

## Antibacterial effects of Azerbaijan Garlic (*Allium sativum*)

Mohammad Khalegi<sup>1</sup>, Behnaz Salahi Eshlaghi<sup>2</sup>, Reza Ghotaslou<sup>3,4\*</sup>

1-Rab-e -Rashidi University, Tabriz, Iran

2-Research Center of Midwife School, Tabriz University of Medical Science, Tabriz, Iran

3- Immunology Research Center, Tabriz University of Medical Sciences, Tabriz, Iran

4- Department of Microbiology, School of Medicine, Tabriz University of Medical Sciences, Tabriz, Iran

\*Corresponding author: [rzgottaslo@yahoo.com](mailto:rzgottaslo@yahoo.com)

(Received 18 June 2016, Accepted 06 September 2016)

### Summary

Bacteria have an important role in human and animal infections and bacterial resistance to antibiotics is a major problem following repetitive antibiotic therapy. Garlic has an antimicrobial activity. The aim of this study was to evaluate *in vitro* antibacterial effects of Azerbaijan garlic against some bacteria. Fifty clinical isolates and five control strains of bacteria included *Escherichia coli*, *Pseudomonas aeruginosa*, *Klebsiella pneumonia*, *Staphylococcus epidermidis* and *Staphylococcus aureus* were selected for the study. The MIC and MBC of garlic on bacteria were determined by microdilution assay. The garlic extract showed diverse activity against the bacteria. The MIC and MBC of garlic on gram-positive bacteria were generally lower than gram-negative bacteria. This study confirms the antimicrobial potential of garlic, and garlic may be appropriate plant for treatment and prevent of infections.

**Keywords:** Antimicrobial effect, Bacteria, Garlic.

### Introduction

Garlic (*Allium sativum*) has been found to avoid many infectious diseases in human and animals. Recently, many modern researches confirmed that garlic has antimicrobial properties and is useful against a wide-range of bacteria, fungi, and viruses (Shuford et al., 2005; Low et al., 2008). Additionally, previous reports have shown a synergistic antibacterial effect when garlic

extract and antibiotics are combined (Li et al., 2015). It is well known that the antibiotic activities of this plant are related to the presence of some bioactive materials (Groppo et al., 2008). However, most former studies had mainly focused on the antimicrobial activities of local garlic (Shuford et al., 2005; Low et al., 2008) while little is known about the potential of Azerbaijan garlic.

Considering the importance of infectious diseases and the increased drug resistance, researchers are searching for some alternative ways to prevent infectious diseases. Therefore, the purpose of this study was to investigate *in vitro* activity of Azerbaijan garlic against some bacteria.

## Materials and Methods

### *Bacterial isolates*

Bacteria were isolated from various clinical samples consisted of; *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumoniae*, *Staphylococcus epidermidis* and *Pseudomonas aeruginosa*. Five strains of bacteria from Persian Type Culture Collection (PTCC) were also tested as control strain including *Escherichia coli* PTCC 1112, *Pseudomonas aeruginosa* PTCC 2381, *Klebsiella pneumoniae* PTCC 1290, *Staphylococcus epidermidis* PTCC 1114 and *Staphylococcus aureus* PTCC 1311.

### *Garlic Extract*

Garlic extract (GE) was prepared as previously described (Lemar et al, 2002). Briefly, 40g fresh garlic cloves were blended in a clean mortar and pressed with gauze. Then, GE was added in sterile saline at a concentration of 40mg/mL and was centrifuged (3000 rpm for 15 min). Finally, it was filtered with a 0.45 mm filter and stored at -20°C until use. All tests were performed in triplicate on three different times.

### *The MIC, MBC*

Microdilution method was used to determine the minimum inhibitory concentration (MIC) of GE (Ghotaslou and Bahrami, 2012). At first, 9 successive

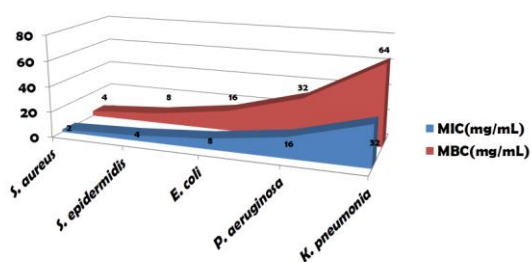
dilutions of GE (1, 2, 4, 8, 16, 32, 64, 128 and 256 mg/mL) were prepared and well 10 and 11 were used as the negative and positive controls for garlic extract, respectively. Each well has been filled with 100 mL Trypticase Soy Broth medium (Merck, Germany) containing various concentrations of garlic extract and different strains of bacteria diluted in 0.5 MC Farland medium and incubated overnight at 37°C. Additionally, the lowest concentration of GE that allows growth of less than 0.1% of the control culture is considered as minimal bactericidal concentration (MBC).

### *Statistical analysis*

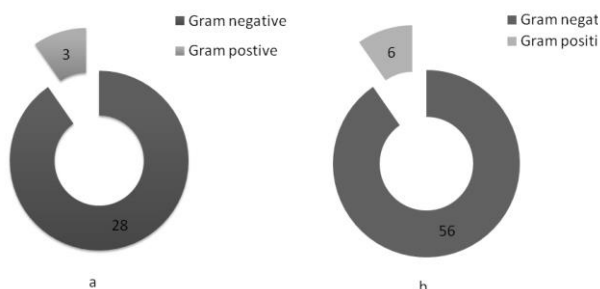
Data were entered into the SPSS software version 18 and the results were analyzed by ANOVA test. In this study  $P \leq 0.05$  was regarded statistically significant.

## Results

The results of MIC and MBC are presented in Fig 1. GE showed the lowest MIC (2 mg/mL) against *Staphylococcus aureus*. The mean MIC against gram-negative bacteria and gram-positive bacteria were 28 and 3 mg/mL, respectively. Additionally, their mean MBC against gram-negative bacteria and gram-positive bacteria were 56 and 6 mg/mL, respectively. One-way ANOVA test showed that the MIC and MBC of GE were significantly different in various gram-positive and gram-negative bacteria (Fig 2).



**Fig 1:** The MIC and MBC (mg/mL) results of garlic against tested bacteria.



**Fig 2:** Comparison the mean MIC and MBC (mg/mL) of garlic between Gram positive and Gram negative bacteria. a, the mean MIC and b, the mean MBC

**Discussion**

Garlic is a vital universal food and it has long been known to have antibacterial effects. It seems that the main antimicrobial activity of garlic is derived from allicin, a sulfur-containing compound that is formed at levels of approximately 3 to 5 mg/g of fresh garlic (Ankri, 1999; Rybak et al., 2008).

In the present study, the comparison of MIC indicates that the lowest MIC was against *Staphylococcus aureus* and *Staphylococcus epidermidis* (2 mg/ml and 4 mg/ml, respectively) and the highest MIC was against *K. pneumonia* and *Pseudomonas aeruginosa* (32 mg/ml and 16 mg/ml, respectively). According to the results of this study, there are also significant differences between gram positive and gram negative bacteria in terms of MIC and MBC ( $P \leq 0.05$ ). Based on the results of MIC values in this study, GE was more effective on gram-positive bacteria in comparison to gram-negative bacteria, whilst MIC of *Klebsiella pneumonia* was eighteen times more than *Staphylococcus aureus*. This may be due to physiochemical features of the cell wall of bacteria, the diameter of peptidoglycan layer and lipopolysaccharide of gram-negative bacteria. Consequently, species-specific bacteria are able to react differently to GE.

The current study showed that GE had potent anti-bacterial effects. Both *in vitro* and *in vivo* studies have shown that GE has decreased the bacterial virulence (Harjai et al., 2010; Smyth et al., 2010; Lee et al., 2011). Harjai et al. (2010) in a study in a mouse model of hospital catheter-related urinary tract infections have demonstrated that oral management with GE has significantly lowered renal bacterial counts and protected mouse kidney from tissue destruction. Smith et al. (2010) reported an improvement in lung function in cystic fibrosis patients with garlic. In contrast to the results of the present study, Lee et al. (2011) described that GE regardless of its antibacterial function enhanced *Streptococcus mutans* attachment on the

orthodontic wire. In the present study, the phytochemical analysis was not evaluated for determination of the dominant chemical compounds of Azerbaijan garlic. It is reasonable to suggest further *in vivo* study to determine the mechanism of the antibacterial effects of garlic that may help to design the new drug to control the spread of resistant bacterial strains among human and animal infections. In conclusion, this study has shown that GE has antibacterial activity against bacteria and may be useful for curing infectious diseases.

### Acknowledgments

We thank the staff of microbiology laboratory, Tabriz University of Medical Sciences for assisting in the project.

**Conflict of interest:** We wish to confirm that there are no known conflicts of interest associated with this publication and there has been no significant financial support for this work that could have influenced its outcome.

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