

Kimchi: A Cross-Vegetable Analysis of Biodiversity & Fermentation



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Abstract

The purpose of this study is to test for variations in biodiversity between kimchi of different vegetable types. Kimchi is a side dish traditionally made with napa cabbage fermented over several days or weeks. We wanted to investigate how different vegetables would cause the fermentation to differ among each other. Therefore, we tested bacterial biodiversity between kimchi made with bok choy, carrot, radish, and cabbage. We believe the biodiversity will be consistent across all vegetables because the conditions among all vegetables are the same: fermentation and marination processes. To test this hypothesis, a methodology consisting of kimchi creation, fermentation, sampling, DNA extraction, quantification, and sequencing was conducted. As a result, we'll be able to deduce whether the type of vegetable affects the biodiversity in kimchi, mainly due to its fermentation process. The primary intention of this study is to comprehend the biodiversity in kimchi when it's made with diverse vegetables, which have varying nutritional, physical, and chemical factors.

Background

Kimchi is a well known dish in Korea. Kimchi is marinated napa cabbage that is fermented for weeks or even months. Kimchi is a versatile dish that can be made with any fruit and vegetable. Notably, *Lactobacillus*, *Weissella*, and *Leuconostoc* have been the primary genera of bacteria across kimchi [2]. These LAB bacteria produce polypeptide nisin and chemical compounds which help preserve and flavor fermented foods [3]. Furthermore, consumption of kimchi - containing LAB bacteria - can increase metabolic activity, prevent disease through antimicrobial compounds, improve gut health [4].

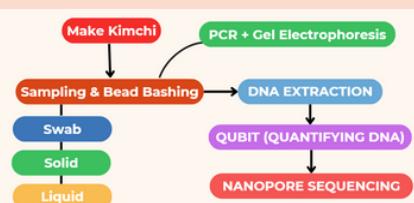
While the role of LAB bacteria has been established as crucial to the fermentation process, flavor profile, and human health, the role of vegetable type in bacterial biodiversity has not been extensively explored [5]. The vast majority of studies have focused simply on napa cabbage despite the 200 variations of kimchi that exist [1]. Kimchi made with different vegetables primarily contain similar LAB bacteria, but vary in terms of specific species, abundance of species, and pH over time [5].



Method and Process Steps

Significant Materials

- ZYMO DNA Prep Kit
- Bead Bashing machine (ZYMOBIOMICS BashingBead buffer)
- Qubit fluorometer
- Nanopore Sequencing machine
- InstaGene Matrix Kit
- Vacuum filtration machine (for liquid DNA extraction)
- Nanopore sequencing software



Kimchi Preparation + Bead-bashing

Kimchi samples were marinated and fermented over the course of two weeks. Samples were bead-bashed on a vortex genie.



DNA Extraction + Qubit

We used DNA binding buffers and filters repeatedly in order to purify the DNA, which was released from the bead bashing tubes. Then the Qubit used a fluorometer to analyze DNA concentrations.



Nanopore Sequencing

DNA was separated into bacterial and nonbacterial DNA using magbinding and amplified to prepare for nanopore sequencing. Then the nanopore machine sequenced DNA by vegetable into different genera.



Research Question: How does bacterial biodiversity compare in fermented kimchi made with different vegetables?

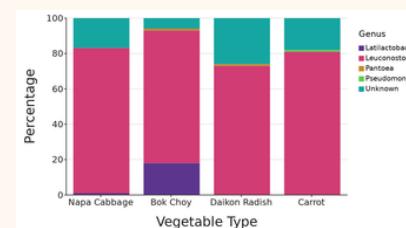
Hypothesis or Criteria for Success

The biodiversity of kimchi is dependent on the vegetable utilized to make the kimchi, because of differences in inherent bacteria present in different types of vegetables.

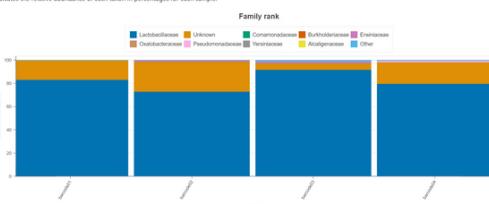
Results

The primary types of bacteria, classified as genera, that were found throughout all our vegetables were *Leuconostoc*, *Lactilactobacillus*, *Pantoea*, and *Pseudomonas*. However, the highest concentration of bacteria that was present throughout all vegetable samples was *Leuconostoc* bacteria.

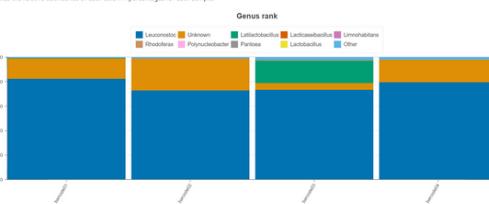
Shannon diversity is a type of index that allows scientists and researchers to compare biodiversities across their samples - higher the number, larger the diversity. In our experiment, bok choy had the highest index score of 0.86 while napa cabbage, daikon radish, and carrot had scores of .51, .65, and .6 respectively. This was a drastic change from the control group of napa cabbage.



Barplot of the 9 most abundant taxa at the family rank in all the samples. Any remaining taxa have been collapsed under the 'Other' category to facilitate the visualization. The y-axis indicates the relative abundance of each taxon in percentages for each sample.



Barplot of the 9 most abundant taxa at the genus rank in all the samples. Any remaining taxa have been collapsed under the 'Other' category to facilitate the visualization. The y-axis indicates the relative abundance of each taxon in percentages for each sample.



Discussion

The differences in bacteria between samples is attributed to inherent changes between vegetable structure and general bacteria. The various structures of the vegetables, including leafy and non-leafy vegetables, may affect the amount of bacteria gathered to utilize as starters for fermentation. Additionally, various vegetables differed in terms of fermentation pace affecting their stage of fermentation during sampling. For instance, carrot, bok choy, and daikon all had a slower fermentation pace than the control group nap cabbage. Daikon radish and bok choy had larger percentages of *Pantoea* bacteria and carrot had larger amounts of *Pseudomonas* which are bacteria associated with each respective vegetable in their early fermentation stages. Furthermore, *Leuconostoc* dominated every vegetable sample as it is a common lactic acid bacteria that suppresses the growth of other bacteria.

Next Steps

Knowing that vegetable causes a difference in biodiversity...

- How does temperature, different amounts of marinade, or water content affect the fermentation process if the type of vegetable is held constant across all samples?

Since Kimchi has numerous health benefits...

- Which type of vegetable based kimchi has the highest potential for probiotic benefits and better gut health?
- A bigger picture...
- Can understanding kimchi fermentation help us learn about microbial communities in different types of fermented foods such as yogurt, kombucha, and sourdough bread?

References

Choi, I., Jung, S., Kim, B., Park, S., Kim, J., & Han, H. (2003). Novel *Leuconostoc citreum* starter culture system for the fermentation of kimchi, a fermented cabbage product. *Antonie Van Leeuwenhoek*, 84(4), 247–253. <https://doi.org/10.1023/a:1026050410724>

Song, H. S., Lee, S. H., Ahn, S. W., Kim, J. Y., Rhee, J.-K., & Roh, S. W. (2021). Effects of the main ingredients of the fermented food, kimchi, on bacterial composition and metabolite profile. *Food Research International*, 149, 110668.

Maoloni, A., Ferrocino, I., Milanović, V., Cocolin, L., Corvaglia, M. R., Ottaviani, D., Bartolini, C., Talevi, G., Belleggi, L., Cardinali, F., Marabini, R., Aquilani, L., & Osimani, A. (2020). The microbial diversity of Non-Korean kimchi as revealed by viable counting and metataxonomic sequencing. *Foods*, 9(11), 1568. <https://doi.org/10.3390/foods9111568>