Genomic Epidemiology of Shigellosis in South Africa

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Background

- Shigellosis is a diarrhoeal disease caused by gram-negative Shigella bacteria

- Is the second leading cause of diarrhoeal death globally

- One third of deaths occur in children <5 years, with risks of long-term health effects

- The greatest disease burden falls on those living in sub-Saharan Africa and Asia

- Whole genome sequence analysis of Shigella underutilised in sub-Saharan Africa

- Antimicrobial resistance a growing obstacle to effective treatment

South Africa
- *S. sonnei* and *S. flexneri* 2a account for nearly 70% of cases

We aim to deepen our understanding of transmission of Shigella and AMR prevalence in South Africa to aid effective public health care policy.

Methods

- 561 South African national surveillance isolates - subjected to whole genome sequencing
  - Illumina HiSeq 4000
  - Collected 2011 to 2015
  - All 9 provinces
  - All ages

- Maximum likelihood phylogentics, based on core-SNPs, to contextualised South African population, predict population structure and infer evolutionary relationships

- Bayesian phylogenetics, also based on core-SNPs, to estimate evolution through time and serotype population dynamics

- Partial AMR phenotyping and full *in silico* phenotype prediction, based on genotype
  - StarAMR and AMR finder, SonneiTyping
  - Using draft genomes

- *In silico* virulence genotyping - virulenceFinder

Results

Shigella flexneri

- Four endemic lineages of *S. flexneri* (Fig. 1A)

- Distinct geographical distributions (Fig. 2)

- Cluster 1 lacks the MDR element SRL = reduced AMR (Fig. 1A)

- Cluster 1 potentially more virulent based on genotype (Fig. 1A)

- Cluster 2 associated with systemic disease

Shigella sonnei

- Identified three endemic lineages of *S. sonnei* (Fig. 1B)

- Evidence of strain importation, some with quinolone resistance

- Clusters highly similar in AMR and geographic distribution

- Cluster 2: higher retention of large virulence plasmid (pINV)
  - linked to virulence gene presence (Fig. 1B)

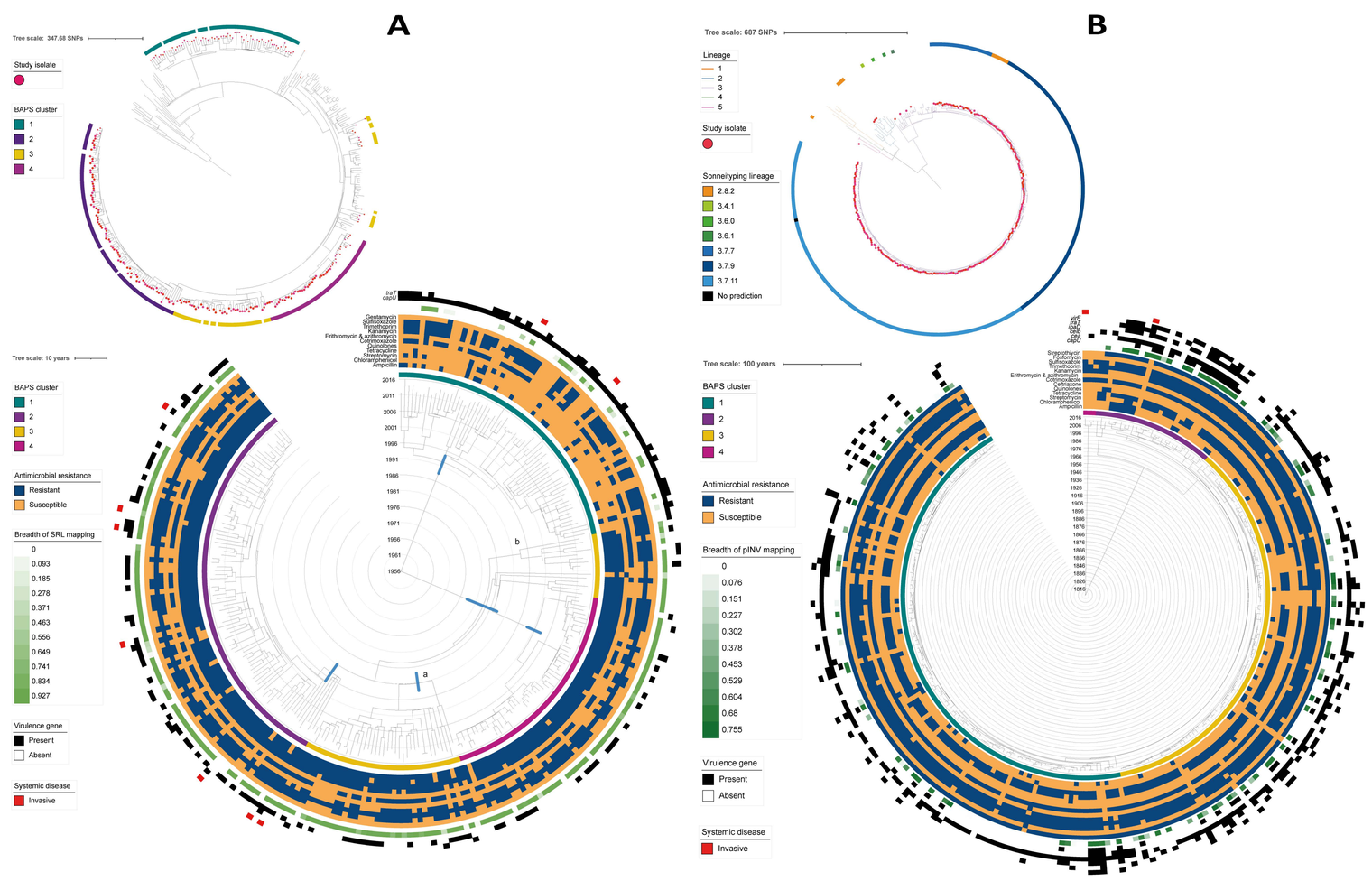

Figure 1. *Phylogenies of South African S. flexneri 2a (A) and S. sonnei (B).*
Maximum likelihood phylogies, with global population reference isolates, are in top left of A and B. Bayesian phylogenies are in bottom part of A and B. Population clusters can be seen on both phylogeny types. Bayesian phylogenies show time scale of evolution, predicted/phenotyped AMR profiles, mapping to the SRL (*S. flexneri* - A) or large virulence plasmid (pINV) (*S. sonnei* - B), presence/absence of virulence genes with uneven distribution i the population, and isolates from blood samples, indicative of systemic disease.

Strain introduction

- Drug susceptible *S. flexneri* 2a cluster 1 likely introduced into South Africa in 1992, after the MDR lineage
  - Coincides with beginning of HIV epidemic (Fig. 3)

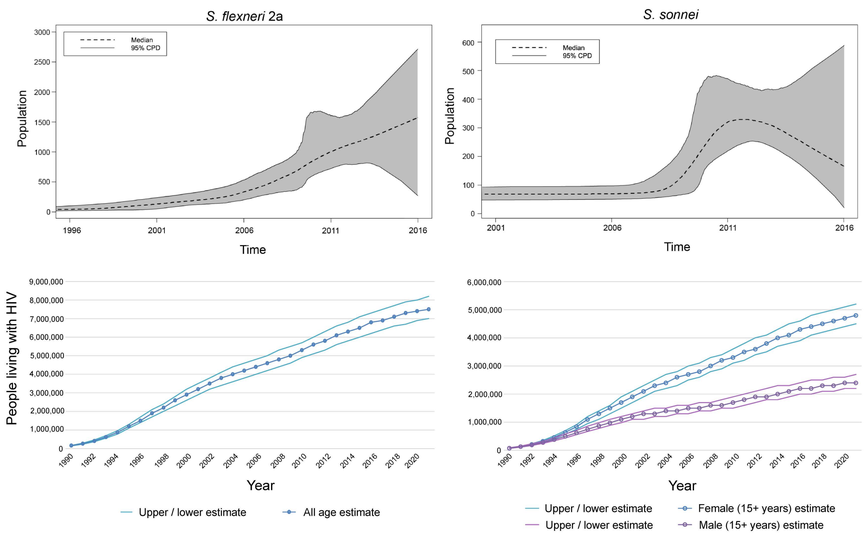

Figure 3. *Estimated population dynamices (top) and number of people in South Africa living with HIV, all ages (left) and 15 years and older (right)*
Estimated population expansion in S. flexneri matches rise in numbers of people livng with HIV, while population increase in S. sonnei started around 2006 when effective HIV treatment was introduced. HIV data from UNAIDS AIDSinfo.unaids.org

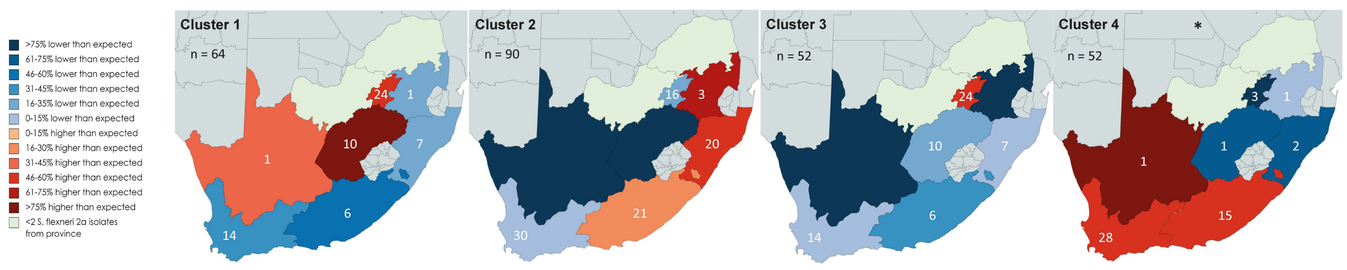

Figure 2. *Distribution of Shigella flexneri 2a isolates within South Africa by populations cluster.*
Provinces coloured according to the percentage difference in number of isolates to the expected number, based on the reported population size in the GERMS-SA annual reports.

Population diversification

- *Shigella flexneri* 2a clusters 2 (1994), 3a (1991) and 4 (2001) emerged from MDR endemic South African strain

- *Shigella sonnei* clusters 1 and 3 also likely emerged in 1996 and 2001, respectively

- Population expansion predicted in both serotypes, coinciding with HIV epidemic (Fig. 3)

Interpretation

- *Shigella flexneri* 2a endemic population is diverse, made up of multiple distinct strains; each with distinct distribution, and AMR and virulenece profiles - supports strain coexistance

- *Shigella sonnei* endemic population highly similar - cluster 2 has higher presence of pINV, suggesting greater retention - smaller population size suggests less successful, maybe due to greater retention of pINV. Instability of pINV previously identified as possible adaptation towards obligate pathogenic lifestyle

- Strain introduction and population expansion and diversification potentially linked to HIV epidemic in the country, though also coincides with the end of Apartheid (1992-1994)

- Adult women more at risk of shigellosis, possibly due to a greater likelihood of a carer role for children and sick relatives than men, though also possible link with HIV as HIV rates are higher in women than men in South Africa (Fig. 3)

MRC Medical Research Council

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References

Khalil et. *al.* (2018) *Morbidity and mortality due to Shigella and enteroinvasive Escherichia coli diarrhoea the Global Burden of Disease Study 1990-2016.* Lancet Infectious Diseases. 18, p:1229-1240.
Group for Enteric, Respiratory and meningeal Surveillance South Africa (2011-2015) *Annual Reports*.

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