Strains of Salmonella associated with invasive disease

Nick Feasey
Enteric vs invasive lifestyles

- Diverse genomic repertoire supporting broad host range
- Genome degradation of so-called "generalist" genes
- Specific additional virulence genes

Genotype

Generalists

Salmonella Typhimurium

Salmonella Enteritidis

Host adapted

Salmonella Dublin

Salmonella Choleraesuis

Host restricted

Salmonella Typhi

Salmonella Gallinarum

Enteric/diarrhoeal disease

Invasive/systemic disease

Low mortality

High mortality

Feasey Lancet 2012
Paediatric iNTS disease in Africa: pre/early HIV era

Nigeria, 1970s
Alausa, Scand J Infect Dis

Kenya 1980s
Wamola, E Afr Med J

Gambia 1980s
Mabey, JID

DRC 1980s
Green, Ann Trop Paeds

Rwanda, 1980s
Le Page, Lancet
iNTS disease AIDS defining & NTS among most common cause of bloodstream infection in SSA

TABLE I - SEROLOGICAL MARKERS AND OPPORTUNISTIC INFECTIONS IN FIVE HETEROSEXUAL BLACK AFRICANS

<table>
<thead>
<tr>
<th>Virus antibody titres (inverse)</th>
<th>CMV</th>
<th>EBV</th>
<th>IGM</th>
<th>Opportunistic infections</th>
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<td>+A. clytostomum apoplasticus;</td>
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<td>Herpesvirus gordini;</td>
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<td>Epstein-Barr viruses;</td>
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<td>Escherichia coli;</td>
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<td>Shigella flexneri;</td>
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<td>Salmonella enteritidis;</td>
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<td>Salmonella paratyphi;</td>
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<td>Salmonella typhi;</td>
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<td>Salmonella typhosa;</td>
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<td>Staphylococcus aureus;</td>
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</tbody>
</table>

North Africa
Two studies covering 14 locations
10 230 patients, 10-3% with BSI
HIV was not reported
All studies were primarily in adults
Three commonest isolates: Salmonella 49-9%
(Salmonella enterica va Typhi 99-0%),
Brucella spp 26-8%, Staphylococcus aureus 7-7%

East Africa
Seven studies covering nine locations
21 317 patients, 7-9% with BSI
18-5% of included patients tested for HIV
23-7% of 3445 tested were seropositive for HIV-1
Four studies were primarily in children and four were primarily in adults
Three commonest isolates: Streptococcus pneumoniae 21-2%
Salmonella 17-8% (non-typhoidal 88-0%),
Escherichia coli 9-5%

West and central Africa
Six studies covering five locations
5887 patients, 12-4% with BSI
5-4% of included patients tested for HIV
63-3% of 319 tested were seropositive for HIV-1 or HIV-2
Five studies were primarily in children and one was primarily in adults
Three commonest isolates: Salmonella 20-8% (non-typhoidal 87-0%),
S pneumoniae 18-9%, S aureus 17-2%

Southern Africa
Seven studies covering five locations
23 893 patients, 9-8% with BSI
5-0% of included patients tested for HIV
59-8% of 1204 tested were seropositive for HIV-1
Four studies were primarily in children and three were primarily in adults
Three commonest isolates: Salmonella 29-0%
(non-typhoidal 97-0%), S pneumoniae 24-0%,
S aureus 9-4%
S. Typhimurium associated with iNTS disease in sub-Saharan Africa have novel MLST: ST313

- Genomic degradation
  - Also seen in S. Typhi and other pathogens becoming host adapted
  - Similar genes to S. Typhi

- Novel Prophage repertoire

- MDR cassette in virulence plasmid
Phylogeny of S. Typhimurium reveals isolates from SSA fall into two highly related lineages.
Novel Prophages: Blantyre Type Prophage 1

S. Typhimurium D23580

A

Gene for O replication protein is a pseudogene preventing viable Gifsy-2
Pseudogene in tail assembly gene preventing viable ST64B
SNP in promoter of operon encoding the antirepressor affects function of Gifsy-1

Single nucleotide allelic exchange

Functional

Reversion of the SNP to P<sub>din-flos<sup>14028s</sup></sub> restores activity of Gifsy-1<sup>10223580</sup>

B

D23580
D23580 P<sub>din-flos<sup>14028s</sup></sub>

10<sup>-1</sup>
Evidence of different behavior of ST313 and ST19

• ST313-td gene on BTP1 – (Herrero-Freson 2014, Owen 2017)
• ST313 stimulate less inflammasome activation than ST19 (Carden 2015)
• ST313 with naturally attenuated flagellin elicits reduced inflammation, replicates in macrophages (Ramachandran 2015)
• Loss of multicellular behavior in ST313 (Singletary 2016)
• Pseudogenization of the Secreted Effector Gene ssel Confers Rapid Systemic Dissemination (Carden 2017)
African S. Typhimurium is more invasive in chickens than global clades.
ST313 not restricted to SAA

- 79/2,888 UK S. Typhimurium in PHE collection are ST313
Lineage I & II isolates associated iNTS disease and travel to Africa

The remainder isolated from stool, prophage diversity, drug susceptible

Satheesh Nair
Public Health England

Ashton & Owen, Manuscript in preparation
African clades of S. Enteritidis

- Genomic degradation
- Novel Prophage repertoire
- MDR cassette in virulence plasmid
- Novel clades have highly conserved accessory genomes

Feasey Nat Gen 2016
Placed in context of PHE collection (~3,000 isolates), there is restriction to SSA
There’s a novel clade of S. Typhi too!

*Emergence of H58 lineage*

*(Or subclade 4.3.1)*

Wong et al Nat Gen 2015
Global dissemination of S. Typhi H58

Emerged ~30 years ago

Malawi
East Africa
South Africa
Western Asia
South Asia
South-East Asia

Fiji

Wong et al Nat Gen 2015
Extended spectrum beta-lactamase producing variants an emerging problem in invasive Salmonella disease

- bla$_{\text{CTX-M15}}$ first reported in S. Typhimurium in Malawi
- Same plasmid subsequently reported in Kenya
- 57% in rural west Kenya
- Emerging problem in S. Typhi
- Has potential to make invasive Salmonella disease untreatable in many settings
Summary

- Novel clades of MDR S. Typhimurium and S. Enteritidis are exploiting the high prevalence of immunosuppressive conditions to cause epidemics of iNTS disease
  - More invasive?
  - Less invasive?
  - Niche adaptation to particular environmental reservoir?

- MDR H58 S. Typhi has established itself globally

- Drug resistance a major and evolving problem
- ESBL particularly frightening
10,000 Salmonella genome project:

Aim: to understand the epidemiology, transmission & virulence of iNTS disease associated Salmonellae

- **Sample Collection**
- **Sample Preparation**
- **DNA Extraction and Sequencing**
- **Results Available through Enterobase**
- **Metadata**
- **Our focus is iNTS associated strains**
- **Salmonella strains from DAC list**

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