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Research Article

Antibacterial and Anti-Urease Activities of Chloropheniramine maleat, Paracetamol and Clarithromycine

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ABSTRACT

This paper presents anti-urease and antibacterial activities of chloropheniramine maleat, paracetamol and clarithromycine. Antibacterial activity of the compounds was investigated against *Salmonella enterica, Enterobacter aerogenes, Bacillus subtilis, Proteus vulgaris, Gordonia rubripertincta, Klebsiella pneumoniae* and *Enterococcus faecalis*. Moreover, anti-urease activity of the compounds was searched. All tested compounds demonstrated antibacterial action with varying degree except for chloropheniramine maleat. Chloropheniramine maleat, paracetamol and chlarithromycine exhibited higher activity than thiourea. The data obtained from the study, chloropheniramine maleate, paracetamol and clarithromycin are thought to be useful as new urease inhibitors and the present study should be supported by further studies. Furthermore, it is thought that paracetamol has antibacterial properties and therefore can be used as an alternative to antibiotics.

Keywords: *Anti-urease, Antibiotic, Antibacterial, Clarithromycine, Chloropheniramine maleat, Paracetamol*

Klorofeniramin Maleat, Parasetamol ve Klaritromisin'in Antibakteriyal ve Anti-Üreaz Aktiviteleri

ÖZET

Bu çalışmada klorofeniramin maleat, parasetamol ve klaritromisin'in antibakteriyal ve anti-üreaz aktiviteleri ile ilgilenmektedir. Maddelerin antibakteriyal aktiviteleri *Salmonella enterica, Enterobacter aerogenes, Bacillus subtilis, Proteus vulgaris, Gordonia rubripertincta*, Klebsiella pneumoniae, *Bacillus megaterium* ve Enterococcus faecalis'e karşı araştırıldı. Bu maddelerin anti-üreaz aktiviteleri de incelendi. Klorofeniramin maleat dışında, test edilen tüm bileşikler değişik oranlarda antibakteriyal etki gösterdi. Klorofeniramin maleat, parasetamol ve klaritromisin tiyoüreden daha yüksek aktivite gösterdi. Çalışmadan elde edilen veriler, klorofeniramin maleat, parasetamol ve klaritromisin yeni üreaz inhibitörü olarak kullanılabileceğini düşünülmüş olup mevcut çalışma daha ileri çalışmalar ile de desteklenmelidir. Ayrıca, parasetamolün antibakteriyel özelliklere sahip olduğu ve bu nedenle antibiyotiklere alternatif olarak kullanılabileceği düşünülmektedir.

Anahtar kelimeler: *Anti-üreaz, Antibiyotik, Antibakteriyal, Klaritromisin, Klorofeniramin maleat, Parasetamol*

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I. INTRODUCTION

The number of multi-drug resistant bacteria has increased. This rise has been linked with indiscriminate utilizing of antibiotics and immunosuppressive medicines. Moreover, in synthetic medicines are not costly and insufficient to cure of illnesses in developing countries [1]. Eventhough resistance to antimicrobial agents amongst important bacterial pathogens throughout the world are increasing, the number of brand antimicrobials which present in market has undergone a stable decrease especially in the past decades [2]. Hence, there is demand to investigate new infection-fighting agents to combat microbial infections [1].

Gastrointestinal diseases like gastritis are commonly caused by *Helicobacter pylori*. This bacterium leads diseases such as urinary stone formation, peptic ulcer, pyelonephritis, and hepatic coma. *H. pylori* habitance in the acidic medium of the stomach is highly dependence on the urease enzyme activity. Urease enzyme converts the stomach medium into a tolerable condition for the bacteria via neutralizing gastric acid through breakdown urea to form carbon dioxide and ammonia [3]. Hence, there is a demand to find brand urease inhibitors to avoid this illnesses. Recently, many medicines have been recommended as urease inhibitors [4].

Clarithromycin is a new semi-synthetic macrolide which derived from erythromycin [5]. Clarithromycin possess bactericidal action towards respiratory pathogens [6]. Chloropheniramine maleate has been utilized to relieve symptoms of cold and cure the allergic illnesses [7]. Paracetamol generally utilized for acute musculoskeletal pains [8].

In the current survey, it was aimed to reveal antibacterial and urease enzyme inhibition activity of clarithromycin, chloropheniramine maleat and paracetamol which are used as active ingredients of some medicines. This is the first study about antibacterial activity of chloropheniramine maleat. Also, this is the first record related to anti-urease activity of clarithromycin, chloropheniramine maleat and paracetamol.

II. MATERIAL AND METHOD

A. CHEMICALS

Chloropheniramine maleat, Paracetamol and Clarithromycine obtained from Atabay pharmaceutical company, Turkey.

B. BACTERIA

Salmonella enterica ATCC 14028 was obtained from Giresun Province Control Laboratory; *Enterobacter aerogenes* CCM 2531, *Bacillus subtilis* IMG 22 and *Proteus vulgaris* FMC 1 were obtained from Fırat University Department of Biology; *Gordonia rubripertincta* (lab isolate) and *Klebsiella pneumoniae* (lab isolate) were acquired from Yeditepe University Department of Genetic and Bioenginneering; *Enterococcus faecalis* was obtained from Rize Tayyip Erdoğan University Department of Biology.

C. ANTIBACTERIAL ACTIVITY

Minimum Inhibition Concentration (MIC) assay was determined with microbroth dilution method [9].

D. ANTI-UREASE ACTIVITY

Urease inhibitor activity was determined using Van Slyke and Archibald's method [10]. Test compounds were prepared at 4 different concentrations (0.00001-0.01 μg/mL). 0.5 mL test compound was mixed with 0.5 mL of urease which prepared with solution (pH = 6.8) in 100 mM 16 mg/mL phosphate buffer. This mixture was incubated at room temperature for 15 min. Then, 0.4 ml phenol red prepared in urea-phosphate buffer was added to the mixture and absorbance was read at 570 nm. Inhibition was calculated from the equation 1 below:

$$\%\ inhibition = \frac{(\mathrm{AO}-\mathrm{A1})}{\mathrm{A0}} X100 \qquad [1]$$

A_0: Absorbance of control
A_1: Absorbance of sample

IC_{50} value of the urease enzyme (the amount of substance required for 50% inhibition of the enzyme) was calculated from the regression equation.

III. RESULTS AND DISCUSSION

A. ANTIBACTERIAL ACTIVITY

MIC defines as the minimum concentration of the antibacterial compound which inhibits the bacterial growth after 24 h [11]. Table 1 shows antimicrobial action of the compounds and standard. Antimicrobial effect was expressed as MIC value. All tested compounds demonstrated antibacterial action with varying degree except for chloropheniramine maleat. The most compound was clarithromycine but all the compounds possess lower activity than ciprofloxacine which used as standard agent. According to obtained results, MIC values of the compounds varied within the range from 0.125 mg/mL to 0.25 mg/mL and from 0.0625 mg/mL to 0.125 mg/mL for paracetamol and clarithromycin, respectively.

Table 1. *MIC values of the tested compounds and standards (mg/mL)*

G (+) / G (-)	Bacteria	Chloropheniramine maleat	Paracetamol	Clarithromycine	Ciprofloxacin
G (-)	*S. enterica*	NA	0.25	0.125	0.00781
G (-)	*E. aerogenes*	NA	0.25	0.0625	0.25
G (+)	*B. subtilis*	NA	0.125	0.0625	0.015625
G (-)	*K. pneumoniae*	NA	0.125	0.125	0.03125

Table 1. (continue). *MIC values of the tested compounds and standards (mg/mL)*

G (+) / G (-)	Bacteria	Chloropheniramine maleat	Paracetamol	Clarithromycine	Ciprofloxacin
G (+)	*G. rubripertincta*	NA	0.25	NA	0.015625
G (-)	*P. vulgaris*	NA	0.25	0.0625	0.00781
G (+)	*E. faecalis*	NA	0.25	0.125	0.0039

NA: No Activity

This is the first record related to antibacterial efficiency of chloropheniramine maleat.

Pathogenic bacteria have been thought as a major reason for mortality in humans. Although pharmaceutical industry have manufactured many brand antibacterial agents in last years but bacterial resistance to these antibacterial agents is public health concern [12].

There are some studies related to antimicrobial activity of paracetamol and clarithromycine. For example, Zimmermann and Curtis (2017) reported that MIC values of paracetamol as 2.5 mg/mL, 5 mg/mL, 2.5 mg/mL, 2.5 mg/mL and 1.25 mg/mL against *Bacillus* spp., *Enterobacter cloacae*, *Escherichia coli*, *Salmonella enterica serovar typhi* and *Staphylococcus aureus*, respectively [13]. In this current study, MIC values were found as 0.25 mg/mL against *S. enterica* and 0.125 mg/mL against *B. subtilis*.

Hussain and Al-Janabi (2010) declared that MIC values of paracetamol as 0.312 mg/mL against *E. coli*, *Salmonella typhi*, *E. cloaceae*, *S. aureus* and *B. subtilis* [14]. On contrast to this study, in our study, MIC values was detected for *E. aerogenes* as 0.25 mg/mL and for *B. subtilis* as 0.125 mg/mL.

Ferrero et al. (1996) reported that clarithromycine possess activity against *S. aureus*, *Streptococcus pyogenes*, *Streptococcus pneumoniae*, *Moraxella catarrhalis* and *Haemophilus influenza* [15]. According to a study which was carried out by Bergman et al (1999) clarithromycine had great activity on *S. pneumoniae* [16].

Different antibacterial results could be explained with using different bacterial strains and different concentration of the test compounds.

B. ANTI-UREASE ACTIVITY

The data for the determination of anti-urease activity of the compounds are given in Table 2. It was found that anti-urease activity rised linearly with the rise in the concentration of the test compounds. Chloropheniramine maleat, paracetamol and chlarithromycine exhibited higher activity than thiourea.

Table 2. *Anti-urease activity of the test compounds (µg/mL)*

Compound	Concentration	% inhibition	IC_{50} value
Chloropheniramine maleat	0.00001	23.24±3.15	0.008575±0.0002
	0.0001	35.45±2.35	
	0.001	46.51±1.91	
	0.01	53.92±1.70	
Paracetamol	0.00001	31.87±2.40	0.006345±0.0002
	0.0001	45.28±1.20	
	0.001	57.45±2.95	
	0.01	73.40±2.48	
Chlarithromycine	0.00001	18.72±2.17	0.00986±0.004
	0.0001	30.22±1.50	
	0.001	39.29±2.82	
	0.01	47.03±2.17	
Thiourea	0.00001	30.78±0.47	0.01052±0.0003
	0.0001	34.90±0.48	
	0.001	38.80±0.13	
	0.01	43.76±1.54	

Acetohydroxamic acid and phosphoramidates which are urease inhibitors have exhibited curative impact, restrictions are linked heavy adverse impacts like psycho-neurological symptoms and teratogenicity so they have restricted their utilizing in the cure of urinary and gastrointestinal tracts disorders. Hence, the exploration for different type of urease inhibitors which possess the lowest side effects has acquire much interest [17].

As far as we know, this is also the first record about anti-urease activity of chloropheniramine maleat, paracetamol and clarithromycin.

IV. Conclusion

This is the first record of anti-urease inhibition of chloropheniramine maleat, paracetamol and clarithromycin. Our results clearly demostrated urease inhibitory effect of the test compounds. According the data obtained from the study demonstrates that chloropheniramine maleat, paracetamol and clarithromycin might be used new urease inhibitor. Also, it was found that paracetamol possess antibacterial property so it may be used as an alternative to antibiotics.

V. References

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