Supplementary material

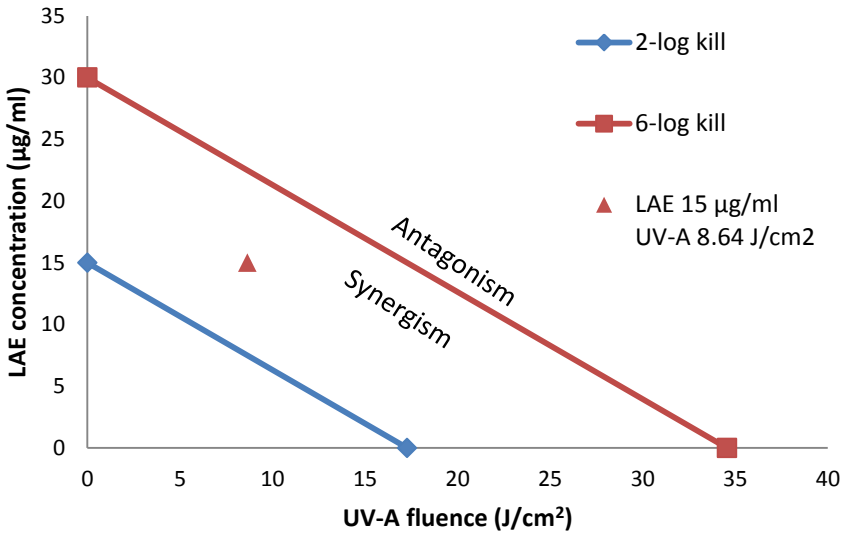
Synergistic combination between LAE and light or mild heat with isobolograms. The results in supplementary Fig. S.1 illustrate the isobole lines for achieving 2-log and 6-log inactivation of *Escherichia coli* using LAE and UV-A light as well as the synergistic antimicrobial combination applied in this study. The y-axis provides the LAE concentration alone required for inactivation of *E. coli* for a fixed time interval. Similarly, intersection of these lines on the x-axis, illustrate the fluence (energy per unit area) of UV-A light required for specific levels of inactivation of target bacteria using UV-A treatment alone. Fluence (J/cm^2) was calculated as a function of time (seconds) multiply by average light intensity for four UV-A lamps (4.8 ± 0.1 mW $\cdot$ cm^{-2}). The results demonstrate that the combination of LAE and UV-A is synergistic compared to individual treatments. Furthermore, the synergistic combination requires half the concentration of LAE and 4-fold lower fluence level of UV-A light to achieve the same 6 log reduction of the inoculated *E.coli* using individual treatments.

The results in supplementary Fig. S.2 illustrate isobole lines for the inactivation of bacteria using heat or LAE. These isobole lines were generated by measuring the individual bactericidal activity of LAE as a function of concentration for a fixed time interval and for the total thermal energy input in the heating equipment. This energy (kJ) was calculated based on the product of the heat capacity of the water bath (1200 W) and treatment time (s) at 55 ºC to inactivate bacteria. The results demonstrate that the combination of LAE and mild heat is synergistic compared to individual treatments. Furthermore, the synergistic combination requires 2.3-fold less concentration of LAE and over 27-fold less thermal energy required to achieve the same 5-log reduction of the inoculated *E.coli* using individual treatments.

Oxidative stress generation from polymyxin B and LAE. Both LAE and polymyxin B were analyzed for their bactericidal activity against *E. coli* O157:H7 at room temperature. As shown in

Fig. S.3a, LAE (15 ppm) demonstrated significant ($P < 0.05$) bactericidal activity (1.5 log reduction) comparing to inoculum size during 30 min incubation, while polymyxin B did not. The LAE bactericidal activity was further neutralized ($P < 0.05$) by glutathione, a common antioxidant: this result implied that the rapid LAE bactericidal activity was related to its oxidative stress genesis. To further confirm this hypothesis, LAE was tested again for its bactericidal activity at 4 °C, since metabolic activity of cells will be slowed down at low temperature, which resulted in attenuated oxidative damage. As shown in Fig. S.3b, no bactericidal activity was observed when *E. coli* cells were treated at 4 °C, indicated that LAE bactericidal activity was correlated with oxidative stress generation. Investigation of oxidative stress generation from polymyxin B was also confirmed by incubating *E. coli* O157:H7 with polymyxin B for 2 hours. As shown in Fig. S.3c, polymyxin B demonstrated significant ($P < 0.05$) bactericidal activity comparing to inoculum size, regardless of glutathione supplementation. In comparison, adding glutathione did not neutralize the bactericidal activity, indicating oxidative stress was not generated from polymyxin B.

List of Figures



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52 **FIG S1**

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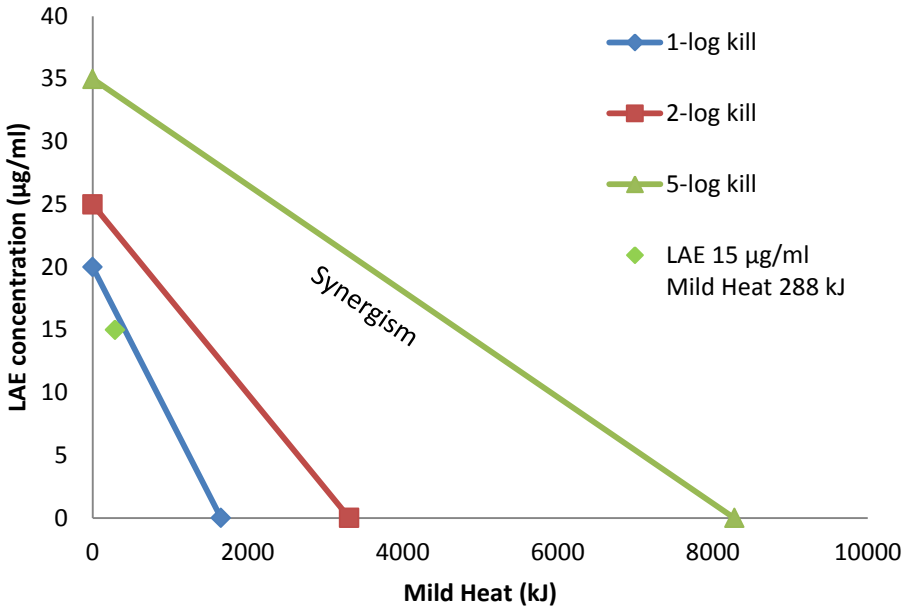

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55 **FIG S2**

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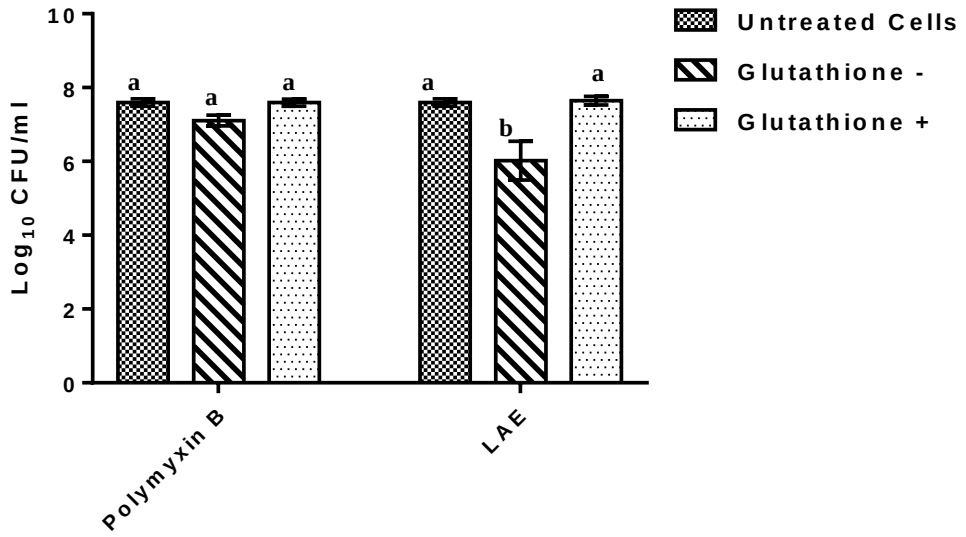
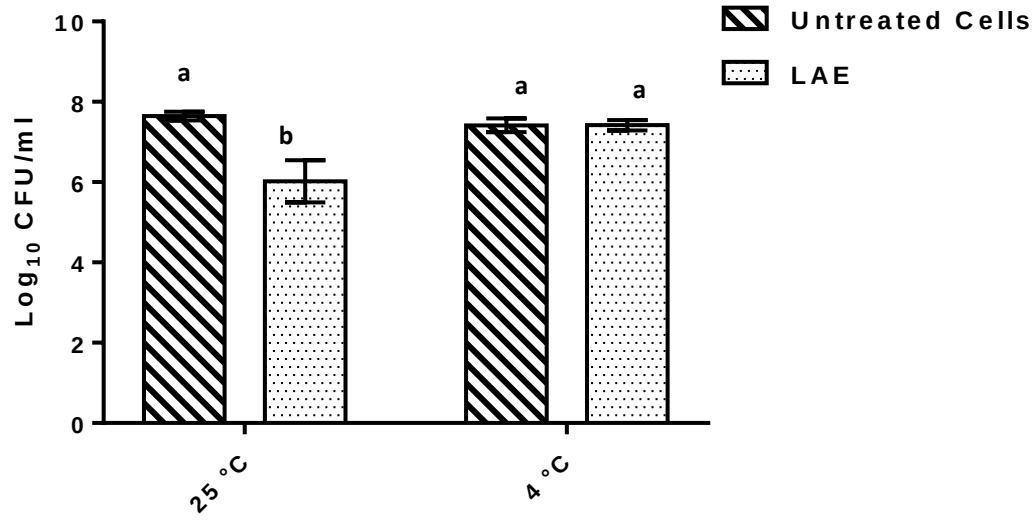
S.3a


Untreated Cells
Glutathione -
Glutathione +
Log10 CFU/ml
10
8
6
4
2
0
a
a
a
a
b
a
Polymyxin B
LAE

S.3b


Untreated Cells
LAE
Log10 CFU/ml
10
8
6
4
2
0
a
b
a
a
25 °C
4 °C

S.3c

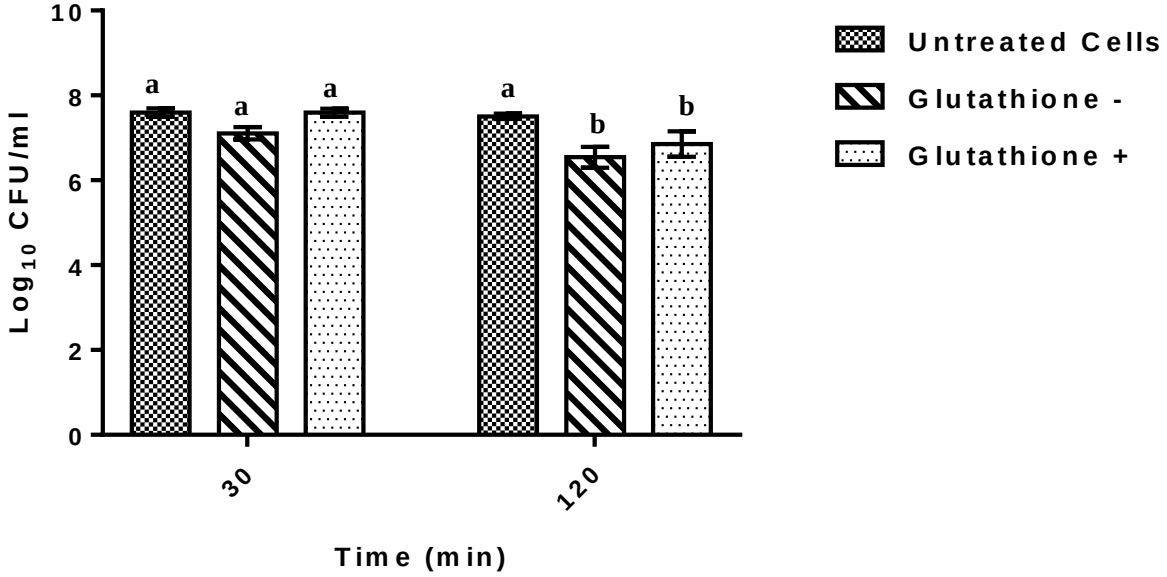

10
8
6
4
2
0
Log10 CFU/ml
a
a
a
a
b
b
30
120
Time (min)
Untreated Cells
Glutathione -
Glutathione +

FIG S3