Blue 470 nm Light Suppresses the Growth of Salmonella enterica and Methicillin-Resistant Staphylococcus aureus(MRSA) In Vitro

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Background and Objective: Emerging evidence suggests that blue light can photo-inactivate some bacteria of clinical importance. Consequently, we tested the hypothesis that 470 nm light can suppress growth of two recalcitrant bacteria, MRSA and Salmonella.

Materials and Methods: We plated 5×10^6 and 7×10^6 CFU/ml USA300 strain of MRSA and 1×10^6 CFU/ml of Salmonella enterica serovars Typhimurium and Heidelberg. Plates were irradiated with 55, 110, 165 and 220 J/cm² of blue light, incubated at 37°C for 24 hours and colony counts determined.

Results: Compared with controls, blue light irradiation produced a significant dose-dependent reduction in the number of colonies formed by each bacterial strain (P < 0.001). Irradiation of 5×10^6 and 7×10^6 CFU/ml MRSA with 55 J/cm² produced 92% (4.6×10^6 CFU/ml) and $86\% (6 \times 10^6 \text{ CFU/ml})$ inactivation respectively, while 110 and 220 J/cm² suppressed each MRSA density 100%. Irradiation of Salmonella Typhimurium with 55 and 110 J/cm^2 suppressed bacterial growth 31% (3.1×10^5 CFU/ml) and 93% $(9.3 \times 10^5 \text{ CFU/ml})$ respectively; while Salmonella Heidelberg was inhibited $11\%~(1.1 imes 10^5~{
m CFU}/$ ml) and 84% $(8.4 \times 10^5 \text{ CFU/ml})$ respectively by the two fluences. Complete inactivation of each Salmonella strain was achieved using 165 or 220 J/cm².

Conclusion: The observed inhibition of Gram-positive (MRSA) and Gram-negative (Salmonella) bacteria suggests the versatility of blue light in bacteria eradication, making it a viable intervention strategy for decontamination of food and environments that harbor such bacteria. Lasers Surg. Med. 47:595-601, 2015.

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Key words: bacterial suppression; blue light; methicillinresistant Staphylococcus aureus; phototherapy; Salmonella enterica Heidelberg; Salmonella enterica Typhimurium

INTRODUCTION

bacteria—methicillin-resistant Staphylococcus aureus (MRSA)-have emerged as a major health concern, causing periodic epidemics worldwide and accounting for approximately 94,000 invasive infections that result in 18,650 deaths yearly in the United States alone [1,2]. Recent estimates suggest that MRSA infection now accounts for 44% of all hospital associated infections in the United States [3]. Of these, community associated MRSA (CA-MRSA), that is, the bacterial strain often acquires from food, person to person contact and from other environmental sources, accounts for as many as 92% [4]. Outbreaks of CA-MRSA have been on the rise [4–6], with pulsed-field type USA300 strain often isolated during such epidemics [7,8]. Consumption of foods contaminated with S. aureus or its antibiotic resistant variant, MRSA, can cause severe toxinmediated illness including gastroenteritis, nausea, vomiting, diarrhea, and abdominal pain [9]. Other disease processes associated with MRSA include skin and soft tissue infections, sepsis, and osteomyelitis [10-12].

Whereas MRSA is a Gram-positive bacterium, Salmonella are facultative anaerobic, Gram-negative, oxidase-negative, rod-shaped bacteria consisting of two prominent species, Salmonella enterica and S. bongori. Worldwide, about 2,400 Salmonella serovars have been identified in humans and farm animals [13,14]. Salmonella infections cause more hospitalizations and deaths than any other food pathogen, and more than one million Americans contract salmonellosis every year due to contaminated food consumption [15]. Outbreaks of S. Heidelberg are common, and particularly associated with consumption of infected chicken, turkey, pork, eggs, milk and cheddar cheese [16]. Animal and human fecal contamination of water and soil is also known to be a

Staphylococcus aureusis a Gram-positive, coagulase positive bacterium commonly found on human skin and in the natural flora of the human nasal passages. In recent times, antibiotic resistant variants of this common

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Fig. 7. Representative culture plates of Salmonella enterica serovar Heidelberg irradiated with 55, 110, 165, and $220 \text{ J/cm}^2 470 \text{ nm}$ light.

some cases, terminated outright, because of reports which suggest that they may be carcinogenic. In addition, bacterial resistance to available chemical sensitizers has been on the rise [44]. At the same time, increasing consumer awareness continues to pressure the food industry to provide the public with fresh microbiologically safe fresh-cut produce; prompting the industry to seek new ways to minimize the use of chemicals concurrently as efforts are made to the reduce the presence of pathogens in food [45]. These emerging trends strongly underscore the significance of our findings that MRSA as well as *S. enterica* serovars Typhimurium and Heidelberg are susceptible to 100% photo-inactivation with 470 nm blue light.

Our results further imply that 470 nm light may serve as an excellent alternative to common traditional food preservative techniques, such as chlorine and hydrogen peroxide solutions used for fresh produce, making blue light a promising, non-thermal and environmentally friendly disinfection technology for freshly cut produce. Other areas of potential application include the suppression of cutaneous MRSA infections by CA-MRSA and HA-MRSA and decontamination of environments that harbor bacteria.

CONCLUSION

Blue 470 nm light inhibits the growth of both Grampositive (MRSA) and Gram-negative (*Salmonella*) bacteria suggesting the versatility of blue light in bacteria eradication. Complete (100%) growth suppression of both bacteria is achieved *in vitro*, using 220 J/cm² energy fluence. These findings suggest that blue 470 nm light is a viable alternative to current methods being used to stem the global epidemic of MRSA and *Salmonella*.

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