Whole Genome Sequencing for routine surveillance, outbreak detection & response in NSW, Australia

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A note about jurisdictional differences (and implications for foodborne outbreaks)

Whole Genome Sequencing (WGS) in Australia

Integration of WGS into routine surveillance in NSW

Cross-jurisdictional lessons and limitations
Context: Australia
Australia to scale
Climate affects FBI in Australia

Hot summers magnify outbreaks due to temperature abuse.

Cold wet winters produce many outbreaks of viral gastroenteritis in major cities, particularly aged care facilities, and child care facilities.

Tropical zone provides optimal growth conditions for foodborne pathogens.

Our native fauna provide reservoirs of infection for many foodborne pathogens.

Our fondness for overseas travel brings home many cases of foodborne disease.
OzFoodNet

- OzFoodNet Site in each jurisdictional health department and the original trial site in Hunter New England
- OzFoodNet Central provides coordination
- Member of Communicable Diseases Network Australia
- OzFoodNet members include:
  - Public Health Laboratory Network
  - Department of Agriculture
  - Food Standards Australia New Zealand
  - National Centre for Epi and Pop Health
Foodborne surveillance in Australia

- State and territory health departments collect notifications of communicable diseases under their public health legislation
  - 6 States + 2 Territories = 8 different Governments involved in the surveillance, investigation and control of foodborne disease

- Food Safety is governed by the Australia New Zealand Food Standards Code and State / Territory legislation

- There is no legislated ‘central body’ for foodborne investigations
WGS in Australia
WGS experience in Australia

- WGS-based surveillance is used in routine national surveillance for *Listeria monocytogenes*

- WGS has also been used in 4 multi-jurisdictional outbreaks investigations (MJOIs) in 2016:
  - *Salmonella* Anatum in mixed salad products ~300 cases
    8 Feb 2016 – 11 May 2016
  - *Salmonella* Saintpaul in mung bean sprouts ~400 cases
  - Listeriosis in delicatessen products, 7 cases
  - *Salmonella* Hvittingfoss in rockmelons ~150 cases
    18 Jul 2016 – 28 Sep 2016
WGS for routine surveillance
Background

- WGS can provide much more detailed information about organisms and their relationships to each other than other typing methods.
- WGS is becoming faster and cheaper to perform.
- Retrospective studies of WGS show it provides better resolution in outbreak settings than traditional bacterial typing methods...

... but does this mean it can be translated into an effective surveillance system?

...and can it be scaled up to meet the requirements for a high-burden enteric pathogen?
Aims

- Determine whether integrating routine whole genome sequencing into surveillance and outbreak investigation has an effect on public health outcomes

- Describe barriers to and enablers of implementation of routine whole genome sequencing for public health

- Focus on three organisms
  - *Mycobacterium tuberculosis* (MTB)
  - *Salmonella Typhimurium* (STm)
  - *Listeria monocytogenes* (Lm)
Implementation plan

- Two year pilot project to evaluate the effect of whole genome sequencing in surveillance of *Salmonella* Typhimurium (STm)

- All *Salmonella* Typhimurium isolates referred to the NSW Enteric Reference Laboratory are being routinely sequenced in parallel with MLVA typing
  - Human isolates
  - Food Authority NSW isolates from environmental and food testing

- WGS cluster information is correlated with epidemiological information to inform an investigation and control actions
WGS project progress

- Routine sequencing of STm began in October 2016
- So far, a total of 1,479 STm isolates have been sequenced, comprising 1,410 human isolates and 69 isolates from food, animal, and environmental sources (~98% of expected human isolates).
- Median turnaround time for specimens:
  - Has been improving overtime, and is occasionally now available before MLVA
  - In September 2017: median turnaround time from specimen collection to WGS report of 25 days (range 17-34 days)
WGS project progress

Aim ≤ 28 days
Of the 1,479 isolates reported, 1,219 (82%) clustered with at least one other isolate. There were 159 SNP clusters containing two or more isolates, with a median cluster size of 3 (range 2-97).

Distribution of cluster size for *Salmonella Typhimurium*
## WGS cluster report

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Workflows – cluster detection

- Cluster identification program
  - reads in SNP cluster report from the lab and extracts patient information from public health database
  - compares results against previous results and reports clusters that are new or increasing in size, with summary statistics

Cluster names are easy to confuse – can give informal names and allocate clusters to one person
Workflows – cluster detection

- For each new or growing cluster:

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Epi curve for cluster 17-0436, by week of specimen collection

Age - cluster 17-0436

- Analysis Variable: age age_at_event_years
- Minimum: 0.0, 50th Pctl: 3.0, Maximum: 63.0

PHUs - cluster 17-0436

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Calculated onset date (week beginning)
WGS cluster 16-0233

Specimen collection date (week beginning)

- 3-16-9-7-523
- 3-16-9-11-523
- 3-17-9-11-523
- Other
Food and environmental isolates

3-16-9-7-523

3-16/17-9-11-523
WGS cluster 16-0233 - lessons

- Including isolates from Food Authority testing in analysis adds to the value of WGS by confirming epidemiological and MLVA links between cases and implicated premises and/or foods.
- SNP trees within a cluster can give more detail about potential sources and guide interview questions.
- However, far from solving the puzzle (as promised!) it can create new questions when new cases are linked by WGS but not epidemiologically.
Overall lessons (1)

1. WGS is excellent for confirming epidemiological links between cases and between sources and cases.

2. When used for prospective cluster detection, WGS identifies a large number of small(er) clusters, which needs additional triage and resources to follow up.

3. Traditional case interviews and epidemiological methods are not helping to solve these clusters as quickly as they do with outbreaks detected through traditional epidemiological methods.

4. Collaboration between public health, laboratory, and food/agriculture is essential.
Overall lessons (2)

5. Case follow-up protocols may need to be revised to get the most value out of WGS.
   - Any data is better than no data
     - Trialng use of novel interview methods: e.g. SMS
   - Surge staff/interviewers
   - In large rapid moving outbreaks it’s not yet fast enough to exclude cases before interview

6. Going forth, need a consistent nomenclature structure
   - SNP analysis has unbeatable resolution but every new isolate changes the tree and potentially the cluster – so can’t store results in traditional surveillance databases
   - SNP analysis in Australia is not yet fast enough or stable enough to detect outbreaks of common pathogens
   - Potential for cgMLST?
Acknowledgements

This project is funded by a NSW Health Translational Research Grant

Project team:

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<th>Partner Organisations</th>
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<th>Affiliates</th>
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<td>Centre for Infectious Diseases and Microbiology-Public Health</td>
<td>Vitali Sintchenko Jon Iredell</td>
<td>Chayanika Biswas, Elena Martinez, Rebecca Rockett, Ranjeeta Menon, Nathan Bachmann</td>
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<td>Communicable Disease Branch, NSW Health Protection</td>
<td>Vicky Sheppeard Kirsty Hope Chris Lowbridge Paula Spokes</td>
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<td>Marie Bashir Institute, University of Sydney and WIMR</td>
<td>Tania Sorrell Grant Hill-Cawthorne</td>
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<td>ICPMR-Pathology West</td>
<td>Dominic Dwyer Sharon Chen</td>
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Special thanks also to:
NSW Public Health Network, NSW Food Authority & OzFoodNet

Questions (or further discussion): [keira.glasgow@moh.health.nsw.gov.au](mailto:keira.glasgow@moh.health.nsw.gov.au)
Further examples
Example: *Salmonella* Anatum MJOI

- Salmonella Anatum in bagged salad products ~300 cases
- 8 Feb 2016 – 11 May 2016 (MJOI dates)
- Rare serotype, able to detect cluster quickly.
- Initial hypothesis generating questionnaires indicated high frequency of consumption of a number of food items including bagged salad.
- Routine shelf life testing at a salad producer detected Salmonella species in packaged product which had been in the market, but passed UB date.
- Microbiological and epidemiological evidence = food recall
- This was the first Salmonella MJOI where WGS was used. The “outbreak sequence” was incorporated into the case definition.
  - Sequences from human and food isolates were indistinguishable.
  - 0-2 SNPs difference in over 100 specimens sequenced
  - It provided confidence that bagged salads were the source of the outbreak despite consumption of several brands and products – eaten at home and out of home
- Case control study conducted in Victoria supported these findings
Example: S. Anatum MJOI

Salmonella Anatum notified by month in Australian States and Territories, NNDSS, 2009-2017 YTD (Analysed 01/08/2017 by Date of Diagnosis, OzFoodNet)
Example: S. Anatum MJOI
Example: *Salmonella* Saintpaul MJOI

- This outbreak began in Dec 2015 in SA and NSW
- Common serotype particularly in Qld often associated with water
- Initial hypothesis generating questionnaires indicated several primary produce products with onions being primarily implicated.
- WGS showed NSW, SA and ACT isolates part of same cluster.
- In Feb cases dropped off in NSW and then rose in SA and NT.
- WGS led us to keep considering it a single related outbreak
- In April new hypothesis of mung bean sprouts
- In late April an environmental sample from an SA sprout grower detected
- WGS showed it to be indistinguishable from cases
Example: S. Saintpaul MJOI
Example: S. Saintpaul MJOI