An Investigation on Antibacterial Effect of Garlic Aqueous Extract on Coliform of Uncooked Hamburger

Hosseini, E. and Shabani, Sh

1Department of Food and Science Technology, Faculty of Agriculture and Natural Resource, Science and Research Branch, Islamic Azad University, Tehran, Iran.
2Department of Food Science and Technology, Science and Research Branch, Islamic Azad University, Tehran, Iran.

Hosseini, E. and Shabani, Sh; An Investigation on Antibacterial Effect of Garlic Aqueous Extract on Coliform of Uncooked Hamburger

ABSTRACT

Coliform bacteria belong to antrobacteriacea family and are of fecal origin. They are also a hygiene indicator for food. Since it is not allowed to use any preservatives in hamburger, in the present research, the antibacterial effect of garlic aqueous extract was studied as a natural antibacterial agent. Aqueous garlic extract was added in 1, 2, 3 cc per 100 gr of hamburger. One group of sample was stored in 0-4°C for a period of 1 and 2 weeks and the other group was stored at -18°C for 1, 2 and 3 months. All the samples were examined in order to determine the most effective ratio of garlic aqueous extract on the growth of coliforms. Statistical results revealed, that in hamburgers containing 2 and 3 cc garlic aqueous extract, the growth of coliforms significantly decreased in treatments stored at refrigerator and freezer temperatures.

Key words: Garlic, Hamburger, Garlic aqueous extract, Coliforms, Antibacterial.

Introduction

The growth of cities and modern life style are associated with the popularity of convenience foods whether in preserved or processed forms or both. The development of convenience foods has resulted in the increasing use of products like hamburgers. Hamburgers are prepared using minced beef or mutton with or without fat and also spices e.g garlic and onion.

The mixture is formed in 100 gr pieces by special mechanical equipment and are frozen after packing in waxed papers.

It is almost certain that the worlds greatest problem is how we should provide enough healthy food for an ever-growing world population [10].

The growing consumption of red meat throughout the world is associated with the popularity of hamburgers as a most important red meat product.

This is because of high nutritional value, taste and easy preparation method and the fact that it is prodused without the use of additives. Microorganisms cause the spoilage of hamburger by growing during storage period and deteriorating its quality.

'Spoilage' develops off-flavor and decreases food acceptability. Although application of antibacterial agents with chemical origins are useful for maintaining quality, extending shelf-life and have lower processing costs and higher productivity but they are not permitted in hamburgers according to legislation.

Garlic is a common ingredient in hamburger manufacturing and imparts a characteristic taste and aroma, it is also a natural additive but no extensive research has been conducted regarding its pharmaceutical properties [9].

Garlic (Allium Sativum) is a bulb and belongs to Liliaceae family. Its close relatives include onion, shallot and leek.

With a history of human use of over 6000 years, garlic is native to central Asia. Garlic is easy to grow and can be grown year-round in mild climate.

There are two subspecies, Ophios corodon or hard-necked garlic and Allium sativum or soft-necked garlic. Garlic is widely used around the world for its pungent flavour as a seasoning in food and also as a medicine in many cultures.

According to remains of Sanskrit hand-writings, Indians were familiar with garlic sauce 5000 years ago and used it for many conditions including weight gaining, digestive disorder, coughs, poor digestion, common cold, tired ness, skin disorder, hemorrhoids, gastrilis, splenalgra, stomach...
pain, parasite, rheumatism, tuberculosis, leprosy, and epilepsy [1].

Hamburger may contain pathogenic microorganism e.g., Salmonellas, Staphylococcus aureus and Escherichia coli which cause typhoid fever, paratyphoid, digestive infection and food poisoning among consumers. They also decrease quality during storage.[3,7,11].

Coliform bacteria belong to antrobacteriacea family and are of fecal origin. They are in short rod shape, single or paired, sporeless, aerobic and anaerobic and mobile which are able to grow at 32-37°C. Lactose fermentation is the most important characteristic of Coliforms which is a basis for it’s distinction and computation. This group of bacterias play an important role in health and hygiene of food[7,11].

Alisin is the most important sulphide compound in garlic which is responsible for it’s antibacterial and anti-oxidant properties. With the formula of c6h100sz, it’s molecular weight is 162.28 g/mol and the metting point is less than 25°C. Alisin inhibit’s proteins by reacting with thiol group. Alisin also prevents synthetic reaction of acetyl co – enzyme i.e it is trapped in a reversible reaction. Enzymes with thiol group which undergo reaction with Alisin include succinic and dehydrogenase, urease, papayan, xanthan oxidase, colin oxidase, hexochinase, colin stearease, triose phosphate dehydrogenase, alcohol dehydrogenase and cysteine protease. It’s capability in penetration through cell membrane and also passive reactions in order to trap radicals and inhibiting enzymes are the most important biological characteristic of Alisin.

Alisin also reacts with those enzymes which are responsible for cell propagation and prevent cell regeneration. The effect of Alisin on RNA polymerase also retards and prevents synthesis of DNA, RNA and protein in microbial cells [2].

Chemical properties of garlic was studied in 1944 for the first time. Using different chemical analysis, Diallyl disulphide; the main component of garlic with sterilized distilled water. In the next stage, garlic was blended and crushed with 200 ml sodium chloride isotonic solution(0.9%) in presence of chopped ice next to flame for providing sterile condition. The mixture was sterilized with ordinary colander and stored at -20°C [4,14].

Results and Discussion

The effect of garlic aqueous solution(1,2 and 3ml) on the growth of Coliforms in hamburger in the refrigeration condition and in frozen condition for 1,2 and 3 months are shown in table 1 and table 2 and 3 respectively.

In figure 1, the effect of different concentrations of garlic aqueous extract on number of Coliforms during different storing times in refrigerator and frozen has been shown in figure 1.

Discussion:

Statistical analysis of first and second week indicate that samples containing 2 and 3 ml garlic aqueous extract decrease Coliforms growth in refrigeration condition significantly (p<0.5). Meantime the effect of 3 ml extract is more than the other samples but is not different from 2 ml significantly. Blank shows no significant difference from 1 ml extract and both have no effect on coliform growth . The results also show that the number of bacterias grown in the first and second week are significantly different so that the number of Coliforms in the first week are significantly less than the second week and are the most in the blank treatment.
Table 1: Number of Coliforms in the samples of first and second week. (Effect of concentration).

<table>
<thead>
<tr>
<th>TREATMENTS</th>
<th>BLANK</th>
<th>1st</th>
<th>2nd</th>
<th>3rd</th>
<th>STANDARD</th>
</tr>
</thead>
<tbody>
<tr>
<td>No.of Coliforms Day(Zero)</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>13</td>
<td>Non fecal Coliforms in small amount is acceptable</td>
</tr>
<tr>
<td>No. Of Coliforms (dilution 10^-6) at first week of storage at 0-4°C</td>
<td>a 10.00±7.00</td>
<td>a 3.00±3.46</td>
<td>b 25.66±38.42</td>
<td>b 5.00±5.29</td>
<td>Non fecal Coliforms in small amount is acceptable</td>
</tr>
<tr>
<td>No.of Coliforms(dilution 10^-6) at second week of storage at 0-4°C</td>
<td>a 15.66±3.78</td>
<td>a 26.33±7.02</td>
<td>b 53.00±31.19</td>
<td>b 84.66±9.07</td>
<td>Non fecal Coliforms in small amount is acceptable</td>
</tr>
</tbody>
</table>

1- Figure are in terms of mean ± standard deviation.
2- Superscripts in the same column indicate significant difference (p>0.05)
3- Number of bacteria are on the basis of cfu/g.
4- Regarding pour plate method, for actual number of coloni, the figure should be multiplied by 1×10^6
5- For significant differences in concentration, the figures should be compared horizontally.

Table 2: Number of Coliforms in the samples of 1, 2 and 3 months. (Effect of concentration).

<table>
<thead>
<tr>
<th>TREATMENTS</th>
<th>BLANK</th>
<th>1st</th>
<th>2nd</th>
<th>3rd</th>
<th>STANDARD</th>
</tr>
</thead>
<tbody>
<tr>
<td>No.of Coliforms Day(Zero)</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>13</td>
<td>Non fecal Coliforms in small amount is acceptable</td>
</tr>
<tr>
<td>No. of Coliforms (dilution 10^-6) at first month of storage at -18°C</td>
<td>a 0.66±1.15</td>
<td>ab 6.33±2.51</td>
<td>c 44.00±25.63</td>
<td>bc 35.00±24.57</td>
<td>Non fecal Coliforms in small amount is acceptable</td>
</tr>
<tr>
<td>No.of Coliforms(dilution 10^-6) at second month of storage at -18°C</td>
<td>a 0.0±0.0</td>
<td>ab 0.0±0.0</td>
<td>c 0.0±0.0</td>
<td>bc 0.0±0.0</td>
<td>Non fecal Coliforms in small amount is acceptable</td>
</tr>
<tr>
<td>No.of Coliforms(dilution 10^-6) at third month of storage at -18°C</td>
<td>a 0.0±0.0</td>
<td>ab 0.0±0.0</td>
<td>c 0.0±0.0</td>
<td>bc 0.0±0.0</td>
<td>Non fecal Coliforms in small amount is acceptable</td>
</tr>
</tbody>
</table>

1- Figure are in terms of mean ± standard deviation.
2- Superscripts in the same column indicate significant difference (p>0.05)
3- Number of bacteria are on the basis of cfu/g.
4- Regarding pour plate method, for actual number of coloni, the figure should be multiplied by 1×10^6
5- For significant differences in concentration, the figures should be compared horizontally.

Table 3: Number of Coliforms in the samples of 1, 2, 3 month. (The effect of time).

<table>
<thead>
<tr>
<th>TREATMENTS</th>
<th>Day zero</th>
<th>First month</th>
<th>Second month</th>
<th>Third month</th>
</tr>
</thead>
<tbody>
<tr>
<td>BLANK</td>
<td>13</td>
<td>b 35.00±24.57</td>
<td>a 0.0±0.0</td>
<td>a 0.0±0.0</td>
</tr>
<tr>
<td>1st</td>
<td>-</td>
<td>b 44.00±25.63</td>
<td>a 0.0±0.0</td>
<td>a 0.0±0.0</td>
</tr>
<tr>
<td>2nd</td>
<td>-</td>
<td>b 6.33±2.51</td>
<td>a 0.0±0.0</td>
<td>a 0.0±0.0</td>
</tr>
<tr>
<td>3rd</td>
<td>-</td>
<td>b 0.66±1.15</td>
<td>a 0.0±0.0</td>
<td>a 0.0±0.0</td>
</tr>
</tbody>
</table>

1- Figure are in terms of mean ± standard deviation.
2- Superscripts in the same column indicate significant difference (p>0.05)
3- Number of bacteria are on the basis of cfu/g.
4- Regarding pour plate method, for actual number of coloni, the figure should be multiplied by 1×10^6
5- For significant differences in concentration, the figures should be compared vertically.

Treatments containing 3 cc garlic aqueous extract shows no significant difference in the first and second week which is an indication of preventing effect of 3 cc treatment on Coliforms growth. In fact garlic aqueous extract in the concentration of 3 cc prevented the growth of Coliforms in the second week and this is an indication of the effect of garlic aqueous extract on the reduction of Coliform growth. Storing the samples for three months indicates that treatment containing 3 cc extract again is significantly different from other treatments regarding its effect on Coliform growth.

The effect of 2 cc extract was similar to 3 cc one and no significant difference was observed between them.

On the other hand, blank treatment as an independent group showed no difference with 2 cc
treatment. The preventing effect of 1 cc treatment was negligible and it had no effect on Coliforms growth. Statistical analysis revealed that there was a significant difference between first month with second and third ones but there was no difference between second and third month. In fact, they were similar.

Rees et al., in 1993 showed that garlic extract has two distinguish effects on useful intestine microflora and harmful bacteria like *E. coli*. It has no effect on useful bacteria but destroys harmful ones.

In an investigation on the effect of garlic extract on *E. coli* and *Lactobacillus casei* it was found that garlic extract is 10 times more effective on *E. coli*. However the reason is not clear but it is believed that this is probably because of difference in membrane nature and also the difference in permeability of membrane against Alisin [8].

![Fig. 1](image.png)

**Fig. 1:** The effect of different concentrations of garlic aqueous extract on number of coliforms during different storing time in refrigerator and frozen.

**References**