Title	Investigation on factors influencing ultrasound-assisted surface decontamination of fresh and fresh-cut vegetables
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Abstract

Although strict practices for controlling the safety of leafy green produce have been implemented in the produce industry, current commercial operations rely on a wash treatment with an antimicrobial agent as the only step for reducing microbial populations on fresh produce. In this study, a systematic approach, starting from understanding the interactions among acoustic energy, produce, sanitizers, and bacteria, as well as the distribution of the ultrasonic field in the treatment channel, was employed to find the answer to the important question: "Can an ultrasound-assisted treatment indeed enhance microbial reduction in a continuous-flow pilot-scale produce washing system?"

The first question that has to be answered for any ultrasound-assisted produce wash is if ultrasound as a form of physical energy will cause produce quality degradation. To address this issue, a determination was made of the threshold of acoustic power density (APD) in an ultrasound treatment allowable without causing unacceptable produce damage immediately after sonication. All subsequent produce wash tests were performed with an APD below this threshold. Next, the effect of ultrasound on the reduction of *Escherichia coli* O157:H7 inoculated on the surfaces of selected produce was measured. The interactions between ultrasound and sanitizers were examined by monitoring the concentration changes of the sanitizers in washing solutions used for fresh-cut lettuce washes. The effects of ultrasound parameters and operational conditions on the uniformity of the acoustic field in the ultrasonic channel, and on surface decontamination of leafy green produce, were also examined. A continuous-flow pilot scale ultrasonic washing system was designed, fabricated, and tested in this study. Further, produce surface characteristics and infiltration pathways were studied. Finally, the application of ultrasonication for surface decontamination of lettuce coring knives was also investigated.

In the tests with the pilot-scale ultrasonic produce washer, ultrasound in combination with chlorine enhanced the reduction of *E. coli* inoculated on spinach by a 0.53 log cycle for batch-leaf washes. Additionally, batch-leaf washes with chlorinated water enhanced the reduction of aerobic plate count by a

0.50 log cycle over a chlorine-only wash. No significant effect of ultrasound was observed for yeast and mold reduction. Ultrasonication enhanced the removal of *E. coli* O157:H7 from baby carrots by 1.24 log cycles and 0.65 log cycle when treated for 1 min and 3 min, respectively. A batch wash with ultrasound also significantly ($p < 0.05$) increased the aerobic plate count reduction for roma tomato surfaces by a 0.75 log cycle over a sanitizer-only wash, while there was only a marginal enhancement in yeast and mold reduction.

A number of new techniques were developed or applied to elucidate the relationships between produce surface characteristics and microbial attachment and removal. In the inactivation tests, the underside of spinach leaves (rough side) was found to provide a better shelter for *E. coli* O157:H7 than the upside. Scanning electron microscope (SEM) and optical profiler mapping were used to provide some insight into this observation. The SEM images and surface profiles of a spinach leaf showed valleys, deeper in the underside than in the upside, harboring and even protecting the cells attached in them. In the effort to quantify the surface roughness of produce, a freeze-drying sample preparation method for confocal laser scanning microscopy was developed which worked well. There were no significant differences in surface roughness among selected vegetables. Moreover, the surface hydrophobicity of the inner surface of spinach leaves was not significantly different from that of the outer surface, and was not affected by ultrasonication. For the first time, the MicroXCT technique was employed in this study to examine the internal structure of a produce sample on the micrometer scale. The MicroXCT images provided a detailed view of the potential infiltration pathways inside a produce tissue that may be used by human pathogens to penetrate fresh and fresh-cut produce. (Abstract shortened by UMI.)