## Synergistic antimicrobial action between food processing methods and food-grade compounds

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We hypothesized that it is possible to accelerate microbial inactivation rate in foods through accumulation of complementary stresses within bacteria by a rational combination of food processing methods and food-grade compounds. We explored synergistic antimicrobial effect between mild natural antimicrobial compounds such as gallic acid, curcumin, cinnamaldehyde and eugenol and three processing methods: ultraviolet (UV)-A light, low and high frequency ultrasound and mild thermal treatments. We also developed mechanistic understanding behind improved microbial inactivation rate. Model foods we tested are fresh produce, liquid beverages and low moisture foods such as powdered infant formula. The bacteria we tested include Escherichia coli O157:H7, Listeria innocua, Salmonella Typhimurium and Cronobacter sakazakii. A combination of ultraviolet-A light with natural compounds significantly accelerates microbial inactivation rate in liquid medium as well as fresh produce surface with as much as 3-5 log reductions within 20 minutes. The antimicrobial treatment is also effective against biofilm. The synergistic antimicrobial action depends on membrane pemeablization by UV-A light that increases the update of antimicrobial compounds, which in-turn produces cascade of antimicrobial effects due to accumulation of oxidative stress. Similar acceleration in microbial inactivation was also observed by combining low (20 kHz) or high (1 MHz) frequency ultrasound with natural compounds on biofilms, liquid foods and fresh produce. We found that the antimicrobial mechanism differed significantly between two ultrasound wavelengths, with low frequency primarily causing membrane damage and high frequency primarily producing oxidative stress within cells. Interestingly, synergistic effect of thermal treatment with antimicrobial compounds in low moisture foods was highly dependent on the water activity and the macronutrient composition of foods. Subsequent analysis with qRT-PCR and biophysical methods demonstrated that bacteria responds to stresses from low water activity and antimicrobial treatment by changing its membrane fatty acid composition that lowers its fluidity, reducing its metabolic activity that lowers uptake of antimicrobial compounds and altering the expression of several stress-response genes such as those responsible for producing osmoprotectants. Thus, our presentation will describe the opportunities and limitations of our approach and advance the understanding of how bacterial survival strategies affect efficacy of synergistic antimicrobial treatments.