ANTIMICROBIAL EFFECT OF SOUR POMEGRANATE SAUCE ON “KISIR”, A TRADITIONAL APPETIZER

Şeniz KARABIYIKLI¹, Duygu KIŞLA², Şahika E. A. GÖNÜL²

¹Gaziosmanpaşa University, Faculty of Engineering and Natural Sciences, Department of Food Engineering, Tokat /TURKEY
²Ege University, Faculty of Engineering, Department of Food Engineering, Izmir/TURKEY
Some applications such as
- modified atmosphere packaging,
- gamma irradiation,
- organic acids,
- ozone treatment,
- heat,
- steam,
- hot water
have been found to be effective for killing pathogens in fresh or processed foods [1].
• However, acidification by using organic acids and natural acidic fruit juices, is another method that is used extensively in food processing to control the growth and survival of spoilage-causing and pathogenic microorganisms [2,3,4].

• The ability of food-borne pathogenic microorganisms about adapting to acidic conditions is a concern in food safety [5].
Pomegranate
Pomegranate:

- Latin language: *Punica granatum*
- Family: Aunicaceae
- Contains: citric acid, carbohydrates, polyphenoles, polysaccharides, vitamins and minerals.
- Consume as: fresh fruit, fruit juice, fruit juice concentrate, marmalade, wine or liquor.
• Pomegranate is native to the Mediterranean region and has been used extensively in the folk medicine of many countries [6].

• Pomegranate which contains a high proportion of polyphenolic compounds (gallocatechins, delphinidin, cyanidin, and pelargonidin), is well known for its therapeutic uses [7].
In recent decades, pomegranate has been studied for many potential uses including: immunomodulation, atherosclerosis/arteriosclerosis, bacterial infection, fungal infection, parasitic infection, periodontal disease, and food poisoning [8,9,10,11].
Sour Pomegranate Sauce
Sour Pomegranate Sauce:

The definition of the “sour pomegranate sauce” in Turkish Standards (TS 12720/2001) is:

“It is a sour food product, produced by clarifying and evaporating of the fruit juice which was obtained by pressing the pomegranate fruits, used for flavoring some foods.”
Traditional pomegranate sour sauce:

1. Pomegranate
2. Washing
3. Granulation
4. Pressing
5. Boiling
6. Cooling
7. Removing of the solid phase
8. Bottling
Industrial pomegranate sauce:

- Homogenization and Bottling
- Coloring agent, Preservatives
- Glucose, Citric acid, Antioxidant agent
- Pomegranate juice concentrate
POMEGRANATE PRODUCTS

• In this study, five different traditional pomegranate sour sauce (called as C, D, E, H, J in experiments) produced in different regions in Turkey and two different industrial pomegranate sauce (called as A and B in experiments) were analyzed.

• All the samples were kept at 40°C in their original packages.
A – commercial pomegranate sauce
B – commercial pomegranate sauce
C – traditional pomegranate sour sauce
D – traditional pomegranate sour sauce
E – traditional pomegranate sour sauce
H – traditional pomegranate sour sauce
J – traditional pomegranate sour sauce

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“Kısıṛ” is a traditional appetizer which is made of bulgur mainly and the other ingredients could be change according to preferences of the consumers.
• “Kısır” is a complicated product because of the different ingredients and each of these ingredients could effect the results of the experiments.

• Because of this reason, “kısr” samples were prepared in 4 main steps to eliminate this and samples were held from each of these steps.
1. The samples were taken from the first step, just consist of *bulgur*, boiled water, spring onion and parsley.

2. In the second step, tomato paste, black pepper and red pepper were added.

3. In the third step, olive oil and salt were added.

4. In the fourth step, fresh lemon juice was added.
10 g samples were placed into a sterile empty glass jar.
• *Staphylococcus aureus* (ATCC-25923) and *Escherichia coli* O157:H7 (ATCC-43895) were obtained from Ege University, Engineering Faculty, Food Engineering Department.
METHOD
The objective of this study is to investigate the antimicrobial effect of the traditional pomegranate sour sauce and industrial pomegranate sauce on “kısır”.
EXPERİMENTS

1. Detection of the antimicrobial effect of the pomegranate sauces (traditional and industrial) on the natural microflora of the “kısır” samples.

2. Detection of the antimicrobial effect of the pomegranate sauces (traditional and industrial) on the “kısır” samples which were inoculated with Staphylococcus aureus and Escherichia coli O157:H7.
NATURAL MICROFLORA

• “Kısır” samples were treated with each pomegranate product to detect the antimicrobial effect of the pomegranate products on natural microflora.
• The jars were left for 0, 5, 10 and 20 minutes at room temperature.
• Decimal dilutions were prepared and surface plated on TSA (Tryptone Soya Agar, pH 7.3±0.2, Oxoid, CM0131).
• The plates were incubated at 37°C for 24-48 hours.
INOCULATION OF THE SAMPLES

• 100 µl of *Staphylococcus aureus* and *Escherichia coli* O157:H7 cultures, adapted to 50 ppm nalidixic acid, spotted on the surface of “kısır” samples entirely.

• Inoculated samples were kept at 22°C for 2 hours and then at 4°C for 22 hours for the attachment of the microorganisms on the surface.
After attachment step, the “kısır” samples were treated with 1 ml of pomegranate product and mixed well. The jars were left for 0, 5, 10 and 20 minutes at room temperature.

Then decimal dilutions were prepared and surface plated on TSAN (TSA which contains 50 ppm nalidixic acid. TSAN; TSA, Oxoid, CM013, Nalidixic acid, Fluka, 70162).

The plates were incubated at 37°C for 24-48 hours.

All the experiments were repeated three times with two parallels.
• The results of the inhibitory effect of the pomegranate products on the natural microflora of the “kısır” samples were shown in the next diagram.
• The results of the inhibitory effect of the pomegranate products on the “kısır” samples which were inoculated with *Staphylococcus aureus* and *Escherichia coli* O157:H7 were shown in the next two diagrams, respectively.
• The results showed that pomegranate products inhibited the growth of the microorganisms on both inoculated and natural microflora of the kıtır samples and it was found that inhibitory effect of all pomegranate products increased by application time.
• However, both traditional and commercial pomegranate products were effective on decreasing the numbers of the microorganisms, traditional pomegranate sour sauces were found to be more effective than the commercial pomegranate sauces, generally.
CONCLUSION

- These results indicate that pomegranate products used as flavoring and acidifying agent, also could prevent food-borne outbreaks related to fresh produce.
References

Thank you
## pH Values

<table>
<thead>
<tr>
<th>Products</th>
<th>pH</th>
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<tbody>
<tr>
<td>A</td>
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<tr>
<td>B</td>
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<tr>
<td>C</td>
<td>2.64</td>
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<tr>
<td>D</td>
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<td>E</td>
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<tr>
<td>H</td>
<td>1.94</td>
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<tr>
<td>J</td>
<td>1.76</td>
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</tbody>
</table>
Usual stock cultures were kept at 4°C on slants of Tryptone Soya Agar (TSA, pH 7.3±0.2, Oxoid, CM0131) and acid adapted stock cultures were kept at the same temperature on slants of TSA which contains 50 ppm nalidixic acid (TSAN; TSA, Oxoid, CM013, 50 ppm nalidixic acid, Fluka, 70162).

Cultures were grown in Tryptone Soya Broth (TSB, Oxoid, CM0129) or TSB supplemented with 50 ppm nalidixic acid (TSBN) and incubated at 37°C for 24 hours.

Cells were collected by centrifugation (5000 rpm, 30 min, 25°C), resuspended in 10 ml of sterile peptone water (0.1%), appropriate dilutions for inoculation were prepared from this suspension.