Whatcom COMMUNITY COLLEGE

The Zesty Experiment: Does Ginger Change Kombucha's Microbial Population?

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Biology 222: Cell and Molecular Biology

Abstract

Ginger has been used as a traditional medicinal antimicrobial for over 5000 years. It has been used in many forms, including, dried, pickled, preserved, crystallized, powdered, ground and grated. This experiment investigates the impact of freshly grated ginger supplementation on the microbiome of kombucha, a fermented tea-base beverage known for its SCOBY(a colony of bacteria and yeast that is used to kick start the fermentation process of Kombucha. We were inspired by a previous lab group, who used turmeric instead of ginger. In their research, they observed the growth of bacterial colonies in kombucha. After investigating this previous lab, we then decided to use ginger because of its previously mentioned, well-known antimicrobial effects and its influence on the microbial composition, diversity, and metabolic activity.

Introduction

As mentioned, kombucha has been acknowledged for its SCOBY. Its SCOBY is rich in bacteria and yeast that is beneficial to the human body in moderation (Al-Mohammadi, Abdul-Raouf, et al, 2021). Ginger has also been used medicinally throughout human history (Ortiz, 2015). How would ginger impact the microbial colonies of the SCOBY within the kombucha? As a result, we observed the various fermentation days with our grated ginger to view if there were any impacts on the microbial colonies of the SCOBY. Using PCR and gel electrophoresis to aid us in analyzing if there were any colonies prevalent. We had three plates in the span of seven days total; one with pure kombucha, ginger, and kombucha with ginger. During our preliminary research of this project, we determined that ginger would inhibit the growth of the bacterial colonies of the SCOBY due to its antimicrobial properties.

Methods

For our materials, we used .29 grams grated ginger, 89 microliters distilled water, 20 ml pure kombucha, pre-warmed hotshot, primers 320F and 817R, 5x neutralization solution another 100 microliters of pure kombucha for the six out of nine agar plates that contained kombucha/kombucha-ginger mixture. For the equipment we used a grater, P1000 pipette, nine agar plates, two 50 ml beakers, microfuge tube, vortex, centrifuge, centrifuge tubes, 1.5 ml test tube, a thermocycler, gel/gel-holder tray. We worked on this experiment for three and a half weeks 02/26/2025-03/24/2025 in Kulshan Hall, biology lab (108), Whatcom Community College. We collected this data during out 11:30pm to 1:25pm lab on Mondays and Wednesdays.

This experiment, conducted over two weeks, aimed to observe microbial growth and perform DNA extraction from kombucha and ginger samples, as well as a mix of the two. On day 0, grated ginger was massed and mixed either with kombucha for one sample, and ginger for another, as well as a sample with kombucha and ginger. Then plated, on labeled MRS InstaT agar plates using the hockey stick method. Plates were incubated at 30 degrees celsius, while the remaining mixtures were stored for further fermentation. On Day 3, Team 1(Scarlet and Sarah) repeated the plating process from Day 1 using the Day 3 fermented samples. Team 2(Gerrit and Cristal) carried out DNA extraction using PCR for Day 3 plates. On Day 7, Team 1 repeated the plating process using Day 7 fermented samples. Team 2 finished the PCR process as well as started it for Day 7 plates. Team 1 then also ran gel electrophoresis for Day 3 samples.

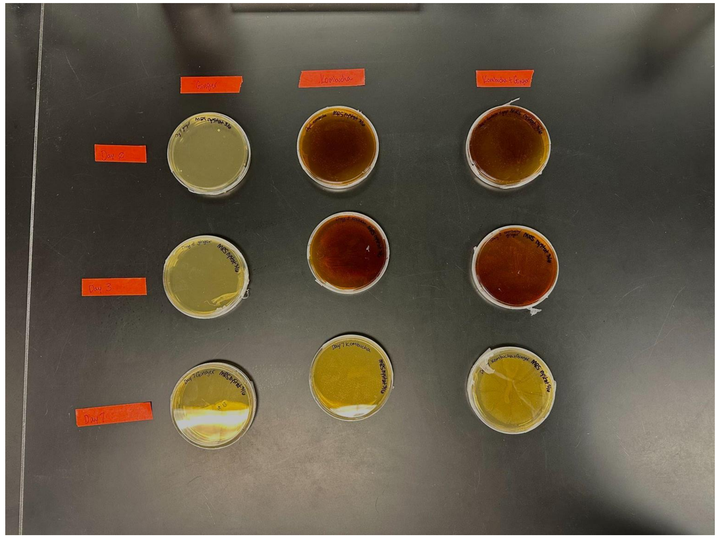

Figure 1. Set of plates with cultures from day 0, day 3, and day 7

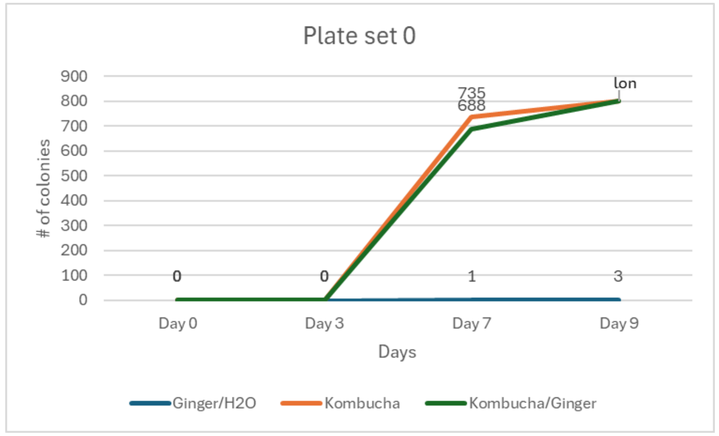

Figure 2. Plate set 0 colony counts

References

Ortiz, Marsunn. "Antimicrobial Activity of Onion and Ginger against Two Food Borne Pathogens Escherichia Coli and Staphylococcus Aureus." *MOJ Food Processing & Technology*, vol. 1, no. 4, 30 Nov. 2015,

Al-Mohammadi, Abdul-Raouf, et al. "Chemical Constitution and Antimicrobial Activity of Kombucha Fermented Beverage." *Molecules*, vol. 26, no. 16, 19 Aug. 2021, p. 5026, https://doi.org/10.3390/molecules26165026.

Discussion

By the end of our experiment we found that ginger infused kombucha showed slightly less growth compared to regular kombucha (668 vs 735) on our day zero plates by day three while very little growth was seen in our ginger and DH2O plate (3). As we progressed with our plating we saw a noticeable increase in the rate of growth between days as our cultures fermented and both our day three and day seven plates were too densely populated to count after 72 hours and were considered lon. When we performed our first run of PCR on our day 0 plates we unfortunately didn't get usable results during our electro gel phoresis stage to send off for PCR. During our second run on day 7 however, we were successful in extracting PCR samples from our plates and were able to send it off for sequencing. After getting our results back we saw a nearly identical makeup of bacteria between our kombucha plate and our kombucha/ginger plate with the majority of the bacteria being some form of a-proteobacteria.

We were hoping that ginger's anti-microbial properties would show a noticeable change in either the rate of microbial growth or the microbial makeup (Ortiz, 2015). We unfortunately couldn't make a definitive conclusion based on our results as our colonies grew much faster than expected and without multiple PCR tests. We were hoping to see if there was a change in the microbiome as the cultures fermented which could have told us if ginger had an effect throughout the fermentation process. If we were to repeat/continue this experiment in the future we would have used a more dilute concentration of kombucha or taken counts at shorter intervals (24 hours instead of 72 hours) to better track the growth of colonies before they turned to lon. In regards to PCR, we would ideally have found a specific type or types of bacteria that are both present in kombucha and affected by ginger. We would then find/use a more specific PCR primer to isolate said bacteria and measure its rate of growth to compare its abundance in relation to the presence of ginger.

We hope that in the future our findings (or lack thereof) can help further research in isolating specific classes of microbes that are both present in kombucha and affected by ginger. We hope that this may help us better understand ginger's specific role in kombuchas microbiome.

Acknowledgements

We would like to acknowledge our instructor Tealia Slagle for allowing us this opportunity and for her encouragement every step of the way.We would also like to thank the lab prep staff Bethany Lolkema, Kris Harrell, and Mark Price for providing materials and guidance.