

Electronic Supplementary Information

**Sonophotocatalysis mediated morphological transition modulates  
virulence and antibiotic resistance in *Salmonella* Typhimurium**

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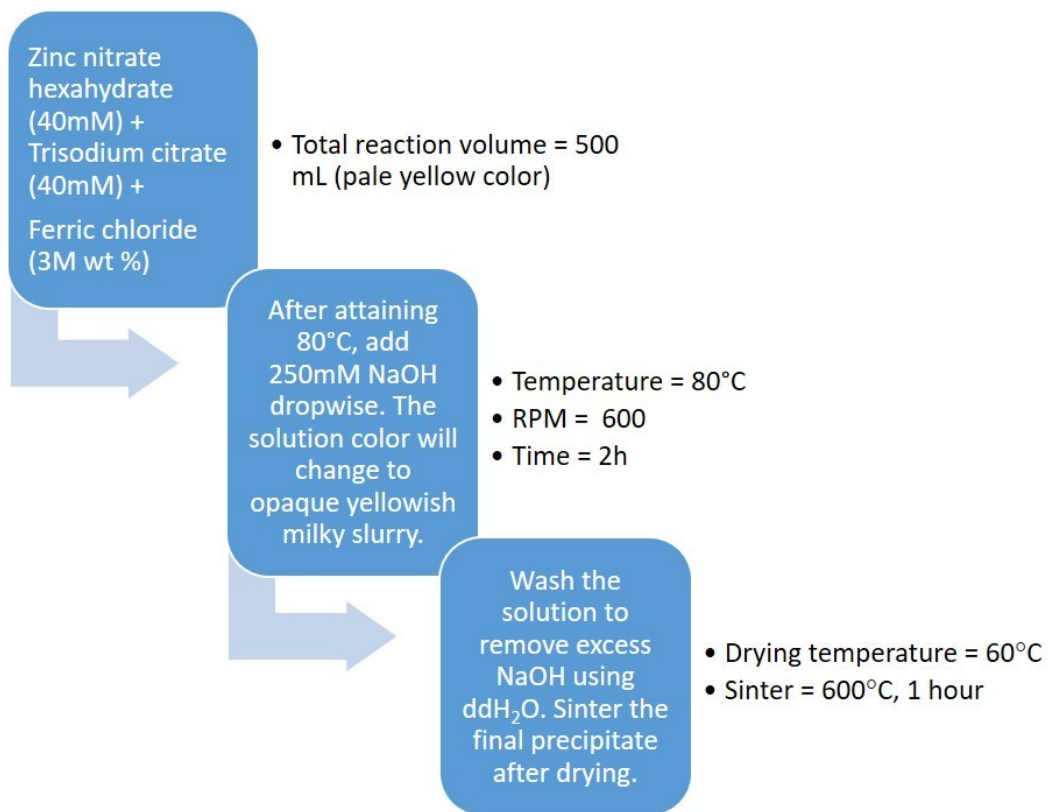
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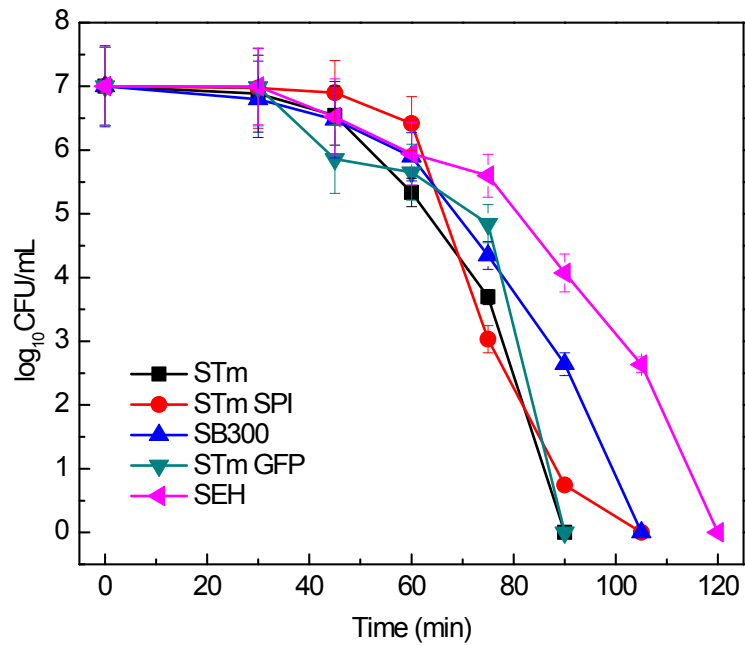
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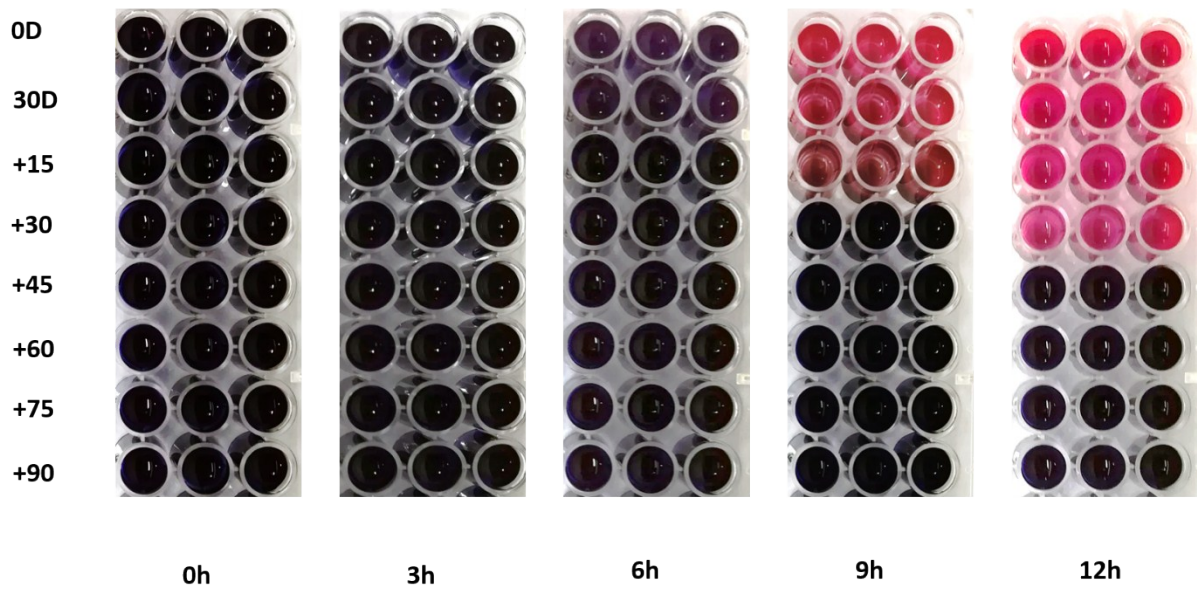
### Synthesis of Fe-ZnO NPs:



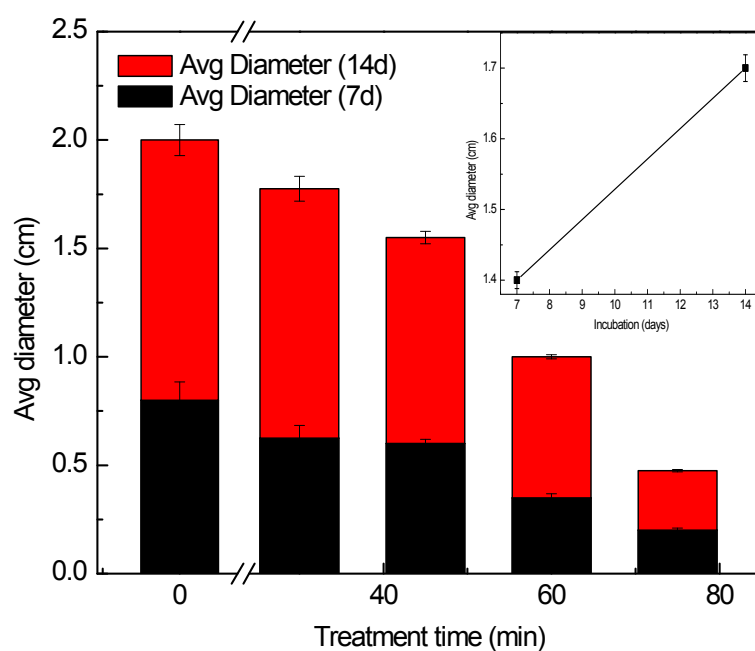
**Supplementary Scheme. 1** Flowchart of Fe-ZnO NPs synthesis



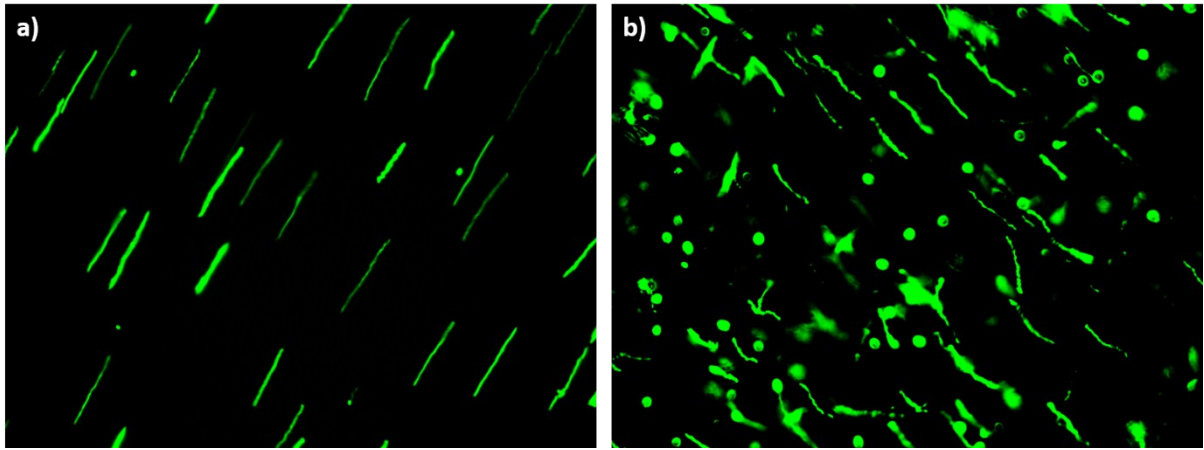
**Fig. S1.** Efficacy of SPC disinfection against different mutants of *S. enterica*. STm- antibiotic resistant mutant, STm SPI- Salmonella pathogenicity island induced STm, SB300- virulent strain of wild type *S. Typhimurium*, STm GFP- Green fluorescence protein tagged mutant, SEH- Hydrogen peroxide resistant mutant of *S. Enteritidis*. (Bacterial loading  $\approx 10^7$  CFU/mL, Fe-ZnO NPs loading = 200 mg/L, Temperature= $35\pm 2^\circ\text{C}$ ). Error bar indicates the standard deviation of replicates (n=3).



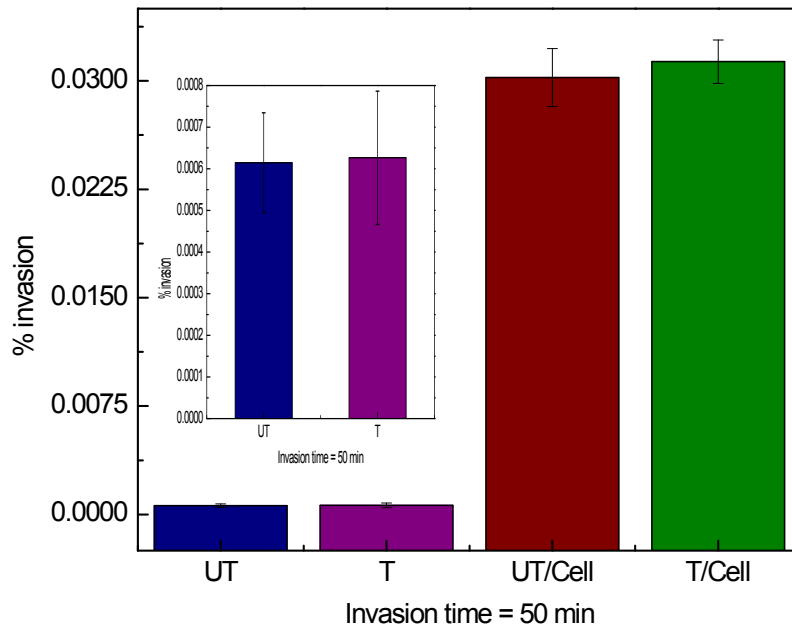
**Fig. S2.** Time dependant analysis of resazurin metabolic assay of STm sampled at different time points of SPC disinfection. Each reaction was performed in triplicate. (Bacterial loading  $\approx 10^7$  CFU/mL, Fe-ZnO NPs loading = 200 mg/L, Temperature =  $35\pm 2^\circ\text{C}$ ).



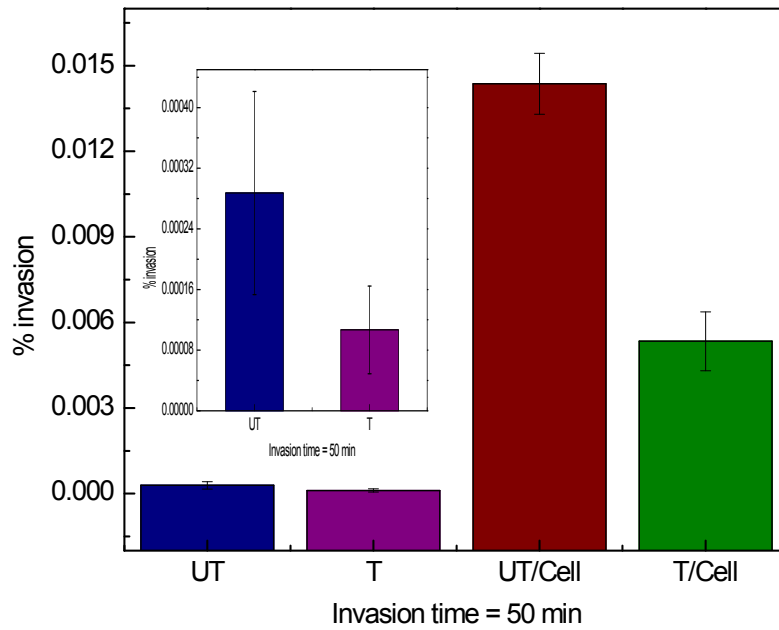
**Fig. S3.** Average colony diameter of STm sampled at different time intervals of SPC disinfection. Diameter was measured after 7 and 14 days of incubation at 37°C. Inset represents the change in diameter of untreated STm. (Bacterial loading  $\approx 10^7$  CFU/mL, Fe-ZnO NPs loading = 200 mg/L, Temperature =  $35 \pm 2^\circ\text{C}$ ). Error bar indicates the standard deviation of replicates ( $n=3$ ).



**Fig. S4.** (a) and (b) shows fluorescent images of GFP tagged untreated and treated (recovered after sublethal SPC - +45 min) STm captured during bacterial motion in the prepared slides, depicting the (a)absence and (b) presence of spheroplast like cells respectively.

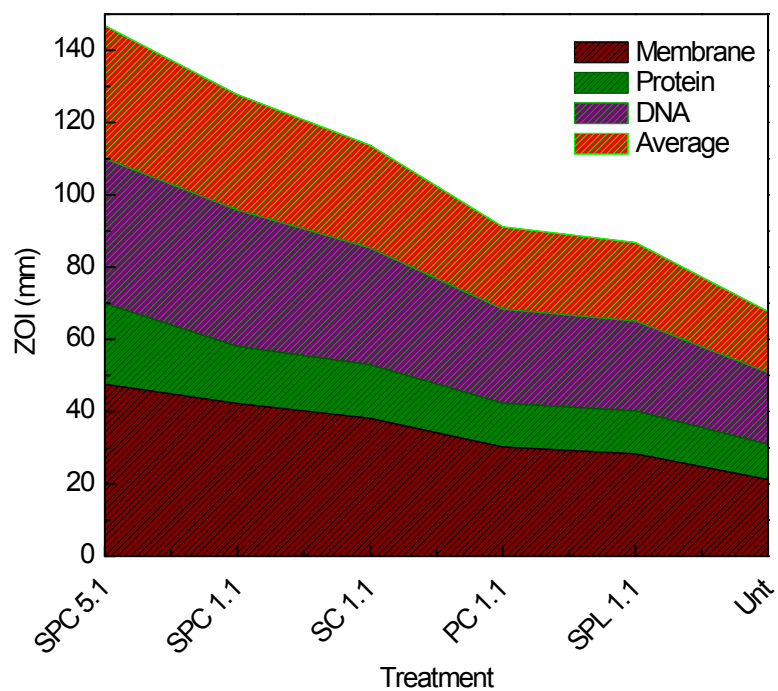


**Fig. S5.** Invasion assay of untreated (UT) and treated (T) STm after sublethal SPC (sampled after +30 min of SPC) in HCT116 colon epithelial cells at MOI=10. Inset represents the difference in total invasion by UT and T STm. Error bar indicates the standard deviation of replicates (n=3).

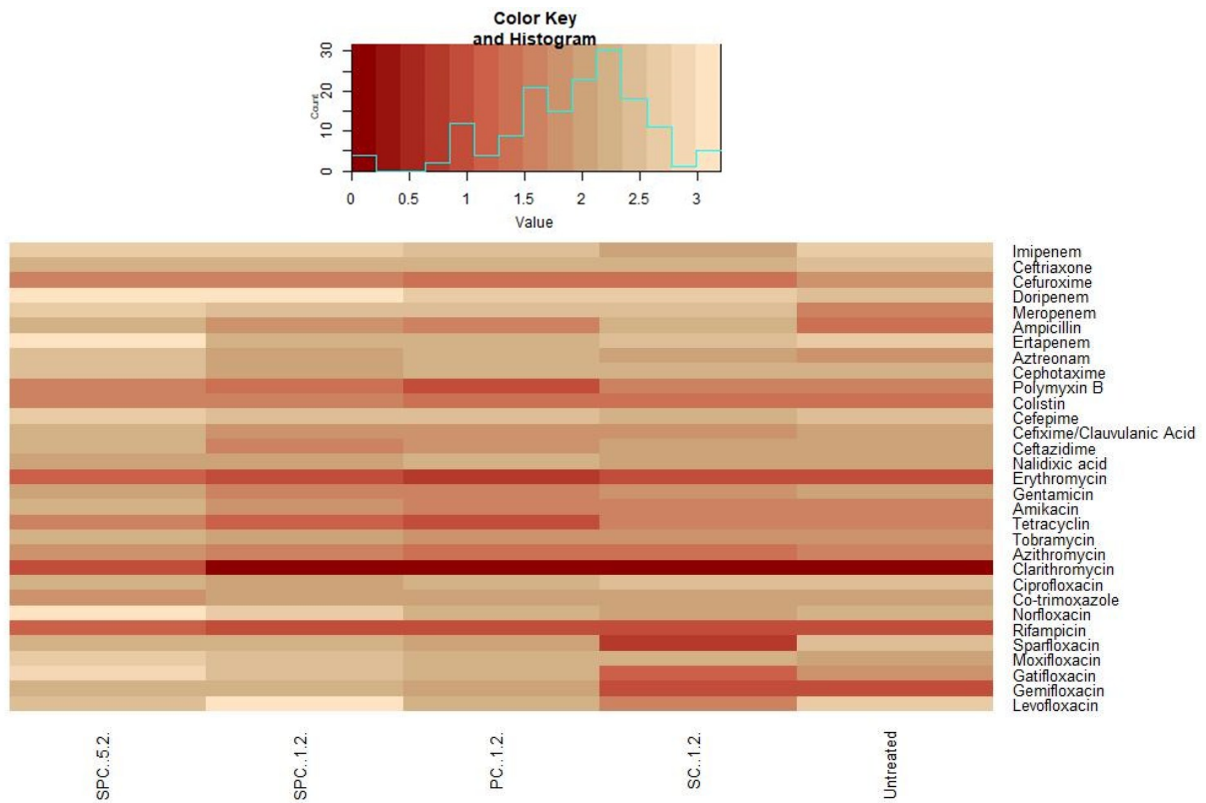


**Fig. S6.** Invasion assay of untreated (UT) and SPC treated SPI induced (T) STm (sampled at +45 min of SPC) in HCT116 colon epithelial cells at MOI=10. Data represents the survived bacterial cells during invasion. Inset represents the difference in total invasion by SPI induced UT and T STm. Total invasion time = 50 min. Error bar indicates the standard deviation of replicates (n=3).

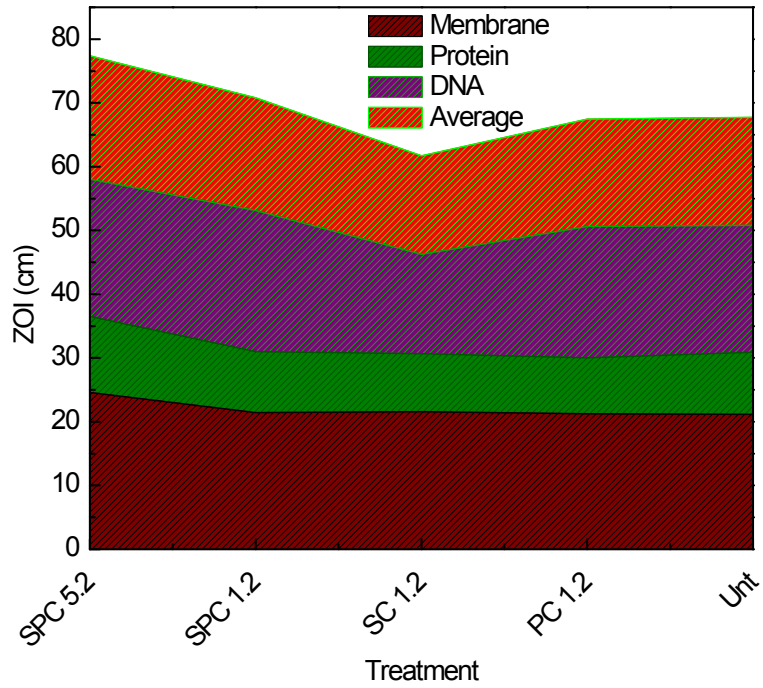




**Fig. S7.** ZOI of antibiotics clustered according to their target (membrane, protein and DNA) after 45min treatment of SPC 5.1 (Sonophotocatalysis 5<sup>th</sup> cycle), and 1<sup>st</sup> cycle of SPC (Sonophotocatalysis), SC (Sonocatalysis), PC (Photocatalysis), SPL (Sonophotolysis) compared with untreated STm. ZOI is measured excluding the diameter of antibiotic disc = 6mm.



**Fig. S8.** Antibiotic resistance/susceptibility profiling of STm recovered after SPC 5.2 (Sonophotocatalysis 5<sup>th</sup> cycle), and 1<sup>st</sup> cycle of SPC (Sonophotocatalysis), PC (Photocatalysis), SC (Sonocatalysis) compared with untreated STm.



**Fig. S9.** ZOI of antibiotics clustered according to their target (Membrane, Protein and DNA) of STm recovered after SPC 5.2 (Sonophotocatalysis 5<sup>th</sup> cycle), and 1<sup>st</sup> cycle of SPC (Sonophotocatalysis), PC (Photocatalysis), SC (Sonocatalysis) compared with untreated STm. ZOI is measured excluding the diameter of antibiotic disc = 6mm.

**Table. S1.** Change in fatty acid composition of sublethal SPC treated and untreated STm

| <b>SI No</b> | <b>Fatty acid</b> | <b>Untreated</b> | <b>Sublethal SPC treated STm</b> |
|--------------|-------------------|------------------|----------------------------------|
| 1            | 12:0              | 3.85             | 3.85                             |
| 2            | 11:0 3OH          | 0.43             | 0.37                             |
| 3            | 13:0              | 0.32             | 0.29                             |
| 4            | 14:0 iso          | 0.13             | 0.25                             |
| 5            | 14:0              | 5.03             | 5.07                             |
| 6            | 15:1 iso G        | 0.41             | 0.45                             |
| 7            | 15:0 anteiso      | 3.27             | 2.86                             |
| 8            | 15:1 w5c          | 0.62             | 0.30                             |
| 9            | 14:0 2OH          | 0.77             | 0.67                             |
| 10           | 16:0 N alcohol    | 0.48             | 0.32                             |
| 11           | 16:0 iso          | 0.29             | 0.30                             |
| 12           | 16:0 anteiso      | 0.35             | 0.33                             |
| 13           | 16:1 w5c          | 0.14             | 0.19                             |
| 14           | 16:0              | 24.34            | 24.13                            |
| 15           | 15:0 2OH          | 0.66             | 0.57                             |
| 16           | 17:1 anteiso A    | 0.61             | 0.77                             |
| 17           | 17:0 anteiso      | 2.25             | 1.93                             |
| 18           | 17:0 cyclo        | 6.38             | 6.29                             |
| 19           | 17:0              | 0.97             | 1.01                             |
| 20           | 18:3 w6c (6,9,12) | 0.66             | 0.61                             |
| 21           | 18:0              | 1.64             | 1.36                             |
| 22           | 19:0 cyclo w8c    | 4.32             | 5.02                             |
| 23           | 19:0              | 0.46             | 0.43                             |
| 24           | 20:2 w6,9c        | 0.92             | 0.47                             |
| 25           | 20:1 w7c          | 0.26             | 0                                |

**Table. S2.** List of antibiotics used in the current study and its major target sites.

| SI No | Antibiotic                | Class or Subgroup                | Target site |
|-------|---------------------------|----------------------------------|-------------|
| 1     | Imipenem                  | B-lactam/ Carbapenem             | Cell wall   |
| 2     | Ceftriaxone               | Lactams/cephalosporin            | Cell wall   |
| 3     | Cefuroxime                | Lactams/cephalosporin            | Cell wall   |
| 4     | Doripenem                 | Lactams/Thienamycins             | Cell wall   |
| 5     | Meropenem                 | Lactams/Thienamycins             | Cell wall   |
| 6     | Ampicillin                | Lactams/Penicillins              | Cell wall   |
| 7     | Ertapenem                 | Lactams/Thienamycins             | Cell wall   |
| 8     | Aztreonam                 | Lactams/Monobactams              | Cell wall   |
| 9     | Cephotaxime               | Lactams/cephalosporin            | Cell wall   |
| 10    | Polymyxin B               | Polymyxin/polypeptide            | Cell wall   |
| 11    | Penicillin G              | Penicillins/B-lactam             | Cell wall   |
| 12    | Colistin                  | Cyclic peptides                  | Cell wall   |
| 13    | Vancomycin                | Glyco/cyclic peptides            | Cell wall   |
| 14    | Methicilin                | Carboxylic acids/ dipeptides     | Cell wall   |
| 15    | Cefepime                  | Lactam/4th gen cephalosporins    | Cell wall   |
| 16    | Cefixime/Clauvulanic Acid | Cephalosporins/alpha amino acids | Cell wall   |
| 17    | Ceftazidime               | Lactams/cephalosporin            | Cell wall   |
| 18    | Chloramphenicol           | Amphenicol/ Nitrobenzene         | Protein     |
| 19    | Nalidixic acid            | Diazanaphthalenes                | Protein     |
| 20    | Erythromycin              | Aminoglycosides                  | Protein     |
| 21    | Gentamicin                | Aminoglycosides                  | Protein     |
| 22    | Amikacin                  | Aminoglycosides                  | Protein     |
| 23    | Tetracyclin               | Tetracyclins                     | Protein     |
| 24    | Linezolid                 | Oxazinanones/ Phenylmorpholines  | Protein     |
| 25    | Streptomycin              | Aminocyclitol glycosides         | Protein     |
| 26    | Kanamycin                 | Aminocyclitol glycosides         | Protein     |
| 27    | Clindamycin               | Lincosamide                      | Protein     |
| 28    | Tobramycin                | Aminocyclitol glycosides         | Protein     |
| 29    | Azithromycin              | Macrolides/Aminoglycosides       | Protein     |
| 30    | Clarithromycin            | Macrolides/Aminoglycosides       | Protein     |
| 31    | Ciprofloxacin             | Quinolines                       | DNA         |

|    |                |                            |     |
|----|----------------|----------------------------|-----|
| 32 | Co-trimoxazole | Sulfonamides               | DNA |
| 33 | Norfloxacin    | Quinolines                 | DNA |
| 34 | Rifampicin     | Macrolactams               | DNA |
| 35 | Sparfloxacin   | Quinoline carboxylic acids | DNA |
| 36 | Moxifloxacin   | Quinoline carboxylic acids | DNA |
| 37 | Gatifloxacin   | Quinoline carboxylic acids | DNA |
| 38 | Gemifloxacin   | Quinoline carboxylic acids | DNA |
| 39 | Levofloxacin   | Quinoline carboxylic acids | DNA |