

Tackling antimicrobial resistance in lower urinary tract infections: treatment options

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Abstract

Urinary tract infections (UTIs) are among the most common infectious diseases occurring in either the community or healthcare settings. A wide variety of bacteria are responsible for causing UTIs, however extra-intestinal pathogenic *E. coli* or ExPEC) remains the most common etiological agent. Since 2000, resistance to antibiotics emerged globally among ExPEC and is causing delays in appropriate therapy with subsequent increased morbidity and mortality. For patients with acute uncomplicated lower UTIs, nitrofurantoin, trimethoprim-sulfamethoxazole, fosfomycin or pivmecillinam should be prescribed for a 1-5 day course depending on the agent used. Single-dose fosfomycin is an excellent option for uncomplicated lower UTIs and has had similar clinical and/or bacteriological efficacy for 3- or 7-day regimens for alternate agents (i.e., ciprofloxacin, norfloxacin, cotrimoxazole or nitrofurantoin). The aim of this review article is to provide an overview on the definitions, etiology, treatment guidelines (including agents for

infections due to antimicrobial resistant bacteria) of lower UTIs and to highlight recent aspects on antimicrobial resistance of ExPEC.

Keywords: Lower UTIs, antimicrobial resistance, treatment, *E. coli* ST131

Introduction

Urinary tract infections (UTIs) are among the most common infectious diseases occurring in either the community or healthcare settings. In North America, these infections account for over 10 million physician visits annually, 1 million emergency department visits, and over 100,000 hospitalizations annually, with an estimated cost of \$1 billion per year in the U.S [1]. UTIs accounts for substantial morbidity, mortality and medical expenses on a global scale and it is estimated that approximately 20% of women over the age of 18 – 29 years will suffer from an UTI during their lifetimes [2]. Moreover, UTIs are the most common bacterial infections found in nursing home residents, accounting for 50% of reported infections in Norwegian nursing homes, 30% to 50% in U.S. nursing homes, and 25% of all infections in the noninstitutionalized elderly in the U.S. Recurrent or relapsing UTIs are especially problematic in many individuals.

A wide variety of bacteria are responsible for causing UTIs, however *Escherichia coli* remains the most common etiological agent [1]. *E. coli* is an incredible versatile bacterium in that it has the ability to colonize numerous hosts (including humans) and forms an important part of the normal intestinal flora. Sometimes *E. coli* acquires pieces of DNA that gives it the ability to cause various diarrheal diseases and is referred to as an intestinal-pathogenic *E. coli*. However, there is another type of *E. coli* that has the capacity to cause infections outside the gastrointestinal system. These types of *E. coli* are referred to as extra-intestinal pathogenic *E.*

coli or ExPEC [3]. Certain factors (referred to as virulence-associated factors) give ExPEC the means to cause infections [4]; they include toxins, adhesins, lipopolysaccharides, capsules, proteases and invasins that are encoded by genes in pathogenicity islands. ExPEC is the most common cause of UTIs in developed and developing countries [5]. Before the 2000's, ExPEC was susceptible to first line antibiotics (e.g. cephalosporins, cotrimoxazole, and fluoroquinolones) that are often used to treat UTIs [5]. Unfortunately, since 2000, resistance to these antibiotics emerged globally and is causing delays in appropriate therapy with subsequent increased morbidity and mortality [5]. This has occurred at a time when the discovery and development of new anti-infective agents with activity against Gram negative bacteria are slowing down.

The aim of this review article is to provide an overview on the definitions, etiology, treatment guidelines (including agents for infections due to antimicrobial resistant bacteria) of lower UTIs and highlight recent aspects on antimicrobial resistance of ExPEC.

Definitions and Etiology of lower urinary tract infections

A UTI is defined as a bacterial infection of the urinary tract; more practically, it is the combination of significant bacteriuria and symptoms consistent with infection e.g. dysuria, frequency, fever [6]. UTIs are often further classified in dichotomies for clarity of discussion. In terms of anatomy, UTIs are divided into lower tract and upper tract infections. Lower tract UTIs, or cystitis, are infections that are limited to the bladder, whereas upper tract UTIs, or pyelonephritis, involve the kidney as well [7].

The differentiation between uncomplicated and complicated UTIs is not as clear cut, but generally uncomplicated UTIs refer to those that occur in a normal urinary tract, both anatomically and neurologically [8]. Conversely, complicated UTIs are characterized by complicating features that causes impairment of urinary flow or of host defenses. These may

include structural abnormalities (e.g. stones, diverticula, urethral valves), functional abnormalities (e.g. neurologic impairment secondary to spinal cord injury or multiple sclerosis), presence of foreign devices (e.g. urinary catheters, ureteral stents), or an otherwise compromised host (e.g. renal transplantation, immunosuppression, pregnancy). UTIs in males are often considered complicated, as are those in diabetic patients [7]. Both uncomplicated and complicated UTI's may be lower or upper tract in location.

Ascension through the urethra is most common route of infection for the urinary tract [1]. The uropathogen is part of the rectal flora, which has a transition period of periurethral and distal urethral colonization. The bacteria, utilizing a suite of virulence factors including adhesins, fimbriae, and pili, can adhere to the uroepithelium and withstand the mechanical forces present during urination.

The most common etiologic agent of uncomplicated UTIs is *Escherichia coli*, implicated in 75-90% of cases [1]. *Staphylococcus saprophyticus* can cause a significant proportion of infections (5-15%) in young women, but is uncommon outside this age group. Other *Enterobacteriaceae*, such as *Klebsiella* spp. and *Proteus* spp., are also important uropathogens. *Enterococcus* spp. and *Streptococcus agalactiae* are commonly isolated; however, these organisms are part of the periurethral flora and recent data suggests that their isolation in uncomplicated UTIs, even in high colony counts, may represent contamination [9].

Complicated UTIs have a broader microbiological range. *E. coli* remains a dominant uropathogen, but higher incidences of other bacteria are seen. A wider range of *Enterobacteriaceae* may be involved, including *Klebsiella* spp., the urease-producers (*Proteus* spp., *Morganella morganii*, *Providencia* spp.), and the other intrinsic AmpC producers (*Enterobacter* spp., *Citrobacter* spp., *Serratia marcescens*) [10]. *Pseudomonas aeruginosa* may

be frequently involved, especially in long-term catheters. *Enterococcus* spp., *S. agalactiae*, and coagulase-negative staphylococci are the most common Gram-positive uropathogens in complicated UTI's, and likely play a more important role in this group than in uncomplicated UTIs [10]. In addition, complicating factors often predispose towards recurrences and multiple courses of antibiotics, raising the likelihood of antimicrobial-resistant organisms [8,10].

Antimicrobial resistance among ExPEC

Antibiotic therapy with cotrimoxazole, cephalosporins, and fluoroquinolones are considered as the treatments of choice for infections due to ExPEC, especially before the 2000s. The development of resistance to the carbapenems among ExPEC is of special concern to the medical community at large, because these agents are often the last line of effective therapy available for the treatment of serious infections due to these bacteria [5]. Recently a new type of colistin resistant mechanism named MCR-1 was described in animals and patients in China and was associated with *E. coli*.

The most important β -lactamases in ExPEC are the extended-spectrum β -lactamases [ESBLs] (e.g. CTX-M types), plasmid-mediated or imported Amp C β -lactamases (e.g. CMY types), and to a lesser extent the carbapenemases (e.g. KPC types, MBLs e.g. VIM, IPM and NDM types, and the oxacilinases e.g. OXA-48-like enzymes) [4]. The NDM, OXA-48 and KPC are the most common carbapenemases among nosocomial and community isolates of ExPEC [11].

The CTX-M β -lactamases

Extended-spectrum β -lactamases (ESBLs) were first described in 1983 and have the ability to hydrolyse the penicillins, cephalosporins and monobactams, but not the cephamycins

and carbapenems [12]. They are inhibited by β -lactamase inhibitors such as clavulanic acid, sulbactam and tazobactam. Although ESBLs have been identified in a range of Enterobacteriaceae, they are most often present in *E. coli* and *K. pneumoniae*. The majority of ESBLs identified in clinical isolates during the 1980s and 1990s were of the SHV or TEM types, which evolved from parent enzymes such as TEM-1, -2 and SHV-1 [12]. A different type of ESBL, named CTX-M (i.e. active against CefoTaXime) β -lactamases, that originated from environmental *Kluyvera* spp, gained prominence in the 2000s with reports of clinical isolates of *E. coli* producing these enzymes from Europe, Africa, Asia, South and North America [13]. Since the mid 2000's CTX-M β -lactamases have also been identified in different members of the Enterobacteriaceae as well as non- Enterobacteriaceae, but remain especially common in *E. coli*, and has become the most wide-spread and common type of ESBL identified worldwide [14].

The CTX-Ms belong to the molecular class A or functional group 2 β -lactamases, and include at least six lineages (i.e. CTX-M-1, CTX-M-2, CTX-M-8, CTX-M-9, CTX-M-25 and KLUC) that differ from each other by $\geq 10\%$ amino acid similarity [13]. Currently, the most widespread and prevalent type of CTX-M enzyme among human clinical isolates of *E. coli* is CTX-M-15. *E. coli* producing this enzyme often belongs to sequence type 131 (ST131) [15]. The intercontinental dissemination of this sequence type has played a major role in the worldwide emergence of CTX-M-15 producing *E. coli*.

CTX-M-producing ExPEC are important causes of hospital and community-onset urinary tract infections, bacteraemia and intra-abdominal infections [13]. The risk factors associated with infections caused by CTX-M-producing *E. coli* include the following: repeat UTIs, underlying renal pathology, previous antibiotics including cephalosporins and fluoroquinolones, previous

hospitalization, nursing home residents, older males and females, Diabetes Mellitus, underlying liver pathology and international travel to high risk areas such as the Indian subcontinent [16].

Global surveys have illustrated an alarming trend of associated resistance to other classes of antimicrobial agents among CTX-M-producing *E. coli* that include trimethoprim-sulfamethoxazole, tetracycline, gentamicin, tobramycin and ciprofloxacin [14]. This has important clinical implications because the fluoroquinolones and cotrimoxazole are popular oral treatments options for UTIs. Fortunately, fosfomycin and nitrofurantoin retain sufficient activity against a high percentage of *E. coli* that produce CTX-Ms [17].

It is evident today that CTX-M-producing *E. coli* is a major player in the world of antimicrobial resistance. A report from the Infectious Diseases Society of America listed ESBL-producing *E. coli* as a priority drug-resistant microbe to which new therapies are urgently needed [18].

AmpC β -lactamases

Enterobacteriaceae with inducible cephalosporinases or AmpC β -lactamases such as *Enterobacter cloacae*, *Citrobacter freundii*, *Serratia marcescens*, *Morganella morganii*, and *Providencia stuartii*, can develop resistance to the oxyimino-cephalosporins and 7- α -methoxy-cephalosporins (i.e. cephamycins e.g. cefoxitin, cefotetan) and monobactams (e.g. aztreonam) by overproducing their chromosomal AmpC β -lactamase [19]. In *Klebsiella spp*, *Salmonella spp.*, and *P. mirabilis*, that lack chromosomal β -lactamases, this type of resistance is usually mediated by plasmid encoded or imported AmpC β -lactamases [20]. *E. coli* is different in that it possesses genes encoding for chromosomal non-inducible AmpC β -lactamases that are regulated by weak promoters and strong attenuators, resulting in low amounts of the cephalosporinase [21]. Surveys of resistance mechanisms in cephamycin-resistant *E. coli*, have identified several

promoter or attenuator mutations, which resulted in the up-regulation and production of the natural occurring chromosomal AmpC β -lactamases [22]. Occasionally, cephamycin-resistant *E. coli* can also produce plasmid-mediated or imported AmpC β -lactamases [19].

AmpC β -lactamases at high levels, hydrolyse penicillins, most cephalosporins, cephamycins and monobactams, but not the fourth generation cephalosporins (e.g. cefepime) and carbapenems (Table 1). Resistance to the fourth generation cephalosporins are caused by point mutations in AmpC β -lactamases and is called extended-spectrum cephalosporinases. AmpC enzymes are not inhibited by “classical” β -lactamase inhibitors, such as clavulanic acid, sulbactam and tazobactam, although boronic acids and cloxacillin have shown to be good inhibitors [23]. The genes are typically encoded on large plasmids containing additional antibiotic resistance genes that are responsible for multi-resistant phenotype, leaving few therapeutic options [21].

Just like ESBL-producing bacteria, *E. coli* with plasmid-mediated AmpC enzymes have mostly been responsible for nosocomial outbreaks on a worldwide basis during the late 1980’s and 1990’s, although the risk factors associated with infection are not as well defined as those associated with ESBL-producing bacteria [21]. In a study reported from Korea, patients infected by plasmid-mediated AmpC-producing organisms had similar clinical features and outcomes to those patients infected with TEM- or SHV-related ESBL producers [24].

It seems that CMY-2 (active on CephaMYcins) is the most common imported AmpC β -lactamase reported in Enterobacteriaceae (especially *E. coli* and *Salmonella* spp.) from different areas of the world [19]. Hospital surveys from Asia, North America and Europe have shown that the DHA types of cephamycinases are mostly present in *Klebsiella* spp from Asia, CMY are

Table 1. Treatment Regimens and Expected Early Efficacy Rates for Acute Uncomplicated Cystitis*

Drug (dosage)	Mean percentage (range)			
	Estimated clinical efficacy ^{ab}	Estimated microbiological efficacy ^b	Common side effects	References
Nitrofurantoin monohydrate/macrocrystals (100 mg twice daily for 5-7 days)	93 (84-95)	88 (86-92)	Nausea, headache	10, 21
Trimethoprim-sulfamethoxazole (160/800 mg twice daily for 3 days)	93 (90-100)	94 (91-100)	Rash, urticarial, nausea, vomiting, hematologic	10, 21
Fosfomycin trometamol (3 g single-dose sachet)	91	80 (78-83)	Diarrhea, nausea, headache	10, 22
Pivmecillinam (400 mg twice daily for 3-7 days)	73 (55-82)	79 (74-84)	Nausea, vomiting, diarrhea	10, 30, 66
Fluoroquinolones (dose varies by agent; 3-day regimen) ^c	90 (85-98)	91 (81-98)	Nausea/vomiting, diarrhea, headache, drowsiness, insomnia	10, 30
β -lactams (dose varies by agent; 305 day regimen) ^d	89 (79-98)	82 (74-98)	Diarrhea, nausea, vomiting, rash, urticarial	10, 14

* Adapted with permission from IDSA/ESCMID Guidelines (10)

^a Efficacy rates refer to cure rates on the visit closest to a 5-9 day period following treatment, and are averages or ranges calculated from clinical trials discussed in the text.

^b Estimated clinical efficacy and microbiological efficacy rates should not necessarily be compared across agents, because study design, efficacy definition, therapy duration, and other factors are heterogeneous. Note that efficacy rates may vary geographically depending on local patterns of antimicrobial resistance among uropathogens.

^c Data on fluoroquinolones are compiled from regimens of ofloxacin, norfloxacin, and ciprofloxacin

^d Data on β -lactams are derived from clinical trials examining second and third generation cephalosporins and amoxicillin-clavulanate.

present in *E. coli* from Asia, North American and Europe while FOX are present in *Klebsiella spp.* from North America and Europe [25-29].

KPC β -lactamases

The most clinical significant of the class A carbapenemase is the KPC-types (i.e. *Klebsiella pneumoniae* Carbapenemase) [30]. KPC-1 (it was later shown to be identical to KPC-2) was reported in the late 1990's from a *K. pneumoniae* isolated in North Carolina, USA and to date, more than 18 different KPC variants have been described [31], even though KPC-2 and -3 remain the most commonly identified variants. These enzymes provide resistance to the carbapenems, cephalosporins, cephamycins and monobactams and are weakly inhibited by “classical” β -lactamase inhibitors such as clavulanic acid and tazobactam (Table 1). KPC β -lactamases (especially KPC-2 and -3) have been described in several enterobacterial species, especially *Klebsiella spp.* and to a lesser extent in *Enterobacter spp.* [30].

Several nosocomial outbreaks, most often due to *K. pneumoniae*, have been reported from North America (especially the USA), South America (Colombia, Argentina), Europe (Greece, Italy, Poland), Asia (China) and Middle East (Israel) [31-33]. KPC-producing bacteria are considered to be endemic in certain parts of the world, such as the North Eastern USA, Puerto Rico, Colombia, Greece, Italy, Israel and China and are important causes of nosocomially-acquired infections in some parts of these countries (Table 1) [33]. *K. pneumoniae* ST258 with KPC-2 and KPC-3 had significantly contributed to the world-wide distribution of this resistance trait [33].

The genes responsible for the production of KPC enzymes are located on transferable plasmids and are associated with the mobile element transposon *Tn4401*, explaining their spread among clinically relevant pathogens [34] (Table 1). Plasmids that encode for KPC enzymes often

contain several genes that encode for resistance to other antimicrobial agents, such as the aminoglycosides, plasmid-mediated quinolone resistance, trimethoprim, sulfonamides and tetracyclines. This ensures that KPC-producing Enterobacteriaceae are often multiresistant to various groups of antibiotics [35].

NDM β -lactamases

A new type of metallo- β -lactamase (MBL), named NDM (i.e. New Delhi metallo- β -lactamases), was described in 2009 in *K. pneumoniae* and *E. coli* recovered from a Swedish patient who was hospitalized in New Delhi, India [36]. NDMs have the ability to hydrolyse a wide variety of β -lactams, including the penicillins, cephalosporins and carbapenems, but not the monobactams (i.e. aztreonam), and are inhibited by metal chelators such as EDTA. NDM-1 shares very little identity with other MBLs, the most similar being VIM-1/VIM-2 with only 32.4% amino acid identity. Since the first description of NDM-1, more than eight variants of this enzyme have been described, with the majority of them originating from Asia [11].

The majority of NDM-1-producing bacteria are broadly resistant to various drug classes and also carry a diversity of other resistance mechanisms (e.g. to aminoglycosides and fluoroquinolones), which leaves limited treatment options [37]. These additional mechanisms include the following: plasmid-mediated AmpC β -lactamases (especially CMY types), ESBLs (especially CTX-M-15), different carbapenemases (e.g. OXA-48-, VIM-, KPC-types), 16S RNA methylases, plasmid-mediated quinolone resistance determinants to quinolones, macrolide modifying esterases, and rifampicin-modifying enzymes. Consequently, Enterobacteriaceae with NDMs remain only susceptible to colistin, fosfomycin and tigecycline [11].

Kumarasamy and colleagues [38], provide compelling evidence that NDM-producing Enterobacteriaceae (mostly *K. pneumoniae* and *E. coli*) are widespread in India and Pakistan.

They also found that many UK patients infected with NDM-producing bacteria had recently traveled to India to undergo several types of medical procedures. The patients presented with a variety of hospital- and community-associated infections with urinary tract infections (UTIs) being the most common clinical syndrome. Recent reports from the subcontinent (including India, Pakistan and Bangladesh) showed that the distribution of NDM β -lactamases among Enterobacteriaceae was widespread through these countries [39,40]: e.g. a hospital in Varanasi in Northern India identified NDM-1 prevalence rate of 6% among *E. coli* (n=528) from outpatients and hospitalized patients between February 2010 and July 2010 [41], 7% among *E. coli* from a major hospital in Mumbai, India [42], whereas 15% (30/200) of in- and outpatients in Rawalpindi, Pakistan carried *E. coli* with NDM-1 in their gut flora [43]. The prevalence of asymptomatic fecal carriage was estimated to have been between 5 to 15 % in these countries [11].

Since 2010 Enterobacteriaceae with NDMs have been reported worldwide from patients with an epidemiological link to the Indian subcontinent [44]. The impact of intercontinental travel as a source of spread of NDM producers has been extensively reported and these enzymes are now one of the most common carbapenemases identified in countries such as Canada, the UK and France [11]. Recent findings suggest that the Balkan states, North Africa and the Middle East might act as secondary reservoirs for the spread of NDMs, which may or may not initially have reached these countries from the Indian subcontinent [11] (Table 1).

Enterobacteriaceae (especially *K. pneumoniae* and *E. coli*) with NDMs, have been recovered from many clinical settings, reflecting the disease spectra of these opportunistic bacteria, including hospital and community-onset urinary tract infections, septicaemia, pulmonary infections, peritonitis, device-associated infections and soft tissue infections

[37,39,44]. The frequent identification of NDMs within *E. coli* is of special concern because this organism is the most common cause of community and hospital onset urinary tract infections and diarrhea [88]. NDM-1-positive bacteria have been recovered from the gut flora of travelers returning from the Indian subcontinent and were undergoing microbiological investigation for unrelated diarrheal symptoms [45]. There is also widespread environmental contamination by NDM-1-positive bacteria in New Delhi [46].

OXA-48-like β -lactamases

The Ambler class D β -lactamases is commonly referred to as OXAs (i.e. “oxacillinases”) and comprises over 400 enzymes and some variants possess carbapenemase activity [93]. These enzymes (also referred to as carbapenem-hydrolysing class D β -lactamases or CHDLs) do not efficiently hydrolyse the oxyimino-cephalosporins or the monobactams (the exception being OXA-163) [37]. The CHDLs possess weak carbapenemase activities that do not confer high level resistance to carbapenems if not associated to other factors such as permeability defects. The majority of CHDLs variants have been identified in *Acinetobacter* spp., but OXA-48 and its derivatives (i.e. OXA-163, OXA-181, OXA-204, and OXA-232) are most often encountered in Enterobacteriaceae [47].

OXA-48 was first identified in 2003 from *K. pneumoniae* isolated in Turkey [48] and since then, bacteria that produce these β -lactamases, have been important causes of nosocomial outbreaks in this country [49]. The first report of OXA-48-producing *K. pneumoniae* outside Turkey was in 2007 from Belgium [50] and bacteria that produce OXA-48 have rapidly spread to several Belgian hospitals [51]. Since then Enterobacteriaceae with OXA-48 have disseminated throughout Europe, and it seems that Turkey and North African countries (especially Morocco and Tunisia [37]) are the main reservoir for these infections. Subsequently several nosocomial

outbreaks of OXA-48-producing *K. pneumoniae*, *E. coli*, and *Enterobacter cloacae* have been reported in France, Germany, Switzerland, Spain, the Netherlands and UK [47].

The spread of OXA-48 is mostly due to a mobile 62.5-kb plasmid that belongs to the Inc L/M replicon group, as well as the presence of *Tn1999* [47]. Interestingly, the 62.5-kb plasmid does not carry additional resistance genes. This plasmid has the ability to efficiently transfer resistance to other enterobacterial species. OXA-48-like enzymes have mostly been identified in *K. pneumoniae* and *E. coli* to a lesser extent and are not inhibited by metal chelators such as EDTA or “classical” β -lactamases inhibitors such as clavulanic acid or tazobactam (Table 1). The production of ESBLs and/or permeability barriers in bacteria that co-produce OXA-48 will increase the level of resistance to the cephalosporins and carbapenems [37].

Several point mutant variants of OXA-48, most often in *K. pneumoniae*, have recently been reported. These included OXA-163, OXA-181, OXA-204 and OXA-232 [11]. OXA-163 differ from the other OXA-48-like enzymes in that its carbapenemase activity is low combined with enhanced hydrolysis of the oxyimino-cephalosporins providing isolates with resistance phenotypes similar to those bacteria with ESBL. OXA-163 was originally identified from enterobacterial isolates (*E. cloacae* and *K. pneumoniae*) recovered in Argentina, and it seems that Enterobacteriaceae with these oxacillinases are common in this South American country [11].

MCR-1: plasmid-mediated colistin resistance

MCR-1, the first transferable plasmid-mediated colistin resistance gene, was reported in ExPEC isolates from food animals, food and patients in China [52]. Since then, detection of MCR-1-positive strains has been reported in several Enterobacteriaceae worldwide. Of clinical concern is the presence of plasmids containing MCR-1 in ExPEC with carbapenemases. Such

isolates have the potential to create pandrug resistant bacteria originating mostly from the community that will nearly be impossible to treat effectively.

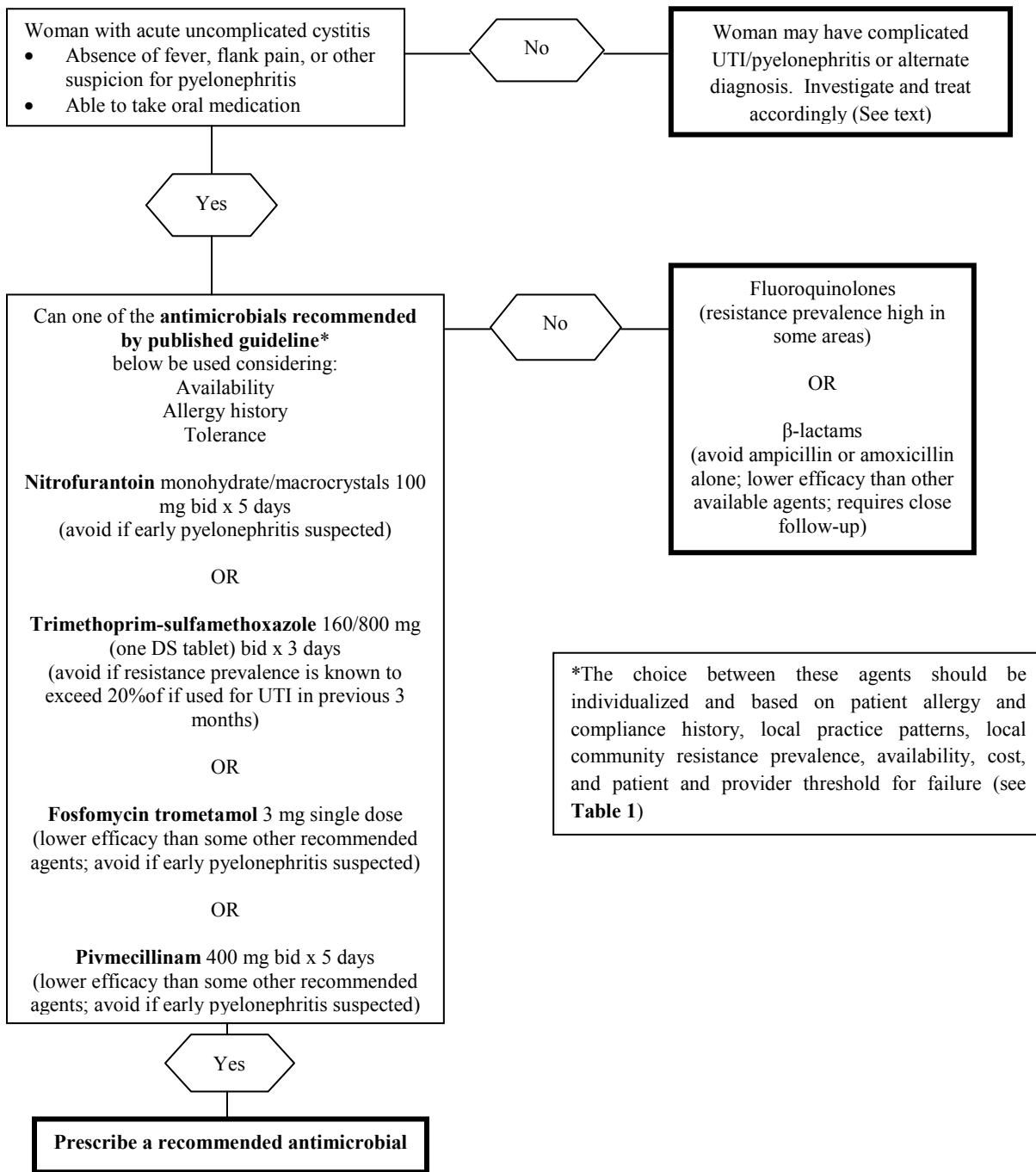
Guidelines for treating lower urinary tract infections

Although acute cystitis is one of the most common infections in ambulatory women, studies show that prescribing practices vary widely despite the publication of treatment guidelines by expert groups [53-55]. The Infectious Diseases Society of America (IDSA) and the European Society for Microbiology and Infectious Diseases initially published clinical practice guidelines for the treatment of acute uncomplicated cystitis in the 1990s [56]. However, in the intervening period, treatment approaches have become more complicated because of the steady rise in antimicrobial resistance amongst uropathogens, particularly *E. coli* [57-61], requiring the IDSA and the ESMID to update their conjoint guidelines in 2010 [62]. Although ampicillin was initially the recommended treatment of choice for uncomplicated lower UTI, this is no longer the case because of the very high prevalence of antimicrobial resistance to both amoxicillin and ampicillin worldwide [63-65]. However, other B-lactams including cefdinir, cefaclor, amoxicillin-clavulanate and cefpodoxime-proxetil in 3-7 day regimens continue to be recommended for treatment of uncomplicated cystitis when susceptibility is confirmed, and other agents cannot be used [62]. However, many of these drugs have not been well studied for the treatment of UTI, and should currently be considered alternative agents as they generally have inferior efficacy and more adverse effects on the host's normal flora (i.e., *Clostridium difficile*) [62,66,67]. The fluoroquinolones including ofloxacin, norfloxacin, ciprofloxacin and levofloxacin are also currently being considered alternative antimicrobials for acute cystitis [62]. Although fluoroquinolones are highly efficacious in a 3-day regimen, there has been a dramatic increase in resistance to these agents in Gram-negative uropathogens in the past decade,

particularly due to the global dissemination of the ESBL-producing *E. coli* ST131, and *Pseudomonas aeruginosa* for which there are currently no other orally available treatment options [57,61,68-70]. Fluoroquinolones also have potentially serious side effects (i.e., polyneuropathy) and an increased risk of ecological disruption of the host normal flora (i.e., *C. difficile*), and should therefore be reserved for more important treatments other than acute cystitis [71,72].

Figure 1 and Table 1 outline the current expert recommendations for the optimal treatment for acute uncomplicated cystitis [62]. For women with acute uncomplicated cystitis who are able to take oral medication, nitrofurantoin (NTF), trimethoprim-sulfamethoxazole (SXT), fosfomicin (FOS) or pivmecillinam (PVM) should be prescribed for a 3-5 day course depending on the agent used. Nitrofurantoin monohydrate/macrocrystals is an appropriate treatment choice due to the minimal resistance [62], efficacy and propensity to cause alteration in the host's flora compared to a 3-day course of SXT [73]. Although SXT is still widely used and recommended for acute cystitis therapy, the caveat is that local resistance rates of uropathogens should not exceed 20%, or if the infecting strain is confirmed to be susceptible [62]. The IDSA/ESMID chose a cut-off of 20% resistance rate for SXT based on clinical, in vitro and mathematical modelling studies with regards to the potential for treatment failure [62]. Fosfomicin-trometamol can also be used if it is available because of minimal resistance and propensity for collateral damage [74,75]. Although fosfomicin may be used for other indications, its use has been restricted to treatment of acute cystitis due to ESBL-producing *E. coli* or *Klebsiella spp.* Fosfomicin appears to have inferior efficacy however, when compared to other standard short course regimens according to data submitted to the US Food and Drug Administration (FDA) [76,77]. Several other recent studies have demonstrated comparable

Figure 1. Recommended Approach for Selection of Appropriate Antimicrobial Therapy for Acute Uncomplicated Cystitis. DS, double-strength; UTI, urinary tract infection. Adapted with permission from IDSA/ESCMID Guidelines (10)



efficacy to other regimens[78]. Finally, Pivmecillinam is an appropriate choice for therapy in some European countries where it is available. This agent is currently not licensed and/or available in North America except through special authorization access. Use of this antimicrobial for acute uncomplicated UTI in Northern European countries for several decades has resulted in minimal resistance and propensity for collateral host damage [79,80]. However, several recent studies have shown that Pivmecillinam may have inferior efficacy compared with other standard short course regimens, which could have been due to under dosing [81,82]. Women with an acute uncomplicated UTI who receive inadequate antimicrobial treatment may develop subclinical pyelonephritis with recurrent symptomatic infections with the same organism [83].

Agents for treating lower urinary tract infections due to antimicrobial resistant bacteria

The emergence of multi-drug resistant Gram-negative bacilli has renewed interest in the use of older antimicrobial agents to treat uncomplicated UTIs, particularly those that have low levels of resistance against β -lactamase-producing *Enterobacteriaceae*. Although nitrofurantoin and SXT have been used worldwide for decades, fosfomycin, temocillin and pivmecillinam are not licensed and/or available in North America and many countries in Europe. As such, there is a paucity of international surveillance data because currently none of these drugs are part of an antibiotic resistant surveillance in developed countries. This section describes agents that have recently become part of the new IDSA treatment recommended for acute uncomplicated UTIs, specifically fosfomycin and pivmecillinam for ExPEC, when resistance to one or more of carbapenems, colistin, aminoglycosides and quinolones are present.

Fosfomycin

This compound has been in use for over forty years, and it has a unique mechanism of action. Fosfomycin-trometamol (FOS) is a phosphonic acid derivative that acts as a bacterial cell wall inhibitor [84,85]. The compound inhibits the UDP-N-acetylglucosamine enolpyruvyl transferase (MurA), an enzyme responsible for ligating phosphoenolpyruvate to the 3'-hydroxyl group of UDP-N-acetylglucosamine in the first step of peptidoglycan synthesis [77,84,86]. The drug enters into the active site of MurA and inhibits the enzyme by covalent binding with the active site, Cys115 [87,88]. Resistance most commonly occurs by decreased permeability, and enzymatic modification of FOS by plasmid-encoded glutathione transferases (encoded by mainly *fosA*, but also *fosB*, *fosC*, and *fosX*)[78,87-89]. Uncommonly, the fosfomycin binding site Cys115 may also be changed via mutations in the genes encoding the hexose phosphate transport (UhpT) and glycerol-3-phosphate transport (GlpT) pathways [78,87]. Resistance to FOS is rare considering the amount of time this compound has been used, due to the compound's unique structure and distinct mechanism of action [87,88]. However, increasing reports of plasmid-mediated FOS resistance signal that this trend may not last. The plasmid-mediated fosfomycin resistance gene *fosA3* in *E. coli* was first described in Japan, and encodes a glutathione S-transferase[90]. Since that time, there have been increasing reports that document detection of plasmid-mediated FOS resistance (*fosA3* and *fosC2*) in multi-drug resistant *E. coli* isolated from livestock and animals from several countries in Southeast Asia, particularly China [91-94]. More recently, plasmid-mediated FOS resistance in *E. coli* has also been reported in a patient in the United States [95].

FOS has a broad spectrum of antibacterial activity, and in vitro testing shows good activity against Gram-negative (*Enterobacteriaceae*) and Gram-positive bacteria

(*Staphylococcus saprophyticus*, *Enterococcus faecalis*, but not *E. faecium*) that commonly cause UTIs [85,96]. A recent systematic review by Falagas *et al* [74], evaluated 17 selected studies of the antimicrobial activity of FOS in isolates of *Enterobacteriaceae* collected after 2000, and its clinical effectiveness in treating not only lower urinary tract infections and other sites (i.e., wounds). Four of these studies involved isolates from Spain [97-100], three from France [101-103], two from the UK [104,105], and two from Thailand [106,107]. The remaining six studies involved isolates from Greece, Hong Kong, Japan, Korea, Turkey or the USA [108-113].

Although most of these studies used either disc diffusion or agar dilution methods, various MIC interpretive guidelines were used including: CLSI (i.e., MIC \leq 64 mg/L) (www.clsi.org), British Society for Antimicrobial Chemotherapy (MIC \leq 128 mg/L) (www.bsac.org), and the Comité de l'Antibiogramme de la Société Française de Microbiologie (MIC \leq 32 mg/L) (www.sfm-microbiologie.org)[74]. In addition, one study from France and another one from Japan did not indicate the breakpoints used. A total of 5057 isolates of *Enterobacteriaceae* [2205 *E. coli*, 764 *K. pneumoniae* and 73 *Enterobacter spp.*] were studied, including 4448 (88%) strains that produced an ESBL. *E. coli* was the most susceptible to FOS of ESBL-producing *Enterobacteriaceae*. At least 90% of *Enterobacteriaceae* isolates with multi-drug resistance were susceptible to FOS using a MIC breakpoint of \leq 64 mg/L including 1604 (96.8%) of ESBL-producing *E. coli* and 608 (81.3%) *Klebsiella pneumoniae*. In the 11/17 studies which used CLSI MIC interpretive criteria, the cumulative susceptibility to FOS of ESBL-producing strains was 3569/3911 (91.3%)[74]. However, ESBL-producing *Enterobacteriaceae* collected from ambulatory patients had a higher rate of susceptibility to FOS by the CLSI criteria (292/297, 98.3%) compared to those recovered from hospitalized patients (1344/1519, 88.5%). Only 4/17 of these studies evaluated the clinical effectiveness of FOS against ESBL-producing *E. coli* in

patients with various risk factors [74]. Cumulatively, treatment with FOS was associated with clinical cure in 75/80 (93.8%) patients[74], but one of these studies reported a lower rate of microbiological clearance (41/52; 78.8%). However, in European countries where this antimicrobial has had widespread use for several decades, susceptibility levels have recently been shown to have substantially decreased, both in strains from ambulatory and hospitalized patients [70].

More recent studies from both North America and Europe have focussed on the use of FOS for lower urinary tract infections, particularly those due to ESBL-producing *Enterobacteriaceae* since a single dose of FOS 3 g achieves a high concentration in urine[77,114], and is well tolerated, with only some nausea and diarrhoea being reported[77,96]. Keating[78] reviewed the results of clinical trials by focusing on the use of single-dose FOS treatment for patients with acute lower urinary tract infections and pregnant women with asymptomatic bacteriuria. Recent randomized trials showed that single-dose FOS had similar clinical and/or bacteriological efficacy to 3- or 7-day regimens for alternate agents (i.e., ciprofloxacin, norfloxacin, cotrimoxazole or nitrofurantoin) in women with uncomplicated UTIs. A single-dose of FOS also had similar bacteriological efficacy to a 5-day course of cefuroxime axetil or a 7-day course of amoxicillin/clavulanic acid in pregnant women with asymptomatic bacteriuria, and similar clinical and/or bacteriological efficacy to a 5-day course of cefuroxime axetil or amoxicillin/clavulanic acid, or a 3-day course of ceftibuten in pregnant women with cystitis[78].

FOS is also increasingly being used in hospitalized patients to primarily treat nosocomial UTIs, particularly for *E. coli* infection due to MDR strains, and in patients where other antimicrobials may be contraindicated due to collateral damage (i.e., *C. difficile*, patient

allergies). A recent study also evaluated the use of FOS among inpatients at a tertiary care hospital in the United States between 2009 and 2013 [75]. The number of patients with UTIs that were treated with FOS substantially increased during this period, from none to 391 in 2013. Of the 537 patients who received FOS for any indication, physician-diagnosed UTI accounted for 74% of prescriptions, another 10% of treatments were due to asymptomatic bacteriuria, and the remainder had other indications including fever, leukocytosis, or sepsis of uncertain etiology[75]. *E. coli* was the most common organism isolated from the 239 patients with UTI, and 20% of these isolates were MDR[75]. Almost all patients received a single-3g-dose of FOS, which resulted in an overall clinical success rate at 48 h of 74.8%. However, among the 89 patients with confirmed UTIs, according to national surveillance definitions, the success rate was much higher (89.9%). Recurrent infections occurred in only 4.3% of cases and the drug was well-tolerated with only a few documented adverse events[75].

A clinical trial is also underway in Spain investigating the clinical non-inferiority of intravenous FOS (4g/6h) compared to meropenem (1g/8h) for treating bacteremic urinary tract infections caused by ESBL-producing *E. coli* [115]. This phase III randomised controlled trial which completes enrollment in 2017, will compare the clinical and microbiological efficacy 5-7 days after treatment, and the safety of these two treatment arms. If FOS is shown to have non-inferior results to the use of a carbapenem, the results may have justified the use of parenteral FOS as an alternate agent for treatment of selected patients with frequent complicated UTIs caused by MDR *E. coli* with susceptibility to this drug.

Pivmecillinam

This compound is the pro-drug of mecillinam (6 β -amidinopenicillanic acid), which is a penicillin derivative that has been used extensively in Scandinavian countries since the 1980s for

treatment of uncomplicated UTIs [116,117]. Mecillinam disrupts cell wall synthesis by inhibiting penicillin-binding protein 2 (PBP2)[118,119], which is responsible for elongation of rod-shaped cells. The mechanism(s) of resistance to mecillinam are currently poorly understood although a recent study showed that mutation in *cysB* preventing the production cysteine was found in clinical isolates. The reduced cysteine levels led to an increased cellular concentration of the ppGpp molecule, which makes the drug's PBP2 target nonessential[120].

Pivmecillinam (PVM) treatment causes minimal disruption to the gastrointestinal microbial flora, and the drug achieves high concentrations in urine [116,117,121]. PVM only has *in vitro* activity against Gram-negative organisms, but its extensive use for uncomplicated UTI has shown that it is safe during pregnancy, and effective for *Staphylococcus saprophyticus* infection because the high urine levels overcome *in vitro* resistance [79,122]. Although the Swedish Reference Group for Antibiotics (www.srga.org) has published breakpoints for the susceptibility testing of PVM against uropathogens, a recent study suggested that these should be revised [122]. Mosen *et al.* [122] recently analyzed data from a prospective, multi-centre, placebo-controlled primary healthcare facility in Sweden of 1143 women with cystitis who were enrolled from 1995-98. The clinical and bacteriological outcomes of PVM treatment were evaluated in this cohort with a specific focus on patients with UTI due to a PVM resistant uropathogen. *In vitro* susceptibility to PVM did not predict clinical or bacteriological outcome of treatment irrespective of the type of uropathogen causing infection.

To date there have been limited reports of the treatment efficacy of PVM for UTIs. Recent studies however have shown albeit in a small number of patients the PVM has bacteriologic and clinical efficacy for treatment of lower UTIs caused by ESBL-producing *Enterobacteriaceae*. Nicolle *et al.*[82] compared 3-days of PVM 400 mg po bid or norfloxacin

400 mg bid treatment of acute uncomplicated urinary infection in women. Bacteriological cure was achieved in 75% of PVM patients compared to 91% of those who received norfloxacin. Early post-therapy clinical cure was achieved in 82% of women who received PVM compared to 88% who received norfloxacin. Although adverse effects were similar for both drugs, norfloxacin was superior for short-course therapy in terms of bacteriological cure, but clinical outcomes were less marked. More recently, Nicolle *et al.* [123] described the successful treatment of a patient with relapsing pyelonephritis due to an CTX-M-producing *E. coli* who was cured with a prolonged course of PVM. Titelman *et al.* [81] recently reported the efficacy of PVM for treatment of lower urinary tract infection caused by ESBL-producing *E. coli* and *Klebsiella pneumoniae* in eight patients. All patients had a good clinical response, but bacteriological cure rates were low (2/8; 25%). However, despite persisting bacteriuria with the same uropathogen, all of the patients remained asymptomatic for 6 months after treatment. The clinical and bacteriological effect of PVM for ESBL-producing *E. coli* and *K. pneumoniae* in UTI was also reported in a larger group of 39 patients who had PVM susceptible isolates [124]. Of the 39 patients enrolled, 30 received a treatment regimen of 400 mg of PVM TID, and the other 9 received 200 mg TID. The overall bacteriological cure rate was 79% and was similar whether the patients received the higher (80%) or lower (78%) antibiotic dosage. Clinical symptoms resolved in 84% of the evaluable patients, and only five patients had relapsing bacteriuria.

Expert commentary

Urinary tract infections (UTIs) are among the most common infectious diseases occurring in either the community or healthcare settings and *E. coli* (extra-intestinal pathogenic *E. coli* or ExPEC) remains the most common etiological agent. During the 1990's, ExPEC was susceptible

to first line antibiotics (e.g. cephalosporins, cotrimoxazole, and fluoroquinolones) that were used to empirically treat UTIs. Resistance to these antibiotics emerged globally and is now causing delays in appropriate therapy with subsequent increased morbidity and mortality. The most important β -lactamases in ExPEC are the extended-spectrum β -lactamases [ESBLs] (e.g. CTX-M types), plasmid-mediated or imported Amp C β -lactamases (e.g. CMY types), and to a lesser extent the carbapenemases (e.g. KPC types, MBLs e.g. VIM, IPM and NDM types, and the oxacilinases e.g. OXA-48-like enzymes). MCR-1, the first transferable plasmid-mediated colistin resistance gene, was reported in ExPEC isolates from food animals, food and patients in China.

The Infectious Diseases Society of America (IDSA) and the European Society for Microbiology and Infectious Diseases initially published clinical practice guidelines for the treatment of acute uncomplicated cystitis in the 1990s. However, in the intervening period, treatment approaches have become more complicated because of the steady rise in antimicrobial resistance amongst uropathogens, particularly *E. coli*, requiring the IDSA and the ESMID to update their conjoint guidelines in 2010. For women with acute uncomplicated cystitis who are able to take oral medication, nitrofurantoin, trimethoprim-sulfamethoxazole, fosfomycin or pivmecillinam should be prescribed for a 1-5 day course depending on the agent used. Single-dose fosfomycin is an excellent option for uncomplicated lower UTIs and had similar clinical and/or bacteriological efficacy to 3- or 7-day regimens for alternate agents (i.e., ciprofloxacin, norfloxacin, cotrimoxazole or nitrofurantoin) in women with uncomplicated UTIs.

Pivmecillinam is available in certain North European countries and is currently not licensed and/or available in North America. Several recent studies have shown that Pivmecillinam may have inferior efficacy compared with other standard short course regimens.

Five Year View

ExPEC will continue to be the most common cause of uncomplicated UTIs on a worldwide basis. Resistance to different classes of antibiotics will continue to increase among global ExPEC due to the expansion of successful drug resistant clones such as *E. coli* ST131 and ST405. This will cause further delays in appropriate therapy of community and hospital onset UTIs with subsequent increased morbidity and mortality. The carbapenemases (e.g. NDMs, KPC and OXA-48), plasmid-mediated resistance to fosfomycin and colistin will most likely expand exponentially among ExPEC in the next few years or so.

The medical community urgently needs effective oral drugs for the treatment of uncomplicated UTIs. Unfortunately, the development of such new anti-infective agents will most likely not be available within the next 5 years.

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Key issues

- Urinary tract infections (UTIs) are among the most common infectious diseases occurring in either the community or healthcare settings.
- A wide variety of bacteria are responsible for causing UTIs, however *Escherichia coli* remains the most common etiological agent.
- Before the 2000's, ExPEC was susceptible to first line antibiotics (e.g. cephalosporins, cotrimoxazole, and fluoroquinolones) that are often used to treat UTIs. Unfortunately since

2000, resistance to these antibiotics had emerged globally and is causing delays in appropriate therapy with subsequent increased morbidity and mortality.

- The most important β -lactamases in ExPEC are the extended-spectrum β -lactamases [ESBLs] (e.g. CTX-M types), plasmid-mediated or imported Amp C β -lactamases (e.g. CMY types), and to a lesser extent the carbapenemases (e.g. KPC types, MBLs e.g. VIM, IPM and NDM types, and the oxacilinases e.g. OXA-48-like enzymes).
- For women with acute uncomplicated cystitis who are able to take oral medication, nitrofurantoin, trimethoprim-sulfamethoxazole (SXT), fosfomycin or pivmecillinam should be prescribed for a 1-5 day course depending on the agent used.
- Single-dose fosfomycin had similar clinical and/or bacteriological efficacy to 3- or 7-day regimens for alternate agents (i.e., ciprofloxacin, norfloxacin, cotrimoxazole or nitrofurantoin) in women with uncomplicated UTIs.
- Pivmecillinam is available in certain North European countries and is currently not licensed and/or available in North America. Several recent studies have shown that Pivmecillinam may have inferior efficacy when compared to other standard short course regimens.

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Highlighted References

Reference 1: Excellent overview on UTIs

Reference 62: excellent guidelines from USA and Europe for treating lower UTIs.

Reference 113: good overview on fosfomycin

Reference 116: good overview on pivmecillinam