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Research Article

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Study of the Impact Antibiotic Combinations Used in Urinary Tract Infections on the Effectiveness of Antimicrobial Therapy

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ABSTRACT

Biofilm-associated urinary tract infections (UTIs) represent a major therapeutic challenge due to their high tolerance to antimicrobial agents and frequent recurrence. The increasing prevalence of multidrug-resistant uropathogens necessitates the evaluation of alternative treatment strategies, including antibiotic combination therapy. The objective of this study was to assess the antibiofilm activity of selected antibiotics used individually and in combination against biofilms formed by clinically relevant uropathogens. The scope of the study included the evaluation of ciprofloxacin, nitrofurantoin, amikacin, and imipenem against biofilms of Escherichia coli, Pseudomonas aeruginosa, Proteus mirabilis, and Enterococcus faecalis isolated from patients with urinary tract infections. Biofilms were formed on polystyrene microtiter plates and exposed to a range of antibiotic concentrations, applied both as monotherapy and in combinations. Biofilm biomass reduction was quantified spectrophotometrically using a crystal violet staining method. The reduction result was expressed as a percentage relative to the positive control, not exposed to antibiotics. The results demonstrated that antibiotic monotherapy produced only moderate reductions in biofilm biomass, with efficacy strongly dependent on bacterial species and antibiotic concentration. Ciprofloxacin and nitrofurantoin reduced biofilms of E. faecalis, P. aeruginosa, and E. coli by up to approximately 45%, while amikacin and imipenem showed variable activity against E. coli and P. mirabilis. In contrast, antibiotic combinations exhibited enhanced antibiofilm effects. The combination of ciprofloxacin with nitrofurantoin resulted in synergistic biofilm reduction, particularly against P. aeruginosa and E. coli. The most pronounced effect was observed for the imipenem–amikacin combination against P. mirabilis, achieving biofilm biomass reduction exceeding 80%. In conclusion, the findings indicate that antibiotic combination therapy is significantly more effective than monotherapy in reducing mature bacterial biofilms formed by uropathogens. These results support the potential clinical utility of rationally selected antibiotic combinations in the management of biofilm-associated urinary tract infections.

Keywords: Biofilm, Urinary Tract Infections, Uropathogens, Ciprofloxacin, Nitrofurantoin, Amikacin, Imipenem

Introduction

Antibiotics are commonly used antimicrobial agents in clinical practice. Antibiotic pressure limits the growth of bacterial cells and their more complex forms [1,2]. Proper use is essential for effective therapy. Too short a period of antibiotic therapy will not allow for achieving therapeutic concentrations and achieving a bacteriostatic effect. It is generally accepted that antibiotic therapy should last 7 days, with the possibility of extending it to 14 days. Overuse and inappropriate use of this therapeutic method contribute to the growth of antibiotic resistance in bacteria [3]. When microorganisms do not respond to treatment, the infection persists and can spread. Moreover, planktonic cells adhere to the substrate and begin to produce a protective extracellular matrix, ensuring high resistance to antibiotics and components of the patient's immune response. These

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multicellular bacterial structures are particularly associated with urinary tract infections [1,2].

Urinary tract infections (UTI) are the most common bacterial infection, which due to anatomical structure is much more common in women than in men [4,5,6]. The term UTI encompasses clinical entities such as asymptomatic bacteriuria, catheter-associated urinary tract infection, and acute cystitis. UTI causes serious consequences and recurrences and can lead to, among other things, kidney damage [6,7]. Colonization of the urinary tract depends on environmental factors such as pH, osmolarity, access to nutrients, oxygen tension, and adhesion site. Urine provides excellent conditions for microbial growth. Urine is usually acidic, and urine flowing through the urethra provides a source of nutrients essential for the growth of the microbiome [8].

Biofilm formation is a key element in the pathogenesis of urinary tract infections (UTIs), particularly catheter-associated infections (CAUTIs), which account for approximately 40% of hospital-acquired infections. Bacterial biofilms significantly contribute to UTI recurrence and increased resistance to antimicrobial therapy. UTIs can be caused by both Gram-negative and Gram-positive bacteria [9,10], with the most common pathogens including Gram-negative rods of the Enterobacterales order; Escherichia coli, Proteus mirabilis, Klebsiella pneumoniae, and Pseudomonas aeruginosa. Among the etiological factors of urinary tract infections, Gram-positive bacteria also include Staphylococcus epidermidis, Staphylococcus aureus, and Enterococcus faecalis [10].

The dominant pathogens isolated from patients are Gram-negative bacilli of the order Enterobacterales. In addition to Escherichia coli strains, bacteria of the species Proteus mirabilis are also important. Increasingly, microorganisms isolated in the etiology of hospital-acquired UTIs include Serratia spp., Enterobacter spp., Klebsiella spp., and non-fermenting bacilli: Pseudomonas spp. and Acinetobacter spp. Among the etiological factors of urinary tract infections, we also distinguish Gram-positive bacteria, such as Enterococcus spp. and Staphylococcus spp. [9,11]. Escherichia coli is the dominant pathogen in urinary tract infections (UTIs), accounting for approximately 80% of uncomplicated infections in humans [12]. Bacilli P. mirabilis is the main pathogen responsible for complicated UTIs, particularly CAUTIs [13]. Opportunistic pathogen as an, P. aeruginosa causes chronic infections. Due to its metabolic diversity and ability to form biofilms on various surfaces, this bacterium is responsible for 10% of CAUTIs in hospitalized patients. The continuous increase in the number of multidrug-resistant (MDR) strains and the lack of innovative, effective drugs create a need to seek new solutions. Sequential exposure of a microorganism to a combination of two antibiotics can reduce the risk of cells acquiring resistance, thus inhibiting their growth and development [1,2]. Synergy with conventional antimicrobial agents can modify and inhibit the mechanisms of acquired bacterial resistance. This may allow for the use of antibiotics at lower concentrations with therapeutic success, potentially reducing the adverse effects associated with their use [14]. The conducted studies tested combinations of antibiotics routinely used in the treatment of urinary tract infections associated with the isolation of Gram-negative bacilli and Gram-positive cocci. These include ciprofloxacin, nitrofurantoin, amikacin, and imipenem.

Nitrofurantoin is a first-line drug for the treatment of UTIs, but it is effective only in the lower urinary tract due to its poor tissue penetration. It is the most commonly used antibiotic for long-term, low-dose prophylactic therapy in patients with recurrent infections [15]. Ciprofloxacin, a fluoroquinolone, is an antagonist to nitrofurantoin. Therefore, they should not be used in combination. Fluoroquinolones are a class of antibiotics to which microorganisms often demonstrate resistance. Nevertheless, they are the drug of choice for the treatment of pyelonephritis and prostatitis due to their excellent penetration into deeper tissues. One of the most common etiological agents of UTIs, E. coli, often develops ESBL resistance, demonstrating resistance to cephalosporins, penicillins, fluoroquinolones, and tetracyclines. An alternative drug is amikacin from the aminoglycoside group, and in the case of multidrug resistance, carbapenems are the choice. A disadvantage of aminoglycoside use is the side effects, which are significantly less common than those of carbapenems, which include imipenem. Furthermore, the recent incidence of microbial intolerance to aminoglycosides has been low. The use of carbapenems in therapeutic strategies stems from the lack of susceptibility of bacterial strains to other drug classes. Therefore, they are most often used in hospitalized patients, but caution should be exercised, as there is a risk of bacterial resistance and the spread of infection with multidrug-resistant strains [16].

Four Drug Combinations were used in the Study

Ciprofloxacin X Amikacin: E.Coli, p. Aeruginosa

The combination of these antibiotics appears particularly promising, especially in the treatment of infections associated with colonization of medical devices. Ciprofloxacin is characterized by high bioavailability and excellent penetration into deeper tissues, including the deepest layers of bacterial biofilm. Moreover, the primary action of ciprofloxacin is based on the inhibition of DNA gyrase. This prevents double-stranded DNA from closing, leading to exonucleolytic degradation. This inhibits the synthesis of proteins, enzymes, and bacterial toxins, leading to bacterial cell death [17]. Amikacin also inhibits bacterial protein synthesis. It exerts its effect by irreversibly binding to the bacterial 30S ribosomal subunit, leading to erroneous mRNA reading [18]. Thanks to this amikacin is the most widely acting aminoglycoside and according to the latest guidelines, recommended for the treatment of acute P. aeruginosa infections. It is also the least nephrotoxic antibiotic [19].

Ciprofloxacin X Nitrofurantoin: E. Faecalis

This pair of antibiotics is not used together in clinical practice. Analyzing their combination against uropathogenic strains is a novel idea. Nitrofurantoin is a promising antibiotic with potent bactericidal properties. Due to its intracellular reduction in bacterial cells to reactive metabolites that damage DNA, RNA, proteins, and cell membranes, it leads to rapid bacterial cell death [20]. Moreover, compared to ciprofloxacin, resistance to nitrofurantoin is significantly less common [21]. Thanks to this combination, the antibiotic complex has the potential to penetrate bacterial biofilms, which would prevent resistance when using ciprofloxacin alone. It is worth mentioning collateral resistance, the acquisition of which can cause mutations in bacterial cells.

The resulting acquired genes can enhance or even inhibit the action of a previously ineffective antibiotic [21].

Imipenem x amikacin: P.mirabilis

Imipenem, a member of the carbapenem group, is a last-resort antibiotic among routinely used antibiotics. These drugs are highly burdensome to the liver and kidneys, causing more side effects than other classes of chemotherapeutic agents. They also exhibit a strong antimicrobial effect by inhibiting cell wall synthesis and inactivating penicillin-binding proteins, ultimately leading to bacterial cell lysis and death [18]. Carbapenems are an effective therapeutic solution, especially when strains are sensitive to them. Unfortunately, in recent years, microbial resistance to even this group of drugs has been increasing and spreading. Combination therapy with carbapenems and aminoglycosides is used for multidrug-resistant infections in immunocompromised patients who have repeatedly received broad-spectrum antibiotics. This therapy can reduce antibiotic doses and limit the development of resistance. [22].

Amikacin X Nitrofurantoin: E.Coli

In vitro studies confirm the synergistic effects of these two antibiotics. Both are used in the routine treatment and prevention of urinary tract infections. These drugs may potentiate each other's effects. Nitrofurantoin generates reactive oxygen species in bacterial cells, which in turn facilitates the penetration of aminoglycosides. Once inside the bacterial cell, their common targets are the ribosomes and disrupt the bacterial cell's genetic expression [23].

The mechanisms of action of the antibiotics included in this study are presented in Figure 1.

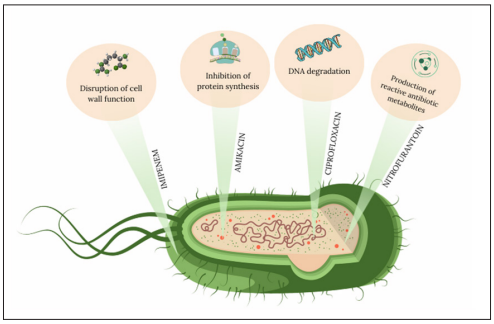

Figure 1: The mechanism of action of the antibiotics [17-20,22]

Materials and Methods

Materials

The study included 200 clinical strains, 50 from each of the following bacterial species: E. coli, P. mirabilis, P. aeruginosa, and E. faecalis. The clinical strains used for the study are part of the collection of the Department of Microbiology, Ludwik Rydygier Medical College in Bydgoszcz, Nicolaus Copernicus University in Toruń. The study included 32 strains: 4 strains of P. aeruginosa, 4 strains of E. faecalis, 16 strains of E. coli and 8 strains of P. mirabilis. Data regarding the results of antibiotic susceptibility testing were downloaded from the Promic IT system (Mori®, Marcin Bogucki). Antibiotic susceptibility was determined as part of microbiological diagnostics performed by staff at the University Hospital No. 1 named after Dr. Antoni Jurasz in Bydgoszcz using an automated method (Phoenix M50, Becton Dickinson).

Methods

The tested strains were stored deep-frozen at -70°C. Before assaying, the strains were subcultured twice onto Columbia Agar with sheep blood and incubated for 24 hours at 37°C. 96-well polystyrene plates were supplemented with 100 µl of MHB (MHB, Mueller Hinton Broth). A series of dilutions of the antibiotics amikacin, nitrofurantoin, ciprofloxacin, and imipenem (Sigma Aldrich) was prepared in sterile tubes by dissolving aliquots of the substances in MHB. A series of 1:2 dilutions was then performed according to the scheme presented in Figure 2 and Table 1. The prepared plates, with the forming biofilm, were incubated for 24 hours at 37°C. Figure 2 shows the general arrangement of the final concentrations obtained in the wells of polystyrene plates used to treat bacterial biofilms and their arrangement. Table 1 shows the scheme of drugs used to treat bacterial biofilms of individual microbial species.

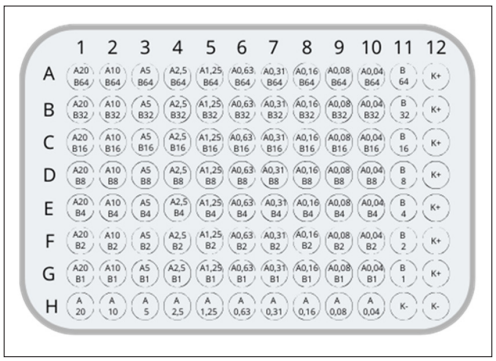

Figure 2: The system of concentrations acting on microorganisms in the study. (K+)-Positive control, (K-)-Negative control, A-drug A concentration in µg/ml, B- drug B concentration in µg/ml. For example A20 -drug A in concentration 20 µg/ml

Table 1: Scheme of drugs that acted on bacterial biofilms of individual microbial species

Microorganism	Antibiotic A	Antibiotic B
Enterococcus faecalis	Ciprofloxacin	Nitrofurantoin
Pseudomonas aeruginosa	Ciprofloxacin	Nitrofurantoin
Escherichia coli (1)	Ciprofloxacin	Amikacin
Escherichia coli (2)	Amikacin	Nitrofurantoin
Proteus mirabilis	Imipenem	Amikacin

Calculation of the Degree of Biofilm Reduction

To obtain the normalized values of the absorbance change of the biofilm mass (AZ) formed in the wells after incubation according to the formula;

$$AZ = \frac{(AK+)-(AB)}{(AK+)x100\%}$$

(AK+) - Absorbance of the positive control
(AB) - Absorbance of the biofilm formed in the plate well

Results

The effect of ciprofloxacin used individually against biofilm of E. faecalis strains (Figure 3)
Reductions in E. faecalis biofilm mass ranged from 12.49% to

47.02%. The lowest reduction was observed at a concentration of 0.04 µg/ml and the highest at a concentration of 1.25 µg/ml.

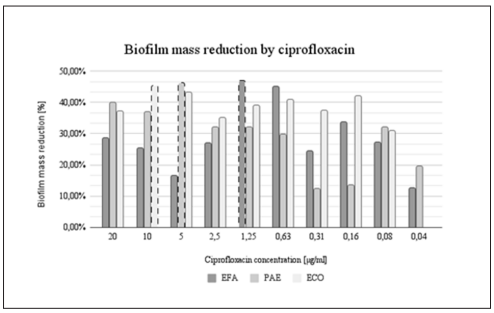

Figure 3: The antibiofilm effect of ciprofloxacin used individually against biofilm of E.faecalis, P. aeruginosa, E.coli

The effect of nitrofurantoin used individually against biofilm of E. faecalis strains (Figure 4)
The biofilm mass reduction ranged from 4.38% to 26.09%. The lowest result was obtained at a concentration of 64 µg/ml, while a 21.79% reduction occurred at a concentration of 32 µg/ml. The highest effect was observed at a concentration of 1 µg/ml.

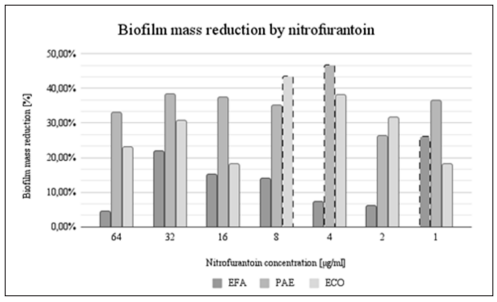

Figure 4: The antibiofilm effect of nitrofurantoin used individually against biofilm of E.faecalis, P. aeruginosa, E.coli

Evaluation of the synergism of ciprofloxacin in combination with nitrofurantoin against biofilm strains of E. faecalis (Figure 5)

The range of reduction was 2.40%–42.13%. The lowest reduction was achieved with the combination of 0.31 µg/ml ciprofloxacin and 16 µg/ml nitrofurantoin. The highest reduction was achieved with the combination of 5 µg/ml ciprofloxacin and 32 µg/ml nitrofurantoin.

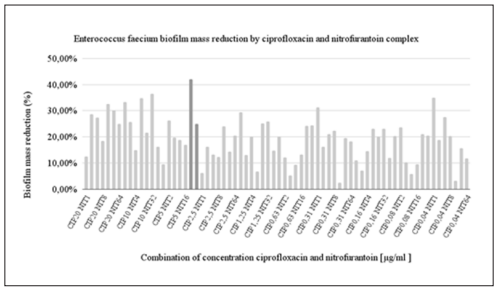

Figure 5: Evaluation of the synergism of ciprofloxacin in combination with nitrofurantoin against biofilm strains of E. faecalis

The effect of ciprofloxacin used individually against biofilm of P. aeruginosa strains (Figure 3)
Biofilm reductions of 12.24%–46.24% were observed. The lowest result was at a concentration of 0.31 µg/ml, and the highest at a concentration of 5 µg/ml. At a concentration of 0.08 µg/ml, the reduction was 32.08%.

The effect of nitrofurantoin used individually against biofilm of P. aeruginosa strains (Figure 4)
The biofilm mass reduction ranged from 26.26% to 46.73%. The lowest value was obtained at a concentration of 2 µg/ml and the highest at a concentration of 4 µg/ml.

Evaluation of the synergism of ciprofloxacin in combination with nitrofurantoin against biofilm strains of P. aeruginosa (Figure 6)
Biofilm reduction ranged from 9.82% to 59.08%. The lowest result was recorded for the combination of 0.31 µg/ml ciprofloxacin and 64 µg/ml nitrofurantoin. The highest result was recorded for the combination of 0.08 µg/ml ciprofloxacin and 64 µg/ml nitrofurantoin.

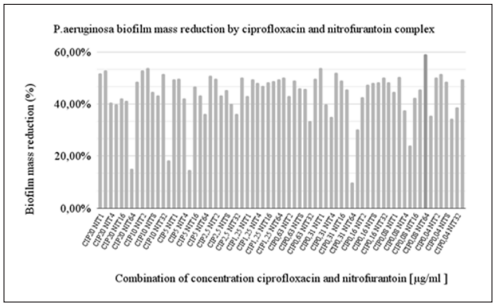

Figure 6: Evaluation of the synergism of ciprofloxacin in combination with nitrofurantoin against biofilm strains of P. aeruginosa

The effect of ciprofloxacin used individually against biofilm of E. coli strains (Figure 3)
The reduction range was 31.02%–45.40%. The lowest value was obtained at a concentration of 0.08 µg/ml, and the highest at a concentration of 10 µg/ml.

The effect of amikacin used individually against biofilm of E. coli strains (Figure 7)
The biofilm reduction ranged from 18.21%–31.39%. The lowest level was observed at a concentration of 8 µg/ml, and the highest at 4 µg/ml.

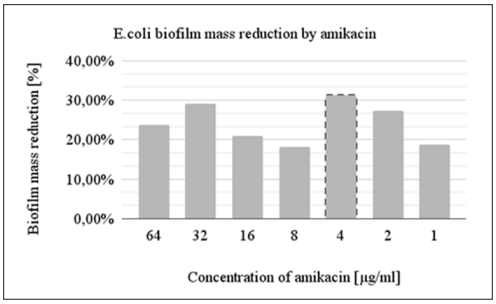

Figure 7: The antibiofilm effect of amikacin used individually against biofilm of E. coli strains

Evaluation of the synergism of ciprofloxacin in combination with amikacin against biofilm strains of E. coli (Figure 8)

The reduction obtained ranged from 20.02%–57.58%. The lowest result was recorded for the combination of 0.08 µg/ml

ciprofloxacin and 64 µg/ml amikacin, and the highest for 5 µg/ml ciprofloxacin and 2 µg/ml amikacin.

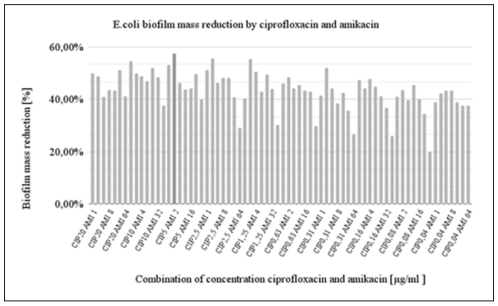

Figure 8: Evaluation of the synergism of ciprofloxacin in combination with amikacin against biofilm strains of E.coli

The effect of nitrofurantoin used individually against biofilm of E. coli strains (Figure 4)
The biofilm mass reduction ranged from 18.07% to 43.43%. The lowest result was achieved at a concentration of 1 µg/ml, and the highest at 8 µg/ml.

Evaluation of the synergism of nitrofurantoin in combination with amikacin against biofilm strains of E. coli (Figure 9)
The range of reduction was 12.35%–63.64%. The lowest result was obtained with the combination of 0.16 µg/ml amikacin and 8 µg/ml nitrofurantoin, and the highest with the combination of 0.08 µg/ml amikacin and 64 µg/ml nitrofurantoin.

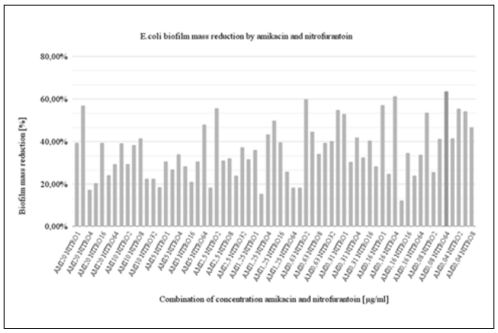

Figure 9: Evaluation of the synergism of nitrofurantoin in combination with amikacin against biofilm strains of E.coli

The effect of amikacin used individually against biofilm strains of P. mirabilis (Figure 10)
The reduction in biofilm mass was 16.79%–49.50%. The lowest result was obtained at a concentration of 1 µg/ml, and the highest at 4 µg/ml.

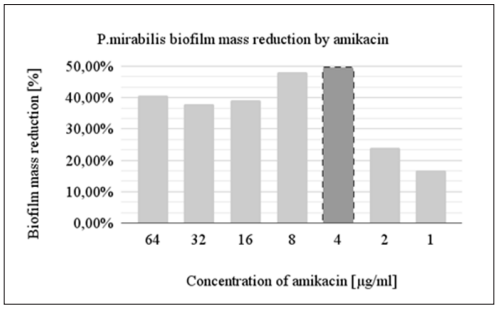

Figure 10: The effect of amikacin used individually against biofilm of P. mirabilis strains

The effect of imipenem used alone against biofilm of P. mirabilis strains (Figure 11)
Reductions ranging from 44.28% to 68.79% were observed. The lowest reduction was observed at a concentration of 0.04 µg/ml, and the highest at a concentration of 5 µg/ml.

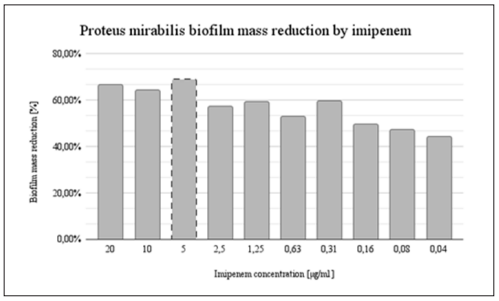

Figure 11: The antibiofilm effect of imipenem used individually against biofilm of P. mirabilis strains

Evaluation of the synergism of imipenem in combination with amikacin against biofilm strains of P. mirabilis (Figure 12)
Reductions in biofilm mass ranging from 26.98% to 82.75% were achieved. The lowest reduction was observed with the combination of 0.08 µg/ml imipenem and 64 µg/ml amikacin. The highest reduction was achieved with 20 µg/ml imipenem and 8 µg/ml amikacin.

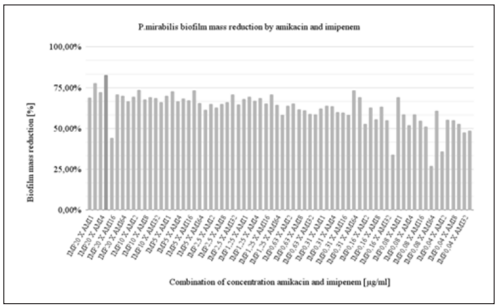

Figure 12: Evaluation of the synergism of imipenem in combination with amikacin against biofilm strains of P. mirabilis

Discussion

This study assessed the activity of ciprofloxacin, nitrofurantoin, amikacin, and imipenem, used alone and in combination, against biofilms of E. faecalis, P. aeruginosa, E. coli, and P. mirabilis isolated from urine. The characteristic structure of biofilms, limited antibiotic diffusion, and metabolic silencing of some cells significantly hinder effective infection eradication and promote their recurrence, which makes research on antibiotic activity in this environment particularly important.

Ciprofloxacin reduced E. faecalis biofilm by 12–47%, with the lowest efficacy observed at a subinhibitory concentration of 0.04 µg/ml. Biofilm susceptibility increased with increasing dose. The limited activity of fluoroquinolones against enterococci results from, among other things, mutations in the gyrA and parC genes and the activity of efflux pumps, as well as the inherent resistance of E [17]. faecalis to many antibiotic groups [23]. These results are consistent with reports that a concentration of 0.38 µg/ml of ciprofloxacin did not significantly affect the amount of biofilm, despite global changes in metabolic gene expression and no changes in esp gene expression [24]. In other studies, the MIC for ciprofloxacin against E. faecalis exceeded 16 µg/ml [25].

Ciprofloxacin demonstrated a 12–46% reduction in P. aeruginosa biofilms, with the highest efficacy at 5 µg/ml. Its activity was

dependent on gyrA and parC mutations, and exposure to concentrations ≥ 4 mg/l resulted in a significant decrease in biofilm viability and changes in gene expression of the toxin-antitoxin and T3SS systems [26,19]. In E. coli biofilms, ciprofloxacin demonstrated a 31–45% reduction, with long-term exposure promoting the rapid selection of resistant mutants, even though sub-MIC may significantly limit biofilm formation [27,28].

Nitrofurantoin showed variable activity against all tested species, which is consistent with its multi-target mechanism of action based on the formation of reactive metabolites [20]. E. faecalis biofilm reduction was 4–26%, with the highest efficacy achieved at low concentrations, and the lack of a dose-effect relationship may be due to modulation of c-di-GMP levels [20]. The efficacy of nitrofurantoin against enterococci isolated from UTI has been confirmed in the literature, however, the presence of NrmA nitroreductase may determine resistance [29,30]. At the same time, it has been shown that both nitrofurantoin and ciprofloxacin can induce or inhibit biofilm formation depending on the strain, even in resistant isolates [21,31].

The combination of ciprofloxacin with nitrofurantoin demonstrated a significant synergistic effect, particularly against P. aeruginosa, where up to 59% biofilm reduction was achieved, with the most favorable effects observed at low ciprofloxacin and high nitrofurantoin concentrations. These results are consistent with the observations of reduced MIC values with combination therapy against UPEC and with the mechanism of increased oxidative stress and DNA damage [32,33].

For E. coli, ciprofloxacin and amikacin monotherapy demonstrated moderate efficacy (31–45% and 18–31%, respectively), while their combination increased biofilm reduction to 58%. The strongest effect was observed with 5 μg/ml ciprofloxacin and 2 μg/ml amikacin. Also, the combination of amikacin with nitrofurantoin showed a strong synergistic effect (>60% reduction), which is confirmed by both in vitro studies and the Galleria mellonella model, although the combination of ciprofloxacin with amikacin alone does not always limit the selection of resistant mutants, and aminoglycoside monotherapy may be insufficient [34,35,2].

The highest therapeutic efficacy was achieved against P. mirabilis biofilms. Imipenem reduced biomass by 44–69%, while in combination with amikacin, efficacy increased to over 80%. The mechanism of synergy results from disruption of cell wall integrity by the carbapenem and facilitated penetration of the aminoglycoside. These data are consistent with the low MBIC values for amikacin against P. mirabilis biofilms and the strong synergy of imipenem and amikacin also against multidrug-resistant strains [35,36].

In summary, the obtained results clearly indicate that antibiotic monotherapy demonstrates only limited effectiveness against mature biofilm forms, whereas combination therapies, based on antibiotics with complementary mechanisms of action, lead to significantly greater biofilm biomass reduction. The bacterial response to the antibiotics used was strongly dependent on both the microbial species (E. faecalis, P. aeruginosa, E. coli, P. mirabilis) and the concentration ratios of the drugs used. Of particular clinical significance is the observed phenomenon in which even very low doses of one antibiotic significantly enhanced the effect of the other, indicating the possibility of effective biofilm eradication while simultaneously limiting the selection of resistant strains and reducing treatment toxicity.

The combination of imipenem and amikacin demonstrated the greatest therapeutic potential against P. mirabilis biofilms-one of the key pathogens of catheter-associated urinary tract infections and complicated infections characterized by the formation of mineralized biofilms. The combination of ciprofloxacin and nitrofurantoin demonstrated equally high efficacy, particularly against P. aeruginosa and E. coli, which are among the most common etiological factors of both uncomplicated and nosocomial urinary tract infections. These results are particularly important in the context of increasing resistance to fluoroquinolones and limited therapeutic options for infections caused by multidrug-resistant strains.

Despite the limitations of using an in vitro model that does not fully reflect conditions in the urinary tract, such as pH variation, urine flow, or the presence of organic components, the data provide strong evidence for further translational research and clinical observations. They indicate that rationally selected combination therapies may be an effective treatment strategy for biofilm-complicated urinary tract infections caused by E. coli, P. aeruginosa, P. mirabilis, and Enterococcus faecalis.

Conclusions

- Antibiotic monotherapy shows limited efficacy against mature biofilms of E. faecalis, P. aeruginosa, E. coli, and P. mirabilis isolated from urinary tract infections.
- The use of combination therapies, based on antibiotics with complementary mechanisms of action, significantly increased the reduction of biofilm biomass compared to single-drug treatment.
- Efficacy was strongly dependent on both the microbial species and the concentration ratio of the antibiotics used, with even low doses of one drug significantly potentiating the effect of the other.
- Combinations of imipenem with amikacin against P. mirabilis and ciprofloxacin with nitrofurantoin against P. aeruginosa and E. coli demonstrated the greatest therapeutic potential.
- The obtained results support the need for further research into combination therapies as an effective treatment strategy for urinary tract infections complicated by the presence of biofilm.

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