

A Multi-Factor Study Deriving a Comprehensive Evaluation of Natural Antibiotic Hybrids Bound to Novel Nanoreactors in an Inorganic Model of an Enterococcus faecalis Infected Jejunum Cross-Analyzed through Static Biofilm Production and Quorum Sensing (Year 5)

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The foundation of this 5-year study has matured from the growing threat of antibiotic resistance and the need for a swift and cost-efficient endogenous stimulus-powered novel drug-delivery mechanism. The research framed synthetic antibiotics and natural antibiotic hybrids in two-type drug-delivery methods against an *Enterococcus faecalis* infected model of the jejunum. The natural antibiotic hybrids entailed concentrations amongst the Piper betel leaf (betel leaf, BL) and Azadirachta indica (neem, N); and BL and Ocimum tenuiflorum (holy basil, HB). Furthermore, amoxicillin represented the synthetic antibiotic group. Data and statistical analysis were conducted to isolate deviations in strain spread, colony growth, and chemical signaling (through biofilm production and quorum sensing or QS through UV-Vis spectrophotometry). Procedures of image analysis (pixel dimensional-analysis) with the cataloging of microbial growth and quantification procedures of QS and biofilm growth validated the hypothesis. Such procedures accompanied series of MANOVA, ANOVA, and paired t-tests (with inferential statistics). The data portrayed that treatments in nanoreactors acquiescent with a 21.875% concentration of Piper Betel leaf illustrated a significant decay of colony dimensions. The average colony size, spread, quantity, biofilm production, and QS assay data of 21.875% BL/HB was 2.121 mm $\pm$ 0.002, 16.232 cm² $\pm$ 0.018, 2453 $\pm$ 1.85, 0.825 OD $\pm$ 0.014, and 0.826 OD $\pm$ 0.011, respectively (significantly lower than without treatment). Representative of the inferential statistics, all of the natural antibiotic hybrids presented a statistically major deviation compared to the synthetic stock group.

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