

## **Microbial Adaptation to Natural and Synthetic Antimicrobials: Implications for Risk Assessment and Understanding the Evolution of Resistance to Antimicrobials**

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### **ABSTRACT**

This research explores the antimicrobial properties of pure frankincense oil (FKO) and investigates microbial adaptation to its prolonged exposure, focusing on resistance development and phenotypic changes. Despite FKO's recognized antimicrobial properties, significant knowledge gaps remain regarding its long-term effects on microbial resistance mechanisms and adaptation. With the global rise in antimicrobial resistance (AMR), it is crucial to understand how natural antimicrobials, such as FKO, influence microbial behavior and whether they drive resistance or unique adaptive behaviors over time. The project aims to explore microbial responses to FKO at both genetic and phenotypic levels, using *Escherichia coli* as a model organism. Initially, the antimicrobial activity of FKO is confirmed through well diffusion assays, followed by determining the Minimum Inhibitory Concentration (MIC) to establish its potency. A selection experiment exposes *E. coli* to sub-lethal concentrations (6.25% v/v) of FKO over several passages to simulate prolonged exposure. The evolved bacteria demonstrate an increase in MIC values compared to the ancestral strain, suggesting the development of resistance. Genomic sequencing of the adapted bacteria will reveal mutations or other genetic changes responsible for resistance, providing deeper insight into the underlying mechanisms of adaptation. In addition, phenotypic assays will be conducted to assess potential cross-resistance to biocides, antibiotics, and other essential oils, to evaluate the broader implications of prolonged exposure to FKO on antimicrobial resistance patterns. This research will contribute to a more comprehensive understanding of FKO's antimicrobial efficacy and its role in the evolution of microbial resistance. It will also offer valuable insights for risk assessment and inform the safe application of natural antimicrobials in clinical and environmental settings. By integrating antimicrobial testing, genomic analysis, and adaptation mechanisms, the study will advance the knowledge of how natural antimicrobials shape microbial resistance in both clinical and ecological contexts.

