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Competitive Inhibition of Methicillin-Resistant Staphylococcus Aureus (Mrsa) By a 4-Strain Lactobacillus Cocktail

L. K. Fuerniss*, C. J. Weissend, A. D. Belk, J. L. Metcalf, M. N. Nair, R. J. Delmore, D. R. Woerner, K. E. Belk, I. Geornaras, and J. N. Martin

Department of Animal Sciences, Colorado State University, Fort Collins, CO, 80523, USA
*Corresponding author. Email: fuerniss@rams.colostate.edu (L. K. Fuerniss)

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Objectives

Antibiotic resistant bacteria are a significant global public health threat. The presence of Methicillin-resistant *Staphylococcus aureus* (MRSA) is a substantial concern in not only human medicine, but also food production. As the mechanisms which promote antimicrobial resistance are better understood, the development of novel strategies to mitigate the presence of resistant bacteria is a priority. Specific to food production, there has been a significant increase in the investigation of solutions which are considered "natural." The objective of this study was to determine the inhibitory effects of a commercially available *Lactobacillus* cocktail against MRSA.

Materials and Methods

Two concentrations of a commercial available probiotic cocktail and 2 concentrations of a 3-strain MRSA cocktail were combined in Brain Heart Infusion broth and stored at 2 temperatures for 5 d. Three ATCC strains of MRSA (from clinical isolates, community acquired isolates, and hospital acquired isolates) were grown from frozen stocks on Brain Heart Infusion (BHI), combined, and purified to make a MRSA cocktail with a final concentration of 10^9 CFU/mL. Similarly, 4 dehydrated *Lactobacillus* strains were rehydrated in BHI broth and combined in proprietary ratios to formulate a probiotic cocktail with a concentration of 10^{10} CFU/mL. Cocktail One was prepared with 10^9 CFU/mL of probiotic and 10^4 CFU/mL of MRSA; Cocktail 2 was prepared with 10^7 CFU/mL of probiotic and 10^2 CFU/mL of MRSA. Both cocktails were stored at either 25 ± 1°C or 37 ± 1°C. Populations of MRSA and probiotic were monitored regularly over a 5-d period. At each 24-h interval during incubation, samples were removed from each cocktail (1 and 2) and storage temperature (25 ± 1°C or 37 ± 1°C) combination, serially diluted, and plated in duplicate onto 2 selective agars–Baird Parker Agar (BPA) for the quantification of MRSA and De Man, Rogosa, and Sharp Agar (MRS) for quantifying the probiotic. Plates were aerobically incubated at 37°C for 24 h (BPA) or anaerobically incubated at the same temperature for 72 h (MRS). As the selective media for the probiotic (MRS) also supports the growth of *Staphylococcus*, the MRS agar was modified with 0.05% cysteine and 0.002% bromophenol blue. The trial was replicated on 3 separate occasions. Analysis was completed using 2-way, mixed-effects ANOVA models fit separately for each cocktail and each agar; least square means were separated with an α level of 0.05.

Results

MRSA populations decreased ($P < 0.05$) in Cocktail One, regardless of storage temperature, and were undetectable after 5 d of storage at 37°C. Conversely, MRSA populations increased ($P < 0.05$) by up to 3 log CFU/mL in Cocktail Two at both temperatures. Probiotic populations in Cocktail One decreased ($P < 0.05$) by up to 2 log CFU/mL over the storage period at either temperature; probiotic populations in Cocktail 2 increased ($P < 0.05$) up to 1.5 log CFU/mL.

Conclusion

Mitigating the presence of MRSA in the environment and in food production facilities can have a significant impact on public health. These results suggest that probiotics could be used as effective inhibitors of MRSA. Additional research is needed to evaluate the efficacy of this probiotic cocktail on the presence of MRSA in an applied setting.

