Microchem Plus and Sporicidin Show Effectivity Against *Coxiella burnetii,* Natural Disinfectants Show Minimum Effect.

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Introduction

- *C. burnetii* is the causative agent of Q fever- a highly infectious disease, causing reproductive issues in animals and severe cases of infection in humans[1].
- Q fever has many agricultural, human, and economic impacts.
- *C. burnetii* exhibits strong environmental resistance by often residing in a spore-like state.[2]
- *C. burnetii* is known to be resistant to heat and many disinfectants, with research in disinfectant response lacking[2].
- We asked the question: will the following disinfectant concentrations be effective on a 7-day stock of *C. burnetii?*
  - 5% Microchem Plus[3]
  - 12.5% Oxivir Five[3]
  - Sporicidin[3]
  - Oregano Oil
  - 75 ug/mL Nisin Z + EDTA[4,5]

Objectives & Hypotheses

Objectives

- Identify disinfectants, natural and chemical, that have not been previously utilized for *C. burnetii* in research settings.
- Utilize disinfectants that have low toxicity, cost, and easy disposal.
- Utilize different amounts of time to determine minimum exposure time needed to result in complete kill if applicable.

Hypotheses

- **H1:** Disinfectants that purposefully target bacterial spore structure will be the most effective in inhibiting growth of *C. burnetii.*
- **H2:** Cultures that are exposed to disinfectants for an extended amount of time will show reduced growth compared to those exposed for shorter periods of time.
- **H3:** Chemical based disinfectants will be more effective in reducing or inhibiting growth than naturally based compounds.

Results

Disinfectant	Effect
5% Microchem Plus	+
12.5% Oxivir Five	-
Sporicidin	+
Oregano Oil	-
75ug/mL Nisin Z + EDTA	-

Figure 2: The effect of disinfectants on 7-day stock of *C. burnetii* on whether they caused complete kill at all titers and time points. **+** represents those disinfectants that produced complete kill, while **-** represents those that did not result in complete kill.

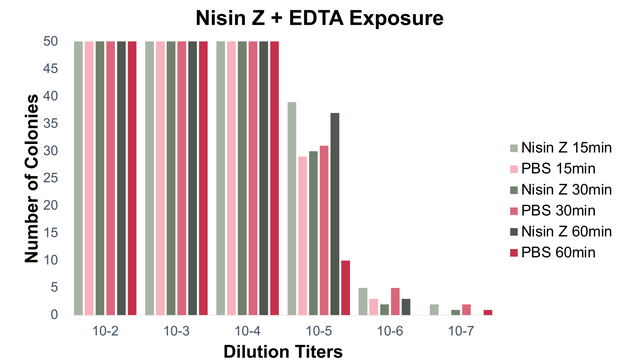

Figure 3: The average number of colonies remaining after exposure to 75ug/mL of Nisin Z + EDTA compared to the average number of colonies remaining after exposure to PBS control. Nisin Z + EDTA data is shown in green while PBS data is shown in pink.

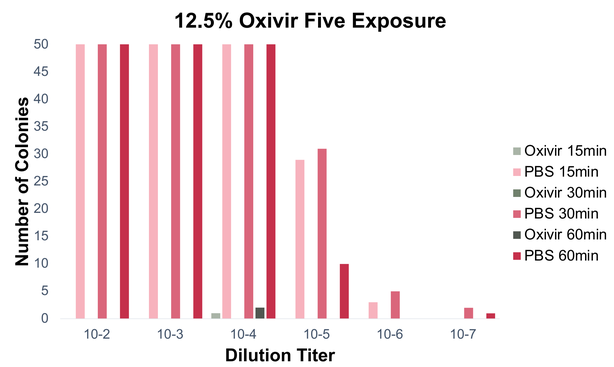

Figure 4: The average number of colonies remaining after exposure to 12.5% Oxivir Five compared to the average number of colonies remaining after exposure to PBS control. Oxivir five data is shown in green while PBS data is shown in pink.

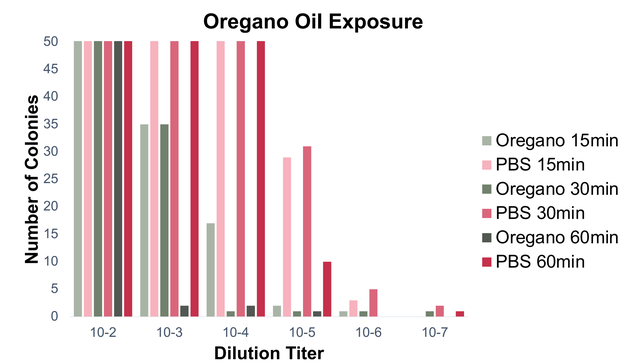

Figure 5: The average number of colonies remaining after exposure to oregano oil compared to the average number of colonies remaining after exposure to PBS control. Oregano oil data is shown in green while PBS data is shown in pink.

Methods

Set-Up

- A 7 day stock of *C. burnetii* was utilized in all rounds of experimentation.

Experimentation

- Disinfectants Tested:
  - 5% Microchem Plus
  - 12.5% Oxivir Five
  - Sporicidin
  - Oregano Oil
  - 75ug/mL Nisin Z + EDTA
- Phosphate buffered saline (PBS) & Ethanol were utilized as negative and positive controls respectively.
- Samples were exposed for times of 15 minutes, 30 minutes, and 60 minutes.
- Samples were serially diluted from 10^{-2} and 10^{-7}
- All samples were allowed to incubate for 10 days before being counted.

Analysis

- 3 trials were run per disinfectant (A, B, & C), averaged for statistical analysis.
- Effectiveness of the disinfectant was determined based on if complete kill occurred at all dilution titers and exposure times.

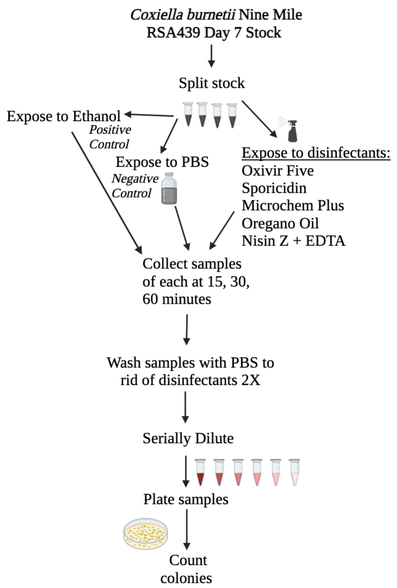

Figure 1: Research design schematic

Discussion

- Microchem plus and sporicidin resulted in complete kill of a 7-day stock of *C. burnetii* at all exposure time points. Since both disinfectants have low toxicity and easy disposal, they would be good to utilize in laboratory settings[3].
- Oxivir five mostly showed complete kill, however, colonies present at the lower dilutions are indicative of possible contamination.
- Oregano oil did not result in complete kill but did have reduced growth compared to the PBS control. This shows oregano oil is slightly bacteriostatic, which could be useful for less infectious cells than *C. burnetii*[2].
- Nisin Z + EDTA did not show any reduced growth compared to the PBS control. This could be due to the fact Nisin Z is more effective on gram-positive bacteria rather than gram-negative which is what *C. burnetii* is[5].

Hypotheses

H1- Supported: The chemical disinfectants microchem plus and sporicidin, normally used on bacterial spores, did cause complete kill. This supports the idea that spore disinfectants will be more likely to be effective[3].

H2- Rejected: Exposure time did not seem to make a difference on level of kill.

H3- Supported: Chemical disinfectants were more effective than natural disinfectants. The chemical disinfectants resulted in complete kill while the natural disinfectants did not.

Conclusions

- Microchem plus and sporicidin were effective in killing *C. burnetii.* Oxivir five showed killing, however, due to possible contamination, should be tested again.
- While oregano oil did show reduced growth, it would not be currently safe to utilize for *C. burnetii,* but, may have uses in other laboratories.
- Nisin Z + EDTA showed no reduced growth.

Future Directions

- Testing disinfectants on different stock days of *C. burnetii* to see if they are still effective even when different *C. burnetii* cell types are present.
- Effective disinfectants should be tested again at lower time points and concentrations to find a minimum required.
- Nisin Z should be tested on other gram-negative bacteria and gram-positive to determine overall effectivity.
- Ineffective disinfectants could be combined with others or at a higher concentration to see if they will ever result in complete kill.
- Effective disinfectants should be tested against the non-attenuated pathogenic strain of *C. burnetii*[6].

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