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ALINE NATÁLIA DE SANTI

Influência do pH na atividade do ciprofloxacino contra *Escherichia coli* isolada em amostras de urina

Maringá
2020

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Dissertação apresentada ao Programa de Pós-Graduação em Biociências e Fisiopatologia do Departamento de Análises Clínicas e Biomedicina, Centro de Ciências da Saúde da Universidade Estadual de Maringá, como requisito parcial para obtenção do título de Mestre em Biociências e Fisiopatologia
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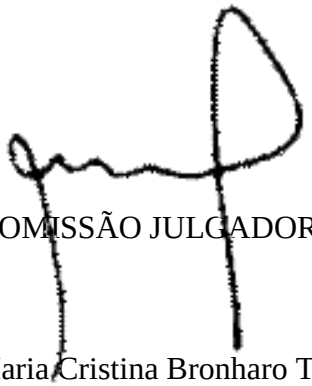

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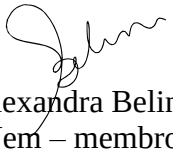

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“A ciência é muito mais que um corpo de conhecimentos. É uma maneira de pensar”.

(Carl Sagan)

INFLUÊNCIA DO pH NA ATIVIDADE DA CIPROFLOXACINA CONTRA *Escherichia Coli* ISOLADA EM AMOSTRAS DE URINA

RESUMO

As infecções do trato urinário (ITU) estão entre as infecções bacterianas mais comuns em humanos, afetando milhões de pessoas na comunidade anualmente. A maioria das ITUs écausada por bactérias Gram-negativas, especialmente espécies da família *Enterobacteriaceae*, com *Escherichia coli* comumente isolada em aproximadamente 70 a 90% dos casos relacionadosa infecções não complicadas do trato urinário. O tratamento das ITUs tem se tornado um desafio tanto pela recorrência dessas infecções quanto pelo aumento gradual das taxas de resistência bacteriana aos antimicrobianos, incluindo o ciprofloxacino. Para que haja eficácia terapêutica nas ITUs, o pH urinário deve ser considerado um fator relevante. O objetivo deste trabalho foi verificar a variação da concentração inibitória mínima do ciprofloxacino contra pH 6,0, 7,0 e 8,0 para a otimização do pH do ambiente, proporcionando um melhor alcance do resultado terapêutico à sua relação com o pH urinário. No período de junho de 2018 a junho de 2019, foram selecionados 106 isolados consecutivos de *E. coli* recuperados de pacientes internados ou atendidos em pronto-socorro ou ambulatório do Hospital Universitário Regional de Maringá. Quando testado em pH 8,0, observou-se que 66% dos isolados foram sensíveis ao ciprofloxacino, com crescimento de até MIC 0,25 mg / L. Um total de 18 isolados resistentes ao ciprofloxacino em pH 6,0 tornaram-se sensíveis quando testados em pH 8,0. Pelos resultados deste estudo, é possível observar que a urina e, em particular, seu pH influenciam a atividade do ciprofloxacino, enfatizando o pH ácido comumente encontrado no trato urinário e que pode diminuir sua atividade.

Palavras-chave: *Enterobacteriaceae*; antimicrobiano; ciprofloxacino; infecção do trato urinário

INFLUENCE OF pH ON CIPROFLOXACIN ACTIVITY AGAINST *Escherichia Coli* ISOLATED IN URINE SAMPLES

ABSTRACT

Urinary tract infections (UTI) are among the most common bacterial infections in humans, affecting millions of people in the community annually. Most UTIs are caused by Gram-negative bacteria, especially species of the *Enterobacteriaceae* family, with *Escherichia coli* commonly isolated in approximately 70 to 90% of cases related to uncomplicated urinary tract infections. Treatment of UTIs has become a challenge both due to the recurrence of these infections and the gradual increase in rates of bacterial resistance to antimicrobials, including ciprofloxacin. For there to be therapeutic efficacy in UTIs, urinary pH must be considered as a relevant factor. The objective of this work was to verify the variation of the minimum inhibitory concentration of ciprofloxacin against pH 6.0, 7.0 and 8.0 for the optimization of the pH of the environment, providing a better reach of the therapeutic result to its relation with urinary pH. In the period from June 2018 to June 2019, 106 consecutive isolates of *E. coli* recovered from patients who were hospitalized or who were seen in emergency care or Regional University Hospital of Maringá outpatient clinics were selected. When tested at pH 8.0, it was noted that 66% of the isolates were sensitive to ciprofloxacin, with growth up to MIC 0.25 mg / L. A total of 18 isolates that were resistant to ciprofloxacin at pH 6.0 became sensitive when tested at pH 8.0. From the results of this study, it is possible to observe that urine and, in particular, its pH influence the activity of ciprofloxacin, emphasizing the acid pH that is commonly found in the urinary tract and which can decrease its activity.

Keywords: *Enterobacteriaceae*; antimicrobial; ciprofloxacin; urinary tract infections

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SUMÁRIO

CAPÍTULO I

1.1 INTRODUÇÃO

As infecções do trato urinário (ITUs) representam um problema global, ocorrendo na comunidade e no âmbito hospitalar[1]. As ITUs estão entre as infecções bacterianas em humanos mais comuns, afetando anualmente milhões de pessoas na comunidade e também nas unidades prestadoras de assistência à saúde[2] e associadas substancialmente a mortalidade e morbidade[3]. Dentre as infecções nosocomiais, cerca de 30% a 50% é representado pelas ITUs[4].

Consideradas entre as mais frequentes causas de internações por infecções em pacientes idosos, as ITUs também são motivo mais comum de prescrição de antimicrobianos na atenção básica[2,5]. A alta incidência está relacionada à vários fatores de risco, como por exemplo, a utilização de cateter, manipulação cirúrgica, diabetes, uso de drogas imunossupressoras e internações prévias[4,6]. Em pacientes hospitalizados, as ITUs possibilitam o desenvolvimento de sepse ocasionando um aumento nas taxas de mortalidade[7,8].

A ITU não complicada é uma infecção ocasionada na bexiga e estruturas relacionadas. Acometem geralmente em pacientes sem anormalidades estruturais e sem comorbidades, como diabetes, imunocomprometidos ou gestantes. As mulheres são mais sucetíveis a desenvolverem infecção urinária 4 vezes mais do que em homens, por terem uretra mais curta[9,10]. Os sintomas podem incluir disúria, frequência urinária ou urgência urinária[11].

A ITU é considerada incomum em homens circuncidados, sendo assim, qualquer ITU masculina é classificada como complicada. Muitos casos de ITU não complicada se resolvem naturalmente, sem necessidade de tratamento, mas muitos pacientes procuram tratamento para os sintomas. O tratamento previne a disseminação para os rins ou desenvolver doenças do trato superior, como a pielonefrite[11, 12].

A urina é um meio ideal para o crescimento de bactérias[11]. Grande parte das infecções do trato urinário (ITUs) são causadas por bactérias Gram-negativas, especialmente espécies da família *Enterobacteriaceae*, sendo a *Escherichia coli* (*E. coli*) comumente isolada[9] em aproximadamente 70 a 90% dos casos relacionados à ITU não complicada[13-15]. Além de *E. coli*, espécies de *Klebsiella*, *Proteus* e *Enterobacter* são frequentemente recuperadas de pacientes comITUs [16].

Para ITUs complicadas, incluindo pielonefrite, a terapia com ciprofloxacino é uma das principais opções contra bactérias Gram-negativas multidroga resistentes devido às elevadas concentrações que pode atingir no trato urinário [17]. O ciprofloxacino está incluído na lista de

medicamentos essenciais da Organização Mundial de Saúde (OMS). Esse antimicrobiano pertence à classe das fluoroquinolonas e, desde 1987, é frequentemente usado para tratar diversasinfecções bacterianas[18].

As fluoroquinolonas apresentam relevância no tratamento de infecções mais graves e septicemia, deste modo, o ciprofloxacino é considerado como um tratamento alternativo, não como um antibiótico de primeira linha, no tratamento de cistite não complicada[19]. O ciprofloxacino é utilizado como terapia empírica de segunda linha em casos de pielonefrite levee moderada ou tratamento de ITU complicada, e como tratamento empírico de terceira linha paracistite não complicada[20].

As propriedades físicas e químicas das quinolonas tem efeitos relevantes para sua farmacocinética e farmacodinâmica[21,22]. O ciprofloxacino tem a capacidade de inibir a proliferação de células eucarióticas por meio de danos ao DNA mitocondrial[23]. Na figura 1 é possível observar a estrutura molecular detalhada do ciprofloxacino e com a seguinte fórmula molecular $C_{17}H_{18}FN_3O_3$.

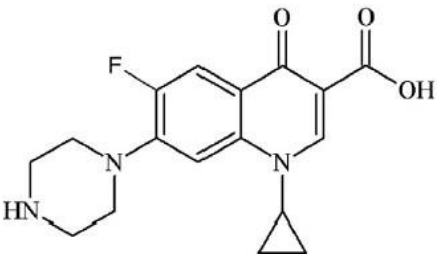

Figura 1: Estrutura molecular do ciprofloxacino. Fonte: Jalil et al., 2015[24].

As fluoroquinolonas com um grupo piperazina em C-7, como o ciprofloxacino, apresentam uma atividade diminuída com a redução do pH. Em pHs altos todas as fluoroquinolonas tendem a ter carga negativa, porém, em pHs mais baixos, as fluoroquinolonas contendo o grupo piperazina são carregadas positivamente e esse fator pode diminuir sua penetração nas bactérias e consequentemente diminuir sua atividade[25]. Da mesma forma que o pH urinário interefere na atividade das fluoroquinolonas, os cátions urinários divalentes como o cálcio e o magnésio também podem diminuir sua atividade[26].

A molécula ciprofloxacino com um pH abaixo de 6,1 é considerada íons positivo (cátions), já com pH de 6,1 a 8,7 são moléculas de íons dipolo, quando o pH é maior que 8,7, a molécula de ciprofloxacino é um íon negativo (ânion)[27]. A molécula deste antibiótico é degradada em soluções do qual o pH seja inferior a 7, devido a alta solubilidade nestas condições(Figura 2).

A prescrição empírica de antibióticos para este tipo de infecção é frequente, por razão da demora na liberação de resultados de cultura e de antibiograma, sendo esta procedência um fator de risco para o desenvolvimento de resistência bacteriana aos antibióticos[28,29]. O tratamento de pacientes com ITUs tem se tornado um desafio devido à alta prevalência e recorrência dessas infecções e ao aumento gradativo das taxas de resistência bacteriana aos antimicrobianos, incluindo o ciprofloxacino[15,16,30-32].

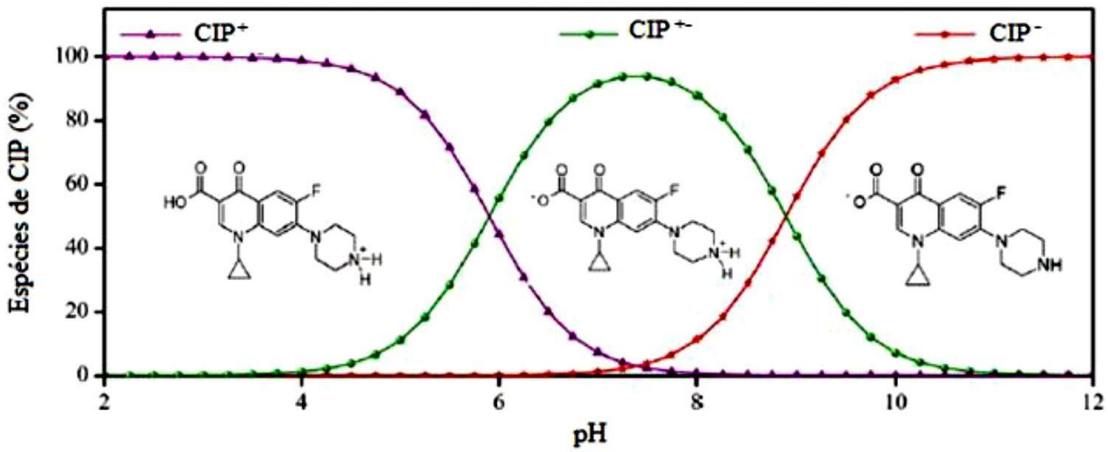

Figura 2: Distribuição das espécies de CIP em função do pH. Fonte: Jalil et al., 2015.

Para que exista eficácia terapêutica, o pH urinário deve ser classificado como um fator de relevância [33]. O pH urinário não afeta apenas o crescimento do uropatógeno, mas também a atividade antimicrobiana de vários antibióticos[34,35].

Dependendo do antimicrobiano, se o pH urinário não for monitorado, a atividade antimicrobiana pode apresentar o mesmo efeito que concentrações urinárias subinibitórias, predispondo ao desenvolvimento de resistência[33]. O pH pode causar um significativo impacto sobre as bactérias, favorecendo várias respostas adaptativas e protetoras alterando, por exemplo, os padrões de expressão gênica de resistência e a fisiologia celular, que influencia a sensibilidade aos antimicrobianos[34].

1.2 JUSTIFICATIVA

Diante do aumento de bactérias Gram-negativas multirresistentes causando ITUs, as dificuldades cada vez maiores no tratamento dessas infecções e a falta de dados na literatura que auxiliem os clínicos a determinarem o melhor recurso terapêutico para as ITUs, observamos a

necessidade de estudos baseados no resgate de antimicrobianos antigos e que verifiquem o impacto do pH na atividade dos mesmos.

Tem sido relatado que as condições fisiológicas, como o pH, apresentam influência na sensibilidade dos microrganismos (begic), porém, poucos estudos atuais relacionam a influência do pH na atividade do ciprofloxacino contra bactérias Gram-negativas isoladas da urina.

Baseados nesse contexto, a partir dos dados que serão coletados neste estudo e considerando a experiência do nosso grupo de pesquisa no assunto, acreditamos que nossos resultados serão importantes instrumentos para auxiliar no direcionamento do tratamento econtrole da resistência bacteriana em infecções urinárias.

1.3 OBJETIVOS

1.3.1 Objetivo Geral

Analisar a influência do pH sobre a atividade do ciprofloxacino contra bactérias gram-negativas isoladas da urina de pacientes atendidos no Hospital Universitário Regional de Maringá.

1.3.2 Objetivos específicos

- Recuperar durante o período de um ano todos os isolados de *E. coli* da urina de pacientes de um hospital ensino;
- Determinar a concentração inibitória mínima do ciprofloxacino em diferentes pHs;
- Investigar o impacto do pH sobre a atividade do ciprofloxacino;
- Buscar dados do pH urinário a partir do exame de urina tipo 1 dos pacientes.

REFERÊNCIAS

1. Wurpel DJ, Totsika M, Allsopp LP, Webb RI, Moriel DG, Schembri MA. Comparative Proteomics of Uropathogenic *Escherichia Coli* During Growth in Human Urine Identify UCA-Like (UCL) Fimbriae as an Adherence Factor Involved in Biofilm Formation and Binding to Uroepithelial Cells. J Proteomics 2016; 131:177-89.
2. Zare F, Rostami FM, Shahsafi M. Prevalence and pattern of antibiotic resistance of gram-negative bacteria isolated from urinary tract infections in patients referring to Neka Laboratories-Iran. Int J BioMed Public Health. 2018; 1(1):30-36.
3. Foxman, B. Urinary Tract Infection Syndromes: Occurrence, Recurrence, Bacteriology, Risk Factors, and Disease Burden. Infect Dis Clin North Am 2014; 28:1-13.
4. Redder JD, Leth RA, Moller JK. Analysing risk factors for urinary tract infection based on automated monitoring of hospital-acquired infection. J Hosp Infect. 2016; 92:397-400.
5. McLellan LK, Hunstad DA. Urinary Tract Infection: Pathogenesis and Outlook. Trends Mol Med. 2016; 22(11):946-957.
6. Saltoglu N, Karali R, Yemisen M, Ozaras R, Balkan II, Mete B. Comparison of community-onset healthcare-associated and hospital-acquired urinary infections caused by extended-spectrum beta-lactamase-producing *Escherichia coli* and antimicrobial activities. Int. J. Clin. Pract. 2015; 69: 766-70.
7. Melzer M, Welch C. Outcomes in UK patients with hospital-acquired bacteraemia and the risk of catheter-associated urinary tract infections. Postgrad Med J. 2013 Mar.
8. Kiffer CRV, Cuba GT, Fortaleza CMCB, Padoveze MC, Pignatari ACC, on behalf of the IRAS Brasil group. Exploratory model for estimating occupation-day costs associated to Hospital Related Infections based on data from national prevalence project: IRAS Brasil Project. Am J Infect Control. 2015 Jan; 4 (1):30-33.
9. Yamaji R, Friedman CR, Rubin J, Suh J, Thys E, McDermott P, Hung-Fan M, Riley LW. A Population-Based Surveillance Study of Shared Genotypes of *Escherichia coli* Isolates from Retail Meat and Suspected Cases of Urinary Tract Infections. mSphere. 2018 Aug 15; 3(4).
10. Sakamoto S, Miyazawa K, Yasui T, Iguchi T, Fujita M, Nishimatsu H, Masaki T,Hasegawa T, Hibi H, Arakawa T, Ando R, Kato Y, Ishito N, Yamaguchi S, Takazawa R, Tsujihata M, Taguchi M, Akakura K, Hata A, Ichikawa T. Chronological changes in epidemiological characteristics of lower urinary tract urolithiasis in Japan. Int J Urol. 2019 Jan; 26(1): 96-101.

11. Richards KA, Cesario S, Best SL, Deeren SM, Bushman W, Safdar N. Reflex urine culture testing in an ambulatory urology clinic: Implications for antibiotic stewardship in urology. Int J Urol. 2019 Jan; 26(1):69-74.
12. Tang M, Quanstrom K, Jin C, Suskind AM. Recurrent Urinary Tract Infections are Associated With Frailty in Older Adults. Urology. 2019 Jan; 123:24-27.
13. Cantón R, Coque, TM. The CTX-M Beta-Lactamase Pandemic. Curr Opin Microbiol 2006 Out; 9(5): 466-75.
14. Gurevich E, Tchernin, D, Schreyber R, Muller R, Leibovitz E. 2016. Follow-up after infants younger than 2 months of age with urinary tract infection in southern Israel: epidemiologic, microbiologic and disease recurrence characteristics. Braz J Infect Dis. 2016; 20:19-25.
15. Campos ACC, Andrade NL, Ferdous M, Santos CC, Correal JCD. Comprehensive Molecular Characterization of Escherichia coli Isolates from Urine Samples of Hospitalized Patients in Rio de Janeiro, Brazil. Front Microbiol. 2018; 16(9):243.
16. Wagenlehner FME, Naber KG. Treatment of bacterial urinary tract infections: presence and future. Eur Urol. 2006; 49(2):235-44.
17. Aldred KJ, Kerns RJ, Osheroff N. Mechanism of quinolone action and resistance. Biochemistry. 2014; 53:1565-1574.
18. Rehman A, Patrick WM, Lamont IL. Mechanisms of ciprofloxacin resistance in *Pseudomonas aeruginosa*: new approaches to an old problem. J Med Microbiol. 2019; 68:1–10.
19. Bartoletti R, Cai T, Wagenlehner FM, Naber K, Bjerklund JTE. Treatment of urinarytract infections and antibiotic stewardship. Eur Urol Suppl. 2016.
20. Cheung A, Karmali G, Noble S, Song H. Antimicrobial stewardship initiative in treatment of urinary tract infections at a rehabilitation and complex continuing care hospital. Can J Hosp Pharm. 2017.
21. Nikaido H, Thanassi D G. Penetration of lipophilic agents with multiple protonation sites into bacterial cells: tetracyclines and fluoroquinolones as example. Antimicrob Agents Chemother. 1993; 37:1393-1399.
22. Sorgel F, Kinzig M. Pharmacokinetics of gyrase inibidores, parte 1: química básica e disposição gastrointestinal. Am J Med. 1993; 94 (Suplemento): 44S-55S.
23. Kloskowski T, Gurtowska N, Olkowska J, Nowak JM, Adamowicz J, Tworkiewicz J, Dębski R, Grzanka A, Drewa T. Ciprofloxacin is a potential topoisomerase II inhibitor for the treatment of NSCLC. Int J Oncol. 2012; 4(6):1943-1949.

24. Jalil, MER, Baschini, M, Sapag, K. Influence of pH and antibiotic solubility on the removal of ciprofloxacin from aqueous media using montmorillonite. Applied Clay Science. 2015; 114:69-76.
25. Lewin CS, Smith J T. 4-Quinolones and multivalent ions. J Antimicrob Chemother. 1990; 26: 149.
26. Mann CK, Yoe JH. Determinação espectrofotométrica de magnésio com sódio 1-azo-2-hidroxi-3- (2,4-dimetilcarboxanilido) -naftaleno-1- (2-hidroxibenzeno-5-sulfonato). Anal Chem. 1956; 28:202–205.
27. Jiang W Changa P, Wanga Y, Tsai Y, Jeana J, Li Z, Krukowski K. 2013. Removal of ciprofloxacin from water by birnessite. J. Hazard. Mater. 2013; 250:362-369.
28. Koch CR, Zimmermann BD, D'agostin J, Ribeiro JC, Machado V, Schnor OH. Resistência antimicrobiana dos uropatógenos em pacientes ambulatoriais, 2000-2004. Rev. Soc. Bras. Med. Trop. 2008; 41(3): 277-81.
29. Adamus-Białek W, Baraniak A, Wawszczak M, Głuszek S, Gad B, Wróbel K, Bator P, Majchrzak M, Parniewski P. O histórico genético da resistência a antibióticos entre cepas clínicas de *Escherichia coli* uropatogênica. Mol Biol Rep. 2018 Out; 45 (5):1055-1065.
30. Pfeifer Y, Cullik A, Witte W. Resistance to cephalosporins and carbapenems in Gram-negative bacterial pathogens. Int J Med Microbiol. 2010; 300(6):371-9.
31. Cuba GT, Pignatari ACC, Patekoski KS, Luchesi LJ, Kiffer CRV. Pharmacodynamic profiling of commonly prescribed antimicrobial drugs against *Escherichia coli* isolates from urinary tract. Braz J Infect Dis. 2014; 8(5):512–517.
32. Garoff L, Huseby DL, Praski AL, Hughes D. Effect of aminoacyl-tRNA synthetase mutations on susceptibility to ciprofloxacin in *Escherichia coli*. J Antimicrob Chemother. 2019; 73 (12): 3285-3292.
33. Cunha BA. An infectious disease and pharmacokinetic perspective on oral antibiotic treatment of uncomplicated urinary tract infections due to multidrug-resistant Gram-negative uropathogens: the importance of urinary antibiotic concentrations and urinary pH. Eur J Clin Microbiol Infect Dis. 2016; 35:521–526.
34. Poole K. Bacterial stress responses as determinants of antimicrobial resistance. J Antimicrob Chemother. 2012; 67(9):2069-2089.
35. Yang L, Whag K, Li H, Denstedt JD, Cadieux PA. The influence of urinary pH on antibiotic efficacy against bacterial uropathogens. Urology. 2014; 84:713. Wl-713.e7.

36. Begic S, Worobec EA. Regulation of Serratia marcescens ompF and ompC porin genes in response to osmotic stress, salicylate, temperature and pH. Microbiology. 2006; 152:485–491.

CAPÍTULO II

MANUSCRITO 1: ”INFLUENCE OF pH ON CIPROFLOXACIN ACTIVITY AGAINST *Escherichia Coli* ISOLATED IN URINE SAMPLES”

Title: Influence of pH on ciprofloxacin activity against *Escherichia coli* isolated in urine samples

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Running title: Influence of pH on ciprofloxacin against *E. Coli*

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Keywords: *Enterobacteriaceae*; antimicrobial; ciprofloxacin; Urinary tract infections

Abstract:

Urinary tract infections (UTI) are among the most common bacterial infections in humans, affecting millions of people in the community annually. Most UTIs are caused by Gram-negative bacteria, especially species of the *Enterobacteriaceae* family, with *Escherichia coli* commonly isolated in approximately 70 to 90% of cases related to uncomplicated urinary tract infections. Treatment of UTIs has become a challenge both due to the recurrence of these infections and the gradual increase in rates of bacterial resistance to antimicrobials, including ciprofloxacin. For there to be therapeutic efficacy in UTIs, urinary pH must be considered as a relevant factor. The objective of this work was to verify the variation of the minimum inhibitory concentration of ciprofloxacin against pH 6.0, 7.0 and 8.0 for the optimization of the pH of the environment, providing a better reach of the therapeutic result to its relation with urinary pH. In the period from June 2018 to June 2019, 106 consecutive isolates of *E. coli* recovered from patients who were hospitalized or who were seen in emergency care or Regional University Hospital of Maringá outpatient clinics were selected. When tested at pH 8.0, it was noted that 66% of the isolates were sensitive to ciprofloxacin, with growth up to MIC 0.25 mg / L. A total of 18 isolates that were resistant to ciprofloxacin at pH 6.0 became sensitive when tested at pH 8.0. From the results of this study, it is possible to observe that urine and, in particular, its pH influence the activity of ciprofloxacin, emphasizing the acid pH that is commonly found in the urinary tract and which can decrease its activity.

Introduction

Urinary tract infections (UTIs) are among the most common bacterial infections in humans, affecting millions of people in the community annually. Considered among the most frequent causes of hospitalizations for infections in elderly patients, UTIs are also the most common reason for prescribing antimicrobials in primary care[1,2].

Most UTIs are caused by Gram-negative bacteria, especially species of the *Enterobacteriaceae* family, with *Escherichia coli* (*E. coli*) commonly isolated in approximately 70 to 90% of cases related to uncomplicated UTI[3,4]. In addition to *E. coli*, species of *Klebsiella*, *Proteus* and *Enterobacter* are often recovered from patients with UTIs[5]. Treatment of UTIs has become a challenge both due to the recurrence of these infections and the gradual increase in rates of bacterial resistance to antimicrobials, including ciprofloxacin[4-8].

Ciprofloxacin is included in the World Health Organization (WHO) list of essential drugs. This antimicrobial belongs to the fluoroquinolone class and, since 1987, has been used frequently to treat various bacterial infections[9]. For complicated UTIs, including pyelonephritis, ciprofloxacin therapy is one of the main options against multidrug-resistant Gram-negative bacteria (MDR-GNB) because they have high concentrations in the urinary tract[10]. Quinolones are structurally similar, but they vary considerably in polarity. The manipulations of the urinary pH change the percentage of this antibiotic with dipolar ion, causing its activity to increase or decrease[11].

For there to be therapeutic efficacy in UTIs, urinary pH must be considered as a relevant factor. In acidic urine, the pH can vary from 4.5 to 6.0 and alkaline from 6.5 to 8.0[12], thus the pH of the fluid where the bacteria are present during the infectious process can vary, assuming an acidic or basic characteristic[5,13]. Urinary pH affects not only the growth of the uropathogen, but also the activity of the antimicrobial agents used in the treatment of UTIs[14,15]. Depending on the antimicrobial, if urinary pH is not monitored, its activity may decrease, predisposing to the development of resistance[12].

In view of the increase in MDR-GNB causing UTIs, the increasing difficulties in the treatment of these infections and the lack of data in the literature to assist in the best therapeutic resource for UTIs, we observed the need for studies to verify the impact of pH on the activity of antimicrobialsIt is known that physiological conditions, such as pH, have an influence on the sensitivity of microorganisms[16], however, few current studies relate the influence of pH on ciprofloxacin activity against *E. coli* isolated from urine.

Thus, the objective of this work was to verify the variation of the CIM (Minimum Inhibitory Concentration) of ciprofloxacin against pH 6.0, 7.0 and 8.0 for the optimization of the pH of the environment, providing a better reach of the therapeutic result to its relation with urinary pH.

Materials and Methods

Bacterial sample

The study was conducted at the Regional University Hospital of Maringá located in the northwest of Paraná state, Brazil. In the period from June 2018 to June 2019, 106 consecutive isolates of *E. coli* recovered from patients who were hospitalized or who were seen in emergency care or Regional University Hospital of Maringá outpatient clinics were selected. They were later stored in the Medical Microbiology Laboratory (MML) of the State University of Maringá, Paraná, Brazil. Only one isolate per patient was included in the study, always being the first isolate. Bacterial isolates were identified and with a sensitivity profile to antibacterial agents using the automated system BD Phoenix™ (BD Diagnostic Systems, Sparks, MD)

Antimicrobial agent

Ciprofloxacin was purchased from LabCompany (Londrina, Paraná, Brazil). The antimicrobial was diluted in water to obtain the solution at a concentration of 16 mg/L (first concentration), always prepared on the day of the experiment.

Determination of the minimum inhibitory concentration

The determination of the minimum inhibitory concentration (MIC) for ciprofloxacin was performed using the dilution agar method as recommended by the Clinical and Laboratory Standards Institute (CLSI)[17] using pH 7.0 and Mueller-Hinton agar (Difco Laboratories, Detroit, MI, EUA). The inoculated plates were incubated in room air at 35 ± 2 ° C for 20 h. The ciprofloxacin sensitivity test was performed for each isolate at pH 6.0; 7.0 and 8.0. The pH adjustment of the culture medium was performed with solutions of sodium hydroxide (NaOH) and hydrochloric acid (HCl) after autoclaving. The MIC of the antimicrobial agent was defined as the lowest concentration that completely inhibits the growth of the organism in duplicate tests. The standard strains *Pseudomonas aeruginosa* ATCC®27853 and *Escherichia coli* ATCC® 25922 were used as controls. Ciprofloxacin concentrations ranged from 0.03 to 16 mg/L. The sensitivity tests were performed in duplicate and the interpretation of MIC results based on the breakpoints for *E. coli* established by CLSI [17].

Secondary data search

The urinary pH data were collected from the patients' urinalysis[18]. The pHs found in the partial urine test ranged from 5.5 to 8.0. These data were correlated with the values of the MICs obtained at the different pHs tested and with the MICs determined by the automated method Phoenix®-BD (Becton Dickinson & Co., Sparks, MD, USA). Of the 106 isolates, the pH data of 90 urine samples were recovered, due to the fact that some samples were not prescribed for urine testing, only culture.

Results

***In vitro* susceptibility**

Table 1 shows the antimicrobial susceptibility profile of 106 E. coli urine isolates to ciprofloxacin at different pH values using the dilution agar methodology. At pH 7.0, the MIC varied between 0.03 to 16 mg/L, ciprofloxacin was highly effective in 55% of the isolates, which were sensitive to the tested antimicrobial. In the automated methodology (Phoenix®-BD), cultures with media at pH 7.0, 73% of the samples were sensitive to ciprofloxacin (Table 2). In

123 contrast, when tested at pH 8.0, it was noted that 66% of the isolates were sensitive to
124 ciprofloxacin, with growth up to MIC 0.25 mg / L. A total of 18 isolates that were resistant to
125 ciprofloxacin at pH 6.0 became sensitive when tested at pH 8.0 (Table 1).

126 **Effect of pH on MIC**

127 **Dilution agar method**

128 The in vitro activity of ciprofloxacin in relation to the alkalinization of the growth culture
129 medium (Figure 1) when tested at pH 6.0, an increase in the number of resistant isolates was
130 observed, reaching 48%, with the variation of MIC 0, 06 to 16 mg / L

131 **Phoenix®-BD automated method**

132 It is possible to observe (Figure 2), that the urinary pH 8.0 isolates all samples of
133 sensitivity to ciprofloxacin, while in urinary pH 6.0, 83% of the isolates showed resistance in
134 relation to ciprofloxacin.

135 **Discussion**

136 Fluoroquinolones with a piperazine group, such as ciprofloxacin, show a progressive
137 decrease in activity when the pH is reduced[19]. The ciprofloxacin molecule with a pH below 6.1 is
138 positive ions (cations), with a pH between 6.1 and 8.7 are dipole ion molecules, when the pH is
139 greater than 8.7, the ciprofloxacin molecule is a negative ion (anion)[20]. From this, it is possible to
140 perceive that in molecules with positive charge has high solubility, the ciprofloxacin molecule
141 will be degraded under pH conditions of the solution below 7[21] (Figure 3).

142 The authors Smith and Ratcliffe suggest that at high pHs, fluoroquinolones tend to be
143 negatively charged and drugs containing negatively charged piperazine, including ciprofloxacin,
144 would have a greater effect. However, at lower pHs such as fluoroquinolones containing
145 piperazine, they are positively charged and this factor can decrease their penetration into bacteria
146 and consequently decrease their activity. In the same way that urinary pH interferes with the
147 activity of fluoroquinolones, divalent urinary cations such as calcium and magnesium can also

148 decrease their activity[22]. Similar to the findings of the present study, where ciprofloxacin was
149 more effective in 70 of the 109 samples, when tested in alkaline pH (Figure 1).

150 In contrast, with lower pHs, fluoroquinolones containing piperazine have a positive
151 charge, and this factor tends to decrease their penetration into bacteria and, consequently, also
152 decreases their activity[19], it is possible to observe (Figure 2) in which when the antimicrobial
153 tested at pH 6.0, there was an increase in MIC, of which 52 isolates from the total of tested
154 samples, presented resistance with MICs ranging from 1 to 16 mg/L. This same effect can be
155 observed in the urine 1 partial pH of the samples, where in more acidic pHs there was an
156 increase in MICs.

157 In a study carried out on the effects of pH on quinolones it was demonstrated that the
158 antibacterial activity of ciprofloxacin against *E. coli* NIHJ JC-2 and *Pseudomonas aeruginosa*
159 ATCC 27853 was affected by urine mainly by its pH. The lower pH decreased ciprofloxacin
160 activity[19]. Some findings indicate that pH factors and the concentration of divalent cations can
161 affect the activities of the drug[19,23].

162 In another study reported[24] that the effect of pH on fluoroquinolone activity against *E.*
163 *coli* depends on the nature of the compounds present in quinolone. Reinforcing positively the
164 results of the present study, since the pHs considerably interfered in the ciprofloxacin activity,
165 mainly in alkaline pH, where it presented greater efficacy.

166 When comparing the result of urinary pH and the result of MICs performed on the
167 Phoenix®-BD automated apparatus, acidic urinary pHs increased MICs compared to alkalis.
168 Considering that the sensitivity tests to antimicrobials used in laboratory routines are carried out
169 at neutral pH, as standardized, we can consider that although some isolates present themselves as
170 sensitive in vitro they may not show the same behavior in vivo mainly for antimicrobials such as
171 quinolones that suffer important effect of pH on its activity.

172 The present study tested and demonstrated an important influence of pH on ciprofloxacin
173 activity, which warns that this fact should be considered at the time of therapy. A simple way is

to know the pH easily measured in the laboratory routine and that would be of great value, for the choice of the antimicrobial, as well as the use of substances can be administered concomitantly with the antimicrobials favoring the therapeutic success of the UTIs.

Conclusion

From the results of this study, it is possible to observe that urine and, in particular, its pH influence the activity of ciprofloxacin, emphasizing the acid pH that is commonly found in the urinary tract and which can decrease its activity, warning that this fact should be considered at the time of therapy. A simple way is to know the pH easily measured in the laboratory routine and that would be of great value, for the choice of the antimicrobial, as well as the use of substances can be administered concomitantly with the antimicrobials favoring the therapeutic success of the UTIs.

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192 **Table 1**. *In vitro* susceptibility by the agar method diluted to pH 6.0, 7.0 and 8.0 for ciprofloxacin in 106 clinical isolates of *E. coli* from urinary
193 tract infections.

													% of isolates		
			Number of isolates with the following MICs (mg/L):										CLSI		
Microorganism	Number of isolates	pH of the test	0,03	0,06	0,125	0,25	0,5	1	2	4	8	16	S	I	R
E. coli	106	6,0	-	1	20	24	9	4	3	9	1	35	42	8	49
		7,0	5	10	33	10	6	2	6	2	5	27	55	6	40
		8,0	31	23	11	5	2	1	1	2	19	11	66	2	32

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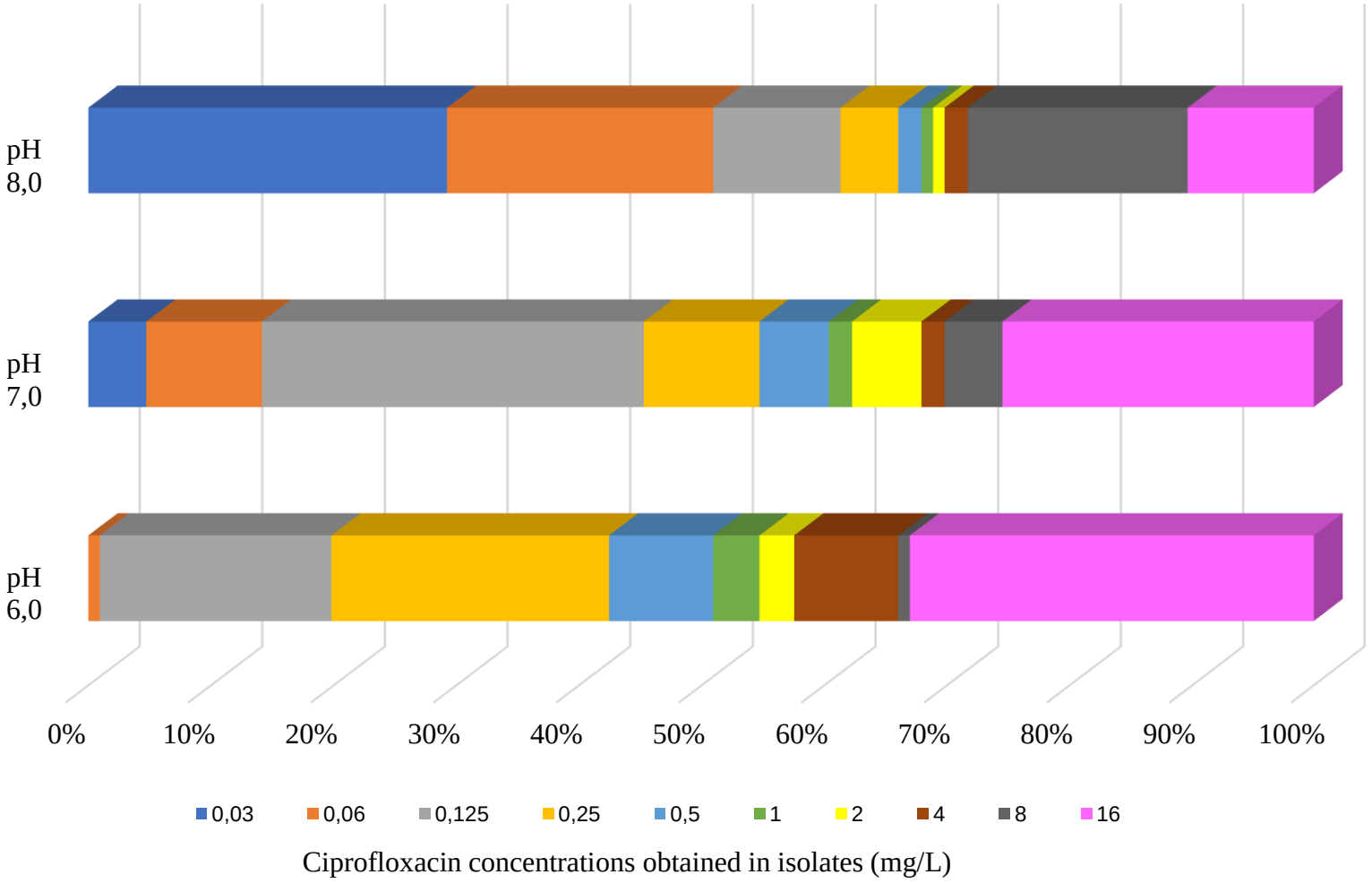
Table 2. *In vitro* sensitivity to ciprofloxacin by the automated Phoenix®-BD method of 90 *E. coli* clinical isolates compared to the urine from which these isolates were recovered.

Microorganism	Number of isolates	pH urinary	Number of isolates with the following MICs (mg/L):					% of isolates CLSI		
			0,124	0,25	0,5	1	2	S	I	R
E. coli	90*	5,0	-	2	-	-	4	33	-	67
		5,5	5	1	1	-	5	50	8	42
		6,0	4	-	-	-	20	17	-	83
		6,5	8	-	2	-	8	44	2	44
		7,0	18	1	1	1	5	73	4	23
		8,0	4	-	-	-	-	100	-	-

Note: * Some isolates were unable to recover the pH data from urinalysis, due to the fact that the test was not performed.

200

201 **Figure 1.** Correlation of pH 6.0, 7.0 and 8.0 with the MIC values obtained by the dilution agar method at the different pHs tested by the % of *E. coli*
202 isolates.



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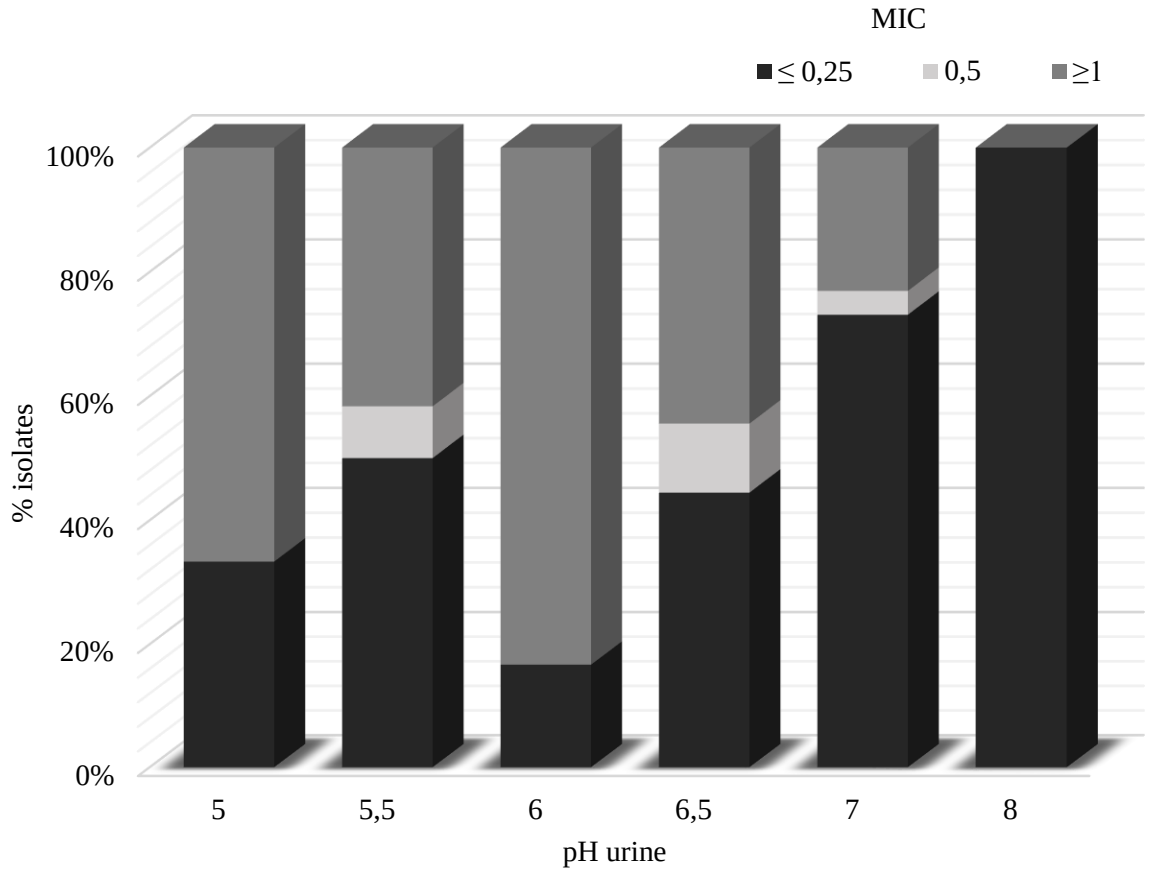
204204

205 **Figure 2.** Correlation of the urinary pHs found in the partial urine test with the MIC values obtained by the automated method at the different pHs
206 tested by the % of *E. coli* isolates.



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210210
211211
212 **Figura 3.** Distribution of ciprofloxacin species as a function of pH.
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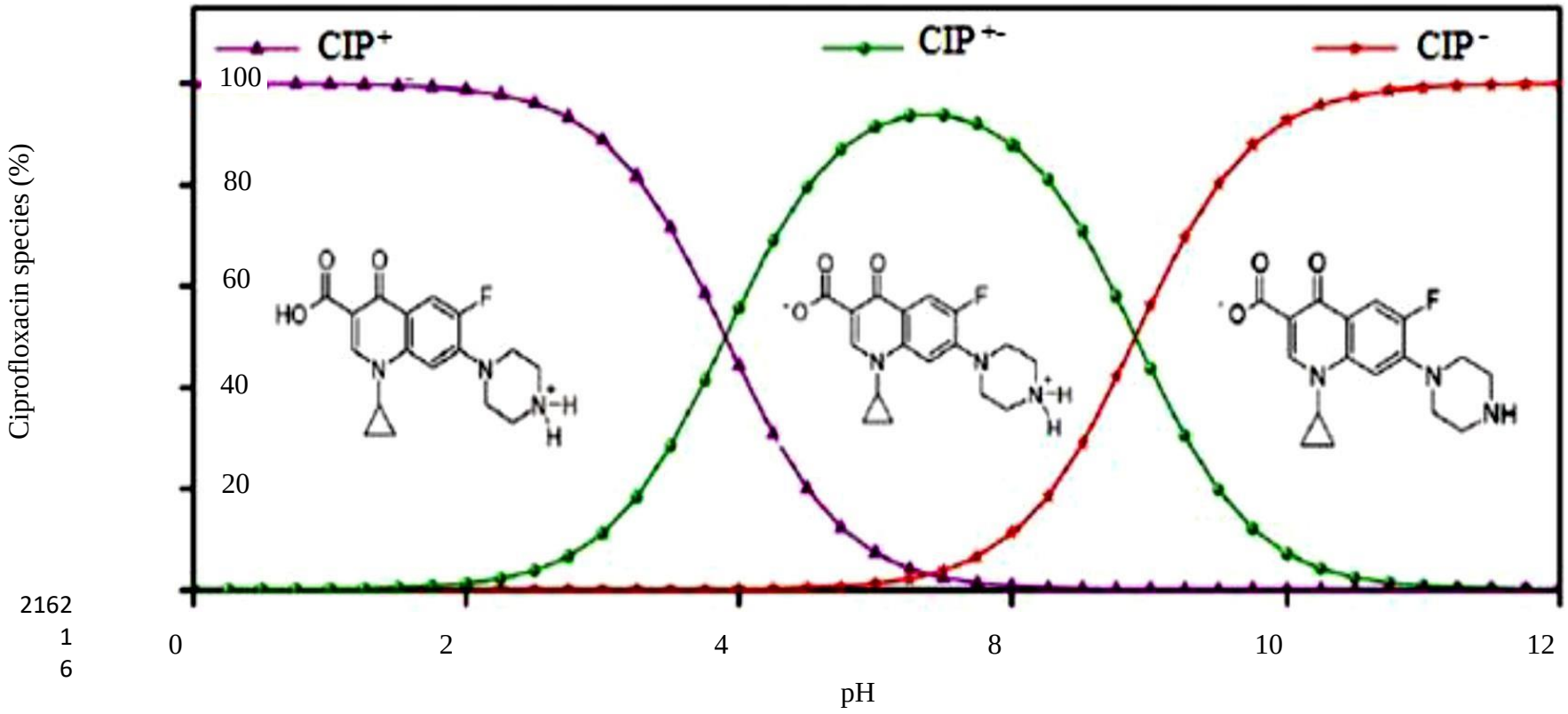

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References

1. McLellan LK, Hunstad DA. 2016. Urinary Tract Infection: Pathogenesis and Outlook. Trends Mol Med 22(11):946-957. doi: 10.1016/j.molmed.2016.09.003.
2. Zare F, Rostami FM, Shahsafi M. 2018. Prevalence and pattern of antibiotic resistance of gram-negative bacteria isolated from urinary tract infections in patients referring to Neka Laboratories-Iran. Int J BioMed Public Health 1(1):30-36. doi: 10.22631/IJBMPH.2018.56097.
3. Gurevich E, Tchernin, D, Schreyber R, Muller R, Leibovitz E. 2016. Follow-up after infants younger than 2 months of age with urinary tract infection in southern Israel: epidemiologic, microbiologic and disease recurrence characteristics. Braz J Infect Dis 20:19-25. doi: 10.1016/j.bjid.2015.09.003.
4. Campos ACC, Andrade NL, Ferdous M, Santos CC, Correal JCD. 2018. Comprehensive Molecular Characterization of Escherichia coli Isolates from Urine Samples of Hospitalized Patients in Rio de Janeiro, Brazil. Front Microbiol 16(9):243. doi: 10.3389/fmicb.2018.00243.
5. Wagenlehner FME, Naber KG. 2006. Treatment of bacterial urinary tract infections: presence and future. Eur Urol 49(2):235-44. doi: 10.1016/j.eururo.2005.12.017.
6. Pfeifer Y, Cullik A, Witte W. 2010. Resistance to cephalosporins and carbapenems in Gram-negative bacterial pathogens. Int J Med Microbiol 300(6):371-9. doi: 10.1016/j.ijmm.2010.04.005.
7. Cuba GT, Pignatari ACC, Patekoski KS, Luchesi LJ, Kiffer CRV. 2014. Pharmacodynamic profiling of commonly prescribed antimicrobial drugs against *Escherichia coli* isolates from urinary tract. Braz J Infect Dis 8(5):512–517. doi: 10.1016/j.bjid.2014.01.008.
8. Garoff L, Huseby DL, Praski AL, Hughes D. 2019. Effect of aminoacyl-tRNA synthetase mutations on susceptibility to ciprofloxacin in *Escherichia coli*. J Antimicrob Chemother 73 (12): 3285-3292. doi: 10.1093/jac/dky356.

9. Rehman A, Patrick WM, Lamont IL. 2019. Mechanisms of ciprofloxacin resistance in *Pseudomonas aeruginosa*: new approaches to an old problem. J Med Microbiol 68:1–10. doi: 10.1099/jmm.0.000873.

10. Aldred KJ, Kerns RJ, Osheroff N. 2014. Mechanism of quinolone action and resistance. Biochemistry 53:1565-1574. doi: 10.1021/bi5000564.

11. Bel FD, Dewulf J, Witte BD, Langenhove HV, Janssen C. 2009.Influence of pH on the sonolysis of ciprofloxacin: Biodegradability, ecotoxicity and antibiotic activity of its degradation products. Chemosphere 77:291–295. doi: 10.1016/j.chemosphere.2009.07.033.

12. Cunha BA. 2016. An infectious disease and pharmacokinetic perspective on oral antibiotic treatment of uncomplicated urinary tract infections due to multidrug-resistant Gram-negative uropathogens: the importance of urinary antibiotic concentrations and urinary pH. Eur J Clin Microbiol Infect Dis 35:521–526. doi: 10.1007/s10096-016-2577-0.

13. Fedrigo NH, Mazucheli J, Albiero J, Shinohara, DR, Lodi FG, Machado ACS, Sy SKB, Tognim MCB. 2017. Pharmacodynamic evaluation of fosfomycin against *E. coli* and *Klebsiella* spp. from urinary tract infections and the influence of pH on fosfomycin activities. Antimicrob Agents Chemother 61:AAC.02498-16. doi: 10.1128/AAC.02498-16.

14. Poole K. 2012. Bacterial stress responses as determinants of antimicrobial resistance. J Antimicrob Chemother 67(9):2069-2089. doi: 10.1093/jac/dks196

15. Yang L, Whag K, Li H, Denstedt JD, Cadieux PA. 2014. The influence of urinary pH on antibiotic efficacy against bacterial uropathogens. Urology. 84:713. Wl-713.e7. doi: 10.1016/j.urology.2014.04.048.

16. Begic S, Worobec EA. 2006. Regulation of Serratia marcescens ompF and ompC porin genes in response to osmotic stress, salicylate, temperature and pH. Microbiology 152:485–491. doi: 10.1099/mic.0.28428-0.

17. Clinical and Laboratory Standards Institute (CLSI). 2020. Performance standards for antimicrobial susceptibility testing. 30th informational supplement M100. Clinical and Laboratory Standards Institute, Wayne, PA.
18. Associação Brasileira de Normas Técnicas. NBR 15268: Laboratório clínico -Requisitos e recomendações para o exame de urina. ABNT. Brasil, 2005.
19. Kamberi M, Tsutsumi K, Kotegawa T, Koichi Kawano, Koichi Nakamura, Yoshihito Niki, Nakano S. 1999. Influences of urinary pH on ciprofloxacin pharmacokinetics in humans and antimicrobial activity *in vitro* versus those of sparfloxacin. Antimicrob Agents Chemother 43(3):525–529. doi: 10.1128/AAC.43.3.525.
20. Jiang W Changa P, Wanga Y, Tsai Y, Jeana J, Li Z, Krukowski K. 2013. Removal of ciprofloxacin from water by birnessite. J. Hazard. Mater, 2013 250:362-369. doi: 10.1016/j.jhazmat.2013.02.015.
21. Jalil, MER, Baschini, M, Sapag, K. 2015. Influence of pH and antibiotic solubility on the removal of ciprofloxacin from aqueous media using montmorillonite. Applied Clay Science 114:69-76. doi: 10.1016/j.clay.2015.05.010.
22. Smith, J. T., and N. T. Ratcliffe. 1986. Effects of pH and magnesium on the activity of 4-quinolone antibacterials. Infection 14(Suppl. 1):31–35. doi: 10.1007/BF01645195.
23. Perez-Giraldo C, Hurtado C, Moran FJ, Blanco MT, Gomez- Garcia AC. 1990. The influence of magnesium on ofloxacin activity against different growth phases of Escherichia coli. J. Antimicrob. Chemother 22:1021–1022. doi: 10.1093/jac/25.6.1021.
24. Aagaard J, Madsen PO, Rhodes P, Gasser T. 1991. MICs of ciprofloxacin and trimethoprim for Escherichia coli: influence of pH, inoculum size and various body fluids. Infection 3(Suppl.):S167–S169. doi: 10.1007/BF01643691.

296 **Table Legends**

297

298 **Table 1.** *In vitro* susceptibility by the agar method diluted to pH 6.0, 7.0 and 8.0 for

299 ciprofloxacin in 106 clinical isolates of *E. coli* from urinary tract infections.

300300

301 This table contains the *in vitro* sensitivity information obtained by the dilution agar method at pH

302 6.0 7.0 and 8.0 tested for ciprofloxacin in 106 clinical isolates of *E. coli* from urinary tract

303 infections by correlating the pH and the sensitivity profile.

304304

305 **Table 2.** In vitro sensitivity to ciprofloxacin by the automated Phoenix®-BD method of 90 *E.*

306 *coli* clinical isolates compared to the urine from which these isolates were recovered.

307307

308 This table correlates the *in vitro* sensitivity to ciprofloxacin obtained by the automated

309 Phoenix®-BD method of 90 *E. coli* clinical isolates compared to the urine from which these

310 isolates were recovered and the pHs of the urine samples from the urine test

311 **Figure 1.** Correlation of pH 6.0, 7.0 and 8.0 with the MIC values obtained by the dilution agar

312 method at the different pHs tested by the % of *E. coli* isolates. Sensitive, MIC of ≤ 0.25 mg/L;

313 intermediate, MIC of 0.5 mg/L; resistant, MIC of ≥ 1 mg/L.

314 **Figure 2.** Correlation of the urinary pHs found in the urine test with the MIC values obtained by

315 the automated method at the different pHs tested by the % of *E. coli* isolates. Sensitive, MIC of ≤

316 0.25 mg/L; intermediate, MIC of 0.5 mg/L; resistant, MIC of ≥ 1 mg/L.

317 **Figure 3.** Distribution of ciprofloxacin species as a function of pH.

318 This figure represents the 3 states of the ciprofloxacin molecule at 3 different pHs that vary

319 below 6.1, when the molecule is positive; 6.1 and 8.7, when the molecule is neutral and above

CAPÍTULO III

3.1 CONCLUSÕES

O aumento de resistência aos antimicrobianos está diretamente ligada à sua prescrição e utilização incorreta. A escolha do amtimicrobiano correto é de extrema importância, pois, ele que impede o desenvolvimento bacteriano. Podemos concluir com os achados do presente estudo que:

- Foi possível observar que a urina e, em particular, o seu pH influenciam na atividade do ciprofloxacino, dando ênfase ao pH ácido que é comumente encontrado no trato urinário e que pode diminuir sua atividade.

- A importante influência do pH sobre a atividade do ciprofloxacino o que alerta para que este fato seja considerado no momento da terapêutica.

3.2 PERSPECTIVAS

Diante disso, é fundamental levar em consideração o pH da urina par a escolha do melhor tratamento. Uma maneira simples é conhecer pH facilmente medido na rotina laboratorial e que seria de grande valia, para à escolha do antimicrobiano, bem como o uso de substâncias possam ser administradas concomitantemente ao antimicrobianos favorecendo ao sucesso terapêutico das ITUs.