

Comparative Genomics of Soil-persistent *E. coli*

Nicholas Waters

Ashleigh Holmes, Florence Abram, Leighton Pritchard, and Fiona Brennan
Department of Microbiology
School of Natural Sciences
National University of Ireland, Galway

September 26, 2017



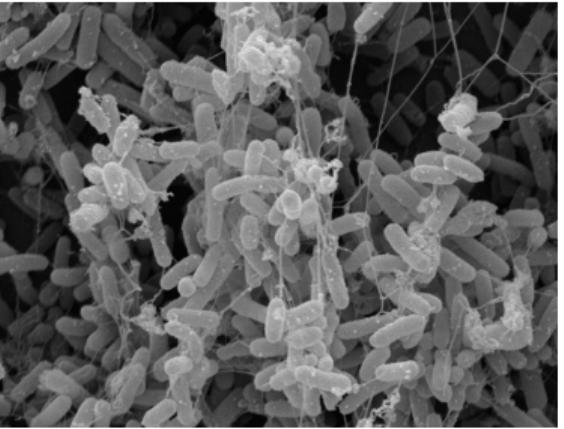
Introduction

- ▶ Overview of soil-persistent *E. coli*
- ▶ Research questions
- ▶ Experimental design
- ▶ Results
- ▶ Next Steps



E. coli: commensal or pathogen?

- ▶ Gram negative
- ▶ Beneficially occurs in the gut
- ▶ Can infect gi and urinary tracts
- ▶ Health burden: ~5 million DALY
- ▶ Used as contamination indicator



¹Image source: NDSU



A Brief History of Soil-persistent *E. coli*

-
- 1886 ···· Escherich: Discovery of *E. coli*
 - 1948 ···· Bardsley: Soil may act as reservoir for *E. coli*
 - 1963 ···· W. and J. Boyd: Cold persistence observed
 - 1972 ···· Evans, et al: Drainage related to coliform counts
 - 1988 ···· Fujioka and Shizumura: Alternative indicators suggested
 - 1997 ···· Texier, et al: Stable populations exist in alpine grasslands
 - 2003 ···· Byappanahalli, et al: Soil persistence is widespread
 - 2010 ···· Brennan, et al: Persistence in maritime temperate soils



Research Questions

Genomic Context

- ▶ Are soil-persistent *E. coli* related?
- ▶ Do soil-persistent *E. coli* possess certain traits?

Virulence

- ▶ Are soil-persistent strains pathogenic?



Experimental Design: Background

Ryan and Fanning: Effects of N and slurry on soils

- ▶ Isolate and protect grassland soil columns
- ▶ Apply treatment, test leachate
- ▶ Last application of slurry in 1998

Brennan, et al: Pathogen survival and transport in soils

- ▶ Two conditions: Cattle slurry and control
- ▶ Compare coliform counts in leachate

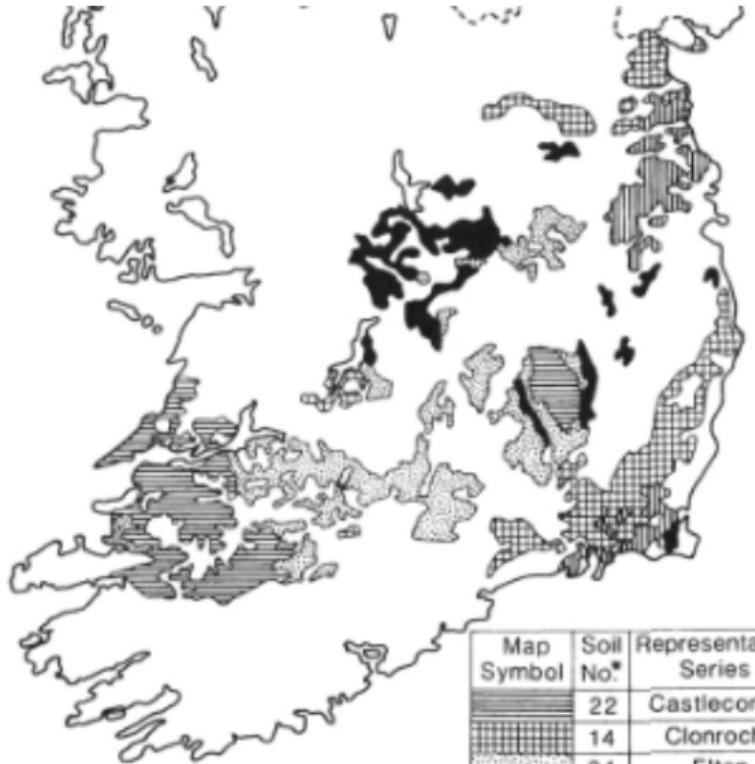


Experimental Design: Current

- ▶ Collect leachate from control lysimeters
- ▶ Enrich for *E. coli*
- ▶ Purify, sequence genomes
- ▶ Compare genomics of soil strains to wider *E. coli* pangenome



Soil Samples



* General Soil Map No's
(2nd edition)

Map Symbol	Soil No.*	Representative Series
Horizontal lines	22	Castlecomer
Diagonal lines	14	Clonroche
Dotted pattern	34	Elton
Black area	30	Oakpark
Vertical lines	40	Rathangan



Lysimeters





Lysimeters





Lysimeters



Lysimeters

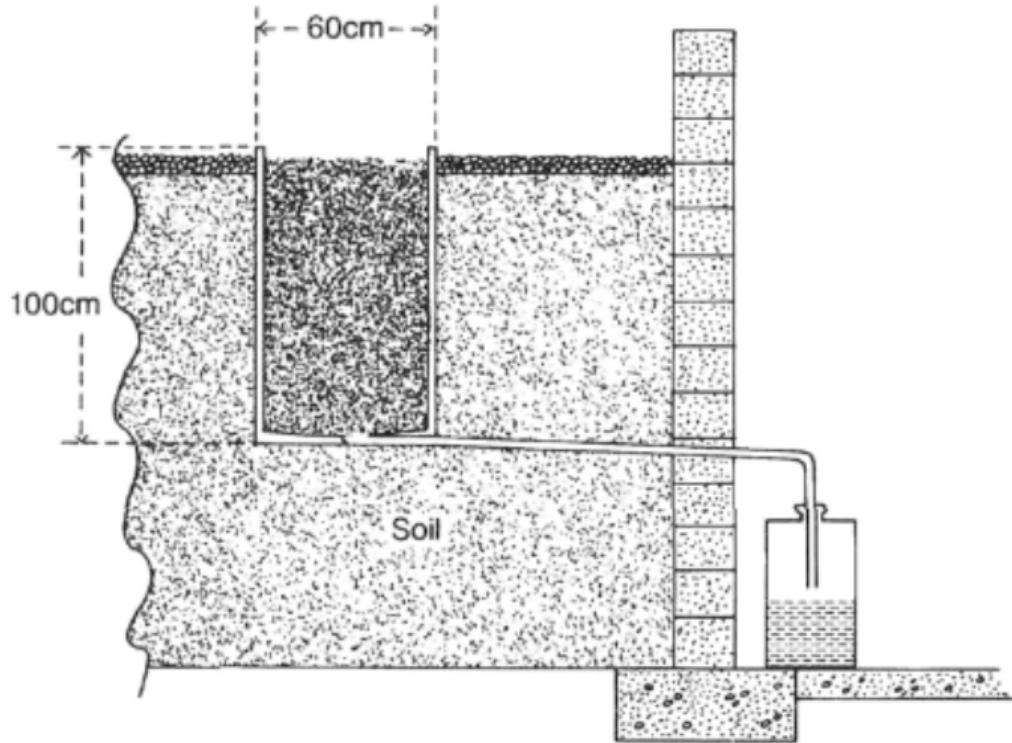
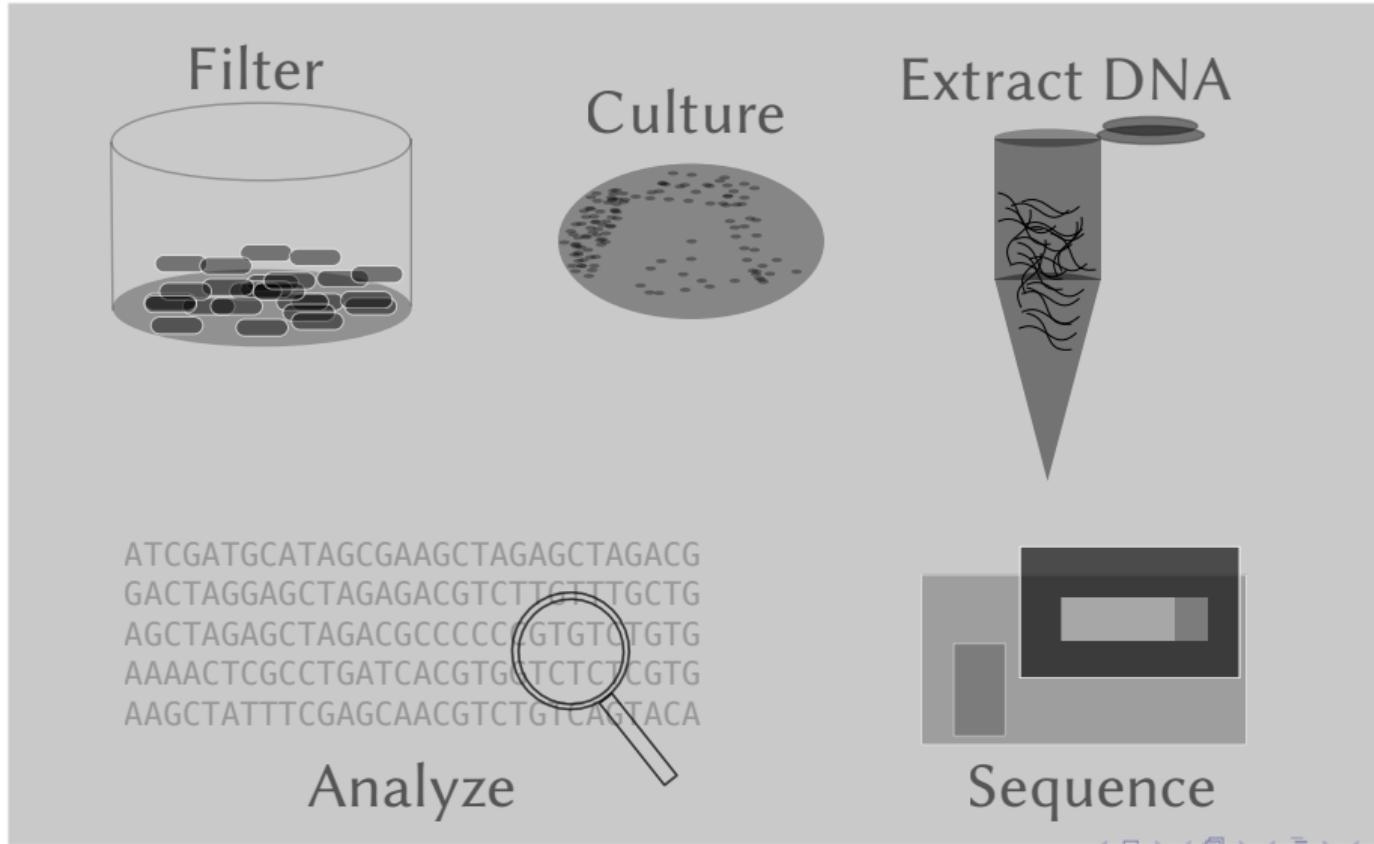


Figure 1: Representation of soil monolith in the lysimeter unit.



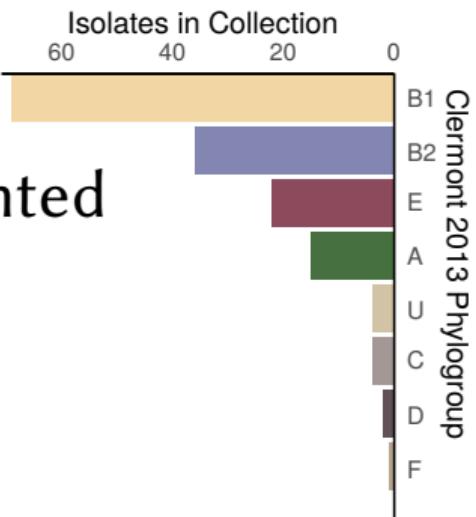
Workflow





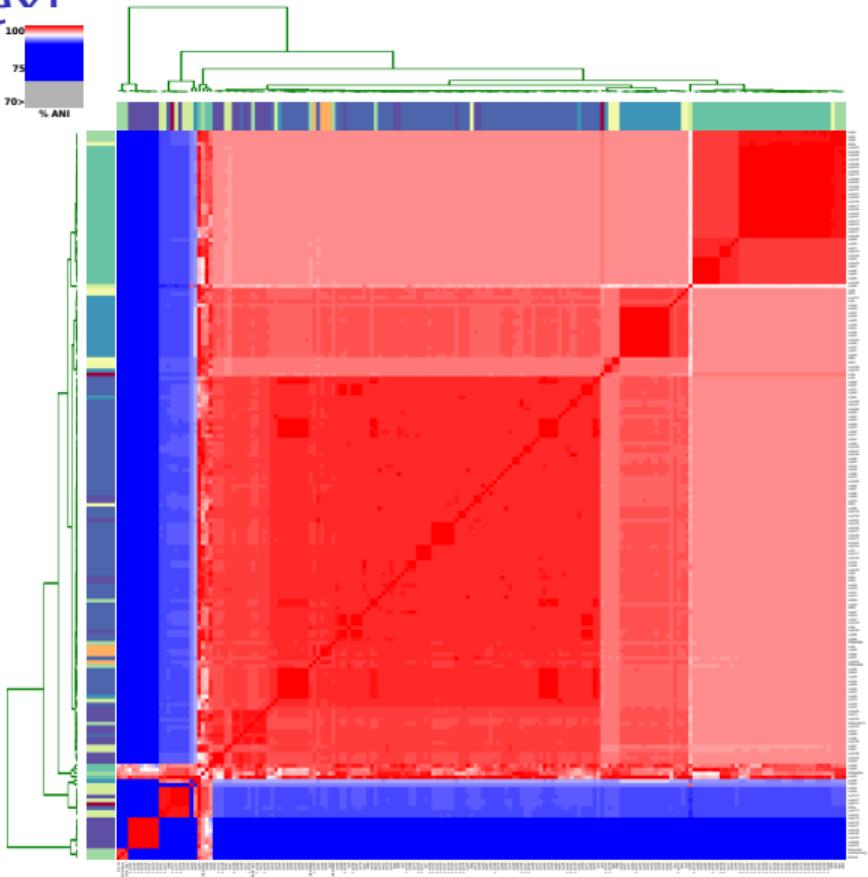
Genomic Context

- ▶ 202 isolates sequenced
- ▶ 153 true *E. coli* passed QC
- ▶ All Clermont phylotypes represented
- ▶ Diverse phenotypes
 - ▶ curli
 - ▶ metabolism
 - ▶ biofilm





Genomic Context⁺



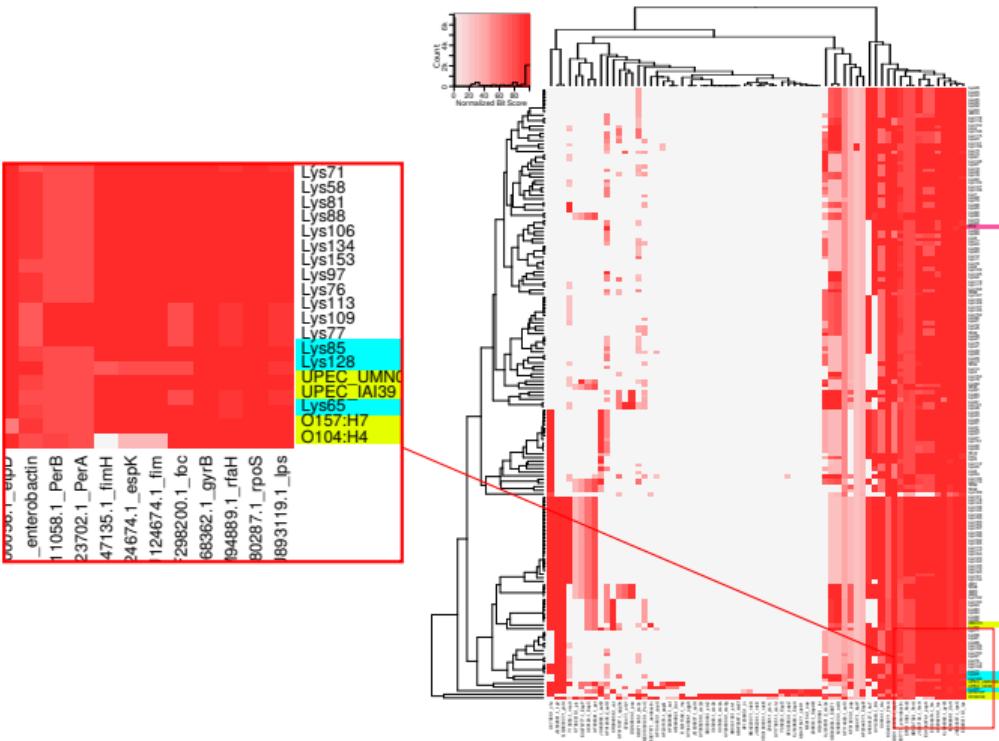


Virulence

- ▶ Search literature for genes implicated in virulence
- ▶ Select representative sequences for ~50 virulence factors
- ▶ Use reciprocal translated blast to find occurrences
- ▶ Filter results, visualize



Virulence Results





Conclusions about Soil-persistent *E. coli*

- ▶ Represent diverse lineages
- ▶ Possess virulence genes, but no *stx* toxins
- ▶ May pose a human health threat
- ▶ Complicate use of *E. coli* as contamination indicator



Next Steps

- ▶ Determine whether virulence genes are functional
- ▶ Explore genomes for markers associated with soil isolates
- ▶ Explore trends potentially relating function to environmental factors



Sources

- Bardsley, D. "A study of coliform organisms in the Melbourne water supply and in animal faeces, with observations on their longevity in faeces and in soil." The Journal of Hygiene, 46(3), 269–79. 1948
- Brennan, et al. "Characterization of environmentally persistent escherichia coli isolates leached from an irish soil." Applied and Environmental Microbiology, 76(7), 2175–2180. 1996
- Boyd, W and J. "Viability of Coliform Bacteria In Antarctic Soil." Journal of Bacteriology, 84. 1963
- Byappanahalli, et al. "Population structure, persistence, and seasonality of autochthonous Escherichia coli in temperate, coastal forest soil from a Great Lakes watershed". Environmental Microbiology, 8(3), 504–513. 2006
- Kirk, et al "World Health Organization Estimates of the Global and Regional Disease Burden of 22 Foodborne Bacterial, Protozoal, and Viral Diseases, 2010: A Data Synthesis." Plos Medicine 2015
- Pruess, B. *E. coli* image. NDSU Agriculture Comm. April 29, 2011
- Ryan and Fanning "Effects of fertiliser N and slurry on nitrate leaching - lysimeter studies on 5 soils." Irish Geography 29(2) 1996



Acknowledgments



OÉ Gaillimh
NUI Galway

NUIG Microbiology

- ▶ Matthias Waibel
- ▶ Stephen Nolan
- ▶ Camilla Thorn



James Hutton Institute, Dundee

Questions?