International Urogynecology Journal (2018) 29:205–210
https://doi.org/10.1007/s00192-017-3528-8

CLINICAL OPINION

Urine trouble: should we think differently about UTI?

Travis K. Price[1] · Evann E. Hilt[1] · Tanaka J. Dune[2] · Elizabeth R. Mueller[3] · Alan J. Wolfe[1] · Linda Brubaker[4,5]

Received: 11 September 2017 / Accepted: 27 November 2017 / Published online: 26 December 2017

Abstract
Urinary tract infection (UTI) is clinically important, given that it is one of the most common bacterial infections in adult women. However, the current understanding of UTI remains based on a now disproven concept that the urinary bladder is sterile. Thus, current standards for UTI diagnosis have significant limitations that may reduce the opportunity to improve patient care. Using data from our work and numerous other peer-reviewed studies, we identified four major limitations to the contemporary UTI description: the language of UTI, UTI diagnostic testing, the *Escherichia coli*-centric view of UTI, and the colony-forming units (CFU) threshold-based diagnosis. Contemporary methods and technology, combined with continued rigorous clinical research can be used to correct these limitations.

Keywords Diagnostics · *Escherichia coli* · Microbiome · Pathogens · Urinalysis · Urinary tract infection

Introduction

Urinary tract infection (UTI) is the most common bacterial infection in adult women, with 50% of women experiencing at least one UTI in their lifetime and as many as 10% having at least one UTI annually. UTI is among the most common reasons for antibiotic treatment [1, 2]. The nomenclature and concepts of UTI, based on the now disproven dogma that the lower urinary tract is a sterile environment, have remained stagnant over many decades. This older dogma has been informed with scientific evidence that some bacteria are present in the absence of urinary symptoms or a positive result of traditional UTI tests (urinalysis or standard urine culture). Although discovery of the urinary microbiota should clearly affect the care of women with UTI, specific clinical changes occur slowly. It is already clear that the widely used standard urine culture methods for detecting urinary bacteria have significant limitations compared with 16S rRNA gene sequencing [3–6] and more sensitive enhanced culture techniques [5–8]. These more sensitive assays have demonstrated that the female urinary bladder contains its own community of microbes, or microbiota. It is increasingly evident that alterations to the microbiota throughout the human body can have an impact on health.

We believe that it is time to advance UTI diagnosis and treatment. The first step in this process is a clear discussion of the limitations of the current standards in the context of the new knowledge about the female urinary microbiota. Although empiric treatment is currently pragmatic and highly effective for symptom resolution in uncomplicated, infrequent UTI, we anticipate that increasing recognition of the collateral effects of this regimen may cause a change in this practice. Our therapeutic goal should be to provide an optimal treatment, with high efficacy and few, if any, undesired effects. We highlight four major limitations in current UTI thinking: the language of UTI, UTI diagnostic testing, the *Escherichia coli*-centric view of UTI, and the colony-forming units (CFU) threshold-based diagnosis. Future research to overcome these limitations using rigorous clinical testing is paramount.

✉ Linda Brubaker
librubaker@ucsd.edu

[1] Department of Microbiology and Immunology, Stritch School of Medicine, Loyola University Chicago, Maywood, IL, USA

[2] Department of Urology, Center for Female Pelvic Health, Weill Cornell Medicine, New York, NY, USA

[3] Departments of Obstetrics & Gynecology and Urology, Loyola University Medical Center, Maywood, IL, USA

[4] Department of Reproductive Medicine, Division of Female Pelvic Medicine and Reconstructive Surgery, University of California San Diego, San Diego, CA, USA

[5] Division of Female Pelvic Medicine and Reconstructive Surgery, Department of Reproductive Medicine, University of California San Diego, La Jolla, CA, USA

The limitations of the language of UTI

The widely used UTI nomenclature is based on a dichotomous clinical scenario—infected or not (with the odd exception of asymptomatic bacteriuria). The traditional view of UTI envisions uropathogens invading a previously sterile environment (the bladder) to cause infection. Antibiotics come to the rescue to rid the person of the invading microbes and the bladder returns to the previously uninfected state. This language is grossly inconsistent with our current scientific knowledge about UTI. More importantly, this language does not recognize changes in the community of microbes that reside in the bladder. This microbial community can have beneficial functions in warding off infection. Furthermore, disruption to the resident microbial community (i.e., dysbiosis) could plausibly result in UTI symptoms, as is the case with several other disorders (see below). In adult women, such dysbioses could result from an invading microbe in the urinary system, an antimicrobial treatment, or some change to the host's metabolism or immune system. Nonetheless, our current language does not place the "UTI-causative microbe invades" concept in the context of noncausative microbes. The goal of UTI treatment should not be to eradicate every microbe in the bladder, especially given the evidence that some members of the urinary microbiota are beneficial and/or protective [5].

The limitations of UTI testing

The Standard Urine Culture (SUC) is currently the diagnostic gold standard for confirming the presence of bacteriuria for UTI diagnosis [9]. Unfortunately, SUC has significant limitations that have resulted in a profound detection bias. This bias has caused fundamental misunderstandings of the bacterial contributions to urinary health and disease.

Typically, SUC is performed in a clinical laboratory by plating 1 μL of urine onto Blood and MacConkey agar plates and incubating aerobically at 35 °C for 24 h. Since the original description of this technique in the 1950s [10], this protocol has been adopted as the standard diagnostic tool for the detection of UTIs, despite numerous limitations reported by many different investigators [11–13]. These limitations include: the inability to detect slow-growing microorganisms, the inability to grow fastidious and non-aerobic microorganisms, the inability to detect microorganisms present at less than 10^3 CFU/mL, and the difficulty of detecting underlying Gram-positive bacteria due to a lack of selective media.

More sensitive culture techniques, such as Expanded Quantitative Urine Culture (EQUC), have repeatedly shown that SUC possesses a 90% false-negative rate [5, 6, 8]. Relative to SUC, this enhanced urine culture protocol uses 100x more urine plated onto several different media and environmental conditions with twice the incubation time [8]. EQUC has provided compelling evidence that almost every adult female studied to date is bacteriuric [2].

In 2016, we demonstrated that SUC even fails to detect "clinically relevant microorganisms" in symptomatic patients [14]. We prospectively enrolled 150 urogynecology patients and dichotomized the group based on their UTI perception by asking, "Do you feel you have a UTI?" (Fig. 1) Transurethral catheterized urine specimens were collected and urinary symptoms were documented using the validated UTI Symptoms Assessment (UTISA) questionnaire [15].

We assessed the microbiota using both SUC and EQUC. In the catheterized urine sample of most women in the UTI cohort (69 out of 75), EQUC detected one or more bacteria that the literature classifies as uropathogens. In these 69 urine samples, EQUC identified a total of 110 uropathogens. In contrast, SUC only detected 50% (55 out of 110) of these uropathogens. Seventy-nine percent (59 out of 75) of the participants in the UTI cohort completed the follow-up UTISA questionnaire. Following clinically selected treatment based on SUC, 59% (35 out of 59) of participants reported symptom improvement, while 41% (24 out of 59) reported no improvement. Half (12 out of 24) of the 24 participants who did not improve had at least one uropathogen detected by EQUC, but not SUC. Collectively, these data show that SUC fails to detect microorganisms that may be contributing to UTI symptoms. Thus, sole reliance on SUC in these patients could lead to a suboptimal clinical outcome. It is reasonable to test whether the use of EQUC would have led to improved symptom resolution.

Other commonly used diagnostic tests are also severely limited. Dipstick and urinalysis tests are commonly used in the replacement of or in conjunction with SUC, but the efficacy of these rapid tests has been questioned, especially for Gram-positive infections [16, 17]. Current studies continue to highlight the limitations of these tests; our own unpublished

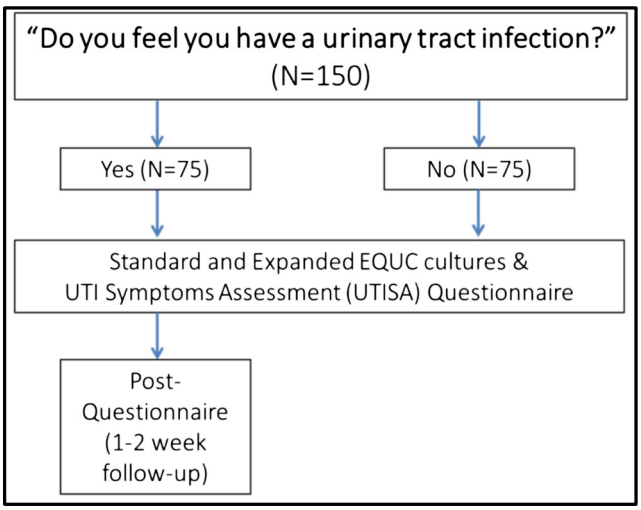

Fig. 1 Patient schematic of Price et al. [14]

data reveal a lack of utility for these tests. For example, we collected transurethral catheterized urine samples from a population of urogynecologically treated women ($N = 199$) with and without lower urinary tract symptoms (LUTS). In this population, 17 out of 199 (8.5%) had a positive dipstick result (i.e., the presence of white blood cells, nitrates, and/or red blood cells). Of these, one (5.9%) had bacteria detected by SUC whereas 13 (76.5%) had bacteria detected by EQUC. In the 182 (91.5%) with a negative dipstick, SUC detected bacteria in 17 samples (9.3%), whereas EQUC detected bacteria in 122 (67.0%). The percentage of bacteriuric samples diagnosed by SUC ($p = 0.23$) or EQUC ($p = 0.63$) were not statistically different in the dipstick-positive or dipstick-negative groups, showing that a positive urine dipstick test has no association with the presence of microorganisms, in this patient population (Price, unpublished data).

The limitations of an *Escherichia coli*-centric view of UTI

Standard Urine Culture was used to establish that *E. coli* is the most common cause of uncomplicated UTI [1, 18]. Yet, other Gram-negative bacteria such as *Pseudomonas aeruginosa* and several species within the Enterobacteriaceae family (e.g., species of the genera *Proteus* and *Klebsiella*) also can cause UTIs [18, 19]. In addition, a few Gram-positive bacteria, like *Staphylococcus saprophyticus* and *Enterococcus faecalis*, as well as some fungi, such as *Candida* sp., have been linked to UTIs [18, 19]. In some patients with complicating factors, the commonality of these organisms depends on other co-morbidities and demographics [19]. The list of uropathogens has grown longer with the availability of more sophisticated identification tools. Several additional microorganisms have now been classified as "emerging uropathogens" [20, 21]. These microorganisms have been found in high colony counts in patients with UTI symptoms and/or acute cystitis, but knowledge regarding their pathophysiology is unavailable.

Given that SUC was designed to detect *E. coli*, its results affect broad epidemiological statements concerning UTI. For example, it is commonly stated that *E. coli* accounts for 80–95% of all uncomplicated UTIs [1, 18]. Indeed, in the Price et al. study (Fig. 1), we showed that most (71%) of the SUC-identified pathogens were *E. coli* (39 out of 55). However, the biases of SUC showed through; it identified only 24% (16 out of 67) of the non-*E. coli* uropathogens in this group of women [14]. Furthermore, although EQUC detected *E coli* in 57% (43 out of 75) of women in the UTI cohort, rarely (8 out of 43) was *E. coli* the only microorganism detected in these samples [14]. Most patients with *E. coli* also had other species (35 out of 43), and frequently these additional species were uropathogens (24 out of 35). Similarly, Wolfe et al. detailed a case study of a woman with a positive SUC of $>10^5$ CFU/mL of *E. coli*. However, 16S rRNA sequencing showed that sequence reads of *Actinobaculum* and *Aerococcus* far exceeded those of *E. coli* [3]. Traditionally, bacteriuria caused by multiple species (i.e., a polymicrobial UTI) has been identified at a higher incidence in the elderly and children under the age of 10 years [22]. These tend to co-occur with multi-species bacteremia [22]. The new data described above suggest that polymicrobial UTI might be both common and frequently overlooked.

The presence of polymicrobial UTIs has been greatly underestimated because the presence of multiple colony morphologies on an SUC typically prompts clinical laboratories to dismiss these as "contamination" [20]. This common practice, combined with the clear screening inadequacies of SUC, has resulted in a flawed, one-species perspective of UTIs. Although it is unlikely that all bacteria associated with a polymicrobial UTI are causative agents of the patient's symptoms, this has not been explicitly tested. It is possible that a bacterial dysbiosis is present. Bacterial dysbioses have been associated with several other health disparities such as bacterial vaginosis (BV) in the vagina [23], and inflammatory bowel disease (IBD) in the gut. Whether urinary dysbiosis can cause UTI symptoms or results in colonization of the UTI causative organism(s) requires further study.

The limitations of a CFU threshold-based diagnosis

Current research calls into question the utility of thresholds in UTI diagnosis and treatment. With the incorporation of SUC in the 1950s, physicians have relied on a threshold of $\geq 10^5$ CFU/mL of urine to distinguish between significant bacteriuria and bacterial contamination. This threshold was set to identify women with pyelonephritis [10]. That diagnostic threshold was subsequently applied to women with acute cystitis. Yet, multiple investigators have demonstrated that 30–50% of women do not meet this threshold, despite symptoms of dysuria and urgency and frequency of urination [24]. Numerous recommendations have since been made to alter this threshold [14, 15, 24]. Stamm demonstrated that for women with acute cystitis, use of $\geq 10^5$ CFU/mL resulted in high specificity, but low sensitivity in detecting bacteriuria, whereas a lower threshold, such as $\geq 10^2$ CFU/mL, had a much higher sensitivity [11]. UTI symptoms and pyuria often persist when bacteriuria at $<10^5$ CFU/mL is left untreated [24] and, in catheterized patients, the presence of bacteria between 10^2 and 10^4 CFU/mL is reported to increase to $\geq 10^5$ CFU/mL if left untreated [12]. All this work, however, predated the newly discovered urinary microbiota.

In a population of women seeking urogynecological care (Fig. 1), we found that a single overall threshold did

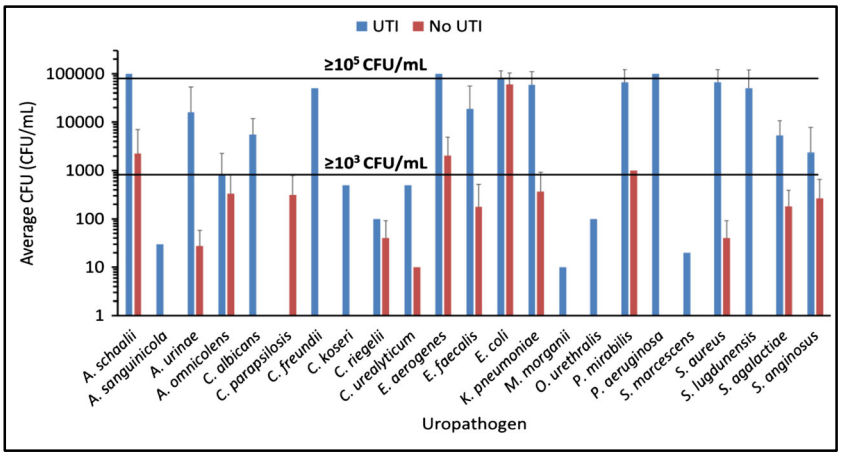

Fig. 2 Average CFU/mL of common and emerging uropathogens [20]

not distinguish between women who self-reported UTI and those who did not. EQUC demonstrated that significant differences in CFU/mL exist between uropathogens identified in women and that the mean CFU/mL for *E. coli* was near 10^5 CFU/mL for both patient cohorts (Fig. 2) [14]. Collectively, these data suggest that not only is the $\geq 10^5$ CFU/mL threshold insufficient to detect most uropathogens, but lowering this threshold does not solve the problem either, and may lead to unnecessary treatment. Importantly, the presence of uropathogens at levels that exceed the threshold, as with *E. coli*, does not necessarily result in symptoms.

Figure 3 shows a schematic of the uropathogen composition and patient symptom resolution of the women in the self-report UTI cohort, grouped by CFU threshold. Ninety-two percent of women (69 out of 75) had a common or emerging uropathogen present. Forty-two percent of these women had a polymicrobial infection (defined

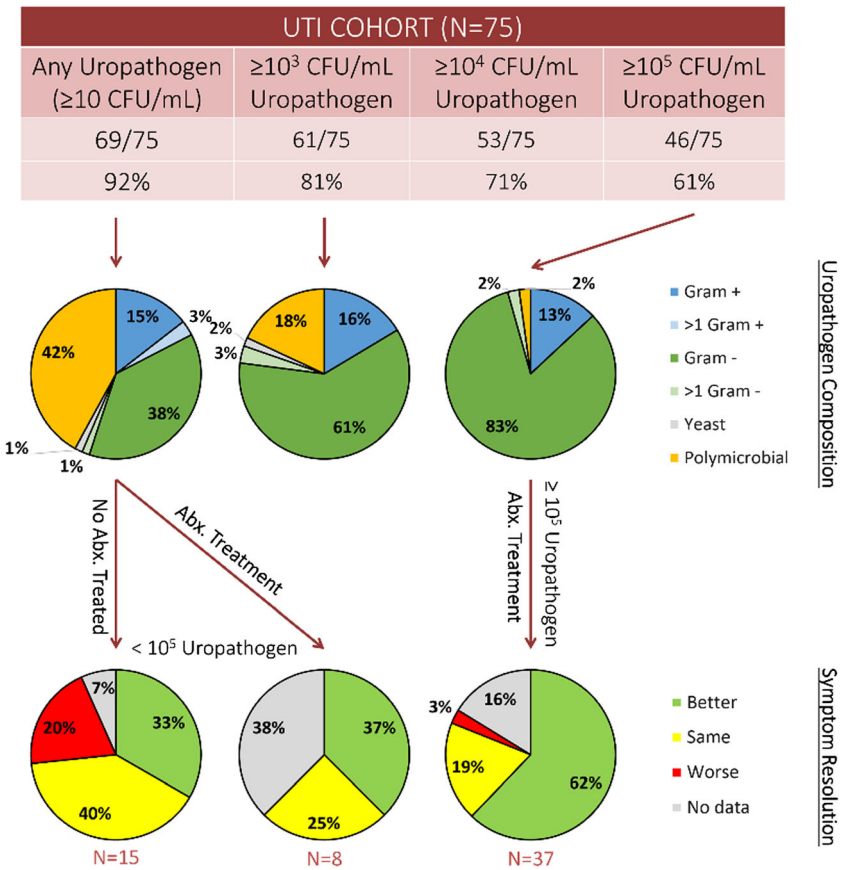

Fig. 3 Schematic of the uropathogen composition and patient symptom resolution of the women in the self-report UTI cohort [20]. *Abx.* antibiotics

here as at least one Gram-positive and one Gram-negative bacterium), whereas 38% had a single Gram-negative bacterium present. Uropathogens were present at $\geq 10^5$ CFU/mL in only 61% of women (46 out of 75). Of the women with at least one uropathogen present at $\geq 10^5$ CFU/mL, 83% had a single Gram-negative bacterium present. Thirty-seven out of 46 of these women were treated with antibiotics and 1 week later, 62% reported feeling "better." 23 women had uropathogens present at $<10^5$ CFU/mL. Fifteen of these 23 women were not given antibiotics, and 1 week later, only 33% reported feeling "better." This finding is certainly possible and entirely consistent with our thoughts about urinary dysbioses. Like other human microbial niches, we expect an individual to be able to restore a dysbiotic niche—i.e., some women may be able to resolve "UTI" without antibiotic treatment. Yet, these data demonstrate that use of the $\geq 10^5$CFU/mL threshold for UTI diagnosis and treatment is not appropriate for all women; specifically, those with polymicrobial or Gram-positive bacterial infections. Importantly, by not treating these patients, their clinical outcome is suboptimal (i.e., fewer reports feeling "better").

Next steps

Research efforts to improve diagnosis are expanding as the limitations of SUC become more apparent. Clinical laboratories can immediately incorporate the EQUC technique; however, clinicians may be challenged to interpret EQUC findings. Clinical trials to develop treatment algorithms based on EQUC findings are needed to promote good antibiotic stewardship to safeguard the overall health and well-being of patients being treated for UTI.

In recent years, other nonculture-based screening assays have been and are being developed. Immunology-based diagnostics, such as the RapidBac, rely on antibody-based detection of common uropathogens at $\geq 10^3$ CFU/mL with high specificity and sensitivity [25]. PCR assays, such as SeptiFast, have been assessed for use on urine samples with mild success [25]. And, several kits using fluorescence in situ hybridization (FISH) previously developed for blood and other specimens are being considered for use on urine for identification of pathogens [25].

Ongoing research will help to evaluate new treatment algorithms that incorporate the presence of previously undetected bacteria in UTI patients. Meanwhile, the role of the SUC must move away from the "gold standard" status. We believe that we will transition away from SUC to more sensitive testing that balances detection of organisms with appropriate therapies, designed to restore and maintain healthy microbial communities.

Conclusion

Should we think differently about UTI? Absolutely. We are entering a new age in UTI diagnosis and treatment. No longer can we define a UTI microbiologically as uropathogens invading a sterile environment. We must now acknowledge that previously ignored populations of bacteria are contributing to urinary health, both positively and negatively. How these findings ultimately change our precise definition of UTI remains to be seen, but will certainly be profound.

The status quo clearly needs to change so that patients benefit from this updated understanding of UTI. Improved treatment algorithms should be able to offer a spectrum of treatment with clear goals that reduce bothersome symptoms, the risk of serious infections and systemic illness, and unwanted collateral effects of UTI therapy. With appropriate adoption of evidence-based research, patients should benefit from more precise diagnosis and targeted treatment with limited deleterious collateral effects. Future research should advance our understanding of the role of the urinary microbiota in the context of both health and disease, and among women of differing demographics.

As with every major transition in clinical care, old habits are slow to fade away and adjustments will be needed as we enter this new era of UTI care. Clinicians need to learn new methods of interpretation of UTI testing, such as EQUC. No single study will provide guidance for all clinical situations and clinical judgment will remain a valued tool in patient care. Although stepping away from long-held clinical patterns of care takes time, our patients deserve better and it is time to improve care for these patients.

Acknowledgements We would like to thank the members of the Loyola Urinary Education and Research Collaborative for their contributions to the work described.

Funding AJW, LB, and ERM have been supported by NIH grants 2 U10 HD41250, U01 DK58229, R21 DK097435, R56 DK104718, P20 DK108268, R01 DK104718, a translational grant from the Falk Foundation and by RFC LU206998 from Loyola University Chicago. AJW has received funding for Investigator Initiated Studies VESI-12D01 and MYRB-15A01 from Astellas Scientific and Medical Affairs Inc. ERM has received funding from an Investigator Initiated Study (VESI-12D01) from Astellas Scientific and Medical Affairs Inc. Our funding sources have played no role in the design or conduct of our studies; collection, management, analysis, and interpretation of our data; or in the preparation, review, or approval of this or any other manuscript.

Compliance with ethical standards

Conflicts of interest Dr Brubaker has received editorial honoraria from UpToDate and Female Pelvic Medicine and Reconstructive Surgery. Dr Wolfe has received research support from Astellas Scientific and Medical Affairs. for urinary microbiome research. Dr Mueller has received honoraria from UpToDate and honoraria and research support from Astellas Scientific and Medical Affairs. Drs Brubaker and Wolfe have received

funding from the NIH and Loyola University Chicago for urinary microbiome research (see funding acknowledgements). Travis Price, Evann Hilt, and Tanaka Dune have no conflicts of interest.

References

1. Hooton TM. Clinical practice. Uncomplicated urinary tract infection. N Engl J Med. 2012;366(11):1028–37.
2. Finucane TE. "Urinary tract infection"—requiem for a heavyweight. J Am Geriatr Soc. 2017;65(8):1650–5.
3. Wolfe AJ, Toh E, Shibata N, Rong R, Kenton K, Fitzgerald M, et al. Evidence of uncultivated bacteria in the adult female bladder. J Clin Microbiol. 2012;50(4):1376–83.
4. Fouts DE, Pieper R, Szpakowski S, Pohl H, Knoblach S, Suh MJ, et al. Integrated next-generation sequencing of 16S rDNA and metaproteomics differentiate the healthy urine microbiome from asymptomatic bacteriuria in neuropathic bladder associated with spinal cord injury. J Transl Med. 2012;10:174.
5. Pearce MM, Hilt EE, Rosenfeld AB, Zilliox MJ, Thomas-White K, Fok C, et al. The female urinary microbiome: a comparison of women with and without urgency urinary incontinence. MBio. 2014;5(4):e01283–14.
6. Thomas-White KJ, Hilt EE, Fok C, et al. Incontinence medication response relates to the female urinary microbiota. Int Urogynecol J 2015;27(5):723–33.
7. Khasriya R, Sathiananthamoorthy S, Ismail S, Kelsey M, Wilson M, Rohn JL, et al. Spectrum of bacterial colonization associated with urothelial cells from patients with chronic lower urinary tract symptoms. J Clin Microbiol. 2013;51(7):2054–62.
8. Hilt EE, McKinley K, Pearce MM, Rosenfeld AB, Zilliox MJ, Mueller ER, et al. Urine is not sterile: use of enhanced urine culture techniques to detect resident bacterial flora in the adult female bladder. J Clin Microbiol. 2014;52(3):871–6.
9. Procop GW, Church DL, Hall GS, Janda WM, Koneman EE, Schreckenberger P, Woods GL. Koneman's color atlas and textbook of diagnostic microbiology. 7th edn. Philadelphia: Wolters Kluwer Health; 2017.
10. Kass EH. Asymptomatic infections of the urinary tract. Trans Assoc Am Phys. 1956;69:56–64.
11. Stamm WE, Counts GW, Running KR, Fihn S, Turck M, Holmes KK. Diagnosis of coliform infection in acutely dysuric women. N Engl J Med. 1982;307(8):463–8.
12. Stark RP, Maki DG. Bacteriuria in the catheterized patient. What quantitative level of bacteriuria is relevant? N Engl J Med. 1984;311(9):560–4.
13. Hooton TM, Roberts PL, Cox ME, Stapleton AE. Voided midstream urine culture and acute cystitis in premenopausal women. N Engl J Med. 2013;369(20):1883–91.
14. Price TK, Dune T, Hilt EE, et al. The clinical urine culture: enhanced techniques improve detection of clinically relevant microorganisms. J Clin Microbiol 2016;54(5):1216–22.
15. Clayson D, Wild D, Doll H, Keating K, Gondek K. Validation of a patient-administered questionnaire to measure the severity and bothersomeness of lower urinary tract symptoms in uncomplicated urinary tract infection (UTI): the UTI symptom assessment questionnaire. BJU Int. 2005;96(3):350–9.
16. Deville WL, Yzermans JC, van Duijn NP, Bezemer PD, van der Windt DA, Bouter LM. The urine dipstick test useful to rule out infections. A meta-analysis of the accuracy. BMC Urol. 2004;4:4.
17. Al Majid F, Buba F. The predictive and discriminant values of urine nitrites in urinary tract infection. Biomed Res. 2010;21:297–9.
18. Foxman B. Urinary tract infection syndromes: occurrence, recurrence, bacteriology, risk factors, and disease burden. Infect Dis Clin N Am. 2014;28(1):1–13.
19. Ronald A. The etiology of urinary tract infection: traditional and emerging pathogens. Am J Med. 2002;113 [Suppl 1A]:14s–9s.
20. Kline KA, Lewis AL. Gram-positive uropathogens, polymicrobial urinary tract infection, and the emerging microbiota of the urinary tract. Microbiol Spectr 2016;4(2). https://doi.org/10.1128/microbiolspec.UTI-0012-2012.
21. Le Brun C, Robert S, Bruyere F, Lanotte P. Emerging uropathogens: point for urologists and biologists. Prog Urol. 2015;25(7):363–9.
22. Siegman-Igra Y, Kulka T, Schwartz D, Konforti N. The significance of polymicrobial growth in urine: contamination or true infection. Scand J Infect Dis. 1993;25(1):85–91.
23. Holmes K, Sparling P, Stamm WE, Piot P, Wasserheit J, Corey L, Cohen M. Sexually transmitted diseases. 4th edn. New York: McGraw-Hill; 2007.
24. Stamm WE. Quantitative urine cultures revisited. Eur J Clin Microbiol. 1984;3(4):279–81.
25. Davenport M, Mach KE, Shortliffe LMD, Banaei N, Wang TH, Liao JC. New and developing diagnostic technologies for urinary tract infections. Nat Rev Urol. 2017;14(5):296–310.