**Sumac (Rhus coriaria L.) Represents a Considerable Antibacterial Activity Against Meticillin Susceptible and Meticillin Resistant Staphylococcus aureus**

Mohammad Mahdi Ahmadian-Attari†, Meyesm Khanlarbeik‡, Mohammad Reza Fazeli†, Hossein Jamalifar‡

†Evidence-Based Phytotherapy and Complementary Medicine Research Center, Alborz University of Medical Sciences, Karaj, Iran
‡Department of Drug and Food Control and Pharmaceutical Quality Assurance Research Center, Faculty of Pharmacy, Tehran University of Medical Sciences, Tehran, Iran

These authors have equal contribution as joint first authors.

**Abstract**

**Background:** Staphylococcus aureus is one of the most important pathogens and the cause of suppuration, abscess formation, a variety of pyogenic infections, and even fatal septicemia in human beings. Isolation of resistant strains of S. aureus especially meticillin-resistant one has elucidated the importance of finding new antibacterial agents. Sumac is one of the important medicinal herbs of Iranian traditional medicine (ITM). Sumac has been traditionally prescribed for some ailments with infectious etiology and therefore, it probably contains antibacterial compounds. Recently, antibacterial activities of sumac against some bacteria were studied by us in our team.

**Objective:** In this article, we aimed to compare the antibacterial activities of sumac against meticillin susceptible (MSSA) and meticillin resistant S. aureus (MRSA) via minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) determination.

**Materials and Methods:** Antibacterial activity of sumac water extract was studied on 19 clinical and 1 standard strain of S. aureus via MIC and MBC determination. Based on their sensitivity to cloxacillin, these bacteria were classified as meticillin-susceptible, intermediate meticillin-resistant, and meticillin-resistant. MICs and MBCs of the extracts in these 3 groups were statistically compared via one-way analysis of variance (ANOVA) followed by a post hoc analysis (Tukey HSD test) using SPSS for Windows (version 15).

**Results:** The results showed that in spite of different susceptibilities to cloxacillin, susceptibility to sumac extract was not different and MICs and MBCs of sumac for all clinical isolates including MSSA and MRSA were similar (P > 0.05).

**Conclusion:** Based on previous studies, sumac extract acts against S. aureus probably through changing the cell wall properties. This activity is similar for both MRSA and MSSA. More investigation on the precise mechanism of action of this extract would be fruitful.

**Background**

Meticillin-resistant Staphylococcus aureus (MRSA), which its resistance is due to the production of penicillin-binding protein (PBP) 2, has been recently emerged as one of the most important nosocomial and community pathogens. According to a European report, MRSA population-weighted mean in European Union is 17.8%, highlighting that MRSA remains a public health priority.1 MRSA is resistant to not only meticillin and other β-lactams but also to many other antimicrobials. This has now resulted in multiresistance of MRSA. Today, some glycopeptides such as vancomycin are widely prescribed for serious MRSA infections; however, the emergence of vancomycin tolerance and resistance has complicated treatment and highlighted the clinical need for new antibiotics which can work against MRSA and other Gram-positive bacteria.2

Sumac (Rhus coriaria L.) is a shrub, inhabiting the regions from Mediterranean Sea to Iran and Afghanistan. Its fruit is widely used in Middle Eastern cuisine especially in Iran and Turkey.3 Sumac fruit has also been mentioned in Iranian traditional medicine (ITM) as a herb with some therapeutic potentials. Traditionally, the powdered fruits have been prescribed as astringent, anti-diarrhea, anti-trachoma, and anti-pus in infectious wounds.4

Inspired from ITM, we previously planned a series
of investigations on sumac antibacterial activities. In these studies, we showed that sumac has considerable antimicrobial activities against some standard and susceptible strains of gram-positive bacteria such as Bacillus cereus, Corynebacterium xerosis, Staphylococcus epidermidis, and S. aureus.

Objectives
The present study was aimed to compare the antibacterial activities of sumac against both meticillin-susceptible S. aureus (MSSA) and MRSA via minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) determination.

Materials and Methods
Plant Material and Extraction
Rhus coriaria L. was obtained from local botanical market and confirmed by Dr. Gholamreza Amin, at Herbarium of the School of Pharmacy, Tehran University of Medical Sciences. One hundred grams of the powder of R. coriaria L. epicarp was soaked in 1000 mL of distilled water at 45°C for 5 days. The extract was concentrated in a rotary evaporator (Heidolph, Germany) and was left to dry in a desiccator. The yield of extract (calculated as weight of dry extract/weight of dry starting material ×100) was 43.6 (w/w). The extract was diluted at desirable concentrations in distilled water and was filter sterilized prior to antibacterial assays.

Bacterial Strains
Staphylococcal strains listed in Table 1 were 19 clinical strains isolated from Shariati and Bouali University hospitals and Danesh Pathobiology Laboratory and identified by conventional morphological and biochemical tests. The standard strain of S. aureus ATCC 6539-P, which was meticillin-sensitive, was stock of the Department of Pharmaceutical and Food Control, School of Pharmacy, Tehran University of Medical Sciences, used in assessing the development of antibacterial resistance induced by the extract. Antibiotic susceptibility was determined by the amount of MIC of cloxacillin, in accordance with guidelines of the National Committee for Clinical Laboratory Standards (NCCLS, 1997). After culture on Mueller–Hinton agar (Merck, Germany), the bacteria were suspended in Mueller–Hinton broth (Merck, Germany) and used for inoculation.

Table 1. MICs and MBCs of Sumac, Cloxacillin, and Vancomycin Against Susceptible, Intermediate Resistant, and Resistant Staphylococcus aureus

<table>
<thead>
<tr>
<th>Clinical Isolates</th>
<th>Rhus coriaria L. (mg/mL)</th>
<th>Cloxacillin (μg/mL)</th>
<th>Vancomycin (μg/mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Meticillin-susceptible S. aureus</td>
<td>2.04</td>
<td>1.5 (3.1)</td>
<td>0.8 (1.6)</td>
</tr>
<tr>
<td></td>
<td>8.15 (16.35)</td>
<td>1.5 (3.1)</td>
<td>3.1 (6.2)</td>
</tr>
<tr>
<td></td>
<td>4.07 (16.35)</td>
<td>1.5 (3.1)</td>
<td>1.5 (6.2)</td>
</tr>
<tr>
<td></td>
<td>0.25 (8.15)</td>
<td>0.8 (1.6)</td>
<td>3.1 (6.2)</td>
</tr>
<tr>
<td></td>
<td>4.07 (16.35)</td>
<td>0.25 (4.07)</td>
<td>1.5 (3.1)</td>
</tr>
<tr>
<td>S. aureus ATCC 6539-P</td>
<td>2.04</td>
<td>2.04</td>
<td>1.5 (3.1)</td>
</tr>
<tr>
<td></td>
<td>3.1 (12.5)</td>
<td>3.1 (12.5)</td>
<td>1.5 (3.1)</td>
</tr>
<tr>
<td></td>
<td>2.04 (16.35)</td>
<td>2.04 (16.35)</td>
<td>3.1 (6.2)</td>
</tr>
<tr>
<td></td>
<td>3.1 (12.5)</td>
<td>3.1 (12.5)</td>
<td>6.2 (6.2)</td>
</tr>
<tr>
<td></td>
<td>2.04</td>
<td>2.04</td>
<td>3.1 (25)</td>
</tr>
<tr>
<td></td>
<td>0.25 (16.35)</td>
<td>0.25 (16.35)</td>
<td>3.1 (6.2)</td>
</tr>
<tr>
<td>Intermediate meticillin-resistant S. aureus</td>
<td>4.07 (28.35)</td>
<td>4.07 (28.35)</td>
<td>25 (50)</td>
</tr>
<tr>
<td></td>
<td>2.04 (16.35)</td>
<td>2.04 (16.35)</td>
<td>12.5 (25)</td>
</tr>
<tr>
<td></td>
<td>8.15 (16.35)</td>
<td>8.15 (16.35)</td>
<td>25 (50)</td>
</tr>
<tr>
<td></td>
<td>0.25 (16.35)</td>
<td>0.25 (16.35)</td>
<td>25 (50)</td>
</tr>
<tr>
<td></td>
<td>4.07 (16.35)</td>
<td>4.07 (16.35)</td>
<td>12.5 (25)</td>
</tr>
<tr>
<td></td>
<td>0.25 (28.35)</td>
<td>0.25 (28.35)</td>
<td>50 (50)</td>
</tr>
<tr>
<td></td>
<td>4.07 (16.35)</td>
<td>4.07 (16.35)</td>
<td>12.5 (25)</td>
</tr>
<tr>
<td></td>
<td>1.02 (8.15)</td>
<td>1.02 (8.15)</td>
<td>25 (50)</td>
</tr>
</tbody>
</table>

Determination of MICs and MBCs
The concentrations of the extract were prepared in the range of 0.25 to 56.7 (mg/mL) in Muller-Hinton broth containing 10⁸ CFU/mL of the bacteria. After incubation at 37°C overnight, the test tubes were examined for possible growth and MICs of the extract were determined as the lowest concentration that ended with no growth. Tubes containing concentrations above the MICs were streaked onto Muller-Hinton agar plates to achieve MBCs of the extract against the bacteria. Vancomycin antibiotic was also used as a positive control.

Statistical Analysis
According to the sensitivity of the bacteria to cloxacillin, the bacteria were divided into 3 groups: meticillin-susceptible S. aureus, Intermediate meticillin-resistant S. aureus, and meticillin-resistant S. aureus. To evaluate
the effectiveness of sumac, MICs and MBCs of the extract in these 3 groups were statistically compared via one-way analysis of variance (ANOVA) followed by a post hoc analysis (Tukey HSD test) using SPSS for Windows (version 15.0).

Results
According to the sensitivity to cloxacillin, the clinical isolates were divided into 3 subgroups: methicillin-susceptible S. aureus, intermediate-meticillin resistant S. aureus, and meticillin-resistant S. aureus (Table 1). The statistical analysis showed that in spite of different susceptibilities to cloxacillin, susceptibility to sumac extract was not different and MICs and MBCs of sumac for all clinical isolates including MSSA and MRSA were similar ($P>0.05$). The effect of vancomycin against all strains of S. aureus was also similar (Figure 1A and 1B).

Discussion
Sumac as an herb with a traditional anti-infective background of use has been recently considered in ethnopharmacological studies. Based on a series of investigations, we published several studies on antibacterial activities of sumac. In these studies, sumac antibacterial activities against both gram-positive and gram-negative bacteria including B. cereus, C. xerosis, S. epidermidis, S. aureus, Escherichia coli, Salmonella typhi, Pseudomonas aeruginosa, and Shigella flexneri were well presented. Other studies completed this list and showed that antibacterial activities of sumac are beyond these bacteria. It has also been shown that the total extract of sumac can be stabilized by autoclaving without any changes in its antibacterial activities. In another article published recently, our team showed the major active compounds of sumac extract against bacteria specifically S. aureus. These activities present sumac as a good candidate to be developed as a final product in pharmaceutical dosage form.

To complete the above-mentioned studies and due to the increasing importance of bacterial resistance, this study aimed to evaluate the behavior of sumac extract in the presence of different MSSA and MRSA strains. The results showed that resistance to meticillin family does not affect antibacterial activity of sumac, as their mechanisms of action probably completely differ from each other.

Gallic acid was recently introduced as one of the active compounds of sumac. Based on the work of Borges and colleagues on the mechanism of action of gallic acid against different bacteria, gallic acid caused irreversible changes in membrane properties through hydrophobicity changes, decrease of surface negative charge, and occurrence of local rupture or pore formation in the cell membranes resulting in consequent leakage of essential intracellular constituents.

The other active compound of sumac, 1,2-dioxo-6-hydroxycyclohexadiene-4-carboxilic acid, belongs to quinones. This class of compounds provides a source of stable free radicals and can form irreversible complexes with nucleophilic amino acids in proteins that often cause their function loss and subsequent cell death. Surface-exposed adhesions, cell wall polypeptides, and membrane-bound enzymes are probable targets of quinone oxidization.

As a conclusion, sumac extract acts against S. aureus probably through changing the cell wall properties. This activity is similar for both MRSA and MSSA. More investigation on the precise mechanism of action of this extract would be fruitful.

Authors' Contributions
Study concept and design: MRF and HJ; Data acquisition: MK and MMAA; Analysis and interpretation of data: MMAA and MK; Drafting of the manuscript: MMAA; Study supervision: MRF and HJ.

Ethical Approval
The research followed the principles of Basel Declaration.

Conflict of Interest Disclosures
The authors declare that they have no conflict of interests.

Financial Support
This article is a part of Pharm D. thesis number 4997, granted by Faculty of Pharmacy, Tehran University of Medical Sciences, Tehran, Iran.
Acknowledgments
The authors would like to thank Professor Gh. Amin and Dr. H. Ashtiani for their valuable and helpful contributions.

References