

ABSTRACT

INTRODUCTION

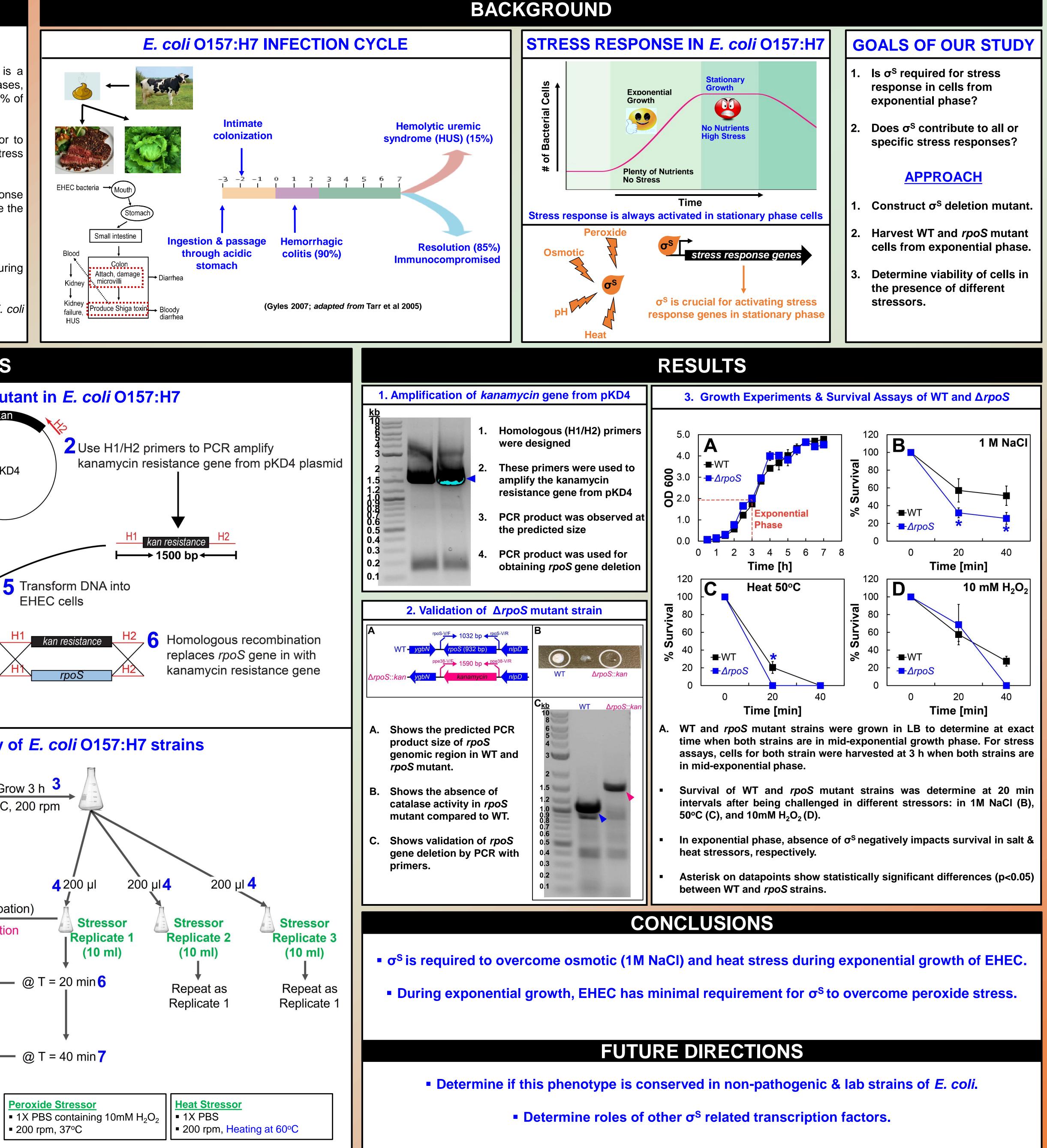
Escherichia coli O157:H7, also known as enterohemorrhagic *E. coli* (EHEC), is a serious food borne pathogen. EHEC causes hemorrhagic colitis in 90% of cases, which then leads to the life threatening hemolytic uremic syndrome (HUS) in ~15% of cases. EHEC is a zoonotic pathogen and humans are an accidental host. A key factor to EHEC being a successful pathogen is due its ability to survive various stress environments in the zoonotic and human hosts. • It is known that the transcription factor σ^{s} is an important activator of stress response during stationary growth phase in EHEC. In this study, we attempted to determine the role of σ^{s} during exponential growth phase in EHEC. RESULTS σ^s is required for survival in the presence of 1M NaCl and high temperatures during exponential growth of *E. coli* O157:H7. σ^s is partially dispensable for survival in peroxide during exponential growth of *E. coli* O157:H7. METHODS **Construction of Gene Deletion Mutant in** *E. coli* **O157:H7** Design H1/H2 primers based on DNA sequence flanking *rpoS* gene in EHEC chromosome pKD4 rpoS **3** Grow EHEC strain Produce Recombinase containing pKM208 plasmid from plasmid pKM208 pKM208 Chromosome Chromosome **Stress Assays to Determine Viability of** *E. coli* **O157:H7 strains** Grow o/n ² Grow 3 h ³ 37°C, 200 rpm 37°C, 200 rpm WT 10 ml LB 5 ml LB or $OD_{600} = 0.05$ **∆**rpoS 450 µl PBS Tubes O T = 0 (before starting stress incubation) Take 50 µl and perform serial dilution 50µl 10-1 Spot 10 µl from each PBS tube onto a 10-2 20hl single LB Plate Take 50 µl, perform -10⁻³ 50µl serial dilution and then **10**⁻¹ 10⁻² 10⁻³ plate on LB 10-4 10-4 **10**⁻⁵ **10**⁻⁶

10-7 10-8	10 ⁻⁵ 10 ⁻⁶ 10 ⁻⁷ 10 ⁻⁷	Take 50 µl, perform ◀ serial dilution and then plate on LB	— @ T =
		 Salt Stressor 1X PBS containing 1M NaCl 200 rpm, 37°C 	Peroxide 1X PBS 200 rpm

The Role of σ^{s} in Stress Response in *E. coli* O157:H7 During Exponential Growth Phase

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