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Comparison of the Effect of Perchlorine, Sodium Hypochlorite, and Electrochemical Method on Disinfection of Vegetables

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ABSTRACT

***Introduction*:** Many vegetables are eaten raw which can lead to the prevalence of food borne diseases if not properly sanitized. The aim of this study was to compare the effect of chlorine, bleach and multioxidant solution in disinfection of edible vegetables.

***Materials and Methods*:** In this experimental study, 100 g of vegetables (lettuce, basil and mint) was disinfected by sodium hypochlorite, chlorine, and multioxidant solution at a concentration of 200 mg/L in 5, 10,15 and 20 min. Microbial load analysis was performed using SPSS-20 software with paired t-tests and ANOVA.

***Results*:** Coliforms reduction in mixed vegetables using chlorine, sodium hypochlorite and electrochemical solution were 1.07-1.47, 0.67-1.39 and 2.17-2.47, respectively. For lettuce, coliforms were reduced to 0.77-1.47, 0.57-1.33 and 2.17-2.43 log/100g. After disinfection of mint using chlorine, sodium hypochlorite and electrochemical solution, coliforms were reduced to 1.07-2.17, 0.95-1.77 and 2.47 log/100g and after Basil disinfection, coliforms were reduced to 0.87-1.87, 0.83-1.39 and 2.47 log/100g, respectively.

***Conclusion*:** With increasing concentrations of disinfectants dose, inactivation of bacteria was done in less time. Electrochemical method is the suitable option to disinfect vegetables at home and emergencies.

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Introduction

The need to consume fresh fruits and vegetables has increased in recent years, and the evidence obtained from various studies has confirmed that the consumption of healthy vegetables can prevent the development of heart disease and some types of cancers, especially gastrointestinal cancers, during the past few years. These foods may be consumed roughly or at least the baking process [1]. Statistics show that the consumption of freshly prepared vegetables is not the same in different countries, varying from 1.3-1.5 kg per person per year, and given that vegetables and fruits are exposed to contaminations from anywhere from the farm to the table, transmission of pathogens is possible. One of the most important factors in contamination of vegetables is the use of fertilizers and wastewater in agricultural land. Many pathogens are considered to be the natural flora of the soil and may be present on the products during harvest [2]. In this way, fresh vegetables and fruits are one of the most important risk factors for human health. Many epidemics are reported each year due to the consumption of vegetables and fresh fruits. Pathogenic agents which are often associated with these epidemics include bacteria (*Salmonella*, *Escherichia coli*, *Listeria monocytogenes*), viruses (neurovirus and *hepatitis A* virus), and parasites (*Cryptosporidium* and *Cyclospora*) [3].

Removing all potentially harmful microorganisms from vegetables is dependent on the health methods (washing and disinfection) use at home by different people [4]. The effectiveness of disinfection in reducing the microbial load of fruits and vegetables depends on the type of product, the type of fruit and vegetable, and also the type of microorganisms. Washing is a major step towards the removal of microorganisms or the reduction of microbial loads[5]. Studies have shown that the washing and disinfection phase reduces up to 1 logarithmic cycle of bacterial populations and 5.1 of the logarithmic cycle of mold and yeast, but in subsequent stages of washing and disinfection, the amount of microbial burden increases again due to secondary contamination[6]. Guidelines recommended by the Ministry of Health and Medical Education of Iran for sanitation of vegetables include washing with water, cleaning (3-5 drops of detergent per liter with a contact time of 5 min) and disinfection of vegetables using 200 mg/L perchlorine with a contact time of 5 min[7]. The detergent, in addition to removing the eggs of the parasites on the vegetables, reduces the hydrophobicity of leafy vegetables and, consequently, increases the contact between the pathogen and the disinfectant and increases its efficiency. Perchlorine can also be combined with organic ingredients in fruits and vegetables and produce toxic substances such as chloramine and trichloromethane, which is harmful to human health. The most commonly used disinfectants are calcium hypochlorite (perchlorine) and sodium hypochlorite (bleach). In different parts of the world, depending on the amount of contamination and the process of vegetable production, different methods are used for the disinfection of vegetables. In some countries, chlorine water is used to disinfect vegetables at various stages, including after harvesting, transportation and processing[8].

In Brazil, the most common method for having healthy and disinfected vegetables is to place vegetables in a solution of sodium hypochlorite at a concentration of 200 mg/L for15 min. The results of a study by Garcia-Gomez et al in Mexico to investigate common vegetable disinfection methods showed that more than 30% of the studied population had used a disinfectant solution with an effective silver component (colloidal silver) for disinfection of vegetables, while 28% of them had washed vegetables with only water and 25% had used sodium hypochlorite to disinfect the vegetables. The rest of the population had used methods such as salt, detergent, salt with lemon, detergent plus iodized solution, and lemon juice. In some countries, including the United States, it is recommended that fruits and vegetables be washed only with water and the use of detergent and any other commercial disinfection is not recommended [9]. Substances used for disinfection may affect the duration of the maintenance of vegetables because they are often chemicals. Investigation of the effect of peracetic acid and sodium hypochlorite on the storage after harvest of a number of fresh fruits and vegetables by Juan Eugenio Alvaro et al (2009) in Spain has been carried out. The results revealed that peracetic acid is more appropriate for washing fruits and does not affect the taste [6]. According to the above mentioned study, it can be stated that the application of traditional methods in making the vegetables and fruits healthy is ineffective on microbial contamination; nevertheless in cases where they have high efficacy, such as using ozone or radiation, its usage is dangerous or difficult for the general population or, in the case of chlorine, it affects the color and the quality of the fruit and vegetable [10]. In order to solve the existing problems, a disinfecting compound produced by electrolysis of salt for vegetables and fruits is used in this study, which, in addition to being simple and effective to use, does not damage the fruit and vegetable tissue. This study attempts to investigate the effect of bleach, perchlorine, multioxidant solution on the microbial quality of ready-to-eat vegetables, and selection of best the disinfectant that has the greatest effect on microbes and increase vegetables storage time.

Materials and Methods

This experimental study was performed on raw edible vegetables including lettuce, basil, and mint. According to field studies, each of the vegetables are studied individually because they have different levels or the effect of disinfect can be different on them. The vegetables were also disinfected in a mixture. In order to conduct this research, 100 g of vegetables were disinfected using different disinfectants: sodium hypochlorite (200 mg/L), perchlorine (200 mg/L), and multioxidant solution (200 mg/L). The disinfection time was 5, 10,15 and 20 min. After disinfection and washing, the microbial load of the specimens was determined. The properties of the commercial solution are such that the water containing 4 g of salt in 4 min is affected by the electrolysis process and the resulting solution is used as a disinfectant. Multioxidant solution contains chlorine, sodium hypochlorite, hypochloric acid, H_2O_2, and ozone. The microbial test was performed using multi-tube fermentation and pour plate method. Data were analyzed by SPSS-20 software using paired t-test and one way ANOVA.

Results

The average of the microbial load in different raw and disinfected samples with different compounds is shown in Figures 1-4. Figure 1 shows the logarithm of inactivating bacteria in mixed vegetables with perchlorine disinfection, sodium hypochlorite, and electrochemical methods. Reduction of coliforms during disinfection of mixed vegetables with perchlorine, sodium hypochlorite, and electrochemical solution was 1.07-7.1, 1.67-39, and 1.7 log/100g, respectively.

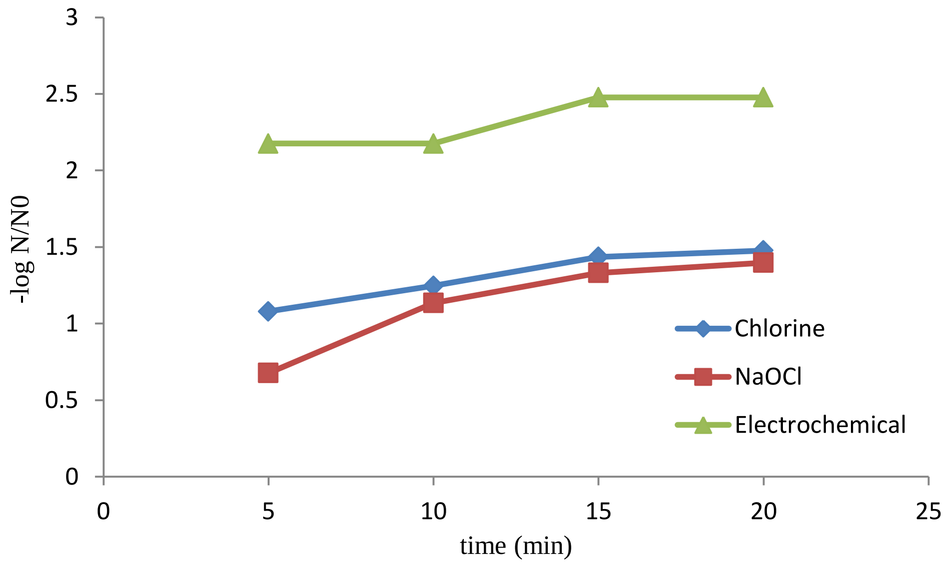

Figure 1: Logarithmic inactivation of bacteria by disinfection of mixed vegetables

In Figure 2, the logarithm of the inactivation of bacteria in lettuce is shown during disinfection with different disinfectants. Reductions of coliforms during lettuce disinfection by perchlorine, sodium hypochlorite, and electrochemical solution were 0.77- 1.77, 0.17- 0.33 and 43.1-4.2-17 log/100g, respectively.

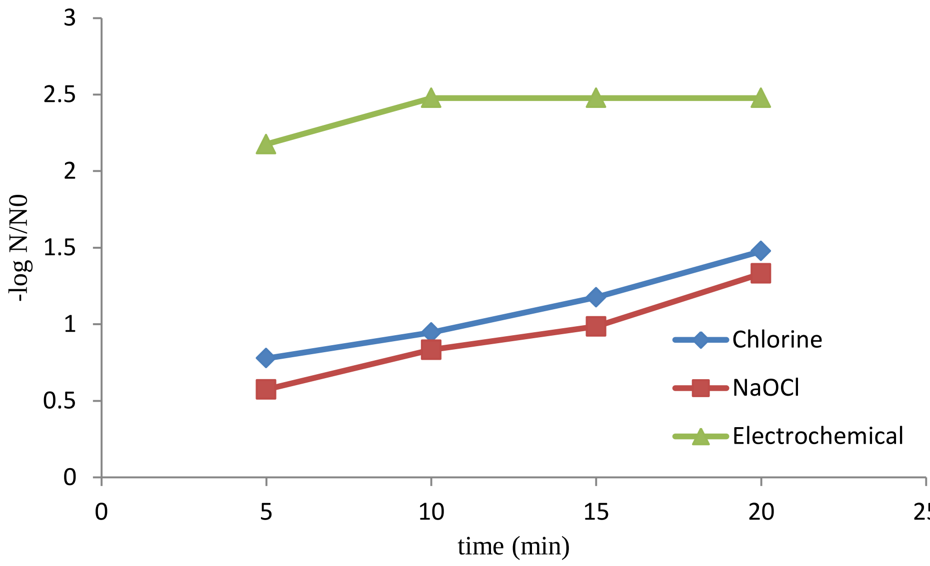

Figure 2: Inactivation of bacteria with lettuce disinfection

In Figure 3, the logarithm of inactivating bacteria in mint is shown during disinfection with different disinfectants. Reduction of chlorophylls during mint disinfection by perchlorine, sodium hypochlorite and electrochemical solution was 1.07-17.7, 0.71- 0.75, and 2.47 log/100 g, respectively.

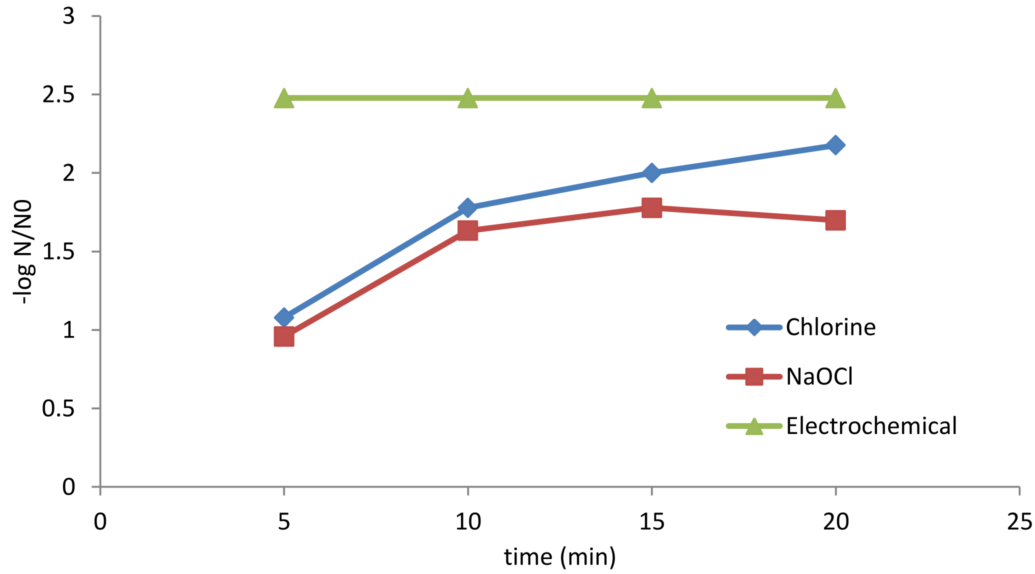

Figure 3: Logarithmic inactivation of bacteria in mint disinfection

In Figure 4, the logarithm of inactivating bacteria in basil is shown during disinfection with different disinfectants. Reduction of chlorophylls during basil disinfection with perchlorine, sodium hypochlorite and electrochemical solution was 0.87-1.87, 0.83-39.8 and 47.2 log/100 g, respectively.

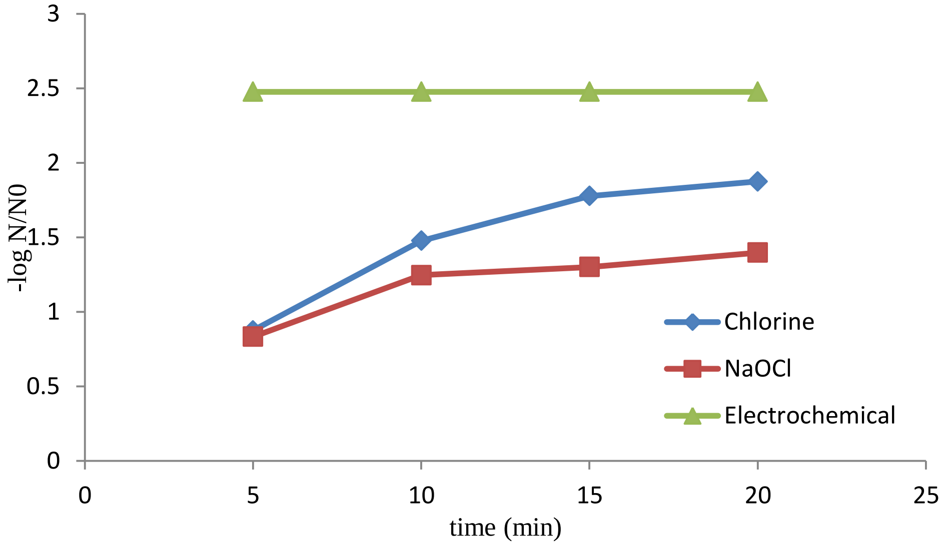

Figure 4: Logarithmic inactivation of bacteria with Basil disinfection

Discussion

In the Middle East, many vegetables are eaten raw because people have found that it has a most nutritional value. This lead to the prevalence of food borne diseases (FBD). In the United States, leafy vegetables are classified by the Food and Drug Administration (FDA) as one of the top ten foods that are at high risk because, according to the Center for Disease Prevention and Control (CDC), about 40% is involved in the spread of food borne diseases [11].

In vegetable farms, animal manure and wastewater are used as fertilizer. Therefore, the big challenge for vegetables and fruits is microbial contamination and parasitic infections. Typically, there are various pathogens of bacteria, viruses, fungi, protozoa, eggs of parasites, etc., on these vegetables which cannot be removed by simple washing [12]. The use of detergents in cleaning fruits and vegetables is one of the most important ways of preventing the transmission of diseases. Chlorine and its compounds are the most common types of disinfectants. Nevertheless, there is a potential for the formation of byproducts such as trihalomethanes, which requires further research.

Planting conditions, water quality and type of fertilizer used in agriculture, transportation conditions and preparation of vegetables in different countries are different and cause changes in the amount of initial microbial load in the product. Hence, HACCP principals should be considered from farm to plate [13].

In this study, after sampling vegetables and transferring them to the laboratory, the average coliform content of raw vegetables was 300 per 100 g while after 5, 10, 15 and 20 min disinfection with perchlorine, average coliform content was reduced to 25, 17, 11 and 10. Using sodium hypochlorite, the rate was reduced to 63, 22, 14 and 12, and with the use of electrochemical solution, the rate was reduced to 2, 2, 1, and 1, respectively. The average load of coliform content in lettuce was reduced by 50, 34, 20 and 10, respectively, after 5, 10, 15 and 20 min, through disinfection with perchlorine. Using sodium hypochlorite, the rate was reduced to 80, 44, 31 and 14 and, in the case of an electrochemical solution, it was reduced to 2, 1, 1, and 1, respectively.

The efficacy of lettuce disinfection based on the conventional method has been carried out by Yarahmadi et al. (2011) in Iran. The results showed that the guidelines of the Ministry of Health as an effective way to improve the health of vegetables could eliminate 98.3% of total coliform and 100% of fecal coliforms on lettuce [14].

The mean coliform content in mint was reduced after 5, 10, 15 and 20 min after disinfection by perchlorine to 25, 5, 3 and 2, respectively. Sodium hypochlorite reduced the coliform content to 33, 7, 5 and 6, while the electrochemical solution reduced it to 1. The mean coliform content of basil after disinfection with perchlorine reduced after 40, 10, 15 and 20 min, to 40, 10, 5 and 4, respectively. Sodium hypochlorite reduced the coliform content to 44, 17, 15 and 12, while the electrochemical solution reduced it to 1. Based on paired t-test, there was a significant difference in the microbial load of vegetables before and after disinfection (p-value < 0.05). The electrolysis solution of edible salt contains multioxidants, including sodium hypochlorite, chlorine dioxide and ozone, which makes it more powerful than other disinfectants. Disinfection of fruits and vegetables with chlorine to inactivate the *Hepatitis A* virus and MS_2 Phage was carried out by Michael J. Casteel et al (2008) in California and the results showed that if the dose of chlorine was initially increased to 200 ppm, the *Hepatitis A* virus inactivation and MSI *E. coli* phage would be more intense and faster [15]. Disinfection of selected vegetables with perchlorine, ozone, ultrasound, and citric acid was performed by Daniela Bermúdez et al (2012) in Washington. Ozone can inactivate bacteria on tomatoes after 3 min. Ozone was effective on green leafy lettuce and finally it was concluded that the concentration of disinfection, intensity of radiation and contact time are important factors for disinfection and removal of microorganisms [16].

An assessment of chlorine, benzalkonium chloride, and lactic acid as a cleanser has been conducted for reducing *Yersinia* and *E.Coli* O157: H7 on fresh vegetables by Velázquez et al (2008) in Argentina.

The results showed that after the disinfection, no pathogens were found in fresh vegetables [17]. One-way ANOVA analysis showed significant differences between disinfection of vegetables by 3 types of disinfectants (p-value < 0.05).

Conclusion

The electrolysis solution of edible salt contains multioxidants including sodium hypochlorite, chlorine dioxide and ozone, which are more efficient than other disinfectants. By increasing the concentration of disinfectants, the bacteria were inactivated in less time. On site production of multioxidant solution using salt electrolysis is a good alternative for disinfection of vegetables at home, disasters and emergency conditions.

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Conflict of interest

The authors declare that there is no conflict of interest.

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