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REVIEW ARTICLE

Antibiotherapy and pathogenesis of uncomplicated UTI: difficult relationships

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Abstract

In a time when conventional antibiotics are becoming increasingly less effective for treatment of infections, the relationship between bacteria and antimicrobial resistance is becoming more and more complicated. This paper provides a current review of studies reported in the literature pertaining to the antibiotherapy of human urinary tract infections (UTI), in a way that helps the reader direct a bibliographic search and develop an integrated perspective of the subject. Highlights are given to (bio)pathogenesis of uncomplicated cystitis. Features associated with the antibiotherapy of UTI such as development of resistance are presented in the text systematically. This review discusses recent advances in the understanding of how the predominant uropathogen *Escherichia coli* interacts with its host and leads to infection; so one can understand some of the reasons behind antibiotherapy failures.

Introduction

Cystitis, infection of the bladder, is responsible for *c.* 95% of all symptomatic urinary tract infections (UTI; Muhldorfer *et al.* 2001), which is one of the most prevalent infectious diseases. A population-based survey in 1995 estimated that annually between 7·3 and 11 million women in the United States of America had at least one presumed UTI treated with an antibiotic (Foxman *et al.* 2000). An estimated 130–175 million cases of UTI occur annually worldwide with huge social costs, and evaluation and management of UTI approximates US$1·6 billion (Foxman 2002). The figures can be higher if costs of self-diagnosed and self-medicated infections (Hooton and Stamm 1997; Gupta and Hooton 2004) are taken into account.

UTI is considered to be uncomplicated when it occurs in patients with urinary tracts that are normal from both a structural and functional perspective or complicated when it occurs in other settings (Hooton 2000; Cohn and Schaeffer 2004). Even episodes of acute uncomplicated cystitis are associated with considerable morbidity, including an average of 6·1 days of symptoms, 2·4 days of restricted activity and 0·4 bed days (Foxman and Frerichs 1985a,b).

Asymptomatic bacteriuria or asymptomatic urinary infection is defined as the presence of a specified quantity of bacteria in an appropriately collected urine specimen obtained from a person without symptoms or signs referable to urinary infection (Nicolle *et al.* 2005). Asymptomatic bacteriuria is common, but the prevalence in populations varies widely with age, sex and other host factors.

Overview of the pathogenesis of UTI

The urinary tract consists of paired organs, the kidneys and two ureters, a bladder and a urethra. The urinary bladder is a muscular organ, hollow and roughly spherical-shaped when filled with urine. The mucosa of the renal pelvis, ureter and urinary bladder is lined by stratified epithelium, the urothelium. Cuboidal-to-columnar cells form the basal layer and the superficial layer is made up of relatively large cells, which may take on an

'umbrella-like' shape owing to their outer convex shape. The urothelium also acts as an active component of the innate immune system, through the production of cytokines, chemokines and secretory immunoglobulin A, sIgA (Mulvey *et al.* 2001; Vaidyanathan *et al.* 2002).

Uropathogens can enter the urinary tract and cause UTI via the ascending route or the haematogenous route. The vast majority of UTI are thought to occur via the ascending route, and bladder infections are much more common than kidney infections (Hooton 2000; Scholes *et al.* 2005). Vaginal colonization is considered to be a prerequisite to bladder infection in women (Stamey and Sexton 1975). Sometimes, these ascending uropathogens reach the kidney and can cause pyelonephritis (in 5% of all UTI). The mechanism of ascension is uncertain, but motility mediated by flagella and pili appears to be important (Wright *et al.* 2005; Lane *et al.* 2007). Risk factors for pyelonephritis are similar to those for cystitis, including sexual intercourse, and genetic factors may play a role (Scholes *et al.* 2005; Rama *et al.* 2005).

Uropathogenic *Escherichia coli* (UPEC) cause 70–95% of all uncomplicated UTI (Hooton 2000) and 50% of healthcare-associated UTI (Hooton and Stamm 1997; Ronald 2002; Cohn and Schaeffer 2004). Other uropathogens causing uncomplicated UTI include *Staphylococcus saprophyticus* in 5–10%, and *Klebsiella pneumoniae*, *Proteus mirabilis*, group B streptococcus, and other bacteria, in less than 5% (Ronald 2002; Nicolle 2003).

Advances in cellular and molecular biology have improved the understanding of the micro-organism–host association, but much is still unknown. It is clear, however, that the strain pathotype is an important determinant of uropathogenicity (Johnson and Russo 2005). Strain pathotypes are characterized by sets of virulence factors that facilitate certain pathogenesis processes (Bekal *et al.* 2003; Marrs *et al.* 2005; Guy 2006). The virulence factors of UPEC are specific properties that confer on bacteria the ability to adhere to the urinary tract, persist and invade the host tissues causing injury (Johnson *et al.* 2005; Rama *et al.* 2005). Such factors also result in avoidance of host defence mechanisms and/or stimulation of a host inflammatory response (Johnson and Stell 2000). UPEC isolated from patients with UTI possess substantially more virulence factors (VF) on average compared with commensal faecal isolates (Johnson *et al.* 2005). The former typically carry large blocks of genes, called pathogenicity-associated islands (PAI), not generally found in commensal faecal isolates and are known to contribute to the pathogenicity of bacteria and their resistance to antibiotics (Hacker and Kaper 2000; Guyer *et al.* 2002; Johnson and Russo 2005). PAI were first described in the human pathogen *E. coli* (Hacker and Kaper 2000; Dobrindt *et al.* 2004). PAI encode a wide range of bacterial virulence factors from adhesins to toxins to host defence avoidance mechanisms (Hacker and Kaper 2000). For example, P fimbriae, which represent important adherence factors of UPEC, are encoded by UPEC-specific PAI. P-fimbrial genes (*pap* or *prs*) are often linked to gene clusters *hly* and *cfn*, respectively encoding the UPEC-specific toxins α-hemolysin and cytotoxic necrotizing factor 1, a linkage that argues for a strong co-evolution of these factors (Hacker and Kaper 2000). Moreover, some genes in plasmids and prophages help bacteria to increase their pathogenicity to a mammalian host. A very good example is the *bor* gene of lambda bacteriophage that confers protection of the bacterial host against serum complement killing (Koch 2007).

The boundary between commensalism and virulence results from a complex balance between the status of the host and the presence and expression of virulence factors in the bacteria (Picard *et al.* 1999).

UPEC attachment to the uroplakins of the host tissue uroepithelium constitutes the first step in the colonization of their hosts by the mannose-sensitive binding of filamentous fimbria. FimH is the adhesin present at the tip of type 1 pili that binds to the mannose-containing glycoprotein receptors expressed on the luminal surface of the bladder (Sussman and Gally 1999; Hung *et al.* 2002).

Studies using mice models have shown that instead of being confined to the lumen of the bladder, UPEC rapidly gain access to the urothelium and are quickly internalized where they proliferate in an intracellular sanctuary. The internalization of adherent UPEC through the FimH adhesin stimulates host cell signalling cascades and leads to the induction of cytoskeletal rearrangements (Mulvey *et al.* 2001). The resulting intracellular bacterial community (IBC) has biofilm-like properties that protect it from the host's innate immune response (Anderson *et al.* 2004; Wright *et al.* 2005) and is inherently tolerant to antibiotics (Cerca *et al.* 2005). Eventually, these IBC detach from the biofilm and disperse themselves into the bladder lumen, resulting in the spread of the bacteria over the epithelial surface, to initiate additional rounds of IBC formation (Anderson *et al.* 2004; Wright *et al.* 2005; Garofalo *et al.* 2007), in a manner reminiscent of a lytic virus cycle (Mulvey *et al.* 2001; Justice *et al.* 2004; Wright *et al.* 2005). Re-entry of detached bacteria into the IBC developmental cascade is marked by slower kinetics (Franco 2005; Wright *et al.* 2005). At this point, bacterial replication ceases and a quiescent reservoir or persistent state is established in the bladder tissue that, in response to nutrient and oxygen limitations in the interior of the microcolonies, can reactivate and trigger a recurrent/persistent bacteriuria (Corbin *et al.* 2001; Wright *et al.* 2005).

In the mouse model, IBC protect UPEC from: (i) innate host defences, such as phagocytosis and clearance by micturition, and (ii) antibiotic treatment. The IBC cascade has not been demonstrated in humans, but a recently published paper demonstrates evidence of IBC-like structures in exfoliated uroepithelial cells of women with acute cystitis (Rosen *et al.* 2007).

Micro-organism VF

Virulence is defined as the ability of an organism to cause disease in a particular host (Rama *et al.* 2005). VF help determine whether or not a strain can invade the host and allow for the survival of an organism in less propitious environments, as urine can be. A variety of virulence genes that encode VF are associated with UPEC (see Table 1), which possess substantially more VF than faecal *E. coli* isolates (Johnson *et al.* 2005). Uropathogenic strains are highly adapted and possess many factors that facilitate bladder colonization and survival in the urinary tract, and often the ability to cause tissue damage (Sussman and Gally 1999). In particular, UPEC infecting young men tend to be highly virulent (Hacker and Kaper 2000; Johnson and Stell 2000; Johnson *et al.* 2005). Table 1 shows the wide diversity of some of the important VF found in UPEC, together with important features associated therewith.

Bacterial adhesins are important and provide UPEC with the ability to adhere to the uroepithelium. Cytolethal distending toxin causes damage to the host. Haemolysin and k-antigen promote phagocytosis. Extracellular

Table 1 Diversity of some of the important virulence factors (VF) found in uropathogenic *Escherichia coli*

VF	Characteristics	References
Adhesins	Serve as ligands for glycoproteins and glycolipids on the surface of uroepithelial cells; prerequisite for penetration of invasive organisms	Rama *et al.* 2005
Pathogenicity-associated islands (PAI)	Genetically linked VF	Johnson and Russo 2005
SPATE (serine protease autotransporters of Enterobacteriaceae) – secreted autotransporter toxin (Sat)	A cytotoxin for bladder and kidney epithelial cells	Guyer *et al.* 2002
Haemolysin	Secreted by haemolytic *E. coli*: (i) lyses human erythrocytes; (ii) contributes to inflammation; (iii) causes tissue injury; and (iv) weakens chemotaxis and phagocytosis	Rama *et al.* 2005
Siderophores – aerobactin and enterochelins	Extracts iron from the host for metabolic activities	Rama *et al.* 2005
traT gene	Confers resistance to serum bactericidal activity	Kanukollu *et al.* 1985
Lipopolysaccharide (LPS) somatic antigen (O-antigen)	Highly immunogenic endotoxin that activates complement via alternate pathway-releasing cytokines, chemokines, etc., leading to an acute inflammatory response	Rama *et al.* 2005
Capsular polysaccharide (K-antigen)	Interferes with O-antigen detection, protecting the bacterium against phagocytosis; poorly immunogenic	Rama *et al.* 2005
Cytotoxic necrotizing factor types 1 and 2 (CNF 1/2)	Members of the family of bacterial toxins that target the Rho family of small GTP-binding proteins	Schmitt *et al.* 1999
Cytolethal distending toxin	Causes progressive cell distending leading to cell death by direct DNA damage of host cells	Mulvey *et al.* 2001
P-fimbriae	The most important and widely studied mannose-resistant adhesins of uropathogenic *E. coli*. These fimbriae bind to the host cells through galactosyl (α,1-4)-galactose-β-dissacharide galbiose receptors, which are part of the P blood group system antigens. P-fimbriated strains are associated with more clinically severe infections	Rama *et al.* 2005
Type-I fimbriae	Mediate mannose-sensitive bacterial attachment to the bladder epithelium via interaction with glycoproteins rich in D-mannose. Type-I fimbriae bind to Tamm-Horsfall-proteins (THP), which often coat uroepithelial cells and prevents bacterial adherence to the urinary mucosa and facilitate the expulsion of bacteria	Rama *et al.* 2005

generation of reactive oxygen species (ROS) is induced by bacterial strains that resist ingestion by phagocytes (Rama *et al.* 2005). Sat is a serine protease that promotes cytopathic effects similar to those inducing vacuolization within the cytoplasm of the human urinary tract (Guyer *et al.* 2002). It is the only known vacuolating toxin of *E. coli* strains isolated in human urine. Recently, it has been classified as a member of the Serine Protease Auto-Transporter of the Enterobacteriaceae (SPATE) family. All phenotypes associated with the SPATE proteins enable bacteria to damage the host and evade the immune response (Guyer *et al.* 2002).

Environmental factors

The urinary tract is an ecological niche where structural, functional and physiological factors play a role in the genesis of infection. Urine is a good bacterial broth with physiological values of pH and osmolarity allowing for rapid bacterial growth. Human urine can promote the growth of UPEC bacteria as its composition includes a variety of nutrients (e.g. glucose, amino acids and uric acid), but it is a poor source of iron (Schwan *et al.* 2002).

However, other components of urine, such as urea and organic acids, may inhibit microbial growth when in high concentrations (Sussman and Gally 1999). Although little is known about the environmental regulation of bacterial toxin production in the urinary tract, haemolysin expression in *E. coli* appears to respond to osmolarity, temperature and anaerobic conditions (Sussman and Gally 1999).

Host factors

Sexual intercourse is one of the most important risk factors associated with the risk of uncomplicated UTI (Hooton 2000). In uncomplicated UTI, anatomy plays an important role with respect to the large difference between men and women in UTI risk. The drier environment surrounding the male urethra, especially with circumcision, prevents the optimal growth of bacteria compared with the female urethra. The antibacterial activity of prostatic secretions in men is also a factor that reduces the risk of UTI in men. In men, there is also a much longer distance between the anus and the urethral meatus (Hooton 2000). Genital microflora plays an important role in women's defenses against invasions, so any alterations (e.g. due to antibiotherapy and its adverse effects upon the genital ecology) can enhance genital colonization with uropathogens and risk of subsequent UTI (Hooton 2000).

Host factors such as the epithelial cell receptivity are also important in the onset of infection (Table 2). For example, *E. coli* binds to vaginal epithelial cells from healthy controls less avidly than to vaginal epithelial cells from women with recurrent UTI (Cohn and Schaeffer 2004). Vaginal cell receptivity also varies as a function of hormonal status. Bacterial adherence tends to be higher earlier in the menstrual cycle and in postmenopausal women as compared with the pre- women or postmenopausal women who are on oestrogen replacement therapy (Cohn and Schaeffer 2004) predisposing them to recurrent UTI (Franco 2005). Presence of diabetes mellitus has been shown to enhance the frequency of recurrent UTI by two- to threefold (Franco 2005). Several approaches can be employed to prevent recurrences of UTI: long-term prophylaxis, postcoital therapy and patient-initiated therapy (Franco 2005).

Determinants of host response to therapy

There are numerous reasons why a patient may not respond to antibiotic therapy, many of which are related to the inappropriate selection of antibiotics for treating UTI (Nicolle 2001). For example, in men with UTI, infection of the prostate requires treatment with antimicrobials that penetrate the prostate. In patients with upper tract infection, nitrofurantoin is inappropriate because it does not achieve adequate tissue concentrations.

Determinants of bacterial response to therapy

Antibiotic resistance complicates the treatment of UTI and is associated with a higher patient morbidity, higher costs of re-evaluation and re-treatment, higher rates of hospitalization and greater use of broader-spectrum antibiotics (Hooton *et al.* 2004). The selective pressure promoted by antimicrobial drugs in the various environmental settings such as nursing homes, hospitals and long-term care facilities, contributes for the selection and spread of resistant clones.

A population of organisms can lose its sensitivity to an antibiotic while the patient is under treatment. The development of new antimicrobial drugs has counterbalanced this trend. However, we are currently approaching a microbial resistance level never attained before, which might pose a serious risk to the effectiveness of the current antibiotic treatments in the near future (Wagenlehner and Naber 2006). An understanding of the mechanisms by which uropathogenic micro-organisms manifest resistance to antimicrobials (both intrinsic and acquired resistances) is necessary to optimize treatment strategies for UTI.

Intrinsic resistance

Intrinsic resistance is an inherent insensitivity of the bacterium to the antibiotic action. Some gram-negative

Table 2 Host susceptibility factors that promote urinary tract infection (UTI)

			References
Gender	Women	Pregnancy	Nicolle *et al.* 2005
		Sexual activity	Foxman *et al.* 2000; Nicolle *et al.* 2005; Nicolle 2001
		Oestrogen deficiency in postmenopausal women	Cohn and Schaeffer 2004
		Genetic predisposition to urinary tract infection – vaginal cell receptivity	
	Men: obstructive uropathy in older men		Nicolle *et al.* 2005
Age	Middle-age: higher incidence in women aged 25–50 years, due to sexual activity		
Indwelling catheters	Chronic indwelling catheters	Use of a long-term indwelling catheter is associated with bacteriuria virtually 100% of the time	Nicolle *et al.* 2005
	Acute indwelling catheters	While catheter remains *in situ* bacteriuria is acquired at the rate of 2–7% per day	Nicolle *et al.* 2005
	Encrustation of biomaterials employed in the urinary tract is a major problem resulting in the obstruction or blockage of catheters or stents		Tunney *et al.* 1997
Diabetics	Women: bacteriuria is more common in diabetic women, with a prevalence of 8–14%, and is usually correlated with duration of diabetics and presence of long-term complications of diabetics		Nicolle 2005
	Men: diabetic men do not appear to have an increased prevalence of bacteriuria, compared with nondiabetic men		Nicolle 2005
Sexual intercourse	The prevalence of bacteriuria among young women is strongly associated with sexual activity		Foxman *et al.* 2000; Hooton 2000; Johnson *et al.* 2005
Patients with spinal cord injuries	Neurogenic bladder with chronic or intermittent catheterization		Ronald 2002; Nicolle *et al.* 2005
Increase in renal/ bladder/prostatic calculi	Causing prostatic hypertrophy and fluxing disorders like turbulent urine flow and urethral obstruction		Nicolle 2001
Iatrogenic genitourinary instrumentation	Women: application of diaphragm		Russell and Love 1991
	Cystourethroscopy		Franco 2005
Care units	Nosocomial environment		
Birth control methods	Oral contraceptives or condoms and diaphragms or cervical caps had a significantly higher incidence of risk of reinfection		Foxman *et al.* 2000
Antibiotic use in the previous 2 weeks of a UTI	Women who reported taking antibiotics during 2 weeks prior to the follow-up survey had a higher rate of second UTI		Foxman *et al.* 2000; Nicolle *et al.* 2005; Franco 2005

bacteria, including Enterobacteriaceae exhibit intrinsic resistance to cephalosporins by producing AmpC β-lactamases (Martinez *et al.* 1998). Another example is the intrinsic resistance of *Pseudomonas* to benzylpenicillin.

Acquired resistance

Acquired resistance represents a major threat to human health because of its high prevalence in major pathogens causing human diseases (Table 3). Acquired resistance is mainly because of: (i) a genetic change in the bacterial genome, which can be the consequence of a mutation of chromosomal target genes, or (ii) exogenous resistance acquired from horizontal acquisition of foreign resistance genes.

Horizontally transferable resistance is because of transfer of: (i) plasmids (R-factor), (ii) integrons and gene cassettes, (iii) bacteriophages, as a prophage (Tinsley *et al.* 2006) and (iv) chromosomal genomic islands, which play a major role in the horizontal transfer of antimicrobial resistance to one or more antimicrobial drugs.

Studies indicate that uropathogens are becoming increasingly resistant to the antibiotics used for the treatment of UTI. As an example, SENTRY, an Antimicrobial

Table 3 Characterization of the resistance to antibiotics

	Type of resistance							
		Acquired						
Antibiotic	Intrinsic	New genes	Gene^{R}	Indications for use	Restrictions to use	Duration of therapy	Resistance (%)	References
Sulfametoxazol-trimethoprim	Not available	Transmission of R-factor, plasmid-mediated	*SulI, SulII* and *Sul3* *dfr*9 and *dfr*1	First-line treatment	Pregnancy	Three days* Ten days* – reduces recurrences	9–15% in many countries; nearly 35% in Portugal and Spain	*Schilling *et al.* 2002; Kahlmeter 2003; Kahlmeter and Menday 2003;
Fluoroquinolones	Not available	In 2006, plasmid-mediated quinolone resistance was reported, with the acquisition of the gene *qnr*	*gyr*A, *gyr*B, *par*C, *qnr*	Allergies to other drugs, in patients with underlying diseases that predispose to serious infection	Pregnancy, children, athletics with high competition Not as first-line therapy	Three days	Less than 5% is United States; southern Europe, 20%; more than 20% in Spain and Portugal	Kahlmeter 2003; Kahlmeter and Menday 2003; Paterson 2006
Nitrofurantoin	Not available	Does not demonstrate R-factor-mediated resistance	Not available	First-line treatment in UTI during pregnancy Should be considered as a fluoroquinolone-sparing alternative to sulfametoxazol-trimethoprim	Can cause acute and chronic pulmonary syndromes	Five or more days	2% among *Escherichia coli* and higher among some other strains	Hooton 2003a; Nicolle *et al.* 2005
β-lactams	β-lactamase production	Transmission of R-factor, plasmid-mediated	*rpo*B, *oxa*2, *pse*1 and bla_{TEM-1} β-lactamase	First-line treatment in UTI during pregnancy Second-line agents	Not available	Seven days	40% ampicillin – Spain, Portugal, Ireland and Luxembourg. 28–59% for cefalotin, amoxicillin/clavulanate	Kahlmeter and Menday 2003; Ponte *et al.* 2005; Paterson 2006
Fosfomycin	Not available	Genes found in cassettes recovered from class 1 integrons**	*Fos A, Fos B, Fos X***	Second-line agents	Not available	Single dose	Not available	**Bernat *et al.* 1999; **Cao *et al.* 2001; Ribeiro *et al.* 2002; **Fillgrove *et al.* 2003

UTI, urinary tract infection.

Surveillance Program that monitored UTI worldwide over a 4-year period between 1997 and 2000 (Turnidge *et al.* 2002), showed an increase in resistance among UTI isolates (Turnidge *et al.* 2002). The frequency of detection of extended-spectrum β-lactamases (ESBL)-producing strains (including *E. coli*) is quite variable but is consistently increasing (Sader *et al.* 1994). The growing levels of resistances to broad-spectrum antibiotics such as fluoroquinolones is recognized as a significant problem in some medical centres and some clinical studies have documented this problem throughout the world (Gales *et al.* 2000).

Strains of community-acquired ESBL causing UTI are often multidrug-resistant, which complicates treatment and are often not detected early (Paterson 2006). The extent of ESBL-producing strains including *E. coli* is quite variable between medical centres and this most likely is related to differing prescription practices and/or infection control practices that can lead to the spread of resistant clones and can in turn spread resistance, through gene transfer, to other related species (Sader *et al.* 1994; Pitout *et al.* 1998; Turnidge *et al.* 2002).

Antimicrobial therapy in uncomplicated UTI

The goal of treatment of an uncomplicated UTI is the resolution of symptoms and sterilization of the urine. Antimicrobial drugs used for UTI therapy should have certain characteristics: (i) they should be active against the most common uropathogens; (ii) they should be excreted in the active form of drug by glomerular filtration into urine; (iii) they should have both adequate concentrations and inhibitory levels in urine (and tissue when treating pyelonephritis); and (iv) they should be active at urinary pH values. Sulfametoxazol-trimethoprim, fluoroquinolones, β-lactams, nitrofurantoin and fosfomycin are the most common antimicrobial agents used in the therapy of UTI (Karlowicz 1997; Gupta *et al.* 2001). Of note, susceptibility breakpoints from the Clinical and Laboratory Standards Institute (CLSI, formerly NCCLS) are based on serum rather than urine concentrations of these antimicrobial agents, except for nitrofurantoin and fosfomycin, which are exclusively used for treatment of cystitis.

Human studies have shown that sulfametoxazol-trimethoprim is less effective against sulfametoxazol-trimethoprim-resistant uropathogens causing cystitis (Gupta *et al.* 2007) or pyelonephritis (Talan *et al.* 2000).

Sulfametoxazol-trimethoprim

Sulfonamides and trimethoprim, also called co-trimoxazole, inhibit the synthesis of tetrahydrofolic acid, which is necessary for DNA synthesis in bacteria. Sulfonamides are structural analogues of the normal substrate *p*-aminobenzoic acid. The enzyme dihydropteroate synthase (*dhps*), an essential enzyme in all living cells, catalyses the formation of dihydropteroic acid in bacteria, a step before tetrahydrofolic acid formation. Sulfonamides act as competitive inhibitors of *dhps*, thereby blocking tetrahydrofolic acid biosynthesis in the bacterial cell. Trimethoprim is a structural analogue of tetrahydrofolic acid and competitively inhibits the reduction of dihydrofolate to tetrahydrofolate by dihydrofolate reductase (*dhfr*). As bacteria are unable to take up folic acid from the environment and are thus dependent on their own *de novo* synthesis, inhibition of folic acid synthesis prevents DNA replication in the bacteria.

Only three genes encoding resistance to sulfonamides are known – *SulI*, *SulII* and *Sul*3 – and the most recent one was discovered only 3 years ago (Guerra *et al.* 2004) (please see Table 3). Mutational changes in the chromosomal gene *folP* that encodes dihydropteroate synthase results in a lowered affinity for sulfonamide (Skold 2001; Fiebelkorn *et al.* 2005). There were 111 altered nucleotides resulting in 30 amino acid changes when *folP* in susceptible and resistant strains were compared (Skold 2001).

Chromosomal resistance to trimethoprim can be because of mutational changes in the intrinsic dihydrofolate reductase *dhfr* gene. Dihydrofolate reductase converts dihydrofolate into tetrahydrofolate, a methyl group shuttle required for the *de novo* synthesis of purines, thymidylic acid and certain amino acids.

Trimethoprim resistance in the 1970s rarely occurred in more than 10% of the UPEC isolates, but in the 1980s increased to 15–20%, with the exception of South America, Asia and Africa where levels of 25–68% were reported (Urbina *et al.* 1989; Wylie and Koornhof 1989; Lamikanra and Ndep 1989).

Guidelines published in 1999 by the Infectious Diseases Society of America (IDSA) recommend sulfametoxazol-trimethoprim as the first-line treatment for acute cystitis. There are numerous other UTI treatment guidelines worldwide that also list sulfametoxazol-trimethoprim as the drug of choice for first-line therapy of acute cystitis. However, resistance to sulfametoxazol-trimethoprim has been increasing, raising concerns about its role as a first-line agent for UTI treatment (Hooton *et al.* 2004; Karaca *et al.* 2005), and leading some clinicians to consider sulfametoxazol-trimethoprim as a first-line agent only in women who do not have underlying risk factors such as older age or diabetes (Gupta *et al.* 2001).

A European survey of a well-defined population of women with acute uncomplicated cystitis (the ECO-SENS Project, conducted during 1999–2000) found

sulfametoxazol-trimethoprim resistance in 9–15% of the *E. coli* isolates in all countries with the exception of Spain and Portugal, where the rate was nearly 35% (Kahlmeter 2003). Over the last decade, the treatment of choice for UTI in Turkey has changed from sulfametoxazol-trimethoprim to fluoroquinolones owing to the rate of resistance to cotrimoxazole and its high level of therapeutic failure (Karaca *et al.* 2005).

The increase in prevalence of sulfametoxazol-trimethoprim resistance among UPEC in the United States is in part related to clonal spread. Clusters of UTI-causing uropathogen clones were identified during the investigation of antibiotic-resistant strains, as they exhibit an concurrent resistance to ampicillin, chloramphenicol, streptomycin, sulfonamides, tetracycline and trimethoprim (ACSSuTTp; Johnson *et al.* 2002). It is possible that many community-acquired UTI occur in epidemiologically linked clusters that are not identified, because urine cultures are often not performed for cystitis and isolates and, when present, are not genotyped or serotyped (Hooton and Samadpour 2005).

Quinolones

Quinolone drugs are bactericidal compounds that act by interfering with DNA and RNA synthesis, and fluoroquinolones inhibit DNA synthesis by inhibiting DNA gyrase, interfering with DNA replication, repair and transcription before ultimately causing bacterial cell death (Karlowicz 1997; Bearden and Danziger 2001). In gram-negative organisms, DNA gyrase is the primary target, whereas in gram-positive bacteria, topoisomerase IV was recently found to be most affected (Bearden and Danziger 2001).

The first clinically available quinolone, nalidixic acid, penetrated poorly into the tissues, and was used only for the therapy of gram-negative UTI. Several newer fluoroquinolones have broader spectrum of activity and more optimal tissue levels and, thus, are used for a wide variety of both uncomplicated and complicated infections. However, the use of fluoroquinolones is contraindicated in pregnant women and children or adolescents, because of potential damage in cartilage formation, high-performance athletes, as they can cause tendonitis and rupture of the Achilles tendon even after a short-term use; and patients with QT interval prolongation or pre-existing rhythm disorders. Although, as a class, fluoroquinolones are not nephrotoxic, their dosage must be adjusted when administered to patients with renal failure.

Fluoroquinolones are often considered agents of first choice in patients with UTI because of the increasing prevalence of resistance of *E. coli* to sulfametoxazol-trimethoprim. Ciprofloxacin, levofloxacin and norfloxacin are increasingly being used, even in uncomplicated UTI. Ciprofloxacin can often be considered the first-choice antimicrobial in UTI patients with allergies to other drugs, in the elderly, in patients with recurrent infections and in diabetics (Schilling *et al.* 2002; Hooton 2003a). However, increasing fluoroquinolone resistance is a serious public health threat, and it is essential that the indiscriminate use of these agents is avoided, if we are to preserve the efficacy of these drugs to critical diseases. For example, the prevalence of fluoroquinolone resistance in UPEC causing uncomplicated UTI is very high in some parts of the world, especially in southern Europe, where it has been reported to be as high as 20% (Kahlmeter 2003).

Fluoroquinolone resistance in Enterobacteriaceae is most frequently caused by chromosomal mutations. More recently, however, plasmid-mediated quinolone resistance has emerged in *E. coli*. The emergence of this new plasmid-mediated quinolone resistance provides a mechanism for the rapid spread of quinolone and multidrug resistance to important members of the Enterobacteriaceae (Hooton 2003b; Paterson 2006).

Given concerns that increasingly widespread use of fluoroquinolones will promote bacterial resistance, their use for the routine treatment of acute uncomplicated cystitis should be discouraged (Kahlmeter 2003; Hooton *et al.* 2004; Gupta *et al.* 2005).

β-lactams

All β-lactams produce their bactericidal effects via inhibition of bacterial cell wall synthesis (Karlowicz 1997).

Aminopenicillins

Ampicillin and amoxicillin have been extensively used in the therapy of cystitis. However, the increased prevalence of resistance of uropathogens to this class of drugs, primarily via β-lactamase production has resulted in decreased usage. β-lactamase is an enzyme produced by some bacteria that inactivates β-lactam antibiotics. Plasmidic β-lactamases are widespread and, of these, TEM-1 is present in 40% of all gram-negative bacterial isolates. The activity of amoxicillin/clavulanic acid against *E. coli* greatly depends on the level of β-lactamase production by the strain. Thus, it is necessary to obtain *in vivo* concentrations of clavulanic acid at sufficient concentrations that inhibit β-lactamase production by *E. coli* at the infection site (Ponte *et al.* 2005).

In recent studies, the prevalence of resistance to ampicillin has ranged from 26% to 38% in the United States (Paterson 2006). The ECO.SENS study showed that *E. coli* is now resistant to ampicillin in more than 40% of all cases in Spain, Portugal, Ireland and Luxembourg (Kahlmeter 2003). Moreover, the number of UPEC strains displaying intermediate resistance to amoxicillin/clavulanic

acid might be increasing, which could have important detrimental consequences in countries such as Spain, where the rates of resistance of *E. coli* to fluoroquinolones is as high as 22·8% from community-acquired UTI isolates (Hooton 2003b; Ponte *et al.* 2005). *Candida* vaginitis is common secondary to the use of aminopenicillins.

Amoxicillin/clavulanic acid has greater activity against resistant UPEC strains, but is more expensive and produces more side effects (such as predisposing the patient for relapses) because these antibiotics greatly alter the normal genital microflora (Hooton 2000). In addition, amoxicillin-clavulanate does not perform, as well as fluoroquinolones, for the treatment of uncomplicated cystitis, even in patients infected with strains susceptible to the drug (Hooton *et al.* 2005).

Cephalosporins

Activity of cephalosporins against *E. coli* increases from the first to the fourth generation of these drugs, but the newer classes are more expensive and there are fewer oral alternatives. However, the number of resistant strains to cephalosporins has been increasing recently (Ribeiro *et al.* 2002). Enterobacteriaceae resistance to third-generation cephalosporins is because of the production of β-lactamases (Paterson 2006). According to the National Nosocomial Infections Surveillance (NNIS) System report in 2003, 5·8% of *E. coli* isolated from patients in intensive care units in the United States, were nonsusceptible to third-generation cephalosporins owing to the production of β-lactamases (Paterson 2006). Cephalosporins are a useful option for the treatment of uncomplicated UTI during pregnancy because of their safety (Nicolle 1996).

Amidinopenicilins

Pivmecillinam is the pro-drug of mecillinam, a penicilin analogue that differs from other β-lactams in that the β-lactam ring is linked to the side chain by an amidino instead of an amindo group (Anderson *et al.* 1976). It possesses activity against gram-negative micro-organisms such as *E. coli* and other Enterobacteriaceae (Graninger 2003), and has higher activity when compared with amoxicillin (Anderson *et al.* 1976). Mecillinam binds only to PBP2 of gram-negative bacilli leading to cell lysis. A study conducted in 16 European countries and Canada found a resistance of *E. coli* of 1·2% (Kahlmeter 2003). The high and increasing levels of resistance among *E. coli* to currently recommended first-line agents for acute cystitis requires a re-evaluation of treatment guidelines. With a low prevalence of resistance, good efficacy (Ferry *et al.* 2007) and not having major effects on the anaerobic intestinal microflora (Edlund and Nord 2000), pivmecillinam is a suitable first-line agent for empirical treatment of acute cystitis.

Nitrofurantoin

Nitrofurantoin has been in clinical use for more than 50 years now, and because antimicrobial levels are not attained in the blood its only indication is for uncomplicated cystitis (Hooton 2003a). It is readily absorbed from the gastrointestinal tract and rapidly excreted by the kidneys. Its mechanism of action is inhibition of bacterial acetyl coenzyme A with the disruption of cell wall formation (Karlowicz 1997). Nitrofurantoin is reduced by bacterial flavoproteins to reactive intermediates, which inactivate or alter bacterial ribosomal proteins and other macromolecules. This inactivation or alteration of bacterial ribosomal proteins and macromolecules inhibits aerobic metabolism and, as a result, the syntheses of bacterial deoxyribonucleic acid (DNA), ribonucleic acid (RNA), cell wall and protein. This antibiotic has maintained excellent activity against almost all *E. coli* strains causing uncomplicated UTI, but some uropathogens, such as *Proteus* and *Klebsiella* are resistant. Nitrofurantoin does not appear in measurable amounts in the stool and, consequently, commensal flora remain unaffected. Moreover, it has recently been shown to be as effective in a 5-day course as sulfametoxazol-trimethoprim in a 3-day course for the treatment of uncomplicated cystitis (Gupta and Hooton 2004). Nitrofurantoin should therefore be considered as a fluoroquinolone-sparing alternative to sulfametoxazol-trimethoprim for uncomplicated cystitis (Hooton *et al.* 2004). There is some concern regarding the safety of nitrofurantoin, because of acute or chronic pulmonary syndromes, but these are rarely seen with short-course regimens used for treatment of uncomplicated UTI (Hooton 2003a).

Fosfomycin

Fosfomycin is a phosphoenolpyruvate analogue that irreversibly inhibits enolpyruvate transferase (*MurA*), which prevents the formation of *N*-acetylmuramic acid, an essential element of the peptidoglycan cell wall. Fosfomycin penetrates into the cells through a system of permeases; any mutation on these permeases can prevent the antibiotic from penetrating the cell. Fosfomycin shows reduced activity at pH >6·4. Resistance to fosfomycin is because of several mechanisms, both chromosomal- and plasmid-borne. Chromosomic resistance can be because of the production of a constitutive enzyme (fosfomycin-glutation-*S*-transferase) that inactivates the antibiotic in the periplasm. Plasmid-mediated resistance is to the result of enzymatic modifications of the antibiotic (Llaneza *et al.* 1985) A 3-g sachet dose is generally effective and well tolerated, with diarrhoea being the most common side effect. Fosfomycin is moderately active against *E. coli* strains and many other uropathogenic micro-organisms

that cause uncomplicated UTI. Urinary levels persist over 48 h, but bacterial resistance may develop quickly. Fosfomycin therapy is one of the most expensive antibiotherapies (Hooton *et al.* 2004).

Conclusions

The increasing frequency of bacterial drug resistance has been attributed to combinations of microbial characteristics, selective pressure of antimicrobial use and both social and technological changes that enhance the transmission of drug-resistant micro-organisms. The overall increase in the life expectancy of human beings and, consequently, of the number of persons with problematic clinical conditions such as the use of urinary catheters and chronic underlying diseases such as cancer, renal insufficiency, immunocompromised (HIV) status and diabetes, have led to a growing number of persons at risk for developing UTI. Prevention and control of bacterial resistance will require new antimicrobial agents, the prudent use of existing ones, new vaccines and enhanced public health efforts to reduce transmission of bacterial resistance.

Antimicrobial treatment of UTI has changed markedly over the last decade. The emergence of bacterial resistance suggests the need for an emphasis on understanding the local patterns of drug resistance, avoiding fluoroquinolones as much as possible to reduce risk of resistance, using sulfametoxazol-trimethoprim as the first-line treatment unless the patient has risk factors to suggest resistance and, e.g. by using more nitrofurantoin as a fluoroquinolone-sparing agent for uncomplicated cystitis.

Future perspectives

Increasing resistance to first-line antibiotics, particularly sulfametoxazol-trimethoprim, has progressively complicated the management of UTI, therefore urging revised treatment approaches. Tailoring the principles reviewed here to individual patients can help the practicing clinician in meeting these challenges. Considerable effort has to be devoted towards reducing the emergence of micro-organisms resistant to antimicrobial drugs, to simplify management, to improve the cost-effectiveness of treatments and to enhance the patient's compliance to the treatment by investigating the efficacy of alternative treatments.

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