

## Demonstration of nucleic acid repair in viruses after UV disinfection using coliphages as surrogates

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## Acknowledgments

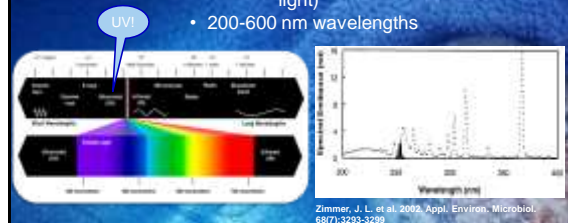
- Connie Chan SMART – Columbia University
- Mentors:
- Karl Linden Ph.D – University of Colorado, Boulder
- Roberto Rodriguez Ph.D – University of Colorado, Boulder
- Sarah Bounty – University of Colorado, Boulder
- And REU program

## Motivation

- Improve water treatment technology
- Develop understanding of high levels of resistance for certain waterborne pathogens

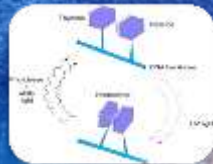
## Background

- Low pressure mercury vapor lamp (monochromatic light)
  - 254 nm wavelength
- Medium pressure mercury vapor lamp (polychromatic light)
  - 200-600 nm wavelengths



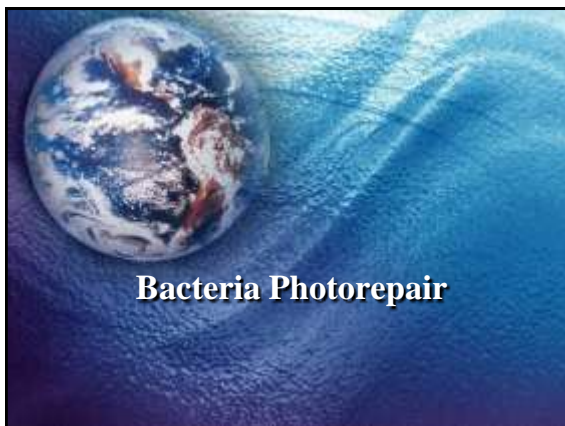
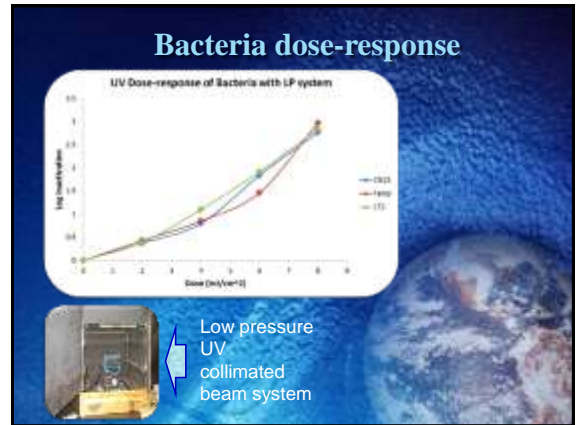
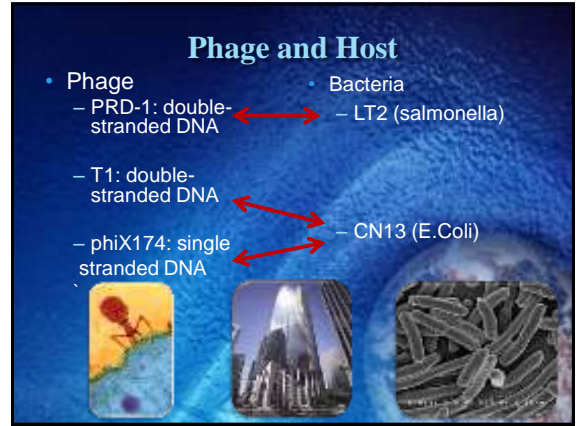
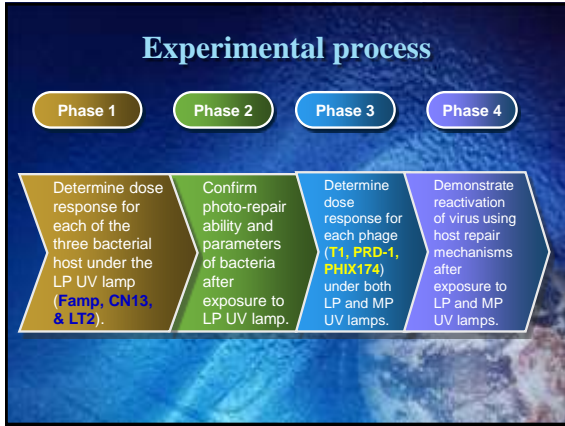
## Background ~ UV damage and repair

- Germicidal wavelength : 200-300 nm
  - Peak DNA absorption at 260 nm
- Photolyase: photo-repairing enzyme
  - Activated at wavelengths 350-450 nm

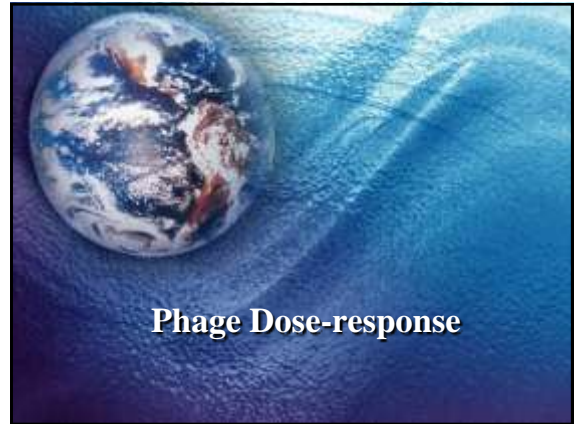
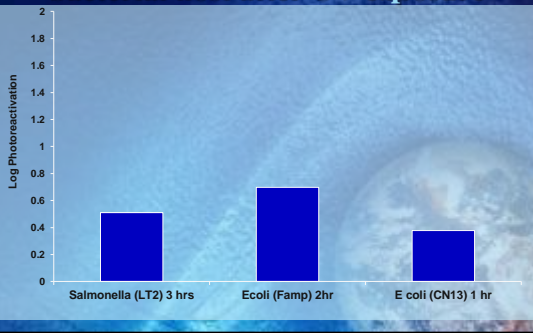


## Objectives

- Determine possible reactivation of viruses utilizing host photorepair mechanisms
- Determine If MP lamps (polychromatic) are more effective than LP lamps (monochromatic) at inactivating phages.



### Maximum photoreactivation of the bacterial host after UV exposure



### Spot plating

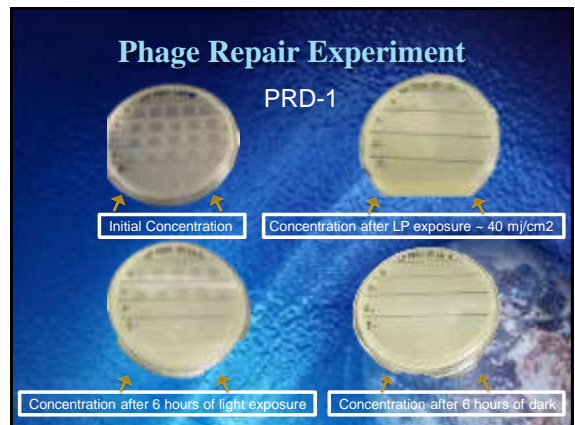
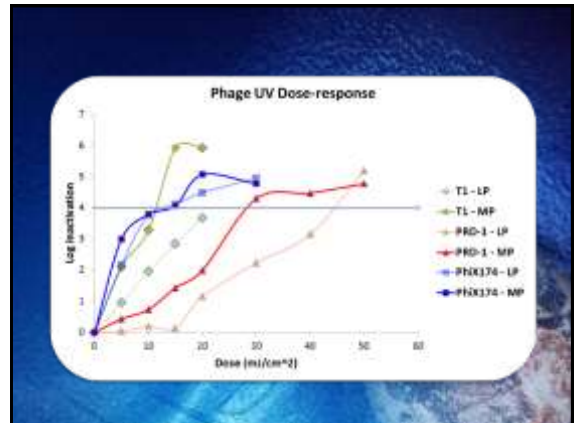
- Saved time and equipment
- 5 dilutions per plate opposed to three plates per dilution



Titering of phage

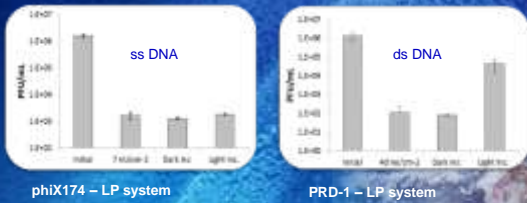
Plating of LT2

Comparison to Double agar layer method:  
 1<sup>st</sup> trial: DAM -  $3.3 \times 10^4$ , 2<sup>nd</sup> trial: DAM -  $5.1 \times 10^9$   
 spot-titer -  $4 \times 10^4$  Spot-titer -  $4.8 \times 10^9$

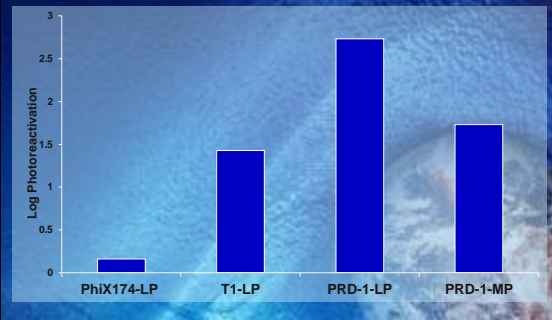


## Phage repair

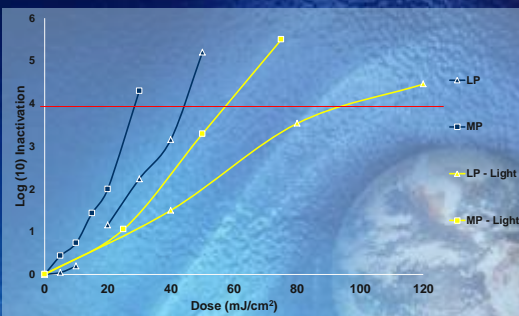
- 6 hours under photo-repair lamp



## Phage photoreactivation



## UV dose-response of phage PRD-1 with and without photoreactivation



## Conclusion

- Phage with double-stranded DNA were able to repair using bacterial host repair-mechanisms, but phage with single stranded DNA were not
- More experiments need to be run to determine if MP lamps are more effective at inactivating PRD-1, T1, and phiX174

## Future work

- Develop dose response curves for Photoreactivation of phage
- Further investigate efficacy of Medium pressure lamps as opposed to Low pressure lamps.

Questions?

## References

Rocky Mountain Laboratories, NIAID, NIH

