Conclusion

Food poisoning caused by *Cl. perfringens* is a preventable condition. If the right checks and balances are built into the food preparation process, then the organism should not have an opportunity to germinate and proliferate. Even if this does occur, thorough reheating of the food at temperatures in excess of 60°C will eliminate the organism and prevent an outbreak of food poisoning. This highlights the importance of appropriate education and compliance of personnel involved in food preparation so that they understand what can happen when they mishandle food or essential equipment breaks down.

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Biography

John Bates is the Chief Scientist in charge of the QHFSS Public Health Microbiology laboratory. This laboratory provides a support arm to the Queensland Health Public Health Units in the investigation of outbreaks of human, water and foodborne disease, as well as providing phenotypic and genotypic data on a wide range of notifiable bacteria in Queensland.

Salmonella and egg-related outbreaks

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Non-typhoidal *Salmonella* infections are a significant public health issue in Australia, with record numbers of both disease notifications and outbreaks being reported in recent years[1,2]. Epidemiology plays an important role in *Salmonella* outbreak investigation, helping to identify raw and minimally cooked eggs as an increasingly common cause for these events. Of particular relevance to disease caused by Australian eggs is *Salmonella enterica* subsp. *enterica* serovar Typhimurium. A crucial element in demonstrating this serovars presence throughout the food chain is the ability to trace suspect eggs to their source. High product turnover makes this challenging but through the adoption of integrated surveillance practices and harmonised laboratory methods, a more effective response may emerge.

Epidemiological investigations of egg-related outbreaks and salmonellosis

Determining the source of infection for a case of salmonellosis is difficult because the volume of cases, coupled with competing public health priorities, means that most cannot be investigated. Consequently much of our understanding of disease causation comes from outbreak investigation. To determine if an outbreak might be attributed to eggs, investigators use both descriptive and analytical epidemiology, findings from environmental health and food safety investigations and the results from microbiological testing of human, food and environmental samples[3].

In outbreaks where the exposure or food vehicle is not immediately apparent, analytical epidemiology may be used to examine disease occurrence and risk. These analyses involve making comparisons between groups of people, e.g. those with a *Salmonella* infection and those without (i.e. case control study) or those who have eaten a particular food and those who not (i.e. retrospective cohort study). Statistical inferences can then be made as to whether a case was more likely to have eaten a particular food or whether having exposure to a particular food carried increased risk of illness. For those cases unable to be investigated by epidemiological or microbiological means, the same attribution to a potential source cannot be as easily made, hence the focus on outbreak cases.

Between 2001–2009, 1,025 foodborne outbreaks were reported in Australia, with *Salmonella* being identified as the causative agent in one-third of these[4]. Among *Salmonella* outbreaks, eggs and egg containing foods were the most commonly identified causes. *S.* Typhimurium was responsible for 90% of these outbreaks, which resulted in some 2300 cases, including over 500 hospitalisations[4]. In the period 2010–2011, this significant morbidity has further increased, with OzFoodNet identifying another 60 egg-related outbreaks, involving over 1000 cases and 180 hospitalisations (*personal communication OzFoodNet Egg Working Group*).

Egg-related outbreaks occur in varied settings, with restaurants and cafes being most frequently identified[1,5,6]. Raw or minimally cooked egg containing sauces (aioli and hollandaise) and desserts (mousse, fried ice cream and tiramisu) are food vehicles commonly involved in these outbreaks[1,5,7]. Such settings are identified because large numbers of people become unwell in a short time, enabling surveillance systems to detect increases in case numbers above expected thresholds or because community members are more likely to alert health officials to issues within commercial premises. Outbreaks in private residences are also common but usually involve fewer cases and are not reported as often. Finally, outbreaks linked to takeaways and bakeries[8,9] are also frequently identified, often being characterised by large case numbers, spread over a wide geographic area. Such characteristics highlight how outbreaks might be more or less likely to be identified due to the setting, potentially biasing our understanding of foodborne illness settings. Nonetheless, outbreak reporting analysis remains our best source of information about foodborne disease while also reducing concerns related to publication bias[10].

During 2001–2009[4], investigators conducted analytical studies for one third of reported outbreaks, with nearly three quarters of these showing an association with eggs or an egg-containing food. Laboratory confirmation of *Salmonella* in an egg-containing food was obtained in 39% of outbreaks. Trace back was conducted in 62% of all outbreaks, with around 70% conducted to the farm level. For farms and processing facilities where on-site testing was conducted, 50% had the outbreak strain recovered in the farm or processing environments.

It is reasonable to assume that for some serovars, the food sources identified during outbreaks will also be responsible for causing sporadic disease[11]. Since 2007, New South Wales (NSW) has used Multi-locus Variable number of tandem repeats Analysis (MLVA) as the primary typing method for S. Typhimurium. MLVA is able to discriminate further within an S. Typhimurium phage type, allowing a better attribution of the source of infection for both sporadic and outbreak cases. For example, during 2012 NSW Health investigated a restaurant outbreak of *S.* Typhimurium MLVA 3-9-9-12-523, an uncommon MLVA pattern. The investigation determined the food vehicle as a raw egg-containing dessert. The New South Wales Food Authority (NSWFA) traced the eggs used back to a specific farm, isolating the outbreak strain from the farm environment and grading facility. Retrospectively, another 30 cases were able to be linked to the egg farm, either via further outbreaks at the restaurant or through consumption of products from a bakery using eggs from the same farm.

Salmonella Typhimurium, *Salmonella* Enteritidis and their control

The epidemiology of egg-related salmonellosis and the efforts to control contamination in laying hens, farm and processing environments differs between Australia and elsewhere internationally. In Australia, *S.* Typhimurium causes the majority of infections, being frequently identified as a cause for foodborne outbreaks, particularly where raw or minimally cooked eggs have been used[1]. In North America and Europe, control efforts have been in response to a problematic and costly *S.* Enteritidis epidemic. Importantly this serovar is not endemic in Australian layer flocks[12]. *S.* Enteritidis differs from other serovars in its capacity for trans-ovarian transmission, i.e. an ability to infect the egg's internal contents. However, *S.* Typhimurium has also been shown to colonise the reproductive tracts of infected hens, though the significance of this remains unclear[13]. Furthermore uncertainty exists around whether vertical or horizontal transmission is more important in *S.* Enteritidis with studies showing shell contamination exceeding that of the internal contents[14].

International poultry control programs have resulted in significant decreases in egg-related salmonellosis[15,16]. These programs employ measures including: on-farm monitoring, diverting contaminated eggs for processing, culling infected flocks, cleaning and disinfection of sheds, through chain refrigeration of eggs, and vaccination of flocks[15,16]. In the United States, these measures apply to *S.* Enteritidis infected flocks whereas the European Union Regulation addresses monitoring and control for both *S.* Enteritidis and *S.* Typhimurium[17]. The absence of *S.* Enteritidis in Australian flocks is fortunate however other serovars still cause egg-related disease. The Australian standard for egg production and processing[18] was developed due to Australia having unacceptably high numbers of cases linked to eggs and inadequate regulatory and non-regulatory measures to prevent illnesses[18].

Eggs, public health and food safety

Egg-related outbreaks result from breakdowns in control measures along the farm to fork continuum. When eggs are epidemiologically associated with illness, the use of food prepared with raw egg is frequently confirmed via observation or interviews with restaurant staff. There is however no strong evidence suggesting any sudden change in preparation and preferences that might explain the increase in outbreaks. What is known is that both egg production and consumption have soared[19]. Given shell egg production could never be *Salmonella* risk free, it is plausible that the volume of contaminated eggs in circulation has risen. While campaigns targeting consumer and food service practices help reduce the incidence of disease, reduction of bacterial contamination in the egg production system would be more effective, as shown with international *Salmonella* control in poultry.

Public health authorities are particularly concerned when *Salmonella* serovars that commonly cause human disease are recovered from egg-laying, grading or processing environments. The Food Standards Australia and New Zealand (FSANZ) Primary Production Processing Standard for Eggs and Egg Products[18] requires producers and processors to identify and control hazards, prohibits the sale of cracked and dirty eggs (unless pasteurised) and requires individual egg stamping to enable tracing. If, as outbreak investigations suggest, increased numbers of contaminated eggs are entering the retail and commercial market supply chains, further evidence of pathogen reduction on-farm, at the grading and at the processing level is required. Prevalence data for contaminated eggs in Australian are limited: estimates were last published in 2005[20] and were based on work from over 10 years ago, well before the observed increase in outbreaks.

Trace back in outbreak investigations

Trace back is a method of determining the source and distribution of a product associated with an outbreak, in addition to identifying the points where contamination could have occurred[21]. It is often difficult to achieve as investigations commence after an initial contamination event. The FSANZ Primary Production Standard for Eggs and Egg Products[18] requires the stamping of all eggs, *theoretically* allowing trace back to individual farms. However in reality this is unlikely to be as useful as eggs and their packaging will likely have been used or discarded before an investigation commences.

Table 1. Laboratory characterisation of *Salmonella* in Australian states and territories.

State/Territory	Serotyping	Phage typing	MLVA	PFGE
Australian Capital Territory	–	–	–	–
New South Wales	✓	–	✓[A]	–
Northern Territory	–	–	–	–
Queensland	✓	–	✓[A]	–
South Australia	✓	✓[A]	✓	–
Tasmania	–	–	–	–
Victoria	✓	✓[A]	✓	–
Western Australia	✓	–	–	✓[A]

[A]Primary method for *S.* Typhimurium sub-classification.

Currently small producers are more likely to have on farm investigations conducted because they have localised operations and less complex supply chains. If an investigation determines 'Brand X' eggs were used by a cafe and those eggs were produced on a single farm, regulators can have greater confidence in conducting on-farm testing for the outbreak strain. However, if Brand 'Z' eggs (produced by a larger company) were used, the trace back investigation can become more difficult. In large operations packaging and processing facilities may be supplied with eggs from dozens of smaller farms. In theory egg stamping will allow trace back to farm level but because of high product turnover in the food sector, leftover eggs, cartons or packaging with bar coding and other identifiers will have been long discarded. Regulators are left in a difficult position: suspicion exists that contaminated eggs have been produced but they are unable to verify this due to a lack of traceability and documentation. OzFoodNet data shows where trace back has been possible and undertaken in a timely manner, the outbreak strain will frequently be found in the farm and processing environments[4].

Integrated surveillance: sharing data from farm to beyond the fork

There is a growing appreciation of the need to integrate surveillance data from on-farm through to the point of sale, including laboratory data derived from human, environmental and animal surveillance[22]. This will require a new level of trust between government and industry, including recognition that reducing foodborne illness is not just a role for health agencies. While OzFoodNet works where possible with regulators and food safety agencies to compare data on human pathogens with other sources, there are drawbacks to the informality of the approach: a lack of access to test results conducted in private laboratory settings and, more worrying, a reluctance to sometimes share information between government agencies, even within the same jurisdiction. The sharing of data for public health purposes and collaboration between government and industry has underpinned the success of the EU *Salmonella* control programs[23].

The role of the microbiology laboratory

The contribution of reference laboratories in the testing of clinical, food and environmental samples is vital. However, the adoption by some but not all jurisdictions, of MLVA typing for the characterisation of *S.* Typhimurium has impeded the ability to rapidly compare data nationally. The predominance of *S.* Typhimurium, particularly in south-eastern Australia, and its responsibility for large numbers of outbreaks, including those due to eggs[7], highlights the value of a coordinated and harmonised laboratory response. Currently smaller jurisdictions forward isolates to their larger neighbours for serotyping. For *S.* Typhimurium, further classification may result in an MLVA profile or a phage type, depending on the typing method used by the receiving reference laboratory. Such issues relating to laboratory characterisation of human isolates are inevitably addressed through OzFoodNet's strong collaborative ties with reference laboratories and related stakeholders. Table 1 shows typing methods employed in Australian jurisdictions.

Conclusion

Salmonella *Typhimurium* is the serovar most responsible for Australia's ongoing '*epidemic* of egg-related outbreaks'. These outbreaks occur most frequently in restaurants and cafés, with raw or minimally cooked egg-containing sauces and desserts being identified as responsible food vehicles, reinforcing the need for ongoing consumer and food service industry education. Although a number of comprehensive and guiding documents exist in Australia[18,24], stronger trace back and regulatory capacity is required. This is unlikely to be achieved through egg stamping; rather improvements in retailer and producer documentation are needed. Nevertheless trace back investigations have provided evidence that *Salmonella* has entered into the wider food chain, reinforcing the need for public health authorities, regulators and those in industry, to develop systems for data sharing and integrated surveillance. Underpinning this should be a goal of targeted reductions in on-farm pathogen prevalence. Finally, while acknowledging laboratories invaluable contribution, the differences in techniques should be flagged as being potentially problematic but by no means insurmountable.

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Biographies

Cameron Moffatt is a communicable diseases epidemiologist currently working with the OzFoodNet network. His particular interests include outbreak investigation and the epidemiology of *Salmonella* infections, notably their link with eggs. Cameron is a graduate of the Australian National University's field epidemiology training program.

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Foodborne campylobacteriosis in Australia

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Thermophilic *Campylobacter* are an important cause of human illness worldwide. *Campylobacter* reservoirs include a wide variety of wild birds, poultry, farm animals, domestic pets and natural water systems. In Australia, infection is mainly associated with foodborne transmission, though other routes of exposure including waterborne and direct zoonotic transmission are not uncommon. Most cases of infection appear to be sporadic in nature, with outbreaks rarely reported. Epidemiological and microbiological evidence suggests chicken meat is the principal source of infection among cases. A recent study estimated there are more than 50,000 cases of *Campylobacter* infection attributed to chicken meat each year in Australia. When outbreaks are detected, they are most often associated with the consumption of poultry, contaminated water and occasionally unpasteurised milk. The lack of recognised foodborne outbreaks of campylobacteriosis could be due to organism-related factors such as the inability of thermophilic *Campylobacter* to multiply on food left at room