Beyond the cloudiness in urinary tract infection: definitions, diagnostics, and strategies for prevention

Bilsen, M.P.

Chapter 1

General introduction

Epidemiology and burden

Urinary tract infection (UTI) is one of the most commonly encountered bacterial infections worldwide. Current estimates suggest that UTI affects over 400 million people annually, which is likely to be an underestimate of the actual global incidence due to underreporting, particularly in developing countries. [1] In the United States, UTI accounted for 8.6 million ambulatory care visits in 2007, with the majority being primary care office visits (59%) and emergency department visits (23%). [2] Similarly, in the Netherlands, UTI is the most common reason for a primary care consultation, with 149 office visits per 1000 patients (translating to 2.6 million office visits) in 2022 alone. [3]

UTI incidence varies by biological sex and age. Premenopausal women are disproportionately affected, with an incidence as high as 0.7 per person-year in a cohort of sexually-active university students. [4] A second peak occurs after the age of 65 years, when incidence increases with advancing age for both men and women, and is highest in women residing in long-term care facilities (LTCF). [5, 6] Predictive factors for UTI in this population include cognitive and functional impairment, previous UTI and urinary incontinence. [6] Among older women, recurrence rates are high. In a cohort study evaluating one-year recurrence rates in 180 women after an index episode, postmenopausal women had higher recurrence rates compared with premenopausal women (53% versus 36%). [7] Even higher one-year recurrence rates (69% and 79%) were found in a larger randomised trial comparing prophylactic strategies in postmenopausal women. [8]

The high incidence and recurrence rate of UTI place a significant socioeconomic burden on society. Women with recent or recurrent UTI consistently demonstrate reduced quality of life scores across both mental and physical domains, with impairments in activities such as sleep, exercise and sexual intercourse. [9, 10] Beyond direct medical expenses related to doctor's visits, laboratory testing and treatment, there are potential indirect costs if symptoms prevent patients from carrying out work-related tasks. Older adults significantly contribute to excessive healthcare costs, as UTI is the second most common suspected infection requiring hospitalisation in this population. [11] With increasing life expectancy the overall burden of UTI is expected to rise substantially.

Spectrum of disease and definitions

UTI is an umbrella term referring to a wide range of clinical phenotypes that differ in terms of site of infection, duration and severity of symptoms and signs. As highlighted in the previous paragraph, UTI occurs in both men and women of all age groups, and each population is characterised by different risk factors. This variety in clinical phenotypes and patient populations is reflected in the various specialties that encounter patients with UTI, including family medicine, emergency medicine, internal medicine, geriatric medicine, infectious diseases, microbiology, urology, gynaecology, and paediatrics.

Acute cystitis refers to a UTI presumed to be confined to the bladder. This phenotype is predominantly observed in women, possibly due to the shorter distance from the urethra to the perineum. Women typically present with new-onset lower urinary tract symptoms, such as dysuria, frequency, urgency and suprapubic pain, while signs of systemic illness, such as fever and rigors, are absent. Symptoms are self-limiting in the majority of patients (although antimicrobial treatment is often initiated in clinical practice) and progression to upper urinary tract infection is rare. [12, 13]

In men, prostatic involvement is common and may occur through bacterial migration from the urethra, intraprostatic urinary reflux, or from direct inoculation following urogenital instrumentation or transrectal biopsy. [14] In addition to urogenital symptoms, men with acute bacterial prostatitis generally present with systemic signs and symptoms. In a randomised trial evaluating the optimal treatment duration of acute bacterial prostatitis, 17% of the included participants had bacteraemia. [15] Approximately 10% of men with acute bacterial prostatitis develop chronic bacterial prostatitis, which tends to recur despite prolonged antimicrobial treatment. [16]

Acute pyelonephritis indicates an upper urinary tract infection involving the renal pelvis and kidney. Population-based studies show that acute pyelonephritis occurs more frequently in women than in men (annual rate of 15 cases per 10.000 women when combining in- and outpatients) with a notable peak in women aged 15 – 35 years, and a second, gradually increasing incidence after 65 years. [17] Acute pyelonephritis typically manifests with systemic signs and symptoms, flank pain and/or costovertebral angle tenderness, although clinical presentations can vary. [18] Despite high rates of bacteraemia (25-40%) mortality is generally low, with

exceptions for older hospitalised patients, in whom mortality rates can exceed 30%. [19–22]

At the end of the severity spectrum lies urosepsis, defined as a life-threatening organ dysfunction caused by a dysregulated host response to UTI. Surviving Sepsis Campaign data show that urosepsis is the second most common cause of septic shock, second only to a pulmonary source. [23] Despite antimicrobial therapy and supportive care, septic shock has a high mortality rate (32%).

Clearly, UTI is not a single type of infection, and as such it has proven difficult to come up with a single definition. For instance, the Centers for Disease Control and Prevention [24], the European Medicines Agency [25] and the U.S. Food and Drug Administration [26, 27] have all proposed definitions with different criteria and interpretations. The different types of UTI are perhaps more aptly described as having family resemblances, a concept first described by early 20th century German philosopher Ludwig Wittgenstein in his book *Philosophical Investigations*. He argues that categories and concepts are not defined by a single set of essential characteristics but rather by a network of overlapping similarities among various members within a category, e.g. like a family sharing traits. While a single definition is not always required in clinical practice, it is crucial in research. The absence of a research definition, also referred to as a reference standard, introduces bias into estimates of diagnostic accuracy and efficacy, affecting the internal validity of a study. [28] Additionally, if different definitions are used across studies, results cannot be readily compared, compromising the external validity of a study.

Pathophysiology: host versus pathogen

Uropathogenic *Escherichia coli* (UPEC) is by far the most common pathogen causing UTI. [29] Other pathogens include *Klebsiella pneumoniae*, *Proteus mirabilis*, and *Staphylococcus saprophyticus*. Enterococci and group B streptococci are frequently isolated from midstream urine, but rarely from urine obtained through single in-out catheterisation, suggesting that these pathogens do not typically cause UTI. [30]

Infection of the bladder mostly occurs through an exogenous route. Uropathogens residing in the gut first colonise the (peri)urethra and subsequently migrate into the bladder. Most research on understanding host-pathogen interactions in UTI has concentrated on infections caused by UPEC. [29] To invade the bladder,

UPEC uses adhesins located on the tip of fimbriae or on the bacterial surface to attach to uroplakins and integrins that coat the most outer layer of the urothelium, i.e. umbrella cells. Recognition of lipopolysaccharide (present on the outer membrane of Gram-negative bacteria) through Toll-like receptors on umbrella cells induces a rapid innate immune response via transcription of pro-inflammatory cytokines and chemokines. While UPEC in the bladder lumen is targeted by recruited neutrophils, antimicrobial peptides and iron-sequestering proteins, internalised UPEC is able to subvert host defences and form intracellular bacterial communities (IBCs) through multiplication. These IBCs are able to survive in the bladder environment due to additional virulence factors such as the toxin α-haemolysin, expediting nutrient acquisition via host cell lysis, and siderophores which facilitate iron uptake. Upon exfoliation of the urothelium due to inflammation and α-haemolysin, bacteria can disperse and invade neighbouring cells. Exfoliation also exposes deeper layers of the bladder epithelium where UPEC can establish quiescent intracellular reservoirs (QIRs), which can remain viable for months and may contribute to recurrences. [29] Although the pathophysiological basis for recurrent UTI in humans remains poorly understood, recent murine studies show that UTI leads to differential bladder tissue remodelling, depending on disease outcome, that affects susceptibility to subsequent UTI episodes. [31, 32] Less is known about the role of adaptive immunity in UTI. Mouse models of bladder infection show that although an adaptive immune response develops, the bacterial burden is only marginally reduced and UTI frequently recurs. [33] Secretory IgA can inhibit adhesion of UPEC to epithelial cells, and in children with acute cystitis, secretory IgA levels in urine are elevated. [34] Recently, sublingual vaccination with a suspension of whole-cell heat-inactivated *E. coli, Klebsiella pneumoniae, Proteus vulgaris,* and *Enterococcus faecalis* has shown remarkable efficacy in a placebo-controlled trial including pre- and postmenopausal women with recurrent UTI. [35] While the exact mechanism of action has not been fully elucidated, both enhanced innate and adaptive (cellular and humoral) immunity seem to play a role. [36]

Currently used diagnostics

The diagnostic approach of UTI differs per clinical presentation (typical or atypical lower urinary tract symptoms, presence of systemic signs), setting (primary care, outpatient clinic, emergency department), population (age group, biological sex, underlying risk factors) and country. Some clinicians do not perform additional

testing in women with classic lower urinary tract symptoms and absence of systemic signs, as the a priori probability of acute cystitis is high. [37] However, lower urinary tract symptoms may be caused by other conditions such as urethritis (*Chlamydia trachomatis, Neisseria gonorrhoeae*) and vaginitis (*Candida* spp., *Trichomonas vaginalis*). As such, additional testing for the presence of pyuria (i.e. leukocyturia) and bacteriuria is often performed to support the diagnosis of UTI.

In clinical practice, particularly in primary care, a urine dipstick is usually applied first, to screen for pyuria and bacteriuria. Among other analytes, the urine dipstick provides semi-quantitative results of leukocyte esterase (an enzyme produced by leukocytes in the urine) and indicates the presence or absence of nitrites. While a urine dipstick is inexpensive, easy to use and provides quick results, there are important drawbacks to note. Both leukocyte esterase and nitrite results may be false-negative due to the presence of other substances such as vitamin C. [38] Moreover, not all pathogens reduce urinary nitrates to nitrites (e.g. *Staphylococcus saprophyticus, Pseudomonas aeruginosa, Enterococcus* spp. do not). Furthermore, leukocyte esterase results correlate poorly with absolute degrees of pyuria. [39] Reported diagnostic accuracy estimates of leukocyte esterase and nitrites for diagnosing UTI vary greatly and are primarily determined by the population under investigation and the reference standard that is applied. In general, nitrites are less sensitive than specific, and leukocyte esterase is more sensitive than specific for UTI. [40-42]

Pyuria can be quantified in different ways. In the late 1960s Mabeck et al. [43] found that a leukocyte excretion rate of 400,000 per hour could distinguish UTI from asymptomatic women. This rate corresponds with a cut-off value of 10 leukocytes/mm^3 in unspun urine. [44] Nowadays, most hospitals quantify pyuria by direct or automated microscopy of (un)spun urine, generally after initial dipstick screening. Automated microscopy reduces variability in centrifugation and resuspension of urine and is more efficient than direct microscopy. [45] In recent years, an increasing number of laboratories are adopting urine flow cytometry for quantification of pyuria. Urine flow cytometers classify and quantify cells in the urine by analysing scattered light emitted by cells passing through a laser beam. While automated microscopy and urine flow cytometry have relatively short turnaround times and a high capacity, they are costly and not available to every clinician. Moreover, reference values for 'significant' pyuria vary in the literature and depend on preanalytic steps in the laboratory, quantification methods and the

studied population. Finally, pyuria may be caused by conditions other than UTI, including interstitial nephritis, urolithiasis and urological malignancies.

Bacteriuria can be determined by spreading the urine onto a plate containing a culture medium and incubating it under various conditions. This approach allows for pathogen identification and quantification, which usually takes 18-30 hours, and antimicrobial susceptibility testing, which may take another 24-48 hours, depending on the use of manual or automated methods. [46] Despite providing valuable information, long turnaround times are an important drawback of urine cultures. Additionally, there is an ongoing debate about the optimal threshold for significant bacteriuria (expressed in colony-forming units (CFU)/mL). While the traditional cut-off value of 10^5 CFU/mL is still applied by some laboratories to avoid misclassification of contamination as UTI, several studies have shown that colony-counts as low as 10^2 CFU/mL in midstream urine are indicative of true bladder bacteriuria (determined by suprapubic aspiration or single in-out catheterisation), at least in symptomatic women with *E. coli* as the causative pathogen. [30, 47] Be that as it may, urine cultures merely indicate bacteriuria, which does not necessarily equate to UTI.

Challenges in older women

In older women, diagnosing UTI presents specific challenges for several reasons. Firstly, symptom assessment is hampered by a higher prevalence of cognitive impairment and indwelling catheters in the older population. The global prevalence of dementia was estimated to be 57.4 million cases in 2019, with a female-to-male ratio of 1.69, and a predicted increase to 152.8 million cases in 2050, mainly driven by population ageing and population growth. [48] Secondly, chronic lower urinary tract symptoms, such as urgency, frequency and urinary incontinence, are common in older women and are difficult to distinguish from non-infectious causes, such as genitourinary syndrome of menopause, and overactive bladder. [49] Most importantly, 20% of community-dwelling and 50% of institutionalised older women have asymptomatic bacteriuria (ASB), defined as the presence of one or more uropathogens ≥ 10^5 CFU/mL in the absence of signs or symptoms attributable to UTI. [50-52] While the pathophysiological basis of ASB has not been completely elucidated, it is thought to arise from an interplay between host factors (e.g. reduced Toll-like receptor 4 expression [53]) and pathogen-specific factors (e.g. reduced adhesive capability of certain *E. coli* strains [54]).

Over 90% of older women with ASB have concomitant pyuria [55]. Consequently, the specificity of both pyuria and bacteriuria for UTI is low in this population, and it can be difficult to distinguish UTI from ASB with current urine diagnostics. As such, inappropriate antimicrobial treatment of asymptomatic pyuria and bacteriuria is very common. Gupta et al. [56] showed that 25% of patients with asymptomatic pyuria on routine preoperative urinalysis (without urine cultures) were treated with antimicrobials, and that the degree of pyuria predicted prescribing of antimicrobials. Moreover, in a study performed in long-term care facility residents with advanced dementia, only 19% of suspected 'UTI' episodes that were treated with antimicrobials fulfilled minimum symptom criteria (suggesting most episodes were actually ASB). [57] In older patients with cognitive impairment, who have difficulty communicating their symptoms, it may be tempting for clinicians to ascribe non-specific symptoms such as confusion or falls to a UTI, especially in the presence of pyuria and bacteriuria. However, the evidence is growing that these non-specific symptoms do not reliably predict actual UTI. [58, 59] More likely, these symptoms indicate normal fluctuations in behaviour or have other causes, such as dehydration and drug-related side effects. Although antimicrobial treatment of ASB may result in short-term microbiological cure, it does not improve survival, nor influence the frequency of subsequent UTI episodes or chronic urinary incontinence in older women. [60-63] In fact, antimicrobial treatment of ASB can lead to adverse drug reactions, interactions and toxicity, which is particularly relevant in a population with high rates of polypharmacy. [62] Moreover, antimicrobial treatment confers an eightfold increased risk of developing *Clostridioides difficile* associated diarrhoea, and it leads to subsequent isolation of multidrug resistant organisms (MDROs) from the urine. [64, 65] Besides being judicious about urine testing in older women with ambiguous symptoms, new diagnostic modalities with the ability to distinguish UTI from ASB are urgently required.

Treatment and prophylaxis in an era of antimicrobial resistance

UTI is typically treated with a course of antimicrobials. The selection of an antimicrobial regimen is primarily dependent on the site of infection, i.e. whether an agent with tissue penetration is required. First-line oral antimicrobials for empirical treatment of UTI without systemic involvement (acute cystitis) include nitrofurantoin, fosfomycin, trimethoprim, and, in some countries, pivmecillinam.

The approach to empirical treatment of UTI with systemic involvement (acute pyelonephritis with or without urosepsis) generally depends on the severity of illness. While outpatients may be treated with oral ciprofloxacin or trimethoprim-sulfamethoxazole, critically ill patients are usually treated parenterally; Dutch guidelines recommend a second or third generation cephalosporin with the option of adding aminoglycosides pending culture results. [66] In patients with recurrent UTI and insufficient efficacy of behavioural modifications, such as increased hydration [67] or postcoital voiding, oral antimicrobial prophylaxis (either daily or postcoital) is often initiated. Continuous antimicrobial prophylaxis is effective in reducing recurrence rates, even in risk groups, such as patients using clean intermittent catheterisation due to urological or neurological comorbidities. [8, 68-70] Antimicrobial options include nitrofurantoin 50 mg or 100 mg daily, and trimethoprim 100 mg daily. However, the most important drawback of antimicrobial treatment and prophylaxis, already pointed out by Alexander Fleming in 1945 [71], is the development of antimicrobial resistance (AMR).

AMR is a rising threat to global health. In fact, based on predictive statistical models, an estimated 4.95 million deaths were associated with bacterial AMR in 2019 alone, with the highest burdens in resource-limited settings. [72] *E. coli* and *Klebsiella pneumoniae (K. pneumoniae)* were the two pathogens responsible for the most AMR-attributable deaths, both common causative pathogens of UTI. AMR surveillance data of 46 European countries, published by the European Centre for Disease Prevention and Control (ECDC) in 2022, showed highest resistance rates in southern and eastern regions, compared with northern and western regions. [73] For *E. coli*, 46% of countries reported ciprofloxacin resistance rates > 25% and 11% of countries reported third generation cephalosporin resistance rates > 50%. Carbapenem resistance was more frequently reported in *K. pneumoniae* than in *E. coli*; 32% of countries reported carbapenem resistance rates > 25%. Not surprisingly, surveillance data published by the World Health Organization (WHO) in 2021 showed a wide range of resistance rates for several antimicrobial classes for *E. coli* and *K. pneumoniae*, with low and middle income countries being disproportionally affected. [74] To support antimicrobial stewardship efforts and to identify antimicrobials with the highest priorities for surveillance of use, the WHO created the Access, Watch, Reserve (AWaRe) classification, in which antimicrobials are categorised into three groups based on the potential to induce and propagate resistance. [75] Multiple agents commonly used in the treatment of UTI (e.g. ciprofloxacin and ceftriaxone) are included in the 'Watch-group' due to

increasing resistance rates. Nosocomial UTI is not infrequently caused by one of the ESKAPE pathogens (i.e. *Enterococcus faecium, Staphylococcus aureus, Klebsiella pneumoniae, Acinetobacter baumannii, Pseudomonas aeruginosa* and *Enterobacter* spp.), which are often difficult to treat due to their resistance to multiple classes of antimicrobials. [76] Finally, as the high incidence and recurrence rates of UTI lead to significant antimicrobial consumption, UTI is an important driver of the silent epidemic that is AMR. It is evident that alternative strategies for the treatment and prevention of UTI need to be explored to reduce the global burden of AMR. In a randomised trial evaluating different management approaches in women presenting to the primary care office with suspected acute cystitis, symptom duration and severity were similar with delayed antimicrobial therapy compared with immediate antimicrobial therapy. [77] In another randomised trial comparing ibuprofen with fosfomycin (with respective placebo dummies in both groups) in women with acute cystitis, two thirds of women in the ibuprofen group recovered without any antibiotics, albeit with a somewhat higher overall symptom burden in the ibuprofen group. [12] Besides behavioural modifications, other non-antimicrobial prophylactic strategies for recurrent UTI include vaginal oestrogen for postmenopausal women, and methenamine hippurate. [78, 79] In one open-label trial showing non-inferiority of methenamine hippurate to oral antimicrobial prophylaxis, the proportion of participants demonstrating resistance to at least one antimicrobial in *E. coli* isolated from perineal swabs was higher in the oral antimicrobial prophylaxis group at 6-12 months. [79] As the gut is a known reservoir for uropathogenic bacteria, Worby et al. [80] collected monthly faecal samples of women with recurrent UTI and controls for metagenomic analysis, and found significantly lower gut microbial richness in women with recurrent UTI. Given that some studies have shown decreased gut microbial richness in patients with intestinal colonisation of MDRO, 'gut sparing' or 'gut restorative' interventions have the potential to reduce the frequency of UTI recurrences and decrease intestinal MDRO colonisation. However, efficacy data for these alternative modalities are sparse.

Outline of the thesis

The overall aim of this thesis is to address unmet needs in the definitions, diagnosis and treatment strategies in UTI, which can contribute to improving health outcomes for individual patients and reducing antimicrobial resistance. This thesis comprises three parts. The first part focuses on the definition of UTI

in research. To evaluate the heterogeneity of UTI definitions in recent studies, a systematic review was performed, which is described in **Chapter 2**. Given the high heterogeneity of study definitions and conflicting research guidelines, a Delphi consensus study involving an international, multidisciplinary panel of UTI experts was conducted to construct a reference standard for UTI research. This consensus study is reported in **Chapter 3**.

The second part of this thesis centres on diagnostic challenges of UTI in older women. **Chapter 4** describes a case-control study including older women with UTI and ASB, in which the diagnostic accuracy of two pyuria quantification methods (automated microscopy and urine flow cytometry) for UTI is determined, and an optimal pyuria threshold for older women is sought. Due to the limitations of current urine diagnostics in older women, the diagnostic accuracy of twelve novel urine biomarkers is evaluated in the same study population, which is reported in **Chapter 5**.

In the third and last part of this thesis alternative strategies for the treatment and prevention of UTI are explored. As stated in the previous paragraph, (systemic) antimicrobial prophylaxis and treatment have important drawbacks. Therefore, we performed a cohort study, described in **Chapter 6**, to assess the treatment satisfaction, long-term safety, and efficacy of a non-systemic antimicrobial prophylactic strategy, i.e. intravesical aminoglycoside instillations. As intestinal MDRO colonisation may precede invasive infection and facilitates spread within communities and hospitals, the efficacy of faecal microbiota transplantation for MDRO decolonisation was assessed in a systematic review in **Chapter 7**.

Chapter 8 provides a summary and a discussion of the results, resulting in a conclusion and views on possible further research.

References

1. Zeng Z, Zhan J, Zhang K, Chen H, Cheng S. Global, regional, and national burden of urinary tract infections from 1990 to 2019: an analysis of the global burden of disease study 2019. World J Urol **2022**; 40(3): 755-63.
2. Schappert SM, Rechtsteiner EA. Ambulatory medical care utilization estimates for 2007. Vital Health Stat 13 **2011**; (169): 1-38.
3. Cijfers huisartsen - Gezondheidsproblemen. Available at: https://www.nivel.nl/nl/nivel-zorgregistraties-eerste-lijn/cijfers-zorg-huisarts-huisartsenpost-paramedici/cijfers-zorg-huisartsen/gezondheidsproblemen. Accessed 1st of July 2023.
4. Hooton TM, Scholes D, Hughes JP, et al. A prospective study of risk factors for symptomatic urinary tract infection in young women. N Engl J Med **1996**; 335(7): 468-74.
5. Ahmed H, Farewell D, Jones HM, Francis NA, Paranjothy S, Butler CC. Incidence and antibiotic prescribing for clinically diagnosed urinary tract infection in older adults in UK primary care, 2004-2014. PLoS One **2018**; 13(1): e0190521.
6. Caljouw MA, den Elzen WP, Cools HJ, Gussekloo J. Predictive factors of urinary tract infections among the oldest old in the general population. A population-based prospective follow-up study. BMC Med **2011**; 9: 57.
7. Ikaheimo R, Siitonen A, Heiskanen T, et al. Recurrence of urinary tract infection in a primary care setting: analysis of a 1-year follow-up of 179 women. Clin Infect Dis **1996**; 22(1): 91-9.
8. Beerepoot MA, ter Riet G, Nys S, et al. Lactobacilli vs antibiotics to prevent urinary tract infections: a randomized, double-blind, noninferiority trial in postmenopausal women. Arch Intern Med **2012**; 172(9): 704-12.
9. Thompson J, Marijam A, Mitrani-Gold FS, Wright J, Joshi AV. Activity impairment, health-related quality of life, productivity, and self-reported resource use and associated costs of uncomplicated urinary tract infection among women in the United States. PLoS One **2023**; 18(2): e0277728.
10. Ennis SS, Guo H, Raman L, Tambyah PA, Chen SL, Tiong HY. Premenopausal women with recurrent urinary tract infections have lower quality of life. Int J Urol **2018**; 25(7): 684-9.
11. Curns AT, Holman RC, Sejvar JJ, Owings MF, Schonberger LB. Infectious disease hospitalizations among older adults in the United States from 1990 through 2002. Arch Intern Med **2005**; 165(21): 2514-20.
12. Gagyor I, Bleidorn J, Kochen MM, Schmiemann G, Wegscheider K, Hummers-Pradier E. Ibuprofen versus fosfomycin for uncomplicated urinary tract infection in women: randomised controlled trial. BMJ **2015**; 351: h6544.
13. Jansaker F, Li X, Vik I, Frimodt-Moller N, Knudsen JD, Sundquist K. The Risk of Pyelonephritis Following Uncomplicated Cystitis: A Nationwide Primary Healthcare Study. Antibiotics (Basel) **2022**; 11(12).
14. Mosharafa AA, Torky MH, El Said WM, Meshref A. Rising incidence of acute prostatitis following prostate biopsy: fluoroquinolone resistance and exposure is a significant risk factor. Urology **2011**; 78(3): 511-4.

15. Lafaurie M, Chevret S, Fontaine JP, et al. Antimicrobial for 7 or 14 Days for Febrile Urinary Tract Infection in Men: A Multicenter Noninferiority Double-Blind, Placebo-Controlled, Randomized Clinical Trial. Clin Infect Dis **2023**; 76(12): 2154-62.

16. Su ZT, Zenilman JM, Sfanos KS, Herati AS. Management of Chronic Bacterial Prostatitis. Curr Urol Rep **2020**; 21(7): 29.

17. Czaja CA, Scholes D, Hooton TM, Stamm WE. Population-based epidemiologic analysis of acute pyelonephritis. Clin Infect Dis **2007**; 45(3): 273-80.

18. Johnson JR, Russo TA. Acute Pyelonephritis in Adults. N Engl J Med **2018**; 378(1): 48-59.

19. Kim Y, Seo MR, Kim SJ, et al. Usefulness of Blood Cultures and Radiologic Imaging Studies in the Management of Patients with Community-Acquired Acute Pyelonephritis. Infect Chemother **2017**; 49(1): 22-30.

20. van Nieuwkoop C, van der Starre WE, Stalenhoef JE, et al. Treatment duration of febrile urinary tract infection: a pragmatic randomized, double-blind, placebo-controlled non-inferiority trial in men and women. BMC Med **2017**; 15(1): 70.

21. Esparcia A, Artero A, Eiros JM, et al. Influence of inadequate antimicrobial therapy on prognosis in elderly patients with severe urinary tract infections. Eur J Intern Med **2014**; 25(6): 523-7.

22. Tal S, Guller V, Levi S, et al. Profile and prognosis of febrile elderly patients with bacteremic urinary tract infection. J Infect **2005**; 50(4): 296-305.

23. Levy MM, Artigas A, Phillips GS, et al. Outcomes of the Surviving Sepsis Campaign in intensive care units in the USA and Europe: a prospective cohort study. Lancet Infect Dis **2012**; 12(12): 919-24.

24. Urinary Tract Infection (Catheter-Associated Urinary Tract Infection [CAUTI] and Non-Catheter-Associated Urinary Tract Infection [UTI]) Events. Available at: https://www.cdc.gov/nhsn/psc/uti/. Accessed 8th of June.

25. Evaluation of medicinal products indicated for treatment of bacterial infections. Available at: https://www.ema.europa.eu/en/evaluation-medicinal-products-indicated-treatment-bacterial-infections Accessed 8th of June.

26. Uncomplicated Urinary Tract Infections: Developing Drugs for Treatment Guidance for Industry. Available at: https://www.fda.gov/regulatory-information/search-fda-guidance-documents/uncomplicated-urinary-tract-infections-developing-drugs-treatment-guidance-industry Accessed 8th of June.

27. Complicated Urinary Tract Infections: Developing Drugs for Treatment. Available at: https://www.fda.gov/regulatory-information/search-fda-guidance-documents/complicated-urinary-tract-infections-developing-drugs-treatment Accessed 8th of June.

28. de Groot JA, Bossuyt PM, Reitsma JB, et al. Verification problems in diagnostic accuracy studies: consequences and solutions. BMJ **2011**; 343: d4770.

29. Flores-Mireles AL, Walker JN, Caparon M, Hultgren SJ. Urinary tract infections: epidemiology, mechanisms of infection and treatment options. Nat Rev Microbiol **2015**; 13(5): 269-84.

30. 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.

31. Hannan TJ, Totsika M, Mansfield KJ, Moore KH, Schembri MA, Hultgren SJ. Host-pathogen checkpoints and population bottlenecks in persistent and intracellular uropathogenic Escherichia coli bladder infection. FEMS Microbiol Rev **2012**; 36(3): 616-48.

32. Russell SK, Harrison JK, Olson BS, et al. Uropathogenic Escherichia coli infection-induced epithelial trained immunity impacts urinary tract disease outcome. Nat Microbiol **2023**; 8(5): 875-88.

33. Lacerda Mariano L, Ingersoll MA. The immune response to infection in the bladder. Nat Rev Urol **2020**; 17(8): 439-58.

34. Butler D, Ambite I, Wan MLY, Tran TH, Wullt B, Svanborg C. Immunomodulation therapy offers new molecular strategies to treat UTI. Nat Rev Urol **2022**; 19(7): 419-37.

35. Lorenzo-Gómez M-F, Foley S, Nickel JC, et al. Sublingual MV140 for Prevention of Recurrent Urinary Tract Infections. NEJM Evidence **2022**; 1(4): EVIDoa2100018.

36. Martin-Cruz L, Sevilla-Ortega C, Benito-Villalvilla C, et al. A Combination of Polybacterial MV140 and Candida albicans V132 as a Potential Novel Trained Immunity-Based Vaccine for Genitourinary Tract Infections. Front Immunol **2020**; 11: 612269.

37. Bent S, Nallamothu BK, Simel DL, Fihn SD, Saint S. Does this woman have an acute uncomplicated urinary tract infection? JAMA **2002**; 287(20): 2701-10.

38. Ko DH, Jeong TD, Kim S, et al. Influence of Vitamin C on Urine Dipstick Test Results. Ann Clin Lab Sci **2015**; 45(4): 391-5.

39. van den Broek D, Keularts IM, Wielders JP, Kraaijenhagen RJ. Benefits of the iQ200 automated urine microscopy analyser in routine urinalysis. Clin Chem Lab Med **2008**; 46(11): 1635-40.

40. Timmermans AE, Walter AEGM, Van Duijn NP, Timmerman CP. De diagnostische waarde van urineonderzoek in de huisartspraktijk. Huisarts Wet **1996**; 39(4): 165-8.

41. Christiaens TCM, De Meyere M, Derese A. Disappointing specificity of the leucocyte-esterase test for the diagnosis of urinary tract infection in general practice. Eur J Gen Pract **1998**; 4: 144-8.

42. Hooton TM, Roberts PL, Stapleton AE. Asymptomatic Bacteriuria and Pyuria in Premenopausal Women. Clin Infect Dis **2021**; 72(8): 1332-8.

43. Mabeck CE. Studies in urinary tract infections. IV. Urinary leucocyte excretion in bacteriuria. Acta Med Scand **1969**; 186(3): 193-8.

44. Stamm WE. Measurement of pyuria and its relation to bacteriuria. Am J Med **1983**; 75(1B): 53-8.

45. Oyaert M, Delanghe J. Progress in Automated Urinalysis. Ann Lab Med **2019**; 39(1): 15-22.

46. 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.

47. 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.

48. Collaborators GBDDF. Estimation of the global prevalence of dementia in 2019 and forecasted prevalence in 2050: an analysis for the Global Burden of Disease Study 2019. Lancet Public Health **2022**; 7(2): e105-e25.

49. Irwin DE, Milsom I, Hunskaar S, et al. Population-based survey of urinary incontinence, overactive bladder, and other lower urinary tract symptoms in five countries: results of the EPIC study. Eur Urol **2006**; 50(6): 1306-14; discussion 14-5.

50. Nicolle LE, Gupta K, Bradley SF, et al. Clinical Practice Guideline for the Management of Asymptomatic Bacteriuria: 2019 Update by the Infectious Diseases Society of America. Clin Infect Dis **2019**; 68(10): 1611-5.

51. Rodhe N, Lofgren S, Matussek A, et al. Asymptomatic bacteriuria in the elderly: high prevalence and high turnover of strains. Scand J Infect Dis **2008**; 40(10): 804-10.

52. Ouslander JG, Schapira M, Fingold S, Schnelle J. Accuracy of rapid urine screening tests among incontinent nursing home residents with asymptomatic bacteriuria. J Am Geriatr Soc **1995**; 43(7): 772-5.

53. Ragnarsdottir B, Samuelsson M, Gustafsson MC, Leijonhufvud I, Karpman D, Svanborg C. Reduced toll-like receptor 4 expression in children with asymptomatic bacteriuria. J Infect Dis **2007**; 196(3): 475-84.

54. Roos V, Schembri MA, Ulett GC, Klemm P. Asymptomatic bacteriuria Escherichia coli strain 83972 carries mutations in the foc locus and is unable to express F1C fimbriae. Microbiology (Reading) **2006**; 152(Pt 6): 1799-806.

55. Boscia JA, Abrutyn E, Levison ME, Pitsakis PG, Kaye D. Pyuria and asymptomatic bacteriuria in elderly ambulatory women. Ann Intern Med **1989**; 110(5): 404-5.

56. Gupta K, O'Brien W, Gallegos-Salazar J, Strymish J, Branch-Elliman W. How Testing Drives Treatment in Asymptomatic Patients: Level of Pyuria Directly Predicts Probability of Antimicrobial Prescribing. Clin Infect Dis **2020**; 71(3): 614-21.

57. Mitchell SL, Shaffer ML, Loeb MB, et al. Infection management and multidrug-resistant organisms in nursing home residents with advanced dementia. JAMA Intern Med **2014**; 174(10): 1660-7.

58. Sundvall PD, Elm M, Ulleryd P, et al. Interleukin-6 concentrations in the urine and dipstick analyses were related to bacteriuria but not symptoms in the elderly: a cross sectional study of 421 nursing home residents. BMC Geriatr **2014**; 14: 88.

59. Sundvall PD, Ulleryd P, Gunnarsson RK. Urine culture doubtful in determining etiology of diffuse symptoms among elderly individuals: a cross-sectional study of 32 nursing homes. BMC Fam Pract **2011**; 12: 36.

60. Abrutyn E, Mossey J, Berlin JA, et al. Does asymptomatic bacteriuria predict mortality and does antimicrobial treatment reduce mortality in elderly ambulatory women? Ann Intern Med **1994**; 120(10): 827-33.

61. Boscia JA, Kobasa WD, Knight RA, Abrutyn E, Levison ME, Kaye D. Therapy vs no therapy for bacteriuria in elderly ambulatory nonhospitalized women. JAMA **1987**; 257(8): 1067-71.

62. Nicolle LE, Mayhew WJ, Bryan L. Prospective randomized comparison of therapy and no therapy for asymptomatic bacteriuria in institutionalized elderly women. Am J Med **1987**; 83(1): 27-33.

63. Ouslander JG, Schapira M, Schnelle JF, et al. Does eradicating bacteriuria affect the severity of chronic urinary incontinence in nursing home residents? Ann Intern Med **1995**; 122(10): 749-54.

64. Das R, Towle V, Van Ness PH, Juthani-Mehta M. Adverse outcomes in nursing home residents with increased episodes of observed bacteriuria. Infect Control Hosp Epidemiol **2011**; 32(1): 84-6.

65. Rotjanapan P, Dosa D, Thomas KS. Potentially inappropriate treatment of urinary tract infections in two Rhode Island nursing homes. Arch Intern Med **2011**; 171(5): 438-43.

66. Optimization of the antibiotic policy in the Netherlands: SWAB guidelines for antimicrobial therapy of urinary tract infections in adults Available at: https://swab.nl/nl/urineweginfecties. Accessed 21st of November 2023.

67. Hooton TM, Vecchio M, Iroz A, et al. Effect of Increased Daily Water Intake in Premenopausal Women With Recurrent Urinary Tract Infections: A Randomized Clinical Trial. JAMA Intern Med **2018**; 178(11): 1509-15.

68. Beerepoot MA, ter Riet G, Nys S, et al. Cranberries vs antibiotics to prevent urinary tract infections: a randomized double-blind noninferiority trial in premenopausal women. Arch Intern Med **2011**; 171(14): 1270-8.

69. Albert X, Huertas I, Pereiro, II, Sanfelix J, Gosalbes V, Perrota C. Antibiotics for preventing recurrent urinary tract infection in non-pregnant women. Cochrane Database Syst Rev **2004**; 2004(3): CD001209.

70. Fisher H, Oluboyede Y, Chadwick T, et al. Continuous low-dose antibiotic prophylaxis for adults with repeated urinary tract infections (AnTIC): a randomised, open-label trial. Lancet Infect Dis **2018**; 18(9): 957-68.

71. Rosenblatt-Farrell N. The landscape of antibiotic resistance. Environ Health Perspect **2009**; 117(6): A244-50.

72. Antimicrobial Resistance C. Global burden of bacterial antimicrobial resistance in 2019: a systematic analysis. Lancet **2022**; 399(10325): 629-55.

73. Surveillance of antimicrobial resistance in Europe. Available at: https://www.ecdc.europa.eu/en/publications-data/surveillance-antimicrobial-resistance-europe-2022-data. Accessed 21st of November 2023.

74. Global Antimicrobial Resistance and Use Surveillance System (GLASS) Report. Available at: https://www.who.int/publications/i/item/9789240027336. Accessed 21st of November 2023.

75. WHO Access, Watch, Reserve (AWaRe) classification of antibiotics for evaluation and monitoring of use, 2021. Geneva: World Health Organization; 2021 (WHO/MHP/HPS/EML/2021.04). Available at: https://www.who.int/publications/i/item/2021-aware-classification. Accessed 25th of November 2023.

76. De Oliveira DMP, Forde BM, Kidd TJ, et al. Antimicrobial Resistance in ESKAPE Pathogens. Clin Microbiol Rev **2020**; 33(3).

77. Little P, Moore MV, Turner S, et al. Effectiveness of five different approaches in management of urinary tract infection: randomised controlled trial. BMJ **2010**; 340: c199.

78. Raz R, Stamm WE. A controlled trial of intravaginal estriol in postmenopausal women with recurrent urinary tract infections. N Engl J Med **1993**; 329(11): 753-6.

79. Harding C, Mossop H, Homer T, et al. Alternative to prophylactic antibiotics for the treatment of recurrent urinary tract infections in women: multicentre, open label, randomised, non-inferiority trial. BMJ **2022**; 376: e068229.

80. Worby CJ, Schreiber HLt, Straub TJ, et al. Longitudinal multi-omics analyses link gut microbiome dysbiosis with recurrent urinary tract infections in women. Nat Microbiol **2022**; 7(5): 630-9.