



Verifying the Ingredients of Convenience Store and more Expensive Salads

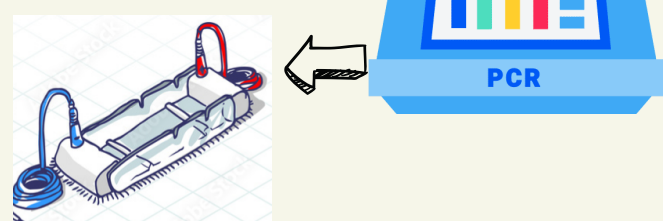
Andy Qian
Stephen Wang
Yvonne Zhang
Mentor: Jeffry Petracca

INTRODUCTION

As salad lovers, salads are an important part of our daily diet. The vegetables and meats in salads can help us to prevent diseases including cancer, diabetes, and cardiovascular disease (Sebastian 2018). However, salads can vary wildly in price even though they seemingly contain the same ingredients. So, we were curious how the price of salads that we eat in Suzhou compared to their ingredients. Food fraud is a severe problem that deceives consumers and introduces threats to food safety (Manning 2016). However, it makes us wonder what makes salad prices different. Do expensive salads really worth their price? Our observations showed that the price differs from 10 CNY to about 100 CNY between different salads. To better understand what makes the price difference, we planned to answer the question by using DNA barcoding technology. DNA barcoding is a system used for species identification by focusing on a specific genetic region used as a barcode. We planned to use DNA barcoding to identify components from salads of different prices. We used the *rbcl* region as barcodes for plants and the *COI* region for animals. We predict that the more expensive the salad is, the ingredients will be richer in diversity and quality, if not, consumers should be careful about making salad purchases.

METHODS

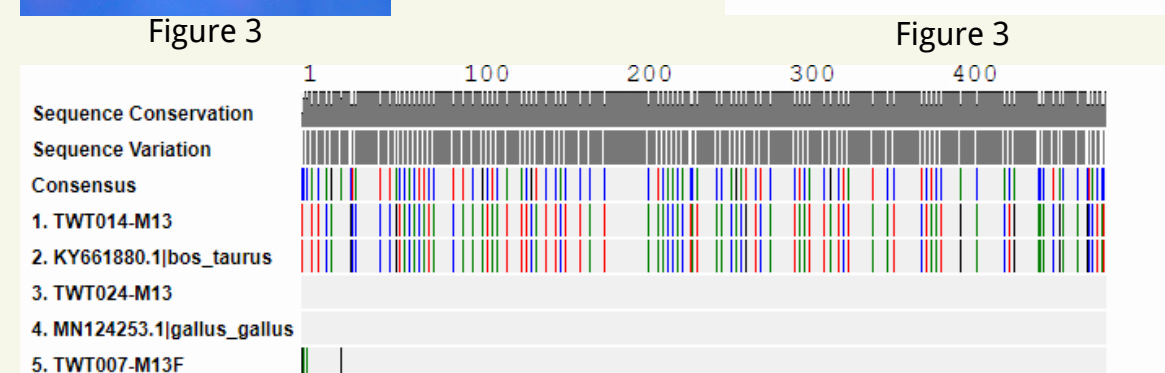
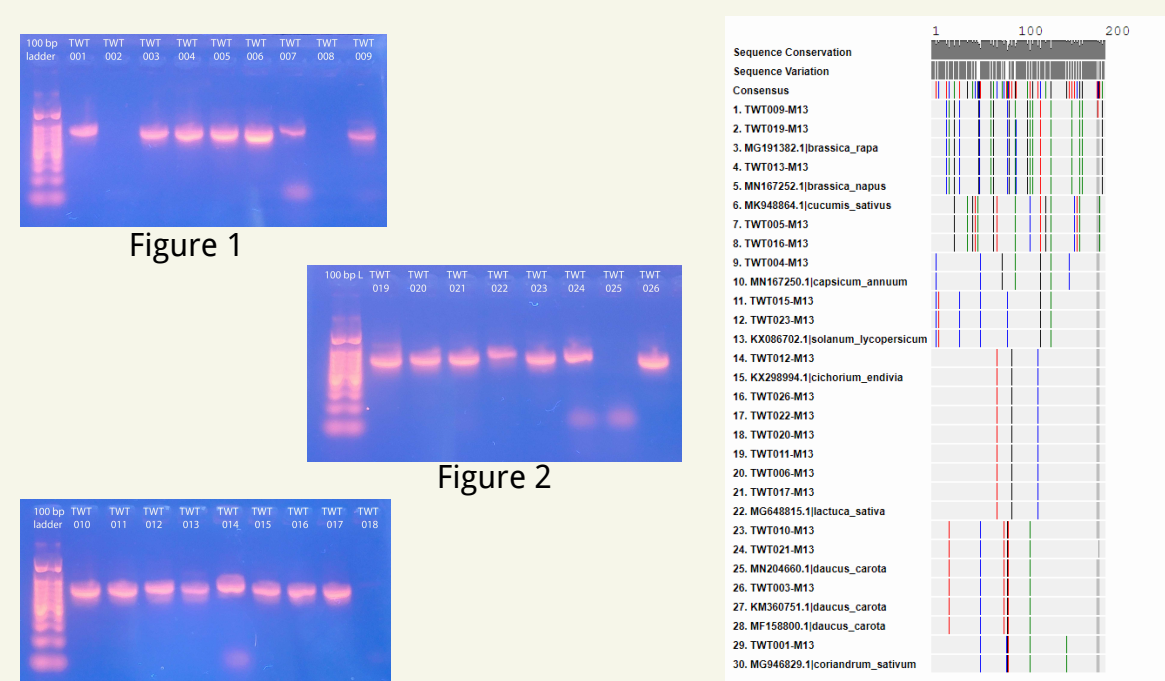
1. We found multiple samples of cheap and expensive salad, and extract DNA from the materials that it contains
2. We used the silica DNA isolation method from the CSH DNALC booklet. (DNALC)
3. We set up a polymerase chain reaction for our samples, which amplified the marker region, making it possible for DNA sequencing. We used the *rbcl* region of the chloroplast for plants and the *COI* region of the mitochondria for animals.
4. We sent samples to a third-party company, Azenta Life Science, for DNA sequencing
5. After receiving the DNA sequence for each sample, it was trimmed and compared to sequences in the GenBank database to see what species this sequence belongs to.
6. We compared the final result to the ingredients chart on the packaging of each product to find out whether food fraud exists.
7. We compare results from convenience store salads and expensive salads and find out which one is a better choice.



ABSTRACT

Food fraud is a serious problem in that sellers deceive consumers by using different ingredients than claimed ingredients. More than money lost, food fraud also proposed threads to the health of consumers. We aimed to test whether expensive salads are worth their price. We collected 5 different kinds of convenience store salads and collected ingredients from 3 of them. The average price of these convenience salads is about 13 CNY. We also collected the ingredients from an expensive salad that costs 80 CNY. We processed of ingredients and used the method of DNA Barcoding to identify the species of these ingredients. To do that, we extracted DNA from each sample and used PCR to amplify *rbcl* for plants and *COI* for animal products. After that, we sent the samples to a third-party company for DNA Sequencing. With our DNA sequence, we processed them using DNA Subway and compared them to data in GenBank to determine which species each sample is. With our results, we can't answer our question because of the limitations of DNA barcoding which is that it can't identify the subspecies or cultivars of samples that were obtained through selective breeding. In addition, it can't determine the freshness of ingredients, which is also a price-determining factor.

RESULTS



After processing the DNA sequence for our samples, and comparing the DNA sequence to data in Genebank, we found the species of our samples. And to compare the sample species we tested and the sample species the manufacturers claimed, we made a table. From our gel phosphoresces images, we can see that all of our samples worked except for samples TWT-002, TWT-008, TWT-018, and TWT-025. You can see the species identified for each sample listed in our table (Figure 5). 17 out of 22 successful sample results have the same family of the ingredient claimed by the manufacturer. 14 out of 22 successful samples have the same species compared to the claimed ingredient by the manufacturer.

Sample ID	Sample specie provided from the salad package	DNA Sequencing	Result	Same family	Same specie
TWT-001	Coriandrum sativum	T	Coriandrum sativum T	T	T
TWT-002	Spinacia oleracea	F	/ /	/	/
TWT-003	Daucus carota	T	Daucus carota T	T	T
TWT-004	Capsicum annuum	T	Capsicum annuum T	T	T
TWT-005	Cucumis sativus	T	Cucumis sativus T	T	T
TWT-006	Brassica oleracea	T	lactuca sativa F	F	F
TWT-007	Gallus gallus	T	Gallus gallus T	T	T
TWT-008	Brassica oleracea	F	/ /	/	/
TWT-009	Raphanus sativus	T	brassica napus F	F	F
TWT-010	Daucus carota	T	Daucus carota T	T	T
TWT-011	actuca sativa	T	lactuca sativa F	F	F
TWT-012	Lactuca sativa longifolia Lam.	T	lactuca sativa T	T	T
TWT-013	Brassica oleracea	T	brassica napus T	F	F
TWT-014	Bos taurus	T	Bos taurus T	T	T
TWT-015	ycopersicon esculentum	T	Solanum lycopersicu F	F	F
TWT-016	Cucumis sativus L	T	Cucumis sativus T	T	T
TWT-017	Lactuca sativa L. var. ramosa Hort	T	lactuca sativa T	T	T
TWT-018	Gallus gallus L.	F	/ /	/	/
TWT-019	Brassica oleracea	T	brassica napus T	F	F
TWT-020	Lactuca sativa	T	lactuca sativa T	T	T
TWT-021	Daucus carota	T	Daucus carota T	T	T
TWT-022	Lactuca sativa	T	lactuca sativa T	T	T
TWT-023	Lycopersicon esculentum var. cerasiforme	T	Solanum lycopersicu F	F	F
TWT-024	Gallus domesticus	T	Gallus gallus T	F	F
TWT-025	Sus domesticus	F	/ /	/	/
TWT-026	Lactuca sativa L. var. longifolia	T	lactuca sativa T	T	T

Figure 5

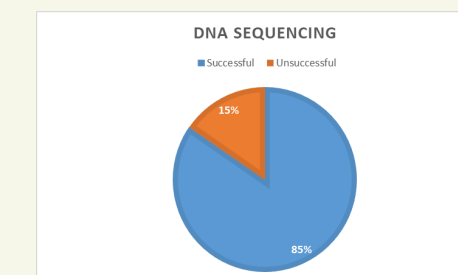


Figure 6

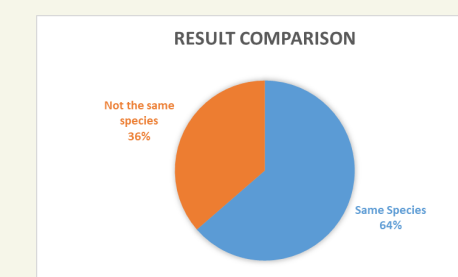


Figure 7

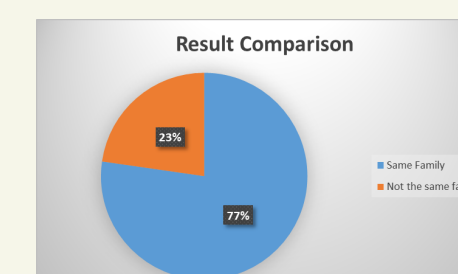


Figure 8

DISCUSSION

From our results, most of the components claimed by the manufacturers are accurate. The lettuce provided by expensive salads was nothing different from lettuce from cheap salads bought in convenience stores. The producer of the expensive salad claimed that they used a specific lettuce called *Lactuca sativa* L. var. *longifolia*. However, DNA barcoding can not identify which subspecies a sample is, so it is impossible for us to identify whether a lettuce's variation is *longifolia* or not. This critical limitation caused us to be uncertain about our hypothesis that costly salads provide us with better ingredients, and to find out about food fraud. Because the technique of selective breeding is largely used among cultivars and domestic animals. For example, there are many subspecies of lettuce that costs different prices. Because we can't determine exactly what subspecies a sample is, so we can't determine whether there's food fraud or not. Another limitation of the barcoding technique is that we can not determine the freshness of an ingredient. However, the freshness of the ingredients used in a salad is also a price-determining factor.

ACKNOWLEDGEMENT

We would like to thank our mentor Jeffry Petracca for tutoring us. His expert advice and encouragements are vitally important for us throughout our project. We also would like to thank Cold Spring Harbor DNA Learning Center and Cold Spring Harbor Asia for providing us with lab materials and the opportunity to do our project.

REFERENCE

"DNA Learning Center Barcoding 101." DNA Learning Center Barcoding 101, dnabarcoding101.org/. Accessed 28 July 2023.
Sebastian, Rhonda S., et al. "Salad Consumption in the U.S. What We Eat in America, NHANES 2011-2014." FSRG Dietary Data Briefs, United States Department of Agriculture (USDA), February 2018.
Manning, Louise, and Jan Mei Soon. "Food Safety, Food Fraud, and Food Defense: A Fast Evolving Literature." Journal of food science vol. 81,4 (2016): R823-34. doi:10.1111/1750-3841.13256