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Evaluation of Sensorial, Chemical, and Microbial Characteristics of Pickled Cucumber Supplied in Shiraz

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ABSTRACT

Background: Cucumber is one of the vegetables that are widely preserved by fermentation in brine. Vegetables such as cucumber are normally not washed in the commercial centers and this causes microbial growth during fermentation. At the beginning of the fermentation process, lactic acid bacteria, depending on the environmental conditions, begin to grow. The aim of this study was to compare the microbial, chemical, and sensorial characteristics of canned and bulk pickles. **Methods**: In this study, a random sampling was implemented from the pickles of three locations in Shiraz city as suppliers of pickles. The sensorial evaluation was also conducted by 60 members of the hospital staff. Data was collected through questionnaires. Sensorial evaluation was performed using a hedonic scale of nine points. For microbial assessment, Iran National Standards 2326 was used. **Results**: The results of the microbiological tests showed that bulk pickles were infected with mesophilic aerobic bacteria at 26.6%, aerobic thermophilic bacteria at 6.6%, and 43.3% by mold and yeast; while canned pickles (with license the Ministry of Health) were infected 13.3% by mesophilic aerobic bacteria. The average pH of the bulk samples based on the national standard was more than the maximum allowed level. The results of the sensorial evaluation of taste, texture, color, and overall acceptability showed that canned pickles had the most overall acceptance and texture acceptance points and bulk pickles had the most points of flavor and color acceptance. **Conclusions**: According to the findings, it seems that the use of herbs in bulk pickles is the main cause of increase in the microbial load and rated the taste, because fragrant fresh vegetables have a greater impact on the microbial load than dried vegetables.

Keywords: *Pickled cucumber; Microbiological; Sensorial, Chemical; Characteristic*

Introduction

Food microbiology is an applied science and the main task of food microbiologists is to increase consumer confidence in the produced food. Achieving this point requires the creation

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and systematic application of microbial ecology of food and food production and the effect of the process on the practical problems. In the past, the only way to keep cucumbers properly was lactic fermentation and storage in saltwater. But then techniques have emerged in which the fermentation techniques do not need fermentation for stabilization of the product. In this regard the earlier technique was based on direct acid adding to the product, using vinegar or acetic acid and then pasteurization, while now adding the acid with the conventional cold storage of the product has been growing. Today, in the United States, where more than half a million tons of cucumber are canned every year, only about 40% is produced by fermentation and the same amount is stored by pasteurization and the remaining is stored through cooling (Mortazavi and Mahounak, 2002).

When the vegetables are placed in the appropriate concentration of sodium chloride solution, naturally fermentation occurs by microorganisms present in the vegetables. Sodium chloride used is about 1–8%. The type and number of microorganisms in the fermentation process are greatly influenced by the concentration of sodium chloride. The integrity of the structure during the fermentation of vegetables is very important. Hence, sodium chloride is essential to prevent softening of the vegetables. In addition, pickled products have been affected by gas corruption and are known as Cucumber Hollow due to the accumulation of CO2 in saltwater during the fermentation procedure (Daeschel and Fleming, 1984). It has been reported that cucumbers should be kept at a temperature of 7.2–10 °C and should be transferred as soon as possible, particularly if they are to be shipped to far locations. If the vegetables are washed with water a few days before the process, their storage time is reduced by half. For this reason, unwashed fruit, stored at 4–5 °C for six days has acceptable quality, while the period is less than four days for washed fruits (Akbudak *et al.*, 2007). Texture is one of the most important qualitative characteristics of pickles and its effect on product acceptance is crucial. In the event that the process of pickle production is carried out under optimal conditions, some changes occur in the texture of primary products which can affect the quality pickle (Rodrigo and Alvarruiz, 1988).

Materials and Methods

Sampling and determining the sample size: From the list of pickle supplementation centers in Shiraz city, three areas were randomly selected for sampling and samples of canned pickled cucumbers were obtained from the selected areas. A total of 60 samples including 30 bulk samples and 30 canned samples were selected in accordance with national standard 2836 (Institute of Standards and Industrial Research of Iran, 1994). Sampling was done for 15 days.

Sensorial analysis: Sixty personnel of Shiraz Ordibehesht hospital were selected for sensorial evaluation of pickled cucumber samples. It should be noted that evaluators were selected based on their previous experiences on the sensorial analysis and taste sensitivity. Samples of pickles were given to the evaluators with the specific codes. The Five–point Hedonic scale (for texture, color, taste and overall acceptability as: Like very much = 9, like moderately = 7, neither like nor dislike = 5, dislike moderately = 3 and dislike very much = 1) was considered (Rajablou *et al.*, 2012).

Bacterial and chemical analysis: In order to evaluate the samples biologically, chemically, and for maximum grading of errors, the national standard was used for pickle samples (Institute of Standards and Industrial Research of Iran, 2007, 2009).

Data analysis: Considering that the test conditions were nonparametric, the means were compared using Kruskal–Wallis post-hoc test at the significance level of 0.05. SPSS version 16 was employed to examine the information and analyze the data.

Results

In this study, in terms of the grading factor and the maximum degree of quality defects, the canned

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pickles with regular consultations were used (Institute of Standards and Industrial Research of Iran, 2007). The canned pickled cucumber is those that have production license from the Ministry of Health and the bulk pickled cucumber is those that is produced traditionally.

Hedonic's scales are used to assist the evaluator in answering questions about the overall acceptability and the acceptance of one or more of that commodity. It is necessary to mention that Hedonic's scales are often stratum. This indicates that the respondent can only choose one of the available degrees or categories and there is no choice between two consecutive stratums. Consequently, discrete data is created by this scale. However, on a linear scale, each person can choose any numeral between two degrees of scale. Therefore, the data created for analysis is of continuous type. Average scores of accepting the taste, texture, color, and overall acceptability of pickles have been presented in **Table 1**. There was a significant difference between the bulk and canned samples; while the canned samples had the highest score of texture and overall acceptance, the bulk samples obtained the highest scores of taste and color ($P < 0.05$). The average chemical and biological characteristics of the samples have been presented in **Table 2**.

Table 1. Average rating acceptance of the taste, texture, color, and overall acceptability of pickles

Kind of pickles cucumber	Taste	Color	Texture	Over all acceptability
Bulk pickled cucumbers	8.41	7.61	6.72	5.2
Canned pickled cucumbers	6.52	5.34	7	7.44
P-value	0.005	0.037	0.029	0.001

Table 2. Microbial and chemical characteristics of pickle samples

Variables	Bulk pickled cucumbers	Canned pickled cucumbers
Average pH	4.78	4.16
Average acidity	0.83	0.91
Average % salt	4.57	2.39
Average % Mesophilic aerobic bacteria	26.60	13.30
Average % Thermophilic aerobic bacteria	6.60	-
Average % Mold and Yeast	43.30	-

Discussion

As much as 40% of the pickles in the world are only used as processed cucumbers (pickles). In the past, cucumbers were fermented in wooden containers or barrels. Usually the containers were filled with pickles and brine and a salt concentration of 8.5% was added to it. But today, pickles are produced in the pasteurized and fermented form and then consumed. However, fermented pickles are divided into two categories of salted and basil. Canned pickles are produced industrially in the food factories and then pasteurized. In the non-fermented process of pickles, microorganisms are inactivated due to the pasteurization; therefore, the pickles should be free from the growth of pathogenic microbes, but pasteurization is not conducted in the pickle fermentation process.

Usually cucumbers immersed in the acidic saltwater are heated in such a way that the inside of the cucumber remains at a temperature of 73.9 °C at least for 15 minutes. Heating and cooling must be done quickly. Pickle fermentation is a complex, variable, and unpredictable process. In general, *Lactobacillus plantarum* is the main acid producing bacteria. But

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Leuconostoc mesenteroides, *Lactobacillus brevis*, *Streptococcus faecalis*, *Pediococcus cerevisiae*, and coliform bacteria are involved in the production of acid. In traditional methods, fermentation time is six–nine weeks, according to the salting method and temperature used. First all salt–resistant species are able to grow in brine cucumbers. Therefore, the types of bacteria which grow in tanks of saltwater is very different and depends on the factors such as the number and types of bacteria remaining in the cucumber or the water used for the preparation of brine, the initial concentration of salt, and the rate of increase to the desired concentration of the fresh and saltwater temperature. It is noteworthy that in most cases the amount of salt in the final product reduces from 6.4% to 4.2% (Abou-Zaid, 2015) Generally, the lower the salt concentration, the more the bacteria will grow and the faster the acid production and more total acidity. In most cases, the mixture of species of the *Pseudomonas* and *Flavobacterium* grow and lead to the product corruption rather than acid production. Bacillus species enter the saltwater through the soil sticking to the cucumbers and their growth is undesirable. The *Lactobacillus plantarum* bacteria increase the acidity in water in high concentrations and at low concentrations of the salt (Mortazavi and Mahounak, 2002). *Lactobacillus plantarum* is known as the desirable lactic bacteria for commercial fermentation of the cucumbers because it is homofermentative and lack of carbon dioxide production from hexose as well as the availability of commercial cultivation for many years (Di Cagno *et al*., 2008).

At 5.3% salt concentration, the growth of lactic acid bacteria increases in comparison with the other microorganisms and overcomes them; while with lower concentrations, infectious bacteria of pickles grow rapidly and corrupt the product . High concentrations of salt (up to 7%) cause decrease in the number of Lactobacillus and at the same time lead to the rapid growth of yeasts resistant to the high concentrations of salt.

The yeast consumes lactic acid produced by Lactobacillus and reduces the amount of acid needed for proper fermentation of the product and pickles and results in the corruption of pickles . At high concentrations of saltwater, the water is removed and the product becomes wrinkled and hollow. Sometimes, due to accumulation of gas inside hollow cucumbers, pickles produce gas and float on the surface. Also, if the salt concentration is low, the product gets slimy, soft, and holds a lot of water. Therefore, the average concentration of salt (5.3%) is desirable (Rajablou *et al.*, 2012).

Changes developed during the corruption of fermented pickles are related to the metabolism of bacteria and yeasts. The ability of *Pichia manshurica* and *Issatchenkia* occidental yeasts in the use of lactic and acetic acids during aerobic metabolism for increasing the pH and chemical reduction of the matrix materials has been confirmed. However, new evidence suggests that *Lactobacillus buchneri* may play an important role in the corruption caused by secondary fermentation of cucumber. Lactic acid degradation during the corruption is influenced by the sodium chloride concentration, pH, and oxygen. *Lactobacillus buchneri* causes lactic acid corruption in the pH = 3.8 and w/w concentration of salt at 6%. Oxygen also has no effect on lactate metabolism by *Lactobacillus buchneri* and consumption of lactic acid is associated with increase in the acetic acid and 1,2–propanediol. *Lactobacillus rapi* is also able to convert 1,2–propanediol to propionic acid and propanol. *Lactobacillus buchneri* is the starting of the corruption under the environmental conditions and *Lactobacillus rapi* may contribute to the production of corrupted metabolites (Johanningsmeier and McFeeters, 2013). The most consumption of lactic acid by *Lactobacillus buchneri* happens at a concentration of 2% NaCl. *Lactobacillus buchneri* causes the initial corruption in a wide range of environmental conditions and *Lactobacillus rapi* may play a syntrophical role in the production of the metabolites related to corruption by *Lactobacillus buchneri* (Di Cagno *et*

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al., 2008). *Lactobacillus buchneri* leads to the corruption of fermented cucumbers through metabolization of the lactic acid, acetic acid, and 1,2–propanediol. However, there is very little information on the unique metabolic ability *Lactobacillus buchneri* and other chemical compounds linked to corruption. *Lactobacillus buchneri* and *Pediococcus ethanolidurans* are able to convert sugars into lactic acid, but under aerobic and anaerobic conditions and initial acid pH, only *Lactobacillus buchneri* is able to use acetic acid in the medium. The formation of secondary products of metabolism of *Clostridium bifermentans* and *Enterobacter cloaceae* is related to a medium with a pH of more than 5.7 In fact, the chemical changes of the first stage of corruption are associated with the oxidative yeasts and *Lactobacillus buchneri* that are able to prevent secondary fermentation (Franco and Pérez-Díaz, 2012). Ninety-two metabolites related to the corruption of fermented pickles have been identified using gas chromatography. During corruption the amounts of mono–disaccharides, amino acids, nucleosides, long–chain fatty acids, aldehydes, and ketones are reduced and the amount of a number of alcohols, botanic acids, and pantenoic acids increases. Most of the metabolic changes induced by lactic acid show that the lactic acid is not the preferred substrate for anaerobic microorganisms during corruption of fermented cucumbers. It is known that citrulline, trehalose and cellobiose may increase activity of *Lactobacillus buchneri* metabolic activity more than other compounds (Johanningsmeier and McFeeters, 2015). In addition to the changes made by the fermentation of microorganisms, consumable cucumbers' quality including cucumbers storage time before the process has a great impact on the quality characteristics of the final product. It is reported that the shelf–life of cucumbers for pickles production under normal atmospheric and 90–95% relative humidity at a temperature of 7 °C is less than 10 days, but using controlled atmosphere of reduced oxygen and increased carbon dioxide levels can increase the shelf–life up to 30 days. The best shelf–life of cucumbers for pickles production is equal to 20 days (Akbudak *et al.*, 2007). Rajablou and colleagues have reported that probiotic pickles containing Lactobacillus obtained less sensorial scores compared to the non-probiotic pickles. Pickles containing vinegar also had more aroma sensory scores, but pickles with no vinegar had more sensorial scores in terms of the tenderness and this means that vinegar pickles can cause tissue tenderness. In general, the overall acceptability of the samples containing *Lactobacillus plantarum* without vinegar is more than the other samples (Rajablou *et al.*, 2012). Internal texture of cucumbers is opaque and light green at the beginning but when they are fermented their color is changed from the light green to olive green or yellowish green and their internal structure is completely transparent. The results of the characteristics of flavor and texture are not consistent with the findings of Rajablou and colleagues because the average pH of bulk samples in this study was higher than canned pickles; however, due to the higher sensorial scores of taste for bulk pickles, vinegar plays a limited role in sensorial scores of pickles and it seems that the use of herbs has had a more active role in relation to the rating of flavor. Also Rajablou and colleagues have reported that vinegar increases the tenderness of pickles, but given that the pH of canned pickles is lower than the bulk samples and the sensorial score of tissue (stiffness) in canned pickles was higher, it suggests that vinegar has no effect on tissue tenderness. Pickles which are prepared using heterofermentative bacteria are harder and denser than those which are produced by homofermentative bacteria (Mortazavi and Mahounak, 2002). Considering that *Lactobacillus plantarum* is one of the homofermentative Lactobacillus with the optimum temperature less than 37 °C and *Lactobacillus buchneri* is a heterofermentative Lactobacillus, *Lactobacillus buchneri* plays an important role compared with *Lactobacillus plantarum* in firmness and increasing the sensorial scores of tissue. Reduction of the number of *Lactobacillus*

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plantarum during shelf–life of fermented foods has been reported by some researchers. Yoon and colleagues reported that during the two weeks' storage at refrigerator temperature, *Lactobacillus plantarum* in fermented cabbage juice reduced from 10^8 cfu/ml to 10^7 cfu/ml and fermented tomato juice slowly decreased over four weeks to 10^6 cfu/ml (Yoon *et al.*, 2004, 2006). McDonald and colleagues also investigated the population of microorganisms in pickles and have reported that the number of lactic bacteria decreased within five days slowly (McDonald *et al.*, 1991). According to the findings and results, it seems that among the lactic bacteria, *Lactobacillus buchneri* reduces less than *Lactobacillus plantarum* during shelf–life. In this study and based on the CODEX STAN 115, the amount of consumed salt and acidity of canned and bulk (traditional) pickles were within the allowed limit (CODEX STAN 115, 1981) and, according to microbiological test results (**Table 2**), it appears that microbial contamination of the bulk pickles is due to secondary pollution during production and packaging. As well as different formulations, insufficient thermal processing and inappropriate packing are the causes of microbial load of canned pickles with mesophilic aerobic bacteria. In general, it can be said that the use of herbs in bulk pickles is the main factor in the increase in the microbial load and taste score because aromatic fresh vegetables, as compared to dried vegetables, due to the water activity and high humidity, have the greater impact on the increase in the microbial load and taste. Also according to the national standard, maximum use of herbs is 5%, but it is much more in the pickles and given that the number of tail and flower remaining on cucumbers in bulk pickles is higher than canned pickles, it can be an acceptable reason for the increased flavor and microbial load. Additionally the use of vinegar in pickles in the optimal ratio cannot have a negative effect on the tissue. Although pickles can cause increased appetite and have a special place in the household, but because they contain salt, they increase the risk of high blood pressure, which is one of the most important risk factors and diet can play a role in preventing it. Patients, who have low–salt diet for a long time, often do not eat much of their food, resulting in weight loss, health problems, and, in some cases, show symptoms of malnutrition. The fermented cucumbers without salt can be prepared using calcium chloride, potassium chloride, or acetic acid and citric acid separately. Of course, the formulation has a great impact on acceptable sensory characteristics and studies show that the use of 7% KCl, 2.5% citric acid, and 5% acetic acid causes a bitter or sour taste that can be improved through submergence in water for 24 hours before consumption or the addition of natural flavors (garlic or paprika) (Johanningsmeier and McFeeters, 2013). Fermented cucumbers need greater desalination in a solution than calcium chloride, and vice versa in sodium chloride cucumbers fermented in a sodium chloride solution step need one step desalination and it is preferred (Wilson *et al.*, 2015). Polygalacturonase causes softening of the pickles gradually. Polygalacturonic acid hydrolysis is recognized according to the fluidity and reduction of the endo– and exo–oligogalactronides groups. Heat treatment of mesocarp tissue in the pasteurized pickles causes concentration of extract rapidly and changes in the pectic materials. Polygalacturonase remaining in cucumber is responsible for the softening process and it seems that depending on the type and amount of polygalacturonase remnant, tissue changes occur (Cho and Buescher, 2012).

Conclusions

The results showed that although the method of preparation of bulk pickles has better flavor and color for consumers, due to higher bacterial load in production, it is not recommended. Also, given that removing the pickles from the shopping cart seems difficult and its salt increases blood pressure, suitable salt substitutes should be used for the production of these products. Therefore, calcium chloride and potassium chloride can be used to produce canned pickles with desirable sensory characteristics instead of sodium chloride.

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Authors' contributions

Sadeghizadeh Yazdi J designed research; Behradkia V conducted research; Sarhadi H analyzed data; Hozoori M and Sadeghizadeh Yazdi J wrote the paper. Hozoori M had primary responsibility for the final content. All authors read and approved the final manuscript.

Conflicts of interest

The authors declare no conflicts of interest.

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