Organic Prevention and Treatment of Mastitis-causing Bacteria in Organic Dairy Herds

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Abstract

Clinical research was conducted to prove the prevention and treatment efficacy of organically approved products through laboratory testing. A controlled environment was used to test the ability of CBD distillate and honey to combat prevalent Mastitis-causing bacteria.

Staphylococcus aureus was found to be susceptible to therapeutic honey and diluted CBD distillate. *E.Coli* was found to be resistant to the tested organic solutions.

This data can be used to develop organic, financially sustainable, and effective treatments for significant diseases like Mastitis in dairy herds.

Introduction

Mastitis is one of the most significant diseases that affect dairy cattle in the United States (Zoetis, n.d.). Several pathogenic bacteria can lead to mastitis in dairy cattle. Yet, organic dairies cannot combat the issue using traditional and effective antibiotics. Based on a review of the literature, little data are available that support the clinical efficacy of alternative products used to prevent or treat mastitis in organic dairy cows (Ruegg, 2009).

Organic dairies need viable prevention and treatment options. Thus, a much-needed first step to correcting this lack of clinical research is to prove the prevention and treatment efficacy of organically approved products through laboratory testing.

During our research, we determined the efficacy of organic CBD distillate and honey products on the prevention and treatment of the mastitis-causing bacteria most prevalent in organic herds.

Materials & Methods

Preparations

Mueller Hinton Broth Tubes Preparation

- 5.25g of dehydrated Mueller Hinton Broth powder
- 250 mL of Deionized Water
- Dissolve with stir bar.
- Pipette 9 mL of broth mixture in small test tubes.
- Autoclave.

Mueller Hinton Agar Preparation

- 38.0g of Mueller Hinton Agar powder
- 1000 mL of Deionized Water
- Dissolve. Autoclave.
- Pour agar solution into new petri dish plates.
- Leave sit for 24 hours.

Thawing Isolated Bacteria Cultures:
E. Coli* & *Staphylococcus aureus

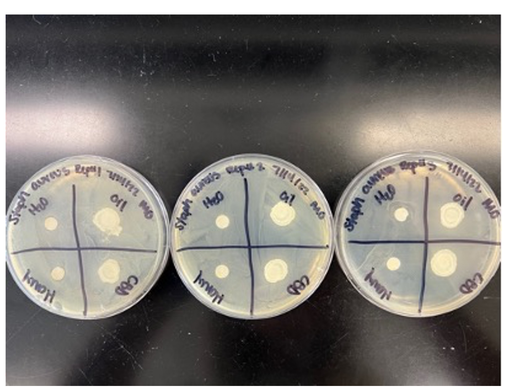
- Inoculation loop
- Quadrant streaking T-streak
- Store in incubator at 37°F.

McFarland 1x10^8 Standard Preparation

- Mueller Hinton Broth tube
- Sterile pipette
- Transfer colonies aseptically from overnight culture.
- Match turbidity.

Overnight Culture Preparation

- Inoculation loop
- 2 Mueller Hinton Broth tubes.
- Flame-to-loop aseptic technique
- Transfer cultures to tubes.
- Incubate at 37°F for 18 hours.



Assays

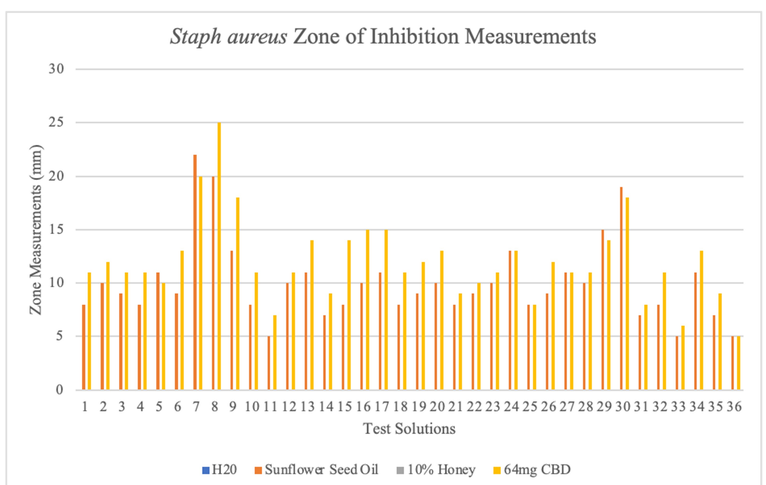
Standard Disc Diffusion Assay

1. 2 Mueller Hinton Agar plates
2. Label for each bacteria.
3. Label with quadrants: H20, Oil, Honey, and CBD.
4. Using a 200μL pipette, transfer 100μL of McFarland Standard on to plate.
5. Ethanol glass spreader, flame in Bunsen burner, cool.
6. Spread 100μL of MF Stnd. across the plate.
7. Warm 10% honey and 64mg CBD mL^{-1} solutions in water bath.
8. Vortex after removal.
9. Prepare 4 sterile plates for each solution.
10. Soak 2 sterile Whatman paper discs per solution.
11. Aseptically place 1 soaked solution disc on to each corresponding plate quadrant. Refer to plates from Step 6.
12. Repeat for each bacteria.
13. Incubate at 37°F for 18 hours.

Zone of Inhibition Measurement

1. Place ruler (mm) in the middle of the soaked paper, so that "0" is in the center.
2. Measure from center of disc to edge of area with zero growth.

Results



Results (cont.)

***No visual representation of *E. Coli* Zones of Inhibition measurement values due to zero value.**

Conclusions

Growth of the bacteria strain, *Staphylococcous aureus,* was inhibited by Sunflower Seed Oil and 64mg CBD. The growth of this bacteria was not inhibited by therapeutic Manuka honey. The fourth test solution, Deionized water, was used as a control variable so no change was anticipated.

Growth of the bacteria strain, *E. Coli*, was not significantly impacted by any of the four test solutions. The Gram-negative condition *E.Coli* can be used to explain the inability of any tested solution to inhibit growth. The presence of lipopolysaccharide in Gram-negative bacteria cell walls make bacteria more antimicrobial resistant, compared to Gram-positive bacteria like *S. aureus*.

Broader Impacts:

This research is the first step in developing an intramammary sealant or treatment that can be used on organic dairies. Finding an organic, financially sustainable, and effective treatment for significant diseases, like Mastitis, in dairy cows will protect farmers' integrity of organic labeling and dairy cattle quality of life. Not only does this project benefit the organic dairy industry, but at the same time, it has a large One Health impact.

Antimicrobial resistance (AMR) is a threat to human health and environmental biodiversity. The revolutionization of organic treatments can give opportunities to farmers to decrease the antimicrobial resistance on their farms.

Acknowledgements

BMS

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