1. Introduction

Various herbs and spices are used to change flavor, aroma and color of food products. They usually have valuable nutritional properties. However, spices can be a source of pathogenic microorganisms that are transmitted via food. During growing, harvesting and postharvest handling, spices are exposed to contamination. Therefore, spices should be treated to reduce hazard. The process of spices by heat or steam is not applicable to all products. It may change their characteristic sensory or functional properties (Sade, Lassila, & Björkroth, 2016). Another method is application of radiation. But this treatment is quite expensive and there is resistance for public acceptance (Grabowski, Strzelczak, & Dąbrowski, 2014). These lead researchers to study alternative methods. In recent years, atmospheric cold plasmas have been raised as a novel decontamination technology (Misra et al., 2014). Non-thermal plasma is ionized gas which can inactivate pathogenic microorganisms existed on material surface. In competition with physical and chemical methods, plasma treatment is regarded as a high-efficiency technique for decontamination of products such as fresh fruit and vegetables, meat, and eggs (Schlüter et al., 2013). Low pressure cold plasma was tested for various nuts. Treatment using air gases for 5 min conducted 1-log reduction of A. parasiticus. 20 min plasma treatment resulted in a 50% decrease in total aflatoxins (Basaran, Basaran-Akgul, & Oksuz, 2008). Decontamination of packed sliced meat caused a reduction of L. innocua between 0.8 to 1.6 log cfu/g (Rød et al., 2012). The background microflora of strawberries treated for 5 min was decreased by 2 log within 24 h of post-cold plasma (Misra et al., 2014). Inactivation of aerobic microorganisms on blueberries was carried out with cold plasma. Reduction ranging of total count was from 0.8 to 1.6 log cfu/g compared to the control after 1 day but plasma effect was not significant for the numbers of yeast and molds (Lacombe et al., 2015).
The aim of this study was to evaluate feasibility of non-thermal plasma application for microbial decontamination of thyme and paprika.

2. Materials and methods
Spices were prepared from Golha Food Industry Complex (Tehran, Iran). Each sample was included 10 g spices distributed into a thin layer in a petri dish. Plasma production was carried out using dielectric barrier discharge method. Figure 1 shows the design of a non-thermal plasma generator system used in this study. The plasma generator includes a high voltage electrode, dielectric barrier and grounded electrode. High voltage electrode is made of aluminum rod 10 mm in diameter, insulated by a polytetrafluoroethylene cylinder to avoid electric shock. Dielectric barrier is a 1 mm thick quartz plate, which is located at the end of the high voltage electrode. Grounded electrode is a stainless mesh attached on the opposite side of quartz plate. The plasma produced near the grounded electrode and diffused away above the sample. The sample placed under the grounded electrode. The cold plasma is created by using a 12 kV pulsed high voltage power supply at approximately 6 KHz. Exposure time for plasma treatment was 5 minutes. The experiments carried out in duplicate and repeated twice. For total bacterial counting the culture medium was sterilized after autoclaving for 15 minutes. Using a micropipette, one milliliter of prepared dilution was poured in plate. Then, 20 to 15 ml of culture medium was added to it and samples were cultured by deep-culture technique and duplicate method. Plates were placed in a warm incubator at 72 ° C for 72 hours. Counting colonies began from the first day and the final countdown was made after the mentioned time. Plates selected for counting included the number of colonies between 30-300 cfu / g. For the mold and yeast count, the culture medium used was 18% glycerol agar. this was sterilized in an autoclave for 15 minutes. Then, using a micropipette, 0.1 ml of dilution was poured in the plate. 15-20 ml culture medium was added and surface culture was carried out by L-shaped glass tubing. Plates were incubated at 25 ° C for 5 to 7 days. The start of counting the colonies was from the first day.

![Figure 1. The design of the implemented system consists of: sample (1), ground electrode (2) and high voltage electrode (3)](image)

**Table 1. Microorganism contamination in treated and untreated spices**

<table>
<thead>
<tr>
<th>Spices</th>
<th>Total count (log cfu/g)</th>
<th>Mold and yeast (log cfu/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control</td>
<td>Treated</td>
</tr>
<tr>
<td>Thyme</td>
<td>2.48</td>
<td>1.3</td>
</tr>
<tr>
<td>Paprika</td>
<td>2.78</td>
<td>2.6</td>
</tr>
</tbody>
</table>

3. Results and Discussion
Table 1 shows effect of applied treatment on contamination of thyme and paprika. It seems bacterial contamination of thyme have been reduced by cold plasma. Other changes were not noticeable. Maybe it is due to the amount of initial contamination and type of microorganisms existed on the spice surface. Morphology of surface can also be effective.

Low pressure cold plasma was used for sterilizing black pepper. The number of microorganisms was decreased after 60 min treatment (Grabowski et al., 2014). The decontamination efficiency of the microwave-powered cold plasma was studied for red pepper powder. Under the vacuum conditions, plasma treatment for 20 min reduced total aerobic bacterial by approximately 1 log cfu/g. The cold plasma integrated
with heat treatment (90 °C, 30 min) was also decontaminated Bacillus cereus spores in a synergistic manner (Kim, Lee, & Min, 2014). The effect of remote plasma on microbial flora of pepper seeds, paprika powder and crushed oregano was investigated. The native microbial flora of pepper seeds and paprika was decreased by more than 3 log after 60 min treatment time. A considerable loss of redness was observed for paprika powder after a treatment time of ≥ 5 min (Hertwig et al., 2015). Plasma jet and microwave-driven remote plasma were applied for black pepper decontamination. Bacterial endospores (S. enterica, B. subtilis spores and B. atrophaeus) was inactivated by 4.1, 2.4 and 2.8 log, respectively, after 30 min remote plasma treatment. Equivalent inactivation levels were not obtained by direct plasma jet (Hertwig et al., 2015). In this research, treatment duration was 5 minute while review of studies on application of cold plasma for spice sterilization indicated that other researcher usually consider longer exposure time. It needs investigating to achieve effective decontamination of thyme and paprika through increase of plasma treatment time. Some studies also used vacuum conditions for non-thermal plasma but in current research, cold plasma was applied under atmospheric pressure which is inexpensive compared to low pressure preparation. Cold plasma can be considered as a useful technique to reduce the microbial load of thyme. Further research was required to replace this method with other technologies. It is also possible to combine non-thermal plasma with other technologies to eliminate some of their limitations.

References

Atmospheric cold plasma inactivation of aerobic microorganisms on blueberries and effects on quality attributes. Food Microbiology 46: 479–484.


