy to obtain/calculate and correlates well with a 24-hour urine collection except at higher levels of proteinuria

Urine Albumin to Creatinine Ratio

There are other problems with urine protein quantification. The proteins excreted vary greatly between individuals. There is no standardized reference for labs to calibrate against, leading to poor inter-lab agreement when quantifying total protein. This holds true for both the 24-hour collection and the spot urine protein to creatinine ratio. The urine albumin to creatinine ratio (ACR) has been suggested to mitigate this problem. The ACR has improved inter-lab precision and is more sensitive at lower levels of urine protein; however, it does miss non-albumin proteins such as immunoglobulins in diseases such as multiple myeloma. Tubular diseases also tend to have a greater proportion of non-albumin proteinuria and may also be underreported. It is important to note that while PCR and ACR are equally valid in predicting disease progression [11], the values are not interchangable and the KDIGO CKD cutoff values are different (Table 5.2).

Table 5.2 Relationship among A1-A3 categories for albuminuria and proteinuria. (Adapted from Kidney International Supplements, Volume 3/Issue 1, Chapter 1: Definition and classification of CKD, Pages No. 19-62, Copyright (2013), with permission from Elsevier.)

	Normal to mildly increased (A1)	**Moderately increased (A2)**	**Severely increased (A3)**
Albumin to creatinine ratio (ACR)			
(mg/mmol)	<3	3–30	>30
(mg/g)	<30	30–300	>300
Protein to creatinine ratio (PCR)			
(mg/mmol)	<15	15–50	>50
(mg/g)	<150	150–500	>500

Transient Causes of Proteinuria

It is important to note that there are transient causes for proteinuria. A patient with a urinary tract infection can have detectable proteinuria which will usually resolve with the treatment of the infection. Fever and heavy exercise can also lead to transient proteinuria.

Orthostatic proteinuria (more common in teenagers compared to adults) is another cause of transient proteinuria. In this benign condition of unknown etiology, there is normal urinary protein excretion during the night but increased excretion during the day. A first void morning specimen will yield a PCR <0.2 while a mid-day void will yield a higher PCR [12]; however, a PCR >1 in a mid-day specimen likely indicates underlying kidney pathology.

Returning to case 1, on closer inspection of her urinalysis at the time of the test 2 months ago, specific gravity on the dipstick was noted to be 1.030 indicating a concentrated urine which can falsely elevate protein concentration. Further history was obtained and the patient endorsed a 5-mile run the morning of her previous clinic visit and urine testing. A repeat urinalysis was performed and was negative for proteinuria. Reassurance was offered. Further workup was not indicated. If the repeat urinalysis had demonstrated proteinuria, the next step would have been to order a urine protein to creatinine ratio to quantify the proteinuria.

Proteinuria Indicative of Intrinsic Kidney Disease: Nephrotic versus Nephritic Proteinuria

Glomerular diseases are typically classified as nephrotic or nephritic. Quantifying the amount of proteinuria in a 24-hour period or estimating it with a spot PCR is a useful first step in narrowing the differential diagnosis in suspected intrinsic renal disease. Proteinuria greater than 3.5 g in a 24-hour period is termed nephrotic. A smaller amount of proteinuria accompanied by hematuria in the form of atypical red blood cells

(RBCs) or RBC casts is termed nephritic. Most intrinsic renal diseases present with either a nephrotic or nephritic clinical picture; however, there may be overlap of the clinical manifestations. Patient demographics, presentation, and history can narrow the differential diagnosis which will help determine the next step in the workup. A definitive diagnosis may require a kidney biopsy. A nephrologist should be consulted in all cases where an intrinsic kidney pathology is suspected.

Case 2: Nephrotic Proteinuria

A 20 year old man presents with worsening lower extremity edema over the past month. Blood pressure is 120/80. He has no pertinent medical history and takes no medications. Review of systems reveals frothy urine.

Urine dipstick and urine microscopy	
Component	**Result**
Color	Yellow
Appearance	Cloudy
pH	6.0
Specific gravity	1.020
Blood	Negative
Glucose	Negative
Ketone	Negative
Protein	4+
Bilirubin	Negative
Urobilinogen	Negative
Leukocyte esterase	Negative
Nitrite	Negative
White blood cells (WBC)	0–5/hpf
Red blood cells (RBC)	0–2/hpf
Squamous epithelial cells	0/lpf
Casts	0/hpf
Bacteria	0/hpf

Nephrotic Syndrome

The diseases that more commonly present with nephrotic-range proteinuria typically involve damage to the filtering mechanism resulting in protein loss which exceeds the reabsorptive capacity of the tubular cells [13]. The epithelial foot processes (podocytes) may be disrupted (effaced) as in minimal change disease and focal segmental glomerulosclerosis (Fig. 5.4), or the integrity of the basement membrane can be disturbed as in membranous nephropathy or C3 glomerulonephritis. Regardless of the mechanism, the end result is a loss of protein through the urine.

The loss of protein (predominately albumin) results in a decrease in intravascular oncotic pressure contributing to edema. The liver increases production of cholesterol as a compensatory mechanism to maintain intravascular oncotic pressure which results in the hypercholesterolemia commonly seen in nephrotic syndrome. Other important circulating proteins are also lost in the urine, including immunoglobulins resulting in an increased infection risk as well as loss of coagulation factors such as anti-thrombin III resulting in an increased risk of thromboembolic events [13].

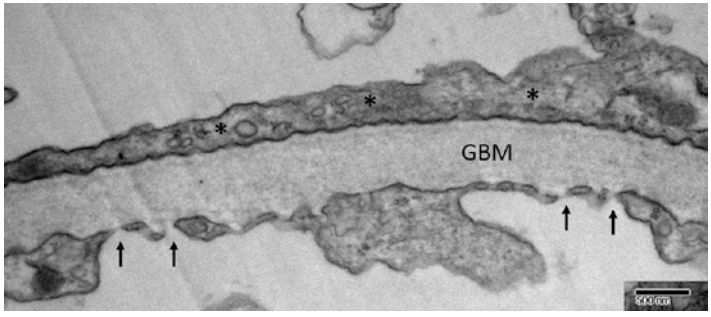

Figure 5.4 Transmission electron micrograph demonstrating complete podocyte foot process effacement. Arrows indicate the endothelial fenestrations of the renal capillaries, GBM is the glomerular basement membrane, and asterisks indicate podocyte foot process effacement. (×30,000). (Courtesy of Danni Holanda MD, University of Iowa, Department of Pathology)

The collective findings of proteinuria >3.5 g/day, hypoalbuminemia, pitting edema, and hypercholesterolemia comprise the classic nephrotic syndrome. Kidney function in terms of glomerular filtration is typically normal, at least initially, in nephrotic causes of proteinuria [14]. A thorough history should be obtained focusing on risk characteristics and classic associations to assist with additional testing considerations (Table 5.3). A definitive diagnosis may require a kidney biopsy. A nephrologist should be consulted in all patients with nephrotic range proteinuria.

Returning to case 2, this patient has heavy proteinuria with pitting edema in the absence of hematuria. Frothy urine is also consistent with heavy proteinuria. The first step is to quantify the amount of proteinuria with a PCR. A thorough history should be obtained for additional risk factors or associations which can help guide additional testing.

Case 3: Nephritic Proteinuria

A 20 year old man presents to clinic with cola-colored urine for 1 week. History is significant for a severe sore throat that he recovered from 2 weeks ago. BP is 170/100.

Urine dipstick and urine microscopy	
Component	**Result**
Color	Yellow
Appearance	Cloudy
pH	6.0
Specific gravity	1.020
Blood	3+
Glucose	Negative
Ketone	Negative
Protein	2+
Bilirubin	Negative
Urobilinogen	Negative

Urine dipstick and urine microscopy	
Component	**Result**
Leukocyte esterase	Negative
Nitrite	Negative
White blood cells (WBC)	0–5/hpf
Red blood cells (RBC)	10–15/hpf
Squamous epithelial cells	0/lpf
Casts	0/hpf
Bacteria	0/hpf

Nephritic Syndrome

The primary feature of nephritic glomerular disease is hematuria. Hematuria originating from the glomerulus is manifested by atypical or dysmorphic RBCs (acanthocytes) and/or RBC casts (see Chap. 10) seen during urine microscopy examination. Nephritic syndrome has less proteinuria (1.0–2.0 g/day) compared to nephrotic syndrome but is more likely than nephrotic syndrome to be accompanied by hypertension as well as altered kidney function in terms of oliguria (<500 mL/day total urine output in an adult) and an increase in serum creatinine. Nephritic causes of kidney disease can be typically classified as immune complex diseases, basement membrane diseases, or pauci-immune vasculitis diseases (Table 5.4).

The capillary beds of the kidneys are susceptible to immune complex deposition as immune complexes can either form directly on the glomerular basement membrane or be deposited there from systemically circulating immune complexes. Immune complexes after deposition can activate the classical complement cascade attracting inflammatory cells which in turn leads to cellular damage and inflammation with subsequent attraction of more inflammatory cells. This is the mechanism of damage in IgA nephropathy, membranoproliferative glomerulonephritis, post-infectious glomerulonephritis and lupus nephritis.

Table 5.3 Nephrotic syndrome: causes, associations and additional testing considerations

Nephrotic Syndrome
>3.5 grams of urine protein per day (nephrotic range proteinuria)
Low serum albumin (hypoalbuminemia)
Swelling (edema)
Increased cholesterol (hypercholesterolemia)

Disease	Typical population	Associations	Testing to consider
Minimal Change Disease (MCD)	Bimodal: #1 nephrotic disease in children, but also seen in adults	Hodgkin's lymphoma, NSAIDs, interferon alpha, dust or pollen allergies	CBC, medication and allergy review, kidney biopsy
Focal Segmental Glomerulosclerosis (FSGS)	African American and Hispanic adults	*APOL1* gene variant, HIV, sickle cell anemia, obesity	*APOL1* gene analysis, HIV, Hgb electrophoresis, kidney biopsy
Membranous Nephropathy (MGN)	Middle-aged Caucasian	PLA2R antibodies, SLE, DM, HBV, NSAIDs, occult tumors	PLA2R antibodies, ANA, dsDNA Ab, C3, C4, Hgb A1C, HBV, Ensure up-to-date cancer screening, kidney biopsy
Membranoproliferative glomerulonephritis (MPGN)	Children and adults (can present with nephritic picture as well)	HBV, HCV, HIV, SLE, CLL, non-Hodgkin's lymphoma, can present as nephritic syndrome: see Table 5.4	HBV, HCV, HIV, ANA, dsDNA Ab, C3, C4, CBC, kidney biopsy
C3 dominant glomerulonephritis (C3GN), Dense Deposit Disease (DDD)	Young adults	Complement mutations	C3, C4, C3 nephritic factor, serum factor H, genetic testing, kidney biopsy
Systemic diseases **Diabetes** **Amyloidosis** **Multiple Myeloma**	Children and adults		Hgb A1C, fat pad biopsy with Congo red stain, SPEP/IFE, UPEP/IFE, serum kappa:lambda ratio, kidney biopsy

ANA antinuclear antibody, *APOL1* apolipoprotein L1, *CBC* complete blood count, *C3* complement C3, *C4* complement C4, *CLL* chonic lymphocytic leukemia, *DM* diabetes mellitus, *dsDNA Ab* DNA double stranded antibody, *HBV* hepatitis B virus, *HCV* hepatitis C virus, *HIV* human immunodeficiency virus, *Hgb* hemoglobin, *NSAIDs* nonsteroidal anti-inflammatory drugs, *PLA2R* phospholipase A2 receptor, *SLE* systemic lupus erythematosus, *SPEP/IFE* serum protein electrophoresis/immunofixation, *UPEP/IFE* urine protein electrophoresis/immunofixation

Table 5.4 Nephritic syndrome: causes, associations and additional testing considerations

Nephritic syndrome
Glomerular hematuria (dysmorphic RBCs and/or RBC casts)
1.0–2.0 g/day proteinuria
Pitting edema
Oliguria
Increased creatinine
Hypertension is common

Disease		Suggestive associations and symptoms	Labs to consider
Immune complex diseases	IgA nephropathy (IgAN) (Berger disease)	• Any age, male predominance • Most common glomerular disease worldwide • Associated with: liver disease/cirrhosis, celiac disease, inflammatory bowel disease, GI tract cancer • In children with systemic manifestations (purpuric skin rash, arthralgias, GI symptoms), consider IgA vasculitis[a]	IgA serum level (neither sensitive, nor specific), liver function tests, albumin, INR, tTg Ab and endomysial Ab, ensure up-to-date GI cancer screening, kidney biopsy
	Membranoproliferative glomerulonephritis (MPGN)	• Any age • Can present as nephrotic syndrome: see Table 5.3 • Associated with: HBV, HCV, HIV, SLE, cryoglobulinemia	HBV, HCV, HIV, ANA, dsDNA Ab, C3, C4, CBC, cryoglobulins, kidney biopsy
	Post-infectious glomerulonephritis (PIGN)	• Any age, PSGN more common in children • Occurs 1-2 weeks after pharyngitis or 2-3 weeks after impetigo • Associated with: group A streptococci, high infection risk (substance abuse particularly IVDU) • Consider endocarditis	C3, ASO, rapid strep test and/or throat culture if symptoms, blood cultures, echocardiogram, kidney biopsy
	Lupus nephritis (LN)	• Any age, but typically between 15-45 years of age • Associated with: SLE (SLE has female predominance but LN occurs equally in men and women) • 30–50% of SLE patient will have renal involvement at presentation • More common in African Americans • Nephrotic range proteinuria possible	ANA, dsDNA Ab, C3, C4, kidney biopsy

Basement membrane diseases	Alport syndrome	• Type IV collagen genetic disease • X-linked is the predominant form of inheritance • Affected males typically have more severe disease • Associated with: sensorineural hearing loss, anterior lenticonus	Genetic testing
	Anti-GBM disease (Goodpasture disease)	• Type IV collagen antibody disease • Rare: 1 case per million per year • Slight male predominance • Associated with: alveolar hemorrhage • Causes rapid loss of kidney function	Anti-GBM antibodies, ANCA, kidney biopsy
Pauci-immune diseases (ANCA Vasculitis)	Granulomatosis with polyangiitis (GPA) (previously known as Wegener vasculitis)	• Any age, but mean age at diagnosis is mid-50s • Necrotizing granulomatous kidney disease • Associate with: frequent history of upper respiratory infections including sinusitis and rhinitis • Alveolar hemorrhage can occur	c-ANCA/PR3, kidney biopsy
	Eosinophilic granulomatosis with polyangiitis (EGPA) (previously known as Churg-Strauss vasculitis)	• Any age, but typically middle-aged individual with a history of new-onset or worsening asthma • Necrotizing granulomatous kidney disease • Associated with: peripheral eosinophilia and mononeuritis multiplex • 50% of patients have cardiac involvement (transient heart block, ventricular hypokinesis, myocarditis)	p-ANCA/MPO, CBC with diff, kidney biopsy
	Microscopic polyangiitis (MPA)	• Any age, but typically middle-aged white males and females • Different from GPA and EPGA in that MPA does not have necrotizing granulomas • Associated with: weight loss and skin lesions	p-ANCA/MPO, kidney biopsy

[a]Previously known as Henoch-Schoenlein Purpura

ANA antinuclear antibody, *ANCA* antineutrophil cytoplasmic antibody, *ASO* anti-streptolysin O, *C3* complement C_3, *C4* complement C_4, *CBC* complete blood count, *CBC w/ diff* complete blood count with differential, *dsDNA Ab* DNA double stranded antibody, *c-ANCA/PR3* cytoplasmic antineutrophil cytoplasmic antibodies/proteinase 3, *GBM* glomerular basement membrane, *GI* gastrointestinal, *HBV* hepatitis B virus, *HCV* hepatitis C virus, *HIV* human immunodeficiency virus, *IgA* immunoglobulin A, *INR* internal normalized ratio, *IVDU* intravenous drug use, *p-ANCA/MPO* perinuclear antineutrophil cytoplasmic antibodies/myeloperoxidase, *PSGN* post-streptococcal glomerulonephritis, *SLE* systemic lupus erythematosus, *tTg Ab* tissue transglutaminase antibody

In the past, renal disease was the most common cause of death of patients with lupus. Although this has been supplanted by heart disease, half of patients with lupus will develop kidney disease. Invariably, all patients with lupus nephritis will have some degree of proteinuria, a significant portion of whom are in the nephrotic range. Patients with lupus nephritis will require biopsy. Treatment and prognosis are based on the histologic findings. As might be expected, patients with nephrotic range proteinuria have significantly more foot process effacement [15].

The second subset of nephritic diseases involves damage to the glomerular basement membrane (GBM). Antibodies can be directed against the GBM and lead to kidney disease. Antibodies directed against the alpha 3 chain of type IV collagen is the underlying pathology of antiglomerular basement membrane (anti-GBM) disease. This antibody can also target the alpha 3 chain of type IV collagen in the alveolar basement membrane of the lung resulting in hemoptysis. Involvement of both kidney and lung in this antibody-mediated disease process is commonly referred to as Goodpasture's disease. Alport disease is a genetic defect or deficiency in one of the three alpha chains that form the type IV collagen matrix of the basement membrane.

The third subset of nephritic diseases is non-immune deposit vasculitis. The numerous blood vessels within the kidney make the kidney a susceptible target for vasculitis. Pauci-immune vasculitis affects the small vessels of the kidney. The absence or paucity of immune complex deposition in vessel walls helps distinguish this vasculitis from immune complex small vessel vasculitis. These vasculitic diseases are associated with the presence of antineutrophil cytoplasmic antibodies (ANCA).

It is important to obtain a detailed history for all the above nephritic conditions as extra-renal signs and symptoms can guide further workup [14]. A definitive diagnosis for a nephritic presentation will likely require a kidney biopsy. A nephrologist should be consulted in all patients suspected of nephritic disease.

Returning to case 3, this patient has proteinuria with significant microscopic hematuria consistent with nephritic syndrome. Red cell casts and dysmorphic red cells are diagnostic but uncommonly seen. IgA nephropathy is the most common cause of nephritic syndrome. Despite the recent pharyngitis in this young man, the advent of widespread antibiotics has made post-streptococcal glomerulonephritis rare. It should be noted that rapid Strep tests and throat culture are insensitive. Renal biopsy is the only definitive method for diagnosis of IgA nephropathy; however, if there are skin findings (palpable purpura), a biopsy of the skin may be taken instead, revealing IgA deposition.

When Is a Kidney Biopsy Indicated?

A kidney biopsy should be obtained in patients with unexplained sub-nephrotic range proteinuria, nephrotic range proteinuria, non-urologic hematuria without a known diagnosis, or for an unexplained progressive rise in serum creatinine. A kidney biopsy is also completed for prognostic purposes in certain kidney disorders. A nephrologist should always make the final determination if a kidney biopsy is indicated.

Minimal change disease is the most common cause of nephrotic kidney disease in children. As a general rule, children who present with nephrotic syndrome are usually not biopsied but are empirically treated for minimal change disease with high dose corticosteroids. If the disease is not steroid responsive or if a significant rise in creatinine occurs, a biopsy is considered as this presentation would be atypical for minimal change disease.

Diabetes mellitus is the number one cause of kidney disease in the United States. Proteinuria in patients with diabetes mellitus is very common so patients with long-standing diabetes are usually not biopsied for proteinuria alone unless the history or symptoms suggest some other underlying kidney disease. Another guide to assist with the determination of

whether to biopsy a patient with diabetes mellitus is the presence of retinopathy. Since 90% of patients with diabetic nephropathy also have diabetic retinopathy (the opposite is not true), the lack of diabetic retinopathy may be a clue that another process is at work.

Case 4: Proteinuria Screening Indications

A 45 year old woman presents to clinic for annual exam and management of diabetes. Medications include maximum dose of metformin and atorvastatin and an 81 mg per day of aspirin. Creatinine today is 1.0 mg/dL which is stable from prior. Hemoglobin A1C is 7.5%. Is there any additional testing which should be performed?

Patients Requiring Routine Screening for Proteinuria

The natural progression of type 1 diabetes mellitus kidney disease begins with hyperfiltration and hypertrophy. Morphologic lesions (GBM thickening, mesangial proliferation and hyalinosis of both the afferent and efferent arteriole) occur often, at first without signs of clinical kidney disease. Microalbuminuria results from continued progression of diabetic renal disease, frequently followed by overt nephropathy. The natural progression of type 2 diabetes mellitus kidney disease is the same as type 1; however, most type 2 patients are not identified until they have had the disease for several years.

Diabetes over time damages the glomerulus and leads to microalbuminuria followed by overt proteinuria. Proteinuria has been associated with kidney disease progression and increased cardiovascular disease risk. For this reason, annual screening for proteinuria is recommended to begin at the time of diagnosis in type 2 diabetes and 5 years after diagnosis of type 1 diabetes [16].

Proteinuria due to hypertension is thought to be at least partially due to increased intraglomerular pressure. Similar to

diabetic nephropathy, treatment targeting proteinuria reduction in hypertensive patients reduces the risk of kidney disease progression, cardiovascular events and all-cause mortality. For this reason, hypertensive patients should be screened yearly for proteinuria and if proteinuria is detected, treatment initiated [5].

Recall that the typical urine dipstick has a low limit of detection for albuminuria (69% sensitive for albuminuria in the range of 30–300 mg/g); therefore, screening of diabetic or hypertensive patients should begin with screening specifically for microalbuminuria and determination of a urine albumin:creatinine ratio (ACR). A normal ACR should be <30 mg/g reflective of <30 mg/day of albumin in the urine. Moderately increased albuminuria (formerly microalbuminuria) is by definition ACR of 30–300 mg/g. Severely increased albuminuria (formerly macroalbuminuria) is >300 mg/g and is essentially overt proteinuria that can usually be detected with a routine urine dipstick. Treatment is indicated for albuminuria at or above 30 mg/g.

Treatment of Proteinuria

Proteinuria detection should lead to initiation of medications that target the renin-angiotensin system such as angiotensin converting enzyme inhibitors (ACEi) or angiotensin receptor blockers (ARB) in order to reduce proteinuria and mitigate disease risk. There is no benefit to administering ACEi and ARB agents concurrently and doing so increases the risk of hypotension, syncope, hyperkalemia, and acute kidney injury [17]. Direct renin blockers (aliskiren) are rarely used in clinical practice because they are no more effective than ACEi or ARB agents and cannot be given concurrently with either of these agents. Direct renin blockers have also been associated with increased risk of stroke in diabetic patients. Treatment should be titrated to the maximum tolerated dose of either an ACEi or ARB while monitoring for hypotension, hyperkalemia and worsening kidney function [5]. An increase in serum creatinine or hyperkalemia may accompany initiation of

ACEi or ARB even at low doses; therefore, checking a basic metabolic profile (BMP) in 1–2 weeks after starting these medications is recommended.

If proteinuria persists despite maximum dose of ACEi or ARB therapy, a non-dihydropyridine calcium channel blocker (diltiazem or verapamil) can be added. These agents have been shown to reduce proteinuria as well; however, the evidence for risk reduction for kidney disease progression and cardiovascular events is not as strong [5]. Newer evidence supports the idea that use of sodium glucose co-transporter 2 (SGLT2) inhibitors such as empagliflozin in diabetics can also reduce proteinuria, slow progression of chronic kidney disease, and improve cardiovascular outcomes [18, 19]. Aldosterone antagonists such as spironolactone and eplerenone have also been found to reduce proteinuria; however these agents have an increased risk of hyperkalemia with no evidence for reduction in progression of kidney disease [20].

Returning to case 4, an in-office urine dipstick analysis was performed but was negative for proteinuria. Urine was subsequently sent for microalbumin analysis and revealed an ACR ratio of 45. Microalbuminuria as indicated by a urine albumin >30 mg/d and patient history of diabetes is an indication for starting an ACEi or ARB even if she is normotensive. A reasonable starting regimen would be lisinopril 5–10 mg per day. Creatinine and potassium should be checked within 1–2 weeks. Urine albumin and urine creatinine should be repeated at subsequent visits and ACEi (or ARB) increased as much as tolerated to minimize albuminuria/proteinuria.

Summary

Proteinuria occurs when there is a defect in the filtration barrier of the kidney. Proteinuria is usually detected initially on routine dipstick analysis. The next step after a positive dipstick for protein is to quantify the amount of protein present in the urine. This can be easily done by ordering a urine pro-

tein to urine creatinine ratio as this ratio correlates well with total 24-hour urine protein excretion. Performing a thorough history/physical examination and knowing how to appropriately order additional testing is important when considering nephrotic and nephritic glomerular disease. A nephrologist should be consulted if nephrotic or nephritic kidney disease is suspected. Patients with diabetes and hypertension are at increased risk to develop proteinuria so should undergo routine screening as persistent proteinuria increases risk for kidney disease progression and cardiovascular events. Screening may initially require specific testing for microalbuminuria given the detection limits of the routine urine dipstick at low levels of proteinuria. Microalbuminuria/proteinuria detection should lead to initiation of medications that target the renin-angiotensin system in order to reduce proteinuria and mitigate disease risk.

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Chapter 6
Urine Dipstick: Urinary Nitrites and Leukocyte Esterase – Dipping into Murky Waters

A. Ben Appenheimer and Bradley Ford

Objectives

1. Differentiate between asymptomatic bacteriuria and symptomatic urinary tract infections
2. Recognize the limitations of urine dipstick nitrite and leukocyte esterase results in assessing for urinary tract infections
3. Identify the causes of false positive and false negative nitrites and leukocyte esterase results

Definitions

Asymptomatic bacteriuria – The isolation of a determined quantitative number of bacteria in a urine speci-

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men obtained from a person without signs or symptoms of infection

Pyuria – Presence of white blood cells in the urine

Sterile pyuria – Presence of white blood cells in the urine in the absence of a positive urine culture

Overview

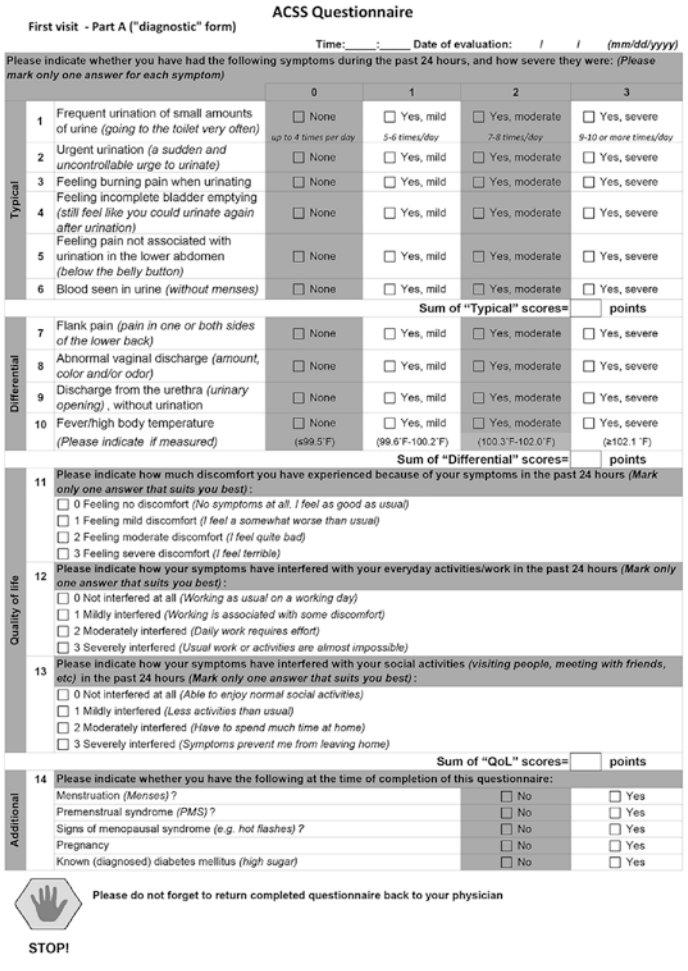
The urine dipstick is used in combination with other clinical and microbiologic data to diagnose urinary tract infections (UTIs). It is usually 'first-line' in diagnosis given the ease of testing, low cost, and ability to use as a point-of-care test. Two components of the dipstick that are utilized in assisting in the diagnosis of UTIs are leukocyte esterase and nitrites. Leukocyte esterase is an enzyme that is released by white blood cells (WBCs) and therefore correlates with pyuria. Some bacteria (such as *E. coli*) convert urinary nitrates that are routinely present in the urine into nitrites, therefore, a positive nitrite test on the urine dipstick is suggestive of the presence of these organisms. Importantly, a positive dipstick result only suggests the presence of bacteriuria and does not help in distinguishing between asymptomatic bacteriuria and symptomatic UTI. False positive and false negative dipstick tests are also common, and consideration should be given to the clinical context when interpreting results. When considering the diagnosis of a urinary tract infection, the dipstick result should therefore be used with careful interpretation of symptoms, urine microscopy, and urine culture along with consideration of other potentially confounding diagnoses such as sexually transmitted infections (STIs) and vaginosis.

Interpretation: Clinical Context

In women who present with a concern for an uncomplicated UTI and either one or more validated symptoms (dysuria, frequency, hematuria, back pain), one sign (costovertebral angle tenderness), or self-diagnosis based on personal experience, the probability of infection is about 50% [1]. In most such cases, this is enough certainty to prescribe antibiotics. A well-validated long-form questionnaire is available that increases the sensitivity and specificity of clinical diagnosis to a similar level as dipstick [2] (Fig. 6.1). Dipstick testing is therefore most useful in cases where clinical symptoms are unclear and where a rapid test may allow consideration of alternate diagnoses during the same visit (notably, STI and vaginosis). Various studies have supported that vaginal discharge or irritation makes vaginosis more likely, reducing the odds of presenting symptoms being due to UTI [1, 3, 4].

Interpretation of the dipstick in terms of positive and negative predictive value is dependent on prior likelihood of disease, so all patients should be screened for clinical signs and symptoms before laboratory testing. Asymptomatic bacteriuria, in particular, is a common non-infectious state where microbiological testing is usually unhelpful [5]. Definitions of common terms and differential findings are included in Table 6.1, illustrating the difficulty inherent to diagnosis of UTI versus related clinical syndromes.

Specimen collection is critical in preventing urine culture contamination, which affects about 1 in 5 urine cultures on average [6]. There are few modifiable risk factors for preventing urine culture contamination. These include instructing the patient as to proper clean catch technique, use of refrigeration or preservative tubes with formic and/or boric acid, cleansing with midstream collection, and using care not to do dipstick testing (which is a nonsterile process) on urine prior



ACSS Questionnaire

First visit - Part A ("diagnostic" form)

Time:_____:_____ Date of evaluation: / / *(mm/dd/yyyy)*

Please indicate whether you have had the following symptoms during the past 24 hours, and how severe they were: *(Please mark only one answer for each symptom)*

			0	1	2	3
Typical	1	Frequent urination of small amounts of urine *(going to the toilet very often)*	☐ None *up to 4 times per day*	☐ Yes, mild *5-6 times/day*	☐ Yes, moderate *7-8 times/day*	☐ Yes, severe *9-10 or more times/day*
	2	Urgent urination *(a sudden and uncontrollable urge to urinate)*	☐ None	☐ Yes, mild	☐ Yes, moderate	☐ Yes, severe
	3	Feeling burning pain when urinating	☐ None	☐ Yes, mild	☐ Yes, moderate	☐ Yes, severe
	4	Feeling incomplete bladder emptying *(still feel like you could urinate again after urination)*	☐ None	☐ Yes, mild	☐ Yes, moderate	☐ Yes, severe
	5	Feeling pain not associated with urination in the lower abdomen *(below the belly button)*	☐ None	☐ Yes, mild	☐ Yes, moderate	☐ Yes, severe
	6	Blood seen in urine *(without menses)*	☐ None	☐ Yes, mild	☐ Yes, moderate	☐ Yes, severe
					Sum of "Typical" scores=	**points**
Differential	7	Flank pain *(pain in one or both sides of the lower back)*	☐ None	☐ Yes, mild	☐ Yes, moderate	☐ Yes, severe
	8	Abnormal vaginal discharge *(amount, color and/or odor)*	☐ None	☐ Yes, mild	☐ Yes, moderate	☐ Yes, severe
	9	Discharge from the urethra *(urinary opening)* , without urination	☐ None	☐ Yes, mild	☐ Yes, moderate	☐ Yes, severe
	10	Fever/high body temperature *(Please indicate if measured)*	☐ None (≤99.5˚F)	☐ Yes, mild (99.6˚F-100.2˚F)	☐ Yes, moderate (100.3˚F-102.0˚F)	☐ Yes, severe (≥102.1 ˚F)
					Sum of "Differential" scores=	**points**

Quality of life	11	**Please indicate how much discomfort you have experienced because of your symptoms in the past 24 hours** ***(Mark only one answer that suits you best)*** **:** ☐ 0 Feeling no discomfort *(No symptoms at all. I feel as good as usual)* ☐ 1 Feeling mild discomfort *(I feel a somewhat worse than usual)* ☐ 2 Feeling moderate discomfort *(I feel quite bad)* ☐ 3 Feeling severe discomfort *(I feel terrible)*
	12	**Please indicate how your symptoms have interfered with your everyday activities/work in the past 24 hours** ***(Mark only one answer that suits you best)*** **:** ☐ 0 Not interfered at all *(Working as usual on a working day)* ☐ 1 Mildly interfered *(Working is associated with some discomfort)* ☐ 2 Moderately interfered *(Daily work requires effort)* ☐ 3 Severely interfered *(Usual work or activities are almost impossible)*
	13	**Please indicate how your symptoms have interfered with your social activities** ***(visiting people, meeting with friends, etc)*** **in the past 24 hours** ***(Mark only one answer that suits you best)*** **:** ☐ 0 Not interfered at all *(Able to enjoy normal social activities)* ☐ 1 Mildly interfered *(Less activities than usual)* ☐ 2 Moderately interfered *(Have to spend much time at home)* ☐ 3 Severely interfered *(Symptoms prevent me from leaving home)*
		Sum of "QoL" scores= **points**

Additional	14	**Please indicate whether you have the following at the time of completion of this questionnaire:**		
		Menstruation *(Menses)* ?	☐ No	☐ Yes
		Premenstrual syndrome *(PMS)* ?	☐ No	☐ Yes
		Signs of menopausal syndrome *(e.g. hot flashes)* ?	☐ No	☐ Yes
		Pregnancy	☐ No	☐ Yes
		Known (diagnosed) diabetes mellitus *(high sugar)*	☐ No	☐ Yes

Please do not forget to return completed questionnaire back to your physician

STOP!

FIGURE 6.1 The Acute Cystitis Symptom Score (ACSS) tool, which (with a threshold of 6 points or greater) demonstrated 94% sensitivity and 90% specificity in a population of women who presented with candidate uncomplicated cystitis. (From Alidjanov JF, et al. [2]. Reprinted with permission of the copyright holders)

Table 6.1 Definitions of terms in this chapter, with expected clinical and laboratory findings

	Clinical findings	Laboratory findings			Clinical pearls
		Urine LE and WBCs + and –	Urine nitrite + and –	Urine culture	
Urinary tract infection (UTI)	Symptomatic; cystitis or pyelonephritis; complicated or uncomplicated	+	+[a]	≥10^5 colony-forming units [CFU]/mL of uropathogen	Nitrite often false negative False-negative culture can occur with antibiotics
Sterile pyuria	Variable	+	±	–	Occurs with: False negative culture Contamination False positive LE Atypical organism STI Primary kidney disease, nephrolithiasis, tumor
Asymptomatic bacteriuria	Asymptomatic	±	±	≥10^5 colony-forming units [CFU]/mL of uropathogen	Only treat in pregnancy or prior to certain urological procedures

(continued)

TABLE 6.1 (continued)

	Clinical findings	Laboratory findings			Clinical pearls
		Urine LE and WBCs + and –	Urine nitrite + and –	Urine culture	
Sexually transmitted infection (STI)	Asymptomatic, mimicking UTI, or with specific findings (e.g. ulcerations)	±	–	–	Test for specific pathogens: Gonorrhea, Chlamydia, Trichomonas, Herpes simplex most common
Vaginosis/ vaginitis	Asymptomatic or symptomatic with thin white vaginal discharge, fishy odor	±	–	–	Specific testing available (whiff-amine test, Gram stain with Nugent score, tests for Candida, Gardnerella and others)

[a]With Gram-negative organisms

to culturing the same sample [6, 7]. See Chap. 3 for detailed instructions for clean-catch urine collection. In children, urine collected on cotton balls or in diapers is considered unacceptable whereas bagged urine is superior but still low quality. In very young children, the rate of false-positive urine cultures from bagged urine nears 100% [8]. Urine obtained by straight catheter is the best commonly feasible sample and suprapubic aspiration, while rarely utilized, remains the gold standard [9, 10]. See Chap. 13 for more details on pediatric urine collection. Of note, urinalysis and microscopy cannot be done from preservative tubes. A common arrangement is therefore to follow best practices in collecting a urine sample into a cup, then transferring urine for culture into a preservative tube. Urine in the cup is then analyzed by dipstick and microscopy separately from the preserved urine, which prevents cross-contamination.

Case 1: Asymptomatic Bacteriuria

You are admitting an 82 year old female from the emergency department for failure to thrive and a mild acute kidney injury. As part of her workup in the emergency department a urinalysis was sent which showed 3+ leukocyte esterase and positive nitrites. Her urine culture eventually turns positive for E. coli. She denies any dysuria, polyuria, increased urinary urgency, suprapubic pain, or flank pain. How should you manage this bacteriuria?

This woman has asymptomatic bacteriuria, a common finding in women and older men. This is especially true in elderly women, where one study showed that 37% of women over the age of 80 had at least one episode of asymptomatic bacteriuria when tested at baseline, 6 months, and 18 months [11]. Aside from a few specific patient populations, such as pregnant women or those undergoing invasive urologic procedures, asymptomatic bacteriuria should not be treated. Neither dipstick testing nor urine microscopy can reliably distinguish between urinary tract infections and asymptomatic bacteriuia, making this purely a clinical distinction.

Returning to case 1, you correctly diagnose this patient as having asymptomatic bacteriuria and decide not to treat. She does not have any signs or symptoms associated with UTIs throughout the hospitalization and her kidney injury improves with hydration.

Case 2: Use of Urine Nitrites in Clinical Diagnosis

A 27 year old female presents to your clinic complaining of dysuria and increased urinary urgency. You order a urine dipstick for further evaluation for a urinary tract infection. This comes back with 2+ leukocyte esterase and positive nitrites. Based on this result, could you predict whether the causative organism would be a gram-negative rod or a gram-positive coccus?

Nitrates are excreted in urine and are reduced to nitrites through the action of the enzyme nitrate reductase. Enterobacteriaceae (Gram-negative uropathogens such as *E. coli*, *Klebsiella* and *Proteus*) produce nitrate reductase, making nitrite a potentially good test for detecting the presence of most typical uropathogens (Table 6.2).

In the nitrite test, nitrite is converted to a diazonium salt that is further coupled with a chromogen that generates color (Fig. 6.2). This is read as a positive test either by eye or by an optical test reader.

Interpretation of nitrite testing is complicated by several factors. Analytical false positive results are rare and contamination with Enterobacteriaceae (i.e., stool flora) is the primary source of spurious positive nitrite results. False positives from exposure of dipsticks to air [17] or colored substances such as phenazopyridine (Fig. 6.2) are possible but not well supported by evidence. Asymptomatic bacteriuria, defined as diagnostic levels of a typical uropathogen in the absence of a symptomatic infection, may return a positive nitrite result as

Table 6.2 Potential uropathogens with nitrate reductase and their relative prevalence in uncomplicated UTIs in adults

Organism	**%**
Escherichia coli	75
Klebsiella spp.	6
Proteus spp.	2
Citrobacter spp.	<1
Enterobacter spp.	<1
Morganella spp.	<1
Providencia spp.	<1
Serratia spp.	<1
Shigella spp.	<1

nitrites do not distinguish between asymptomatic bacteriuria and UTI. False negative results are common, usually due to the weak activity of nitrate reductase. As this conversion takes up to 4 hours [10, 11], a bladder incubation time of less than 4 hours significantly affects the nitrite result [12]. This makes a long dwell time of urine in the bladder essential, requiring submission of a first-morning urine. However, this requirement is rarely met in practice. In one well-controlled comprehensive study of the performance of dipsticks for the diagnosis of a UTI, patients were instructed to submit first-void urines, but only 5% managed to comply even in a controlled setting [13, 14].

Along these lines, "lack of dietary nitrate" (often implying lack of vegetable intake) is often cited as a potential reason for false-negative urine nitrite testing. This idea dates to the original description of nitrite testing in dipstick form for the diagnosis of UTI [13, 14]. Nitrates and nitrites are interconverted in vivo, and both are widely used as texture enhancers and preservatives of meats, cheese and beverages, and occur as a normal component of drink-

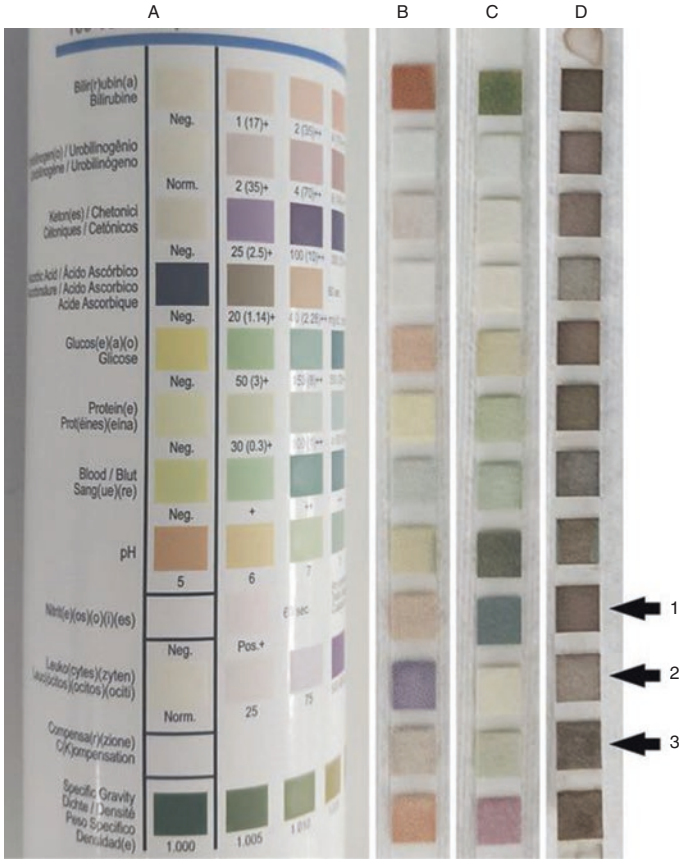


FIGURE 6.2 Dipstick tests commonly come with colored indices for manual interpretation, often on the container itself (**A**). Dipstick (**B**) has positive nitrite [B1] and leukocyte esterase (LE) [B2]. Dipstick (**C**) has negative LE [C2] and a green false-positive nitrite [C1] due to deliberate overexposure to air. Dipstick (**D**) is uniformly false positive due to blood, a state that is obvious to automated readers that check the blank "compensation" row [3] for intrinsic urine color

ing water. In clinical studies, reduction in dietary nitrate requires controlled feeding with exclusion of many common foods [16]; "lack of dietary nitrate" therefore remains ill-defined in practical terms and is not screened for prior to dipstick testing.

Many studies have evaluated the test characteristics of nitrites in predicting UTIs in patients. While the test performance varies somewhat by patient population [15], the test performs similarly in most populations, including men [16]. In general, the presence of nitrites has a very high specificity, up to 98%, [15–17] meaning the presence of nitrites alone can be used to rule in a urinary tract infection in the presence of symptoms [18]. While nitrites have a high specificity, their sensitivity is low, ranging from 45% to 60% [15]. Since nitrites are only formed by Enterobacteriaceae, they will be negative if the urinary tract infection is caused by an organism outside of this family, such as *Staphylococcus saprophyticus*, which may cause up to 11% of UTIs in young, sexually active women [19]. Other common atypical pathogens such as *Pseudomonas* (75% of which are nitrate positive), *Streptococcus agalactiae* ("Group B Strep"), *Staphylococcus aureus*, *Corynebacterium urealyticum*, and *Aerococcus spp.* [20] are likely to be negative by the dipstick nitrite test as well [21]. Other potential causes of false negative nitrite tests include dilution of urine through administration of fluids and/or oral intake prior to providing the sample, recent urination, extraordinary cases of low urinary excretion of nitrates due to reduced dietary intake [22], and a decreased urinary pH [23].

Returning to case 2, based on the positive nitrite you presume that this UTI is caused by a Gram-negative rod in the Enterobacteriaceae family. The patient is started on nitrofurantoin and urine culture grows Klebsiella. Her dysuria and urinary urgency improve and she completes her course of therapy.

Case 3: Use of Urine Leukocyte Esterase in Clinical Diagnosis

You are evaluating a 72 year old female for neutropenic fever with history of chronic lymphocytic leukemia (CLL) currently receiving bendamustine and rituximab. Her absolute neutrophil count (ANC) is 90 cells/mm^3, and her fevers have been as high as 39 °C. She is started on empiric cefepime and her initial workup includes a chest x-ray, UA, and blood cultures. Her CXR was normal, UA showed negative nitrite and negative leukocyte esterase, and urine microscopy showed 3 WBCs. Blood cultures remain no growth. She does complain of some mild dysuria. Since her UA and microscopy were negative this was not reflexed to culture. Would you be comfortable that a urinary tract infection had been ruled out? Why or why not?

Leukocyte esterase is an enzyme derived from neutrophils. Therefore, this is a test designed to indirectly detect the presence of neutrophils in the urine and can be falsely negative in patients with neutropenia. In this test, an indoxyl ester is cleaved by neutrophil esterase, and the resulting indoxyl is reacted to form a colored dye. The test therefore does not distinguish between intact and lysed neutrophils.

Pyuria is classically defined by quantitative microscopy. Cutoffs for a "normal" number of WBCs per high-power field or cubic millimeter on microscopy most commonly range from less than or equal to 5 [24] or 10 [25]. Dipstick leukocyte esterase is able to detect about 10 leukocytes per cubic millimeter at its lower limit of detection ("trace" positivity; Fig. 6.2) [26], making any degree of dipstick positivity "abnormal". Beyond this concept, there is no standard for a degree of leukocyte esterase positivity (or WBC count) that optimally defines UTI.

As a stand-alone test, leukocyte esterase has less utility than nitrites given relatively low and variable sensitivity and specificity. In a meta-analysis the sensitivity of this test varied from 48% to 86%, and specificity varied from 17% to 93% depending on several factors including the setting, level of care, and population [15] (Tables 6.3 and 6.4). In a study

Table 6.3 Sensitivity and specificity of leukocyte esterase testing for the diagnosis of urinary tract infections

Study	Population	Sensitivity (%)	Specificity (%)
Deville et al. (2004) [17]	All (meta-analysis)	48–86	17–93
Koeijers et al. (2007) [18]	Men	78	59

Table 6.4 Subgroup analyses for the sensitivity and specificity of leukocyte esterase in the diagnosis of urinary tract infections or bacteriuria

Study characteristic	N	Sensitivity (95%CI)	Specificity (95%CI)
Population			
Non-urological	33	0.62 (0.54–0.71)	0.70 (0.60–0.81)
Urological	2	0.86 (0.68–1.00)	0.93 (0.81–1.00)
Non-urological studies			
Bacteriuria	8	0.56 (0.38–0.82)	0.61 (0.41–0.90)
UTI	25	0.64 (0.56–0.74)	0.73 (0.63–0.85)
Setting			
Family physician	6	0.87 (0.83–0.92)	0.36 (0.21–0.64)
Outpatient	12	0.50 (0.35–0.68)	0.80 (0.72–0.88)
Emergency	3	0.56 (0.41–0.75)	0.86 (0.74–0.99)
Inpatient	11	0.66 (0.60–0.73)	0.81 (0.74–0.88)
Level of care			
Community	1	0.63 (0.08–1.00)	0.85 (0.82–0.88)
Primary care	10	0.76 (0.60–0.98)	0.46 (0.32–0.68)

(continued)

Table 6.4 (continued)

Study characteristic	N	Sensitivity (95%CI)	Specificity (95%CI)
Secondary care	8	0.48 (0.33–0.71)	0.83 (0.73–0.93)
Tertiary care	14	0.62 (055–0.70)	0.84 (0.80–0.89)
Reader of test			
Family physician	2	0.86 (0.71–1.00)	0.17 (0.04–0.62)
Nurse	7	0.67 (0.58–0.79)	0.65 (0.37–1.00)
Clinician	2	0.34 (0.18–0.64)	0.90 (0.68–1.00)
Lab worker	17	0.59 (0.47–0.73)	0.81 (0.76–0.87)

(Modified with permission from [17])

evaluating UTIs in men, the sensitivity was 78%, the specificity was 59%, and the results of the leukocyte esterase test did not significantly alter diagnostic accuracy of nitrite alone [16] (Table 6.3). Positive results for leukocyte esterase can occur with contamination of the specimen, vaginosis, and STIs (Table 6.1). Because most UTIs are in women and leukocyte esterase is a component of vaginal secretions, contamination with vaginal secretions is probably the most common generator of false positive leukocyte esterase results. Tomas and colleagues [27] performed a study illustrating the extent of this problem in practice, in which emergency department providers were allowed to practice as usual while a comprehensive microbiological workup was performed in the background. In this study, 92% of 264 symptomatic women had a positive urinalysis (primarily leukocyte esterase) which led physicians to give a presumptive diagnosis of UTI in 66% of the patients. However, only 48% of those diagnosed with a presumptive UTI had a positive urine culture, demonstrating an overtreatment of presumptive UTIs when using leukocyte esterase as a marker. In addition, 15% of all patients with vaginosis and 40% of STI cases were misdiagnosed as UTIs.

Other causes of sterile pyuria include chronic interstitial nephritis, nephrolithiasis, uroepithelial tumors, intra-abdominal inflammatory processes adjacent to the bladder, and the presence of atypical organisms such as *Mycoplasma*, *Ureaplasma,* or tuberculosis (see Chap. 8). False negatives can occur secondary to a dilute sample, low-level bacteriuria that does not meet the technical criteria for a UTI, male gender, neutropenia, or catheter-associated infections [28].

Returning to case 3, you notice that a reflex urine culture was not performed because of the negative urinalysis. However, since she is neutropenic you know that leukocyte esterase and urine microscopy could both be falsely negative as these require neutrophils in the urine. You ask for a urine culture even though the UA and micro were negative and this eventually grows Pseudomonas. You decide to continue to treat with cefepime until the susceptibilities are available.

Additional Approaches to Interpreting Urine Dipsticks

Given the low sensitivity of these tests independently and the low specificity of LE, studies have also looked at combining the tests to improve these test characteristics. Considering either a positive nitrite or leukocyte esterase as a positive test, sensitivity increases up to 68–88% [15, 17, 27]. Studies have varied on the recommendations for the interpretation of a dipstick that is negative for both nitrite and leukocyte esterase, with some saying it is sufficient to rule out infection [15, 17] while others suggest further testing is needed [16, 29, 30]. In reality, the negative predictive value of dipstick testing depends on the pre-test probability of infection.

Given the limitations discussed above, more recent studies have evaluated various algorithmic approaches which include some combination of dipstick findings, clinical features, and urine culture, with variable sensitivities and specificities [2, 18, 31]. One study evaluated two different algorithms, one

of which was based purely on dipstick results and the other based purely on clinical factors. Using the 'dipstick rule', a positive test was one with *either* a positive nitrite *or* both positive blood and leukocyte esterase. This yielded a sensitivity of 75–77% and a specificity of 66–70% [30, 32]. Given their rate of positive cultures, this correlated with a positive predictive value of 81% and a negative predictive value of 57–65%, performing better than a rule based solely on clinical factors that required two or more of the following: moderately severe dysuria, moderately severe nocturia, offensive urine smell, or cloudy urine.

Summary

When interpreting urine dipsticks, a positive nitrite value is highly suggestive of the presence of Gram-negative bacteria in the urine. However, the role of leukocyte esterase remains less clear. In the setting of a low pre-test probability for infection, the presence of bacteria in the urine is unlikely if both nitrites and leukocyte esterase are negative. The results of a dipstick only suggest the presence of bacteria and in isolation should not have any bearing on distinguishing between symptomatic UTI, asymptomatic bacteriuria, and alternate diagnoses.

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Chapter 7
Urine Dipstick: An Approach to Glucosuria, Ketonuria, pH, Specific Gravity, Bilirubin and Urobilinogen – Undeniable Chemistry

Puja T. Pape, Victoria J. A. Sharp, and Jessica Rockafellow

Objectives

1. Recognize common pathological conditions that lead to the presence of glucose and ketones in the urine
2. Gain familiarity with common causes of false positive and false negative results seen in urine glucose and ketone dipstick results

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3. Identify systemic illnesses that may be associated with acidic or alkaline urine and distinguish these from benign causes of shifts in urine pH
4. Learn to apply urine specific gravity measurements to clinical scenarios
5. Describe association of pre-, intrinsic, and post-hepatic jaundice with urobilinogen and urine bilirubin

Overview

Dipstick urinalysis for glucose, ketones, pH, specific gravity, bilirubin and urobilinogen is an important tool available to clinicians for screening and diagnosis of several systemic disorders. This chapter will provide an in-depth look at the chemical reactions behind these tests, the clinical significance of a positive or negative result, and suggested next steps in evaluation and management. Reasons for false positive and false negative results will also be discussed.

Case 1: Urine Dipstick Positive for Glucose

A 45 year old obese female presents to her primary care provider complaining of being thirsty and urinating a lot, especially at night.

Urine dipstick and urine microscopy	
Component	**Result**
Color	Yellow
Appearance	Clear
pH	6.0
Specific gravity	1.015
Blood	Negative
Glucose	2+
Ketone	Negative

Urine dipstick and urine microscopy	
Component	**Result**
Protein	Negative
Bilirubin	Negative
Urobilinogen	Negative
Leukocyte esterase	Negative
Nitrite	Negative
White blood cells (WBC)	0–5/hpf
Red blood cells (RBC)	0–2/hpf
Squamous epithelial cells	2/lpf
Casts	0/hpf
Bacteria	0/hpf

Glucosuria is the excretion of detectable amounts of glucose in urine. In an otherwise healthy individual, nearly all glucose is reabsorbed at the proximal convoluted tubule of the kidney through active transport. In pathologic states, as seen in diabetes mellitus, the threshold of reabsorption is overwhelmed and glucose will be detectable in urine [1]. This occurs in states of systemic hyperglycemia at glucose levels greater than or equal to 160 mg/dL [2].

Reagent test strips are specific for glucose and do not detect other types of sugars [3, 4]. The chemical reaction requires the oxidation of glucose by the enzyme glucose oxidase to produce gluconic acid and hydrogen peroxide (H_2O_2). Hydrogen peroxide reacts with chromogen, which is facilitated by peroxidase enzyme, and causes a color reaction that is indicated on the test strip (Fig. 7.1).

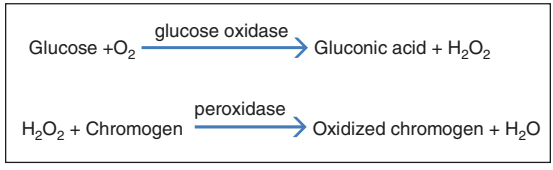

Figure 7.1 Chemical reaction of urine dipstick for glucose [5]

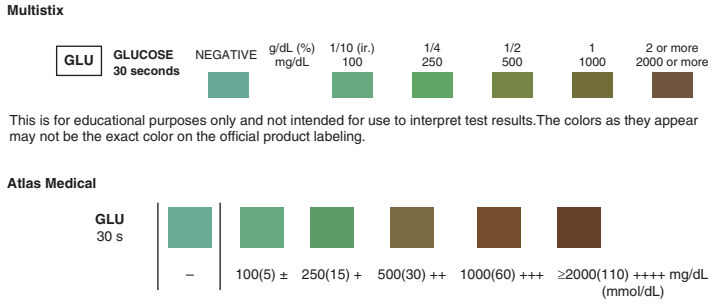

FIGURE 7.2 Urine glucose dipstick results. Sources: Siemens Healthcare Diagnostics Inc and Atlas Medical. Used with permission

The degree of oxidation of chromagen determines the final color on the urine dipstick with the result ranging from green to brown. A qualitative positive is detected faster (as soon as 10 seconds) than quantitative positive (detectable at 30 seconds) [4]. The color change seen may vary by different brands of reagent test strips (Fig. 7.2). Sensitivity of the test depends on different manufacturers but can be detected at concentrations as low as 75–125 mg/dL with test strips such as Multistix [6]. The results are typically reported as a range from negative to 3+, with a higher positive number indicating a higher glucose concentration in the urine. A urine dipstick result of 1+ glucose commonly indicates a urine glucose concentration of greater than 250 mg/dL, while 2+ glucose indicates a glucose greater than 500 mg/dL and 3+ glucose indicates a glucose level of greater than 1000 mg/dL [7].

What Is the Differential Diagnosis for Glucosuria?

Urine glucose testing, similar to many of the other components of dipstick urinalysis, is interpreted depending on timing of the sample. The first step in evaluation of glucosuria is typically to distinguish if the patient is hyperglycemic or normoglycemic. In healthy patients, glucosuria can be present after a high glucose content meal. Historically, fasting urine and blood glucose have been used as screening tests for dia-

betes mellitus. It is important to note that a fasting morning urine sample can contain glucose from a patient's evening meal if they have not yet voided overnight or the morning of testing. For this reason, a second sample should be obtained for the most accurate result [1]. Glucosuria can be associated with systemic hyperglycemia, such as in cases of diabetes, steroid induced hyperglycemia, or Cushing's disease. The most common cause of glucosuria is diabetes mellitus.

Patients with type II diabetes are often asymptomatic. If symptomatic, classic symptoms include polyuria, polydipsia, nocturia, blurred vision, fatigue and occasionally weight gain or loss. Glucosuria can also be seen in pregnancy with gestational diabetes.

Non-diabetic causes of glucosuria include pancreatitis, pancreatic cancer, acromegaly, Cushing syndrome, hyperthyroidism, pheochromocytoma, CNS damage or stress. This primarily occurs as a result of hormone-mediated production of glucose through the breakdown of glycogen and inhibition of insulin secretion [1]. Glucosuria with normal serum glucose is associated with renal disorders due to poor reabsorption of glucose at the renal tubules, such as in Fanconi syndrome, heavy metal poisoning, and in pregnancy at levels below the cut off for the diagnosis of gestational diabetes [1, 2]. Selective sodium glucose co-transporter 2 (SGLT2) inhibitors have been used clinically to improve glycemic control in diabetics in an insulin independent manner by blocking proximal tubular glucose reabsorption, thereby resulting in glucosuria.

The sensitivity of urine glucose testing is dependent on the specific gravity of urine, with sensitivity improving at a lower specific gravity [3]. At a higher specific gravity, a urine glucose will be detected with a higher rate of false positives. Low temperatures decrease sensitivity of the test [1]. As with several other urine dipstick tests, improper storage of urine and test strips can cause either false negatives or false positives [2]. Table 7.1 shows sources of false positive and false negative results that can interfere with interpretation of urine glucose testing.

Table 7.1 Causes of false positive and false negative results on glucose reagent strip

False positive	False negative
Improper storage of sample or test strips [2], presence of oxidizing agents in urine [2, 3], levodopa [1, 8], ketones in urine [1, 8]	Improper storage of sample or test strips [2], high concentration of uric acid or vitamin C [1, 8], tetracyclines [3]

Returning to case 1, the patient had several symptoms of hyperglycemia. The next step in her evaluation would be a fasting serum glucose level. Her fasting level was 140 mg/dL (>126 mg/dL) and her hemoglobin A1c was 7.3 (> 6.5), meeting the criteria for diabetes mellitus. Treatment was initiated with lifestyle modifications (diet, exercise, weight reduction) and additional pharmacologic therapy will be added if needed.

Case 2: Urine Dipstick Positive for Ketones

An 11 year old with type I diabetes mellitus presents to your clinic with his parents for chief concern of vomiting. They report he has been vomiting for the past two days and has been unable to keep solid food down.

Urine dipstick and urine microscopy	
Component	**Result**
Color	Yellow
Appearance	Clear
pH	6.0
Specific gravity	1.020
Blood	Negative
Glucose	2+
Ketone	2+
Protein	Negative

Urine dipstick and urine microscopy	
Component	**Result**
Bilirubin	Negative
Urobilinogen	Negative
Leukocyte esterase	Negative
Nitrite	Negative
White blood cells (WBC)	0–5/hpf
Red blood cells (RBC)	0–2/hpf
Squamous epithelial cells	0/lpf
Casts	0/hpf
Bacteria	0/hpf

Ketonuria is the presence of ketones in urine. In normal circumstances, carbohydrates are broken down into glucose and used as the body's primary source of energy. When carbohydrates are not available, fat serves as an alternative source of energy. Fat is then broken down into carbon dioxide and water and ketones are not detectable in urine; however, with incomplete fat metabolism, ketone bodies are excreted into the urine. The primary ketone products present are acetone, acetoacetic acid (acetoacetate) and beta-hydroxybutyric acid [1, 2]. In clinical practice, the fruity smell of a patient's breath with ketosis represents the presence of acetone, while acetoacetate can be detected in the urine and beta hydroxybutyrate can be measured in the serum.

The chemical reaction for urine detection of ketones occurs when ketones (mainly acetoacetate), which are not normally present in urine, react with nitroprusside and glycine (Fig. 7.3). This reaction produces a color change of dark pink to purple with a positive result [4]. Of note, urine dip-

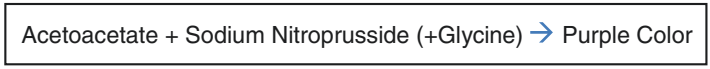

Figure 7.3 Chemical reaction of urine dipstick for ketones [5]

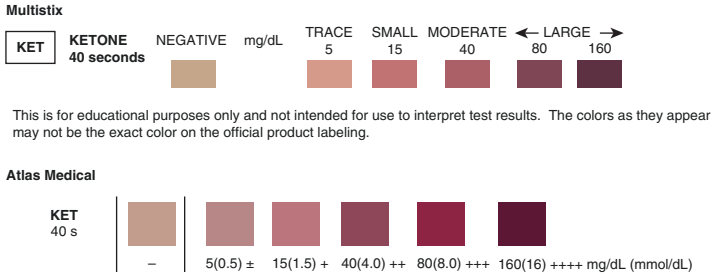

FIGURE 7.4 Urine dipstick for ketones results. Sources: Siemens Healthcare Diagnostics Inc and Atlas Medical. Used with permission

stick tests detect the presence of acetoacetic acid, but do not detect acetone or beta-hydroxybutyric acid.

Ketones are detectable in urine before they are detectable in serum [6]. Ketones can be detected at a sensitivity as low as 5–10 mg/dL of acetoacetic acid [6]. As with glucosuria, ketones are detected on urine dipstick from a scale of negative to 4+, with higher degrees of positivity reflecting a higher ketone concentration in urine [7] (Fig. 7.4).

What Is the Differential Diagnosis for Ketonuria?

The presence of ketones is indicative of a variety of conditions associated with increased fat metabolism. The presence of ketones occurs with diabetic ketoacidosis and can reflect an insulin deficiency [1, 2]. This is one of the most common causes of ketonuria. Additional common conditions that should be in the differential for ketonuria include starvation ketosis, alcoholic ketosis or a low carbohydrate diet. Interestingly, the ketogenic diet has become a fad diet for weight loss and the urine is commonly monitored for the presence of ketones. Carbohydrate intake is strictly limited, which causes the body to rely on fat metabolism for its primary energy source. Ketonuria also occurs with acute febrile illnesses or vomiting [1, 2, 8]. Ketones may be present after

strenuous exercise or with certain inborn errors of amino acid metabolism [1]. Table 7.2 provides a summary of common conditions that may cause positive ketones and suggested further supporting laboratory evaluation.

Ketonuria can be falsely detected with use of medications that have L-dopa metabolites, such as captopril, levodopa, or methyldopa [2, 3]. Large amounts of dyes, particularly red dye or phthalein dyes, can cause false positives [1–3]. Large amounts of phenylketone in urine produce a false positive result [2, 3]. False negatives, as with several other urine dipstick tests, occur without prompt evaluation of the sample [1, 2]. This occurs due to the conversion of acetoacetic acid to acetone [3]. There are several causes of false positive and false negative results on urine reagent strips for ketones (Table 7.3).

Table 7.2 Conditions associated with positive ketones on urinalysis

Potential etiology of ketonuria	Suggested further testing to support diagnosis
Insulin deficiency (DKA) [1, 2]	Serum glucose, serum electrolytes, beta hydroxybutyrate, anion gap
Starvation ketosis (anorexia, low carbohydrate diet, acute febrile illness or vomiting) [1, 2, 8]	Serum electrolytes, prealbumin
Alcoholic ketoacidosis	Serum ethanol level, osmolar gap

Table 7.3 Causes of false positive and false negative results on urine ketone reagent strip

False positive	False negative
Use of certain Medications (captopril, levodopa, methyldopa) [2, 3], presence of certain dyes [1–3], presence of large amounts of phenylketones [2, 3]	Delay in interpretation of sample (conversion of acetoacetate to acetone) [1, 2]

In patients with type I diabetes mellitus, the presence of urine ketones is an early indicator of an insulin deficiency. When untreated, this can progress to volume depletion, electrolyte imbalances, diabetic ketoacidosis and ultimately diabetic coma and death if untreated [1]. This makes urine ketone testing an important clinical tool in the management of type I diabetics.

Returning to case 2, in addition to ketonuria, a serum glucose is noted to be 350 with an elevated beta-hydroxybutyric acid of 2.0 mmol/L (normal range 0.4–0.5 mmol/L, diabetic ketoacidosis range 1.5–3.5 mmol/L) [5]. *The diagnosis of diabetic ketoacidosis is confirmed and the patient is sent to the ED for further management with insulin and IV fluids.*

Case 3: Urine Dipstick with Low Urine pH

A 45 year old male with history of gout presents to ED in Tucson, AZ with severe right flank pain, low grade fever, nausea and vomiting.

Urine dipstick and urine microscopy	
Component	**Result**
Color	Yellow
Appearance	Cloudy
pH	5.0
Specific gravity	1.010
Blood	1+
Glucose	Negative
Ketone	Negative
Protein	Negative
Bilirubin	Negative
Urobilinogen	Negative

Urine dipstick and urine microscopy	
Component	**Result**
Leukocyte esterase	Negative
Nitrite	Negative
White blood cells (WBC)	0–5/hpf
Red blood cells (RBC)	0–2/hpf
Squamous epithelial cells	0/lpf
Casts	0/hpf
Bacteria	0/hpf

The urine pH is routinely included in most basic dipstick urinalysis testing. In general, a urine pH of 7 is considered neutral, with a urine pH of <7 indicating acidic urine and a urine pH of >7 indicating basic urine. On average, urine tends to be acidic (pH 6), although the range of pH varies from 5 to 8 [2]. Given the variety of factors that can affect urine pH that must be taken into account when interpreting the test, there are no set normal parameters for testing [1]. This wide range of values can be suggestive of physiologic changes related to diet, but also can reflect pathologic states. The urine pH is reported using a broad range of colors based on a double indicator system, to include the entire range of possible values [4]. The pH is commonly sorted into increments of 0.5 based on color [7] (Fig. 7.5).

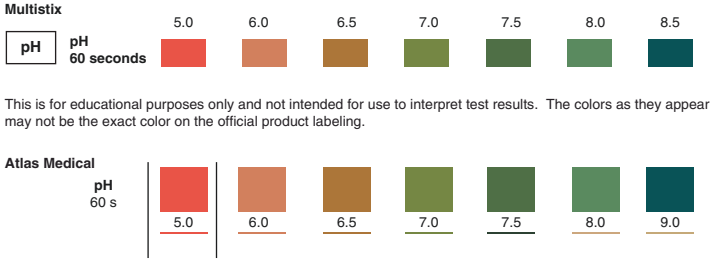

Figure 7.5 Urine pH dipstick results. Sources: Siemens Healthcare Diagnostics Inc and Atlas Medical. Used with permission

What Are the Clinical Implications of Urine pH?

A pH of 5 reflects a more *acidic* urine. Most variation in urinary pH is benign and is related to variation in diet. More acidic urine is seen with high intake of meat and certain fruits such as cranberries. The timing of specimen collection may also impact urinary pH. A morning specimen is typically slightly acidic [1]. Acidic urine could also be indicative of the presence of certain bacteria, such as *E. coli*, which is an acid-producing bacteria. Uric acid urinary calculi are also associated with an acidic urinary pH. *This should be high on the list of differential diagnoses for the patient in our case.*

A urinary pH close to 8 reflects a more *alkaline* urine. Benign dietary causes include a vegetarian diet or a diet high in citrus fruit. Urine collected after meals is typically more alkalotic. It is important to note that an improperly collected specimen may be falsely alkalotic; if improperly stored, urine urea will convert to ammonia.

In clinical practice, urine pH is an important test to determine management of nephrolithiasis. Obtaining a urine pH is recommended by the American Urological Association after the first episode of renal or ureteral stones. In addition, more alkaline urine with a pH >7 can indicate the presence of a urease producing bacteria, like *Proteus species*, that can lead to a higher probability of struvite stones [11]. Urine pH may help support a diagnosis of a systemic acid-base disorder in certain clinical scenarios [1]. Caution should be taken in ruling out a systemic acid-base disorder on the basis of urine pH alone, as renal disorders may lead to inability of the kidneys to properly regulate the pH of the urine [1]. Urine pH may also periodically be monitored to assess the therapeutic dose of potassium citrate which works by making the urine less acidic. This treatment is part of the pharmacological management of patients with either uric acid urinary calculi or calcium oxalate urinary calculi associated with hypocitraturia (see Chap. 12).

Case 4: Urine Dipstick with Low Specific Gravity

A 54 year old male presents to clinic with intense thirst and excessive urination.

Urine dipstick and urine microscopy	
Component	**Result**
Color	Red
Appearance	Cloudy
pH	6.0
Specific gravity	<1.005
Blood	Negative
Glucose	Negative
Ketone	Negative
Protein	Negative
Bilirubin	Negative
Urobilinogen	Negative
Leukocyte esterase	Negative
Nitrite	Negative
White blood cells (WBC)	0–5/hpf
Red blood cells (RBC)	0–2/hpf
Squamous epithelial cells	0/lpf
Casts	0/hpf
Bacteria	0/hpf

To maintain homeostasis of body fluids and electrolytes, the kidneys vary the volume of excreted urine and the concentration of solute in the urine. Specific gravity is an indicator of urine concentration. It is a ratio of urine density to density of an equal volume of pure water (1.000) at the same temperature [3].

The minimum specific gravity that urine can attain is approximately 1.002. The maximum specific gravity that urine can attain is equal to that of the hyperosmotic renal medulla, approximately 1.040. Most random specimens fall between 1.010 and 1.030 [3]. *In case 4, the patient's urine specific gravity is abnormally low. Is a urine dipstick accurate in measuring urine specific gravity?*

There is some disagreement concerning the use of reagent strips to determine specific gravity because they do not measure the "true" or total solute content but only ionic solutes. On the other hand, it is actually the ionic solutes that are of diagnostic value because they are involved in the concentrating and secreting activities of the kidneys [1, 3].

This test is based on the pKa change of certain pretreated polyelectrolytes in relation to ionic concentration, which is a surrogate marker of specific gravity. The more ions in a specimen, the more the pH will change and the higher the specific gravity will become. The colors on the reagent strip will range from deep blue green in urine of low ionic concentration to green and yellow-green in urine of increasing ionic concentration [10]. Readings can be made in 0.005 intervals by careful comparison with the color chart [3] (Fig. 7.6).

An abnormal specific gravity result can be confirmed by a urinometer or a refractometer. A urinometer is a hydrometer that is calibrated against a specific temperature, usually 20°C. It is based on buoyancy and uses displacement to esti-

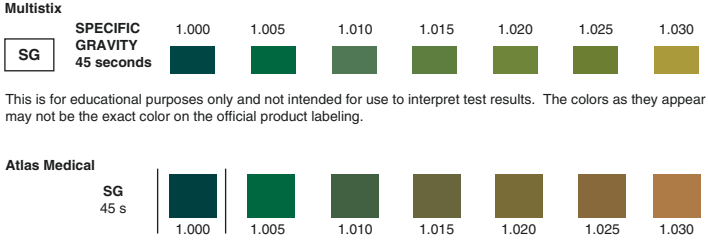

FIGURE 7.6 Urine specific gravity dipstick results. Sources: Siemens Healthcare Diagnostics Inc and Atlas Medical. Used with permission

mate specific gravity. Thus, the higher the specific gravity of a specimen, the higher the urinometer will float. If the urinometer is not calibrated to 20°C then specific gravity can be corrected by adding 0.001 for every 3°C over 20°C, or subtracting 0.001 for every 3°C below 20°C.

Alternatively, the specimen can be tested in a refractometer, although this is not routinely done. The refractometer measures the refractive index of the solution, which is the ratio of the velocity of light in air to the velocity of light in solution. The path of light is deviated when it enters a solution, and the degree of deviation is proportional to the density of the solution. The refractometer is temperature-compensated for temperatures between 60°F and 100°F and therefore requires no correction within that range. The refractometer requires only one drop of urine which gives the method an advantage over the urinometer. Because of the larger volume (at least 15 ml) of sample that is required for the urinometer, it is often necessary to report out a specific gravity as "qns" (quantity not sufficient), but the refractometer eliminates this problem [1, 3]

What Is the Clinical Relevance of the Urine Specific Gravity?

A urine specific gravity can aid the clinician in assessing a patient's hydration status and the kidney's ability to dilute or concentrate urine. Clinical context is important when evaluating specific gravity. A summation of the specific gravity values and potential pathology it is associated with can be found in Table 7.4.

The ultrafiltrate that enters Bowman's space of the glomeruli has the same specific gravity as protein-free plasma (1.010). As the ultrafiltrate passes through the nephrons, solutes and water are selectively absorbed and secreted (see Chap. 1). If the tubules are unable to perform these functions, the urine specific gravity will remain fixed at 1.010. This condition is called *isothenuria* and may indicate significant renal dysfunction. Therefore, it is important to note that specific gravity between 1.005 and 1.035 on a random urine sample is

Table 7.4 Potential etiologies of alterations in specific gravity in disease states

Urine specific gravity	Potential etiology	Physiology
<1.010	Compulsive/excessive water intake	Washout of medullary concentration gradient
	Diabetes insipidus (central, nephrogenic)	Water diuresis
1.010–1.020	Acute tubular necrosis	Tubular dysfunction
	Sickle cell disease	Medullary ischemia and micro-infarction
	CKD/ESRD	Inability to adequately concentrate or dilute urine
	Chronic hypercalcemia	Impairment of NaCl reabsorption (interfering with countercurrent mechanism) and insensitivity of collecting tubule to ADH
>1.020	True pre-renal azotemia	Low intravascular volume
	Congestive heart failure/ liver failure	Low effective arterial blood volume
	SIADH	Free water retention
	Dehydration	Low water intake

Abbreviations: *CKD* chronic kidney disease, *ESRD* end-stage renal disease, *NaCl* sodium chloride, *ADH* antidiuretic hormone, *SIADH* syndrome of inappropriate antidiuretic hormone secretion

considered normal *only* if kidney function is normal [1, 3, 9]. Patients with potential isosthenuria include those with end stage renal disease (ESRD), chronic obstruction, acute tubular necrosis (ATN), renal tubular acidosis (RTA), hypercalcemia or chronic tubulointerstitial renal disease.

Specific Gravity <1.010

Hyposthenuria is defined as a specific gravity less than 1.010. *In case 4, the patient's specific gravity is therefore interpreted*

Table 7.5 Causes of falsely high and falsely low urine specific gravity

Falsely high	Falsely low
Glucosuria (e.g., diabetes mellitus or IV glucose administration), proteinuria, IV contrast, urine contamination, and LMW dextran solutions [1, 3, 9]	Alkaline urine [10]

as hyposthenuric at 1.005. Hyposthenuria occurs when nephrons selectively absorb solutes and excrete water to create dilute urine [1, 3, 9]. A low specific gravity therefore indicates pathology in which there is increased water intake or a urinary concentration problem manifested as a water diuresis. Examples include diuretics and inadequate secretion/action of antidiuretic hormone (ADH) (central vs nephrogenic diabetes insipidus). It may also indicate pathology in which the kidney loses the ability to concentrate urine appropriately, such as chronic pyelonephritis [12]. Additionally, falsely low specific gravity can be associated with alkaline urine; therefore, it is important to correlate urine specific gravity with urinary pH [10] (Table 7.5).

Specific Gravity >1.010

Hypersthenuria is defined as a specific gravity of greater than 1.010. Most normal random specimens fall in this range. Hypersthenuria indicates a state in which nephrons selectively absorb more water than they excrete to create concentrated urine. It therefore can indicate pathology in which there is decreased water intake or poor water diuresis. Examples include dehydration and syndrome of inappropriate antidiuretic hormone secretion (SIADH). States of low effective arterial blood volume also cause the same effect, despite the body being in a hypervolemic state, such as would occur in congestive heart failure or liver failure. False elevations in specific gravity can occur with glucosuria (e.g. diabetes mellitus or IV glucose administration), proteinuria, IV contrast, urine contamination, and low molecular weight

(LMW) dextran solutions [1, 2, 9] (Table 7.5). In these circumstances the urine specific gravity does not correlate with the urine osmolality. While under normal conditions urine specific gravity can be used as a guage of urine concentrating ability, it should not exclude obtaining a urine osmolality when evaluating pathologic conditions, such as hyponatremia or diabetes insipidus (see Chap. 15).

Returning to case 4, while the patient waited patiently in clinic for his urine studies to be completed and interpreted, he was noted to have taken in 4.2 L of water and urinated roughly 4 L. The differential diagnosis in the context of his polydipsia, polyuria and dilute urine could include compulsive water consumption or diabetes insipidus. He was referred to nephrology for further testing (see Chap. 15).

Case 5: Urine Dipstick Positive for Bilirubin

A 25 year old female with a past medical history of frequent urinary tract infections presents to clinic with abdominal pain. She frequently has abdominal pain with urinary tract infections, but concedes that this is "way worse." Your nurse decides to check a urine dipstick before patient is roomed. On your initial evaluation you note mild scleral icterus. She is clutching the right side of her abdomen.

Urine dipstick and urine microscopy	
Component	**Result**
Color	Amber
Appearance	Clear
pH	6.0
Specific gravity	1.015
Blood	Negative
Glucose	Negative
Ketone	Negative

Urine dipstick and urine microscopy	
Component	**Result**
Protein	Negative
Bilirubin	3+
Urobilinogen	Negative
Leukocyte esterase	Negative
Nitrite	Negative
White blood cells (WBC)	0–5/hpf
Red blood cells (RBC)	0–2/hpf
Squamous epithelial cells	0/lpf
Casts	0/hpf
Bacteria	0/hpf

When hepatocytes are unable to excrete excess amounts of conjugated bilirubin into the bile or when biliary stasis occurs, bilirubin is secreted into the blood. Unlike unconjugated bilirubin, conjugated bilirubin is not bound to protein and is therefore easily filtered through the glomerulus and excreted into the urine. The urine dipstick test for conjugated bilirubin has a sensitivity of 0.5–1.0 mg/dL [1].

This test is based on the binding of conjugated bilirubin to diazotized salts (Fig. 7.7) on the test pad in a strong acidic environment to produce a colored compound that is various shades of tan or magenta. Results are reported as negative, small, moderate, or large, or as negative, 1+, 2+, or 3+ [3] (Fig. 7.8).

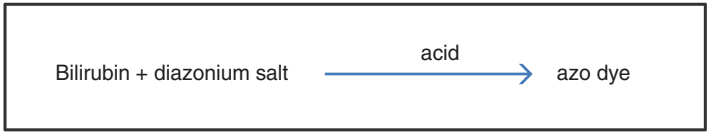

Figure 7.7 Chemical reaction of urine dipstick for bilirubin [3]

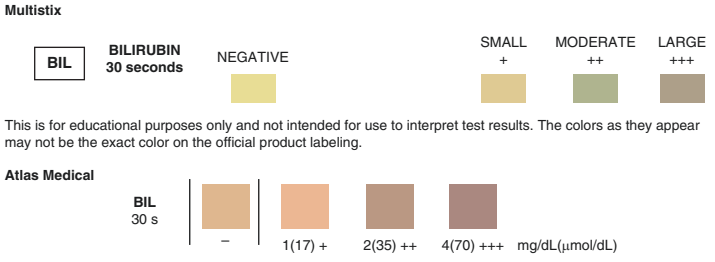

FIGURE 7.8 Urine bilirubin dipstick results. Sources: Siemens Healthcare Diagnostics Inc and Atlas Medical. Used with permission

Positive dipstick tests are confirmed with the Ictotest, which is more sensitive and specific than the urine dipstick test. Ictotest is a tablet test that uses a similar chemical reaction but a different test environment. Urine is placed on an absorbent test mat that captures substances within the urine. The reagent tablet is then placed on top of the absorbed urine and water is added to the tablet. The water dissolves the solid diazonium salt and acid in the tablet so that they run onto the mat. The reaction of conjugated bilirubin with the diazonium salt in the acid environment results in the formation of a blue ring around the dissolving tablet. All urines should be tested by this method, even when a positive dipstick result does not occur, whenever the patient has a history of liver problems or suspected liver disease. Normal adult urine contains about 0.02 mg/dL of bilirubin, which is not detectable by even the most sensitive methods [10].

What Is the Clinical Relevance of Bilirubinuria?

A positive test for urine bilirubin confirms conjugated hyperbilirubinemia. Raised conjugated bilirubinemia (with bilirubinuria) is associated with hepatocellular disease, cirrhosis, viral and drug induced hepatitis, biliary tract obstruction (e.g. choledocholithiasis), pancreatic causes of obstructive jaun-

Table 7.6 Causes of false positive and false negative results on urine bilirubin reagent strip

False positive	False negative
Highly pigmented urine from phenazopyridine compounds, indicant (intestinal disorders), or metabolites of etodolac [1, 3]	Ascorbic acid (vitamin C), aged sample (conjugated bilirubin hydrolyzes to unconjugated bilirubin at room temperature), rifampicin, and exposure to UV light (converts bilirubin to biliverdin) [1, 3]

dice (e.g. carcinoma of the head of the pancreas) and recurrent idiopathic jaundice of pregnancy. It is important to note that false elevations can occur in highly pigmented urine from phenazopyridine compounds, indican (intestinal disorders) or metabolites of etodolac [1, 3].

A negative urine bilirubin is a normal result. False negatives can occur with the presence of ascorbic acid (vitamin C), aged sample (conjugated bilirubin hydrolyzes to unconjugated bilirubin at room temperature), rifampicin and exposure to UV light (converts bilirubin to biliverdin) [1, 3] (Table 7.6).

A positive test can provide an early indication of liver disease and is often detected before the patient exhibits jaundice. It is especially useful when combined with urobilinogen. *Therefore, the best way to interpret this patient's bilirubinuria is to also evaluate for the presence of urobilinogen.*

What is the Clinical Significance of Urobilinogen?

Unconjugated bilirubin is converted to urobilinogen by intestinal bacteria in the duodenum. Most urobilinogen is excreted in the feces or transported back to the liver and converted into bile. The remaining urobilinogen (<1%) is excreted in the urine. Urobilinogen is normally present in the urine in low concentrations (0.2–1.0 mg/dL or <17 micromol/L) [3]. Most dipsticks use para-dimethylami-

nobenzaldehyde in a strongly acid medium to test for urobilinogen. A positive reaction produces a pink-red color [10] (Figs. 7.9 and 7.10).

Urine urobilinogen is a very sensitive but non-specific test to evaluate for liver damage, hemolytic disease, and severe infections. A positive urobilinogen dipstick finding can be confirmed with the Watson-Schwartz test. The Watson-Schwartz test is a qualitative test that adds an Ehrlich reagent directly to the urine sample. Ehrlich reagent contains para-dimethylaminobenzaldehyde and thus can act as an indicator for the presence of urobilinogen [10].

Normal values for urobilinogen are 1 mg/dL or less. False negatives can be attributed to high concentrations of nitrite in the urine or improper preservation of the sample leading to photo-oxidation from urobilinogen to urobilin. Additionally, patients receiving broad spectrum antibiotics may have alterations of the normal bacterial flora in the intestines and will excrete little or no urobilinogen in their urine because urobilinogen cannot be formed in their intestines [1, 3, 9].

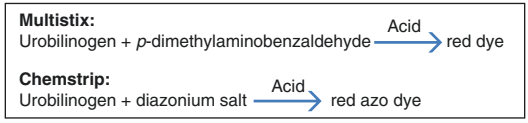

Figure 7.9 Chemical reaction of urine dipstick for urobilinogen [3]

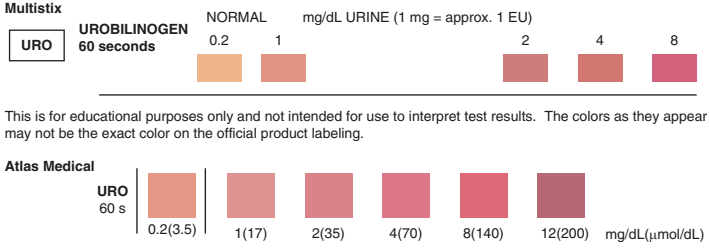

Figure 7.10 Urine urobilinogen dipstick results. Sources: Siemens Healthcare Diagnostics Inc and Atlas Medical. Used with permission

Table 7.7 Causes of false positive and false negative results on urine urobilinogen reagent strip

False positive	False negative
Intestinal obstruction	High concentrations of nitrite in the urine or improper preservation of the sample, broad spectrum antibiotics [1, 3, 9]

Urobilinogen is present in increased concentrations (>1 mg/dL) in the urine in patients with liver disease or hemolytic disorders. Examples include cirrhosis, infective hepatitis, extravascular hemolysis, hemolytic anemia, pernicious anemia, malaria and hepatitis secondary to infectious mononucleosis. False positives are uncommon, but may be seen in intestinal obstruction. With obstruction, significant quantities of urobilinogen may be absorbed from the intestine; thus the urine levels will increase [1, 3, 9, 10] (Table 7.7).

What is the Connection Between the Urine Bilirubin Test and the Urine Urobilinogen Test?

With hepatocellular jaundice, urine bilirubin may or may not be elevated, but urobilinogen will be elevated. Causes of hepatocellular jaundice include cirrhosis, hepatitis, and hepatic toxins. With post-hepatic jaundice, urine bilirubin will be markedly elevated and urine urobilinogen will be normal. Post-hepatic jaundice is typically caused by biliary obstruction. In pre-hepatic jaundice, urine bilirubin is negative, but urine urobilinogen will be markedly elevated. Pre-hepatic jaundice is typically caused by hemolytic disease (Table 7.8).

Returning to case 5, the patient's urine dipstick findings are consistent with bile duct obstruction. Additional laboratory tests were obtained showing: a creatinine of 1.3 indicating a mild acute kidney injury, an AST/ALT in the 200s indicating hepatocellular injury, markedly elevated alkaline phosphatase in the low 1000s, and a total bilirubin of 2.5 consistent with bile duct obstruction. A right upper quadrant ultrasound

Table 7.8 Use of urine bilirubin and urine urobilinogen in the evaluation of jaundice

	Urine bilirubin	Urine urobilinogen
Hepatocellular jaundice	+ or –	++
Post-hepatic jaundice	+++	–
Pre-hepatic jaundice	–	+++

obtained in the ED confirmed the diagnosis of biliary tree obstruction and she was referred to gastroenterology and general surgery.

Summary

There were multiple components of the dipstick urinalysis discussed in this chapter that are valuable to providers in the diagnosis, assessment and management of patients. The presence of glucose in urine can be a marker of a hyperglycemic state or renal disease, and next steps in management of a positive urine glucose is to obtain a serum glucose level. Glucosuria in combination with ketonuria is a common finding in both type 1 and type 2 diabetics with poor control and can be a marker of diabetic ketoacidosis.

In regard to urine pH, several benign causes and dietary changes can be associated with more acidic or alkaline urine. It is important to keep in mind there are pathological conditions, such as *E. coli* UTI associated with acidic urine or *Proteus* UTI associated with alkaline urine, that can be reflected in the urine pH.

The dipstick urinalysis also reports specific gravity values, which can be useful in evaluation of disorders that alter the ability of the kidney to adequately concentrate or dilute urine, and giving information regarding the hydration status of the patient.

Urine bilirubin can be associated with hepatic damage, and its interpretation with urobilinogen can help distinguish if this is due to hepatic, pre-hepatic or post-hepatic damage.

The urobilinogen additionally is a sensitive test that should raise suspicion for hemolytic anemia in the right clinical scenario.

Urine test results should always be interpreted in the clinical context of why they were ordered. They should also be interpreted with caution in cases where false positive or false negative influencers may be present.

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Chapter 8
Urine Microscopy: The Burning Truth – White Blood Cells in the Urine

Andrew M. Vitale and Gina M. Lockwood

Objectives

- Describe the clinical implications for different types of leukocytes on urine microscopy
- Describe differential diagnoses of pyuria, both infectious and "sterile" pyuria
- Describe evaluation and management for pyuria associated with urinary tract infections
- Describe evaluation and management for sterile pyuria
- Describe clinical presentation and appropriate evaluation for genitourinary tuberculosis
- Describe specialized urine testing available to detect urogynecologic and uroenteric fistulae

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Definitions

- **Pyuria** – Presence of white blood cells in the urine (in this chapter defined as >5 white blood cells/high power field)
- **Sterile pyuria** – Presence of white blood cells in the urine in the absence of a positive bacterial urine culture, as determined by means of aerobic laboratory techniques (on a 5% sheep-blood agar plate and MacConkey agar plate)

Overview

Pyuria, or presence of white blood cells (WBC)s in the urine, is most commonly associated with a urinary tract infection (UTI), but a UTI is not the only cause of pyuria. Isolated pyuria in the absence of bacteriuria can represent many inflammatory processes in the urinary tract, anywhere from the glomerulus to the urethra, or even the genital tract. It is important for the provider to understand reasons for the presence of pyuria, as treatment of its associated conditions can range from outpatient observation to urgent nephrectomy. This chapter will discuss nuances of the analysis of pyuria in the context of clinical presentation to develop an appropriate differential diagnosis. Initial evaluation and management of urinary tract infection, sterile pyuria, genitourinary tuberculosis, and urinary tract fistulae will be discussed.

Case 1: Symptomatic Infectious Pyuria

A 59 year old female presents to her primary care physician complaining of 5 days of painful urination, urinary frequency, intermittent right flank pain, subjective fevers, and nausea. She has a history of diabetes mellitus on metformin, recurrent UTI, pyelonephritis, and nephrolithiasis.

On examination, she is febrile to 39°C and has mild tachycardia to 107 beats per minute. She has right costovertebral angle tenderness.

Labs (pertinent findings):

- *Complete blood count with differential: WBC Count 17,000 (predominantly neutrophils)*
- *Basic metabolic panel: within normal limits*

Urine dipstick & urine microscopy	
Component	**Result**
Color	Yellow
Appearance	Cloudy
pH	6.0
Specific gravity	1.010
Blood	2+
Glucose	Negative
Ketone	Negative
Protein	Negative
Bilirubin	Negative
Urobilinogen	Negative
Leukocyte esterase	3+
Nitrite	2+
White blood cells (WBC)	>50/hpf
Red blood cells (RBC)	7–10/hpf
Squamous epithelial cells	0/lpf
Casts	0/hpf
Bacteria	Many/hpf

What would the next steps be in her evaluation, and what are the differential diagnoses for her pyuria?

Leukocytes can commonly be identified under low power magnification and always under high power magnification. Their presence usually indicates infection or inflammation in the genitourinary tract. It is common to identify 1–2 leukocytes/high power field (hpf) in circumcised men and up to 5 leukocytes/hpf in women and uncircumcised men in whom the urine sample may be contaminated by skin or vaginal secretions. Although there is no universally agreed upon or standardized definition for pyuria, the preferred definition considered to be most clinically significant is 10 leukocytes/mm^3 of midstream urine by counting chamber or >5–10 WBC/hpf on examination of urine sediment after centrifugation (see Chap. 3) [1, 2]. Aged leukocytes (small and wrinkled in appearance) can be present in normal vaginal secretions, but fresh leukocytes more accurately indicate pathology.

Types of Leukocytes Found in the Urine

Neutrophil

Neutrophils are by far the most common type of white blood cell found in the urine. These cells are easily recognized under a microscope (Fig. 8.1). They are round with a lobed, centrally-located segmented nucleus and granular cytoplasm. In the majority of patients their presence indicates a UTI, but their presence can also be attributed to contamination from genital secretions or genitourinary inflammation [3].

Eosinophil

The presence of greater than 1% of all leukocytes in the urine being eosinophils (eosinophiluria) is considered clinically significant [4]. Their identification requires Hansel staining in fresh urine, and this stain is not routinely performed for urinalysis [3]. These cells have a characteristically bilobed

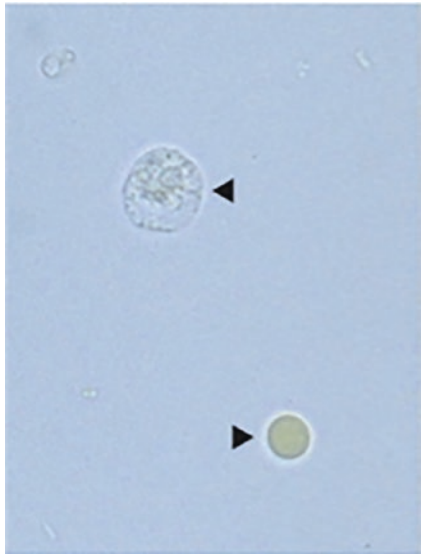

Figure 8.1 A neutrophil seen on urine bright field microscopy (top arrow); bottom arrow showing red blood cell for size and morphology comparison; Reprinted with permission from Elsevier [5]

nucleus. They are most commonly seen in the urine secondary to hypersensitivity reactions [1, 6]. Eosinophilic cystitis is a rare, distinct clinical entity in which eosinophils can be found throughout all layers of the bladder wall, manifesting with irritative lower urinary tract symptoms, like urgency, dysuria, and urinary frequency [1]. This entity is seen in children more commonly than in adults, and its appearance can mimic bladder neoplasm. Staining for eosinophils is usually only performed when eosinophilic cystitis is suspected. Small numbers can be seen with bacterial urinary tract infections. Additionally, they can be found in chronic pyelonephritis, urinary schistosomiasis, and prostatitis [3].

Lymphocyte

This type of leukocyte requires special stains for identification and has a very large nucleus that fills the majority of the cell. Small lymphocytes are normally present in the urine. Their presence is also typical in those with chyluria [3].
Causes of bacterial pyuria (UTI)

Clinical Approach to Pyuria

When leukocytes are identified in the urine, the provider should attempt to distinguish between infection in the urinary tract and other causes. Urine culture should be performed to confirm presence of bacteria in the urine if UTI is suspected. In the absence of a UTI, pyuria can indicate a contaminated urine sample, an atypical infection, or inflammation in or near the genitourinary tract. Traditionally, any number of squamous cells or epithelial cells on urine microscopy was thought to be associated with contamination from the genital tract or perineal skin. However, multiple studies have refuted this concept, showing that squamous cells are actually very poor predictors for contamination [7, 8]. See Table 8.1 for differential diagnoses of pyuria on urine microscopy.

Returning to case 1, urine culture should be performed given fever, tachycardia, leukocytosis, and pyuria. Given her presentation, pyelonephritis is suspected. An acute stone episode can also cause pyuria, but in the absence of concomitant infection, fever is unlikely.

Case 2: Symptomatic Infectious Pyuria

The patient in case 1 has a urine culture performed for suspected urinary tract infection. While awaiting culture results, should any additional testing be performed (serologic, radiologic)? Should antibiotic treatment be started empirically?

Table 8.1 Differential diagnoses for pyuria

Causes of bacterial pyuria (UTI)
Cystitis
Uncomplicated cystitis
Complicated cystitis
Emphysematous cystitis
Urethritis
Periurethral abscess
Ureteritis
Pyelonephritis
Xanthogranulomatous pyelonephritis
Emphysematous pyelonephritis
Renal/perirenal abscess
Bacterial epididymitis
Bacterial prostatitis
Causes of sterile (abacterial) pyuria
Atypical UTI
Viral (adenovirus, BK polyomavirus)
Fungal (*Candida* species)
Parasitic (*Trichomonas vaginalis*, *Schistosoma haematobium*)
Genitourinary tuberculosis
Sexually-transmitted infection
Gonococcal/non-Gonococcal urethritis
Gonococcal/non-Gonococcal epididymitis
Genital herpes
Human papilloma virus
Human immunodeficiency virus

(continued)

Table 8.1 (continued)

Inflammatory genitourinary and systemic disorders
Tubular interstitial disease
Acute interstitial nephritis
Interstitial cystitis (bladder pain syndrome)
Urolithiasis (renal, ureteral, bladder, urethral)
Chronic nonbacterial prostatitis (chronic pelvic pain syndrome)
Genitourinary malignancy
Genitourinary fistula
Analgesic nephropathy
Kawasaki disease
Systemic lupus erythematosus
Iatrogenic
Indwelling catheter
Intermittent catheterization
Foreign body (transvaginal mesh, ureteral stent)
Radiation cystitis
Strenuous exercise

The vast majority of patients in whom pyuria is present will be diagnosed with an infection of the urinary tract. Urine dipstick can act as a screening tool for pyuria and UTI (see Chap. 6). Cloudy/turbid/milky appearance on gross examination of urine can indicate the presence of white blood cells. In a patient with positive leukocyte esterase (LE) on urine dipstick (with or without the presence of nitrite), urine microscopy should be obtained for confirmation of presence of leukocytes. Leukocyte esterase is an indoxyl esterase enzyme released from white blood cell granulocytes [3]. Its presence suggests current or recent WBCs in the urine. Nitrites are not a specific marker for WBCs in the urine and can be present without pyuria. Many species of gram-negative bacteria convert nitrates to nitrites, so this test acts as a screening tool for bacteriuria (see Chap. 6).

Table 8.2 Sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV) of leukocyte esterase, nitrite and urine leukocytes > 5/hpf for urinary tract infection [1, 4, 6]

	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)
Leukocyte esterase	47–95	64–92	43–56	82–91
Nitrite	8–95	98	_____	_____
Leukocytes on urine microscopy	90–96	47–50	56–59	83–95

Urine leukocyte esterase, nitrite, and microscopy for leukocytes have variable sensitivity and specificity in the diagnosis of UTI (Table 8.2). Causes of a false negative LE include but are not limited to an increased urine specific gravity, urobilinogen, glycosuria, as well as ingestion of large amounts of vitamin C (ascorbic acid). Extrinsic factors can alter the number of WBCs in the urine (e.g., hydration status, collection method, and the centrifuge process), which can alter accuracy for diagnosis of UTI with positive leukocytes on microscopy. However, in the absence of microscopic pyuria, the diagnosis of a UTI should be questioned unless culture proven. When microscopic pyuria is confirmed and a clinical diagnosis of infection is suspected, this typically should be confirmed with urine culture.

A UTI is diagnosed when clinical signs and/or symptoms are combined with the presence of a known uropathogenic bacteria (positive urine culture). Most UTIs occur secondary to ascending infection. The urinary tract is typically devoid of pathogenic bacteria, however bacteria can move from the perineal region into the urethra and bladder leading to a bladder infection, or cystitis (see Chap. 11).

Acute uncomplicated UTI (cystitis) is a common problem affecting otherwise healthy women and is associated with considerable short-term morbidity (lower urinary tract symptoms) but generally no long-term morbidity [9]. There is debate regarding whether routine urine culture is needed for a symptomatic woman with recent signs and symptoms consistent with acute cystitis. Some advocate that in this population, typical symptoms and a urinalysis positive for pyuria,

bacteriuria, or hematuria, or a combination of these can be treated empirically without culture [10]. However, urine culture should be obtained in those in whom a diagnosis of cystitis is in doubt or in those with recurrent cystitis. Urinary tract infections are less common in men and usually signify a complicating factor such as bladder outlet obstruction leading to incomplete bladder emptying. Urinary tract infections can also cause infection of the epididymis and prostate.

Approximately 50% of lower urinary tract infections ascend into the upper urinary tract (ureter, renal pelvis, major and minor calyces) causing pyelonephritis. Hematogenous seeding of infection to the kidney is rare in otherwise healthy patients; however, when this does occur it is usually with gram positive organisms (e.g., *Staphylococcus aureus*). Pyelonephritis occurs more commonly in diabetics or other immunocompromised patients, as well as those with anatomic abnormalities of the genitourinary tract (vesicoureteral reflux) or nephrolithiasis [11].

Patient symptoms alone can usually differentiate between the presence of a lower urinary tract infection (cystitis) and upper urinary tract infection (pyelonephritis). Cystitis usually presents with irritative lower urinary tract symptoms such as

First Line Therapy	Nitrofurantoin monohydrate/macrocrystals 100 mg twice daily x 5 days (avoid if pyelonephritis suspected) Trimethoprim-sulfamethoxazole 160/800 mg (1 double-strength tablet) twice daily x 3 days (avoid if resistance locally > 20% or if used for UTI in last 3 months) Fosfomycin trometamol 3 gm single dose (avoid if early pyelonephritis suspected)
Second Line Therapy (Patient unable to use first line therapy because of unavailability, allergy, or intolerance)	Ciprofloxacin 250 mg every 12 hours x 3 days OR Ciprofloxacin 500 md daily x 3 days

Figure 8.2 Infectious Diseases Society of America Approach to empiric antibiotic treatment in an uncomplicated UTI. For acute, uncomplicated cystitis, patient must not have fever, flank pain, or other suspicion for pyelonephritis. To utilize first or second line therapies, patient must be able to tolerate oral medication [20, 27]

dysuria, urinary frequency, gross hematuria, urgency, and feelings of incomplete bladder emptying. Figure 8.2 outlines most recent Infectious Diseases Society of America recommendations for empiric antibiotic treatment of an uncomplicated urinary tract infection in a female. The choice between agents should be individualized and based on patient allergy and compliance history, local practice patterns, local community resistance prevalence, availability, cost, and patient and provider threshold for failure. For symptom control phenazopyridine (pyridium) is a unique medication used to relieve discomfort associated with UTIs but can also be used to relieve irritation of the lower urinary tract mucosa from other causes. It is an azo dye excreted in the urine and exerts a topical analgesic effect on the mucosa of the urinary tract without antibacterial effects. It is generally used for dysuria and urinary urgency caused acutely by urinary tract manipulation or infection, but it can only be used for two days, as toxic metabolites can accumulate. Because it is a dye, it produces red/orange discoloration of the urine.

Pyelonephritis is a clinical diagnosis associated with fever, flank pain, and leukocytosis. Basic serologic studies are warranted in suspected pyelonephritis including complete blood count (CBC) and basic metabolic panel (BMP). Urine culture should always be obtained. Blood cultures are positive in about 25% of cases of uncomplicated pyelonephritis in women, and the majority replicate the urine culture and do not influence decisions regarding therapy. Therefore blood cultures should not be routinely obtained for the evaluation of uncomplicated pyelonephritis in women. Blood cultures should be performed in men and women with systemic toxicity or in those requiring hospitalization or with risk factors such as pregnancy [12].

Imaging can be useful if pyelonephritis is suspected based on symptoms but in the absence of pyuria or bacteriuria. This situation can occur in the setting of concomitant obstruction (e.g., ureteral calculus) and infection that causes urine stasis in one region of the urinary tract. Imaging should also be obtained if the patient is not responding to culture appropriate antibiotics. In that situation renal abscess or concomitant obstruction should be considered. Both renal bladder sono-

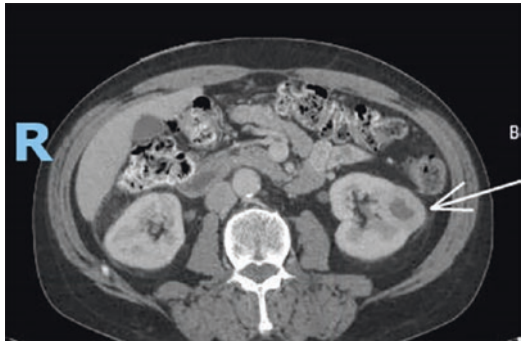

Figure 8.3 Left interpolar renal abscess in a male patient as seen on CT of the abdomen with IV contrast; this abscess resolved with antibiotic therapy, not requiring surgical drainage (Courtesy of University of Iowa, Department of Urology)

gram and CT of the abdomen and pelvis with and without IV contrast can identify pyelonephritis, stones or renal abscesses (Fig. 8.3), but if intervention is needed for abscess drainage or treatment of stones, a CT should be performed for surgical planning. A renal abscess is usually associated with flank or abdominal pain, fevers, nausea, vomiting, and sometimes lower urinary tract symptoms. Renal abscess is often misdiagnosed clinically as pyelonephritis [13]. Additionally, xanthogranulomatous pyelonephritis should be suspected in a patient with symptoms of pyelonephritis and history of nephrolithiasis. This is a rare, severe, chronic infection that typically results in diffuse renal destruction and the need for nephrectomy. It is more common in diabetics and immunocompromised patients than in the healthy population. This entity should also be recognized on CT of the abdomen and pelvis.

Treatment of pyelonephritis consists of oral or intravenous antibiotics for 7–14 days. Treatment should be started empirically while awaiting urine/blood cultures to prevent renal scarring and sepsis. Intravenous antibiotics are necessary in situations in which a patient cannot tolerate oral intake due to nausea and vomiting, or in which urinary tract organisms

are resistant to all oral antibiotic options. CT or fluoroscopy-guided percutaneous drainage and/or a prolonged antibiotic course may be needed in the setting of a renal abscess. The presence of an obstructing stone in the presence of a UTI or renal/peri-renal abscess requires urology consultation. An obstructing stone with concurrent pyelonephritis requires urgent urinary tract decompression with ureteral stent placement or nephrostomy tube placement.

Returning to case 2, this patient should be started on empiric therapy while awaiting urine culture results to prevent morbidity. Blood culture is not required. Given her history of urolithiasis, it is prudent to obtain a renal sonogram or CT to rule out concurrent obstruction from a urinary tract stone.

Case 3: Sterile Pyuria

A 33 year old male presents to his primary care physician complaining of 3 days of urinary frequency, urgency, occasional burning, and a feeling of lower abdominal pressure. He returned 1 month ago from a 6-month mission trip to India. Of note, he has a history of rheumatoid arthritis on chronic immunosuppression.

Urine dipstick & urine microscopy	
Component	**Result**
Color	Yellow
Appearance	Clear
pH	7.0
Specific gravity	1.015
Blood	Negative
Glucose	Negative
Ketone	Negative
Protein	Negative

Urine dipstick & urine microscopy	
Component	**Result**
Bilirubin	Negative
Urobilinogen	Negative
Leukocyte esterase	2+
Nitrite	Negative
White blood cells (WBC)	>20/hpf
Red blood cells (RBC)	8–10/hpf
Squamous epithelial cells	1–2/lpf
Casts	0/hpf
Bacteria	0/hpf

A urine culture resulted as no growth.

"Sterile" or "abacterial" pyuria is the persistent finding of white blood cells in the urine in the absence of bacteria on standard urine culture. Population-based studies have shown that up to 13.9% of women and 2.6% of men are affected by sterile pyuria [14]. It can indicate inflammation of the urinary tract or autoimmune disease in the absence of infection (Table 8.1). Sterile pyuria must be distinguished from urinary contamination or an incorrectly collected specimen (See Chap. 3). The next step in evaluation of sterile pyuria is a repeat clean-catch or catheterized urine specimen. Women and men should not be screened for pyuria, but persistent pyuria does necessitate evaluation. See Fig. 8.4 for the evaluation of sterile pyuria.

Nephrolithiasis anywhere in the GU tract can lead to pyuria, not only from stasis-induced ascending infection but also localized urothelial inflammatory response in the absence of infection. Urothelial tumors can lead to chronic inflammation and pyuria. Kawasaki disease is a rare, acute self-limited vasculitis usually seen in children that can lead to coronary artery aneurysms. It can also manifest as sterile pyuria, microscopic hematuria, and proteinuria with renal involvement.

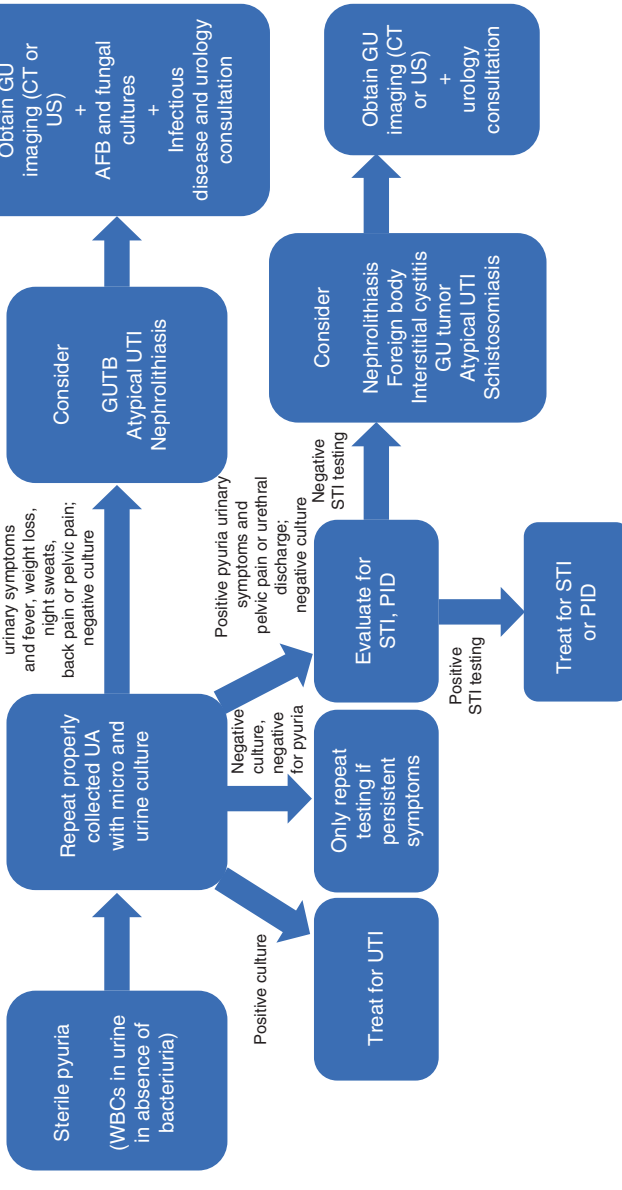

FIGURE 8.4 Evaluation for sterile pyuria. *GUTB genitourinary tuberculosis, AFB acid-fast bacilli, STI sexually transmitted infection, PID pelvic inflammatory disease*

Analgesic nephropathy can cause sterile pyuria in association with chronic interstitial nephritis and renal papillary necrosis [15]. Transient presence of leukocytes in the urine can occur with fevers or strenuous exercise [4].

Sterile pyuria does not always indicate absence of any infectious source. Sterile pyuria can be identified in fungal (*Candida albicans*), parasitic (*Trichomonas vaginalis, Schistosoma haematobium*), and viral (genital herpes or advanced human immunodeficiency virus) infections or genitourinary tuberculosis. Urethritis or vaginitis from sexually-transmitted infections should always be kept in mind when lower urinary tract symptoms are present with positive leukocyte esterase, pyuria, and negative urine culture. This is especially true in patients of reproductive age with symptoms not characteristic for UTI and with urethral or vaginal discharge. *Neisseria gonorrhoeae* and *Chlamydia trachomatis* are the most common offending organisms. Culture and hybridization tests that require urethral swab specimens are available, but nucleic acid amplification tests (NAAT)s performed on urine are preferred because of their non-invasive nature and higher sensitivity (see Chap. 16).

Genitourinary Tuberculosis

Genitourinary tuberculosis (GUTB) is an entity classically associated with sterile pyuria and deserves special mention. In men, symptoms are most commonly from urethritis, mimicking a urinary tract infection. In women, symptoms may be absent initially and then develop into pelvic inflammatory disease. GUTB accounts for 27% of cases of tuberculosis in the United States each year and can affect all of the genitourinary organs. In the US, approximately 10,000 cases of active TB were reported in 2012, but fortunately the incidence has been declining since the early 1990s [16]. Moreover, the frequency at which GUTB occurs depends on the community studied. Those in developed nations with pulmonary TB will have

GUTB in 2–10% of cases versus 15–20% in developing nations. GUTB develops via hematogenous spread with seeding of the GU organs (most common), ascending from the lower urinary tract, and contiguous spread/direct inoculation from nearby organs [16].

If a patient is diagnosed with sterile pyuria (aerobic and anaerobic cultures have proven negative), GUTB should be considered. An increased index of suspicion for GUTB is necessary in patients who have recently traveled out of the country (*like case 3*) or live with immigrants. It should also be strongly considered in the immunocompromised.

Mycobacterium tuberculosis can sometimes be identified on standard urine culture, but in one study, these bacteria were only present in acid-fast bacilli (AFB) culture 37% of the time [15]. The current gold standard for the diagnosis of GUTB is urine acid fast bacilli culture and stain, with 3–6 consecutive early morning urine samples recommended for highest yield (increases sensitivity to 80%) [17]. Unfortunately, results can take upwards of 4–6 weeks to return [16]. When diagnosing extrapulmonary TB, NAATs via polymerase chain reaction can increase sensitivity to 87% to 96%. Although this technique can decrease time to results by 1–2 days and detect low bacillary loads, it is not widely used at this time [15].

Finally, when GUTB is suspected, radiographic studies (e.g., CT with IV contrast and delayed images or high resolution ultrasound) should be completed to evaluate the upper and lower urinary tract, as strictures have been noted from the renal pelvis down to the ureterovesical junction in as many as 60–84% of cases (Fig. 8.5). Even if the patient has no respiratory symptoms, it is prudent to evaluate the pulmonary system since up to 20% of patients with GUTB will have concomitant pulmonary TB [16]. Management of uncomplicated GUTB is the same four-drug regimen used for pulmonary TB: isoniazid, rifampin, pyrazinamide and ethambutol [18].

Returning to case 3, given this patient's recent international travel as well as immunocompromised state, genitourinary

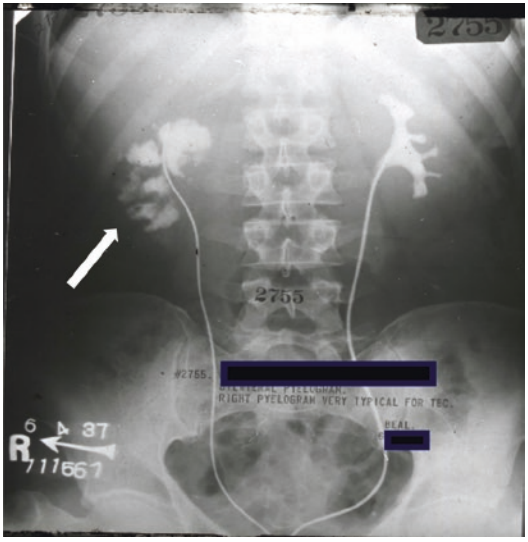

FIGURE 8.5 Retrograde pyelogram from 1937 depicting "feathering" of right renal calyces affected by GUTB; blacked out text contains patient information. (Courtesy of Charles Hawtrey, MD, University of Iowa, Department of Urology)

tuberculosis should be high on the list of differential diagnoses for sterile pyuria. Early morning AFB stain and culture should be obtained, as well as imaging of the GU tract.

Case 4: Genitourinary Fistulae

A 58 year old female presents to your office for a postoperative visit after undergoing hysterectomy 4 weeks ago for uterine fibroids. She has recovered well but since the time of foley catheter removal from the bladder she reports she has been leaking urine. This urine leakage is worse with increased activity. She is sure this is just occurring because of her increased age and having had multiple children, but she did not have these symptoms prior to surgery.

Urine dipstick & urine microscopy	
Component	**Result**
Color	Yellow
Appearance	Clear
pH	7.0
Specific gravity	1.010
Blood	2+
Glucose	Negative
Ketone	Negative
Protein	Negative
Bilirubin	Negative
Urobilinogen	Negative
Leukocyte esterase	2+
Nitrite	Negative
White blood cells (WBC)	10/hpf
Red blood cells (RBC)	5–10/hpf
Squamous epithelial cells	1–2/lpf
Casts	0/hpf
Bacteria	0/hpf

Urine culture obtained is consistent with mixed flora. What differential diagnoses should be considered, especially given this patient's recent surgery? What type of evaluation would be helpful in determining the etiology of her leakage?

Urogynecologic Fistulae

A urogynecologic fistula is a cause of sterile pyuria and often continuous urinary incontinence. Generally speaking, a fistula is any abnormal connection between two or more epithelium or mesothelium lined body cavities or the skin surface. Most

fistulae between the female genital and urinary tracts in industrialized countries are iatrogenic in nature, but they can also result from childbirth (the most common cause worldwide), congenital anomalies, malignancy, inflammation and infection, radiation, surgery, trauma, foreign bodies, ischemia, and other causes [21].

A vesicovaginal fistula is the most common acquired fistula of the urinary tract. In industrialized nations, the most common etiology is injury during gynecologic surgery, and the incidence after hysterectomy is estimated at 0.1–0.2% [22]. The most common complaint is constant urinary drainage per vagina, although small fistulae can cause positional wetness that can be mistaken for stress urinary incontinence. In addition to history and physical examination including speculum examination, traditional urine studies can be performed, although they can give very nonspecific findings like microscopic hematuria and pyuria. Urine culture should be obtained to rule out concomitant infection. A vesicovaginal fistula can be confirmed by instilling methylene blue dye or indigo carmine into the bladder per urethra and observing for discolored vaginal drainage directly or with insertion of vaginal gauze.

Up to 12% of postsurgical vesicovaginal fistulae have an associated ureteral injury or ureterovaginal fistula [23]. A "double dye test" may confirm a genitourinary fistula and identify or distinguish between a vesicovaginal fistula, urethrovaginal fistula, or ureterovaginal fistula. During this test, a tampon is placed in the vagina. Oral phenazopyridine is given, and blue dye is instilled into the bladder per urethra. If the tampon is colored yellow/orange at the proximal end, a ureterovaginal fistula is suggested. Blue discoloration in the midportion is suggestive of vesicovaginal fistula, and blue discoloration at the distal end is indicative of urethovaginal fistulae or incontinence. Endoscopy and/or fluoroscopic studies are used to confirm diagnosis and localize the fistula. Other appropriate urine studies for a urogynecologic fistula include urine culture and cytology if a malignant source is suspected. Creatinine level of vaginal discharge being consistent with urine can also diagnose a urogynecologic fistula.

Because of the high rate of associated upper tract involvement, renal imaging is often necessitated.

Vesicoenteric Fistulae

Vesicoenteric fistulae can also cause sterile pyuria. Abnormal connections between the bowel and urinary tract can result from bowel disease like diverticulitis, colorectal carcinoma, Crohn's disease, or from radiation, infection, or trauma. Symptoms can originate from the genitourinary or gastrointestinal tracts. Initial symptoms associated with vesicoenteric fistulae are often nonspecific urinary complaints including pneumaturia (air passed with urination), frequency, urgency, suprapubic pain, recurrent urinary tract infections, and hematuria [22]. Gastronintestinal symptoms include fecaluria and tenesmus.

A diagnosis of vesicoenteric fistula can often be made based on clinical history. Urinalysis can show hematuria and pyuria with possible polymicrobial culture growth. CT is the imaging modality of choice, if necessary, but it can lack sensitivity and specificity. An adjunct test for confirmation of vesicoenteric fistula in difficult cases is oral intake of 50 g activated charcoal, which will then appear in the urine as black particles. Alternatively the poppy seed test can be helpful. Poppy seeds are ingested orally (1.25 ounces in 12 ounces water or 6 ounces yogurt), then urine is collected for 48 hours and examined for poppy seeds [24]. Neither of these tests will localize the fistula. Additionally, the *Bourne test* [25] can be performed after a nondiagnostic barium enema. The next voided urine following the enema is centrifuged and then examined under fluoroscopy (Fig. 8.6) [26]. Radiodense particles in the urine are considered diagnostic for a vesicoenteric fistula.

Returning to case 4, this patient's history and urinalysis are consistent with a urogynecologic fistula. Although her symptoms are exacerbated by activity, new onset stress urinary incontinence is unlikely. She should undergo evaluation for both vesicovaginal as well as ureterovaginal fistula.

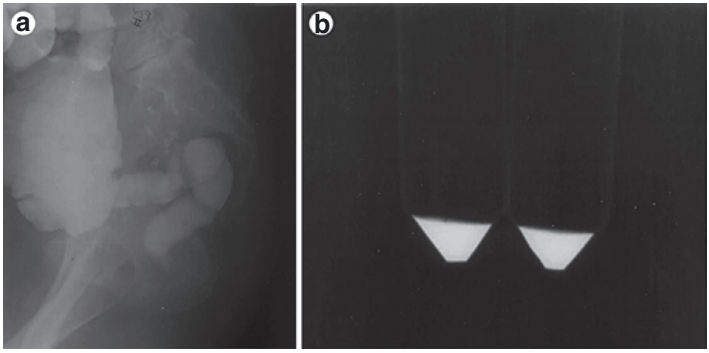

Figure 8.6 Diagnosis of a vesicoenteric fistula. (**a**) Barium enema study with no evidence of uroenteric fistula. (**b**) Subsequent centrifuged urine (Bourne test) showing radiodensity, consistent with vesicoenteric fistula [26]; Reprinted from [26], with permission from Elsevier

Summary

Pyuria is most commonly used as a screening measure for diagnosis of urinary tract infection with good sensitivity. Pyuria, however, is not specific for UTI, and in the absence of infection, other diagnostic possibilities must be considered. The differential diagnosis of pyuria is vast, including atypical infections, sexually-transmitted infections, inflammatory disorders of the urinary tract, genitourinary fistulae, systemic inflammation, and even genitourinary malignancy. In patients with sterile pyuria, urology referral is indicated. Genitourinary tuberculosis is the classic disease state associated with sterile pyuria, and although much less common in modern developed countries, it is still prevalent throughout the world and remains a pertinent clinical entity.

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Chapter 9
Urine Microscopy: Seeing Red – Understanding Blood in the Urine

Christopher Meier and Gina M. Lockwood

Objectives

- Describe the microscopic appearance of normal and abnormal red blood cells in the urine and their possible clinical implications
- Describe the differential diagnosis and basic evaluation of a patient identified as having microscopic or gross hematuria
- Differentiate glomerular from non-glomerular causes of hematuria
- Identify patients with risk factors for malignancy or other causes of hematuria

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- Understand indications for referral to nephrology and/or urology for further evaluation and management of hematuria

Overview

This chapter addresses the finding of blood in the urine (hematuria) on both a microscopic and gross level. Formulation of an appropriate differential diagnosis is reviewed, with consideration of the patient's clinical history and microscopic appearance of red blood cells (RBC)s. Important guidelines regarding further diagnostic workup, including reasons for referral, are also discussed.

Case 1: Microscopic Evaluation of Red Blood Cells

A 55 year old male presents to the clinic for weak urinary stream for the last 6 months. Urine dipstick was obtained and positive for blood so microscopic urinalysis was performed.

How would this urinalysis be interpreted? What pertinent information can be obtained from examination of red blood cells on microscopy?

Urine dipstick & urine microscopy	
Component	**Result**
Color	Yellow
Appearance	Clear
pH	6.5
Specific gravity	1.015
Blood	1+
Glucose	Negative
Ketone	Negative
Protein	Negative
Bilirubin	Negative
Urobilinogen	Negative

Urine dipstick & urine microscopy	
Component	**Result**
Leukocyte esterase	Negative
Nitrite	Negative
White blood cells (WBC)	0–5/hpf
Red blood cells (RBC)	6–10/hpf
Squamous epithelial cells	0/lpf
Casts	0/hpf
Bacteria	0/hpf

Microscopic hematuria is defined by the presence of RBCs on microscopic examination of the urine. Since a small number of RBCs may enter the urine under normal conditions in healthy individuals [1, 2], the threshold established for a diagnosis of microscopic hematuria by the American Urological Association is three or more RBCs per high power field [3]. As false positive results for blood often occur with dipstick testing (see Chap. 4), a dipstick positive for blood must be confirmed with microscopic testing to confirm a diagnosis of microscopic hematuria**.** Gross hematuria is defined by the presence of visible blood or blood clots in the urine.

For the purpose of evaluating RBCs in the urine, the specimen may be prepared with an automated system or manually. If performed manually, the specimen should be centrifuged with the sediment resuspended in a standardized volume of supernatant as described in Chap. 3 [1, 4]. The urine is then examined microscopically under high power. Urine samples should ideally be reviewed within 2–3 hours after collection [1, 4]. While refrigeration may assist with preserving the specimen for a more extended period of time, this can also lead to precipitation of other substances. RBCs may lyse in urine with a low specific gravity, alkaline urine or with delay in examination [1, 4].

When viewed microscopically, RBCs may be described as isomorphic or dysmorphic. Isomorphic cells are of normal size (average around 6–7 μm) and shape (biconcave discs) (Fig. 9.1) [1, 4]. Dysmorphic cells, on the other hand, are of irregular shape and contour, and are generally of glomerular origin, where they may be deformed by passage through the glomerular basement

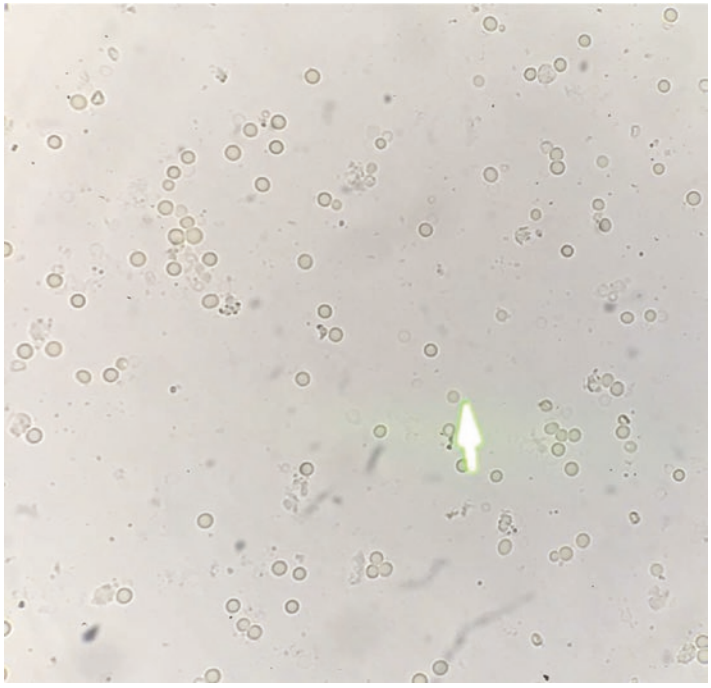


FIGURE 9.1 The normal appearance of red blood cells under high power magnification on bright field microscopy. (Courtesy of University of Iowa, Department of Nephrology)

membrane. They may also sustain subsequent additional damage as they pass through the tubular system of the nephron, where they are subjected to changes in pH and osmolality [5]. The specific proportion of dysmorphic RBCs that define hematuria as glomerular is not established. Glomerular origin is suggested by an increased number of red blood cell casts (see Chap. 10) or acanthocytes (spur cells), which are a specific subtype of dysmorphic RBCs characterized by thorn-like projections. It is accepted by some that if at least 5% of the urinary RBCs are acanthocytes, the source of hematuria is most likely glomerular in origin [4–6].

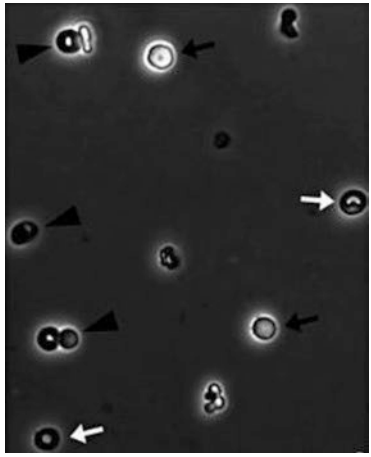
Some other rare RBC variants such as elliptocytes, dacryocytes and sickle cells may also be detected in the urine and

generally reflect systemic pathology. In concentrated urine, normal RBCs may become crenated (contracted) through the loss of intracellular water, leading to smaller cells with rough appearing edges [1]. See Table 9.1 for examples of RBC variants on microscopy, as well as possible causes of their presence.

Returning to case 1, this patient has microscopic hematuria based on microscopic urine analysis. Red blood cells should be analyzed for isomorphic versus dysmorphic appearance. With his history of weak stream, a nonglomerular cause of hematuria is likely, such as benign prostatic hypertrophy or urethral stricture. Bladder cancer should also be ruled out.

TABLE 9.1 Rare red blood cell variants on urine microscopy

Type	Characteristic appearance	Associated pathology
Acanthocyte (Spur cell)	Ring-shaped RBC with blebbed protrusions	Glomerular hematuria



Isomorphic (black arrows) and dysmorphic (white arrows) RBCs, and acanthocytes (arrowheads) on phase contrast microscopy

(continued)

Table 9.1 (continued)

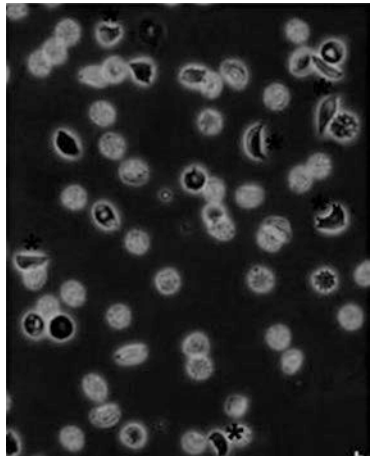
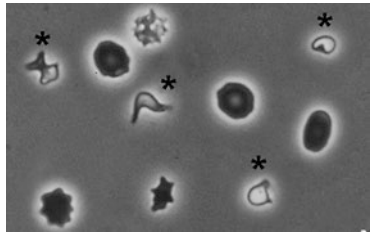
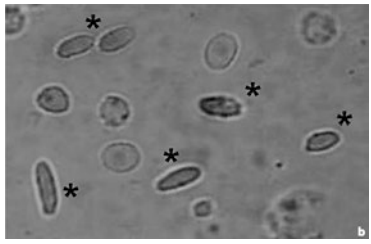
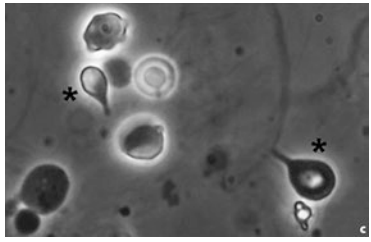
Type	Characteristic appearance	Associated pathology
Sickle cell	Sickle, crescents, holly leaf, pecked contour  Sickle cells (asterisks) with isomorphic crenated RBCs on phase contrast microscopy	Sickle cell disease/ trait
Anisocyte	Variation in RBC size	Anemia (various etiologies)
Poikilocyte	Variation in cell shape (includes schistocytes – fragmented RBCs)  Schistocytes and poikilocytes (asterisks) with isomorphic crenated and non-crenated RBCs on phase contrast microscopy	Anemia (various etiologies)

Table 9.1 (continued)

Type	Characteristic appearance	Associated pathology
Elliptocyte	Elongated, cigar-like shape  Elliptocytes (asterisks) with isomorphic RBCs on bright field microscopy	Hemolytic anemia
Dacrocyte	Tear drop  Dacrocytes (asterisks) on phase contrast microscopy	Anemia secondary to systemic lupus

Images reprinted with permission from Tesser Poloni JA, Bosan IB, Garigali G, Fogazzi GB. Urinary red blood cells: not only glomerular and nonglomerular. Nephron Clin Pract. 2012;120:c36–41. Copyright © 2012 Karger Publishers, Basel, Switzerland

Case 2: Differential Diagnosis of Microscopic Hematuria

A 38 year old female presents to the emergency department with a 24-hour history of right flank pain and nausea. She has no lower urinary tract symptoms. She has significant right costovertebral angle tenderness on examination, but physical examination is otherwise unremarkable. Based on her history and urinalysis results, what are possible causes of her symptoms?

Urine dipstick & urine microscopy	
Component	**Result**
Color	Yellow
Appearance	Clear
pH	6.5
Specific gravity	1.020
Blood	3+
Glucose	Negative
Ketone	Negative
Protein	Negative
Bilirubin	Negative
Urobilinogen	Negative
Leukocyte esterase	1+
Nitrite	Negative
White blood cells (WBC)	2–4/hpf
Red blood cells (RBC)	>50/hpf
Squamous epithelial cells	0/lpf
Casts	0/hpf
Bacteria	0/hpf

Evaluation for microscopic hematuria is necessary, regardless of whether there are associated symptoms. Table 9.2 details the most common etiologies of microscopic hematuria. Sometimes a definitive cause is not established, and the hematuria is deemed idiopathic. Differential diagnoses for microscopic and gross hematuria are generally similar but reflect varying degrees of disease.

History, physical exam, and urine testing can help to distinguish between the glomerular and non-glomerular causes of hematuria (Table 9.3). The hallmark of glomerular disease is excretion of protein in the urine (see Chap. 5). Glomerular diseases with dysmorphic RBCs and proteinuria on urinaly-

Table 9.2 Common etiologies of microscopic hematuria

Diagnosis	Frequency (%)
Idiopathic	43–68
Urinary tract infection	4–22
Benign prostatic hyperplasia	10–13
Urinary calculi	4–5
Bladder cancer	2–4
Renal cystic disease	2–3
Medical renal disease	2–3
Kidney cancer	<1
Prostate cancer	<1
Urethral stricture disease	<1

Table 9.3 Differential diagnosis for glomerular and non-glomerular causes of hematuria [12, 13]

Glomerular causes	Non-glomerular causes
Diagnoses causing nephritic syndrome	*Infection/Inflammation*
IgA nephropathy	Cystitis, pyelonephritis, urethritis
Membranoproliferative glomerulonephritis	Sexually transmitted infection
Post-infectious (including post-streptococcal) glomerulonephritis	Atypical urinary tract infection (e.g., Schistosomiasis)
Lupus nephritis	Radiation cystitis
Alport syndrome	Interstitial cystitis
	Drugs
	Cyclophosphamide (hemorrhagic cystitis)

(continued)

Table 9.3 (continued)

Glomerular causes	Non-glomerular causes
Anti-glomerular basement membrane disease	Anticoagulation (e.g., warfarin)
Pauci-immune diseases	*Calculi*
Granulomatosis with polyangiitis	Renal, ureteral, bladder
Eosinophilic granulomatosis with polyangiitis	Asymptomatic crystalluria (hypercalciuria, hyperuricosuria)
Microscopic polyangiitis	*Benign prostatic hyperplasia*
Diagnoses causing nephrotic syndrome	*Obstruction*
Minimal change disease	Urethral/ureteral stricture
Focal segmental glomerulosclerosis	Ureteropelvic junction obstruction
Membranous nephropathy	Posterior urethral valve
Membranoproliferative glomerulonephritis	*Tumor/neoplasm*
C3-dominant glomerulonephritis	Kidney, ureter, bladder, prostate, urethra
Dense deposit disease	*Gynecologic*
Systemic diseases	Endometriosis
Diabetes mellitus	Menstrual contamination
Amyloidosis	Atrophic vaginitis
Multiple myeloma	*Other anatomic/structural causes*
	Cystic renal disease
	Urethral diverticulum
	Urogynecologic or uroenteric fistula
	Arteriovenous malformation
	Renal artery/vein thrombosis
	Miscellaneous causes
	Blunt trauma

Table 9.3 (continued)

Glomerular causes	Non-glomerular causes
	Recent urinary tract instrumentation (e.g., catheterization, cystoscopy)
	Sexual trauma
	Vigorous exercise
	Sickle cell disease
	Loin pain hematuria syndrome

sis, as well as oliguria, renal dysfunction and often hypertension, are consistent with nephritic syndrome. Examples include lupus nephritis and membranoproliferative glomerulonephritis. Diseases like minimal change disease that cause nephrotic syndrome less commonly have hematuria but have increased proteinuria, hypoalbuminemia, and edema. See Chap. 5 for further information regarding proteinuria and glomerular disease.

Symptoms suggesting non-glomerular causes of hematuria include flank or abdominal pain, or lower urinary tract symptoms like frequency, urgency, or dysuria. Generally, RBCs on urine microscopy are isomorphic.

Neoplasms anywhere along the urinary tract may cause hematuria, including tumors of the kidney parenchyma, renal collecting system, ureter, bladder, prostate, and urethra. The majority of causes of microscopic hematuria are benign, with an underlying malignancy discovered in less than 5% of individuals who undergo a complete workup [7]. However, microscopic hematuria may be one of the first signs of advanced malignancy. There is an increased likelihood of malignancy in individuals who have also experienced gross hematuria [7]. Risks factors that may increase the likelihood of identifying a malignant lesion include older age, male gender, history of tobacco use, exposure to certain chemicals, dyes or carcinogenic agents, history of chronic indwelling catheter or tube and prior radiation exposure (see Chap. 4).

Vigorous exercise is implicated in some cases of microscopic hematuria, and it is considered to be a diagnosis of exclusion. Exercise-induced hematuria may occur in both contact sports, from direct trauma to the kidneys, as well as non-contact sports. Mechanisms by which this is thought to occur in the absence of trauma or contact include a relative decrease in renal blood flow that correlates with the need for increased flow to the skeletal muscles, heart, and lungs with heavy exercise. This may lead to hypoxia within the nephrons of the kidney, and the resultant increase in glomerular permeability can lead to an increase in red blood cells in the urine. This may be further contributed to by constriction of the efferent arterioles, which causes an increased pressure in the glomerular capillaries [8]. Additionally, bleeding may also originate from the bladder, which may result from repeated impact of the mobile posterior wall of the bladder against the fixed bladder base, as can occur in long-distance runners [8].

In cases of gross hematuria, the timing of the hematuria within the urinary stream may help to elucidate the source. Initial hematuria, occurring at the beginning of the urinary stream, suggests urethral origin of hematuria, like urethral stricture disease. Total hematuria, occurring throughout the urinary stream, typically suggests a bladder, ureteral, or renal source. Terminal hematuria generally results from bleeding of the lower urinary tract in the bladder trigone, bladder neck, prostate, or urethra [7].

Color of urine can also differentiate between glomerular versus non-glomerular causes. Glomerular causes of hematuria tend to produce brown, tea-colored, or "cola-colored" urine [8]. Gross hematuria that produces blood clots is virtually always non-glomerular in nature. The shape of the blood clot can indicate anatomic location of the source of bleeding. For example, long, thin clots suggest a renal or ureteral source of bleeding, while large, round clots, indicate a bladder source.

Returning to case 2, this patient most likely has obstructing urolithiasis given her flank pain and microscopic hematuria. Pyelonephritis should also be considered. A glomerular cause of hematuria is not suspected given symptoms, lack of proteinuria, and lack of RBC casts. Urine culture should be obtained, as should imaging of the genitourinary tract with renal ultrasound or CT.

Case 3: Evaluation of Microscopic Hematuria

A 70 year old female presents with recurrent dysuria and urinary frequency. She has a history of hypertension and coronary artery disease for which she takes metoprolol, aspirin, and clopidogrel. She has been treated empirically for two urinary tract infections over the past year. Urinalysis and urine microscopy are obtained.

What would be the next step in evaluation? Would evaluation differ if this patient presented with gross hematuria?

Urine dipstick & urine microscopy	
Component	**Result**
Color	Yellow
Appearance	Clear
pH	7.0
Specific gravity	1.020
Blood	1+
Glucose	Negative
Ketone	Negative
Protein	Negative
Bilirubin	Negative
Urobilinogen	Negative
Leukocyte esterase	1+
Nitrite	Negative
White blood cells (WBC)	10/hpf
Red blood cells (RBC)	25/hpf
Squamous epithelial cells	2/lpf
Casts	0/hpf
Bacteria	0/hpf

Indications for Evaluation of Hematuria

According to the American Urological Association (AUA) guidelines, a single urine examination demonstrating microscopic hematuria is considered sufficient for proceeding with further workup "in the absence of an obvious benign cause." [3] As RBCs may appear in the urine only intermittently, even in the presence of a malignant lesion, repeat testing to confirm the presence of microscopic hematuria is not required. Although positive dipstick testing for hematuria prompts follow-up microscopic evaluation, if microscopy does not confirm the presence of three or more RBCs/hpf, further evaluation is not necessary unless clinical suspicion is high, in which case repeat microscopy may be warranted.

When microscopic hematuria is attributed to a gynecologic or non-malignant urologic etiology, urinalysis should be repeated following resolution of the suspected cause, especially if hematuria has been a recurrent finding. For example, in the event that a urinary tract infection is suspected as the cause of microscopic hematuria, this should be confirmed with a urine culture, with repeat microscopic evaluation of the urine following an adequate course of treatment . It is considered prudent to repeat the UA at least 3 weeks after completion of treatment in order to allow sufficient time for the microsopic hematuria to resolve but also within a period of 3 months so as not to delay diagnosis. If hematuria persists, it should then be evaluated according to risk [3, 7]. If microscopic hematuria persists, evaluation should be performed, as this approach may limit delays in diagnosis. If menstrual bleeding is occurring at the time of sample collection, UA may be repeated after menstruation has ceased, or a catheterized specimen may be obtained. Of note, microscopic hematuria in patients taking anticoagulant and antiplatelet agents still necessitates a standard workup. Additionally, recent urinary tract instrumentation does not preclude the need for hematuria evaluation if hematuria is persistent after instrumentation.

Evaluation Protocols

See Fig. 9.2 for an algorithm for the workup of microscopic hematuria. The most recent AUA and Society of Urodynamics, Female Pelvic Medicine & Urogenital Reconstruction (SUFU) guidelines risk stratify patients with microscopic hematuria into low, intermediate and high risk groups based on risk factors for urologic malignancy, as shown in Fig. 9.3. Evaluation should be individualized based on findings on history and physical examination as well as assessment of these risk factors. A serum creatinine (with estimation of GFR) should be obtained to help identify underlying renal disease and guide choice of imaging if indicated. Presentation with gross hematuria classifies a patient as high risk for urologic malignancy and necessitates cystoscopy and imaging of the upper urinary tract [3].

The choice of imaging modality for evaluation of microscopic or gross hematuria takes into consideration multiple

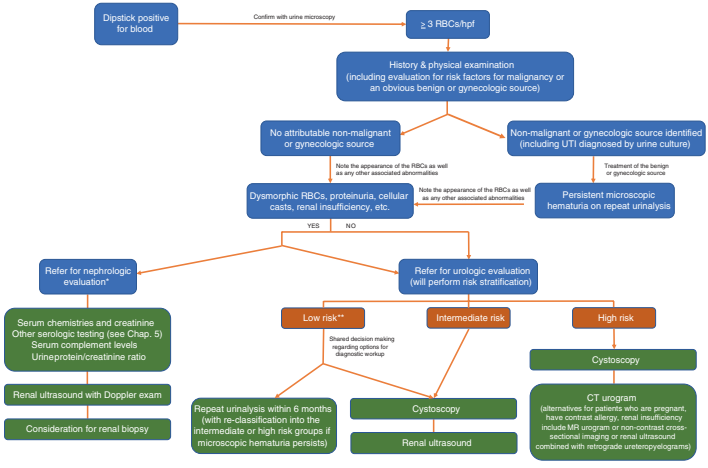


FIGURE 9.2 Algorithm for the evaluation of Microscopic Hematuria. *AUA guidelines recommend risk-based urologic evaluation even if nephrologic evaluation is performed for suspected medical renal disease. **For low risk patients, shared decision making is required for understanding the risks and benefits of hematuria evaluation. If a primary care physician is not comfortable having this discussion, urology referral should be made

Risk Stratification for Microscopic Hematuria Evaluation		
Low risk	Intermediate risk	High risk
women < age 50 OR men < age 40 AND <10 pack-year smoking history AND 3-10 RBC/hpf on UA AND no prior episodes of microscopic hematuria	women age 50-59 OR men age 40-59 OR 10-30 pack-years smoking OR 11-25 RBC/hpf on UA OR previously low risk with recurrent/persistent microscopic hematuria and no prior evaluation (with 3-25 RBC/hpf on UA) OR any additional risk factors for urothelial carcinoma (per the AUA guidelines, these "include but are not limited to irritative lower urinary tract voiding symptoms, history of cyclophosphamide or ifosfamide chemotherapy, family history of urothelial carcinoma or Lynch Syndrome, occupational exposures to benzene chemicals or aromatic amines, history of chronic indwelling foreign body in the urinary tract")	men ≥ age 60 OR women ≥ age 60 OR > 30 pack-years smoking OR > 25 RBC/hpf on UA OR previously low risk with recurrent/persistent microscopic hematuria and no prior evaluation (with > 25 RBC/hpf on UA) OR history of gross hematuria

Figure 9.3 Risk stratification for urologic malignancy per AUA/SUFU guidelines [3]

factors. Although renal ultrasound has a decreased sensitivity for detecting upper tract urothelial carcinoma compared to computed tomography (CT), the likelihood of this diagnosis is low in patients who do not have risk factors. Thus, ultrasound is considered the preferred modality in some patients. Benefits of ultrasound include low cost, lack of ionizing radiation and intravenous contrast; however image quality may be affected by technician and patient body habits.

Patients with gross hematuria or microscopic hematuria and risk factors should undergo a multi-phasic CT, unless contraindicated. CT urogram is the most sensitive and specific imaging study for this purpose and includes non-contrast and contrast phases, as well as a delayed excretory phase. Magnetic resonance urography (MRU) is an alternative imaging choice in patient with iodinated contrast allergy or renal insufficiency. When contraindications exist to the aforementioned imaging

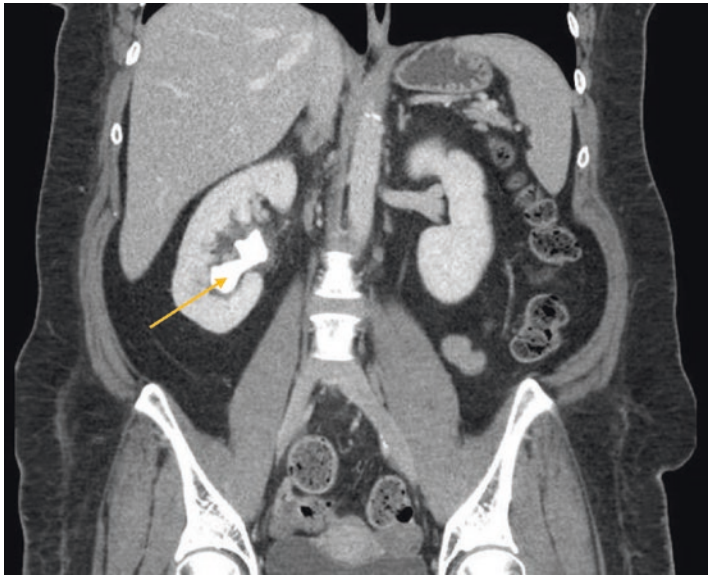


FIGURE 9.4 Non-contrast CT scan (coronal reconstruction) demonstrating partial staghorn stone (arrow) within the lower pole of the right kidney in a patient undergoing workup for microscopic hematuria. (Courtesy of University of Iowa, Department of Urology)

protocols (e.g., GFR less than 30), non-contrast cross-sectional imaging or renal ultrasound may be combined with retrograde ureteropyelograms to complete evaluation of the upper urinary tract [3]. There are some special considerations in certain patient populations. In patients with a family history of renal cell carcinoma or genetic syndrome associated with development of renal tumors (e.g., von Hippel-Lindau, Birt-Hogg Dube, tuberous sclerosis), upper urinary tract imaging should always be performed. Additionally, given that the majority of pregnant patients will fall into low or intermediate risk groups, renal ultrasound is the best study for pregnant women with consideration for CT or MRU after delivery if indicated [3]. Figures 9.4 and 9.5 show examples of upper urinary tract imaging commonly obtained by urologic providers.

As the majority of cancers diagnosed from hematuria evaluation are bladder cancers, which often are not detectable by

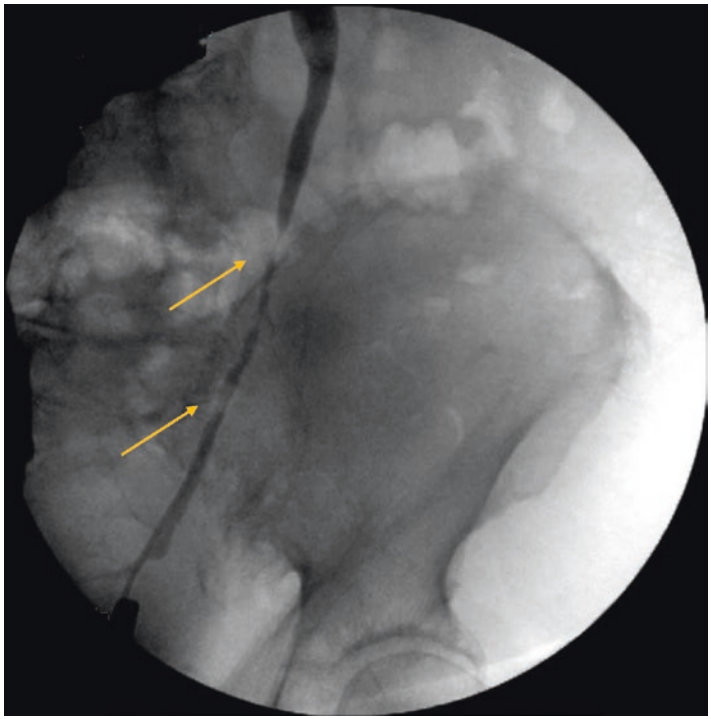


FIGURE 9.5 Retrograde ureteropyelogram showing filling defects in ureteral lumen (arrows). Subsequent ureteroscopy was performed, and at the location of the more proximal arrow, there was a papillary soft tissue mass. Note the dilation of the ureter proximal to this site as a result of the partial obstruction from the mass. (Courtesy of University of Iowa, Department of Urology)

imaging alone, cystoscopy remains an important diagnostic tool. Urologic evaluation of the lower urinary tract includes a cystoscopy in those considered intermediate or high risk, with consideration for cystoscopy in low risk patients based on clinical judgement and shared decision-making with the patient [3]. This remains the most reliable means of evaluating for lesions within the bladder and urethra. Patients considered low risk should be offered the option of proceeding directly to cystoscopy and renal ultrasound versus repeating the urinalysis within 6 months with plans to perform cystoscopy and imaging if hematuria persists. Persistent hematuria reclassifies a patient as intermediate or

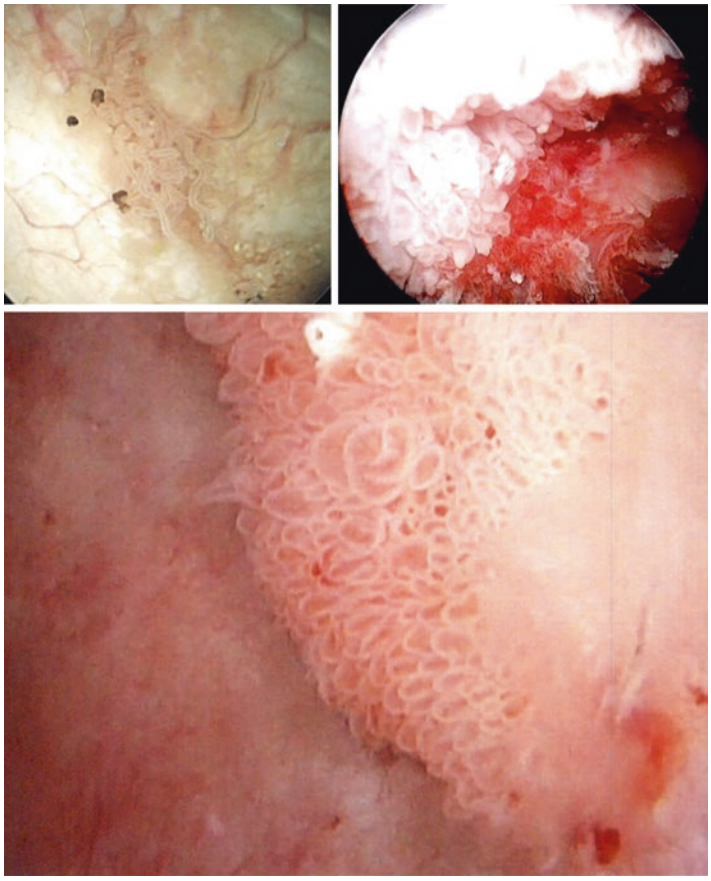


FIGURE 9.6 Photographs of various papillary bladder tumors as seen on cystoscopy; note the frondular appearance of urothelial carcinoma in the bladder with associated bleeding and friability. (Courtesy of University of Iowa, Department of Urology)

high risk. Figure 9.6 shows multiple bladder lesions consistent with malignancy, as found on cystoscopy.

Urine cytology is not recommended as part of the routine workup for hematuria, although this may be considered in select patient populations, including those with risk factors for malignancy and prior negative hematuria evaluation, particularly those with irritative voiding symptoms like urgency or dysuria [3, 7] (see Chap. 14).

Nephrology evaluation should be obtained when dysmorphic RBCs are present, or in the presence of other indicators of medical renal disease such as proteinuria, casts, or evidence of reduced GFR (e.g., abnormal serum creatinine). Renal biopsy is the only way to definitively diagnose a glomerular cause of hematuria, although this may not be necessary in all patients with microscopic hematuria. Nephrology may obtain further serologic studies and determine if biopsy is indicated. A random urine protein/creatinine allows for a quantitative measure of protein excretion [8] (See Chap. 5). The need for concurrent urology referral is controversial if the patient is diagnosed with glomerular disease, although it is still recommended by the AUA. Data is sparse on the incidence of concomitant glomerular and non-glomerular disease, but both referrals should be considered, especially in the presence of risk factors for genitourinary malignancy.

Specific guidelines for the management of hematuria in the setting of trauma are more complex and may take into account other variables, including the mechanism of injury, associated injuries and clinical status. Traumatic hematuria is therefore beyond the scope of discussion for this chapter and should prompt urologic consultation. Additional considerations are also made in pediatric patients with microscopic and gross hematuria (see Chap. 13).

Returning to case 3, the patient presented with several voiding complaints and was noted to have microscopic hematuria. The remainder of the urinalysis is notable for positive leukocyte esterase, as well as pyuria and bacteriuria.

It is important to note that although urinary tract infections may cause varying degrees of hematuria, bladder tumors may cause similar urinary symptoms. Women, in particular, may experience a delay in bladder cancer diagnosis, often being treated with multiple courses of antibiotics [9, 10]. *Accordingly, in settings in which hematuria and irritative voiding symptoms occur in the absence of a positive urine culture, it is prudent to consider early urology referral. Pelvic floor dysfunction could also cause this scenario. While awaiting urologic evaluation, a urine culture should be obtained, with appropriate treatment of infection. It is also important to note that the use of antiplatelet or anticoagulant medications does not preclude the*

need for further evaluation. Given this patient's risk factors for urologic malignancy, gross hematuria would warrant the same evaluation as microscopic hematuria.

Follow-Up and Management

Management of gross or microscopic hematuria is dictated by the clinical scenario, which varies significantly by cause. If a specialist is involved, he or she will outline an appropriate follow-up plan if necessary. Risk factors for both urologic and nephrologic disease (i.e., smoking cessation for bladder cancer) should be evaluated and minimized.

Protocols for repeat testing should be individualized. Patients who have completed a negative nephrologic and/or urologic hematuria evaluation should have a repeat UA performed within 12 months. This testing approach also applies to those with negative UA after treatment of a non-malignant urologic or gynecologic source. If this UA is negative, no specific follow up is warranted. If there is persistent or recurrent hematuria, repeat evaluation should be strongly considered [3]. There are some conditions that may cause persistent microscopic hematuria that do not require active treatment, like benign prostatic hyperplasia, vaginal atrophy, and pelvic organ prolapse. In these cases, shared decision-making should be used to decide whether to perform repeat evaluation, also taking into account new or changed risk factors. If there is a significant increase in the degree of microscopic hematuria, the development of gross hematuria or new symptoms, additional evaluation should be performed.

Summary

Patients may present with hematuria under a variety of circumstances, ranging from asymptomatic, incidentally detected microscopic hematuria to symptomatic or gross hematuria. It is important for providers to understand the possible underlying causes of hematuria and distinguish between them by performing a thorough history and physical examination and microscopic urinalysis. Most hematuria will require specialist evaluation unless definitively attributed to a benign cause, such as UTI.

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Chapter 10
Urine Microscopy: The Utility of Urinary Casts in Patient Care – Practical and Useful Tips for Busy Clinicians

Stephanie J. Houston, M. Lee Sanders, and Lyndsay A. Harshman

Objectives

- Describe the physiology of urinary cast formation
- Delineate the proper laboratory process for performing a urine microscopic analysis in order to view a urinary cast
- Discuss the classification of urinary casts and the relationship of casts to renal pathology

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Overview

Clinical urinary assessment should include a microscopic review of urine sediment for cast morphology. Casts are generally a marker of tubular injury and serve to represent the presence of a variety of disease processes. The presence of casts can also inform the healthcare provider about disease states and allow for monitoring in certain renal conditions.

The Physiology of Urinary Cast Formation

Urinary casts seen under the microscope tend to be cylindrical in shape because they form within the renal tubules (Fig. 10.1). The precise anatomic location for ongoing cast formation is the distal convoluted tubule and/or the collecting duct. Casts are generally not formed within the proximal convoluted tubule or loop of Henle. The reason for this anatomic distinction is that casts are composed of an external

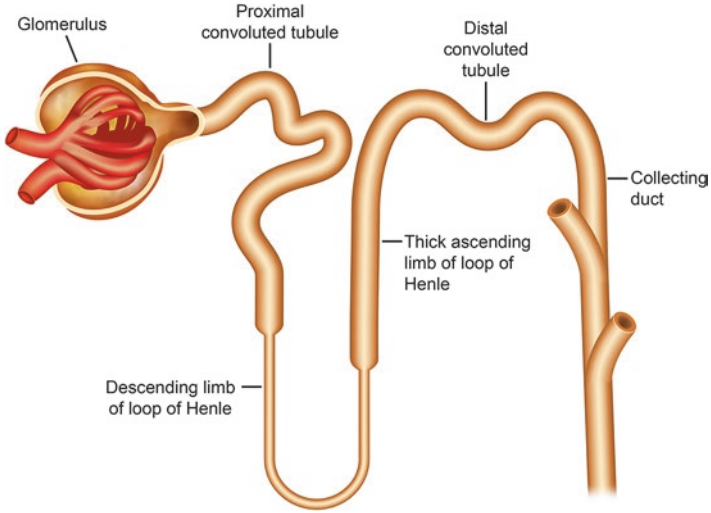

Figure 10.1 Cast formation occurs in the distal convoluted tubule and/or the collecting duct. (Courtesy of Teresa Ruggle, University of Iowa)

backbone of uromodulin (Tamm-Horsfall) protein which is secreted mainly in the distal convoluted tubule. Factors that favor cast formation are low flow rate/urinary stasis, high salt concentration and low urine pH [1].

The differentiation of casts in renal pathology primarily focuses on the internal material adhering to the uromodulin matrix. If glomerular and/or tubular damage has occurred, material resulting from this damage can be present within the tubule at the time of cast formation essentially trapping this material within the cast. Casts in these cases can provide a non-invasive "liquid biopsy" to aid in determining the cause of kidney injury and provide a snapshot of the milieu of the tubule at the time of cast formation [2]. It is important to note that even if the glomerulus is functioning properly and there is no tubular damage, casts may still form under the right physiologic conditions but these casts will be void of any internal material.

Case 1: Assessment of Urine Casts in the Clinical Setting

A 38 year old male presents to the emergency room after 3 days of nausea, vomiting and diarrhea. His labs indicate a mild acute kidney injury, but you feel this is likely due to volume depletion from poor oral intake. Your plan is intravenous fluids with an antiemetic and monitor for improvement with likely discharge home. When you mention the acute kidney injury, this is very concerning to him as he has a family history of kidney disease although he does not remember the exact etiology. You order a urine specimen for analysis in order to put him more at ease. At discharge, the patient informs you that his urine specimen has been sitting by his bed for the last 3 hours. He asks, "Will that alter the results of the urine test?"

The identification of urinary casts by microscopy is highly dependent on laboratory preparation of the specimen. Urine should be prepared in a systematic manner (see Chap. 3) for reliable assessment of casts. This preparation begins with the collection of a urine sample which is ideally a fresh clean catch specimen. Care must be taken in preparation of the

urine as casts may be disintegrated if the manual agitation or suction/expulsion is performed too vigorously [3].

A single drop of prepared urine is placed on a standardized glass slide with a cover slip. Examination by microscopy should begin on low power then transitioned to high power. Casts are generally most visible at the cover slip edges [3].

Urine specimens should ideally be analyzed within 2 hours of collection. Standing urine becomes progressively more alkaline over time as urea is broken down generating ammonia. This ammonia generation leads to an increasing alkaline environment which dissolves casts and promotes cell lysis. This is why urine that is allowed to sit for hours before microscopic analysis or urine obtained from a Foley bag is generally not acceptable for cast analysis. If immediate specimen examination cannot be performed, urine can be preserved by refrigeration for up to 6 hours before examination for casts [4].

Returning to case 1, you regrettably inform the patient that the delay in analysis of the urine specimen may indeed alter the results. You discuss that ideally a fresh clean catch specimen should be analyzed within 2 hours of collection. After additional discussion with the patient, the patient decides he will discuss the need for any additional testing with his primary care provider.

Case 2: Elevated Creatinine in a Young Female

A 15 year old female was admitted to the hospital for treatment of community acquired pneumonia with hypoxia. She was treated with ceftriaxone. Over the next 5 days, serum creatinine rose from 0.7 mg/dL to 2.8 mg/dL despite respiratory improvement. Urinalysis showed 1+ blood, 1+ leukocyte esterase, negative nitrite, and a freshly obtained sample examined immediately by microscopy showed a single white blood cell cast.

Urinary casts can be broadly classified as cellular, noncellular, and pigmented types. It is important to note that the presence of cells within a cast is specific to an intrarenal etiology (Table 10.1).

Table 10.1 Quick reference of commonly observed cellular and non-cellular casts

Cast type	Composition/appearance	Disease etiologies/clinical correlates
Cellular casts		
	RBC cast (red blood cells contained within the cast)	Associated with nephritic syndrome (see Table 5.4 in Chap. 5)
	WBC cast (white blood cells contained within the cast)	Associated with renal interstitial inflammation (pyelonephritis, acute interstitial nephritis, tuberculosis)
	rTEC cast (renal tubular epithelial cells contained within the cast)	Associated with tubular epithelial destruction (cytomegalovirus infection) and ischemic injury
Non-cellular casts		
	Hyaline cast (acellular cast that consists of solidified Tamm-Horsfall protein which appears transparent and empty)	Associated with volume depletion or renal disease (when several casts are present), but the presence of a small number of hyaline casts can be normal
	Granular cast (acellular cast resulting from either cellular cast degradation or inclusion of aggregate plasma proteins)	Associated with nephrotoxic and ischemic injury (acute tubular necrosis)
	Waxy cast (acellular cast that is the final stage of cellular cast degradation)	Associated with advanced chronic kidney disease
	Lipid cast (acellular casts that contain lipid droplets)	Associated with nephrotic syndrome (see Table 5.3 in Chap. 5)

Cellular Casts

Red blood cell (RBC) casts (Fig. 10.2)

The presence of RBC casts on microscopy usually indicates that glomerular injury is present. Dysmorphic red blood cells, also called acanthocytes, are also a characteristic marker of glomerular hematuria. The presence of RBC casts and dysmorphic RBCs can also be useful for surveillance of patients with known glomerular disease (i.e., lupus glomerulonephritis and small-vessel vasculitis) to gauge response to therapy and disease recurrence [3].

RBC casts consist of easily visible erythrocytes within a tubular cast matrix. Fresh RBC casts may appear visibly

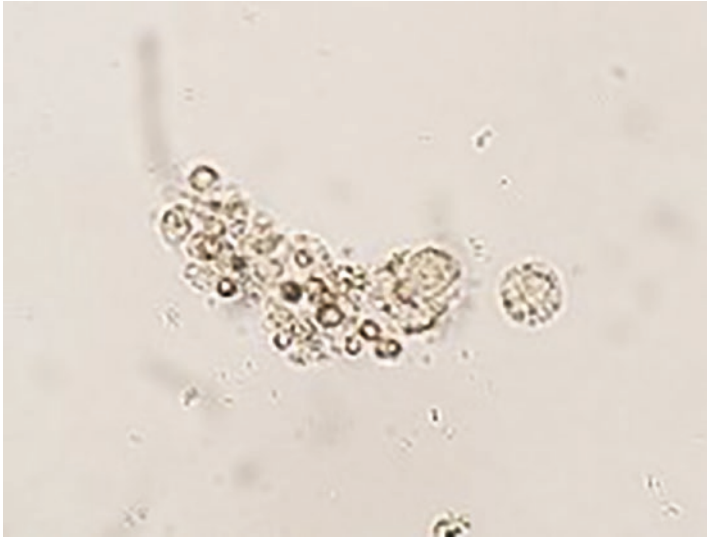

FIGURE 10.2 Red blood cell (RBC) cast. (Courtesy of University of Iowa, Division of Nephrology)

brown; however, with time, the heme pigmentation degrades within the erythrocyte and the coloration dissipates. Thus, an RBC cast may begin to appear like a granular cast as the cast ages and the erythrocytes lose cellular integrity [3].

White blood cell (WBC) casts (Fig. 10.3)

Documentation of WBC casts in the urine suggests the possibility of an interstitial kidney disease such as parenchymal infection, an inflammatory processes, pyelonephritis, and less often glomerulonephritis. WBC casts may also occur in the setting of acute interstitial nephritis (AIN); however, case series data document that only between 5% and 14% of patients with AIN have WBC casts in their urine sediment despite active clinical evidence of disease [5, 6].

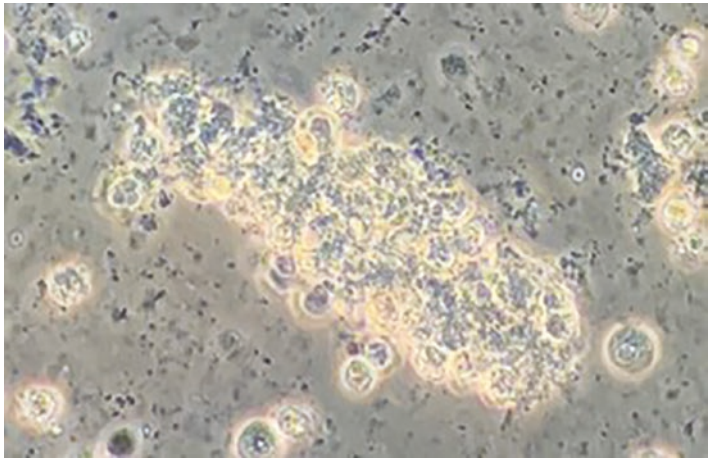

Figure 10.3 White blood cell (WBC) cast. (Courtesy of University of Iowa, Division of Nephrology)

Renal tubular epithelial cell (rTEC) casts (Fig. 10.4)

Formation of rTEC casts may occur in the setting of ischemic or nephrotoxic tubular injury leading to acute tubular necrosis with sloughing of the renal tubular epithelium. rTEC casts have also been observed in infections, such as cytomegalovirus, known to cause direct injury to renal tubules [3].

Returning to case 2, acute interstitial nephritis (AIN) was a leading diagnostic possibility for the acute kidney injury with ceftriaxone being the most likely drug responsible. Antibiotic therapy for the pneumonia was changed to levofloxacin. She also received IV steroids for the AIN with gradual improvement in serum creatinine from 2.8 mg/dL to 0.8 mg/dL over the next 5 days.

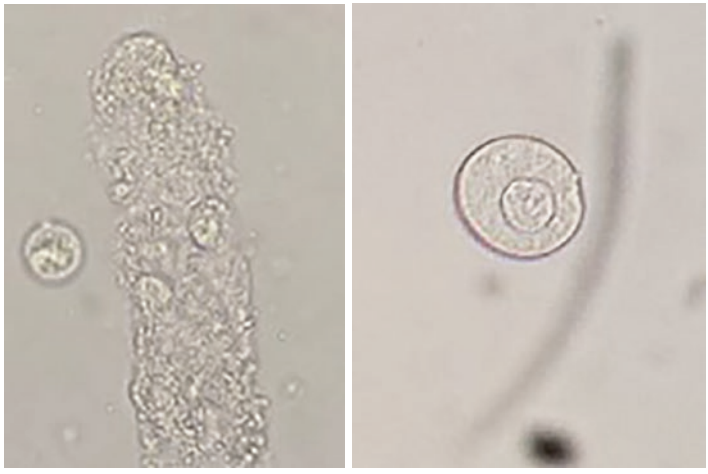

FIGURE 10.4 Renal tubular epithelial cell (rTEC) cast (left) and magnified renal tubular epithelial cell (right). (Courtesy of University of Iowa, Division of Nephrology)

Case 3: Acute Kidney Injury in a Hospitalized Patient

A 21 year old collegiate soccer player was admitted to the hospital after collapsing during soccer practice. She appeared severely volume depleted and received aggressive fluid resuscitation. Labs revealed acute kidney injury. A urine sample examined under microscopy revealed many hyaline and granular casts consistent with acute tubular necrosis (ATN). Over the next few days her serum creatinine peaked and then began to down-trend. What did the casts in this case tell us about the renal pathology?

Non-cellular Casts

Hyaline casts (Fig. 10.5)

Hyaline casts have a transparent, empty appearance on microscopy. An occasional hyaline cast (defined as less than three to five casts per high power field) in an otherwise bland

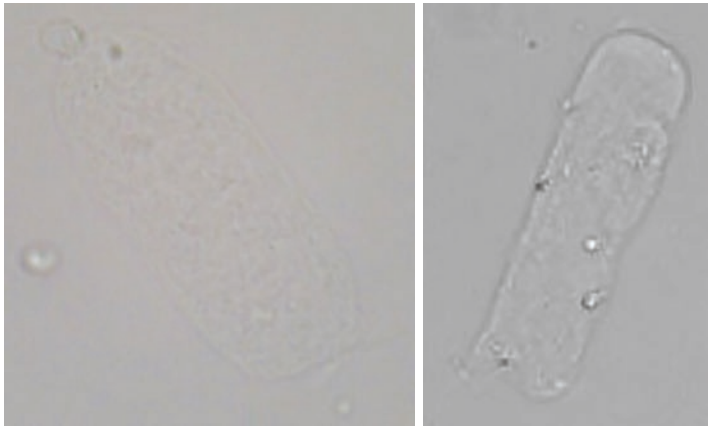

FIGURE 10.5 Hyaline casts. (Courtesy of University of Iowa, Division of Nephrology)

urine can be normal and occur in healthy individuals. The presence of numerous hyaline casts however is associated with renal hypoperfusion states, diuretic therapy, and concentrated urine [3].

Granular casts (Fig. 10.6)

One urinary marker of renal tubular injury is the presence of granular casts. These casts are composed of a cast matrix of uromodulin surrounding degenerated cellular casts. One variation of granular casts is that of a "muddy brown cast". This is a coarse, granular cast with a brown coloration due to tubular damage/necrosis. These muddy brown (granular) casts are thought to be pathognomonic of "acute tubular necrosis" (ATN) [3].

Waxy casts

A waxy cast is not necessarily an independent cast etiology but can be thought of as progression of granular cast degen-

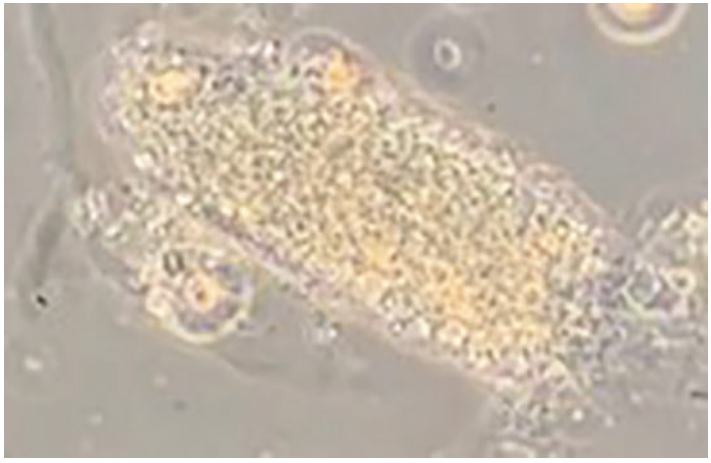

Figure 10.6 Granular cast. (Courtesy of University of Iowa, Division of Nephrology)

eration. Although nonspecific, these casts are often associated with advanced chronic kidney disease when seen on microscopy [3].

Lipid casts

In the setting of nephrotic-range proteinuria, the urine sediment may contain free lipid droplets. When these free lipid droplets become embedded in an uromodulin cast matrix, a lipid cast is formed [3].

Pigmented Casts

Pigmented casts can be formed either endogenously or exogenously. Endogenous formation of pigment casts may occur due to breakdown of cellular material such as hemoglobin (hemolytic anemia), myoglobin (rhabdomyolysis) or bilirubin (liver disease). Exogenous formation of yellow-orange pigment casts may occur secondary to drug ingestion. Phenazopyridine is the active ingredient in a widely used over the counter bladder analgesic and is the most notable drug to cause pigment cast discoloration [3]. A summary of pigment cast relevance in clinical application is found in Table 10.2.

Practical Clinical Use of Urinary Casts

The presence of urinary casts can trigger the clinician to specific diagnostic considerations in patient care. Given that prerenal AKI and ATN are the most common causes of AKI in hospitalized patients, differentiating between the two can be important in determining management.

Urine sediment is a useful tool to inform the diagnosis of ATN. Perazella et al. described a urinary sediment scoring system to distinguish ATN from prerenal AKI [7]. In patients with a high pretest probability of ATN, the presence of casts or rTEC inform a high positive predictive value and low

Table 10.2 Overview of commonly observed pigmented casts encountered in clinical practice

Cast type	Composition/appearance	Disease etiologies/clinical correlates
Bilirubin	Bile (acellular casts that contain bile/bilirubin)	Associated with severe liver dysfunction "Bile cast nephropathy" typically occurs when total bilirubin > 20 mg/dL Poorly understood mechanism as bile/bilirubin may contribute to renal tubular injury by direct toxicity, tubular obstruction, or a combination of both
Myoglobin	Myoglobin (globular, red-brown casts with coarse granular appearance that contain myoglobin)	Associated with rhabdomyolysis (necrosis of muscle cells) Serum creatinine kinase (CK) is usually five times the upper limit of normal Myoglobin can precipitate with Tamm-Horsefall proteins (especially in acidic urine) to form intracellular casts that obstruct the renal tubules Urinalysis will test positive for blood, but RBCs will be absent on urine microscopy (see Chap. 4)
Hemoglobin	Hemoglobin (acellular casts that contain hemoglobin)	Associated with hemolytic anemia or renal parenchymal bleeding May result from degradation of RBCs within a RBC cast (see Table 10.1) Heme pigment has a direct renal tubulotoxic effect Urinalysis will test positive for blood, but RBCs will be absent on urine microscopy (see Chap. 4)

negative predictive value for diagnosis of ATN. Conversely, in patient populations with a low pretest probability of ATN (e.g., prerenal AKI), a lack of casts or rTEC on urinary sediment lends a negative predictive value of 91%. Appropriate use of urine sediment in this case setting not only was a low-cost option to inform patient care but also prevented the patient from moving to an unneeded renal biopsy.

Similarly, a simplified acute kidney injury cast scoring index has been piloted to standardize urine microscopy and grade the degree (none, rare, moderate, sheets) of epithelial cell and granular casts present on urine microscopy. The scoring system was found to be reliable with good inter-observer agreement. The cast scoring index was then used to evaluate renal outcomes in patients with a clinical diagnosis of ATN. Patients without renal recovery had a higher cast scoring index as compared to patients who recovered kidney function [8].

Returning to case 3, the young female had both hyaline casts and granular casts on microscopic examination of the urine. The presence of many hyaline casts reflected poor renal perfusion likely from volume depletion. Her volume depletion was severe as the presence of granular casts reflected tubular injury. The granular casts were likely "muddy brown" in color signifying ATN. The serum creatinine in AKI due to mild to moderate ATN is usually noted to increase abruptly at first but then peak and level off followed by slow improvement. Severe ATN may require dialysis and have a prolonged recovery phase.

Case 4: Elevated Creatinine with Systemic Symptoms and Positive Urine Sediment

A previously healthy 13 year old female presented with intermittent fever, migratory erythematous rash of face and extremities, fatigue, headaches, and diarrhea. She was found on evaluation to have pancytopenia, positive antinuclear antibodies (ANA), and acute kidney injury with elevated creatinine. Urinalysis showed 2+ blood, 3+ protein with WBCs, RBCs, many hyaline casts, granular casts, and red blood cell casts on

microscopy. The elevated creatinine and presence of urinary sediment led to the decision to proceed with renal biopsy which revealed diffuse proliferative lupus nephritis. She was started on treatment with steroids and cyclophosphamide.

Lupus nephritis

Examination of urinary sediment in lupus nephritis can be informative not only during initial diagnostic work-up but also monitoring treatment response. Red blood cell and white blood cell casts are commonly found reflecting glomerulonephritis and inflammation, respectively. Hebert et al. studied a cohort of 17 known patients with systemic lupus erythematosus and found that the appearance of red blood cell or white blood cell casts was observed before or at the onset of 35 of 43 lupus nephritis relapses (sensitivity 81%) [9].

Returning to case 4, this young female with a new diagnosis of diffuse proliferative lupus nephritis should be longitudinally followed by checking not only her serum creatinine but also her urinalysis with microscopic evaluation.

Summary

The presence of urinary sediment can provide vital supplemental information regarding underlying renal pathology and inform practical patient management. **Case 1** focused on the importance of urine collection and preparation for potential cast visualization. **Cases 2** and **3** highlighted the importance of urinary cast evaluation to assist with diagnosis of the appropriate renal pathology. **Case 4** demonstrated that microscopic examination for casts can also assist with disease monitoring. In conclusion, a skillful and motivated urine sediment examination can assist with glomerular disease diagnosis as well as potentially allow for earlier detection of recurrent disease [10].

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Chapter 11
Urine Microscopy: Clouding Over – Bacteria, Yeast, Parasites and Zika

Bradley Ford, Wendy Fiordellisi, Victoria J. A. Sharp, and A. Ben Appenheimer

Objectives

- Discuss techniques for obtaining a proper sterile urine culture
- Recognize the limitations of urine Gram stains
- Recognize the indications for screening and treating asymptomatic bacteriuria
- Explain the diagnostic tests used to diagnose trichomoniasis and genitourinary schistosomiasis
- Indicate the exposures associated with urinary *bacillus Calmette-Guérin* (BCG) infection

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- Differentiate the diagnostic tests utilized in diagnosing genitourinary tuberculosis
- Recognize the importance of the clinical setting and risk factors when interpreting yeast in the urine
- Identify indications and techniques for Zika virus testing

Definitions
Vaginitis: *Inflammation or infection of the vagina.* Symptoms include pruritus, vaginal discharge, or vaginal odor. Common causes include *Candida* (aka 'yeast infection'), *Trichimonas* and bacterial vaginosis.
Urethritis: *Inflammation or infection of the urethra.* Symptoms include dysuria, frequent urination and urgent urination. This can be associated with sexually transmitted infections and can be difficult to differentiate from acute cystitis based on symptoms alone.
Cystitis: *Inflammation or infection of the bladder, often caused by a bacterial infection.* Symptoms include urinary urgency, burning with urination, increased frequency of urination, hematuria, and pelvic discomfort.
Asymptomatic bacteriuria: *The isolation of a determined quantitative number of bacteria in a urine specimen obtained from a person without signs or symptoms of a urinary infection* [1, 2].

Overview

Urine microscopy, whether done by Gram stain, concentrated wet-mount, or automated microscopy, is an attractive diagnostic procedure because of its rapidity and potential for narrowing the differential diagnosis to particular kinds of infectious etiologies. This chapter reviews the performance characteristics of microscopic examination in the case of typical uropathogens as well as special cases such as fungal and parasitic infections of the urinary tract.

Case 1: Bacterial Urinary Tract Infection (UTI)

You are seeing a 32 year old female in clinic with concern for a UTI. She has never had a UTI before but is complaining of dysuria, urinary urgency, and urinary frequency.

Urine dipstick & urine microscopy	
Component	**Result**
Color	Yellow
Appearance	Cloudy
pH	6.0
Specific gravity	1.015
Blood	Negative
Glucose	Negative
Ketone	Negative
Protein	Negative
Bilirubin	Negative
Urobilinogen	Negative
Leukocyte esterase	2+
Nitrite	Positive
White blood cells (WBC)	41/hpf
Red blood cells (RBC)	0–2/hpf
Squamous epithelial cells	3/lpf
Casts	0/hpf
Bacteria	0/hpf

Based on this information alone, what is the most likely pathogen causing her UTI?

Uropathogens are primarily Gram-negative bacteria with *E. coli* predominating (Table 11.1). Gram-positive uropathogens such as *Streptococcus agalactiae, Staphylococcus saprophyticus, Staphylococcus aureus, and Enterococci* share with Gram-negative uropathogens their ability to express adhe-

Table 11.1 Prevalence of uropathogens in adult uncomplicated, adult complicated, and pediatric UTI [3–5]

Organism	Gram stain appearance	Uncomplicated UTI, adult (%)	Complicated UTI, adult (%)	Pediatric*(%)
Escherichia coli	GNR	75	65	80
Klebsiella spp.	GNR	6	8	8
Staphylococcus saprophyticus	GPC	6	None	None
Enterococcus spp.	GPC	5	11	2
Streptococcus agalactiae (GBS)	GPC	3	2	[a]3
Proteus spp.	GNR	2	2	4
Pseudomonas aeruginosa	GNR	1	2	2
Staphylococcus aureus	GPC	1	3	[a]3
Candida spp.	Yeast	1	7	[a]3

(Courtesy Dr. Bradley Ford, University of Iowa, Department of Pathology) *GNR* Gram-negative rods, *GPC* Gram-positive cocci, *GBS* group B strep. Choice of antibiotic will depend on local susceptibility patterns and patient history. Recommend consulting local antibiogram and previous patient cultures

[a]Collectively, *Staphylococci, Streptococci*, and various Gram-negative rods, other than *E. coli* and *Klebsiella*, cause 3% of pediatric UTIs

*The pediatric values shown are approximations of the adult pathogens from different sources, and therefore do not add precisely to 100%

sive factors that promote binding to uroepithelial and kidney cells along with abiotic surfaces. Gram-negative organisms tend toward higher virulence and most often express toxins, immune evasion molecules, and other virulence factors that promote their success as uropathogens [6]. Pyelonephritis is caused primarily by these same uropathogens and descending UTI can occur in special cases, most commonly with *Salmonella, S. aureus and Candida spp.,* and rarely with other disseminated infections with organisms such as *Cryptococcus*.

About 1 in 5 urine cultures on average are contaminated [7]. Contamination with stool creates false positive culture results with potential uropathogens such as *E. coli*, while contamination with vaginal flora introduces lactobacilli, streptococci and other bacteria that are more easily identified as contaminants. Attention to collection technique, specimen transport, and acquisition of a straight catheter specimen when clean collection is in doubt will provide the most reliable sample (see Chap. 3).

Kidney stones are a uniquely difficult situation for medical management (see Chap. 12). Urea-splitting organisms such as *Proteus* and coagulase-negative *Staphylococci* commonly form stones, though other hard-to-identify urea splitters and *Ureaplasma urealyticum* (which cannot be routinely cultured) can as well. Culture of stones themselves is not routinely done but may be useful [8]. In many cases the number of colonies of urea-splitting organisms recovered in culture does not meet the usual threshold for workup, and alerting the laboratory that you are looking for such pathogens will generate a more complete identification and susceptibility testing profile.

Susceptibility testing at present does not account for the concentration of antibiotics achievable in the urine except for nitrofurantoin and fosfomycin [9]. As some drugs typically used to treat uncomplicated UTIs concentrate in the urine, results do not correlate well with clinical outcomes in these cases. Therefore, susceptibility testing should be just one of many factors taken into consideration when selecting an antibiotic to treat an uncomplicated UTI. Susceptibility test-

ing is explicitly recommended against for most Gram-positive organisms, including *S. saprophyticus*, but may be performed in special situations by request.

Overview of Laboratory Testing

The Clinical Laboratory Improvement Amendments of 1988 (CLIA) set rules for how laboratory testing should be performed, and who can perform it. Laboratory tests are broadly divided into three categories: "high complexity", "moderate complexity" and "waived". "High complexity" and "moderate complexity" tests can be performed with strict oversight; "waived" tests can be performed by anyone with minimal training as long as manufacturer instructions and good laboratory practices are followed. Because non-waived testing requires a CLIA certificate and adherence to CLIA quality standards as defined by the Code of Federal Regulations (including undergoing extensive inspections), waived tests are more accessible to independent clinics. Costs of these tests accordingly vary widely, from a few dollars to hundreds of dollars apiece depending on the test, whether further workup is required, and whether the test is performed in-house or contracted to a fee-for-service laboratory (see Chap. 2).

The Food and Drug Administration (FDA) maintains a list of waived tests [10] which includes dipstick testing and simple tests for trichomoniasis. Wet-mount "provider-performed microscopy" (PPM) procedures such as examination of a direct specimen for *Trichomonas*, or for unstained elements such as bacteria and yeast, require a CLIA certificate issued by the Centers for Medicare and Medicaid Services. This makes even simple, unstained microscopy a level of testing that is beyond the capability of most outpatient clinics. Essentially all procedures in this chapter are therefore classified as high complexity and are typically done in centralized laboratories with trained personnel. For unusual diagnoses that are not time critical (parasites and mycobacteria), refer-

ence laboratory testing from commercial providers is available.

Urine Culture Collection Best Practices

Urine samples are broadly divided into sterile (suprapubic aspiration as gold standard, with straight catheterization a more practical alternative) and nonsterile (clean catch midstream urine being the most typical collection). In the laboratory, sterile samples are plated using ten times the amount of urine to increase sensitivity. Specificity (i.e., lack of contamination) follows from the quality of the sample, which is higher with more standardized and invasive sterile urine collections. A great deal of attention has therefore been paid to optimizing quality of clean catch urine samples.

Broad surveys [7, 11] have defined refrigeration and/or preservative tubes, male sex, outpatient status, provision of written and verbal instructions, not pre-screening urine with dipstick testing, and central processing of samples (as opposed to handling of one urine sample by multiple laboratories for different tests) as factors associated with a lower likelihood of contamination. In practice, refrigeration and/or preservative tubes and provision of instructions for collection are the only easily modified factors at the point of care, apart from subjecting the patient to a straight catheterization.

The elements of a clean catch midstream urine collection are, broadly, attention to sterility and handwashing, perineal cleansing, labial spread or foreskin retraction, and voiding a small amount into the toilet before catching a midstream urine into a sterile container such as a urine cup [12–14, reviewed in 15]. From there, the sample may be refrigerated within half an hour and/or transferred to a preservative tube (see Chap. 3). There are several problems with clean catch collection: 1) it is physically difficult to perform, and provision of accurate written and verbal instructions is not always done because of complexity and barriers to communication, 2) a first morning urine culture is almost never obtained under these circumstances but

is best for non-culture diagnostic testing such as nitrite, and 3) a first void (as opposed to a midstream) urine is often contaminated for culture but is necessary for testing for many sexually transmitted infections such as chlamydia or gonorrhea, making diagnosis of an infection difficult to address adequately with one urine sample.

Gram Stain

Gram stain is a technique that is universally performed in microbiology laboratories. It relies on the ability of Gram-positive bacteria to take up crystal violet stain (blue-purple) which is trapped in Gram-positive but not Gram-negative cells with iodide. Gram-negative cells are stained pink-red with a safranin counterstain in the last step, and Gram stained slides containing primary patient sample or a smear of cultured organism are then examined at high power to determine the presence, color/shape and quantity of bacterial cells as well as the presence and approximate degree of neutrophil response to infection. Gram stain of unspun urine samples using a calibrated 10-microliter amount of urine and often a calibrated-circle slide has been promoted as a means of rapidly ruling out UTI in difficult cases, primarily pediatric patients. For example, Williams and colleagues [11] showed that Gram stain had a sensitivity of about 91%, which compared similarly to combined leukocyte esterase and nitrite (88%). Given the similar sensitivities, it follows that Gram stain is unlikely to detect many more cases than dipstick analysis alone. In fact, in a pediatric population Cantey et al. showed that over 1000 Gram stains would have to be done to detect one additional case of UTI over leukocyte esterase and nitrite analysis [12]. One possible benefit of urine Gram stain is the ability to immediately obtain microbiologic information about the potential pathogens based on their gram staining and morphology [13] (Fig. 11.1).

An additional complication of Gram stain is that it is high-complexity testing according to CLIA and therefore cannot be done in most clinics, which have only a moderate complexity provider-performed microscopy certification, if any. In

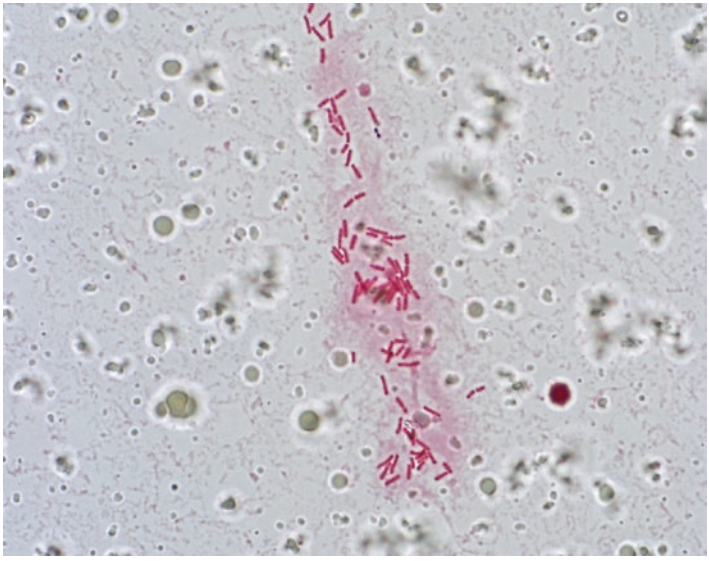

FIGURE 11.1 Gram stain (100× objective) illustrating *E. coli* found in the direct Gram stain of urine collected from a woman with typical symptoms of UTI. A likely contaminant (Gram-positive diplococci resembling *Enterococcus spp.*) is present at top center. (Courtesy of Dr. Bradley Ford, University of Iowa, Department of Pathology)

contrast, dipstick is considered a "waived" test that can be performed essentially everywhere with minimal additional training. In addition, Gram stain is more labor intensive than dipstick testing. Therefore, direct urine Gram stains are rarely performed in clinical practice (not to be confused with Gram stains of organisms grown in urine cultures, which do have clinical utility and are routinely run in high-complexity labs).

Manual and Automated Microscopy

Manual microscopic analysis involves creation of a ten-fold concentrated sediment that is examined at 400× magnification for casts, crystals, bacteria, and other formed elements.

"Particle analyzer" systems such as the Sysmex UF-100 and Beckman Coulter iq200 [14, 15] have been in use since the mid-1990s and more recent iterations such as the UF-1000i [16, 17] have a specific channel for the detection of bacteria. Bacteria are very small, which presents a challenge for optical detection and distinction from debris. A typical approach to utilization of these instruments as screening tests for bacteriuria maximizes sensitivity as a rule-out for UTI but compromises specificity such that about one in four samples test false-positive [17]. Unlike urine Gram stain, these automated instruments (typically available only in hospital settings) are not labor-intensive to use and can accurately perform counts of white cells, red blood cells, and epithelial cells as well as other formed elements of the urine in automated or semi-automated fashion. These instruments are therefore a valuable way to triage patients into more- or less-likely categories regarding UTI status, as well as to triage toward other diagnoses related to other formed elements in the urine such as white and red cells [18].

The presence of bacteria on automated microscopy seems to perform similarly to urine dipstick results [19, 20]. It is often used in conjunction with other diagnostic tests in evaluating for UTIs, most often nitrites and leukocyte esterase, and can serve as a convenient automated substitute for dipstick testing in centralized, high-complexity laboratories.

Returning to case 1, urine culture returned positive for E coli, the most common cause of urinary tract infections. The patient was treated with nitrofurantoin with improvement of her symptoms.

Case 2: Asymptomatic Bacteriuria

You are seeing a 32 year old female in your clinic today for a routine physical. She is healthy and has no symptoms. Your new nurse collected and sent a UA.

Urine dipstick and urine microscopy	
Component	**Result**
Color	Yellow
Appearance	Cloudy
pH	6.0
Specific gravity	1.015
Blood	Negative
Glucose	Negative
Ketone	Negative
Protein	Negative
Bilirubin	Negative
Urobilinogen	Negative
Leukocyte esterase	2+
Nitrite	Positive
White blood cells (WBC)	20/hpf
Red blood cells (RBC)	0–2/hpf
Squamous epithelial cells	1/lpf
Casts	0/hpf
Bacteria	0/hpf

Should your nurse have collected a urine sample as part of the patient's routine physical exam? Now that you have these results, should you treat the patient for a UTI?

Asymptomatic Bacteriuria

Urine has traditionally been considered a sterile substance; however, urine can be a good medium for growth of bacteria that enter the urinary tract. While this sometimes leads to infections of the bladder, lower urinary tract, and/or kidneys, bacteria can be found in the urine of a person without dys-

uria, urinary frequency, urinary urgency, suprapubic pain, flank pain, or fever. Urine is now also thought to have its own microbiome consisting of a resident bacterial community, some of which should not be taken as evidence of infection [21]. "Asymptomatic bacteriuria" is defined as the isolation of a determined quantitative number of bacteria in a urine specimen obtained from a person without signs or symptoms of a urinary infection [1, 2].

Asymptomatic bacteriuria can be diagnosed only if the urine sample is collected, stored, and transported in a way to minimize contamination and bacterial growth (see Chap. 3). Bacteriuria is diagnosed in women if a bacterial strain in counts of $\geq 10^5$ cfu/mL is isolated from two consecutive voided urine specimens [1, 2]. This two specimen recommendation comes from literature which states that 10–60% of women who initially have a positive specimen do not continue to have a positive specimen on repeat screening. A second specimen is generally not collected in practice before treatment. In men, a bacterial strain isolated in counts of $\geq 10^5$ cfu/mL from one voided specimen is sufficient. In a catheterized specimen from a male or female, bacterial counts need only be $\geq 10^2$ cfu/mL. Pyuria, or white blood cells in the urine, can exist in both infectious and non-infectious conditions. Pyuria coinciding with asymptomatic bacteriuria is not an indication for treatment.

Asymptomatic bacteriuria is common, particularly in certain populations. In women, the prevalence of asymptomatic bacteriuria increases with age, up to 16% in women aged 70 or older [22]. Women who are sexually active have more bacteriuria than those who are not, and diabetic women have more bacteriuria than nondiabetic women [23]. Asymptomatic bacteriuria is rare in young men, but up to 19% of men >70 years old have asymptomatic bacteriuria [22]. Nearly 50% of elderly patients in long-term care facilities [22] and over 50% of patients with spinal cord injuries have asymptomatic bacteriuria [24]. Prevalence is highest in patients with long-term indwelling urinary catheters, practically 100% of whom have bacteriuria [25]. Their urine cultures are often

polymicrobial and grow *Pseudomonas aeruginosa* and urease-producing organisms. In addition to antimicrobial therapy sometimes not providing a benefit, there are risks associated with overtreatment including antimicrobial resistance, adverse drug effects, *Clostridioides difficile* infection (CDI), and UTI shortly after therapy [2].

Screening persons for asymptomatic bacteriuria is appropriate if there is a risk of adverse outcomes associated with asymptomatic bacteriuria that can be prevented with antimicrobial treatment. The Infectious Diseases Society of America has developed evidence-based recommendations for which patients to screen and treat for asymptomatic bacteriuria (Table 11.2) [2].

Returning to case 2, based on guidelines, a UA in an asymptomatic healthy female should not have been obtained. Since she is asymptomatic and otherwise healthy, she warrants no further evaluation or treatment.

Case 3: Urethritis/Vaginitis with Parasites

A 23 year old sexually active female comes to your clinic complaining of vaginal discharge. In the past 6 months she has had multiple sexual partners and does not reliably use condoms. The discharge is significant and grey in appearance, but is without a significant odor. A 10% potassium hydroxide prep (KOH prep) is done which causes a foul fishy odor. A saline wet prep is done immediately and multiple motile flagellated protozoans are seen. What is the most likely diagnosis and what is the sensitivity of the wet prep in this case?

Two trichomonads can be found in urine samples: *Trichomonas hominis* and *Trichomonas vaginalis*. These species are practically impossible to differentiate by morphology yet have vastly different clinical significance. *T. vaginalis* is a pathogen and a cause of urethritis and/or vaginitis while *T. hominis* is a nonpathogen whose presence reflects stool contamination. Trichomonads are motile and have a lateral

Table 11.2 Indications to screen and treat for asymptomatic bacteriuria

Patients in whom it is indicated to screen and treat for asymptomatic bacteriuria	Patients in whom it is not recommended to screen or treat for asymptomatic bacteriuria	Patients in whom it is unclear whether to screen or treat for asymptomatic bacteriuria
Pregnant women: Screen at least once during early pregnancy. Treat with antimicrobial therapy for 4–7 days Patients undergoing endoscopic urologic procedures during which mucosal trauma is anticipated: Screen prior to procedure. If positive, culture and target antimicrobial therapy, short course (1–2 doses) prophylaxis initiated 30–60 min prior to the procedure recommended	Infants and children Premenopausal, non-pregnant, healthy postmenopausal women Patients with diabetes Elderly, either living in the community or in a facility Catheterized patients whose catheter remains in place <30 days or long term Patients with renal transplant >1 month or non-renal solid organ transplant (SOT) Patients with spinal cord injury Patients undergoing elective non-urologic surgery Patients undergoing surgery for placement of artificial urethral sphincter or penile prosthesis implant and those with implanted urologic devices should not be screened but should receive pre-op prophylaxis	Elderly with functional/cognitive Impairment, with delirium or fall, no GU symptoms or systems signs of infection: Assess for other causes and observation. If fever and systemic signs of severe infection without localizing source, treat with broad spectrum antibiotics directed at urinary and non-urinary sources High-risk neutropenia (absolute neutrophil count <100 cells/mm^3): ≥7 days duration after chemotherapy Patients at the time of indwelling catheter removal

Adapted from [1, 2]

membrane and flagella that make them easily apparent in fresh sample; however, because of their size (15 micrometers) they have a tendency to be obscured by WBCs and epithelial cells. Further, at 10 minutes after collection 20% of samples will be false negative due to loss of motility and alteration of morphology. Routine urine microscopy is capable of detecting *Trichomonas* but because transport time is essentially always delayed beyond the time where they are morphologically recognizable, its sensitivity is very low. At 2 hours most samples will be false negative by microscopy [26], rationalizing the trend away from wet-mount microscopy for diagnosis of *Trichomonas* infection [27]. One study showed a sensitivity of wet mounts in detecting trichomonads of just 60–70% when compared to culture-based methods [28]. Molecular techniques can also distinguish *T. vaginalis* and *T. hominis* and are more sensitive than wet-mount microscopy [29], further rationalizing a trend away from microscopy. Recent CDC guidelines express a preference for amplified nucleic acid methods (e.g. polymerase chain reaction (PCR)) over culture, non-amplified methods, and antigen testing for diagnosis of *Trichomonas* infection [30]. When ordering *Trichomonas* testing, it may be clear what method the test employs to detect *Trichomonas*; if not, providers should query the laboratory as laboratory methods to diagnose *Trichomonas* vary greatly in their effectiveness.

Another parasitic infection that can occasionally be detected in the urine is schistosomiasis, which is caused by trematode flukes of several species, only one of which *(S. haematobium)* has a tropism for the genitourinary tract. This species commonly excretes eggs into the bladder wall, causing painless hematuria. Infection with this species of parasite has been associated with an increased risk of bladder cancer [31]. In endemic areas (sub-Saharan Africa and areas along the Nile river), urine dipstick analysis for blood is a sensitive test for *S. haematobium* infection [32]. In nonendemic areas where prior probability of infection is low, the finding of hematuria on microscopy or dipstick analysis is less specific and diagnosis is made by urine microscopy looking for eggs (Fig. 11.2).

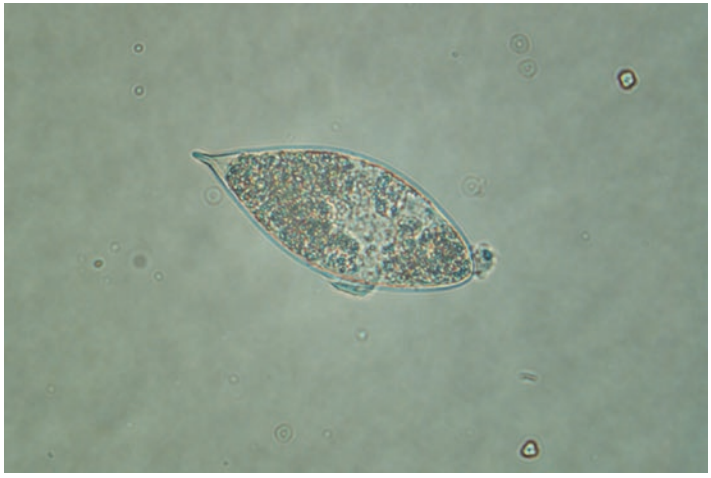

FIGURE 11.2 *Schistosoma haemotobium* egg (with characteristic terminal spine at left) observed in the concentrated, unstained wet mount of urine collected from a 16 year old Ethiopian patient with painless hematuria. 10× objective; the egg measures approximately 150 microns in length. (Courtesy of Dr. Bradley Ford, University of Iowa, Department of Pathology)

For diagnosis of schistosomiasis by detection of eggs in urine, the time of collection should be near noon and the minimum volume for analysis by centrifugal concentration or filtration should be 10 ml [33, 34] *S. haematobium* secretes eggs according to a circadian rhythm, with a peak around noon. One presumes that this is tied to the likelihood of the host being up and about at noon and proportionally more likely to urinate the eggs into a body of water containing its intermediate host, snails of genus *Bulinus*. Microscopy for schistosomes is an unusual specialty test that is unlikely to be performed in many laboratories. Before ordering, physicians should consult their laboratory for the best way to order the test and collect and transport the specimen.

Returning to case 3, this clinical presentation is consistent with Trichomonas vaginalis. This tends to lead to a profuse

discharge that can have variable color and either a mild fishy odor or no odor at all. Application of KOH can lead to an increase in this fishy odor. This case was diagnosed based on the clinical features and positive saline wet prep (which is specific but only has a 60–70% sensitivity as above). She was treated with 500 mg of metronidazole twice daily x 7 days.

Case 4: Urinary Tract Disease due to *Mycobacterium*

An 85 year old man with multiple medical problems, including a history of bladder cancer, presents to your clinic for the first time, complaining that he has had a UTI "for a year" but is otherwise in his usual state of health. He reports that he is here today because he has tried several other doctors who placed him on "several different" antibiotics to no effect. When asked about urine culture, he says that he has given a sample each time and "nothing came of it". What element of this man's history would be most useful to elicit?

This is a case consistent with cystitis due to biological treatment of bladder cancer with *bacillus Calmette-Guérin* (BCG). BCG is a member of the mycobacterium tuberculosis family, and when administered into the bladder can cause a variety of late-presenting infections. In this case, the patient has presented with a common therapy-related infection long after his last contact with his treating urologist, necessitating a careful history before ordering a diagnostic mycobacterial urine culture.

BCG is an attenuated strain of *Mycobacterium bovis*, a *Mycobacterium tuberculosis* complex species, that originated as a vaccine strain and was later adapted for use as immunotherapy against non-muscle-invasive urothelial cancers [35]. Intravesical instillation of live attenuated BCG is used to treat such cancers but is prone to causing chronic cystitis and sterile pyuria [36] which must be treated as an infectious pro-

cess when the symptoms persist beyond 48 hours after BCG instillation. In areas where tuberculosis is not endemic, genitourinary infection with mycobacteria is otherwise rare, making the decision to order mycobacterial culture of the urine dependent on careful elicitation of a history of intravesical BCG therapy, often in the distant past. Because BCG is a member of the *Mycobacterium tuberculosis* complex, mycobacterial culture is the diagnostic test of choice and these should be incubated for up to 6 weeks given the slow growth of this organism.

In patients who come from areas endemic for tuberculosis (TB), genitourinary tuberculosis is a common cause of extrapulmonary TB [37] (see Chap. 8). The historical gold standard for testing for genitourinary TB is an Acid-Fast Bacilli (AFB) culture but PCR is now being utilized as well. While not as sensitive, PCR is much faster than AFB cultures (which can take 6 weeks or more) [38]. However, urine TB PCR is not approved by the FDA for detecting TB in the urine and it is not commercially available. First morning voided specimens for AFB smear are sometimes done but these have a low sensitivity [37] and do not distinguish between TB and nontuberculous mycobacteria, making them less useful, especially when PCR is available.

Because mycobacterial numbers are low in urinary tract infection and stains are prone to false-positive results, Gram stain will not detect mycobacterial infection and other stains are not commonly performed. Because routine culture will not detect slow-growing mycobacteria, these diagnoses are only likely to be made if high clinical suspicion results in order of an AFB culture.

Returning to case 4, this man was diagnosed with BCG cystitis secondary to biologic treatment for his bladder cancer. This organism is intrinsically resistant to pyrazinamide and treatment was initiated with rifampin, isoniazid, and ethambutol pending additional susceptibility testing.

Case 5: Candiduria

You receive a call from a nursing home regarding an 82 year old female with multiple medical problems including diabetes with a neurogenic bladder. She has recently had difficulty with urinary retention and has had an indwelling urinary catheter for the last 4 weeks. The nurse noted a foul smell of her urine today and sent off a urinalysis and urine culture.

Urine dipstick and urine microscopy	
Component	**Result**
Color	Yellow
Appearance	Cloudy
pH	6.0
Specific gravity	1.015
Blood	Negative
Glucose	Negative
Ketone	Negative
Protein	Negative
Bilirubin	Negative
Urobilinogen	Negative
Leukocyte esterase	1+
Nitrite	Negative
White blood cells (WBC)	45/hpf
Red blood cells (RBC)	0–2/hpf
Squamous epithelial cells	1/lpf
Casts	0/hpf
Bacteria/other	Yeast present

What is the most likely pathogen based on that microscopy? What is the clinical significance of this finding? What features of the history could affect that clinical significance?

The significance of yeast in the urine found on microscopy or culture depends on the presence of symptoms and certain risk factors. Candiduria (*Candida* species in urine, regardless of clinical significance) is common due to contamination, colonization, ascending infection and (rarely) descending infection arising from hematogenous spread to the bladder. Treatment of asymptomatic candiduria is recommended only in special circumstances such as extremely low birth weight, neutropenia, or prior to certain urological procedures [39]. In many cases, removal of indwelling catheters results in resolution of asymptomatic candiduria. Most cases of ascending infection occur in the setting of urinary obstruction or the presence of foreign bodies. Given poor urinary penetration of echinocandins, established treatment options for symptomatic candiduria (same symptoms associated with bacterial UTI, namely dysuria, polyuria, urniary urgency and suprapubic or flank pain) are essentially limited to fluconazole, flucytosine, and amphotericin B. However, even with poor urinary penetration of the drugs, the reports on the efficacy of caspofungin and micafungin are mixed [40], possibly because of uncertainty over whether cases of candiduria represent true infections or transient self-resolving colonization. The specific choice of the drug depends on susceptibility patterns of different *Candida* species and the comorbidities of the patient.

Because susceptibility testing for *Candida spp.* is not routinely done and breakpoints do not account for urinary excretion of drugs, choice of therapy is often driven by identification of an intrinsically fluconazole-resistant yeast (Table 11.3). In general, for those isolates that are susceptible to fluconazole, this is the optimal choice due to its urinary penetration. Caspofungin does not penetrate well into the urine and data for its efficacy are mixed [39]. For fluconazole-resistant organisms, amphotericin deoxycholate (as opposed to liposomal amphotericin) is recommended due to improved urinary levels and penetration into the renal parenchyma [39]. *Candida auris* is an emerging pathogen that may be resistant to all antifungal drugs, and attention to local epidemiology is

Table 11.3 General susceptibility trends for *Candida* species affected by both patient and geographic factors. (Consult your local antibiogram for geography-specific data).

	Fluconazole	Amphotericin[1]	Caspofungin[2]
Candida albicans	++	+	++
Candida auris[3]	0	±	++
Candida glabrata	±[4]	++	++
Candida krusei	0	++	++
Candida lusitaniae	++	0	++
Candida parapsilosis	++	++	+
Other Candida sp.[5]	++	++	++

[1]For pyelonephritis, use deoxycholate formulation for better penetration into renal parenchyma.
[2]**Caspofungin susceptibility is nearly universal in vitro, but echinocandins to not adequately penetrate the urine or renal parenchyma.**
[3]*Candida auris* has variable susceptibility patterns. Most are resistant to fluconazole and susceptible to caspofungin with some variability. Susceptibility to amphoterici B is variable.
[4]*C. glabrata* is reported as "susceptible dose dependent rather than susceptible as higher doses are necessary to hit therapeutic targets. Depending on local patterns, a significant minority can be fluconazole resistant.
[5]Additional species include dubliniensis, tropicalis, guilliermondii, etc.; these are generally similar to *C. albicans*.
Modified from Gilbert DN, et al. The Sanford Guide Web Edition, Antifungal Activity Spectra, accessed via http://webedition.sanfordguide.com, January 17, 2020 (Antimicrobial Therapy, Inc.)

necessary to risk-stratify patients at risk for acquiring this difficult to identify *Candida* species [41].

Cryptococcuria is a secondary manifestation of disseminated disease with *Cryptococcus neoformans* (or *Cryptococcus gattii*, which was formerly classified as a subset of *C. neoformans*) [42]. Disseminated Cryptococcal infection is rare, and cryptococcuria is a rare complication, so few cases are described in the literature. However, because cryptococcuria is a manifestation of an often unappreciated disseminated infection, its detection can present an opportunity to reduce mortality for immunocompromised patients. Cryptococcal cells look similar enough to *Candida* on urine microscopy that only identification of suspect colonies in culture will make this diagnosis. Disseminated cryptococcal infections are initially treated with amphotericin B (with the addition of flucytosine if CNS involvement is suspected).

Returning to case 5, this patient was diagnosed with asymptomatic Candiduria and treatment was deferred as he was not neutropenic and no urgent urologic procedures were planned.

Case 6: Zika Virus

A pregnant 25 year old female at 20 weeks gestation presents to your office complaining of a new pruritic rash on her legs. She just returned from an extended vacation in Brazil.

In this case, an infection with Zika virus should be considered.

Zika is a mosquito-transmitted single stranded RNA virus that has been detected in various parts of the world (Central and South America, India, parts of Asia and Africa and in the United States in Texas and Florida). Transmission can be through sexual contact, *in utero* from mother to fetus or by blood transfusion (however the last has not been confirmed).

Many people are asymptomatic when infected with the Zika virus. Symptoms, if present, include headache, low-grade fever, pruritic maculopapular rash, arthralgias, myalgias and conjunctivitis. They are usually mild, present for 2–14 days, and resolve within 2–7 days. Treatment consists of rest, fluids, analgesics and antipyretics. Currently there is no antiviral medication available for treatment [45]. The CDC recommends women who are trying to conceive and women who are pregnant not travel to any area with risk of transmission [44]. The infection may lead to Congenital Zika Syndrome syndrome which consists of multiple congenital anomalies including microcephaly, eye damage, loss of joint range of motion, clubfoot, and increase in muscle tone [46].

To test for the Zika virus, a nucleic acid amplification test (NAAT) by polymerase chain reaction (PCR) is used to directly detect viral genomic material. Specifically, the Zika Triplex PCR test is used to test individuals with clinical signs and symptoms of Zika virus infection. Zika virus RNA can be detected in a urine specimen for up to 14 days from symptom onset and indicates active infection. The Triplex test can also

detect Zika virus RNA, Chikungunya virus RNA and Dengue virus RNA on serum, whole blood or cerebrospinal fluid specimens. This test can be run in state public health laboratories and the Centers for Disease Control and Prevention. It is a qualitative test so does not provide quantitative number of organisms present. Concomitant IgM serum testing should also be performed. Zika IgM antibodies can persist beyond 12 weeks after the infection which makes it difficult to distinguish if the infection occurred during the current pregnancy or prior to it [47, 48].

Returning to case 6, this patient did not have any other symptoms consistent with Zika virus infection and her urine and serum tests were negative. She was given the recommendation to not travel to any areas with risk of Zika virus transmission while pregnant or while trying to conceive.

Summary

Gram stain of urine is a much discussed subject with little clinical utility relative to its intuitive appeal. Automated instruments perform little better but serve to automate rapid screening in some contexts. General screening and treatment of patients with asymptomatic bacteria should only be undertaken in special circumstances. Incidental detection of *Trichomonas* in urine is possible, though Trichomoniasis is commonly considered intentionally in the workup of sexually transmitted infections and microscopy is not the preferred method for its detection. Other parasitic, mycobacterial, fungal and viral infections of the urinary tract are uncommon and are difficult to diagnose without careful consideration of patient history and pathogen-specific laboratory methods.

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Chapter 12
Urine Microscopy – Urine Made Crystal Clear

Courtney Yong, Chad R. Tracy, and Lisa M. Antes

Objectives

- Identify normal and pathologic crystalluria and when to seek expert consultation
- Understand the basis for medical management of recurrent kidney stones and how medications can alter urinary parameters to affect stone formation
- Identify what constitutes complicated versus uncomplicated stone cases and when to seek surgical consultation
- Recognize less common causes of crystalluria

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Overview

This chapter describes the diagnostic value of crystalluria on urinalysis/urine microscopy as well as subsequent measures for management of urinary lithiasis. We also discuss reasons for consultation to nephrology or urology, initial diagnostic workup, and basic tenets of treatment including medical and surgical management. Finally, the important considerations for management of inpatients or those who are critically ill with an acute and/or infectious stone episode are described.

Case 1: Incidental Hematuria and Crystalluria

An otherwise healthy 30 year old male presents to his primary care physician for a pre-employment physical. On urinalysis, he is found to have microscopic hematuria and crystalluria. Subsequent microscopic hematuria workup is negative (see Chap. 9).

While crystals in the urine, also known as crystalluria, may be incidentally noted on random urinalysis, the recommended method for testing for crystalluria involves having the patient in a fasting state and evaluating the whole volume of the first voided morning urine sample within 2 hours of collection at room temperature [1]. Microscopic urinalysis should be performed in conjunction with dipstick analysis. Phase contrast microscopy should also be utilized to further evaluate crystalluria as well as identify other urinary characteristics (see Chap. 3).

The formation of urinary crystals is dependent on several factors, including urine pH, which is important in determining which crystals will precipitate in a urine sample (see Chap. 7). Uric acid and cystine stones will form in acidic urine, with uric acid stones readily forming at pH < 6.5 and cystine stones forming at pH < 8.5, whereas calcium phosphate and struvite stones will form in urine with pH > 6.5 [1, 2]. Crystal forma-

tion is also highly dependent on urine concentration, with lower urine volumes and higher concentrations encouraging crystal formation. Finally, there are several crystal inhibitors that are naturally found in urine, such as citrate, magnesium, and Tamm-Horsfall proteins, which discourage crystallization [2]. Some of these parameters, such as citrate and magnesium, can be evaluated by direct testing of the urine, while others, such as Tamm-Horsfall proteins cannot.

Crystalluria can be found in both pathologic and normal states. Approximately 15–20% of asymptomatic, healthy patients with no history of kidney stones will have crystalluria on urinalysis, which is only slightly lower than the rate of 27% seen in patients with a history of kidney stones [3, 4]. Therefore, the diagnostic value of isolated crystalluria on urinalysis is limited. The presence of crystalluria on urinalysis, however, should prompt a more thorough history to evaluate for any personal or family history of kidney stones as well as for other comorbidities that would increase the risk of stone disease, such as inflammatory bowel disease, metabolic syndrome, type 1 renal tubular acidosis and sarcoidosis, among others [5].

Crystal shape is largely dependent on crystal composition, and many crystals have unique shapes that can identify them and assist in diagnosis (Table 12.1). While some crystals, such as calcium oxalate and uric acid can be found in healthy patients, others, such as cystine and magnesium ammonium phosphate, are pathognomonic for certain pathological conditions and should prompt further workup.

Cystinuria is a hereditary, autosomal recessive disease of impaired proximal tubule reabsorption of cysteine, ornithine, lysine, and arginine from the urine, leading to increased urinary excretion. As cysteine is the least soluble of these amino acids, the crystals are prone to aggregate and form cystine crystals, which are distinctive under the microscope as having a hexagonal shape and may be used to monitor the activity of the disease (Table 12.1) [2]. In a retrospective study, cystine crystals were identified in 83% of patients with known cystin-

uria and were found in only 0.25% of normal controls [6]. Cystine crystalluria should prompt referral to urology or nephrology for evaluation.

Magnesium ammonium phosphate (struvite) stones are caused by urinary infection, always with urease-producing bacteria [2]. Discovery of struvite crystals, which are coffin lid-shaped, is pathognomonic and the patient should undergo a urine culture and imaging to evaluate for struvite nephrolithiasis (Table 12.1) [1].

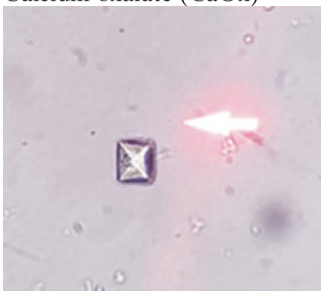
Returning to case 1, to determine the need for further evaluation of crystalluria, you would want to obtain a more complete medical history. This includes asking about personal stone history, family stone history and risk factors for stone formation.

Case 2: Kidney Stones and Crystalluria

The same healthy 30 year old man with crystalluria is found to have a history of multiple episodes of spontaneous kidney stone passage on additional questioning.

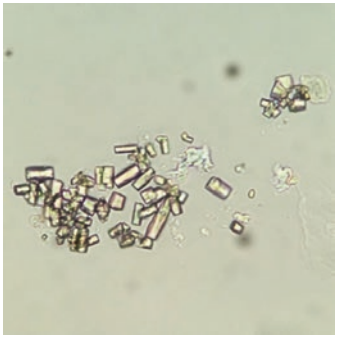
The incidence of nephrolithiasis has increased significantly over the last several decades [7]. The lifetime risk of developing kidney stone disease is estimated to be between 1–15% with variability according to certain demographics [2]. Risk factors for development of nephrolithiasis include living in geographically warmer climates, dietary factors including low fluid intake or high sodium intake, and metabolic syndrome characteristics (especially obesity and impaired fasting glucose) [7]. While men are more likely to develop kidney stones than women, the incidence of stones in women has been increasing [7].

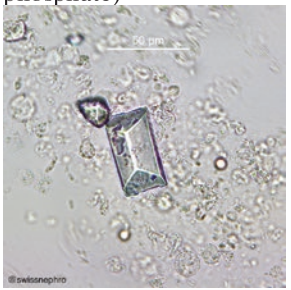
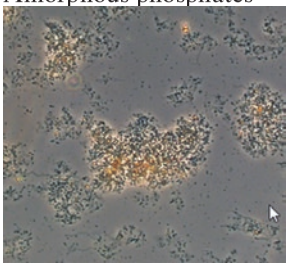
TABLE 12.1 Crystal characteristics with associated conditions and urine Litholink findings

Stone type and urine microscopy	Characteristics	Associated conditions	Potential findings on Litholink
Calcium oxalate (CaOx) 	Polarize Any urine pH **Monohydrate**: dumbbell, ovoid or long **Dihydrate**: octahedral (shown in picture)	Healthy individuals with high oxalate consumption or excess vitamin C, idiopathic hypercalciuria, ethylene glycol intoxication, primary hyperparathyroidism	Hypercalciuria Hyperoxaluria Hypocitraturia Hyperuricosuria High urine sodium High supersaturation of CaOx Low urine magnesium

(continued)

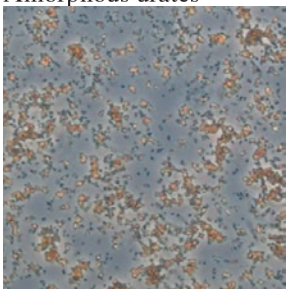
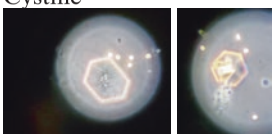
TABLE 12.1 (continued)

Stone type and urine microscopy	Characteristics	Associated conditions	Potential findings on Litholink
Uric acid 	**Monosodium urate**: Slender pencil like prisms (ends not pointed) **Uric acid**: pH < 5.7 Most common diamond shaped but can be barrel shaped, 6 sided or rosette Polarize	Gout, cytotoxic drugs, tumor lysis syndrome, medullary cystic kidney disease, GI losses of alkali, myeloproliferative disorders, low carbohydrate/high protein diets	Hyperuricosuria Hypercalciuria

Staghorn (struvite or triple phosphate) 	Most common crystal in alkaline urine 3–6 sided prism (coffin lid) Struvite = magnesium ammonium phosphate Some have calcium carbonate apatite	Infections with urease splitting organisms in upper urinary tract	High urine ammonium
Amorphous phosphates 	Appear in alkaline urine as small granulations, without polarization	May be seen in vegetarian diets or diets rich in phosphates. May also be seen in association with calcium phosphate crystalluria.	Litholink evaluation not usually necessary

(continued)

Table 12.1 (continued)

Stone type and urine microscopy	Characteristics	Associated conditions	Potential findings on Litholink
Amorphous urates 	pH 5.7–7.0 Irregular granules Sand like or brick dust color	May be found in association with uric acid crystalluria	Litholink evaluation not usually necessary
Cystine 	Colorless hexagonal plates may be heaped Typically seen in acid urine (pH < 8.5) Do not polarize Can confirm with cyanide nitroprusside reaction	Congenital cystinosis (genetic disorder) or cystinuria	Cystinuria

(Calcium oxalate and cystine crystals courtesy of University of Iowa, Division of Nephrology and Department of Urology)(Uric acid, struvite, amorphous phosphate and amorphous urate crystals courtesy of Florian Buchkremer)

Crystals found in the urine of stone formers can have diagnostic value. In patients with a history of calcium oxalate or cystine stones, those with crystalluria on ≥ 50% of morning urinalyses have been found to have significantly greater risk of a recurrent stone episode within 3 years [8, 9]. In addition, in recurrent stone formers with a history of crystalluria, clearance of urinary crystals on urinalysis after initiation of dietary modifications or medical therapy can indicate relative success of therapy and a decreased risk of new stone growth or recurrent stone events [1, 10].

Case 3: Metabolic Stone Disease

The patient in case 2 is referred to the multidisciplinary Metabolic Stone Clinic, which consists of nephrologists, urologists, and dieticians. Because of his history of recurrent stones, it is recommended that he undergo a metabolic evaluation.

The timing of metabolic stone evaluation is under debate. While it is generally agreed that patients who have had two or more stone episodes should be considered for metabolic stone evaluation, there is conflicting evidence as to whether or not first time stone formers should be referred for evaluation [2]. Generally, patients with recurrent stones, as well as those with known risk factors for stone disease, should be considered for referral (Table 12.2) [11]. Additionally,

Table 12.2 Quick Referral Guides for Crystals/Stones

Quick urology referral guide
1. Staghorn calculi
2. Stones >4 mm needing extraction
3. Bladder stones
4. Symptomatic nephrolithiasis
Quick referral guide for nephrology or metabolic stone clinic
1. New stone former with risk factors
2. Recurrent stones for initial management and follow-up
3. Hereditary/genetic stone disease (cystine, hypercalcemic hypercalciuria)

pediatric patients should generally be evaluated with metabolic analysis, as they are at higher risk for treatable metabolic abnormalities [2]. In a retrospective study by Eisner et al., the rates of metabolic abnormalities found on 24-hour urine analysis were similar between first time stone formers and recurrent stone formers, which supports early referral to a Metabolic Stone Clinic [12, 13]. Additionally, up to 50% of first time stone formers will experience a recurrent stone event within 5–10 years [14, 15].

Metabolic evaluation for nephrolithiasis includes blood tests and 2 random 24-hour urine collections with the patient on their baseline diet. The blood tests include electrolytes with calcium, creatinine and parathyroid hormone (PTH) if serum calcium is elevated. The urine studies include urine sodium, potassium, creatinine, uric acid, calcium, citrate, oxalate, pH and total volume. In addition, if the patient underwent a procedure for stone extraction, or if the patient collected a passed stone, the stone should be sent for stone composition analysis. The findings of each of these tests will inform potential therapy.

Most stone formers will benefit from dietary modification including increased hydration and low salt and animal protein intake. Patients should be instructed to increase fluid intake with a goal 24-hour urine volume of at least 2.5 liters, to lower their sodium intake to <50 mmol (1150 mg)/day, and decrease their animal protein intake to <52 g/day [2, 11, 16]. While all patients will benefit from decreasing animal protein, the type of protein (and uric acid content) may also be important in stone prevention [17]. Patients with calcium stones should be advised to maintain normal amounts of calcium (30 mmol (1200 mg)/day) and vitamin D in their diets, as restricting dietary calcium has been shown to paradoxically increase the risk of stone disease [16]. Additional dietary modification or addition of medical therapy should be informed by the results of the metabolic analysis.

Metabolic management of stone disease is largely based on dietary modifications as well as medications that can change urine parameters. Dietary modifications such as a low protein diet to minimize uric acid secretion or a low methionine diet to minimize cystine secretion can also help to alter

Table 12.3 Summary of metabolic treatment of stone disease

Metabolic abnormality	Commonly associated stone types	Dietary/Medical management
Hypercalciuria	Calcium oxalate Calcium phosphate	Low sodium diet Normal dietary intake of calcium Normal dietary intake of vitamin D Thiazide diuretics
Hyperuricosuria	Calcium oxalate	Low protein diet Low sodium diet
	Uric acid	Urinary alkalinization Consider allopurinol if unable to maintain low protein diet
Acidic urine	Uric acid Cystine	Low protein diet Potassium citrate
Hypocitraturia	Calcium oxalate	Increased citrate intake in diet with citrus juices (lemonade, orange juice) Potassium citrate
Hyperuricemia (blood)	Calcium oxalate	Low protein diet Low sodium diet
	Uric acid	Allopurinol

Recommended for all stone formers: high fluid intake (>2.5 L urine output per day), low sodium, and low protein diets

urine parameters and prevent stone formation [2]. Thiazide diuretics can decrease urinary calcium excretion and are often used to treat hypercalciuria. Table 12.3 summarizes the metabolic treatment of stone disease, which incorporates dietary modifications and medical therapies. Specific medications with dosages commonly used in management of stone disease are summarized in Table 12.4.

Hyperuricosuria or hypercalciuria associated with elevated levels of serum uric acid can be managed with allopurinol, a xanthine oxidase inhibitor, to decrease serum uric acid [2]. Other parameters, such as urine pH, can be altered by

Table 12.4 Medications used in the metabolic management of stone disease [2]

Drug classification	Medication name	Typical treatment dose	Potential side effects
Thiazide diuretics	Hydrochlorothiazide	25 mg po BID	Hypokalemia, hyperuricemia, alkalosis, hypocitraturia
	Chlorthalidone	25–50 mg po daily	
	Indapamide	2.5 mg po daily	
Xanthine oxidase inhibitor	Allopurinol	300 mg po daily	Rash, myalgia
Alteration of urine pH	Potassium citrate	20 mEq po BID or TID	Gastrointestinal symptoms, hyperkalemia Increased risk of calcium phosphate stones if pH > 7.0

prescribing potassium citrate, which will alkalinize the urine and can be used to prevent uric acid and cystine stones, both of which precipitate in acidic urine. Once metabolic management is initiated, a follow up 24-hour urine study can be performed several months later to ensure appropriate urinary response to treatment.

The 24-hour urine analysis for the patient in Case 3 is shown in Fig. 12.1.

	Ref. Range and Units	
STONE STUDIES, URINE		
Ammonia, Stone, Urine	Latest Range: 15 - 60 mmol/d	***39***
Body Weight (kg)	Latest Units: kg	***86.2***
Ca/kg Body Weight	Latest Range: <4 mg/kg	***3.8***
Ca/Creatinine	Latest Range: <140 mg/g	***153*** ▲
Calcium, Stone, Urine	Latest Range: <250 mg/24H	***323*** ▲
Calculi Composition	No range found	
Calculi Description	No range found	
Calculi Number	No range found	
Calculi Size (mm)	Latest Units: mm	
Chloride, Stone, Urine	Latest Range: 70 - 250 mE q/d	***146***
Citrate, Stone, Urine	Latest Range: >450 mg/d	***400*** ▼
Creat/kg Body Weight	Latest Range: 18 - 24 mg/kg	***24.6*** ▲
Creatinine, Stone, Urine	Latest Units: mg/d	***2,120****
Cystine Date	No range found	***7/5/13***
Cystine Screening	Latest Range: Negative	***Negative***
Magnesium, Stone, Urine	Latest Range: 30 - 120 mg/d	***152*** ▲
Calculi Mass	Latest Units: mg	
Oxalate, Stone, Urine	Latest Range: 20 - 40 mg/24H	***25***
pH, Stone, Urine	Latest Range: 5.8 - 6.2	***6.666*** ▲
Phosphorus, Stone, Urine	Latest Range: 0.6 - 1.2 g/d	***1.256*** ▲
Potassium, Stone, Urine	Latest Range: 20 - 100 mmol/d	***63***
Sodium, Stone, Urine	Latest Range: 50 - 150 mmol/d	***154*** ▲
Stone, Analysis Order	No range found	
Sulfate, Stone, Urine	Latest Range: 20 - 80 mE q/d	***69***
Supersat CaOxalate	Latest Range: 6 - 10	***3.07*** ▼
Supersat CaPO4	Latest Range: 0.5 - 2	***2.09*** ▲
Supersat Uric Acid	Latest Range: 0 - 1	***0.10***
Urea Nitrogen, Stone, Urine	Latest Range: 6 - 14 g/d	***11.15***
Uric Acid, Stone, Urine	Latest Range: <0.800 g/d	***0.655***
Volume, Stone	Latest Range: 0.50 - 4.00 L/D	***3.21***
Protein Calabolic Rate	Latest Range: 0.8 - 1.4 g/kg/d	***1.0***

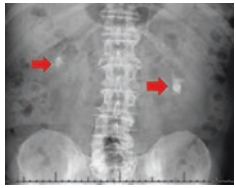
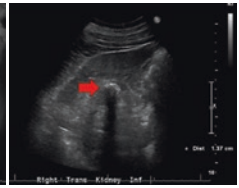
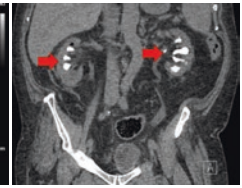
FIGURE 12.1 24-hour urine testing results` for the patient in Case 3

Overall, medical metabolic management of stone disease is highly effective. Several studies have shown a significant improvement in stone outcomes with metabolic therapy, with an estimated 70–80% reduction of subsequent stone formation [18–20]. In addition, a previous study showed that metabolic evaluation and treatment of stones does not necessarily need to be done in a specialized clinic, and similar beneficial results can be achieved in a smaller clinic with appropriate resources [21].

Recurrent stone patients should be routinely followed with imaging such as kidneys, ureters, bladder x-ray (KUB), renal ultrasound, or CT scan for evaluation of new stone formation (Table 12.5) [11]. Low dose, non-contrast CT scan is the best initial test to diagnose kidney stones with sensitivity and specificity of 95% and 97%, respectively. By comparison, ultrasound has an 84% sensitivity and 53% specificity, and KUB has a 57% sensitivity and 76% specificity [22]. In terms of cost-effectiveness, CT scan is about 10 times the cost of KUB, whereas ultrasound is 5 times the cost of KUB [22]. For children and women who are pregnant, ultrasound should be the first line imaging study for renal pathology due to the relative risk of radiation in these patients. MRI has a sensitivity of 82% and specificity of 98%, is less sensitive than CT for detecting calcifications and is 30 times more expensive than KUB. As such, it is not typically used in stone disease except in special circumstances such as pregnancy [22].

Patients with crystalluria and hematuria should have a full urologic evaluation including a CT urogram and cystoscopy. A CT urogram is a 3-phase evaluation that includes a non-contrast phase to evaluate for the presence of stones, as well as a contrast phase to evaluate for renal masses and a delayed phase to evaluate the collecting system for any filling defects or tumors. The interval and modality of follow up imaging after diagnosing recurrent stone disease or after stone passage is patient and practitioner dependent, but patients are typically followed every 6–12 months with KUB or renal ultrasound for evaluation of interval stone changes. Notably, some stones such as uric acid stones are radiolucent on KUB and require CT scan or ultrasound for evaluation [23]. Rarely, stones caused by

Table 12.5 Comparison of KUB, US, and CT as imaging modalities for the detection of kidney stones (indicated with red arrows)

KUB	Ultrasound	Non-contrast CT
Pros:	**Pros:**	**Pros:**
• Widely available • Low cost • Minimal radiation	• Noninvasive • No radiation, can be used safely in pregnancy • Widely available • Low cost	• Excellent sensitivity (95%) and specificity (97%) • Best evaluates ureteral stones • Determines stone density and accurately measures size
Cons:	**Cons:**	**Cons:**
• Limited sensitivity (57%) and specificity (76%) • Does not evaluate for hydronephrosis • Does not visualize radiolucent stones	• Lower sensitivity (84%) and specificity (53%) • Difficult to evaluate for ureteral stones	• Radiation exposure; limited use in pregnancy • High cost (10× cost of KUB)

urinary precipitation of drugs such as indinavir can be radiolucent on both KUB and CT scan; these can be evaluated clinically or possibly assessed with delayed-phase CT with IV contrast to evaluate for filling defects in the renal collecting system [23]. Timing of intervention for non-obstructing stones discovered on surveillance imaging is variable and is again patient and practitioner dependent, but several factors, such as stone size, location, rate of stone growth, and number of stones can inform decision making. The majority of patients with

symptomatic stone disease pass the stone spontaneously and do not experience kidney dysfunction. Keep in mind that the serum creatinine may remain normal in the acute setting if the obstruction is unilateral. Obstructive uropathy from a stone, however, can cause acute kidney injury and may also produce irreversible chronic injury, particularly if the obstruction is not relieved or the patient has recurrent episodes of obstructive uropathy. In addition, certain stone types such as cystine stones are more likely to result in CKD in adulthood.

Returning to case 3, the patient's urinary volume is adequate with 3.21 L/day. However, he has hypercalciuria and hypocitraturia, which could be contributing to his stone formation. If no contraindications, this patient should be started on a thiazide diuretic, which would decrease his urinary calcium, and potassium citrate, which would increase his urinary citrate. It should be recommended that he continue high fluid intake and maintain normal amounts of dietary calcium.

Case 4: Acute Stone Episode

The patient in case 3 does well on medical management for some time. He then presents to the ED 2 years later with acute left sided flank pain. The patient has no fevers or costovertebral angle tenderness. Urinalysis is negative for nitrites and leukocyte esterase, but shows hematuria with no pyuria on microscopic analysis. Creatinine is normal. Non-contrast CT scan reveals a 5 mm left proximal ureteral stone with mild associated hydronephrosis. His pain is well controlled on oral pain medications, and he tolerates clear liquids without vomiting.

The rate of spontaneous stone passage decreases the larger the stone, with stones >5 mm having a significantly lower passage rate than smaller stones [24, 25]. Per the American Urological Association (AUA) guidelines, in patients whose pain is well controlled and who have uncom-

plicated distal ureteral stones 5 mm or less, a trial of medical expulsive therapy is warranted [26]. One medication recommended as a component of medical expulsive therapy is tamsulosin, an alpha-1 blocker that is thought to encourage stone passage by relaxation of ureteral musculature. While some studies have shown improved stone passage with the alpha blocker tamsulosin, others have found no benefit [27–29]. However, the current AUA and European Association of Urology (EAU) guidelines still recommend the use of alpha blockers for medical expulsive therapy, in addition to oral pain control with NSAIDs or narcotics [26, 30]. Patients should also be closely followed by a urologist or primary care physician, if their physician is comfortable with the management of stones, with subsequent imaging and with straining of their urine to confirm stone passage for up to 4–6 weeks [26].

For patients with larger stones, uncontrollable pain, presence of infection, a solitary kidney or for those who fail to pass their stones, surgical management should be offered. Renal drainage with either ureteral stenting or percutaneous nephrostomy tube placement can be offered in the acute setting to relieve pain or infection and should be considered emergently as primary treatment if there is infection associated with an obstructing renal stone. For ureteral stones, ureteroscopy with lithotripsy is first line management [31]. Shock wave lithotripsy (SWL) can be offered to patients with renal stones 20 mm or less in size for upper and middle pole stones and 10 mm or less in size for lower pole stones. While SWL is less invasive, the stone free rate is less than that of ureteroscopy [31]. For stones >20 mm, percutaneous nephrolithotomy (PCNL) or ureteroscopy may both be viable treatment options [31].

Returning to case 4, the patient should be sent home with medical expulsive therapy and plans for close follow up with a urologist with re-imaging to confirm stone passage. If the stone does not pass, he could then be offered a surgical procedure for removal. Metabolic testing should be performed after the stone has been passed or surgically removed and the urinary system has had time to heal from any interventions performed.

Case 5: Infectious Stone

A 67 year old female with a history of recurrent urinary tract infections presents to the ED with fevers, tachycardia, hypotension, and flank pain. Initial bloodwork shows leukocytosis and acute kidney injury with creatinine 3.2 mg/dL from baseline 1.0 mg/dL. Urinalysis is positive for leukocyte esterase and nitrites, and microscopic analysis shows pyuria, bacteriuria, and coffin lid-shaped crystals. Non-contrast CT scan shows a right sided staghorn stone with associated hydronephrosis (Fig. 12.2).

In the setting of infection, obstructing stones, whether large staghorn stones as in this case or small ureteral stones, can be fatal. Thus, sepsis from a presumed urological source should include evaluation for upper urinary tract obstruction. When a patient presents with urinary obstruction and infection, urgent urology consultation should be obtained for drainage of the collecting system with either a ureteral stent or a percutaneous nephrostomy (PCN) tube [26]. Urine culture in this population is imperative as they are at increased risk for atypical organisms and frequent prior medical exposures may increase their risk of resistant organisms [32]. Definitive surgical management of stones should be deferred until the patient has stabilized and the infection is under control with antibiotics, as lithotripsy performed during active infection can induce sepsis and can be fatal.

Struvite stones (magnesium ammonium phosphate and calcium carbonate-apatite) form secondary to infection with urease-producing organisms, which break down urinary urea into ammonia, leading to increased supersaturation of ammonium and elevated urinary pH. While *Proteus spp.* are the most common bacteria found in struvite nephrolithiasis, at least 14 other bacteria, including *Staphylococcus spp.*, *Klebsiella spp.* and *Corynebacterium spp.*, may lead to struvite stone formation [32]. In addition to the causative organism, patients with an infected stone are at an increased risk of superinfection with other bacteria such that empiric antibi-

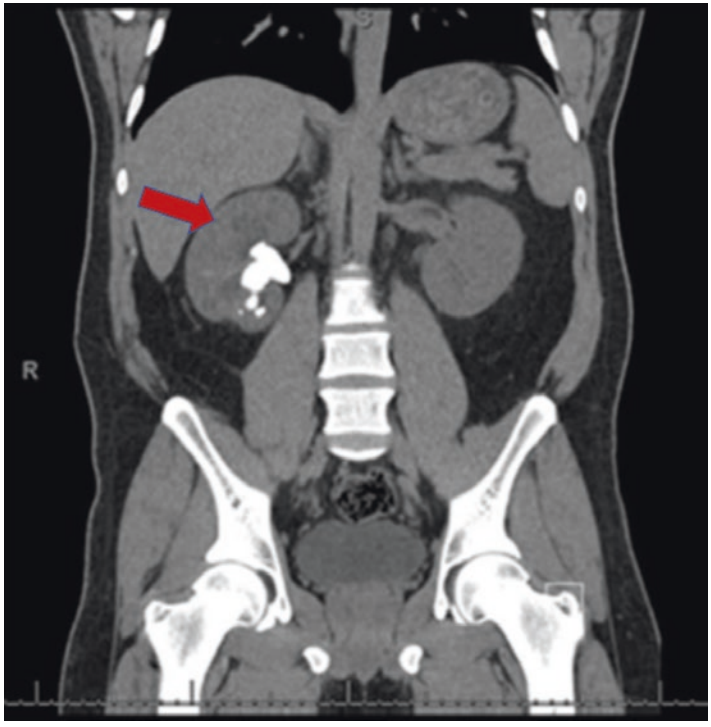


Figure 12.2 Non-contrast CT showing right-sided staghorn stone associated with hydronephrosis (indicated with red arrow). (Courtesy of University of Iowa, Department of Urology)

otic coverage in this setting should broadly cover both Gram-negative and Gram-positive organisms [32].

Other Considerations Regarding Crystalluria

While crystalluria can be found in both healthy subjects and in patients with nephrolithiasis, there are several other less common diagnoses to consider when a patient presents with crystalluria.

Bladder stones account for approximately 5% of urinary calculi and crystalluria can be seen in the setting of bladder

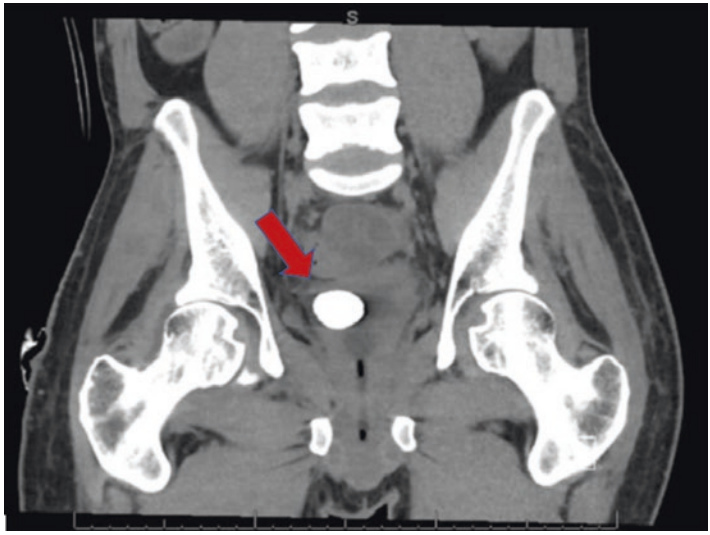


Figure 12.3 Non-contrast CT showing a patient with 2 cm bladder stone (indicated with red arrow). (Courtesy of University of Iowa, Department of Urology)

stones without kidney stones [33] (Figs. 12.3 and 12.4). In developed countries, bladder calculi predominantly affect adult males, and the most common presenting symptom is acute urinary retention. They are often associated with previous episodes of urinary retention as well as prior episodes of renal stones including prior renal colic and prior passage of renal stones [33]. Bladder stones can also be seen in endemic areas where malnutrition and low protein diets are common. Children from endemic areas are most commonly affected by bladder stones. Endemic bladder stone composition is predominantly ammonium acid urate or uric acid [34]. Treatment of bladder stones involves stone removal as well as subsequent prevention by either relieving bladder obstruction or improving the quality of life and diet of the endemic population. Patients with bladder stones should be referred for urological evaluation as they are likely a symptom of severe bladder outlet obstruction and urinary stasis, which may require additional therapy.

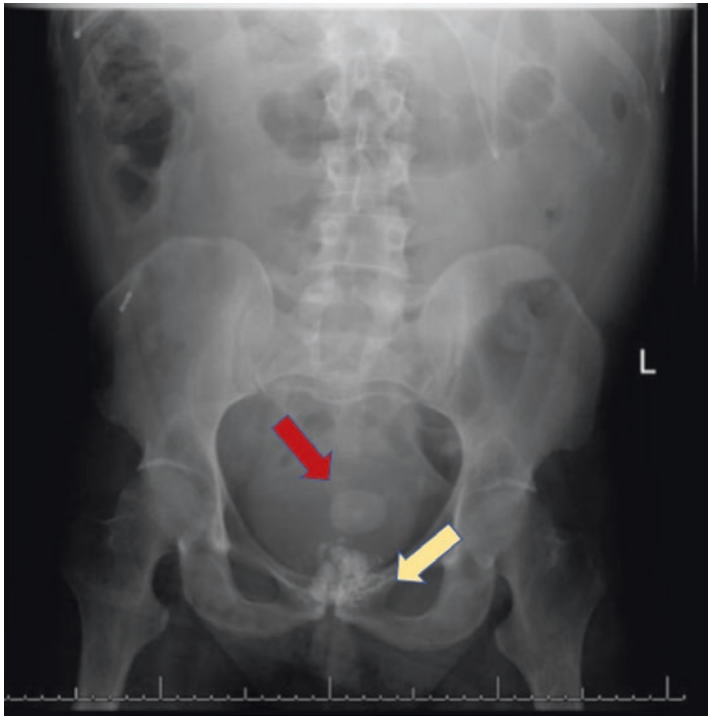


FIGURE 12.4 KUB showing a patient with 3.3 cm bladder stone (indicated with red arrow) and extensive prostatic calcifications (indicated with yellow arrow). (Courtesy of University of Iowa, Department of Urology)

Crystalluria can also be found in certain unique inpatient situations. Ethylene glycol poisoning should be suspected in a patient with altered mental status, osmolar gap metabolic acidosis, acute kidney injury (AKI), and calcium oxalate crystalluria. A previous study determined that the predominant form of calcium oxalate crystalluria in the setting of ethylene glycol poisoning is calcium oxalate monohydrate, which is characterized by oval or dumbbell shaped crystals [35]. More recently, it has been determined that this calcium oxalate crystalluria is the primary mechanism of acute kidney injury and subsequent renal failure in ethylene glycol poisoning (Fig. 12.5) [36].

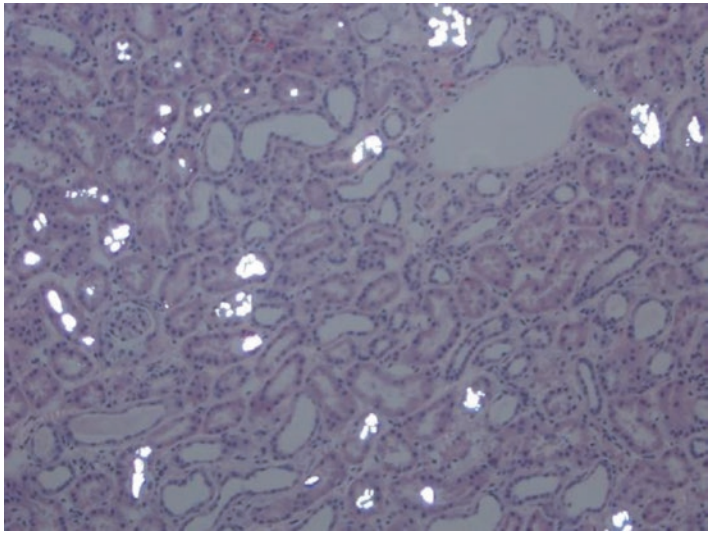


FIGURE 12.5 Intratubular polarizable crystalline materials consistent with oxalate crystals in an autopsy kidney section from a young man ingesting ethylene glycol. H&E staining; 400× magnification. (Courtesy of Dr. Dao-Fu Dai, University of Iowa, Division of Nephrology)

Several medications have been shown to precipitate in the urine as crystals of the drug itself. While drug induced nephrolithiasis accounts for <0.5% of all renal stones, patients who are on these medications can be at significantly increased risk for drug induced crystalluria and nephrolithiasis [37]. Indinavir, a protease inhibitor used to treat HIV, can precipitate in the urine and can cause crystalluria or nephrolithiasis in up to 50% of patients [37]. Several antibiotics have been shown in case reports to cause crystalluria, including sulfamethoxazole (6% of patients) Fig. 12.6, amoxicillin (18% of patients) and ciprofloxacin (rare, case reports) [38–40]. Triamterene, a potassium-sparing diuretic, is well known to cause stones in approximately 1/2000 patients treated with the drug [41, 42]. Finally, treatment of severe hyperuricemia with a xanthine oxidase inhibitor, such as allopurinol, may lead to the development of xanthine stones [43].

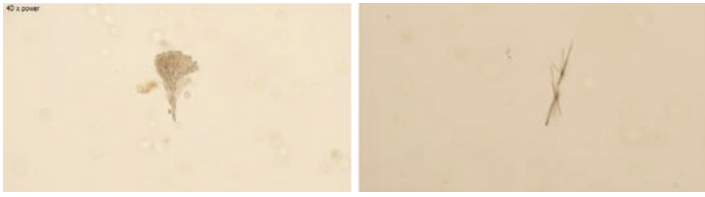


Figure 12.6 Urine microscopy showing sulfamethoxazole crystals. (Courtesy of University of Iowa Hospitals and Clinics)

Summary

Evaluation of crystalluria can be a useful diagnostic aid. While crystalluria can be found in healthy patients, it can also be used to identify metabolic pathology such as cystinuria, as well as determine the risk of recurrent stone disease in patients with a kidney stone history. Therefore, the presence of crystalluria should prompt a more thorough history and evaluation of the patient. Management of patients with kidney stones or other pathologic crystalluria requires a multidisciplinary approach with a combination of medical, metabolic, and surgical management for both treatment and prevention.

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Chapter 13
Urine Testing in Children: Little People, Big Challenges

Gina M. Lockwood and Douglas W. Storm

Objectives

- Identify recommended methods of urine collection in children of various ages
- Describe normal and abnormal values on urinalysis in children
- Understand differential diagnoses of proteinuria, microscopic/gross hematuria and urinary tract infection in children, as well as their initial evaluation and management
- Identify "red flags" in children which indicate clinically significant hematuria or proteinuria
- Identify which children with abnormal urine testing should undergo nephrology and/or urology evaluation

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Overview

It is estimated that at least 2% of boys and 7% of girls will be diagnosed with a urinary tract infection (UTI) by 6 years of life [1]. Primary care physicians obtain urine testing in children not only to diagnose UTI, but to investigate symptoms like hematuria, dysuria and urinary incontinence, similar to the adult population; however, collection of urine in children presents unique challenges and pitfalls, especially in the child who is not toilet-trained. Additionally, the differential diagnosis, evaluation and management for UTI and other common pediatric urinary abnormalities like hematuria and proteinuria differ significantly from adults. In this chapter, strategies for successful urine collection in a child will be discussed. Common urinary abnormalities in children will also be explained, focusing on the differences in testing thresholds for positive results in children compared to adults, as well as generation of pediatric-specific differential diagnoses.

Case 1: Urine Collection Methods in Children

A 6 month old male presents to the outpatient clinic with a fever of 39.5°C. His only medical history is that of prenatally-diagnosed left hydronephrosis. He is more fussy than usual. He has no upper respiratory symptoms. He is eating and drinking less than normal, but he does continue to make wet diapers. On examination, he has a normal ear examination, normal mucous membranes, clear lung sounds, and a soft abdomen. He is uncircumcised. A urinary tract infection is suspected.

Infants and young children are unable to urinate on demand, thus urine collection for even simple testing can become complex. For the child who is not toilet-trained, the least invasive method of urine collection is via diaper pad or collection bag affixed to the perineum. Bagged specimens can be performed for urine testing not affected by bacterial contamination but must be used with significant caution when

Table 13.1 Sensitivity and specificity for diagnosis of UTI via positive urine culture for different modes of urine collection

Type of specimen collected	Sensitivity (%)	Specificity (%)
Perineal bag	Up to 100% [36]	63% [3]
Clean catch[a]	75–100% [4, 5]	57–100% [4, 5]
Catheterized[a]	95% [37]	99% [37]

[a]Using suprapubic aspiration as "gold-standard"

obtained for diagnosis of a UTI. Urine cultures obtained with bagged urine specimens have a high false positive rate because of sloughing of perineal, preputial, vaginal and rectal flora into the specimen. Sensitivity and specificity of different methods of urine collection for urine culture in children are shown in Table 13.1.

Because of its poor specificity, urine collection for diagnosis of UTI via bagged specimen is only considered useful when it rules out a UTI, i.e. if dipstick and/or urinalysis is completely negative. Bagged urine specimens cannot be relied upon as the only test to positively diagnose a UTI. Obtaining a bagged urine can be considered as screening for UTI before moving to more invasive urine collection methods, as only a small proportion of urine cultures obtained in small infants and children will ultimately be positive [2].

The American Academy of Pediatrics recommends urethral catheterization or suprapubic aspiration for urine collection in non-toilet trained children less than 24 months of age when UTI is suspected [3]. Disadvantages of these methods include invasiveness, need for skilled personnel, potential for trauma and potential for introduction of infection. Suprapubic aspiration via a 22-gauge needle is considered the gold standard for sterile urine collection, but its use is often not practical given the need for performance by a skilled practitioner and possible need for ultra-

sound guidance. Although considered unacceptably invasive by some, it may be necessary in boys with tight phimosis or girls with dense labial adhesions [4, 5].

Urethral catheter insertion is much more commonly performed and should be performed in a sterile fashion. The use of intraurethral and/or topical lidocaine 2–10 minutes prior to catheterization has been shown to reduce discomfort and distress from catheterization [6–8]. See Table 13.2 for appropriate catheter size by age.

Urethral catheterization in girls often requires a two-person technique with one person exposing the urethral meatus and the other person inserting the catheter. The normally recessed urethral opening and surrounding anatomic landmarks can most easily be identified by placing gentle traction on each of the labia majora outward and slightly lateral. The tendency with a one-handed technique is to pull the labia too far laterally, which can cause significant pain and trauma, making subsequent catheterization more difficult. Once the catheter is inserted, the first few drops of urine obtained should not be collected for specimen, as they are the most likely to be contaminated.

Urethral catheterization in the uncircumcised male can present a unique challenge. If the foreskin can be retracted behind the glans (head) of the penis, this should be sterilized and the catheter inserted. However, especially in infants, the foreskin may not retract completely. The foreskin should be retracted as much as possible to reveal the urethral meatus

Table 13.2 Appropriate urethral catheter size by patient age

Age (y)	Catheter French size (F)
0–5	5–8[a]
5–10	8–10
10–14	10
>14	10–14

Adapted from Ref. [38]

[a]Many infants will require 5–6 F catheter placement. Children >2 y are generally able to tolerate 8 F catheter

prior to catheterization, but the foreskin should never be forcefully retracted, causing pain or bleeding.

Although not an option in children prior to toilet training, a clean-catch urine sample is also a non-invasive method of urine collection. It is more reliable in older girls and circumcised boys but still has a much higher chance of contamination than suprapubic aspiration or catheterization. Young toilet-trained children are often unable to urinate in a small, sterile specimen cup without assistance. Often a large, non-sterile collection container is used for pediatric patients, i.e., a urine "hat;" however, using a non-sterile collection container increases risk of contamination. Additionally, there is debate regarding use of sterile wipes in children for obtaining clean-catch specimens. Using wipes is thought to potentially increase the risk of contamination.

When considering the method of obtaining urine for testing in a child, especially for culture, the risk of false positive results must be weighed against the risk of harm from procedural invasiveness. It is especially important to obtain accurate culture results in children with febrile urinary tract infections or recurrent urinary tract infections to prevent long term urinary tract sequelae like renal scarring. In children with anatomic genitourinary abnormalities like hydronephrosis or vesicoureteral reflux, the decision to proceed with surgery is often based on the number of UTIs the child has had. After urine is properly collected, the subsequent handling and analysis of urine tests is no different in children than in adults (see Chap. 3).

Returning to case 1, a catheterized urine specimen should be obtained to achieve accurate diagnosis, especially in the presence of an uncircumcised penis, which increases urine sample contamination.

Case 2: Hematuria in the Child

A 5 year old healthy, potty-trained female comes to the outpatient clinic complaining of a 1 month history of burning with urination. She has no fevers, abdominal pain or flank pain.

Neither she nor parents have seen discoloration of the urine. Her physical examination is normal. She has no history of prior UTI.

Urine dipstick & urine microscopy	
Component	**Result**
Color	Yellow
Appearance	Clear
pH	6.5
Specific gravity	1.015
Blood	2+
Glucose	Negative
Ketone	Negative
Protein	Negative
Urobilinogen	Negative
Leukocyte esterase	Negative
Nitrite	Negative
White blood cells (WBC)	0–2/hpf
Red blood cells (RBC)	>50/hpf
Squamous epithelial cells	0–2/hpf
Casts	None
Bacteria	Few

Hematuria in children has very different diagnostic implications than in the adult population, given the rarity of pediatric urinary tract malignancy. Although there is no accepted guideline, microscopic hematuria is usually considered to be >5 RBCs/hpf, in the absence of infection in children, compared to ≥3 in adults [9]. Blood on urine dipstick should always be confirmed with urine microscopy. Microscopic hematuria, whether asymptomatic or symptomatic, should prompt repeat urine testing over a period of 2–3 weeks, and if persistent on

two to three additional samples warrants further evaluation [10]. Isolated microscopic hematuria in pediatric patients is very common, usually self-limited and benign in nature [11, 12]. The incidence of microscopic hematuria on screening urinalysis in evaluated children ranges from 0.5% to 3%, and is often idiopathic [12]. Gross hematuria is less common, and a cause is found approximately two thirds of the time.

The three most common causes of isolated microscopic hematuria in children are thin basement membrane disease (15.2%), IgA nephropathy (10.4%), and hypercalciuria (7.7%). The three most common causes among children with microscopic hematuria and associated proteinuria are IgA nephropathy (44.3%), thin basement membrane disease (12.8%), and mesangial proliferative glomerulosclerosis (8.9%) [13]. The most common cause overall of microscopic or gross hematuria in the child is hypercalciuria, thus a random urine calcium/creatinine should be obtained in all patients with gross hematuria and those with persistent microscopic hematuria. Hypercalciuria may be asymptomatic or have associated dysuria, and it can be diet-related but is most often idiopathic. A calcium/creatinine of greater than 0.2 in children greater than 5 years and greater than 0.4 in children 2–5 years is considered abnormal [14]. A diagnosis of hypercalciuria is treated in most with high fluid intake and modest sodium restriction. It is considered a risk factor for nephrolithiasis. Diagnosis of hypercalciuria warrants pediatric urology referral and renal bladder ultrasound.

Although it is not as common, urethrorrhagia (benign urethrorrhagia or idiopathic urethrorrhagia) is an entity seen almost exclusively in pediatric male patients. It is usually self-limited. It is described as spotting of blood in the underwear after urination or terminal hematuria (blood at the end of the urine stream) following normal, clear urination. Its etiology is unclear, but some attribute it to voiding dysfunction or meatal stenosis. It has a high rate of spontaneous resolution (up to 92% in symptomatic adolescent males) [15]; however, urethral stricture disease has been reported on evaluation for urethrorrhagia in 14–60% of patients [16, 17]. General treatment recommendations include optimization of bladder and

bowel habits, including timed voiding and increased fluid intake.

When establishing a cause of microscopic or gross hematuria in the child, as in adults, it is important to differentiate between glomerular and non-glomerular causes (Table 13.3). A thorough history can help to do this. In a child with tea or cola-colored urine, history of recent pharyngitis or respiratory illness or systemic complaints like joint pain or swelling, there should be an increased index of suspicion for glomerular hematuria. In children with urinary symptoms like dysuria, weak urine stream, terminal hematuria, abdominal or back pain, non-glomerular causes like UTI, urolithiasis, or urethrorrhagia should be strongly considered.

Glomerular causes of hematuria often yield red blood cell casts and dysmorphic red blood cells on urine microscopy. Proteinuria also indicates a glomerular cause of hematuria. It is important to note that a large number of red blood cells in the urine may lead to low positive levels of protein on urine dipstick because of red blood cell lysis and release of hemoglobin into the urine [18]. Thus, it is important to quantify proteinuria with a random urine protein if glomerular disease is suspected. Importantly, all patients with microscopic or gross hematuria and elevated urine protein should undergo pediatric nephrology consultation. Additionally, patients with hypertension or other signs of systemic disease most likely have glomerular hematuria and should be evaluated further and managed by pediatric nephrology. Initial evaluation usually includes quantification of urine protein, serum chemistries, blood counts, complement components C3 and C4, antistreptolysin O, and other serologic testing [19] (see Chap. 5). Patients with suspected non-glomerular disease may undergo urinary tract imaging and/or cystourethroscopy by a pediatric urologist.

Figures 13.1 and 13.2 outline basic initial evaluation for the child with isolated asymptomatic microscopic hematuria or gross hematuria, respectively. In general, microscopic hematuria warrants a watchful waiting approach in the absence of concerning history or physical examination find-

Table 13.3 Differential diagnoses of gross and microscopic hematuria in children

Non-glomerular	Glomerular
Common causes	*Common causes*
Hypercalciuria	Thin basement membrane disease (benign familial hematuria)
Urinary tract infection	IgA nephropathy
Bacterial	Membranoproliferative glomerulonephritis
Trauma	Poststreptococcal glomerulonephritis
Perineal, urethral	
Less common causes	*Less common causes*
Exercise	Alport syndrome
Atypical urinary tract infection	Lupus nephritis
Viral, tuberculosis, fungal	Henoch-Schönlein purpura
Urolithiasis	Rapidly progressive glomerulonephritis
Urethral stricture disease	
Benign urethrorrhagia	
Tumor	
Wilms tumor, rhabdomyosarcoma, urothelial carcinoma	
Nutcracker syndrome	
Drugs	
Penicillin, sulfa, anticonvulsants, aspirin, colchicine, cyclophosphamide, indomethacin	
Sickle cell disease	
Coagulopathy	
Renal vein thrombosis	
Congenital anomaly	
Ureteropelvic junction obstruction, posterior urethral valve, polycystic kidney disease	
Papillary necrosis	
Vascular malformation	

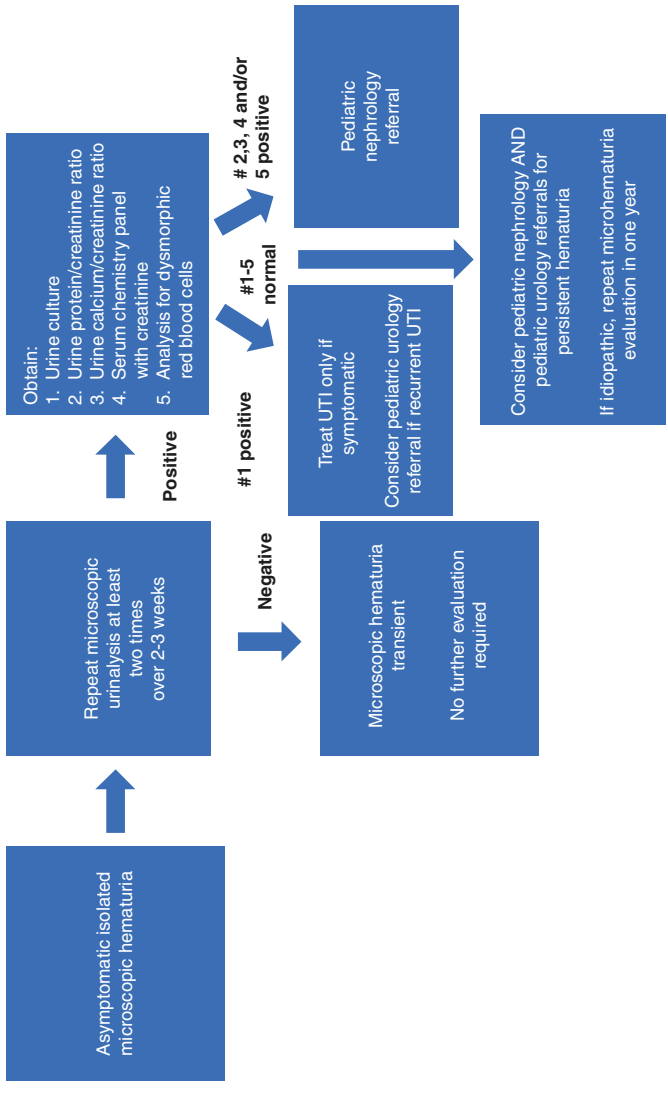

FIGURE 13.1 Algorithm for evaluation of microscopic hematuria in a child

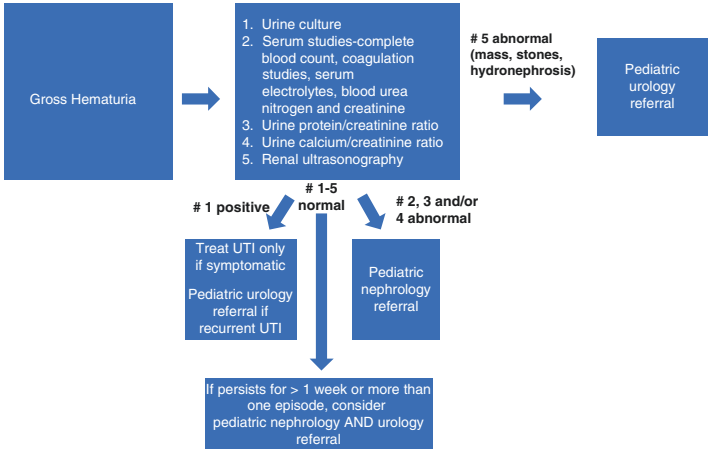

FIGURE 13.2 Algorithm for evaluation of gross hematuria in a child

ings (pain, fever, abdominal mass, edema, lower urinary tract symptoms, arthralgia, rash, hypertension, preceding or concomitant upper respiratory illness) and/or proteinuria. Gross hematuria requires a more aggressive evaluation.

Returning to case 2, this patient dysuria and microhematuria should have her urine tested for culture, calcium to creatinine ratio, protein to creatinine ratio and microscopic analysis for dysmorphic RBCs. She should also have a serum basic metabolic panel obtained. Given no protein seen on urine dipstick, a non-glomerular cause like hypercalciuria is most likely.

Case 3: Proteinuria in the Child

A 12 year old athlete presents for his annual sports physical. He has no complaints. His blood pressure is 138/89, but the rest of his vital signs are within normal limits. Abdominal exam in normal. Due to hypertension for his age, a urinalysis is obtained.

Urine dipstick & urine microscopy	
Component	**Result**
Color	Yellow
Appearance	Clear
pH	6.0
Specific gravity	1.010
Blood	Negative
Glucose	Negative
Ketone	Negative
Protein	2+
Bilirubin	Negative
Urobilinogen	Negative
Leukocyte esterase	Negative
Nitrite	Negative
White blood cells (WBC)	0–2/hpf
Red blood cells (RBC)	0–2/hpf
Squamous epithelial cells	0–2/hpf
Casts	None
Bacteria	None

Whether caused by congenital or acquired disorders (glomerulonephritis, renal scarring, longstanding diabetes mellitus) that disrupt the filtration barrier of the glomerulus, clinically relevant protein excretion in the urine occurs when the glomerular filtration barrier is altered (see Chap. 5). A new episode of isolated asymptomatic proteinuria in a child can be transient (caused by fever, strenuous exercise, cold or heat exposure, administration of epinephrine, emotional stress, seizures). If a persistent abnormality, it can be from a benign cause, like orthostatic proteinuria, or the result of serious renal disease [20]. Estimated prevalence of isolated asymptomatic proteinuria in children ranges from 0.6% to 6%. Orthostatic proteinuria accounts for 60% of isolated proteinuria in children and an

even higher percentage in adolescents [20]. Thus, it is important when evaluating proteinuria in children to obtain early morning urine samples. Recumbent urine collection for protein will be negative by dipstick with protein levels less than 100 mg/8–12 hours (or urine protein/creatinine <0.2). Upright collection will be positive by dipstick with protein levels 300–900 mg/12–16 hours (see Chap. 5). Daily urine protein varies with body mass as well as renal maturity. Urinary protein levels in children based on age are shown in Table 13.4 [21].

The American Academy of Pediatrics has discontinued its recommendation for routine screening urinalysis, as it has not been found to be cost-effective [22]. If screened, healthy children with low protein levels on dipstick are usually found to have transient proteinuria or false positive results. Evaluation should be focused on those with medical illness, systemic symptoms (hypertension, edema) and/or moderate to high levels of proteinuria, as these children are more likely to harbor renal disease. A urine protein/creatinine above 3.0 in a child is considered nephrotic range, consistent with glomeru-

Table 13.4 Normal pediatric urinary protein excretion

Age	Mean protein excretion mg/m²/day (Range)	Protein/creatinine ratio (mg/mg)
Premature infant (<30 days)	182 (8–377)	0.7
Term infant (<30 days)	145 (68–309)	0.7
2 months to 4 years	100 (37–244)	0.55–0.7 (<12 months) 0.4 (1–2 years) 0.3 (2–3 years)
5–10 years	85 (21–234)	0.2
>10 years	63 (22–181)	0.15–0.2

Adapted from [39] with permission from Elsevier
Data from Miltenyi M. Urinary protein excretion in healthy children. Clin Nephrol 1979;12:216–21 and Guignard J-P, Santos F. Laboratory investigations. In: Avner ED, Harmon WE, Niaudet P, editors. Pediatric nephrology. 5th ed. Philadelphia: Lippincott, Williams & Wilkins; 2004.

lar disease, and is often accompanied by salt and fluid retention, as well as edema [23]. Additionally, proteinuria is an independent risk factor for chronic kidney disease in children.

Differential diagnoses for proteinuria in children are similar to that in adults (see Chap. 5); however, clinically significant kidney disease in children skews toward congenital abnormalities causing renal dysplasia or urinary tract obstruction. Reflux nephropathy in children deserves specific mention, as this can present with asymptomatic proteinuria with or without hypertension in older children or adolescents, after going undiagnosed for years. Vesicoureteral reflux, or retrograde flow of urine from the bladder to the ureter and kidney, is a common congenital urinary tract abnormality that can lead to recurrent episodes of pyelonephritis, renal scarring and more rarely, renal dysplasia (reflux nephropathy). Reflux nephropathy is defined by the presence of tubulo-interstitial inflammation and fibrosis that can lead to end-stage renal disease [24]. A careful history should be obtained regarding past antibiotic use or history of urinary tract infections if this is suspected. Renal ultrasonography can show asymmetry in renal size or parenchymal defects that point toward the diagnosis of renal disease secondary to vesicoureteral reflux (Fig. 13.3).

Persistent, non-orthostatic proteinuria (elevated urine protein/creatinine on two early morning voided samples) warrants further evaluation and should be considered pathologic until proven otherwise. Evaluation includes 24-hour urine protein quantification, serum chemistries, serum creatinine, serum albumin, serum lipids, complete blood count and nephrology consultation [19]. Renal biopsy may ultimately be warranted. In most instances of pathologic proteinuria due to kidney disease, treatment is aimed at curing the underlying disease.

Orthostatic and transient proteinuria are benign and do not require treatment. In acute or self-limited processes like urinary tract infection or febrile illness, proteinuria may persist for a short time period beyond the acute disease, but does not require specific therapy unless worsening.

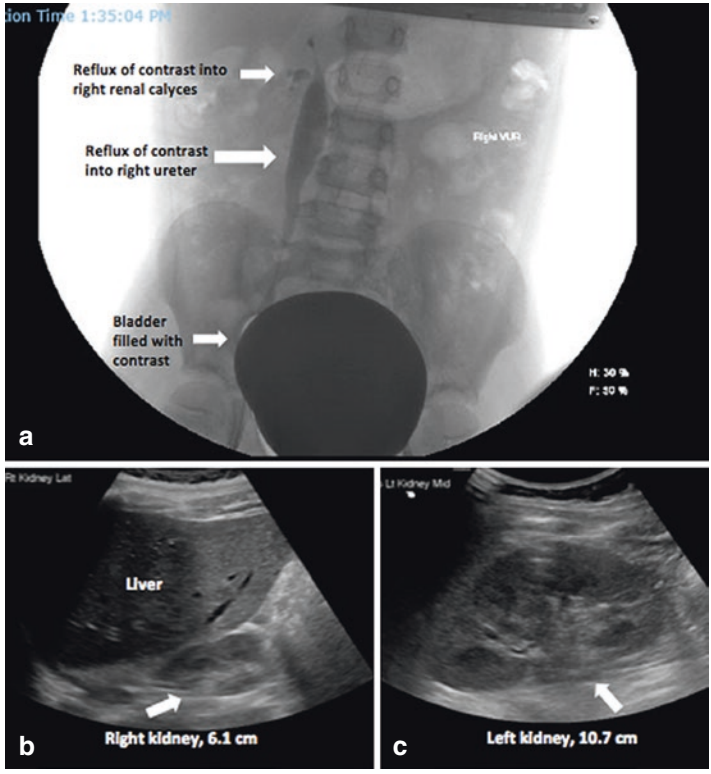

FIGURE 13.3 (**a**) Voiding cystourethrogram from 9 year old patient with right vesicoureteral reflux (**b**) Renal ultrasonography showing small echogenic right kidney adjacent to liver (**c**) Hypertrophied left kidney in same patient; mean renal length in normal 9 year old child 9.0 cm (Images courtesy of University of Iowa Hospitals and Clinics)

Returning to case 3, this patient will likely require nephrology referral after quantification of urine protein and confirmation of hypertension to rule out glomerular and renovascular disease.

Case 4: Urinary Tract Infection in the Child

A 7 year old potty-trained child presents to the outpatient clinic with a 1 week history of daytime urinary incontinence and urinary frequency. She denies dysuria and hematuria. She has been diagnosed with three culture proven urinary tract infections in the last year, which presented in a similar manner. She is afebrile, and vital signs are stable. She has mild suprapubic tenderness to palpation but no costovertebral angle tenderness.

Urine dipstick and urine microscopy	
Component	**Result**
Color	Yellow
Appearance	Cloudy
pH	6.0
Specific gravity	1.020
Blood	Trace
Glucose	Negative
Ketone	Negative
Protein	2+
Bilirubin	Negative
Urobilinogen	Negative
Leukocyte esterase	1+
Nitrite	Positive
White blood cells (WBC)	25–50/hpf
Red blood cells (RBC)	0–2/hpf
Squamous epithelial cells	0–2/hpf
Casts	None
Bacteria/other	2+

Urine culture obtained shows > 100,000 colonies Escherichia coli/hpf

As in adults, diagnosis of a urinary tract infection in children requires not only proof of organisms in the urinary tract (usually via urine culture), but also confirmation that the organism is pathogenic (validated by symptoms or evidence of immune response in urine or blood tests). Asymptomatic bacteriuria does not require antibiotic treatment, nor is screening for it recommended in children with no anatomic abnormalities of the genitourinary tract [25, 26].

In children less than 24 months in whom a UTI is suspected, fever is often the only symptom, but symptoms can also include irritability, poor feeding, jaundice, failure to thrive, vomiting, diarrhea, abdominal distention and/or foul-smelling urine [27, 28]. A UTI should be considered in any febrile infant, especially in the absence of another source of infection or in the presence of a known urinary tract abnormality like hydronephrosis or vesicoureteral reflux. The first year of life is the only time in which the risk of UTI is greater in boys than it is in girls. Additionally, during the first year, the risk of UTI in the uncircumcised boy is almost 10 times that in a circumcised boy [29–31].

In children older than 24 months, symptoms classically associated with adult UTIs may become more apparent as children begin to verbalize their complaints. Dysuria, incontinence, changes in voiding habits, nocturnal enuresis, flank and abdominal pain can all be reported and increase the likelihood of UTI diagnosis [32]; however, it must be considered that these symptoms are nonspecific for UTI and are also often see in children with bowel and bladder dysfunction, urinary calculi, and vulvovaginitis in the absence of UTI.

Sexual abuse in children is not thought to significantly increase rates of urinary tract infection, but there is a high rate of urinary tract symptoms in children who have been sexually abused, including dysuria, incontinence and genital discomfort [33]. Urine testing in suspected victims of sexual abuse is usually limited to ruling out sexually transmitted infection.

A history of febrile urinary tract infections or recurrent symptomatic afebrile urinary tract infections in a child should

not be considered normal. UTIs can signify anatomic abnormalities of the genitourinary tract, including obstruction or vesicoureteral reflux. Recurrent episodes of pyelonephritis in a child can ultimately lead to renal scarring, as well as chronic kidney disease and hypertension. Febrile UTIs, as well as UTIs associated with nausea, vomiting, or flank pain, should be considered as pyelonephritis and treated empirically in children to prevent renal scarring. Thus, urinary tract imaging and/or pediatric urologic consultation is warranted in children with history of a single febrile urinary tract infection or recurrent afebrile urinary tract infections.

One febrile urinary tract infection or recurrent afebrile urinary tract infections warrant obtaining renal bladder ultrasound, given its availability, low cost, and minimally-invasive nature. This practice is recommended by the American Academy of Pediatrics and National Institute for Health and Clinical Excellence [34]. Ultrasound can detect abnormalities that indicate urinary stasis and thus predispose to infection, like hydronephrosis and bladder diverticuli. Ultrasound can also identify nephrolithiasis, which may act as a reservoir for urinary tract infection. The appropriate role of voiding cystourethrogram and renal nuclear scintigraphy continues to be investigated, and their use is outside the scope of this chapter. Their use is best determined by a pediatric nephrologist or urologist.

Treatment of both febrile and afebrile UTIs in children is similar to that in adults. The most common pathogenic organisms in children are similar to those in adults and include *Escherichia coli, Proteus mirabilis*, and *Klebsiella* species. Uncomplicated cystitis can be treated with a short, three-day antibiotic course based on local antibiograms. Pyelonephritis requires at least a 7-day, and sometimes up to 14-day course of oral or intravenous antibiotics, based on the patient's clinical status. Ciprofloxacin should be used with caution in children because of concerns for abnormal cartilage development, and tetracyclines are avoided in children less than 8 years of age because of the risk of permanent tooth discoloration.

There is an increased presence of *Enterococcus* species in infants, which should be recognized and considered when

making an antibiotic choice. Amoxicillin is often the antibiotic of choice in infants less than 2 months of age, and trimethoprim-sulfamethoxazole should not be used until after this period because of the risk of bilirubin displacement leading to unconjugated hyperbilirubinemia and kernicterus. The decision to use antibiotic prophylaxis for recurrent urinary tract infections is usually best left to pediatric urology and/or nephrology. Risk factors for recurrent urinary tract infections in children include infrequent voiding, inadequate fluid intake, poor toileting posture, constipation, nephrolithiasis and anatomic genitourinary abnormalities. Children with recurrent UTIs should be educated and counseled on behavioral modification to alleviate modifiable factors [35].

Returning to case 4, this child should be treated for her UTI based on culture sensitivities. She should be referred to pediatric urology to rule out anatomic causes of recurrent UTI or functional bowel and bladder dysfunction.

Summary

From collecting urine to interpreting urine test results, there are many nuances to pediatric urine testing. Special care should be taken by the primary care practitioner to understand differential diagnoses of abnormal urine testing in children, given that management strategies differ greatly from adults. Additionally, caution should be taken in interpreting potentially contaminated specimens in the pediatric patient because of high rates of false positive results, especially in the diagnosis of urinary tract infection.

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Chapter 14
Urine Based Tests in the Diagnosis of Genitourinary Cancers

Morgan Schubbe, Laila Dahmoush, and Kenneth G. Nepple

Objectives

- Describe the role of urine-based tests in bladder cancer diagnosis and surveillance
- Understand novel methods of urine testing for bladder cancer and the clinical scenarios in which use of these tests is appropriate
- Describe the role of urine-based tests in prostate cancer diagnosis and management
- Understand novel methods of urine testing for prostate cancer and the clinical scenarios in which use of these tests is appropriate

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Overview

This chapter will discuss some of the currently available urine markers for bladder cancer and prostate cancer and the appropriate clinical scenarios in which each test may be used. Given the complexity of urine testing for malignancy, urologists are best equipped to order these tests and interpret the results in the clinical context of each patient. The tests presented here should not be ordered in the primary care setting.

Testing for Genitourinary Cancer

Genitourinary malignancies are some of the most frequently diagnosed cancers, with prostate the most common and bladder the fourth most common cancer diagnosed in American men [1]. Long-term follow up for these diseases is invasive and expensive. There has been a trend to develop urinary markers that may serve as accurate, non-invasive, inexpensive ways to evaluate for these cancers or to monitor patients on surveillance. An ideal diagnostic test should have both a high specificity and sensitivity, that is the ability to correctly identify those patients who do not have the disease and thus avoid unnecessary costly workup, and to correctly identify those patients who do have the disease and require further treatment. Table 14.1 gives an overview of available urine testing used in the management of bladder and prostate cancers.

Case 1: Urine Cytology for Bladder Cancer

A 75 year old male with a 50 pack-year smoking history presents to his primary care physician with complaints of urinary frequency and mild dysuria over the past 3 weeks.

Table 14.1 General information for urinary bladder cancer tests including approximate cost, availability, and result turnover

Diagnostic test	Uses	Sensitivity (%)	Specificity (%)	Approximate cost	Availability	Result turnover
Cytology	• Bladder cancer surveillance	20–50	80–100	$662	Most hospital pathology labs	1–2 business days
	• Bladder cancer diagnosis	43	95			
Urovysion® FISH	• Bladder cancer surveillance • Hematuria workup • Equivocal cytology	60	87	$1000	Most academic centers, community hospitals may require mail out	Days to weeks, depending on how often the assay is run at a given hospital or if mail out is required
lmmunocyt/ uCyt+	• Bladder cancer surveillance • Bladder cancer diagnosis • Adjunct to cytology	72.5	65.7	$820	Some academic centers, community hospitals may require mail out	Days to weeks, depending on how often the assay is run at a given hospital or if mail out is required

(continued)

Table 14.1 (continued)

Diagnostic test	Uses	Sensitivity (%)	Specificity (%)	Approximate cost	Availability	Result turnover
Cxbladder Triage	• Bladder cancer diagnosis in low risk patients	98 NPV		$2995	Mail out	5 business days
Cxbladder Detect	• Bladder cancer diagnosis in high risk patients	81.8	85.1	$2995	Mail out	5 business days
Cxbladder Monitor	• Bladder cancer surveillance	93	97	$2995	Mail out	5 business days

Urovysion® FISH is manufactured by Abbott Laboratories.
Immunocyt/uCyt+ is manufactured by Scimedx, Inc.
The Cxbladder tests are manufactured by Pacific Edge, Inc.
NPV negative predictive value

He has no recent history of genitourinary trauma or instrumentation:

Urine dipstick and urine microscopy	
Component	**Result**
Color	Red
Appearance	Cloudy
pH	6.2
Specific gravity	1.015
Blood	3+
Glucose	Negative
Ketone	Negative
Protein	Negative
Bilirubin	Negative
Urobilinogen	Negative
Leukocyte esterase	Trace
Nitrite	Negative
White blood cells (WBC)	0–5/hpf
Red blood cells (RBC)	25–50/hpf
Squamous epithelial cells	8/lpf
Casts	0/hpf
Bacteria	Few/hpf

Urine culture is negative. Would obtaining a urine sample for cytology be an appropriate next step in this patient's evaluation?

Evaluation of microscopic or gross hematuria in the absence of a benign cause such as menstruation or genitourinary instrumentation often warrants referral to a urologist and/or nephrologist (see Chap. 9). Asymptomatic microscopic hematuria is indicative of underlying urinary tract malignancy in approximately 3.3% of patients [2]. Thus, the most recent American Urological Association (AUA) guidelines for hematuria evalu-

ation require cystoscopy as well as upper tract imaging for most patients to rule out genitourinary malignancy.

Urine cytology may be used in the initial workup of primary hematuria but is not recommended by AUA guidelines as a necessary component of the evaluation of asymptomatic microscopic hematuria [2]. Urine cytology is the most widely available urinary screening test and has long been part of standard bladder cancer surveillance protocols. Urine may be collected as a voided specimen or as a bladder wash/barbotage through the cystoscope. Barbotage (collected by vigorously flushing and withdrawing urine through a 60 cc syringe) increases urothelial sloughing, thus increasing the number of cells in the sample, but may miss sampling the prostatic urethra. Voided urine will sample the urothelial cells of the prostatic urethra and should be collected after the second morning void as cells in the bladder may degenerate overnight making interpretation difficult. Ideal urine collection volume is at least 20–30 mL. The sample is then analyzed by a pathologist who reports on the presence of atypical cells or malignant cells consistent with high grade urothelial carcinoma (Fig. 14.1).

Given the well known difficulty of recognizing low grade urothelial carcinomas in urine cytology, negative specimens are currently reported as: "negative for high grade urothelial carcinoma." Urine cytology testing is similar to the cytopathology evaluation of a Pap test (Pap smear) in gynecologic cancer.

In the surveillance setting, cytology has high specificity overall, ranging from 80–100%, but has poor sensitivity of 20–50% [3]. Sensitivity does improve with increased grade of tumor, ranging from 4–31% for low grade tumors and 38–84% for high grade tumors but can still miss clinically significant tumors [3]. It is also susceptible to high interobserver variability amongst pathologists [4].

Similarly in a primary hematuria evaluation, cytology has a specificity of approximately 95% and sensitivity of approximately 43% [5]. Sensitivity improves significantly when combined with upper tract imaging, increasing up to 90% with CT urogram and 66.7% with renal ultrasound; however, urine cytology is still appropriate only as an adjunctive test and does not preclude the need for cystoscopy [5].

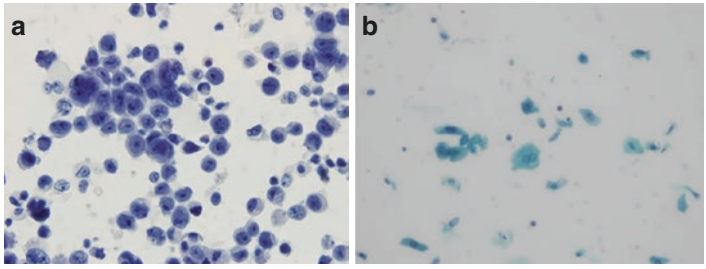

FIGURE 14.1 (**a**) Voided urine sample with malignant high-grade urothelial carcinoma cells, identified with Papanicolaou stain. These cells show large nuclei with a high nuclear-to-cytoplasmic ratio. There is dense chromatin within irregular nuclear borders. Many cells will also demonstrate prominent nucleoli. The background of the slide also demonstrates inflammatory or necrotic debris. (**b**) Normal urine cytology showing normal superficial urothelial cells with large round nuclei (commonly seen in pairs), central and prominent nucleoli, and abundant granular cytoplasm. Other small cells seen may represent lymphocytes (normal finding). (Courtesy of Laila Dahmoush, MD, University of Iowa, Department of Pathology)

Additional pitfalls of cytology include false negative or false positive results which may subject patients to unnecessary invasive procedures, as well as the diagnostic dilemma associated with indeterminate "atypical" results. False negative cytology results can be due to inadequate sample collection, paucity of cells or presence of inflammatory cells or other debris that obscure accurate interpretation. False positive cytology results may be due to recent instrumentation, therapeutic changes associated with radiation or other treatment, such as with intravesical BCG (*bacille Calmette-Guérin*) for high grade bladder cancers. Given the variable quality and accuracy of cytology interpretation even at academic centers, this is a test that should be reserved for use by urologists and should not be routinely ordered by primary care providers.

Returning to case 1, given the red color of this patient's urine and 25–50 RBCs on urinalysis, he has gross hematuria. A urine culture should be obtained to rule out UTI, but if negative, or if hematuria persists after treatment for positive culture, espe-

cially given his significant smoking history, he should undergo urology consultation. Cystoscopy and upper tract imaging should be performed. Urine cytology could be obtained as a voided specimen or during cystoscopy as an adjunctive test to direct further management if the diagnosis is in question.

Case 2: Urinary Biomarkers for Bladder Cancer

The patient from Case 1 is seen by a urologist for hematuria workup. He undergoes CT urogram which is unremarkable for obvious pathology. Cystoscopy shows some patchy red areas in the bladder, and cytology from the bladder wash returns as positive for high-grade urothelial carcinoma. Subsequent bladder biopsies are positive for urothelial carcinoma in situ (high grade superficial bladder cancer) and he is scheduled for a 6-week induction course of intravesical BCG. At this point, his bladder cancer treatment and surveillance will be managed by the urologist.

Several novel biomarkers in the urine have been investigated in recent years to be used for initial diagnosis of genitourinary malignancy. Urine tests are also routinely used in the ongoing surveillance of bladder cancer patients to detect recurrence or help assess response to intravesical treatments. The following is an overview of some of the currently available markers used in the management of bladder cancer.

Urovysion®/FISH

The Urovysion® FISH assay is a lab test performed on urine collected from a voided specimen or bladder wash at time of cystoscopy (Table 14.1). The assay uses fluorescence in situ hybridization (FISH) to detect common DNA aberrations associated with urothelial cancers, specifically aneuploidy in chromosomes 3, 7, 17, and the 9p21 locus. Chromosomal aberrations are present in the majority of urothelial cancers with alterations in chromosome 9 the most common, occurring in

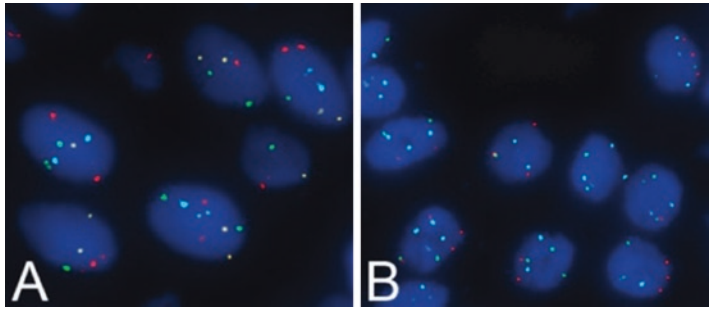

Figure 14.2 FISH results showing gain of at least 2 chromosomes or a homozygous loss of a chromosome. Both pictures A and B show abnormal FISH results. Each numbered chromosome (3, 7, and 9) is labeled with a different colored fluorescent marker and is normally present in pairs (two copies of each color). Any gain (more than 2 copies) or loss (1 copy) of a chromosome is considered abnormal [9]. Figure used with permission from the Creative Commons Attribution License

nearly 50% of urothelial cancers [6]. Results are then manually reviewed by trained technologists and are reported qualitatively as "positive" or "negative" (Fig. 14.2). There has been some data suggesting that quantitative results may also play a role in risk stratification [10].

Urovysion® FISH has an overall sensitivity of approximately 60% and specificity of 87% [7]. The test has FDA approval for use in both detecting recurrent tumors in patients with a history of bladder cancer as well as for detection of bladder cancer in patients presenting with hematuria but no prior history of bladder cancer. FISH may also be useful when cystoscopy does not identify a tumor and cytology results are equivocal [11]. This assay is still utilized in conjunction with standard cystoscopy and upper tract evaluation and should not be used as the sole test in bladder cancer evaluation.

Immunocyt/uCyt+

Immunocyt/uCyt+ (Table 14.1) is a urine-based laboratory test that uses three fluorescently labeled monoclonal antibodies (LDQ10, M344, CEA 19A211) to detect malignant

urothelial cells shed in the urine. M344 and CEA 19A211 have been found to be expressed in up to 90% of superficial bladder tumors [12]. Overall, Immunocyt/uCyt+ has sensitivity of 72.5% with specificity of 65.7% [13]. When used with standard cytology, the two tests have a combined sensitivity of 83.3% with improved detection of low grade tumors [13]. Immunocyt/uCyt+ is not recommended for independent use and can be used in combination with standard cystoscopy and cytology for bladder cancer diagnosis and surveillance.

Cxbladder

Cxbladder is a trio of urine-based laboratory tests intended to help detect the presence or absence of bladder cancer (Table 14.1). Samples are collected via mid-stream catch. The assays measure gene expression levels of five messenger RNA (mRNA) biomarkers including MDK, HOXA13, CDC2, IGFBP5, CXCR2 that are involved in various stages of cell proliferation and migration and are found in higher concentrations in the urine of patients with bladder cancer. The calculated relationship between the quantified gene expressions provides a probability of the presence of urothelial cancer in each appropriately selected patient.

Cxbladder Triage is designed as a diagnostic test to potentially replace cystoscopy for evaluation of low risk patients presenting with hematuria. It also identifies patients who still require standard hematuria workup. Genotypic information is combined with phenotypic information (age, gender, smoking history, degree of hematuria) in a model designed to predict patients with a low probability of urothelial cancer. The result will provide a calculated gene expression value that predicts the probability of the presence of urothelial cancer in a low risk patient. Cxbladder Triage has shown promising results in a validation study performed by its manufacturers with NPV of 98%, however more research is required before it can be considered for widespread clinical adoption [14].

Cxbladder Detect is designed as an adjunctive test to predict the probability of bladder cancer in patients presenting

with gross hematuria and with more risk factors for malignancy, and is meant to be used with standard cystoscopic evaluation. Again, the result provides a calculated gene expression value that predicts the probability of urothelial cancer in a higher risk population. It has an overall sensitivity of 81.8% and specificity of 85.1% and has been shown to detect up to 97% of high-grade tumors and 69% of low-grade tumors [15].

Cxbladder Monitor is designed for use in the surveillance setting in patients with a history of primary or recurrent urothelial cancer. It is to be used in conjunction with cystoscopy. In an internally validated study, the test had a sensitivity of 93% and negative predictive value of 97% for ruling out recurrent bladder cancer [16]. Even when stratified by tumor grade, Cxbladder Monitor maintained 97% sensitivity for high-grade tumors and 85% for low-grade tumors [16]. While more studies are needed, Cxbladder Monitor does show promise in potentially decreasing the number and frequency of cystoscopies required in the surveillance of bladder cancer.

In summary, the AUA Guidelines for asymptomatic microscopic hematuria do not recommend use of cytology in initial hematuria evaluation, and also discourage the use of biomarkers in place of cystoscopy for ongoing bladder cancer surveillance; however, biomarkers may be useful in surveillance or assessing response to intravesical treatments. Markers such as Urovysion® FISH and Immunocyt/uCyt+ may also be used in the setting of equivocal cytology [8].

Returning to case 2, after treatment with BCG, urinary biomarkers could be used as adjunctive tests in ongoing surveillance of his cancer.

Case 3: Urinary Biomarkers for Prostate Cancer

A 62 year old male has been followed by his urologist for many years for elevated prostate specific antigen (PSA). His current PSA is 8.4 ng/mL (elevated) and digital rectal exam

(DRE) is normal with no enlargement of the prostate. He has undergone 2 previous transrectal ultrasound (TRUS) guided prostate biopsies with benign results. He has no family history of prostate cancer. The patient is reluctant to undergo a third prostate biopsy. Multiparametric MRI is discussed as an option for testing, but the patient is claustrophobic. Is there any additional testing that may help determine if an additional biopsy is warranted?

PSA screening for prostate cancer has long been a controversial topic. PSA is a substance produced by prostatic tissue and can be elevated for a number of reasons including prostate cancer, but also inflammation, infection (as in cases of urinary tract infection or prostatitis), recent catheter placement, or simply a large prostate gland (as with benign prostatic hyperplasia or BPH).

Prostate cancer diagnosis has traditionally relied on prostate biopsy, with biopsy prompted by elevated PSA, abnormal digital rectal examination, or a combination of the two. There are a large number of patients, as the patient highlighted in Case 3, with a history of elevated PSA but with a negative prostate biopsy. Prostate biopsy has inherent risks including urinary retention, hematuria, hematochezia, hematospermia, and erectile dysfunction which are typically minor and self-limiting. There is also a risk of infection or life-threatening sepsis that can occur despite appropriate antibacterial prophylaxis, with international meta-analyses reporting hospitalization rates of 0–6.3% [17]. Prostate biopsy can also miss a cancer diagnosis, if cancer is in a region that is not adequately sampled. Multiparametric MRI is becoming increasingly utilized in the diagnosis of prostate cancer, especially in patients in whom PSA and biopsy results are incongruous or who are on active surveillance regimens. However, MRI is expensive and contraindicated in some patients.

There are no definitive guidelines for management of patients with elevated PSA and negative prostate biopsy, and urologists rely on clinical judgment in combination

Table 14.2 General information for urinary prostate cancer tests, including approximate cost, availability, and result turnover

Diagnostic test	Uses	Prostate biopsy reduction (%)	Approximate cost	Availability	Result turnover
Progensa® PCA3	Men >50 with prior negative biopsy, now indicated for repeat biopsy	37–77	$802	Mail out	1–2 weeks
Select MDx®	Elevated PSA, prior to first prostate biopsy	40	$500	Mail out	5 business days

Progensa® PCA3 is manufactured by Hologic, Inc.
SelectMDx® is manufactured by MDxHealth.

with shared decision making with each patient to decide who should undergo continued surveillance versus repeat biopsy. This has prompted research into potential biomarkers that may be used to help predict the presence of prostate cancer. The following prostate biomarkers are intended for use in the clinical decision-making process between the patient and the urologist and are not recommended in the primary care setting. Table 14.2 gives an overview of these biomarkers.

Progensa® PCA3

Prostate cancer antigen 3 (PCA3) is a prostate-specific mRNA biomarker that is overexpressed in prostate cancer. The Progensa® PCA3 assay is designed to detect the overexpression of PCA3 in a urine sample. The patient undergoes a digital rectal examination, including three gentle strokes per prostate lobe to release sufficient prostate cells into the urine. The urine sample is then collected immediately after the DRE and sent to a PCA3 lab. The assay measures the ratio of PCA3 RNA molecules to PSA RNA molecules and reports this as a score. PCA3 score is not affected by prostate volume. The score is then used together with other patient specific clinical information to determine the need for repeat biopsy. A PCA3 score <25 predicts a decreased likelihood of positive prostate biopsy. A meta-analysis of PCA3 studies reported reduction in prostate biopsies ranging from 37–77% based on score cutoffs from 20 to 35, but that reduction in biopsies missed between 9% and 22% of prostate cancers [18]. Progensa® PCA3 is currently FDA approved for use in men over the age of 50 who have had at least one prior negative biopsy and who would be indicated for repeat biopsy based on the current standard of care. This test is most useful as a risk stratification tool and should be used together with clinical history, PSA, and DRE to ultimately decide when repeat biopsy is warranted.

SelectMDx®

SelectMDx® is a urine-based assay that measures the expression of two mRNA cancer-related biomarkers, HOXC6 and DLX1, that are upregulated in prostate cancer [19]. This test helps to determine a patient's risk of having prostate cancer, and when used in conjunction with clinical history, can help inform the urologist whether or not to proceed with prostate biopsy. A midstream urine sample is collected after digital rectal examination. Results are reported as either "increased risk" or "very low risk" for clinically significant prostate cancer. Preliminary studies have shown approximate sensitivity of 91% and NPV of 94% with avoidance of up to 40% of prostate biopsies and only 1–2% chance of missing a high-risk prostate cancer [20, 21]. SelectMDx® is validated to be used prior to first prostate biopsy but has not yet received FDA approval and is not recommended by current AUA guidelines.

Returning to case 3, both PCA3 and SelectMDx testing could be used as adjunctive testing to help risk stratify this patient, determining his likelihood of clinically significant prostate cancer.

Summary

Patients are living longer with chronic diseases such as bladder and prostate cancers that require long-term surveillance. Bladder and prostate cancers are two urologic diseases that require intensive lifelong follow up that can be invasive, expensive, and place patients at risk of infection or other harms. Several companies and researchers have been working on developing urinary biomarker assays that are non-invasive and less expensive than other testing, that may obviate the need for cystoscopies or biopsies in appropriately selected patients. Despite promising research, none of these tests have yet replaced the current standards of care, and should be used only to provide adjunctive information.

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Chapter 15
Kidney Excretions: The Lyter Side of Urine

Jeremy Steinman, Carly Kuehn, and Lisa M. Antes

Objectives

- Understand the categories to classify hypokalemia, the differential diagnosis in each category and the use of urine potassium to help narrow the differential
- Understand the relevance of hypokalemia associated with hyperaldosteronism and initial testing considerations
- Recognize Type I, II and IV renal tubular acidosis (RTA), some common causes of each and the implications for obtaining a urine anion gap when evaluating patients with suspected RTA

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- Use urine osmolarity to sort polyuria into a solute or water diuresis and describe common causes of each
- Understand the basic physiologic principles behind a water deprivation test
- Understand the limitations and use of fractional excretion of sodium and fractional excretion of urea in patients with acute kidney injury

Overview

The evaluation of serum electrolytes is part of a basic laboratory workup for various conditions and clinical signs or symptoms. Urine testing can be a powerful tool to further delineate some conditions not clearly understood from history, physical exam and serum electrolyte evaluation alone. This chapter shows the clinician the physiology behind metabolic conditions commonly evaluated with urine electrolytes (hypokalemia, renal tubular acidosis, acute kidney injury) and how urine testing aids in evaluation (polyuria).

Case 1: Hypokalemia Evaluation

A 20 year old female with no past medical history presents to your clinic for an evaluation of muscle cramps. She has no other complaints. BP is 113/80 and physical exam is unremarkable. A basic metabolic panel (BMP) reveals a potassium (K) of 2.9 mEq/L (normal 3.5–5 mEq/L). What else would you ask her and how would you proceed with the evaluation?

Hypokalemia is a common issue encountered in both the primary care and inpatient settings. The differential diagnosis is broad, but can be narrowed with a few key clinical clues. There are **4 main categories**:

Cause of hypokalemia	Common example
1. Increased renal losses of potassium	Diuretic use
2. Increased extrarenal losses of potassium (mostly GI tract)	Diarrhea (e.g., enterocolitis)
3. Decreased dietary intake	Malnourishment
4. Transcellular shifts	Continuous insulin infusions

Renal losses are most easily evaluated with assessment of urine K excretion. In this regard, it is important to understand the context and the response expected by the kidneys. In the face of low serum potassium, the kidney should be conserving (or reabsorbing) potassium and the urine potassium value should therefore be low (< 20 mEq/day or a random urine specimen with < 15mEq/L). If the urinary potassium is high or within the normal range in the face of hypokalemia, this represents pathologic potassium loss in the urine.

Renal losses comprise a diverse category, including medication associated urine K losses, osmotic diuresis, renal tubular defects and mineralocorticoid excess states [1]. A thorough medication history is imperative when evaluating renal losses of potassium, as several drugs are known to cause renal K wasting. In addition, several medications are known to cause hypomagnesemia (Table 15.1), which in turn can contribute to hypokalemia. Magnesium (Mg) facilitates K reabsorption by the kidney. Checking a serum Mg level is a simple way to explore this possiblity. Patients with medication associated hypokalemia may have concomitant acid-base disorders, as is the case with diuretic therapy or normal acid-base status, as is the case with proton pump inhibitors (PPI). Osmotic diuresis (e.g., hyperglycemia) can result in renal K losses as well.

Two important causes of excessive renal potassium losses are type I and II renal tubular acidosis (RTA) and hyperaldosteronism. These can be differentiated by evaluation of acid-base status and blood pressure. Patients with RTA have

Table 15.1 Medications associated with hypomagnesemia

Renal Mg losses
Diuretics (loop and thiazide type)
Aminoglycosides
Calcineurin inhibitors
Cisplatin/carboplatin
Foscarnet
Amphotericin B
GI Mg losses
Proton pump inhibitors

a non-anion gap metabolic acidosis. Patients with hyperaldosteronism have hypertension and either a normal acid-base status or metabolic alkalosis. There are also rarer conditions, both genetic and acquired, in which patients have a syndrome of primary renal potassium losses, metabolic alkalosis and salt wasting: Bartter and Gitelman syndromes are the most common (see Chap. 1). While the lab abnormalities may mimic hyperaldosteronism, these latter conditions are characterized by low or normal blood pressures and not hypertension.

Extra-renal losses of potassium can be seen with GI tract losses of K either from the upper tract or lower tract. Upper tract losses include nasogastric tube drainage or vomiting resulting in volume depletion and increase in aldosterone; aldosterone then increases urinary losses of K. Such a patient may also have a concomitant metabolic alkalosis. Lower tract causes such as diarrhea or surreptitious laxative use can also be considered, and may have an associated non-anion gap metabolic acidosis (NAGMA).

It is always important to assess **diet** when evaluating patients with hypokalemia. Normal intake of potassium ranges from 80 to 120 mEq per day in patients with nor-

mal renal function. Patients with malnutrition as a cause of hypokalemia are likely to have other abnormal parameters, including unexplained weight loss, a low serum blood urea nitrogen (BUN) and low serum phosphorus, as well as low prealbumin.

Transcellular shifts may occur for a variety of reasons. In the hospitalized setting, transcellular shifts can occur in those receiving excessive beta-2 agonism (e.g., albuterol use in status asthmaticus) or continuous insulin infusions used to treat diabetic ketoacidosis. The exchange of potassium between the extracellular space and skeletal muscle is mediated by specific membrane transporters (sodium-potassium ATPase = Na^+-K^+ pump). Another rare type of transcellular shift, hypokalemic periodic paralysis, causes muscle cramps after a high carbohydrate meal in certain populations due to a fall in blood K levels. Other causes of transcellular shifts result from increased cellular formation (e.g., red blood cell proliferation post vitamin B12 therapy or white blood cell proliferation post granulocyte colony stimulating factor treatment) or hypothermia related hypokalemia in critically ill patients.

Returning to case 1, this young woman with normal blood pressure had labs with persistent hypokalemia and serum bicarbonate of 28 mEq/L (normal 23–25). Her urine potassium was >40 mEq/L (consistent with renal loss). Her serum magnesium was 2.0 mEq/L (normal range 1.5–2.5 mEq/L). She was on no medications. Since the primary physician felt this was a renal loss of potassium, the patient was referred to nephrology clinic. Patient underwent a diuretic screen to exclude surreptitious use of diuretics, which was negative. She was suspected to have Gitelman syndrome, a condition in which there is a defect in the thiazide sensitive NaCl cotransporter. She was given potassium supplements and told to ensure adequate intake of dietary sodium chloride, and was referred for further genetic testing (done in specialized centers).

Case 2: Hypokalemia Associated with Hypertension

A 21 year old female with a blood pressure of 152/84 presents to your clinic. Her labs are as follows: sodium 141 meq/L (normal 135–145 mEq/L), potassium 3.2 mEq/L (normal 3.5–5 mEq/L), chloride 101 mEq/L (normal 95–107 mEq/L), bicarbonate 33 mEq/L (normal 23–25 mEq/L), BUN 13 mg/dL (normal 7–20), creatinine 0.7 mg/dL (normal 0.5–1.1). What are the next steps in evaluation?

While 90–95% of cases of hypertension (HTN) are termed essential or primary, secondary HTN may be suggested by **symptoms** (e.g., flushing and sweating suggestive of a pheochromocytoma), **physical exam findings** (e.g., renal bruit suggestive of renal artery stenosis) or **laboratory abnormalities** (e.g., unprovoked hypokalemia suggestive of hyperaldosteronism). Secondary HTN should also be considered in patients with resistant HTN, a severe or accelerated course of HTN, early or late onset HTN or specific anti-hypertensive intolerances. It is important to consider secondary causes because these imply an underlying, potentially correctable cause (Table 15.2).

Table 15.2 Evaluation of secondary causes of hypertension [2]

Etiology	Signs and symptoms	Screenings tests/ findings
Renal parenchymal disease	Edema, HTN	Elevated serum creatinine or decreased eGFR. Abnormal urine sediment (cells, casts) Abnormal urine dipstick (proteinuria, hematuria)

Table 15.2 (continued)

Etiology	Signs and symptoms	Screenings tests/ findings
Renovascular Hypertension (Renal Artery Stenosis)	Previously well controlled HTN, now uncontrolled or resistant; recent onset HTN in elderly with vascular disease or in very young (esp females with fibromuscular dysplasia); increased risk in smokers or those with extensive vascular disease; possible abdominal renal bruit	Continued rise in serum creatinine with initiation of RAS blocking agents. US may show disparity in kidney size. Renal dopplers may show elevated resistive indices and parvus tardus waveforms
Drug-Induced	Active NSAID use or catecholamine releasing drugs (cocaine, amphetamines)	Drug screening
Aldosterone Excess	Unprovoked hypokalemia	Abnormal aldosterone:renin ratio (ARR> 20); abnormal response to sodium loading
Pheochromocytoma	Flushing, palpitations, paroxysms of HTN (labile), diaphoresis	Abnormal urinary fractionated catecholamine excretion (metanephrines and normetanephrines); abnormal plasma free metanephrines

(continued)

Table 15.2 (continued)

Etiology	Signs and symptoms	Screenings tests/ findings
Cushing's Syndrome	Central obesity, striae, muscle weakness, moon facies, elevated blood glucose, fluid retention	Increased 24-hour urinary cortisol; positive low dose dexamethasone suppression test or midnight salivary cortisol
Thyroid under or overactivity	Tachycardia, weight loss, anxiety, elevated SBP for overactive thyroid vs bradycardia, weight gain, fatigue, elevated DBP for underactive thyroid	Abnormal TSH and sometimes abnormal free T4
Obstructive Sleep Apnea (OSA)	Snoring, interrupted sleep, daytime somnolence, stout neck, obesity	Abnormal polysomnography (sleep study), Sleep Apnea Clinical Score with nighttime pulse oximetry
Coarctation of the aorta	Brachial:femoral pulse differential/ delay, systolic bruits in back/chest, arm to leg SBP difference > 20 mmHg	Imaging of chest (rib notching); abnormal ECHO (children) or MRI (adults)

ACE-i angiotensin converting enzyme inhibitors, *ARB* angiotensin receptor blockers, *ARR* aldosterone:renin ratio, *DBP* diastolic blood pressure, *ECHO* echocardiogram, *eGFR* estimated glomerular filtration rate, *Hypertension* hypertension, *MRI* magnetic resonance imaging, *NSAIDs* nonsteroidal anti-inflammatory drugs, *SBP* systolic blood pressure, *T4* thyroxine, *TSH* thyroid stimulating hormone, *US* ultrasound

Returning to case 2, HTN in an otherwise healthy young woman should prompt consideration of a secondary cause of HTN. Additional history is elicited and a thorough physical exam is performed. She is taking no medications that can elevate blood pressure (e.g., herbal supplements, sympathomimetics, oral contraceptives). Her body mass index (BMI) is 21 and she has no symptoms or body features to suggest OSA. Similarly, she has no symptoms or exam findings of cortisol or thyroid hormone excess. She has no arm to leg BP difference. The main differential diagnosis for a secondary cause of her HTN would rest between renovascular HTN and hyperaldosteronism. The latter is particularly suggested by HTN associated with unprovoked hypokalemia and metabolic alkalosis.

Case 3: Hyperaldosteronism Diagnosis and Evaluation

The patient in case 2 had a random urine K of 80 mEq/L, which indicates an inappropriately high K excretion in the face of hypokalemia. This supports the previous suspicion that her metabolic abnormalities and hypertension are due to some form of hyperaldosteronism, resulting in renal K wasting.

The next step in evaluation would be to obtain labs for plasma aldosterone concentration (PAC) and plasma renin activity (PRA). Her PAC returns at 35 ng/dL (reference range, upright: 4–31 ng/dL) and her PRA returns at 0.4 ng/mL/h (reference range, upright: 0.5–4.0 ng/mL/h). Her PCA: PRA ratio is 70. With a high PAC and a suppressed renin activity, primary hyperaldosteronism should be considered. (Table 15.3) Typical cut-offs to consider this entity are a PAC > 15 ng/dL and a PAC: PRA ratio of > 20. The latter ratio is also called an aldosterone-renin ratio (ARR) [3].

It is important to note that many medications can interfere with the ARR (Fig. 15.1) Medications that impair renin release, like NSAIDs and β-blockers, may elevate the

Table 15.3 Evaluation of aldosterone/renin axis

Disorder	Aldosterone	Renin	Examples
Primary hyperaldosteronism*	High	Low	**Aldosterone producing adenoma** **Bilateral idiopathic hyperaldosteronism** Familial hyperaldosteronism
Secondary hyperaldosteronism	High	High	**Renovascular HTN** (atherosclerotic RAS, fibromuscular dysplasia) Renin secreting tumor
Pseudohyperaldosteronism[a]	Normal	Normal	**Cushing's syndrome** (pituitary Cushing's, adrenal overproduction, ectopic ACTH secretion)
	Low	Low	**Exogenous mineralocorticoids** Liddle syndrome Glycyrrhizic acid (found in black English licorice) Apparent mineralocorticoid excess syndrome

Commonly encountered diagnoses are bolded
RAS renal artery stenosis, *ACTH* adrenocorticotropic hormone
*PAC/PRA must be greater than 20 and PAC must be higher than 15 ng/dL to diagnose primary hyperaldosteronism (*PAC* plasma aldosterone concentration, *PRA* plasma renin activity)
[a]Pseudohyperaldoseronism can be characterized by normal aldosterone/normal renin or low aldosterone/low renin conditions

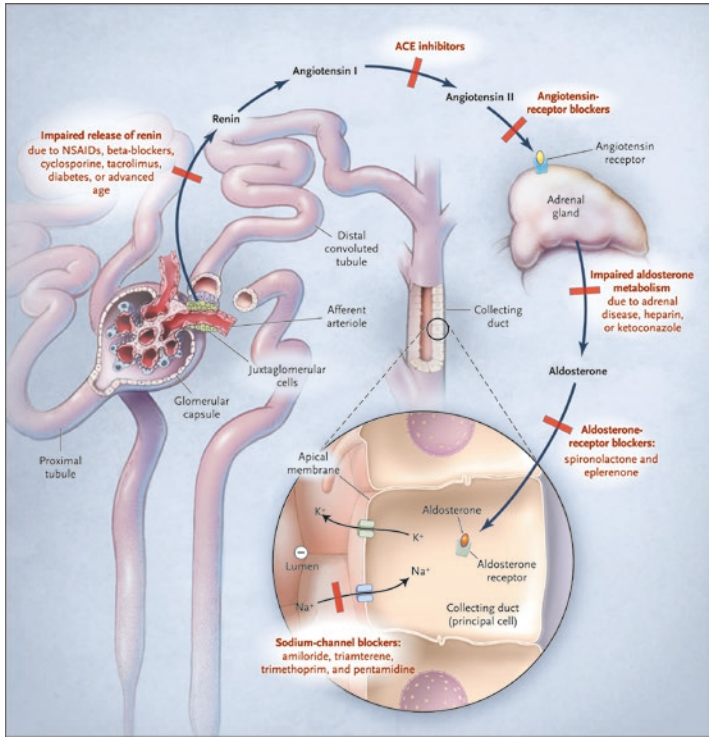


Figure 15.1 Medications acting on Renin-Angiotensin-Aldosterone System. This figure illustrates the normal Renin-Angiotensin-Aldosterone System (RAAS). The red rectangles show locations where drugs/medications/diseases/conditions can disrupt the pathway. The first step in the RAAS is the release of renin from the juxtaglomerular cells in the afferent arteriole in response to low blood volume (e.g., true volume depletion) or low effective arterial blood volume (e.g., congestive heart failure). Renin then acts as a proteolytic enzyme to convert angiotensinogen to angiotensin I. Under the influence of ACE, angiotensin I is converted to angiotensin II. Angiotensin II has a myriad of effects, including enhancing aldosterone release form the adrenal gland. The round circle highlights the site of action of aldosterone in the principal cell of the collecting duct, where it acts to reabsorb sodium and secrete potassium. In addition to low blood volume/low effective arterial blood volume, aldosterone release can also be stimulated by high plasma potassium levels. (From Ref. [4], Copyright @ 2004 Massachusetts Medical Society. Reprinted with permission from Massachusetts Medical Society)

ARR. Diabetes or advanced age have a similar mechanism. Medications that may lower the ARR include ACE-inhibitors (by blocking the conversion of angiotensin I to angiotensin II) and ARBs (by blocking the action of angiotensin II at its receptor). In addition, other drugs can lower the ARR by impairing aldosterone metabolism (eg, heparin) or by blocking the aldosterone receptor (eg, spironolactone). Interpretation of the ARR must take this into account.

In patients in whom there is a high clinical concern for hyperaldosteronism but lab criteria are not met (i.e., PAC is not >15 and PAC:PRA is not >20), a salt loading test (saline suppression test) can be done. Typically this involves infusion of 2L of normal saline (NS) intravenously over 4 hours. The PAC and PRA are then repeated; an adequate test to interpret results would require a urine Na > 200 mEq. If the patient has hyperaldosteronism, administration of NS would not suppress the PAC and PRA. (See box for specifics of this testing).

Saline Suppression Test (Salt Loading to confirm suspicion of hyperaldosteronism)

This can be achieved by either infusion of normal saline (NS), generally 2L over 4 hours, or with oral salt loading. Patients are instructed to add 1 flat teaspoon of salt to their daily food intake and to consume salty foods like potato chips, pretzels and pickles for 72 hours. Urine is collected after 3 days to ensure urine sodium > 200 mEq/24 h to ensure test validity and then a PRA and PAC are obtained. Many centers prefer the saline infusion as it is practically easier to administer and can ensure the validity without a 24 hour urine collection; however, this test is more expensive. A test is thought to be positive if the aldosterone remains unsuppressed (i.e., remains elevated) and the renin continues to be suppressed (i.e., remains low) [3].

Returning to case 3, as this patient had biochemical evidence for primary hyperaldosteronism, cross-sectional imaging with a CT scan or MRI is indicated to help elucidate whether an adrenal adenoma is present. Adrenal vein sampling can also be considered in the right clinical context but is typically recommended by and performed by specialists.

Case 4: Renal Tubular Acidosis

A 60 year old male with type 2 diabetes mellitus on lisinopril 20 mg daily with CKD stage 3 (with baseline creatinine 1.7 mg/dL) presents to his physician with labs significant for sodium 136 mEq/L (normal 135–145 meEq/L), potassium 5.2 mEq/L (normal 3.5–5 mEq/L), chloride 108 mEq/L (normal 97–107 mEq/L), bicarbonate 18 mEq/L (normal 22–26 mEq/L) and normal albumin. What further evaluation would you pursue to understand the etiology of his metabolic acidosis?

Abnormal serum laboratory values can often lead to questions of what testing may be needed next. If labs differ dramatically from prior values, early recheck of the lab is most helpful to ensure that the lab value is consistently abnormal and not due to a processing error. *For case 4, we first would investigate whether the acidosis is a high anion gap metabolic acidosis (HAGMA) or a normal anion gap metabolic acidosis (NAGMA).*

The anion gap is determined by the following equation:

$$\text{Serum Anion Gap} = \left[\text{Na}^+\right] - \left[\text{Cl}^-\right] - \left[\text{HCO}_{3^-}\right]$$

which in this patient is calculated to be 10 (using case values, 136 − 108 − 18 = 10). The normal anion gap (AG) is 10–12 and thus this person has a NAGMA.

NAGMA causes can be recalled using the ACCRUED mnemonic. A NAGMA mainly occurs through (1) loss of bicarbonate in the GI tract (2) impairment of acid excretion

or excessive loss of bicarbonate in the kidneys, or (3) either acid gain or dilutional acidosis due to rapid administration of high volumes of sodium chloride (Table 15.4). A Medication Administration Record (MAR) typically assists with the latter, however if the history does not provide obvious delineation between the first two, then a urine anion gap can be helpful.

$$\text{Urine Anion Gap} = \left[Na^+ \right] + \left[K^+ \right] - \left[Cl^- \right]$$

The urine anion gap is essentially an "ammonium detector" for the urine [7]. Ammonium cannot be directly measured in the urine by clinical labs, but its presence can be inferred by checking a urine anion gap. On a typical Western diet, we generate about 100 mEq of nonvolatile acid each day (acids other than carbon dioxide) that we must excrete to stay in acid-base balance. Ammonia (NH_3) is produced and secreted

Table 15.4 Causes of non-anion gap metabolic acidosis using the ACCRUED mnemonic (number after etiology refers to pathophysiologic cause of NAGMA described in text)

Mnemonic	Etiology	Examples
A	Acid infusion (3)	Hyperalimentation
C	Chronic kidney disease (2)	Diabetic nephropathy
C	Carbonic anhydrase inhibitor (2)	Acetazolamide
R	Renal tubular acidosis (2)	Type IV RTA
U	Ureteral diversion (1)	Ileal conduit
E	Expansion/Extra chloride (3)	High/rapid volume sodium chloride infusion
D	Diarrhea (1)	Viral gastroenteritis

by the proximal tubule and serves as one of the principal urinary buffers. Ammonia (uncharged) traps the hydronium ion (H^+) excreted by the kidney in the urine as charged ammonium (NH_4^+). Urine NH_4^+ and urine chloride (Cl^-) bond to form a soluble salt ($NH_4^+Cl^-$) that is excreted in the urine.

In cases of a NAGMA resulting from loss of bicarbonate from the GI tract, the urine anion gap will be negative. In these instances, the kidneys will function normally and try to compensate for the NAGMA by excreting more acid. This attempt at NAGMA compensation will result in more $NH_4^+Cl^-$ in the urine resulting in a higher chloride measurement. Looking at the urine anion gap equation, the higher than normal urine chloride measurement leads to a negative urine anion gap. Simply stated, the kidney is working, and the problem is the GI tract when the urine anion gap is "neGUTive."

In cases of a NAGMA resulting from either renal impairment of acid excretion or excessive loss of bicarbonate, the urine anion gap will be positive (generally > 20). In these instances, the kidneys are not functioning appropriately and are likely the culprit of the metabolic acidosis. This is likely the result of a renal tubular acidosis (RTA).

There are some caveats to the urine anion gap that should be noted. To avoid confusion, use of the urine anion gap calculation should only be used in the setting of a NAGMA. Normal patients without a NAGMA have a positive urine anion gap due to normal physiologic excretion of $NH_4^+Cl^-$ and should not mistakenly be considered to have an RTA. The interpretation of the urine anion gap may also be difficult in states of volume depletion, in proximal RTA (type II RTA) or in the presence of excess negative charges (e.g., beta-hydroxybutyrate).

Returning to case 4, the following urine tests were obtained: Urine sodium 110 mEq/L, urine potassium, 13 mEq/L, urine chloride: 95 mEq/L. Urine anion gap was calculated to be 28 (using case values 110 + 13 – 95 = 28), which in the face of a NAGMA is consistent with an RTA.

RTA occurs as a result of the failure of the kidney to reabsorb all the filtered bicarbonate, failure to synthesize new bicarbonate to keep up with daily metabolic demands (protein metabolism from our dietary daily acid load consumes bicarbonate and must be replaced) or failure to excrete acid. Table 15.5 lists the 3 general types of RTA, types I, II and IV. Two features to differentiate the RTAs are the serum potassium and urine pH. If serum potassium is elevated, the patient most likely has a type IV RTA. If normal or low, the patient may have a type I or II RTA. If the urine pH >5.5, the RTA is a distal or type 1 RTA. The urine pH in proximal or type 2 RTA can be variable depending on the severity of the acidosis. In mild cases, urine pH will be relatively alkaline (urine pH >5.5), but in severe cases will be acidic (urine pH <5.5).

Type I or distal RTA arises from the inability to secrete H^+ ions into the urine in the distal nephron for two possible reasons. First, H^+ ion channel disruption can occur in autoimmune conditions (e.g., Sjogren's syndrome or SLE) or genetic conditions (e.g., Liddle syndrome). Alternatively, membrane permeability may be altered due to medications like amphotericin B, topiramate, or lithium [5, 6]. Patients with type I RTA may have a propensity to form calcium phosphate stones or develop nephrocalcinosis. Type 1 or distal RTAs often require nephrology consultation.

Type II or proximal RTA is the result of inability to reabsorb bicarbonate in the proximal tubule, resulting in wasting of bicarbonate. The differential of type II RTA includes Fanconi syndrome, which is a generalized proximal tubular dysfunction resulting in urine loss of glucose, bicarbonate and phosphates, amino acids, uric acid and potassium. This may be caused by both genetic mutations and medications such as tenofovir or ifosfamide [7], but also may be associated with clinical entities like amyloidosis and multiple myeloma [5]. A nephrology consult may be necessary in these patients for further testing to delineate these conditions. Lastly, practitioners should be aware of patients presenting with osteomalacia who might need consideration for an underlying type

Table 15.5 Renal tubular acidosis (RTA) types, lab abnormalities, causes and clinical features

RTA type	Location of disorder	Dysfunction	Serum K/ HCO3 (mEq/L)	Urine pH	Differential diagnoses	Other clinical features
I	Distal tubule	Low H^+ secretion	Low/<12	>5.5	Autoimmune (Sjogren's syndrome, SLE), Drugs (amphotericin B, lithium, topiramate)	Calcium phosphate stones, nephrocalcinosis, hypocitraturia
II	Proximal tubule	Low HCO_3 resorption	Low/14–18	>5.5, but may be <5.5 in severe cases	Fanconi Syndrome, Mutiple myeloma, Amyloidosis, Drugs (carbonic anhydrase inhibitors, tenofovir, topiramate, ifosfamide), Familial/Hereditary, Heavy metal poisoning	Osteomalacia, rickets
IV	Collecting duct	Low aldosterone activity or level	High/~18	Usually <5.5	Diabetes, Urinary obstruction, Drugs (ACE-i, NSAIDs, cyclosporine)	Hyperkalemia

Abbreviations: *HCO3* bicarbonate, *ACE-i* angiotensin converting enzyme inhibitor, *NSAIDs* nonsteroidal anti-inflammatory drugs, *SLE* systemic lupus erythematosus
Normal serum HCO_3 = 23–25 mEq/L; Normal serum K = 3.5–5 mEq/L

II RTA. Treatment of RTA due to a medication effect may involve discontinuation of the contributing medication, but this often requires multi-disciplinary conversation to assess risk-benefit to the patient.

Type IV RTA is the most common RTA providers will encounter. Aldosterone may be less effective or deficient resulting in poor excretion of potassium and retention of hydronium ion in the collecting duct, resulting in a NAGMA with hyperkalemia [8]. The common causes of type IV RTA are urinary obstruction, chronic kidney disease, medications like ACE-i, and diabetes mellitus (even with mild nephropathy).

Returning to case 4, the presence of hyperkalemia and an RTA in the context of diabetes should prompt the clinician to suspect type IV RTA. The clinical history of chronic kidney disease and exposure to ACE-i would be consistent with this as well.

Treatment of Type 4 RTA

A decision to treat the metabolic derangements in type IV RTA depends on the severity of the metabolic acidosis and the degree of the hyperkalemia. To better address the hyperkalemia, it is important to understand the regulation of potassium excretion in the kidney and the role of the Renin-Angiotensin-Aldosterone System (RAAS) (Fig. 15.1). While potassium is freely filtered at the glomerulus, about 90% undergoes reabsorption before reaching the distal nephron. The handling of potassium in the kidney is very unique in that potassium can be secreted into the distal nephron, depending on physiological needs. Two important regulators of this process are aldosterone and the amount of sodium delivered to the distal area of the nephron. In the principal cell of the collecting duct, under the stimulation of aldosterone, sodium is reabsorbed through a sodium channel on the urine, or lumen, side of the membrane. This creates more electronegativity

in the lumen (due to sodium travelling as a positive charge into the cell) and creates a favorable gradient for potassium to be secreted from inside the cell into the urine through a potassium channel. How much potassium excreted is also dependent on distal sodium delivery to that area of the nephron. Thus in situations of low sodium delivery, as in volume depletion, potentially less potassium will be secreted.

There are many drugs that can interfere with potassium excretion by disrupting any of the pathways along the RAAS (see red rectangular markings in Fig. 15.1). Examples include: NSAIDs can impair renin release; ACE-i and ARB can impair production of angiotensin II or the effect of angiotensin II at its receptor (respectively); heparin can impair aldosterone biosynthesis/metabolism in the adrenal gland; spironolactone can block the aldosterone receptor in the principal cell; trimethoprim can block the sodium channel on the lumen side of the principal cell. Patients who may be particularly prone to developing hyperkalemia associated with these drugs include those with underlying chronic kidney disease, diabetes mellitus, advanced age or states of true or effective circulating volume depletion. Further risk is imposed by diets high in potassium or use of salt substitutes that contain potassium. Thus a careful assessment of diet, volume status and medications that may impair urinary K secretion is necessary in evaluating hyperkalemia.

In a patient with diabetes mellitus and type IV RTA with proteinuria, it can get even more complicated. The goal is to reduce proteinuria to slow progression of renal disease and perhaps reduce cardiovascular risk. ACE-i or ARBs are common anti-proteinuric therapies, however their use may be limited by hyperkalemia or their implementation impaired by pre-existing hyperkalemia (see Chap. 5). As illustrated in Fig 15.1, it is important to discontinue all unnecessary agents like NSAIDs or trimethoprim that can cause hyperkalemia. Initiation of anti-proteinuric therapies may first require additional strategies to lower serum K level, focusing on potassium balance: decreasing intake of potassium, facilitating transcellular shifts of potassium or enhancing urinary potassium excretion.

Physiologic Focus	Strategy to facilitate lowering K
Diet	The patient can meet with a nutritionist to learn how to create a low potassium, low carbohydrate diet
Transcellular K shift	Excellent *control of diabe*tes Supplemental bicarbonate may help facilitate shift of K intracellularly and may help correct acidosis (chronic acidosis may be associated with progression of kidney disease, muscle wasting and osteoporosis)
Facilitate renal K excretion	Use of a thiazide or loop diuretic may be helpful to facilitate K excretion if the patient is hypertensive or fluid overloaded

With any of the above changes, it is important to recheck potassium within 2 weeks. Persistent elevations in K despite these changes may require expert consultation with a nephrologist.

Returning to case 4, he was started on furosemide, as he was also mildly hypervolemic. The furosemide helped to decrease his serum K to 4.9 mEq/L and he was able to continue his lisinopril. On discharge he was also started on sodium bicarbonate tablets with a nephrology referral to follow up on his type IV RTA, hyperkalemia, and chronic kidney disease.

Case 5: Evaluation of Polyuria

A 40 year old woman has been in the ICU for the past week following a trauma. Her urine output for the last several days has been 6 liters per day. Her serum sodium is 150 mEq/L (normal 135–145 mEq/L). What is the next step in diagnosis/management?

The case highlights the challenge of determining the cause of significant polyuria. Most define polyuria as >3 L of urine per day. Polyuria that is not addressed can lead to multiple medical problems. These include severe electrolyte derange-

ments and hypovolemia, and if the patient is unable to keep up with the water losses, severe dehydration can occur.

One must first determine whether polyuria is a water or solute diuresis, which can be evaluated with urine osmolality obtained on random urine specimen (Fig. 15.2). If the urine osmolality is <100 mOsm/L, then the polyuria is due to a water diuresis. If the urine osmolality is greater than 300 mOsm/L, then the polyuria is due to a solute diuresis.

History is crucial in determining the cause of the water diuresis. If the patient endorses compulsive water drinking, this may be consistent with primary polydipsia. In these individuals the polyuria is the normal response to habitually high water intakes. A water diuresis may also be due to diabetes inspidius (DI), which may be central or nephrogenic. [9]. Central DI is considered following intracranial insult (e.g. hemorrhage, trauma, or procedures) or primary pituitary deficiency (especially granulomatous disease such as sarcoidosis or tuberculosis). Nephrogenic DI can be considered in the context of chronic diuretic use, lithium therapy, or concomitant hypercalcemia. The polyuria in diabetes insipidus is inappropriate, thus the patient urinates alot and as a consequence then drinks alot.

In contrast to water diuresis, typified by a low urine osmolality, a urine osmolality >300 mOsm/kg suggests the diuresis is a solute diuresis. The solutes may be electrolytes (e.g. sodium) or nonionic compounds (e.g., glucose, urea, mannitol). The differential includes:

1. **high solute load**
   - large volume saline infusions
   - mannitol
2. **urea, particularly BUN >100**
   - renal failure
   - parenteral nutrition with protein content >100 g per day
3. **glucose (diabetes or glucose infusions)**
   - excessive serum glucose
   - use of sodium glucose co-transporter 2 inhibitors (SGLT2 inhibitors), the "-flozin" medications

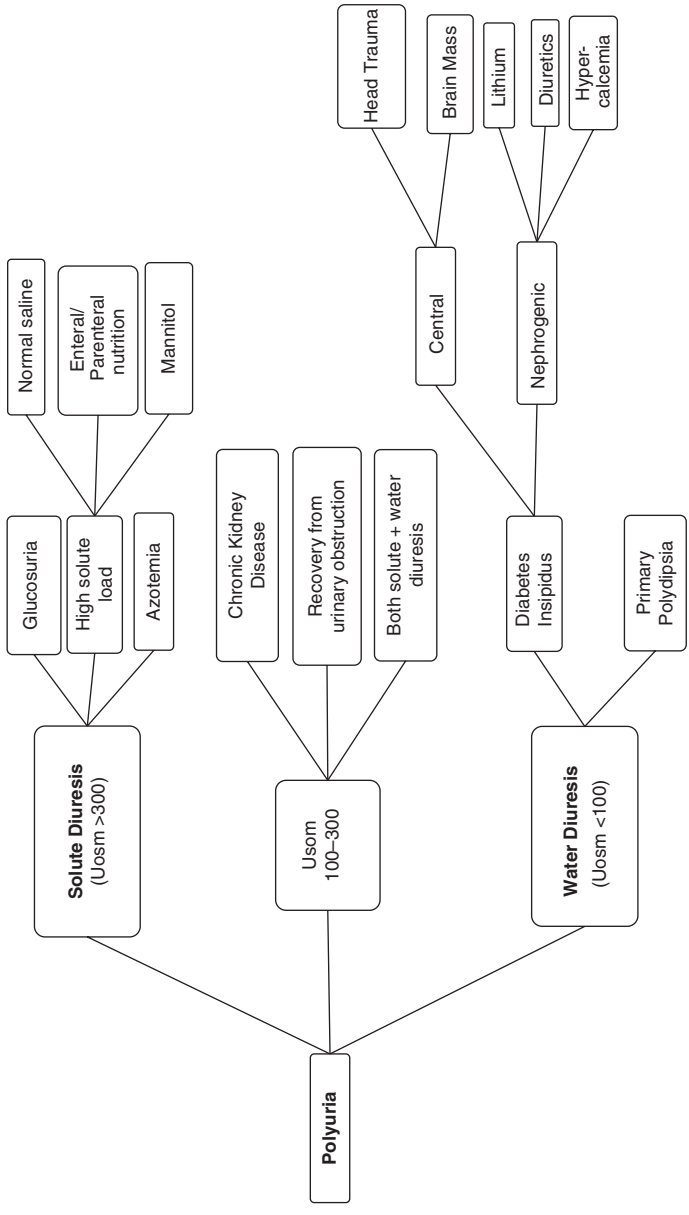


FIGURE 15.2 Polyuria: Solute versus water diuresis based on urine osmolality and appropriate differential diagnosis

Review of the MAR may help identify excess solutes or medications causing glucosuria. Routine labs (like blood glucose or blood urea nitrogen) are helpful at elucidating hyperglycemia and uremia, respectively. The most common causes of solute diuresis clinically are large volume saline infusions and hyperglycemia.

If the cause of the solute diuresis is still unclear or there are multiple causes, it is possible to determine the exact solute causing the solute diuresis with a 24-hour urine collection of electrolytes (sodium, potassium) and nonionic compounds (glucose, urea nitrogen).

$$\text{Urine Osmolality} = 2\left[U_{Na} + U_{K}\right] + \left(\text{UUN} / 2.8\right) + \left(\text{Urine glucose} / 18\right)$$

UUN = urine urea nitrogen; U_K *= urine K;* U_{Na} *= urine Na*

A difference between calculated and measured osmolality suggests the presence of a nonionic agent like mannitol [9].

Polyuria with urine osmolality between 100–300 mOsm/kg is most commonly seen in patients with CKD isosthenuria, which is the inability to concentrate urine (see Chap. 7). It may also occur in situations of simultaneous excess water and solute intake, a partial DI or in patients recovering from urinary obstruction.

Returning to case 5, the patient's urine osmolality was elevated to >600 mOsm/kg. Review of recent medications revealed no new medications, however review of her intake/output (I/O) documentation showed that she had 5 L of normal saline per day over the past few days and she was euvolemic. As this appeared to be an appropriate solute diuresis, the team discontinued her saline and her urine output decreased to 2 L per day and her serum sodium normalized.

Case 6: Polyuria due to Water Diuresis

A 60 year old man is admitted after a head trauma to neurosurgical service and has been quite thirsty. He has been urinating 6L/day with stable serum sodium of 144 mEq/L (normal 135–145 mEq/L). His urine osmolality is measured and is <100 mOsm/kg. He is unsure of his medications. What is the next step in diagnosis and treatment?

Case 6 highlights a water diuresis. If urine osmolality is not available, a low urine specific gravity is a useful surrogate (see Chap. 7). The first line test, typical for any medical patient, is a clear history and physical exam; in this patient, given his intra-cranial trauma, this likely represents central DI but we cannot exclude nephrogenic DI. If the clinical context is unclear, one can consider a **water deprivation test** (pattern of results seen in Fig. 15.3a), which assesses the patient's ability to concentrate the urine when fluids are withheld [6]. Under normal circumstances, in response to dehydration, antidiuretic hormone (ADH) is secreted, resulting in conservation of water by the kidney. The result is the production of a small amount of concentrated urine, with a urine osmolality of >800 mOsm/kg. Similarly, patients with mild primary polydipsia will be able to concentrate their urine when deprived of fluids. An inability to maximally concentrate the urine with dehydration alone will be observed in patients who lack ADH (central DI), lack ADH responsiveness (nephrogenic DI) or have severe primary polydipsia. These last groups of patients can be administered a desmopressin challenge to further characterize and distinguish the etiology of their polyuria (Fig. 15.3b).

The basic principle of the water deprivation test involves baseline data including patient body weight, serum sodium concentration, plasma and urine osmolalities and ADH level. Then careful measurements of body weight, urine volume, and urine and plasma osmolalities every 1-2 hours are performed in the clinic setting under a state of water deprivation. It is crucial to monitor patients very closely

during the testing period as they can become profoundly volume depleted.

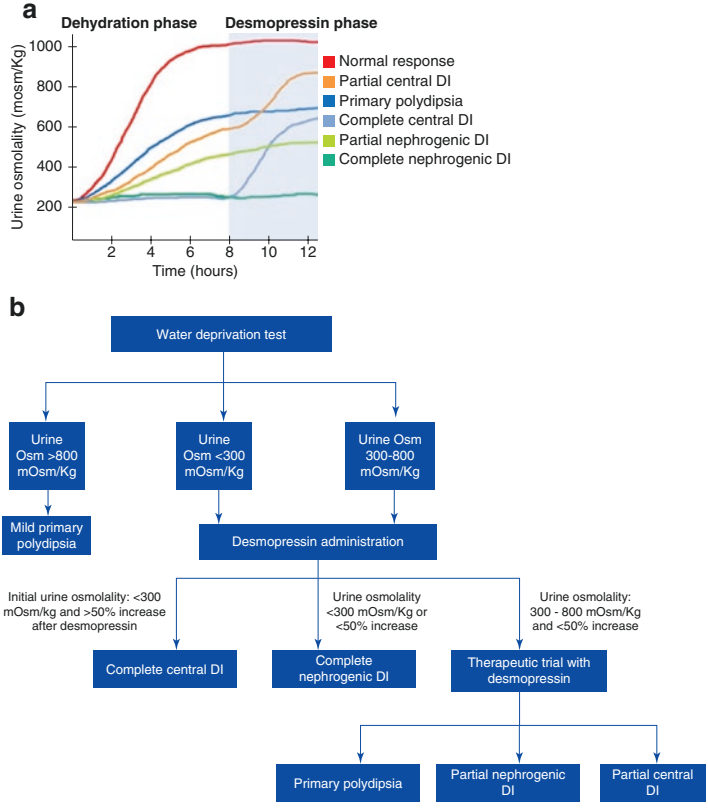
The test is continued until one of several endpoints is reached:

1. the patient has lost 3% of body weight.
2. the patient can achieve a urine osmolality of >600 mOsm/kg with water deprivation alone, indicating that both ADH release and effect are intact.
3. the plasma osmolality exceeds 295–300 mOsm/kg or the sodium concentration is at or exceeds 145 mEq/L with water deprivation alone, indicating that both ADH release and effect are intact.

If the plasma osmolality reaches >300 mOsm/kg and the urine osmolality remains <600 mOsm/kg, or if the urine osmolality on 2–3 successive hourly measurements is stable despite a rise in plasma osmolality, then desmopressin (DDAVP) is administered. A patient with central DI will have a consistently low urine osmolality (<300 mOsm/kg) with water deprivation, but will increase to >600 mOsm/kg after DDAVP administration, since they are responsive to ADH but simply lack the hormone (orange and light blue lines, Fig. 15.3a). It is classified as partial or central based on how much concentration occurs in the dehydration phase. Central DI is classically seen in association with a hypothalamic/pituitary insult (after pituitary surgery or trauma for example). A patient with nephrogenic DI will exhibit low urine osmolality with water deprivation (<300 mOsm/kg) with only partial response to ADH since ADH is secreted normally, but the kidney is less responsive to ADH in this condition (light and dark green lines, Fig. 15.3a). Nephrogenic DI is again classified as partial or complete based on the ability to concentrate urine in the dehydration phase of the test. A classic cause of nephrogenic DI is chronic lithium therapy. A patient with primary polydipsia (dark blue line, Fig. 15.3a) should be able to concentrate their urine to >600 mOsm/kg with the water deprivation part of the test alone [6]. Primary

polydipsia classically is associated with certain mental health conditions, like schizophrenia.

Returning to case 6, he was found to have a urine osmolality of 80 mOsm/kg. When deprived of water, his urine osmolality increased to 90 mOsm/kg. When serum sodium was 146 mEq/L (>145 mEq/L) and plasma osmolality was 305 mOsm/kg (>295 mOsm/kg) DDAVP was administered. After DDAVP was given, his urine osmolality increased to 650 mOsm/kg and the urine output decreased. This is consistent with complete central DI as the cause; the etiology was felt to be secondary to his



brain trauma. For treatment, access to free water was restored and desmopressin 100 mcg twice daily was administered. With this treatment his serum sodium concentration improved to 138 mEq/L and his urine output decreased to 2–3 L per day. In the setting of intracranial trauma, the central DI may be transient, so close monitoring of patient in the outpatient setting is required to prevent hyponatremia.

Fractional Excretion of Sodium and Urea "Do's and Don'ts"

Under normal circumstances, of the approximate 25,000 mEq of sodium the kidney filters daily, less than 1% is excreted in the urine. The kidney has a remarkable ability to reabsorb almost all the filtered sodium daily through the actions of sodium transport in the kidney tubules (see Chap. 1). When

FIGURE 15.3 (**a**) Graphical representations of the water deprivation test for diabetes insipidus [6]. (**b**) Algorithm for Diagnosis of the Main Types of Polyuria Using Results from Water Deprivation Testing and Desmopressin Administration. [Modified with permission from from 6]. The normal response to dehydration is a rise in the urine osmolality to > 800 mOsm/kg (red line). In patients with primary polydipsia, with water deprivation the urine will concentrate to ranges between 300 and 800 mOsm/kg depending on the severity of the problem (in milder forms will be able to concentrate the urine more); there is only a small further increment in urine osmolality (dark blue line) after desmopressin administration. In patients with complete DI (central or nephrogenic), during the dehydration phase the urine osmolality stays < 300 mOsm/kg with a concomitant rise in plasma osm to > 300; patients with complete central DI will have >50% increase in urine osm after desmopressin administration (light blue line), while patients with complete nephrogenic DI will have a <50% increase due to insensitivity to the hormone (dark green line). In patients with partial central (orange line) or nephrogenic DI (light green line), the urine osmoality after water deprivation increases to usually 300–600 mOsm/kg, with <50% increase in urine osmolality after desmopressin administration.

the kidney tubules are damaged or injured, more sodium will appear in the urine. The test that calculates how much of the filtered sodium appears in the urine is called the Fractional Excretion of Sodium (FeNa). The FeNa is often used clinically to differentiate pre-renal acute kidney injury (AKI) from acute tubular necrosis (ATN).

The FeNa is calculated by the following equation:

$$\text{FeNa}: \left(U_{Na} \times P_{cr}\right) / \left(P_{Na} \times U_{cr}\right) \times 100$$

P = plasma, U = urine, Na = sodium, cr = creatinine, Fe = fractional excretion

The values are obtained from a simultaneous lab draw for plasma sodium and creatinine and a random urine sample for urine sodium and creatinine. The general principle is that a FeNa <1% indicates prerenal causes and a FeNa >2% is suggestive of ATN. A value in between may be suggestive of a transition from pre-renal to ATN [10].

It is important to know the limitations of this equation:

1. **Do not use in nonoliguric patients.** It can only be used in patients who are oliguric (i.e., urine output <400–500 ml daily). The sensitivity and specificity change markedly if, for example, the patient is making 2 L urine daily and should therefore not be used in nonoliguric patients.
2. **Do not use in patients with AKI superimposed on CKD.** The values in patients with underlying CKD can be misleading. The remaining nephrons need to manage the daily solute load with less functioning nephrons and thus the values may be higher and may not be interpretable in CKD.
3. **Do not use in proximity to diuretics.** If a diuretic is given just prior to sampling, error may occur as loop diuretics block sodium reabsorption and more sodium will appear in the urine, i.e., the FeNa may be high. The sensitivity and specificity change markedly when using this as a tool in patients with AKI who have received diuretics [11].

Some conditions are known to be associated with a low FeNa that are not pre-renal in nature. These include early contrast nephrotoxicity (due ot vasoconstriction) and acute glomerulonephritis (due to sodium avidity). In these conditions the FeNa is poorly reflective of the actual cause of the AKI. Similarly, the FeNa should not be checked after large amounts of normal saline are administered. For any clinical condition, however, a thorough history and physical exam and putting the AKI in the clinical context is the most helpful. Tools such as the FeNa are used to help support the clinical suspicion but should never replace it and should be interpreted in that light.

An alternative equation is the fractional excretion of urea (FeUrea), performed the same way but just with urea checked in substitution for the sodium. The values for the equation are <35 for identifying pre-renal and >50 for ATN.

$$\text{FeUrea}: \left(U_{\text{Urea}} \times P_{\text{cr}}\right) / \left(P_{\text{cr}} \times U_{\text{Urea}}\right) \times 100$$

P = plasma, Fe = fractional excretion, cr = creatinine

Summary

Overall, urine electrolytes are powerful diagnostic tests when used in the correct setting to help taper differentials. They can help further characterize incidental lab findings, such as a urine potassium in a patient with hypokalemia, but more importantly they can help guide treatment, as is the case for obtaining urine osmolality in the evaluation of acute polyuria. While these tests are not always at the front of our minds, they are overall fairly inexpensive ways to limit the financial and emotional burden of further testing and in some cases help our patients' quality of life.

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Chapter 16
Other Common Uses for Urine Screening in Clinical Practice: Substance Use Disorders, Antipsychotic Adherence, Sexually Transmitted Infections

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Objectives

- Describe the usefulness of urine drug screening in clinical practice as well as employment physicals
- Discuss the causes and potential ramifications of false positive and false negative urine drug screen results

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- Describe clinical settings to consider a urine screen specific for diuretic abuse
- Discuss the potential utility of urine screening for antipsychotic compliance in clinical practice
- Define urine screening recommendations for the two most common sexually transmitted infections: *Neisseria gonorrhoeae* and *Chlamydia trachomatis*

Overview

Urine testing is routinely used for screening purposes in clinical practice. Indications for urine screening previously covered in this book are to assist in the diagnosis of various nephrologic and/or urologic diseases. Three other common uses include urine tests to screen for substance use disorders, to confirm antipsychotic medication adherence and to identify sexually transmitted infections in at risk populations.

Case 1: Urine Screening for Substance Use Disorders - Urine Drug Screen

A 30 year old male is a new hire at a local factory. He is required to perform a urine drug screen as part of the hiring process. You are the newest partner hired at the medical clinic responsible for performing the new hire physicals and overseeing the urine drug screen process. He inquires, "I have heard this drug test isn't always accurate. Is that true?"

Drug testing can be completed on various biological specimens other than urine, including blood, hair, saliva, sweat, nails and even meconium; however, urine is the most commonly obtained specimen due to its noninvasive route and ease of sample collection. Steps should be taken to reduce potential tampering of urine specimens collected for drug screening, including direct observation when indicated as common chemicals have been used for adulteration of the

sample to avoid drug detection. If the direct observation method of collection is not used, blue dye should be placed in the toilet prior to the test to ensure water from the toilet is not submitted as the sample. Collection of at least 30 mL of urine in a sealed tamper-resistant container is the standard. The temperature and pH of the urine sample collected should be measured and be between 90–100°F and have a pH in the 4.5–8.5 range to confirm a fresh sample has been obtained. Dilution of the urine through excessive water consumption, diuretics, or direct addition of water to the urine sample can decrease the urine drug concentration and make a negative result more likely; therefore excessively dilute samples (SG ≤1.001) should be rejected [1].

Initial urine drug screens are performed using immunoassay technology. Immunoassays use antibodies to detect the presence of drug metabolites or classes of drug metabolites in the urine. These screens have high (>90%) sensitivity and allow for a large number of specimens to be analyzed quickly. Unfortunately, immunoassays can detect substances with similar characteristics leading to false-positive results (Table 16.1) [2].

Table 16.1 Drugs that may cause false positive results in immunoassay urine drug screen testing

Test drug or drug category	Drugs that may cause false positive results
Cocaine	Topical anesthetics containing cocaine, coca tea leaf products
Amphetamine	Amantadine, aripiprazole, benzphetamine, brompheniramine, bupropion, chlorpromazine, desipramine, ephedrine nasal inhaler, fluoxetine, labetalol, metformin, methylphenidate, phentermine, phenylephrine, phenylpropanolamine, promethazine, pseudoephedrine, ranitidine, ritodrine, selegiline, thioridazine, trazodone, trimethobenzamide, trimipramine

(continued)

Table 16.1 (continued)

Test drug or drug category	Drugs that may cause false positive results
Phencyclidine (PCP)	Ciprofloxacin, dextromethorphan, diphenhydramine, doxylamine, gatifloxacin, ibuprofen, imipramine, ketamine, lamotrigine, levofloxacin, meperidine, mesoridazine, naproxen, ofloxacin, thioridazine, tramadol, venlafaxine
Tetrahydrocannabinol (THC)	Dronabinol, efavirenz, hemp oils, ibuprofen, naproxen, sulindac, pantoprazole
Opioid	Ciprofloxacin, dextromethorphan, diphenhydramine, doxylamine, gatifloxacin, levofloxacin, ofloxacin, poppy seeds, quetiapine, quinine, rifampin
Barbiturate	Ibuprofen, naproxen
Benzodiazepine	Oxaprozin, sertraline
Lysergic acid diethylamide (LSD)	Amitriptyline, bupropion, diltiazem, fluoxetine, labetalol, methylphenidate, sertraline, trazodone
Methadone	Chlorpromazine, clomipramine, diphenhydramine, quetiapine, thioridazine, verapamil
Tricyclic antidepressant (TCA)	Carbamazepine, cyclobenzaprine, cyproheptadine, diphenhydramine, hydroxyzine, quetiapine

Gas chromatography/mass spectrometry (GC-MS) or liquid chromatography/tandem mass spectrometry (LC-MS/MS) are considered the gold standard for confirming a positive result on immunoassay testing. This confirmatory method can identify specific molecular structures and distinguish individual drugs in a class, as well as quantify the amount of

a drug or substance present in the sample. These confirmatory tests must be performed by highly trained personnel as these tests are time consuming as well as costly. The confirmation test is usually reserved for positive initial drug screens and should always be conducted when legal, forensic or employment outcomes are in question [3].

The rationale for the urine drug screen as well as the intended use of the result should always be considered prior to ordering the test, as interpretation of drug screening results can be difficult. A true confirmed positive result indicates only that the person was exposed to the drug recently. This exposure could have been willingly or unknowingly and the positive test result may not reflect drug abuse. A positive test result can corroborate clinical suspicion but provides no information on whether the drug in question is contributing to the current clinical presentation [4]. In contrast, a negative drug screen does not rule out substance intoxication or a substance induced encephalopathy. A negative screen could be due to an ingested substance that is not part of the typical drug screen, such as cathinones (bath salts) [5].

The most commonly ordered drug screens are for cocaine metabolites, amphetamines, phencyclidine, marijuana metabolites and opiate metabolites [1, 2]. It is important to know the manufacturer specifications for the urine drug screen test that you are using as some routine opiate urine screening tests are designed to detect only morphine metabolites and would not detect other opioid class medications. An expanded panel would need to be ordered in order to detect other commonly used opioids including fentanyl, hydrocodone, methadone, meperidine, oxycodone, buprenorphine and tramadol [2]. The same is true for certain sedative-hypnotic drugs. The urine metabolite of clonazepam, 7-aminoclonazepam, is not detected by many benzodiazepine assays, nor are related sedative-hypnotics such as zolpidem. Alprazolam and lorazepam may also go undetected in some urine drug tests. Some manufacturers offer separate testing for these drugs so knowledge of the manufacturer specifications for the particular drug screen test is essential [6, 7].

It is important to remember that most drugs have a window of detection in the urine for 1–3 days; therefore, a negative drug screen result does not necessarily exclude occasional or even frequent drug use. On the other hand, some long-lived drugs and metabolites such as methadone and buprenorphine may be detected for up to 14 days after use. Another example of variable drug detection time is the delta-9-tetrahydrocannabinol (THC) component for marijuana users, as single use is detectable up to 3 days where heavy sustained use remains detectable for several weeks [1, 2, 8, 9].

Positive drug screen results and in particular false positive results can cause significant unintended harm. Examples of such unintended harm include ineligibility for being hired at a new job, suspension from work or loss of employment, unnecessary changes to pain medication regimens, disqualification from rehabilitation programs, potential criminal charges, social service investigations for pediatric patients who test positive, ineligibility for organ transplantation, and loss of trust from healthcare professionals. A large number of over-the-counter and prescribed medications also cross-react with drug detection assays (Table 16.1). The manufacturer specifications for the urine drug test may provide information on cross-reactivity thresholds. The interpretation of the result should always be taken in context with a complete list of the prescribed, over-the-counter, and herbal medications taken by the test subject [10–24].

Returning to case 1, you inform the man who presented for a new hire physical that urine drug screens are highly sensitive and sometimes have unintended false positive results. You reassure him, however, that if the initial urine immunoassay drug screen is positive, a separate confirmatory test will be performed before reporting any results to his new employer. You also ensure that you have a complete and accurate medication list for the man, including any over-the-counter medications he is taking so the urine drug screen results can be interpreted accurately.

Case 2: Urine Screening for Diuretic Abuse

A 17 year old female is brought to your clinic by her mother. The patient continues to complain of ongoing muscle cramps and weakness. She tells you that 2 days ago she fainted. You note she appears frail. She has excoriations on the posterior aspect of the second and third digits of her right hand. You order a basic metabolic profile which reveals severe hypokalemia. You suspect she suffers from bulimia nervosa but the mother is insistent her daughter would never subject herself to that behavior. The patient denies any substance abuse and even consents to testing. Her mother also provides consent for the daughter to be tested. What urine drug screen would be beneficial to confirm your suspected diagnosis in this patient?

Another class of medications with abuse potential is diuretics. Diuretics increase salt and water elimination from the body via the kidney. The target end result is weight loss from increased fluid removal through increased urination. This is the intended benefit in conditions such as heart failure; however, diuretics can sometimes be abused. Diuretics can be used as a source of purging in eating disorders for the desired weight loss effect as illustrated in the case above. Diuretics will also sometimes be abused by athletes in an attempt to flush out performance enhancing substances or to lose weight to make a certain weight class for an athletic event [25].

Consequences of diuretic use include symptoms of thirst and muscle cramps/weakness from sodium and potassium imbalance. Orthostatic hypotension and fainting may also result from diuretic use due to sodium imbalance. Diuretics can cause profound hypokalemia. Patients prescribed diuretics are often simultaneously prescribed oral potassium supplements to address the expected hypokalemia; however, in cases of abuse, these individuals often are not taking potassium supplements which results in severe hypokalemia. Severe hypokalemia can result in muscle paralysis as well as severe cardiac complications such as palpitations and arrhythmias and can even result in death.

A urine “diuretic screen” can be ordered as an in-house or send out lab depending on provider location. Several diuretics from both the loop and thiazide/thiazide-like diuretic classes

can be screened for in the urine. Diuretic identification is performed by High Performance Liquid Chromatography/Tandem Mass Spectrometry (HPLC-MS/MS) [26].

Returning to case 2, based on the clinical scenario, you have a high suspicion for diuretic abuse and order a diuretic drug screen. The result returns positive for furosemide. You discuss the positive result with the girl and her mother at a follow-up visit and she confirms she has been using furosemide for water weight loss. She tells you that she did not think it was a big deal as all it does is make you pee and lose weight. You discuss the harmful effects of diuretic abuse and refer her for counseling.

Case 3: Urine Screening for Antipsychotic Adherence

A 35 year old female is seen in your clinic for the first time to establish care with a new mental health provider. She takes chlorpromazine for schizophrenia. You have obtained her outside medical records and have reviewed them in preparation of the new patient visit. The records indicate suspicion of past non-compliance. Your interaction with the patient leads you to the conclusion that she is currently compliant with therapy. Is routine urine screening indicated going forward?

Partial adherence and nonadherence to antipsychotic medications are unfortunately common. It has been estimated that as high as 50% of patients are nonadherent to antipsychotics namely due to the side effects of these medications. Patient self-reporting and clinician estimates of compliance are often inaccurate [27, 28]. Laboratories now offer urine testing for antipsychotic medications allowing for a noninvasive means for testing adherence. While detection thresholds and actual drugs tested vary by company, testing is generally available for the majority of antipsychotic medications currently in use. Testing is usually performed on a random urine sample using liquid chromatography-mass spectrometry. An initial positive qualitative analysis screening is followed by confirmatory quantitative testing [29, 30].

Urine antipsychotic testing is not in universal use but it has found utility among some community psychiatrists. This testing

serves not only as a means of compliance monitoring in these practices but also as a catalyst for conversations about adherence with patients. In 2017, Cohen et al. [31] released guidelines on the use of urine antipsychotic drug testing to assess medication adherence. These guidelines are summarized in Table 16.2.

The impact and feasibility of testing were felt to be most appropriate for certain patient types during the initial evaluation. Patients who present for their initial evaluation with symptoms of a serious mental illness with no previously established diagnosis or patients with a known serious mental health illness who have risk factors for poor treatment adherence (elderly, homeless or have a co-occurring substance use disorder) should undergo urine monitoring. Repeat urine monitoring at a subsequent visit should be done to address any concerns from initial urinary monitoring, if clinical dete-

Table 16.2 Clinical recommendations for urine monitoring of antipsychotic medications

At the time of the initial patient mental health evaluation, conduct urine monitoring for antipsychotic medications in a patient with:
No previously established diagnosis
Conduct urine monitoring for an established diagnosis of serious mental illness in a patient who has one or more of the following risk factors:
Poor treatment adherence
Homelessness
Co-occuring substance use disorder
Elderly
During continuing mental health treatment, conduct repeat urine monitoring of antipsychotic medications:
If issues of concern regarding results of previous urine monitoring
If clinical deterioration or inadequate therapeutic response
If substantial change in social situation of the patient (a change in the level of care, living environment, health care provider, or pharmacy) that may require medication reconciliation
Periodically at set or random intervals depending on clinical indication but at least annually among patients with a prior normal test and no indications of deterioration or risk

rioration occurs or there is an inappropriate therapeutic response observed. Repeat monitoring should also be considered if a change in the social situation of the patient occurs such as change in the level of care, living environment, health care provider or pharmacy that may require medication reconciliation. Periodic urine monitoring at set or random intervals depending on clinical need should be performed with at least annual monitoring in patients with a prior normal test and no indications of deterioration or risk [31].

The guidelines also give recommendations in regard to urine monitoring method, patient education and patient feedback for urine antipsychotic testing. Urine collection should be conducted at the site where the medication is prescribed. Patients should receive either written or verbal education prior to urine monitoring testing on the importance of psychotropic medication adherence as well as the role and cost of urine monitoring. Patients should be provided access to urine results when they become available as well as verbal feedback from the provider within a clinically appropriate timeframe [31].

Care must always be taken in interpreting results of urine antipsychotic medication screening. Case reports have noted false positive test results in patients without a current prescription who previously took a medication 3 years prior to testing. This result was likely due to ongoing medication release from fat stores. The risk of false positive results increases for patients who are obese and/or have had a past long-acting antipsychotic injection. Open dialogue and transparency of test results are of utmost importance when clinical scenarios such as these occur [32].

Returning to case 3, you decide at this initial visit not to perform a urine screen for antipsychotic adherence. Her schizophrenia appears clinically to be under adequate control. The patient is well-groomed and dressed appropriately. She is not homeless and denies any substance abuse. You discuss the importance of medication compliance in the treatment of her mental illness and schedule a follow-up appointment for ongoing management.

Case 4: Urine Screening for *Neisseria gonorrhoeae* and *Chlamydia trachomatis*

An 18 year old female presents to your clinic to establish care. She is requesting oral contraceptive pills as she is currently sexually active in a new relationship. As part of your visit, you counsel her that while oral contraception can prevent pregnancy, it will not prevent sexually transmitted infections. She denies any current symptoms but inquires if there is any routine testing recommended for sexually active females her age besides invasive Pap testing and recurring blood draws.

Chlamydia and gonorrhea are the most commonly reported sexually transmitted infections in the United States and urine screening tests have become widely available. Most men and women with genital chlamydia do not experience symptoms, thus warranting routine and regular sexually transmitted infection (STI) screening. Gonorrhea is usually accompanied by urogenital discomfort and discharge. The advent of urine-based tests has increased the acceptance of STI screening among patients and providers since it allows for routine specimen collection without pelvic examination or a swab of the urethra [33].

Most screening efforts target women, as untreated chlamydia or gonorrhea can lead to complications such as pelvic inflammatory disease, ectopic pregnancy, infertility and chronic pelvic pain. The US Preventive Services Task Force (USPSTF) recommends screening for gonorrhea and chlamydia annually in sexually active women aged 24 years and younger and in older women who are at increased risk for infection. Increased risk factors for gonorrhea and chlamydia include the following: new or multiple sex partners, a sex partner with concurrent partners or a sex partner with an STI, inconsistent condom use among persons who are not in mutually monogamous relationships, previous or concurrent STI, and exchanging sex for money or drugs. Prevalence is also higher among incarcerated populations, military recruits and patients receiving care at public STI clinics. Screening for these STIs is effective because they are curable. Intrauterine

or perinatally transmitted STIs can have grave effects for pregnant women and their fetuses. Pregnant women under the age of 25 or with risk factors are routinely screened at their initial prenatal visit with third trimester screening recommended for at-risk patients [34].

Clinical Pearl:
The US Preventive Services Task Force (USPSTF) recommends screening for gonorrhea and chlamydia annually in sexually active women aged 24 years and younger and in older women who are at increased risk for infection.

STI screening recommendations differ between men who have sex exclusively with women and men who have sex with other men. For men who have sex with other men, it is reasonable to perform screening on at least an annual basis given the high prevalence rates of these STIs in this population. Routine screening is not recommended for HIV-uninfected heterosexual men for STIs unless they have a history of STIs. It is reasonable to screen heterosexual men at risk for STIs in areas of high prevalence, such as at STI or adolescent clinics and correctional facilities, if resources allow. More frequent screening is indicated at 3-month intervals for those at particularly high risk for STIs, including those with multiple or anonymous partners. Routine STI screening of HIV-infected patients in order to reduce the spread of STIs is warranted, particularly because STIs, in turn, can increase HIV transmission [35].

Bacterial cell cultures were considered to be the gold standard because of superior specificity and ability to detect a very small number of chlamydial organisms; however, nucleic acid amplification tests (NAATs) are now the most sensitive and specific tests for detecting chlamydial infections

and have become the standard diagnostic and screening test. Diagnosis in women can be made by testing urine or from vaginal/endocervical swabs. Vaginal swabs have been shown to have higher detection rates than urine testing and are the preferred method of testing for women. Diagnosis in men can be made by testing urine or from urethral swabs. Detection rates in urine are similar to detection rates from urethral swabs; therefore urine testing is the preferred method of testing for men. Men should not urinate for 1–2 hours before the urine sample is collected, and urine should be obtained from the first part of the urine stream. It is important to note that NAATs may remain positive for up to 3 weeks following treatment [36].

The sensitivity and specificity of the urine tests are both greater than 85%. A positive test generally means that a patient has the STI. Positive results warrant treatment with antibiotics as well as counseling regarding safe sex practices to prevent re-infection. Sex partners should also be notified, examined and treated for the STI. Positive results must be interpreted carefully in low prevalence populations as false-positive results may occur more frequently than true-positive results in this setting. False-positive tests can have severe psychological and relationship consequences. A negative test result does not exclude the possibility of infection in the following instances: improper specimen collection, concurrent antibiotic therapy, and low numbers of organisms in the specimen [34].

Returning to case 4, you discuss with your young sexually active female patient that chlamydia and gonorrhea are the most commonly reported sexually transmitted infections. While gonorrhea often causes symptoms, chlamydia is usually asymptomatic. She requests to be screened for chlamydia. The screen is positive. She has no reported medication allergies, so you treat her with 1 gram of oral azithromycin in the clinic. You also counsel her that her sexual partner should come to the clinic to be treated as well.

Summary

Urine testing is available and can be routinely used for screening purposes for substance use disorders, to detect diuretic abuse, to confirm antipsychotic medication adherence and to identify sexually transmitted infections in at risk populations. Positive urine drug screens should be confirmed and always interpreted in the context in which they were ordered, whereas a negative drug screen does not necessarily eliminate the possibility of abuse. Urine antipsychotic testing is not in universal use, but it has found utility among some community psychiatrists as testing serves not only as a means of compliance monitoring but also as a catalyst for conversations about adherence. Chlamydia and gonorrhea are the most common reported sexually transmitted infections in the United States; therefore the USPSTF has recommended screening annually in all sexually active women aged 24 years and younger and in older women who are at increased risk for infection.

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Chapter 17
Urine Tests: Solidifying Concepts – Questions and Answers

M. Lee Sanders, Lisa M. Antes, Victoria J. A. Sharp, and Gina M. Lockwood

Chapter 1. Urine: The Golden Elixir of Life

1. Most of the sodium filtered at the glomerulus is reabsorbed by which of the following:

   A. Collecting Duct
   B. Loop of Henle
   C. Proximal convoluted tubule
   D. Distal convoluted tubule

   Answer: C. Sodium is the main ion reabsorbed by the kidney. Approximately 65% is reabsorbed in the proximal convoluted tubule, 25–35% in the loop of Henle, 5–10% in the distal convoluted tubule and 1–3% in the collecting duct.

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2. Hereditary or acquired dysfunction of the Na^+Cl^- symporter in the distal convoluted tubule leads to which syndrome:

   A. Bartter
   B. Gitelman
   C. Liddle

   Answer: B. Hereditary or acquired dysfunction of the Na^+Cl^- symporter in the distal convoluted tubule results in Gitelman syndrome. This disorder has clinical features (hypokalemia, metabolic alkalosis, hypocalciuria) similar to those seen in patients given a thiazide diuretic. Hereditary or acquired dysfunction of the $Na^+K^+2Cl^-$ symporter in the thick ascending limb of the loop of Henle results in Bartter syndrome. This disorder has clinical features (hypokalemia, metabolic alkalosis, hypercalciuria) similar to those seen in patients given a loop diuretic. A hereditary or acquired activating mutation of the epithelial sodium channel (ENaC) in the collecting duct results in Liddle syndrome which is a disorder with clinical characteristics similar to those of a high aldosterone state (excessive sodium reabsorption, hypervolemia, hypertension, hypokalemia).

3. The average human adult produces how much urine volume in a day?

   A. 50–100 mL
   B. 400–500 mL
   C. 1000–2000 mL
   D. 3000–4000 mL

   Answer: C. The amount of urine produced per day depends on the state of hydration, activity level and overall health of the individual. Average urine production for adults is usually between 1–2 L per day. Producing too much or too little urine requires medical attention. Polyuria is a condition of excessive urine production (>3 L/day). Oliguria is the production of less than 400–500 mL/day and anuria is the production of less than 50–100 mL/day.

Chapter 2. Follow the Money: Costs, Reimbursement, and Regulations of Urine Based Testing

1. A patient visits his primary care practitioner at her clinic office, which bills as a hospital outpatient department. The patient is assessed, and the practitioner determines that the patient needs a urine-based test. She has the option of ordering between two tests that are clinically equivalent. The practitioner learned yesterday that her organization has contracted with a new outside lab. She has heard from peers that this lab has excellent quality, and that they negotiated a large discount on testing fees. The patient informs the practitioner that his insurance coverage recently lapsed. The practitioner's organization has a generous charity care program. What factors should the practitioner consider in deciding which test to order?

   A. Clinical equivalency
   B. Cost of test
   C. Patient's insurance coverage
   D. All the above

   Answer: D. Clinical equivalency relates to the test results being the same, whether performed internally or at an outside laboratory. The location where the test is performed has a direct effect on the cost of the test. As a hospital outpatient department, there is a facility fee associated with testing done in the practitioner's office, and the steep discount in the contract with the outside lab may outweigh this. It is also important to consider the patient's insurance coverage. The patient may be eligible for charity care, which could cover his entire bill or just a small portion of it. The patient will likely be charged a higher rate for this test than an insured patient, since insurance companies are able to negotiate discounted rates for testing and self-pay patients must pay the chargemaster rate for a test.

2. A patient visits her primary care practitioner and reports that she took an at-home pregnancy test that indicated she is pregnant. She has been trying for several months to get pregnant and would like confirmation. The patient has a private payer well known to the provider who will reimburse either a urine test or a blood test in full. What factors should the provider consider in ordering this test?

   A. Clinical equivalency
   B. Cost of test
   C. Shared decision-making
   D. All the above

   Answer: D. In this case, since the blood test is more accurate than a urine test, the results are not clinically equivalent. In considering cost, a urine test is less expensive than the blood test; however the patient would bear none of the cost for either test. With shared decision making, the patient may be in a better position than the provider to determine the trade-off between cost and accuracy.

3. Private payers generally reimburse the highest rates for urine-based tests performed in which location?

   A. Hospital outreach laboratories
   B. Independent laboratories
   C. Physician office laboratories
   D. Purchased by patient/performed at home

   Answer: C. Private payer reimbursement is generally highest for physician office laboratories, followed by hospital outreach laboratories and lowest for independent laboratories. Private payers do not, in general, reimburse for urine tests purchased by patient/performed at home.

Chapter 3. Going with the Flow: Proper Urine Testing Methods for Clinical Practice

1. A 30 year old healthy woman complains of urinary frequency and dysuria for 2 days. She has gross hematuria as well. A urinary tract infection is suspected. Which type of

specimen should be collected for a routine urinalysis and urine culture?

A. First morning voided specimen
B. Midstream "clean-catch"
C. 24-hour timed collection for blood and urine electrolytes
D. Suprapubic aspirate

Answer: B. Midstream "clean catch" would be the most appropriate type of specimen collection to perform a routine urinalysis and urine culture. The time of day, as in a first morning specimen, is not as important for a routine urinalysis and culture compared to having a fresh specimen. A 24-hour collection could lead to a contaminated specimen. A suprapubic aspirate could be sent for a routine urinalysis and culture if that was the only way to get a specimen. A clean-catch voided specimen is easier to obtain with less risk.

2. A 55 year old female complains of urinary frequency. Urine dipstick testing obtained in clinic using midstream clean-catch technique is positive for 1+ blood, 1+ leukocyte esterase, and is nitrite negative. What is the next step in evaluation/treatment of this patient?

A. Treat for 3 days with oral Ciprofloxacin for urinary tract infection
B. Treat for 7 days with oral Ciprofloxacin for urinary tract infection
C. Repeat urine dipstick testing using catheterized urine specimen
D. Obtain urine microscopy

Answer: D. Confirmatory urine microscopy should be obtained given the high incidence of false positive results on urine dipstick. There is not enough evidence on urine dipstick yet to treat for urinary tract infection, as her symptoms could have other causes. A catheterized urine specimen is not necessary unless significant contamination is strongly suspected.

3. A 30 year old female presents to your office for her annual physical exam. She is currently asymptomatic and her last normal menses (4 days duration) ended 1 day ago. She is trying to conceive. In counseling her for best results, if she performs a home urine pregnancy test, all the following are true, *except*?

   A. Concentrated, first-morning urine sample should be used
   B. The test should be performed 15 days after the start of the last normal menses
   C. The test should not be performed before the first day of missed menses
   D. She will have to call your office to get the results

   Answer: B. For best results, a home urine pregnancy test should not be performed before the first day of missed menses, or approximately 15 days after conception, although some claim accuracy earlier. The test should be performed on a concentrated, first morning urine sample. The test results are indicated on the test strip so there would be no need to call the office to get the results. Answer B is not true because 15 days after the start of the last normal menses would be around the time of ovulation/conception, and the test should not be performed until approximately 15 days after conception.

Chapter 4. Urine Dipstick: Blood - The Spectrum of Red

1. What does a speckled pattern on a urine dipstick for blood reagent strip indicate?

   A. Egg consumption
   B. Intact red blood cells
   C. Myoglobin
   D. Hemoglobin

   Answer: B. Reagent testing pad that is not uniform in color and appears blotchy, spotty or speckled is a good indication of intact red blood cells. Myoglobin and hemoglo-

bin both produce a positive reaction on the reagent strip, indicated by a solid color change. Egg consumption does not produce a reaction on the reagent strip.

2. A urine dipstick for blood reagent strip detects all the following, *except*?

   A. Free hemoglobin
   B. Free myoglobin
   C. Intact red blood cells
   D. Platelets

   Answer: D. The urine dipstick for blood reagent strip detects free hemoglobin, free myoglobin and intact red blood cells but not platelets.

3. Possible etiologies for dark reddish color urine include all the following, *except*?

   A. Red food coloring
   B. Hemoglobinuria
   C. Beet consumption
   D. Myoglobinuria

   Answer: A. Red food coloring consumption does not change the color of urine. Dark reddish colored urine can be caused by eating beets but the urine dipstick for blood and urine microscopy remain negative. Both hemoglobin and myoglobin can change the color of urine to red and cause a positive urine dipstick but with a negative urine microscopy for red blood cells.

Chapter 5. Urine Dipstick: Proteinuria – Causes, Consequences and Diagnostic Approach

1. A positive urine dipstick for protein should lead to which of the following:

   A. Immediately repeating the test to determine if correct
   B. Observation and recheck in 3 months
   C. Referral to a nephrologist

D. Quantification of the urine protein amount

Answer: D. If a urine dipstick is positive for protein, the next step is to quantify the amount of protein. This can be done by checking a urine protein to creatinine ratio which corresponds very well with a 24-hour urine protein measurement. Repeating the test immediately is usually not indicated unless collection error or a transient cause of proteinuria (urinary tract infection, fever, recent heavy exercise) is suspected. Proteinuria can be an initial clue that an underlying kidney disease is present, so mere observation is not recommended. Low level proteinuria can be managed by a primary care provider. Referral to a nephrologist may be indicated if the proteinuria is believed to be due to an intrinsic renal disease. Expedient referral to a nephrologist should occur if there is compromised renal function in addition to the proteinuria.

2. The following characteristics are components of nephrotic syndrome, *except*:

A. Urine protein excretion >3.5 grams per day
B. Hypoalbuminemia
C. Hypercholesterolemia
D. Hypertension

Answer: D. Nephrotic syndrome is usually defined as a constellation of symptoms including high amounts of urine protein (albumin) excretion (>3.5 g/day), hypoalbuminemia, hypercholesterolemia and pitting edema. Hypertension is more common with nephritic syndrome.

3. Which of the following patients should be screened for proteinuria? More than 1 answer may be correct.

A. 50 year old man with a new diagnosis of hypertension
B. 32 year old woman with a new diagnosis of type 2 diabetes
C. 15 year old girl with a new diagnosis of type 1 diabetes

D. 65 year old man with coronary artery disease and prior myocardial infarction
E. 50 year old man with morbid obesity and a strong family history of early heart disease
F. 50 year old man with chronic kidney disease stage G3b, but no diabetes or hypertension

Answers: A, B, F. All patients with a diagnosis of hypertension, type 2 diabetes (at the time of diagnosis) or type 1 diabetes (5 years after diagnosis) should be screened for proteinuria. Type 2 diabetic patients are screened at the time of diagnosis because it is presumed that they have had diabetes for years prior to diagnosis, while type 1 diabetics are typically identified and diagnosed soon after the disease manifests. If screening is negative for overt proteinuria with a typical urine dipstick, you should screen specifically for microalbuminuria. If the patient is positive for either microalbuminuria or proteinuria, treatment should include an angiotensin converting enzyme inhibitor (ACEi) or angiotensin receptor blocker (ARB). In patients D and E, although proteinuria predicts progression of coronary artery disease and chronic kidney disease, there are no formal recommendations for screening in this population. These patients should certainly be screened for hypertension, and if hypertensive, then screening for proteinuria should occur.

Chapter 6. Urine Dipstick: Urinary Nitrites and Leukocyte Esterase – Dipping into Murky Waters

1. You are seeing a 27 year old female who presents to your clinic complaining of burning with urination and increased frequency of urination. She is currently sexually active and does not use protection consistently. You order a urinalysis and it shows 1+ leukocyte esterase and negative nitrite. Testing is negative for gonorrhea and chlamydia but the urine culture comes back positive. An infection with which

of the following organisms would explain the negative nitrite?

A. *Escherichia coli*
B. *Staphylococcus saprophyticus*
C. *Klebsiella pneumoniae*
D. *Proteus mirabilis*

Answer B. Nitrate, which is uniformly present in the urine, is converted to nitrite by an enzyme called nitrate reductase. This enzyme is produced by a family of Gram-negative rods called Enterobacteriaceae, of which examples include *Klebsiella, Proteus,* and *E coli spp*. (thus options A, C and D would have a positive nitrite test). *Staphylococci* are Gram-positive cocci and are not in the *Enterobacteriaceae* family. Therefore, if the urine is infected with this organism, the nitrite test would be expected to be negative. Other organisms that can infect urine and do not have nitrate reductase include other *Staphylococci* and *Enterococcus.*

2. You are seeing a 76 year old female in your clinic who is asking for a urine culture because of a change in color of her urine. You decide to order a urinalysis and it shows 2+ urobilinogen, 2+ leukocyte esterase, and positive nitrite. Urine culture grows 100,000 colonies/ml of *Escherichia coli*. She denies any fevers, chills, flank pain, dysuria, polyuria, difficulty urinating, or any other associated symptoms. What is your next step in management?

A. Await susceptibility data, then treat based on the results
B. Start Ciprofloxacin and await susceptibilities
C. Start trimethoprim/sulfamethoxazole and await susceptibilities
D. Do not treat with antibiotics regardless of susceptibilities

Answer D. This patient has asymptomatic bacteriuria. The only symptom that she reports is a change in color of her urine, a finding that has not been associated with urinary tract infections in the literature. While her urinalysis is positive both for leukocyte esterase and nitrite, these do not discriminate between asymptomatic bacteriuria and

a true urinary tract infection. In the absence of symptoms, treatment should be deferred, even with a positive dipstick and positive culture in this patient population.

3. You see a 39 year old female with little past medical history who is coming in with burning with urination that started a few days ago. She is sexually active and uses protection 'most of the time'. She denies any history of sexually transmitted infections (STIs). You decide to order a urinalysis and it comes back with positive nitrite and 3+ leukocyte esterase. Microscopy shows 50 WBCs. Which of the following is most predictive of a positive urine culture?

   A. Positive nitrite
   B. 3+ leukocyte esterase
   C. 50 white blood cells on microscopy
   D. History of unprotected sexual intercourse

   Answer: A. Of the options listed above, a positive nitrite has the highest specificity and therefore the highest positive predictive value for detecting bacteria in the urine. Some studies show specificity as high as 98% with a range of 85–98%. As leukocyte esterase is more prone to false positives, the specificity is much lower. Common causes of false positives include contamination of the specimen, vaginosis, and STIs. Her history of unprotected sexual intercourse puts her at risk for STIs but is not predictive of a urinary tract infection.

Chapter 7. Urine Dipstick: An Approach to Glucosuria, Ketonuria, pH, Specific Gravity, Bilirubin and Urobilinogen - Undeniable Chemistry

1. All the following may cause a false-positive glucose reaction on the reagent pad, *except*:

   A. High concentration of vitamin C (ascorbic acid)
   B. Improper storage of sample or test strips
   C. Presence of oxidizing agents in the urine

D. Ketones in the urine

Answer: A. Improper storage of sample or test strips, presence of oxidizing agents in the urine and ketones in the urine can all produce a false-positive glucose reaction. A high concentration of ascorbic acid in the urine can produce a false-negative glucose reaction.

2. Ketonuria may be caused by all the following, *except*:

A. Alcoholic ketoacidosis
B. Vomiting
C. Diabetic ketoacidosis (insulin deficiency)
D. High carbohydrate diet

Answer: D. Alcoholic ketoacidosis, vomiting, insulin deficiency and low carbohydrate diet can cause ketonuria. A high carbohydrate diet does not usually cause ketonuria.

3. Hepatocellular jaundice could be associated with all the following, *except*:

A. Negative urine bilirubin
B. Positive urine bilirubin
C. Negative urine urobilinogen
D. Positive urine urobilinogen

Answer: C. Hepatocellular jaundice is usually associated with a positive or negative urine bilirubin and a positive urine urobilinogen. Urine urobilinogen is a very sensitive but non-specific test to evaluate for liver damage, hemolytic disease, and severe infections and as such would be positive in a patient with hepatocellular jaundice.

4. Urine pH above 8 may be caused by all the following, *except*:

A. A vegetarian diet
B. Diet high in citrus fruit
C. *E. coli*, an acid-producing bacteria

D. *Proteus*, a urease producing bacteria

Answer: C. Urinary pH close to 8 reflects a more *alkaline* urine. Pathologic causes of urinary alkalosis can include the presence of certain bacteria such as *Proteus*, a urease producing bacteria, or renal tubular acidosis. Benign dietary causes of urinary alkalosis include a vegetarian diet or diet high in citrus fruits (Answers A and B), Answer C is the correct answer because the presence of certain bacteria such as *E. coli*, an acid-producing bacteria, can be indicative of acidic urine, not an alkaline urine.

Chapter 8. Urine Microscopy: The Burning Truth – White Blood Cells in the Urine

1. Which of the following screening tests has the highest sensitivity for diagnosis of a urinary tract infection (UTI)?

   A. Urine dipstick for leukocyte esterase
   B. Urine dipstick for blood
   C. Urine microscopy for leukocytes
   D. Urine microscopy for red blood cells

   Answer: C. Pyuria, or the presence of leukocytes on microscopic urinalysis, is a strong objective indicator of a UTI. Sensitivity ranges from 90% to 96%. A diagnosis of UTI is rare in the absence of pyuria. Leukocyte esterase usually detects the presence of WBCs in the urine, and in turn infection, but it is not as accurate as identification of WBCs on microscopy. Red blood cells can be present in UTIs, but not every UTI is associated with hematuria.

2. Which of the following is not considered a cause of sterile pyuria?

   A. *E. coli* urinary tract infection
   B. Urinary tract stone
   C. Genitourinary tuberculosis

D. Gonococcal urethritis

Answer: A. An *E. coli* urinary tract infection would yield a positive bacterial urine culture, so pyuria would not be considered "sterile." The rest of the choices are considered causes of sterile pyuria. Although genitourinary tuberculosis and gonococcal urethritis are both infections, they should not lead to a positive urine culture on standard culture medium. Urolithiasis causes sloughing of leukocytes in the urinary tract even in the absence of infection.

3. A 50 year old female presents with a 1 month history of urinary incontinence that she describes as a constant, low-volume dribble in the underwear. Volume increases with movement from a seated to standing position, as well as with laughing or coughing. She underwent an abdominal hysterectomy 6 weeks ago for uterine fibroids. She has no abdominal pain, and her abdominal incision is healing well. She has a normal speculum and pelvic examination. Renal ultrasound is obtained showing moderate hydronephrosis of the left kidney. A "double dye" test is performed, and upon removal of the tampon from the vagina, there is an orange/yellow discoloration on the proximal ½ of the tampon but is otherwise dry. What diagnosis does this clinical picture suggest?

A. Vesicovaginal fistula
B. Ureterovaginal fistula
C. Colovesical fistula
D. Urethrovaginal fistula

Answer: B. Ureterovaginal fistula is suspected for multiple reasons. Unilateral hydronephrosis on ultrasound indicates an obstruction affecting one kidney and ureter. A vesicovaginal fistula, however, could not be ruled out by this study, as it could also affect one kidney if near the ureteral orifice. The double dye test indicates urine from the ureter staining the upper 1/2 of the tampon after administration with oral phenazopyridine. There is no concomitant evidence of a bladder or urethral fistula

given no blue dye on the tampon after intravesical instillation of methylene blue. A "double dye" test would not yield a positive result in the setting of a colovesical fistula.

Chapter 9. Urine Microscopy: Seeing Red – Understanding Blood in the Urine

1. Which of the following findings on urine microscopy most strongly suggests a glomerular origin for hematuria?

   A. Spherocytes
   B. Sickle cells
   C. Acanthocytes
   D. > 50 RBCs/hpf

   Answer: C. Acanthocytes, or spur cells, are a specific type of dysmorphic RBC that suggests deformation with passage through the glomerular basement membrane. A large percentage of acanthocytes in the urine suggests glomerular source of hematuria. Spherocytosis suggests intravascular hemolysis. A high RBC concentration is more commonly associated with bleeding from a non-glomerular origin like genitourinary malignancy. Sickle cells suggest sickle cell anemia.

2. A 55 year old male patient with history of type 2 diabetes mellitus and tobacco use undergoes a comprehensive medical evaluation by his primary care provider. He reports nocturia three times nightly as well as daytime urinary frequency. Urinalysis is notable for 5 RBCs/hpf and is otherwise unremarkable. He has had no recent trauma or urethral instrumentation. What is the most appropriate next step in management?

   A. Repeat urinalysis and microscopy to confirm diagnosis of microscopic hematuria
   B. Prescribe empiric course of antibiotics for possible prostatitis
   C. Obtain retroperitoneal ultrasound

D. Refer to urology

Answer: D. Based on American Urologic Association guidelines, a single episode of microscopic hematuria warrants prompt urologic evaluation. This is especially true in a patient with risk factors like smoking. A urinalysis could be repeated to confirm the diagnosis, but it is not needed for referral. Urine culture should also be obtained given irritative lower urinary tract symptoms, as urinary tract infection or prostatitis are also possible. Upper tract imaging is warranted. CT urogram is preferred to renal ultrasound if renal function is normal to best characterize the upper tracts.

3. A 16 year old male presents with a 3 day history of "cola-colored" urine following a "nasty cold" for which he stayed home from school for 1 week. Urinalysis shows >50 RBCs/hpf. Given this information, what is the most likely cause of his hematuria?

A. Urothelial carcinoma
B. Ingestion of dye from too much coca cola
C. IgA nephropathy
D. Wilms tumor

Answer: C. Given the brown discoloration of this patient's urine alone, a glomerular cause of hematuria is suggested. This patient's age and recent illness are consistent with IgA nephropathy or post-infectious glomerulonephritis. Non-glomerular causes of urine like malignancy usually yield bright red discoloration of the urine. Ingestion of dye is not suggested as this patient does have RBCs on his urine microscopy in addition to discoloration.

Chapter 10. Urine Microscopy: The Utility of Urinary Casts in Patient Care – Practical and Useful Tips for Busy Clinicians

1. Factors that favor urinary cast formation include all the following, *except*:

   A. Low flow state
   B. Low urine pH
   C. High urine pH
   D. High salt concentration

   Answer: C. Factors that favor urinary cast formation include low urinary flow state/rate through the tubules, a high salt concentration in the urine and a low urine pH. A high urine pH (alkaline urine) is detrimental to urinary casts. Standing urine becomes progressively more alkaline over time as urea is broken down generating ammonia. This ammonia generation leads to an increasing alkaline environment which dissolves casts and promotes cell lysis.

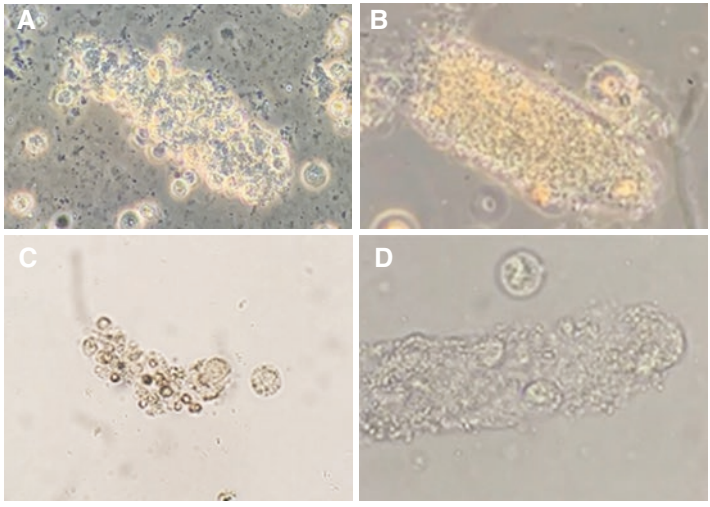
2. Urinary cast generally form in which anatomical part of the nephron?

   A. Glomerulus
   B. Proximal convoluted tubule
   C. Loop of Henle
   D. Distal convoluted tubule

   Answer: D. The precise anatomic location for urinary cast formation is the distal convoluted tubule and/or the collecting duct. Casts are generally not formed within the glomerulus, proximal convoluted tubule or loop of Henle. The reason for this anatomic distinction is that casts are composed of an external backbone of uromodulin (Tamm-Horsfall) protein which is secreted mainly in the distal convoluted tubule.

3. Match the cast type with the appropriate cast image below.

   Red blood cell cast
   White blood cell cast
   Renal tubular epithelial cell cast
   Granular cast



Answers:

Red blood cell cast: C
White blood cell cast: A
Renal tubular epithelial cell cast: D
Granular cast: B

Chapter 11. Urine Microscopy: Clouding Over – Bacteria, Yeast, Parasites and Zika

1. You are seeing a 37 year old male in your office for painless gross hematuria. His urinalysis is remarkable for 3+ blood with 15 RBCs noted on microscopy. He is originally from Egypt and immigrated to the Unites States 2 years ago. You

look at the microscopy and see what resembles an egg. What is the most likely cause?

A. Zika virus
B. Tuberculosis
C. Schistosomiasis
D. Trichomoniasis
E. Ascaris

Answer C. This is a case of painless gross hematuria caused by Schistosomiasis. The species that affects the genitourinary tract is *Schistosoma haematobium* and is found mostly along the Nile River or in sub-Saharan Africa. This can cause painless hematuria by excretion of the eggs into the bladder wall, but it is also associated with bladder cancer, so cystoscopy should be considered if the diagnosis is made. None of the other options would show the presence of eggs in the urine.

2. You receive a call from a nursing home regarding a 75 year old male with multiple medical problems including benign prostatic hyperplasia who has had an indwelling urinary catheter for the last 2 weeks. The nurse noted a foul smell of his urine today and sent off a urinalysis and urine culture. The urinalysis showed 2+ leukocyte esterase and negative nitrite. The urine microscopy was notable for 45 WBCs and yeast. Per the report from the nurse, he is feeling well overall and not having any fevers, chills, or abdominal pain. What is the most appropriate next step in his care?

A. Urine culture
B. Start fluconazole
C. Start amphotericin
D. Start nitrofurantoin
E. Monitor clinically

Answer E. This is asymptomatic Candiduria which rarely requires treatment. Foul smell is not considered an indication for treatment of a urinary tract infection. While he does have a positive UA and microscopy, this alone

does not warrant treatment in an asymptomatic patient. If he was undergoing a urologic procedure, treatment should be considered.

3. A 25 year old diabetic female has asymptomatic bacteriuria on routine urine studies. The next step in management is:

   A. Urinary tract imaging
   B. No treatment
   C. Antibiotics alone
   D. Antibiotics followed by cystoscopy
   E. Antibiotics followed by annual screening urinalyses

   Answer B. The isolated presence of asymptomatic bacteriuria in adults is not known to cause harm; however, these patients are at an increased risk for developing a symptomatic urinary tract infection at some point in their lives. The presence of diabetes, although a risk for development of other conditions (e.g. cardiovascular disease, peripheral neuropathy, renal insufficiency), in and of itself does not warrant screening for asymptomatic bacteriuria. Those who do benefit from screening and subsequent treatment of asymptomatic bacteriuria include patients who will be undergoing a urologic procedure that requires entry into the urinary tract and pregnant females.

Chapter 12. Urine Microscopy: Urine Made Crystal Clear

1. You incidentally find crystalluria on an otherwise healthy patient's urine with no history of stones and no concerning risk factors. The patient is worried about this "abnormal" finding. What is the incidence of crystalluria in healthy patients?

   A. 5–10%
   B. 15–20%
   C. 30–35%

D. 40–45%
E. 55–60%

Answer: B. Approximately 15–20% of asymptomatic, healthy patients with no history of kidney stones will have crystalluria on urinalysis. Crystal formation in the urine is dependent on many physical factors including urine pH, the degree of saturation in the urine (i.e., concentration) and whether inhibitors of crystal formation are absent.

2. A 20 year old male has intermittent flank pain and has noticed "sand" passing in his urine. The flank pain resolves after the sand passes. His mother also has a history of kidney stones. A urinalysis shows microscopic hematuria and hexagonal-shaped crystals. The next best step is:

   A. Reassurance and hydration
   B. Give pain medication and tamsulosin and follow up with a KUB Xray
   C. Referral to the metabolic stone clinic
   D. 7 days of antibiotics per urine culture results
   E. CT urogram and referral to urology

   Answer: C. Hexagonal crystals on urinalysis are pathognomonic for cystinuria, a genetic cause of kidney stones; therefore, the patient should be referred to urology/nephrology or a metabolic stone clinic for evaluation and treatment for stone prevention. He also should have imaging to quantify his current stone burden and assess the need for surgical intervention.

3. A 60 year old female presents to the emergency department with fevers and left flank and abdominal pain. She is tolerating clear liquids. CT abdomen/pelvis shows a 5 mm left distal ureteral stone with associated hydronephrosis. Creatinine is 1.2 mg/dL and urinalysis is positive for leukocyte esterase and nitrites. The next best step is:

   A. Consult urology for urgent ureteral stent placement
   B. Discharge with a course of ciprofloxacin for complicated pyelonephritis

C. Admit to the hospital for pain control and monitoring of fevers
D. Discharge with pain medications, ciprofloxacin, and tamsulosin and set up close follow up with urology
E. Consult urology for urgent lithotripsy

Answer: A. Urinary obstruction with associated fevers and concern for sepsis secondary to urinary source should be urgently evaluated by urology for decompression of the urinary tract. This can be a life-threatening situation, and patients can decompensate quickly. Drainage of the urinary tract with either ureteral stent or a percutaneous nephrostomy (PCN) tube placement is essential for management.

Chapter 13. Urine Testing in Children: Little People, Big Challenges

1. A 1 year old male is brought to the emergency department with a 2day history of fevers as high as 39°C, decreased oral intake, and increased fussiness. He is otherwise asymptomatic. He has a history of one episode of pyelonephritis in the past necessitating hospital admission. Physical examination including respiratory, ear examination, and abdominal examination are unremarkable. He is uncircumcised, but his foreskin retracts to reveal his urethral meatus. What is the most appropriate method to obtain a urine specimen in this child for testing?

A. Clean-catch midstream specimen
B. Bagged urine specimen
C. Suprapubic aspirate
D. Catheterized urine specimen

Answer: D. Catheterized urine specimen is the most appropriate urine collection method for this clinical scenario, and it is recommended by the American Academy of Pediatrics in children under 24 months in whom a urinary tract infection is suspected. A clean-catch urine specimen cannot be obtained in a child who

is not toilet-trained. Bagged urine specimens, although non-invasive, have a high risk of contamination, especially in an uncircumcised patient. If this child is inaccurately diagnosed with pyelonephritis, it may lead to expensive and invasive testing unnecessarily. Although suprapubic aspirate would yield a specimen with low risk for contamination, it is more invasive than urinary catheterization and requires skilled personnel as well as possible imaging guidance.

2. A 5 year old boy presents with a 1 week history of cola-colored urine. He has had no recent illnesses or trauma. On examination in the office, you note that he has periorbital edema and bilateral lower extremity pitting edema. Urine dipstick shows positive blood and protein. Random urine protein to creatinine at the time of the visit is 3.6. What is an appropriate next step?

   A. Repeat urine protein to creatinine ratio in the first morning voided sample within 1 week
   B. Obtain two to three urinalyses with microscopic analysis over the next two to 3 weeks
   C. Refer patient to pediatric nephrology
   D. Treat empirically with oral antibiotics

   Answer: C. Given this child's systemic symptoms, proteinuria and hematuria, prompt nephrologic evaluation is warranted, even with a single urinalysis. Further quantitative urine protein studies may be performed. Although microscopic urinalysis should be obtained to confirm findings, there is enough clinical information to warrant nephrology referral. Urinary tract infection is not suspected based on clinical history and urinalysis results.

3. True or False: Circumcision decreases the risk of urinary tract infection (UTI) in male infants.

   Answer: True. Circumcision decreases the risk of a UTI by approximately tenfold during the first year of life. Risk of pyelonephritis in an uncircumcised boy within the first year of life is approximately 1–2%. Thus, circumcision should be considered in children at high risk for

recurrent pyelonephritis, like those with posterior urethral valve or severe vesicoureteral reflux. After the first year, the risk of UTI is similar between circumcised and uncircumcised boys.

Chapter 14. Urine Based Tests in the Diagnosis of Genitourinary Cancers

1. A 63 year old male with a 50 pack-year smoking history presents to his primary care physician for 6 weeks of lower urinary tract symptoms including frequency and dysuria. A urine dipstick is obtained and is negative for nitrites and leukocyte esterase but shows 1+ blood. Which of the following is the most appropriate next step?

   A. Referral to urology for hematuria workup
   B. Send a urine culture and await results prior to treatment
   C. Order urine cytology
   D. Perform microscopic urinalysis
   E. Start tamsulosin 0.4 mg daily

   Answer: D. This patient with significant smoking history and 6 weeks of irritative lower urinary tract symptoms is at increased risk of urothelial cancer. A urine dipstick examination is not sufficient for diagnosing microscopic hematuria, and formal microscopic examination is required. If microscopic exam reveals ≥3 RBC/hpf, he requires referral to a urologist for hematuria evaluation with a CT urogram and cystoscopy. A urine culture should be obtained, but hematuria should also be confirmed. Cytology has a limited role in initial diagnosis of bladder cancer. Tamsulosin for benign prostatic hypertrophy is appropriate in some patients with such symptoms as hesitancy and weak stream but would not be first line in this patient.

2. A 65 year old male presents to a urologist for microscopic hematuria evaluation. He has no smoking history, no known occupational exposures, and no irritative voiding symptoms. Which of the following tests would most accurately determine his need for further workup including cystoscopy and CT urogram?

   A. Urine cytology
   B. Fluorescence in situ hybridization (FISH)
   C. Cxbladder Triage
   D. Cxbladder Detect
   E. Cxbladder Monitor

   Answer: C. This is a patient presenting with incidental microscopic hematuria and no known risk factors for development of bladder cancer. Cxbladder Triage is a non-invasive test that will help determine his probability of bladder cancer and thus help decide if he warrants further evaluation with cystoscopy. Cxbladder Detect can be useful to diagnose cancer in high risk patients, unlike this patient. Cytology, FISH, and Cxbladder Monitor are more useful in surveillance and when other testing is equivocal.

3. A 75 year old male with a history of urothelial carcinoma-in-situ presents for his first office cystoscopy following 6 weeks of induction with bacille Calmette-Guérin (BCG). His bladder appears normal on cystoscopy. Bladder wash is obtained, and cytology returns with a report of "atypical cells". Which of the following additional tests can help stratify the need for further evaluation?

   A. Repeat cytology
   B. Cxbladder Detect
   C. Fluorescence in situ hybridization (FISH)
   D. SelectMDx
   E. Cxbladder Triage

   Answer: C. Cytology can take several weeks to normalize in patients undergoing BCG treatments. In patients with indeterminate cytology, adjunctive tests such as

FISH can be helpful in predicting the presence of residual cancer versus lingering treatment effect. Cxbladder Detect and Cxbladder Triage are more useful in new hematuria evaluation. SelectMDx is a marker for prostate cancer.

4. A 60 year old male has been followed by his urologist for a history of elevated prostate specific antigen (PSA). He has had one prior negative transrectal ultrasound-guided biopsy 2 years ago. His current PSA is 6.7 ng/mL (normal <4.5 ng/mL) and his digital rectal exam is normal. His prostate size is estimated at 65gm (mild enlargement). There is a family history of prostate cancer in his father and a paternal uncle. Which of the following statements is true?

   A. PCA3 testing is appropriate for this patient who has had one prior negative biopsy
   B. A PCA3 score < 25 rules out the presence of prostate cancer
   C. PCA3 score is not appropriate for this patient as his prostate size is >50gm
   D. The patient may collect his urine sample at home and send to the lab for PCA3 testing
   E. SelectMDx would likely stratify this patient as "intermediate risk"

   Answer: A. In a patient >50 years old with a prior negative biopsy, PCA3 testing can help risk stratify and determine the need for repeat biopsy. PCA3 is not affected by prostate size. The sample must be collected in the clinic after digital rectal examination. While a score < 25 predicts a higher likelihood of negative biopsy, PCA3 cannot completely rule out the presence of cancer. SelectMDx stratifies patients only as either "increased risk" or "very low risk" for clinically significant prostate cancer.

Chapter 15. Kidney Excretions: The Lyter Side of Urine

1. A patient is seen by her primary care physician for cramps. An electrolyte panel is drawn and the potassium (K) level returns at 2.5 mEq/L (normal 3.5–5 mEq/L) while she is still in the office. She has no chronic medical problems other than gastrointestinal reflux disease. Her serum creatinine is normal, plasma glucose is normal and she has no acid-base disturbances. Her BP is a bit on the low side at 95/60. Which of the following would be a correct statement regarding the etiology of this patient's hypokalemia?

   A. A high urine K (>20 mEq/L) would indicate appropriate renal response to hypokalemia
   B. A high urine K (>20 mEq/L) would make you suspicious that diarrhea is the cause of the hypokalemia.
   C. She is likely surreptitiously using insulin, causing a transcellular shift of K
   D. A magnesium (Mg) level should be checked and if low prompt a review of any recent medication changes

   Answer: D. While history and physical exam remain important in evaluation of electrolyte disturbances, the etiology of hypokalemia can be divided into 4 main categories: increased renal losses of K, increased extrarenal losses of K, decreased dietary intake/poor absorption, and transcellular shifts. In the face of hypokalemia, the kidney should be acting to conserve K and thus the urine K should be low (option A is incorrect). For any extrarenal causes of hypokalemia (diarrhea), the urine K should be low (option B is incorrect). If the patient is currently using insulin and is non-diabetic, you would expect the serum glucose to be low, but this patient has a normal glucose. A serum Mg can be very helpful in elucidating the cause of possible medication induced hypokalemia, as might be seen with proton pump inhibitors. On further questioning,

she had started taking a proton pump inhibitor over-the-counter 1 month prior to this encounter for her ongoing reflux symptoms.

2. A 60 year old female is seen for hypokalemia evaluation. She has had some mild diarrhea, but the diarrhea improved 3 days ago. She has been asymptomatic but adding more potassium to her diet has not improved the serum K. Her medications include atorvastatin for hyperlipidemia, topiramate for mood disorder, lisinopril for hypertension and rare use of ibuprofen for headaches. Her labs revealed sodium 142 mEq/L (normal 135–145 mEq/L), potassium 3.0 mEq/L (normal 3.5–5 mEq/L), chloride 109 mEq/L (normal 95–107 mEq/L), bicarbonate of 15 mEq/L (normal 23–25 mEq/L) and albumin 4 g/dL (normal 3.5–4.8 g/dL). Urine anion gap analysis on a random urine specimen revealed the following: sodium 60 mEq/L, potassium 80 mEq/L and chloride 50 mEq/L. What is the most likely cause of the patient's hypokalemia?

   A. Topiramate
   B. Diarrhea-related potassium losses
   C. NSAIDs
   D. Lisinopril

   Answer: A. This patient has a non-anion gap metabolic acidosis (NAGMA) and hypokalemia. Two main differentials of a NAGMA are diarrhea and renal tubular acidosis (RTA); while clinical history is important, calculation of the urine anion gap may assist in differentiating the 2 possible etiologies. Recall the formula for calculation of the urine anion gap is: Urine Anion Gap = $[Na^+] + [K^+] - [Cl^-]$. In this patient the urine anion gap was calculated to be +90 = (60 + 80–50). A positive value would make an RTA likely. Topiramate is a likely culprit (option A) and is known to cause both type I and type II RTAs. Although NSAIDs and lisinopril can cause RTAs, they are both associated with a

type IV RTA which is characterized by hyperkalemia (options C and D are incorrect). Diarrhea would result in a negative urine anion gap (option B is incorrect).

3. A 42 year old male patient with hyponatremia of 130 mEq/L (normal 135–145 mEq/L) is evaluated in clinic. Past medical history includes hypertension treated with lisinopril 5 mg daily and depression treated with fluoxetine 40 mg daily. The patient notes urinating a lot when questioned, and on a 24-hour urine collection made 7 L of urine in a day (normal 1–2 L/day). The patient's urine osmolality is 85 mOsm/kg. The patient undergoes a water deprivation test and at the end of the test, the urine osmolality concentrates to 700 mOsm/kg and the serum sodium increased to 135 mEq/L. What is the next step?

   A. Give desmopressin (DDAVP) as this may be diabetes insipidus
   B. Limit the patient's free water intakes as he likely has primary polydipsia
   C. Stop lisinopril as it is causing free water retention
   D. Stop the fluoxetine as it is causing free water retention

   Answer: B. This patient has polyuria (urine volume > 3 L/day). The next step in evaluation of polyuria is to determine if the diuresis is due to water or solutes. The low urine osmolality (<100 mOsm/kg) indicates a water diuresis. The water deprivation test has 2 phases: a dehydration phase in which fluid is restricted and the patient is monitored closely both clinically and with labs and the desmopressin administration phase. A rise in the urine osmolality to normal range with the water deprivation test is consistent with a diagnosis of primary polydipsia (option B). There is no indication to administer desmopressin as the patient has been able to concentrate their urine to normal range with dehydration alone (option A is incorrect). There is no indication of any medication side effects in this case (options C and D are incorrect).

Chapter 16. Other Common Uses for Urine Screening in Clinical Practice: Substance Use Disorders, Antipsychotic Adherence, Sexually Transmitted Infections

1. The typical urine drug screen is directed at detecting all the following, *except*:

   A. Amphetamines
   B. Marijuana
   C. Opiates
   D. Ecstasy

   Answer: D. The typical urine drug screen is directed at detecting cocaine metabolites, amphetamines, phencyclidine, marijuana metabolites and opiate metabolites.

2. Urine monitoring of antipsychotic medication use at the time of initial patient mental health evaluation should be considered in all the following instances, *except*:

   A. New diagnosis
   B. Homeless
   C. Substance use disorder
   D. Young age

   Answer: D. Urine antipsychotic testing is not in universal use, but it has found utility among some community psychiatrists. Patients who present for their initial mental health evaluation with symptoms of a serious mental illness with no previously established diagnosis (new diagnosis) or patients with a known serious mental health illness who have risk factors for poor treatment adherence (homeless, co-occurring substance use disorder, elderly) should undergo urine monitoring.

3. Untreated chlamydia or gonorrhea can lead to which of the following complications in women?

   A. Pelvic inflammatory disease
   B. Ectopic pregnancy
   C. Infertility

D. Chronic pelvic pain
E. All the above

Answer: E. Untreated chlamydia or gonorrhea can lead to complications such as pelvic inflammatory disease, ectopic pregnancy, infertility and chronic pelvic pain. Given these complications and the fact that both chlamydia and gonorrhea are treatable sexually transmitted infections (STIs), the US Preventive Services Task Force recommends screening for chlamydia and gonorrhea annually in sexually active women aged 24 years and younger as well as in older women who are at increased risk for infection. Increased risk factors for infection include new or multiple sex partners, a sex partner with concurrent partners, a sex partner with an active STI, inconsistent condom use among persons who are not in mutually monogamous relationships, previous or concurrent STI history and exchanging sex for money or drugs.

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