Kimchi: Determining the rapidity of acidification depending on temperature

Daniel Park¹, Bobby Sidhu², Fred Shaw³

Lead Author, School of Health Sciences, British Columbia Institute of Technology, Burnaby, BC
Supervisor, School of Health Sciences, British Columbia Institute of Technology, Burnaby, BC
Contributor, School of Health Sciences, British Columbia Institute of Technology, Burnaby, BC

Abstract

Background: The increasing number of kimchi consumers in Metro Vancouver raises food safety concerns over the kimchi being out in the ambient temperature. Although kimchi is known to have lactic acid producing bacteria as its normal flora, environmental health officers have no specific reference to the change in pH with respect to time. The purpose of this study was to understand the rate at which kimchi ferments at different temperatures and determine whether kimchi is a hazardous food or not.

Methods: Freshly made kimchi at researcher's residence were divided into two groups; 4 °C and 25 °C. 30 samples for each set with equal amounts were left at these two different conditions. PH and temperature were measured at the time of separation and for three weeks weekly using the Waterproof Palm pH analyzer. **Results:** There was a steeper decline in the 25 °C set compared to 4 °C. It took some time between 22 hours and 34 hours for 25 °C set to show a drop in pH. On the other hand, 4 °C set did not show a significant decline in pH within the time period of the experiment.

Keywords: Kimchi, pH, Waterproof Palm pH analyzer, fermentation, acidification

Introduction – What is Kimchi?

Kimchi is a culturally-defining Korean dish that involves the fermentation process to produce acidic vegetables with a variety of spices. Koreans, on average, consume 40 pounds of kimchi per person annually (Magnier, 2003). The major ingredients include cabbage and/or radish, salt, powdered red pepper, onions, sugar, salted seafood, grounded garlic and various fruits and vegetables. Kimchi is fermented by naturally occurring bacteria of genera Lactobacillus and Leuconostoc (Lee, Chun, Hector, et al. 1997). For the fermentation process, it is widely accepted in Korea to dig a large hole in the ground and plant crocks full of kimchi for fermentation until the taste matches one's preference. It is covered by the matching lid heavy enough to limit any local wild animal from disturbing it.

Although this is a common practise in Korea due to historical and cultural practices, it raises questions with respect to food safety. A major safety concern in the kimchi industry is the process of fermentation. Contributing factors affecting the fermentation are pH, temperature, starting micro flora, ingredients, and methods of producing industrial kimchi.

Environmental Health Officers (EHOs), during their inspections, are not aware of the possible risk fermented kimchi poses to the public. Kimchi is a food product that is consumed in many restaurants in Canada. As there are different food safety risk factors around kimchi, pH tests and microbial numeration techniques have been chosen to analyze the association of fermentation to change in pH in two different temperature scenarios. Questions about public health concerns regarding kimchi plays a significant role in carrying out this study.

Kimchi, as explained, is a fermented food product that may be considered as a potentially hazardous food. The fermentation process of kimchi occur at different temperatures and the concerns arise when the process occurs at temperature that is considered to be in the danger zone, which is between 4 and 60 degrees Celsius (PHAC. 2012). The process of making kimchi involves manual handling and these activities can introduce harmful microorganisms into the food. Cross contamination of unwanted microbes should be prevented through proper training, a satisfactory level of personal hygiene and a sanitized food preparation area.

Concerns regarding the procedure of making Kimchi are: production is primarily done by workers who acquired recipes that run down their families, and every person making kimchi has their own unique way of preparing and fermenting it. In addition, most of the preparation steps are done on the ground due to the inconvenience of carrying heavy materials. Before even considering the microbial matters, environmental health officers must educate producers with respect to safe food preparation techniques.

Microbial health implications must be considered once physical component is dealt with. As stated above, fermentation of kimchi is done in variety of ways, each depending on consumer's taste. Some prefer to ferment fast at room temperature and some prefer to put it in a specialized kimchi refrigerator to have slower fermentation, which affects taste. In all methods, however, kimchi is still prone to harbour microorganisms. Recall that naturally occurring bacteria in kimchi called Leuconostoc mesenteroides produces lactic acid to reduce the pH as the fermentation progresses down to 4.2 or less (Lee, 2009). Keeping this in mind, the question now becomes: How long does it take to bring the pH down to an acceptable level of 4.6 or below at two different temperature to be not considered a potentially hazardous food?

Literature Review

Controlled fermentation of kimchi using naturally occurring antimicrobial agents

The research entails the study of micro flora forming in kimchi during fermentation and its effect when over-acidification occurs during distribution and storage. The over-acidification extends the shelf-life of kimchi; however, as it progresses, decrease in quality is a major concern in the industry. An investigation was conducted to find ways to delay the over-acidification by adding several ingredients (herbs, tea leaves, fruits and vegetable) that do not belong to kimchi's normal recipe (Kim, Bang, et al. 2012). The addition of naturally occurring antimicrobial agents (herbs, leaves, fruits and vegetables) to inhibit lactic acid forming bacteria is a viable method to slow down acidification process but it also has a disadvantage in that it may have a negative effect in smell and taste. Hurdle technologies are applied which involve using combinations of low concentrations of different antimicrobial agents to control undesirable sensory changes. At the end, they found no significant influence on smell and taste changes in kimchi after applying the ingredients and antimicrobial agents (Kim, Bang, et al. 2012).

The focus of Kim's research was to develop an appropriate hurdle technology using the results. The relevance of this study to the future study to be conducted is significant due to a few reasons. The addition of antimicrobial agents hindering acidification is the opposite of an environmental health officer's perspective. Considering environmentally healthy food, low pH must be ensured for human consumption when left unattended for a long period of time at ambient room temperature. Also, the hurdle technology was applied in the study conducted by Kim, et al. Microbiologically stable food, especially for fermented food products, require variety of factors for preservation. Preservation involved three hurdles (temperature, pH and water activity) to analyze the correlation among these factors.

Characterization of Pathogenic Escherichia coli Strains linked to an Outbreak Associated with Kimchi Consumption in South Korea, 2012

In September 2012, an outbreak occurred in Gyeonggi province, South Korea, affecting 1200 students and food handlers from 7 schools. The causative agent is pathogenic *E. coli* in contaminated kimchi. Investigations and sampling tests from individuals with the illness identified EAEC and ETEC genotypes which have 98% homology with the bacteria from kimchi. Environmental investigation was performed to identify sources of *E. coli*; suspected sources were water tanks, cafeteria, cooking equipment, and the water supply. An epidemiological investigation was done and determined that on September 6th, 421 students

from one of the 7 schools reportedly became ill and additional 772 students from the other 6 nearby school became ill on September 6^{th} to 10^{th} . The outbreak confirmed that it is pathogenic *E. coli O120 EAEC, O120 ETEC*, and *O99 ETEC*; however, the source of the pathogen was inconclusive (Cho, et al, 2014).

Pathogenic E. coli outbreak is a serious concern. The outbreak research done was significantly relevant since it contained both kimchi and its potential risk of harbouring the disease. Pathogenic E. coli is transmitted via fecal-oral route. It could possibly be direct person-to-person contact or indirect food contamination. Production of kimchi involves extensive food handling compared to other dishes since the mixing needs to be done by hand (unless it is industrially produced from a manufacturing plant). As such, there are numerous ways for pathogens to be introduced. The outbreak investigation, however, does not include the fermentation process of the kimchi that was served to the students. The temperature of the fermentation as well as the pH level of the kimchi served was not included in the study. Therefore, the correlation of this review was limited to the mode of transmission, genotypes of E. coli and the risk of kimchi-borne outbreak. Nonetheless, this outbreak investigation provided informative methods of conducting a food-borne outbreak investigation and its application for EHOs in the field.

Kimchi fermentation and characteristics of the related lactic acid bacteria

A review of different factors affecting the naturally occurring microflora in kimchi provides different aspects of each factor discussed: temperature, raw ingredients, salt concentration, and starter cultures.

In this particular review, temperature and its effect on fermentation process was discussed. Although kimchi is usually fermented at lower temperature for better ripening and slower acidification, there are cases where some do ferment kimchi at higher temperature for faster acidification (Jung and Jeon, 2014). Noticeable change was observed when kimchi was fermented at 20°C; pH dropped drastically. However, slower decrease in pH was noted at 10°C compared to the higher temperature. Maximum total acid produced in kimchi at 20°C and 15°C was 1.6% (Mheen). Salt concentration was another factor to consider. The salt concentration during fermentation was usually around 2-3% which is optimal for normal microflora. Acidification of kimchi slowed down at higher concentration than the optimal range.

Restaurants sour on rules over kimchi

Lidea Park, the owner of Duck Hyang restaurant in Queens, received 7 violation points during her restaurant inspection in June 2011. The violation points were due to five pounds of kimchi sitting at room temperature and going over the city health authority's 41-degree Fahrenheit temperature requirement for cold foods. The health department demanded that after kimchi is made (kimchi-making procedure is not the health concern), kimchi has to be treated like any other cold food item and must be treated and stored as such; 41-degrees Fahrenheit or lower. The owner of Korean restaurants were forced to shift their ways of life and ways of preparing staple food of their cultural cuisine.

Health department was showing concern that kimchi, as it is not very well known to them, may pose public health risks. Restaurant owners and health officials were frustrated because they could not come to an understanding of kimchi to the same level. The relevance of this article to the subject of kimchi was crucial to understand because it showed the hardship EHOs are going through in different parts of United States and Canada. With a better understanding of the fermentation steps, such unfortunate cases and complaints may be avoided.

World's Healthiest Foods: Kimchi (Korea)

For its nutritious value, kimchi was nominated as one of the world's healthiest food. Although there were other opinions on consumption of kimchi causing gastric cancer (Nan. Et al. 2005), kimchi still holds greater benefits to the public and is a dish that require more understanding by environmental health officers Metro Vancouver.

Gastric Cancer Epidemiology

A research was conducted to understand the onset of gastric cancer in South Korea. Under the dietary factors, authors noted that kimchi is responsible for 20% of sodium intake. "Case-control studies on the intake level of kimchi and gastric cancer risk generally showed an increased risk among subjects with high or frequent intakes of kimchi" (Shin, et. al. 2011).

Methods

Production of Kimchi

Chinese cabbages and white radishes were washed thoroughly, removing all the dirt and other debris. Then, cabbage and radishes were put into a large container filled with water mixed with 3 kg of salt. The length of salting process depended on the amount of salt mixed with water (Family Member. 2014). For 3 kg of salt, researcher's family member preferred to have it overnight, roughly 10 hours long.

While pickling continues, kimchi sauce was prepared. Into a large container, 500g of diced green onions, 1.5kg of powdered chili peppers, 1kg of ground garlic, 100g of ground ginger, 450g of fermented anchovy sauce, 700g fermented shrimp sauce, 5 teaspoons of brown sugar, 1 diced onion, and a liter of glutinous rice broth were added. This broth was prepared by adding small amount of powdered glutinous rice into water and boiling if for several hours to produce semi-clear broth. The purpose for this broth was to soften and to add water to the sauce so that adequate mixing is possible as well as for easier administration to the cabbages and radishes (Family Member. 2014).

After salting has completed, cabbages and radishes were washed with fresh water to remove any salt content on the surface of the vegetables in order to avoid too much saltiness. Cabbages and radishes were mixed with the prepared seasoning. Seasoning was applied to each leaf of the cabbages until all the sauces were adequately spread evenly (Seoulistic. 2014). The prepared kimchi was, then, cut into small portions for the study.

This study required 60 containers to separate kimchi into two different categories; 4°C and 25°C. The containers needed to be large enough to hold sufficient amount of kimchi for testing and small enough keep them in the refrigerator. Small plastic containers (2oz) with lids were purchased from Costco for this research. Lids were important since they create similar conditions for kimchi in normal environment (Family Member. 2014). It is common to hold kimchi in closed containers and leave them in the refrigerator or outside for fermentation.



The set prepared on the left was put in the kimchi-refrigerator and the set prepared on the right was left in a room at a constant temperature of 25 degrees Celsius.

Proposed Standard Method

The proposed standard method to be used in this research is to analyze the pH of same kimchi in two different conditions using a Waterproof Palm pH meter. Prepared kimchi was divided into two groups: non-refrigerated and refrigerated. Each group contained 30 samples. Non-refrigerated kimchi was sitting at 25°C, and refrigerated kimchi was sitting at 4°C. Kimchi samples at 25°C were stationed in a room with constant air flow to maintain temperature and the room was not disturbed unless for measurement purposes. Kimchi samples at 4°C were positioned away from the entrance as kimchi-refrigerators have additional cover to prevent air from escaping. Each sample was tested for pH at various points in time (2h, 4h, 6h, 8h, 10h, 22h, 34h, and 46h) and pH analyzer was used to produce most valid and reliable data for statistical analysis.

Results

Two sets of 30 samples of kimchi underwent descriptive and inferential statistical tests to analyze their statistical significance. Descriptive statistical analysis found that the 25°C set, compared to 4°C set, showed a decline in pH level after 22 hours.

Time	Mean pH @	Mean pH @
	$4^{o}C$	$25^{\circ}C$
2hr	5.71	5.66
4hr	5.72	5.71
6hr	5.73	5.72
8hr	5.70	5.71
10hr	5.72	5.72
22hr	5.71	5.72
34hr	5.72	<mark>4.85</mark>
46hr	5.70	<mark>4.21</mark>

The significance level at $\alpha = 0.05$ at the room setting was 0.0000. With this information, the researcher was able to infer that the null hypothesis is rejected at the room setting. On the other hand, the significance level at $\alpha = 0.05$ at the fridge setting was 0.2817; therefore, the null hypothesis was not rejected.

Juxtaposing results from each setting, the researcher was able to determine the effect of temperature on pH variance in kimchi. Higher temperature was related to faster and more drastic change in pH.

Set-	R-	Cor-	Τ-	Sig. lvl
ting	Squared	rela-	value	of T
		tion		
Room	0.8313	-0.91	-34.2	0.0000
Fridge	0.0049	-0.07	-1.08	0.2816

Descriptive Statistics

Statistical analysis was done with real data. The results were recorded at 2 hr, 4 hr, 6 hr, 8 hr, 10 hr, 22 hr, 34 hr, and 46 hr after the kimchi was made. As time went by, the mean and the median for the set left out in 25°C showed a steady but much faster decrease in the pH level. Whereas the set left in the refrigerator showed, although a decreasing patter, much slower change compared to the set in the ambient temperature. The standard deviation for the pH at 4°C (fridge) was much narrower than the pH at 25°C (room), which indicated that data points were closer to the mean at 4°C.

	Standard deviation	Mean	Max/Min
pH @ Room	0.5411	5.4143	5.84/4.07
pH @ Fridge	0.0476	5.7161	5.80/5.63

It was clearly visible that the decrease in pH was much greater and faster when kimchi was held at 25°C compared to 4°C. Note that the difference in the means of pH at room temperature and fridge temperature is not big. This was due to pH dropping drastically at later points in time rather than showing a gradual change over time.





Inferential Statistics

Inferential statistics were used to make educated judgments of the probability that the observed difference between groups were a dependable one and extrapolated the knowledge onto a more generalized statement (Trochim. 2006). The proposed statistics strategy used for this research was a linear regression and correlation test.

Two linear regression and correlation test were conducted; then the results were juxtaposed to elaborate statistical significance of the findings.

Two sets of hypotheses were set:

- Null hypothesis @ room temperature (H_{OR}): In 46 hours, there will be no change in pH at room temperature.
- Alternate hypothesis @ room temperature (H_{AR}): In 46 hours, there will be a change in pH at room temperature.
- Null hypothesis @ fridge temperature (H_{OF}): In 46 hours, there will be no change in pH at fridge temperature.
- Alternate hypothesis @ fridge temperature (H_{AF}): In 46 hours, there will be a change in pH at fridge temperature.

Interpretation of the Statistical Analysis

pH at Room temperature $(25^{\circ}C)$

Using 240 observations in this dataset, the equation of the straight line relating pH and time is estimated to be pH = (5.9562) + (-0.0328) xtime. The value of R-Squared, the proportion of the variation in pH that can be accounted for by variation in time, is 0.8313. The correlation between pH and time was -0.9117. A significance test that the slope was zero resulted in a t-value of -34.2420. The significance level of this t-test was 0.0000. Since 0.0000 < 0.0500, the null hypothesis is rejected. The estimated slope is -0.0328. The lower limit of the 95% confidence interval for the slope is -0.0347 and the upper limit is -0.0309. The estimated intercept is 5.9562. The lower limit of the 95% confidence interval for the intercept is 5.9141 and the upper limit is 5.9983.

pH at Fridge temperature $(4^{\circ}C)$

Using 240 observations in this dataset, the equation of the straight line relating pH and time is estimated to be pH = (5.7197) + (-0.0002) xtime. The value of R-Squared, the proportion of the variation in pH that can be accounted for by variation in time, is 0.0049. The correlation between pH and time is -0.0698. A significance test that the slope is zero resulted in a t-value of -1.0791. The significance level of this t-test is 0.2816. Since 0.2816 > 0.0500, the null hypothesis cannot be rejected. The estimated slope is -0.0002. The lower limit of the 95% confidence interval for the slope is -0.0006 and the upper limit is 0.0002. The estimated intercept is 5.7197. The lower limit of the 95% confidence interval for the intercept is 5.7107 and the upper limit is 5.7287.

Discussion

There have been limited number of attempts to understand the acidification process of kimchi in terms of when and how fast the pH decreases in a course of the fermentation. Results from this study showed that kimchi set at room temperature (25°C) had a steady level of pH. However, drastic drop in pH resulted in between 22hr and 34hr after it was made.

A research conducted in 2012 by Kim, Bang, et al., described different processes to control fermentation of kimchi using naturally occurring antimicrobial agents. They added herbs, leaves, fruits and vegetables to inhibit lactic acid forming bacteria. This research employed different hurdle technology to control acidification of kimchi (Kim, Bang, et al. 2012); however, it did not include temperature and time. Their research, if incorporated time and temperature to understand the rapidity of acidification, may have shown different aspects of the hurdle technology. Nevertheless, they were able to show the inhibition of fermentation process with the additions of antimicrobial agents.

In a review by Jung and Jeon in 2014, temperature and its effect on fermentation process was discussed. Noticeable changes were observed when kimchi was fermented at 20°C; pH dropped drastically. However, slower decrease in pH was noted at 10°C compared to the higher temperature. Similar results were shown with this study (Jung, Jeon. 2014); however, this review did not note on the rapidity of the fermentation.

The main difference from previous researches from the current one is that both studies mentioned above did not include time component of fermentation.

The focus of this study was to understand kimchi and its fermentation processes in specified situations (temperature in this particular case). With evidences support by previous research, this study was able to synthesize a result that kimchi, as time passes, will ferment at a different rate when exposed to different environments. However, the degree of differences in fermentation did depend on temperature. Comparable to Jung and Jeon's research in 2014, two different sets of temperatures were employed which lead to similar trend but different quantitative results.

Unfortunately, there were limited number of studies regarding kimchi fermentation and related pathogenic microorganisms. As a result, there are

no guidelines and/or legislations for EHOs to reference to when dealing with kimchi during inspections. The results of this study, although not very informative in terms of specific pH change points, can be the basis of a development of policies and public health practices. Different tools can be developed to connect with operators with the appropriate knowledge of what and how important kimchi is to the Korean culture.

Recommendations

There are no specific guidelines or sections in guidelines about kimchi in BC health authorities. However, following the general standards, kimchi should be stored at 4 °C or lower to ensure safety of the public. Specific to different types of kimchi, all included ingredients in the process of making kimchi should be listed and managed in a proper manner. Operators also need to consider different options for storing kimchi other than in room temperature. Once kimchi's pH drops to or below 4.6, it is not considered a potentially hazardous food; therefore, it can be kept in the room temperature.

EHOs, with the findings of this study, can approach operators to discuss safe consumption of the ethnic dish. The spark of the discussion will be how long can kimchi be out in the room temperature. Since there are only a few resources to fully grasp the pH change during fermentation of kimchi, EHOs would have to use discretion to decide whether the food is safe or not at the time of inspection. Tools to measure pH onsite are recommended.

Limitations

Calibration for Dimchae dual compartment kimchi refrigerator was not plausible at the time of study due to the fact that sending this product back to the manufacturer for calibration only incurred increase in cost and waste in time. In order to overcome this deficiency, the refrigerator was tested for accuracy of automated temperature display with non-electronic mercury thermometers used for freezers and walk-in coolers in food service establishments.

Another possible error is that the production of kimchi relied heavily on producer's recipe. Although it followed the traditional way of making kimchi, slight variations were still present. Ingredients used to make kimchi varies significantly depending what type it is. As a completely different set of ingredients can be used to produce a different type of kimchi, the results from this study generalized kimchi as one-and-only product. If a different type of kimchi was used, there would be a possibility of different results.

Many improvements were required; an example included the independent variable, time. This study could have produced better results if the time gap between measurements were consistently 2 hours. The results show that pH drop began to occur sometime between 22hr and 34 hr. In addition, safe pH limit of 4.6 was established sometime between 34 hours and 46 hours. With more frequent measurements, more accurate data set could have been collected to show at what time the pH actually began to decrease.

Sampling itself raised concerns as well. Each set of measurements took more than 30 minutes. This meant that samples from the fridge had to be constantly moved around. Although the results did not reflect this problem, faster measurement methods could have been sought to decrease the time it takes to analyze all the samples. Moreover, when measurements were taken, the last sample was always measured 30 minutes after the first sample. The consequences of this pattern were not identified; however, there is a possibility that skewed data were collected.

The largest limitation of this study was time. Time was a crucial component of this research since it was the only controlled variable other than temperature. Time was also an aspect that EHOs would look for when answering the questions, is kimchi a hazardous food. Unfortunately, taking measurements every 2 hours were not feasible. As a result, broad and generalized data sets were collected. With this result, environmental health officers would have a difficult time educating and coercing food service establishment operators how kimchi can be handled onsite. In order to close the gap between the two parties, EHOs would need studies that show more relevant results that can be employed in the field of public health.

Conclusion

Kimchi is commonly left out in ambient temperature or in a kimchi fridge. Unfortunately, lack of knowledge on the food and kimchi fridge technology, concerns the public of potential health risks. Descriptive statistical analysis found that the 25°C set, compared to 4°C set, showed a decline in pH level after 22 hours. Only between 34 hours and 46 hours it showed a decrease in pH significant enough to be not considered a hazardous food. Juxtaposing results from each setting, the researcher was able to determine the effect of temperature on pH variance in kimchi. Higher temperature was related to faster and more drastic change in pH. With these information, the researcher was able to infer that the kimchi is not safe to be stored at room temperature since it can harbour pathogenic microorganisms such as ETEC. Storage at 4 °C or lower is recommended until pH drops to or below 4.6 as suggested by BCCDC.

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Competing Interests

The authors declare that they have no competing interests.

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