Salmonella risk profile of UK-produced hen shell eggs

Area of research interest: <u>Foodborne pathogens</u> Project status: Completed Project code: G1000072 Authors: Erin Lewis, Victoria Cohen, Charlotte Evans, Iulia Gherman Conducted by: Food Standards Agency DOI: <u>https://doi.org/10.46756/sci.fsa.rpp424</u>

Salmonella risk profile of UK-produced hen shell eggs: Executive summary

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Background

<u>A previous risk assessment</u> from the Advisory Committee on the Microbiological Safety of Food (ACMSF) in 2016 concluded that due to the significant reduction in the risk from *Salmonella* in UKproduced hen shell eggs produced under a recognised farm assurance scheme (Lion Code or equivalent), the risk to consumers from eggs produced under these schemes was 'very low'. This risk assessment led the FSA and FSS to update their consumer advice on the consumption of eggs in 2017, stating that vulnerable groups could consume raw or runny eggs produced within an assurance scheme.

This risk profile will examine the current situation of Salmonella in UK-produced table eggs, and the factors that may influence the current risk of *Salmonella* in UK-produced eggs and highlight any that have changed since the risk assessment provided by the ACMSF in 2016.

Hazard Identification

Non-typhoidal *Salmonella* is widespread in domestic and wild animals, and the environment, and can readily pass through the food chain. *Salmonella* is estimated to be one of the leading causes of foodborne disease in the UK. Eggs and egg products were the food type most commonly associated in *Salmonella* outbreaks in the UK between 2015-2020.

Exposure assessment

The prevalence of all *Salmonella* serovars in laying flocks in the UK in the period 2017-2021 was similar to 2009-2016 levels. The prevalence of regulated *Salmonella* serovars and *Salmonella* Enteritidis has roughly doubled in the 2017-2021 period. Levels of regulated *Salmonella* serovars still remain within the <u>National Control Programme (NCP)</u> requirements of 2% - in 2021, this was 0.23% in layers tested.

An EU ban on the use of formaldehyde-based products in animal feed was implemented in 2018 and was suggested as a contributing factor to an increase of *Salmonella* seen in broiler flocks but further evidence is needed to support this.

Eggs can be contaminated with *Salmonella* via direct contamination during formation, or indirectly after the egg has been laid. The major extrinsic factors of penetration of eggs by *Salmonella* are the bacterial strain, temperature differential, moisture, number of organisms present, and storage conditions. The main intrinsic factors are shell defects, cuticle and porosity of the eggshell. Cross contamination can occur at many points along the supply chain. During processing and packaging, potential areas of risk include those where the eggs are in close contact with each other, such as in the egg grader and rollers, where bacteria can be transferred from one egg to another. Additionally, an experiment involving inoculated cartons show Salmonella will transfer to eggshells from cartons.

Cross contamination can also occur during the breaking and processing of eggs before consumption - breaking eggs can transfer Salmonella to the hands or surfaces and whisking can spread Salmonella up to 40cm from the bowl.

Consumption of raw or runny eggs was investigated in the FSA Food and You 2 survey, with 61% of respondents never eating raw eggs, 22% never eating less-than-thoroughly cooked (LTTC) eggs and 8% never eat thoroughly cooked eggs.

Hazard characterisation

Symptoms of *Salmonella* infection can range from asymptomatic carriage to severe diarrhoea. This is usually self-limiting, however it can be more severe in vulnerable groups, including the elderly, pregnant women, the young, and immunocompromised, leading to systemic infection and death.

The median infective dose required to infect 50% of the population (ID50) for non-typhoidal *Salmonella* is high but varies between serovar and food vehicle, with a study by Teunis et al.,

2010, suggesting the ID50 to be as low as 1.09 x 101 colony-forming units in eggs.

Temperature can affect the viability of *Salmonella* on eggshells, with cold temperatures decreasing the growth of *Salmonella* in the egg yolk and albumen but increasing the survival of *Salmonella* on the eggshell surface.

Between 2015 and 2019, a total of 954 confirmed cases of salmonellosis were investigated as part of outbreak investigations and determined to be associated with consumption of eggs and/or egg products, in a total of 15 reported investigations. This is a similar number of cases per year as reported in the ACMSF risk assessment (2016). The majority of outbreaks (10/15) reported fewer than 45 cases. The largest outbreaks occurred in 2016 (158 cases), 2017 (162 cases) and 2018 (259 cases). Data collected is provisional, and a full dataset has been requested from UKHSA. Two outbreaks of around 100 cases each were linked to Lion Code eggs. The ACMSF risk assessment had found only one small outbreak in 2009 linked to Lion code eggs.

Since the ACMSF report, the implementation of whole genome sequencing for *Salmonella* surveillance by the UK public health agencies has become routine. This has increased the sensitivity and specificity of case ascertainment in outbreak investigations, and confidence in source attribution.

Overall, analysis of *Salmonella* in UK-produced hen shell eggs does not indicate a need for a risk assessment at this time. Certain uncertainties and data gaps were identified that it would be beneficial to address before the commissioning of future risk assessments.

Salmonella risk profile of UK-produced hen shell eggs: Lay summary

This risk profile assesses new evidence regarding *Salmonella* associated with UK-produced hen shell eggs including food made using raw/ less-than-thoroughly-cooked hen eggs. This will determine whether there is a need to review the ACMSF risk assessment on shell eggs published in 2016. *Salmonella* Entertidis is the primary hazard associated with eggs.

Overall, the prevalence of *Salmonella* in adult laying flocks has not changed, but detection of *Salmonella* Enteritidis has roughly doubled in recent years. The percentage of flocks positive for regulated *Salmonella* serovars remains within requirements of the *Salmonella* National Control Programs.

Since the 2016 ACMSF report was published, a ban on the use of formaldehyde-based products in animal feed was implemented in 2018. This has been suggested as a possible contributor to the increased isolations of *Salmonella* in the broiler sector from 2018 but there is no evidence to support this (uncertainty).

Outbreaks associated with *Salmonella* in eggs in the UK show a similar number of cases per year, however, unlike previous years, two large outbreaks have been associated with Lion Code eggs. The implementation of whole genome sequencing for Salmonella surveillance by the UK public health agencies has now become routine in outbreak investigations.

Overall, analysis of *Salmonella* in UK-produced hen shell eggs does not indicate a need for a risk assessment at this time.

Salmonella risk profile of UK-produced hen shell eggs: Statement of purpose

1.1 Background

1.1.1 Previous risk assessment

Historically, FSA advice has been for at risk or vulnerable groups to avoid the consumption of raw or LTTC (for example, runny) eggs. This advice was due to high levels of human infection with Salmonella Enteritidis and was reported on by the Advisory Committee on the Microbiological Safety of Food (ACMSF) in 1993 and 2001 (ACMSF, 1993 and ACMSF, 2001). Decreases in the number of cases and outbreaks of Salmonella Enteritidis triggered the FSA to commission a risk assessment of the microbiological safety of eggs in the UK. This was completed in 2016, and concluded that there had been a significant reduction in the risk from Salmonella in UK-produced hen shell eggs, in particular eggs produced under the Lion Code (or equivalent) egg assurance scheme (ACMSF, 2016). Therefore, the risk to consumers from eggs produced under these schemes was assigned a risk level of 'very low' (very rare but cannot be excluded), with 'low' uncertainty (there are solid and complete data available; strong evidence is provided in multiple references; authors report similar conclusions), whilst the risk from shell eggs not produced under these schemes was considered 'low' (rare, but does occur) risk. In addition, the ACMSF concluded that for eggs produced under the Lion Code (or a demonstrably equivalent) egg assurance scheme, it would be possible to serve raw or lightly cooked eggs to individuals who may be more vulnerable to infection, including pregnant women, infants, young children, and elderly people.

Due to this change in the risk level of eggs, in 2017 the FSA and FSS updated their consumer advice on the consumption of eggs. In addition, they issued communications targeting consumers, and worked with specialist poultry vets to produce a matrix with criteria that egg assurance schemes would need to meet to be considered 'very low' risk for *Salmonella*. The purpose of the matrix is to enable independent assessment of industry schemes. The first scheme to fulfil these criteria was Laid in Britain, and the guidance was amended in July 2021 to reflect this (Food Standards Agency, 2022).

Detections of *Salmonella* on laying hen farms started to increase in 2017, and the use of whole genome sequencing (WGS) has increased the sensitivity and specificity of case ascertainment in outbreak investigations and confidence in source attribution. There have been several persistent outbreaks of *Salmonella* linked to exposure to UK produced eggs since 2017, as well as several large persistent outbreaks linked to imported eggs since 2015. Given these emerging concerns, this risk profile will examine the current situation of *Salmonella* in UK-produced table eggs, describing and defining the factors that may influence the current risk of *Salmonella* in UK-produced eggs and highlight any factors that have changed since the risk assessment provided by the ACMSF in 2016. This will be used to assess whether the assumptions underpinning the ACMSF risk assessment remain appropriate and consider whether an updated risk assessment is necessary.

1.1.2 Summary of policy

The Retained Regulation EC No. 2160/2003, which came into force on 21st December 2003, intends to limit the occurrence of certain zoonotic infections at the primary production level by implementing species-specific *Salmonella* National Control Programs (NCPs). The NCP for commercial laying flocks of chickens implements the monitoring and controls that are required in

order to meet the target for reduction in *Salmonella* prevalence that has been set in <u>Retained</u> <u>Regulation (EU) No. 517/2011</u> (amended in 2019 by <u>Retained Regulation (EU) 2019/268)</u>. The goal is for no more than 2% of adult laying hen flocks to test positive for regulated *Salmonella* serovars each year. S. Enteritidis and S. Typhimurium, including monophasic strains of S. Typhimurium, are the controlled serovars. All commercial egg producing flocks with 350 or more birds are included in the *Salmonella* in Laying Flocks NCP. Separate laws enforce the NCP, each country has their equivalent Control of *Salmonella* in Poultry Orders (CSPO): in England <u>The</u> <u>Control of *Salmonella* in Poultry Order 2007 (legislation.gov.uk), Scotland <u>The Control of</u> <u>Salmonella</u> in Poultry (Scotland) Order 2008 (legislation.gov.uk) and Wales <u>The Control of</u> <u>Salmonella</u> in Poultry (Wales) Order 2008 (legislation.gov.uk). The EC reduction objective only applies to controlled serovars in adult laying flocks, unless a flock is linked to a human foodborne disease outbreak (<u>Retained Regulation (EC) No. 1237/2007</u>, amending <u>Retained Regulation (EC)</u> <u>No. 2160/2003</u>). The CSPO establishes sampling and recording criteria for both in-rear and adult flocks, and APHA keeps track of the outcomes of testing in both age groups (APHA, 2021).</u>

The legislation stated above together with <u>Regulation (EC) No 852/2004</u>, and the support of the NCP programme, aims to reduce the risk to consumers when eating raw or less-than-thoroughly cooked eggs. Due to the contamination being monitored through the sampling and control programmes, risk management actions can be taken to protect consumers.

1.2 Question to be addressed

This report reviews the recently available evidence concerning *Salmonella* associated with UKproduced hen eggs (including food made using raw/lightly cooked UK hen eggs, not further sufficiently heat treated, for example, mayonnaise, sauces, desserts) to determine whether there is a need to review the ACMSF risk assessment published in 2016 (ACMSF, 2016).

1.3 In scope

Data and consideration in scope of this risk profile include:

- all consumers, including vulnerable consumers: infants, children, pregnant women, and elderly people as defined in the ACMSF report (2016).
- consumer's behaviours and consumption patterns.
- UK laying flocks.
- UK hen shell eggs.
- Food made using raw/lightly cooked UK hen eggs, not further sufficiently heat-treated for example, mayonnaise, sauces, desserts.
- Risk associated with using eggs and inappropriate handling and/or hygiene at all parts of the commercial egg production and supply chain including risks arising from cross contamination:
 - o feed
 - farms
 - transporting
 - packing centres
 - caterer or other food businesses preparing food using raw/lightly cooked UK hen eggs not further sufficiently heat treated for example, mayonnaise, sauces, desserts
 retail
 - ∘ retail
- trends in UK egg supply.
- identifying changes in UK industry practices, their impact on risks associated with *Salmonella* in eggs and actions taken by assurance schemes to mitigate those risks.
- data on isolations, incidents and outbreaks associated with UK eggs and outcomes of investigations into those outbreaks.

• information from EU and other countries where this informs the risk profile and the impact of practices/developments in the UK.

1.4 Out of scope

Data and considerations out of scope of this risk profile include:

- excluded consumers: severely immunocompromised individuals such as those undergoing transplant surgery etc. who will shave a highly specialised and restricted diet that will not include foods such as eggs.
- non-Salmonella related risks (including all other pathogens and antimicrobial resistance).
- eggs of other species (meaning non-chicken).
- non-UK eggs.
- non raw/lightly cooked egg and eggs products, meaning food that is sufficiently cooked (inclusive of powdered or pasteurised egg).
- risks arising from processing, catering, domestic practice, or behaviour as follows:
- sources other than the egg itself (for example, contamination of a mayonnaise by *Salmonella* originating from pork meat used at a caterer)
- risk of insufficient heat treatment of a product that was originally intended to be sufficiently cooked (for example, pasteurised eggs where the pasteurisation failed).

As this is a risk profile only, no assessment of the risk, or change in risk levels or uncertainty since the ACMSF risk assessment (ACMSF, 2016), will be undertaken.

Salmonella risk profile of UK-produced hen shell eggs: Hazard identification

Non-typhoidal *Salmonella* enterica (henceforth referred to as *Salmonella*) is widespread in domestic and wild animals and can readily pass through the food chain (World Health Organisation, 2022). Human cases of *Salmonella* are generally contracted through the consumption of contaminated food, primarily foods of animal origin (mainly eggs, meat, poultry, and milk), although other foods, including green vegetables, have been implicated with cases (World Health Organisation, 2022). Prevention of *Salmonella* requires control of the bacteria through the food chain, through animal feed, vaccination, processing, and manufacturing measures.

Salmonella is one of the leading causes of foodborne disease in the UK, with an estimated 31,601 foodborne cases in 2018, taking into account underreporting (Holland and Mahmoudzadeh, 2020). In 2019 there were 8,398 laboratory confirmed cases of *Salmonella* in the UK, primarily caused by S. Enteritidis and S. Typhimurium (UK Health Security Agency, 2021). This is also reflected in EU data (Table 1).

Across the EU for the year 2020, *Salmonella* was the causative agent in 694 foodborne outbreaks with 3,686 cases of illness (European Food Safety Authority and European Centre for Disease Prevention and Control, 2021). Salmonella was a common source of foodborne outbreaks across the UK between 2015-2020 at 27% (68/251) of total foodborne outbreaks. Eggs were implicated in 26% (18/68) of these foodborne outbreaks, making them the most common food vehicle associated with *Salmonella* outbreaks across the 5-year period (DEFRA, 2021).

 Table 1: Top five serovars of Salmonella that cause human disease in the EU in 2020 and their primary animal sources. Data adapted from European Food Safety Authority and

European Centre for Disease Prevention and Control (2021)

Serovar	Primary source
S. Enteritidis	Broilers, layers and eggs
S. Typhimurium	Broiler and pig
Monophasic S. Typimurium	Pig and broiler
S. Infantis	Broiler sources
S. Derby	Pig

The top food vehicle associated with outbreaks of Salmonella in the EU in 2020 were eggs and egg products, which were associated with 44% (37) of outbreaks (European Food Safety Authority and European Centre for Disease Prevention and Control, 2021). Of these, 25 outbreaks were associated with S. Enteritidis contamination of eggs.

The external surface of raw shell eggs may become contaminated with *Salmonella* via i) contamination with faeces after laying ii) during the laying process where the reproductive tract is contaminated. *Salmonella* may additionally migrate through the shell surface into the interior of the egg. S. Enteritidis is considered the primary serotype for cases associated with eggs and may be more able to transfer into the interior of eggs (Ricke, 2017).

Salmonella in eggs is controlled through various strategies, but primarily through the prevention of Salmonella infection of layer hens, as once hens are infected, the subsequent infection of eggs is too complex a system to control (Pande et al., 2016). Prevention of infection in layer flocks is done through a variety of interventions, including environmental control strategies and vaccination of hens (Ricke, 2017). There are a number of schemes in the UK that cover the eggs production chain, using food safety controls that prevent contamination of eggs with Salmonella to reduce the risk to consumers. This includes the British Lion Scheme (British Lion Eggs, 2022) and the Laid in Britain Scheme (Laid in Britain, 2022). These schemes require increased levels of controls compared to legislative requirements, including vaccination of hens against Salmonella, increased testing of eggs and the egg production chain and regular audits of the production system. Labelling of these eggs, for instance with the British Lion Stamp, helps inform vulnerable consumers that these are part of specific food safety schemes.

An ACMSF report (ACMSF, 2016) concluded that eggs produced under the Lion Code scheme and demonstrably equivalent schemes (later broadened to include Laid in Britain) would pose a 'very low' risk with a 'low' degree of uncertainty to UK consumers. In 2017, the FSA deemed raw or less-than-thoroughly cooked eggs and egg products as safe to consume by certain vulnerable groups due to the additional control measures put in place by these schemes. Other UK hen shell eggs that are not under one of these schemes pose a 'low' risk to public health.

Salmonella risk profile of UK-produced hen shell eggs: Exposure assessment

3.1 The egg supply chain - summary

The steps in the egg supply chain are outlined below in Table 2 and consider the key risks of each stage of the supply chain from farm to consumer. Eggs can become contaminated with *Salmonella* at various stages of the egg supply chain. They can be contaminated via direct

contamination, where the egg is infected during the formation, or indirectly after the egg has been laid. The indirect contamination can occur during the egg production process, storage, handling or food preparation, making the source difficult to identify and manage (Whiley and Ross, 2015).

Once an egg becomes contaminated, there are several points in the egg supply chain (Table 2) at which cross contamination can occur, increasing the volume of contaminated eggs and therefore the potential risk to the final consumers.

Table 2: Risk pathway depicting stages of egg production & risks associated with them in the UK adapted from (British Lion Eggs, 2021)

Stage of supply chain	Key risk (s)
On farm: Cage, Barn, Free-range including organic	Caged flocks can have higher prevalence of <i>Salmonella</i> Pests and wildlife can spread contamination within housed birds Infected feed/bedding/water/farm staff Introduction of infected birds/point of lay pullets Introduction of <i>Salmonella</i> via staff, farm visitors/ equipment such as lorries and catcher crews and egg crates for collection Insufficient cleaning may allow contamination to persist Continued production of Class B eggs by positive flock where there are other flocks on site not subject to restrictions
Transport of eggs to packing centre	Transport trays have been found to be contaminated with Salmonella Incorrect temperature controls can lead to condensation on the eggshells, which can encourage internalisation of Salmonella.
Processing and packaging: Grading machine, dirt detector, crack detector, UV system, Weighing, blood detector	Areas where the eggs come into contact can transfer <i>Salmonella</i> for example, production line belts – this is more likely when the eggs are wet UV will potentially decrease <i>Salmonella</i> contamination levels; therefore, the lack of this step may increase risk Contaminated environment of packing centre Cross contamination from infected staff
Eggs shipped to retail	Temp control failure - Lion brand eggs must be kept at a constant temperature below 20 degrees.
Retail and consumer handling	Increase in temperature may increase <i>Salmonella</i> growth within the egg Cross contamination in processes such as whisking, and contamination of surfaces Raw egg products need to follow correct de-activation processes which may be difficult in-home kitchen Handling and cross contamination of cooking surfaces where external contamination exists on egg shell

3.2 On farm

3.2.1 On-farm detections and data trends

The ACMSF's 2016 report provided a summary of long-term trends of Salmonella Enteritidis detections in broilers (Figure 1). A 70% decrease in detected S. Enteritidis in flocks occurred in 1994, and is thought to have followed wide-scale implementation of vaccination programs (Lane et al., 2014).

Figure 1: Trends in the reporting of incidents of Salmonella enterica serovar Enteritidis in chickens in Great Britain versus laboratory reporting of human S. enterica serovar Enteritidis infections, England and Wales, 1985–2011 (Lane et al., 2014).

The trend of low numbers of S. Enteritidis incidents has continued in recent years, with 27 isolations in 2018, 48 in 2019, 31 in 2020 and 9 in 2021 in UK chicken (APHA, 2022). A rise in Salmonella in laying hen flocks would lead to a rise in Salmonella detections in eggs and egg products (Lane et al., 2014).

Figure 2, Table 3 and Table 4 show the prevalence of Salmonella in laying hen flocks from 2009 to 2021.

During 2020, operators continued to take NCP samples as they usually do, and APHA official samples were also taken as usual. There may have been a reduction in BEIC surveillance and sampling due to the COVID-19 pandemic (APHA, 2021).

Since the ACMSF risk assessment in 2016, the overall prevalence of Salmonella in laying hen flocks remains the same, although there have been fluctuations year on year (Table 3, Table 4). The number of adult laying hen flocks that tested positive for any Salmonella was lowest in 2016 at 0.58% (22/ 3793). There was a peak in 2019 of 1.20% prevalence of any Salmonella spp., dropping down to 0.96% in 2021.

Figure 2: Prevalence of Salmonella in laying hen flocks tested under NCP in GB 2010-2021 (APHA, 2022).

Since the ACMSF report in 2016, there has been an overall increase in the proportion of regulated serovars and S. Enteritidis in laying flocks (Table 4). The increase in number of flocks testing positive for S. Enteritidis in 2019 and 2020 compared to previous years could also be influenced by the occurrence of risk-based enhanced sampling by APHA and BEIC on some premises (APHA, 2021). However, these remain well within the flock positive target of 2% under the NCP testing regulations.

In 2016, 0/3793 NCP eligible laying flocks were positive for regulated serovars (including S. Enteritidis). In 2017, 6/3906 (0.15%) flocks were positive for regulated serovars, all of which were S. Enteritidis. In 2018, the proportions reduced, even with an increase in the number of eligible flocks tested, the number positive for regulated serovars was 4/4106 (0.09%), 3 of which were S. Enteritidis. In 2019, there was a peak where 16/4014 (0.40%) were positive for regulated serovars and 14 of these were S. Enteritidis. In 2020, 14/3959 (0.35%) were positive for regulated serovars and 11 of these were S. Enteritidis. In 2021, there was a slight reduction in the proportions testing positive for regulated serovars at 9/3977 (0.23%), with 3 of these testing positive for S. Enteritidis (Table 3).

Year	Total NCP eligible Flock # Adult Laying Hens	Positive for regulated serovars	Positive for SE	Positive total (adult)
2009	4197	10	7	67
2010	4099	11	6	41
2011	3865	7	5	29
2012	3777	3	1	32
2013	3,687	3	2	37

Table 3: Adult laying flocks tested and positive for Salmonella, 2009 – 2021 (AHVLA, 2009, 2010, 2011, 2012; APHA, 2019b, 2019a, 2021, 2022).

Year	Total NCP eligible Flock # Adult Laying Hens	Positive for regulatedserovars	Positive for SE	Positive total (adult)
2014	3,704	2	2	36
2015	3,674	1	0	22
2016	3,793	0	0	22
2017	3,906	6	6	28
2018	4,106	4	3	26
2019	4,014	16	14	48
2020	3,959	14	11	42
2021	3,977	9	3	38

Table 4: A summary of NCP testing results for laying flocks broken down into two time periods – 2009-2016, as would have been observed in the ACMSF 2016 risk assessment, and 2017-2021.

2009-2016	All tested	All Salmonella	Regulated Salmonella	S. Enteritidis
Flocks	30,796	286	37	23
% of tested	100%	0.93%	0.12%	0.07%

2017 - 2021	All tested	All Salmonella	Regulated Salmonella	S. Enteritidis
Flocks	19.962	182	49	37
% of tested	100%	0.91%	0.25%	0.19%

FSA has been notified of several incidents of S. Typhimurium detection on layer farms in recent years – however, comparable data is not available prior to 2019, so a trend cannot be inferred.

3.2.2 Factors affecting Salmonella prevalence and levels in flocks

The ACMSF 2016 report concluded that the factors affecting levels of *Salmonella* were the introduction of National Control Programs, the combination of vaccination, rodent control and improved farm hygiene standards, together with the removal of traditional battery cage systems, which resulted in virtual eradication of infection. It highlighted that the introduction of vaccination for *Salmonella* Enteritidis in the egg industry was associated with a large reduction in human cases. A search of PubMed was completed in order to identify new evidence for factors that affect the prevalence and levels of Salmonella in flocks and also to provide context for the levels of *Salmonella* seen in the UK. Since 2016 the oldest flocks have increased from 60/70 weeks to 80/90 weeks old with a move to free-range egg production compared to caged hens (APHA, personal communication).

3.2.2.1 General

In Crabb et al., 2019, the overall finding from across multiple flocks was that the risk of contamination declined as the flocks aged. The environmental prevalence of Salmonella spp. remained similar as the flocks aged, but the prevalence in eggs varied significantly. The study also looked at pooling egg samples and found that a high prevalence in pooled eggs did not guarantee the detection of positive individual egg components in that subsample. This outcome indicates that factors other than simple environmental cross contamination are important in contributing to egg contamination and that physiological status of the hens such as variation in body weight and egg production was contributing to egg contamination (Crabb, Gilkerson and

Browning, 2019). The birds were monitored up to 50 weeks in age and the vaccination status of the flocks included is not known.

3.2.2.2 Housing

There are diverse and sometimes conflicting results on how housing system of flocks may affect the prevalence of *Salmonella* but in general, lower levels of S. Enteritidis are associated with non-cage systems (EFSA BIOHAZPanel et al., 2019). It was also suggested that the change from conventional cages to enriched cages <u>Council Directive 1999/74/EC</u> could have been associated with the initial reduction of *Salmonella* prevalence in laying hens that occurred from 2009 until 2013 as this required the laying houses to be totally cleared for refurbishment, but no studies have specifically investigated this change. The literature reviewed in this EFSA document showed that stress inducing conditions, an increase in stoking density and larger farms resulted in an increased persistence and spread of *Salmonella* within laying flocks (EFSA BIOHAZPanel et al., 2019).

There are numerous methods employed to control Salmonella in laying flocks which revolve around maintaining clean henhouses. One method is cleaning flock housing between flocks, but the effectiveness of this cleaning is very variable (Whiley and Ross, 2015). Another is the control of pests and wildlife as these can transmit Salmonella between flocks and can re-contaminate clean hen housing. Biosecurity measures and the design of housing systems can mitigate this spread via wild animals (APHA, 2021).

Due to a recent avian influenza outbreak, an Avian Influenza Prevention Zone came into force across England on 3 November 2021, and additional housing measures came into force across the UK on 29 November 2021 which were further updated on 29 March 2022 and 2 May 2022. These measures meant that it was a legal requirement for all bird keepers across the UK (whether they have pet birds, commercial flocks or just a few birds in a backyard flock) to keep their birds indoors or fully netted area and follow strict biosecurity measures to limit the spread of and eradicate the disease. This may have an effect on the *Salmonella* spread in flocks. The Avian Influenza Prevention Zone was lifted on 16 August 2022.

3.2.2.3 Salmonella vaccination of hens

Salmonella vaccination of hens increases their immunity to Salmonella (Arnold et al., 2012), prevents and reduces the intestinal colonisation of bacteria, and results in reduced faecal shedding and eggshell contamination (EFSA, 2004). It also acts to reduce bacterial colonisation in reproductive tissues (EFSA, 2004). In the UK, there is evidence that the introduction of vaccination in the broiler/ breeder sector, along with improved hygiene and biosecurity, has been crucial in breaking the cycle of persistent farm hatchery contamination and spreading of S. Enteritidis infection (ACMSF, 2016).

The vaccines currently licensed for use in GB are split into live-attenuated and inactive (Table 5). Since the ACMSF report in 2016 there have been two new vaccines licensed for use in the UK including the live vaccine Nobilis SE Live Lyophilisate for use in drinking water in 2018. The onset of immunity is 14 days after the first vaccination and 4 weeks after the third with the duration of immunity lasting for 60 weeks after completion of the 3 vaccine schedule (Veterinary Medicines Directorate, 2021b). The second is the inactive vaccine Nobilis Salenvac ETC suspension for injection for chickens, licensed in 2020. This can be injected into the birds from 6 weeks of age with the onset of immunity after the second vaccination being 4 weeks. The duration of immunity after the second vaccine varies between strains but for S. Enteritidis duration is 48 weeks (evidenced by challenge) and 90 weeks (evidenced by serology) (Veterinary Medicines Directorate, 2020).

A study by Arnold et al. (2014) found that vaccinated flocks continue shedding *Salmonella*, and nearly all the birds had positive cloacal swabs regardless of vaccination after a challenge dose. However, vaccination did decrease the amount of Salmonella found on the eggshells, and resulted in a 55% and 21% reduction for the two monophasic strains of S.?Typhimurium and a 28% reduction for the S. Enteritidis strain (Arnold et al., 2014). A study by Crouch et al., (2020) investigated the efficacy of the inactive trivalent vaccine Nobilis Salenvac ETC in broiler breeder hens. Under field conditions, this vaccine induced an immune response, producing specific antibodies against the three component serovars (S. Enteritidis, S. Infantis and S. Typhimurium), persisting at high levels until the hens were at least 56 weeks of age (39 weeks after second vaccination). There was a significant reduction in colonisation of the organs and intestines after a challenge dose (108 colony-forming units, CFU/ bird) with a reduction of 63.2% faecal shedding and 44% organ invasion of S. Enteritidis (Crouch et al., 2020).

Table 5: Currently approved Salmonella vaccines for use in poultry populations in GB adapted from source (Veterinary Medicines Directorate, 2021a).

Name	Active substance	Live/inactivated	Target species	Date of issue
Nobilis Salenvac	S. Enteritidis	Inactivated	Chickens	1995
Nobilis Salenvac T suspension for injection for chickens	S. Enteritidis and S. Typhimurium	Inactivated	Chickens	2000
AviPro Salmonella Vac T	S. Typhimurium	Live	Chickens	2002
Cevac Salmovac Lyophilisate for use in drinking water	S. Enteritidis and S. Typhimurium	Live	Chickens	2003
Gallimune SE + St, Water-in oil emulsion for injection	S. Enteritidis and S. Typhimurium	Inactivated	Chickens	2007
AviPro Salmonella Duo Lyophilisate for use in drinking water	S. Enteritidis and S. Typhimurium	Live	Chickens, ducks and turkeys	2011
Nobilis SE Live Lyophilisate for use in drinking water	S. Enteritidis and S. Typhimurium	Live	Chickens	2018
Nobilis Salenvac ETC suspension for injection for chickens	S. Enteritidis, S. Infantis and S. Typhimurium	Inactivated	Chickens	2020

3.2.2.4 Feed

Feed can harbour *Salmonella*, which can contaminate the eggs, either via the layer or through the environment. Since the 2016 ACMSF report was published, a ban on the use of formaldehydebased products in animal feed was implemented in 2018. This has been suggested as a possible contributor to the increased isolations of *Salmonella* in the broiler sector which was up by 27% in 2020 compared with 2019. The NCP data for layers, however, has shown a decrease in *Salmonella* positive flocks in 2020 and 2021 compared with 2019 (APHA, 2021), though this could have confounding factors such as an avian influenza season causing birds to be kept indoors. Further information is needed to confirm if the removal of formaldehyde were the cause, as there may have been a replacement to the formaldehyde which would maintain low levels of *Salmonella* in feed.

A study by Brooks et al., (2021) comparing the rate of infection in broiler chicken tissue after the consumption of feed inoculated with *Salmonella*, found that both serovars used, S. Enteritidis and S. Heidelberg, were able to infect the tissues. S. Enteritidis infections were more prevalent than S. Heidelberg at 68% and 9% respectively, infecting at least one tissue. S. Enteritidis infected a larger proportion of tissues at 13.1% compared with 0.7% with S. Heidelberg (Brooks et al., 2021). This study suggests that contaminated feed could potentially cause Salmonella infection in chickens (uncertainty).

3.2.3 UK assurance schemes

Lion Code and Laid in Britain are currently the only egg assurance schemes in the UK, and under the FSA advice, the only eggs that are suitable to be consumed less-than-thoroughly cooked by vulnerable groups. The Lion Code and Laid in Britain codes of practice have been updated since 2016 and are outlined in Section 7.1.

Since 2016, changes have been put in place by British Egg Industry Council (BEIC) and APHA in response to incidents, and the COVID-19 pandemic. Changes in practice for assurance schemes include (personal communication, FSA, 2022):

- improving the efficiency of vaccines. Over time this could reduce the Salmonella prevalence in flocks and reduce the risk of contamination in or on eggs
- implementing enhanced sampling for flocks over the age of 72 weeks
- all Lion Code flocks sampled over a 9-months programme, working with producers and Defra approved labs to increase the quality of sampling and the sensitivity of detection
- circulating a new instruction to depopulate Lion Code laying flocks when found with a regulated serovar
- extending the sampling to the environment of packing centres that are now used as a sentinel of Salmonella presence
- taking rapid restrictive action in packing centres in a case of a positive detection while waiting for further tests.

In 2020, during the COVID-19 pandemic, BEIC stopped live visits to sites and adopted remote video audits with trained auditors who were familiar with the sites. Operator official NCP samples and APHA official samples were taken as normal (personal communication, APHA, 2022).

3.2.4 Transmission routes of Salmonella into eggs

Salmonella transmission in eggs can be vertical or horizontal. In vertical transmission, Salmonella is introduced from ovaries or oviduct tissue that are infected to eggs, prior to the formation of the shell. Certain Salmonella serovars have the potential to colonise and infect hen ovaries or oviduct tissues. This could result in Salmonella being transferred to the yolk or albumen before the shell or membranes are formed (Hudson et al., 2015). The Salmonella serovar that typically exhibits vertical transmission is S. Enteritidis, although it is still considered a rare event. Other serovars such as S. Heidelberg, S. Infantis, and S. Typhimurium may also be vertically transmitted to some extent, but this is very variable according to strain and the management of laying flocks (ACMSF, 2016).

In hens, the intestinal, urinary, and reproductive processes all share a common orifice and therefore the egg can get contaminated (typically faecal) when laid. Horizontal transmission is typically a result of this faecal contamination on the shell surface, or contamination through environmental vectors, such as farm environment. Shell damage, such as cracks, can accelerate this process.

Vertical transmission is considered to be the major route of *Salmonella* contamination and is more difficult to control, while horizontal transmission can be effectively reduced by adequate disinfection of the environment (WHO/ FAO, 2002).

Since 2016 there has been no evidence or research found to show that new serovars of *Salmonella* have been linked to the contamination in or on shell eggs (uncertainty). However, in 2020, S. Mikawasima (a single isolate that was not related to the human outbreak strain), S. Ramatgan and S. Stanleyville were isolated from chickens in GB for the first time. Other unusual serovars included S. Bredeney (from laying hens), which was last isolated from chickens in 2014 and S. 2003; S. Stourbridge (from broilers), which was last isolated from chickens in 2014 and S. Schwarzengrund (from laying hens) and S. Braenderup (from broiler), neither of which had been

isolated from chickens in GB since 2015 (APHA, 2021). There is no evidence that these serovars can infect or persist in the egg and therefore the risk to consumers of LTTC eggs are unknown (uncertainty).

3.2.5 Penetration of eggshells by Salmonella#

Penetration of eggs with *Salmonella* is complicated, with a wide range of factors affecting the penetration throughout the egg supply chain. The major extrinsic factors are bacterial strain, temperature differential, moisture, number of organisms present, and storage conditions. The main intrinsic factors are shell defects. Cuticle and porosity had been considered major factors though current evidence suggests that bacteria are checked in their movement by structural modifications in the mammillary layer (Messens, Grijspeerdt and Herman, 2005). Microorganisms may enter the egg through pores or fissures in the shell, potentially contaminating the contents (Hudson et al., 2015).

Horizontal transmission can occur when eggs are placed in an environment which can create a negative gradient, drawing any fluids or micro-organisms across the surface of the shell and into the egg. Other transmission routes for micro-organisms are cracks or shell damage that are small enough to go unnoticed. Bacteria that enter the shell in this way are likely to be trapped by the shell membrane and will only multiply in adequate conditions. Should this occur, it would be expected that spoilage of the egg contents would be noticed (visual or olfactory abnormalities) but it could be that the bacterial density is insufficient for spoilage to be obvious (uncertainty) (ACMSF, 2016).

In a study by Messens et al. (2007), eggs from free-range hens (6%) and generic white eggs (16%), were better at resisting *Salmonella* penetration than generic brown eggs (30%), and organic and omega-3–enriched eggs (34%). A second experiment in this paper looked at effect of two types of feed on eggshell penetration (standard feed and a feed with a corn-cob mix). Eggs from hens kept in aviaries and fed corn-cob mix were more frequently penetrated (62%) than eggs of hens given standard feed (48%) (although this was not significant). Further work is needed to confirm that feed can play a role in penetration of *Salmonella* (Messens et al., 2007). No newer papers on these topics were found, therefore we do not know if these rates would be comparable to current egg production (uncertainty).

Indirect contamination of the egg contents has the highest incident between 15 minutes and 3 hours post laying when at 25°C. Experimental refrigeration of eggs at 4°C significantly decreased penetration by *Salmonella* (Miyamoto et al., 1998). Lower temperatures increase survival of S. Enteritidis on eggshells, but also limit internal growth (Khan et al., 2021). Fluctuations in temperatures cause rapid increases in *Salmonella* (Okamura et al., 2008). Incorrectly controlled changes in temperature may lead to condensation forming on the egg, which in turn increases the likelihood of *Salmonella* internalisation within the egg (Gradl et al., 2017).

A study by Lin et al., (2021) found that a novel serovar, *Salmonella* Hessarek, has emerged in salmonellosis linked to eggs and egg products in Australia and other parts of the world. As there is limited research on this serovar, their study focused on the penetration of S. Hessarek into table eggs at different temperatures. Penetration into egg contents was significantly higher in cold (eggs stored at 5°C) compared with warm conditions (eggs stored at 25 degrees) (Lin et al., 2021).

3.3 Processing and packaging

The 2016 ACMSF report acknowledged that egg exteriors could be contaminated via either direct contact or indirectly by packaging materials. Since 2016 an increased number of packing centres have made a change from single use cardboard trays to transport eggs, to re-usable plastic trays,

which should be disinfected between use. This could lead to cross-contamination between eggs if the plastic trays are not washed adequately and could be linked to outbreaks that have occurred at packing centres, but further investigation is needed to confirm this (uncertainty) (APHA, personal communication).

A study by Musgrove et al., (2009) showed that the transportation trays used to move the eggs from farm to packaging centre can become contaminated with *Salmonella* which could contaminate eggs. However, this study was performed in the USA where the vaccination of laying hens is not widespread so the extent this pathway may contribute to contamination in UK eggs is unknown (uncertainty). The study's focus was on Enterobacteriaceae, but they also analysed isolates to determine the genre, and *Salmonella* along with other bacteria were identified in the isolates tested.

Other potential areas of risk are areas where the eggs are in close contact with each other such as during packing and in the egg grader and rollers where bacteria can be transferred from one egg to another. A study by Regmi et al., (2021) found that transfer of S. Enteritidis from the eggshell surface to the egg carton or packing material could occur (uncertainty). They inoculated wells of egg cartons with S. Enteritidis and uninoculated eggs were placed into the cartons at 4°C and 25°C. 8 eggshell samples tested positive for S. Enteritidis, 6 from polystyrene foam packaging and 2 from plastic cartons. This indicates a possible risk of contamination from packaging material to egg shell surface (uncertainty) (Regmi et al., 2021).

Investigations of two outbreaks of human S. Enteritidis cases in recent years in the UK has revealed that dissemination of the pathogen could occur at packing centres (Section 4.5).

3.4 Retail, catering and consumer handling

Food safety concerns at the retail, catering and consumer level highlighted by the ACMSF in 2016 include poor hygiene and preparation practices, failure to observe Best-before dates, and inappropriate storage that encourages condensation on eggshells.

External contamination present on the outer surface of the eggs poses risks in relation to cross contamination of the egg contents, the food handlers' hands and the food preparation environment. Cross contamination can additionally occur during the breakage and processing of eggs. Research by Humphrey et al. (1994) showed that breaking inoculated eggs by hand led to contamination of the hands and whisking of eggs led to distribution of *Salmonella* over 40cm away from bowl, which survived for 24 hours (Humphrey, Martin and Whitehead, 1994). The prevalence of this in domestic settings however is difficult to ascertain (uncertainty).

Effective food hygiene such as effective cleaning and the avoidance of cross-contamination within food processing environments is important for reducing the risk from cross-contamination. While much training focuses on food handlers in larger scale food processing and catering kitchens, good practice advice is likely to be of value to any and all domestic food handlers, especially if they are involved in the preparation of food for higher risk groups or individuals (ACMSF, 2016).

There has not been a significant change identified in consumer or retail handling since the ACMSF report in 2016. An outbreak of S. Enteritidis in Spain in 2021 involving two cases of illness suspected that cross-contamination with unclean eggs in the fridge was the source of infection (uncertainty) (European Centre for Disease Prevention and Control, European Food Safety Authority, 2022). This shows the importance of ensuring hygienic practices while handling eggs.

An FSA study on UK shell eggs in 2003 found a contamination prevalence with *Salmonella* of 0.34% (Food Standards Agency, 2004). No recent studies on the occurrence of *Salmonella* in UK retail eggs have been conducted.

3.5 Egg products

Egg products are defined as 'processed products resulting from the processing of eggs, or of various components or mixtures of eggs, or from the further processing of such processed products' (Retained Regulation EC No 853/2004). Processing can include heating, smoking, curing, drying, marinating, etc., to manufacture products such as refrigerated liquid egg, powdered egg, or frozen egg products. These are widely used in sauces, desserts, pasta, processed meats and fish.

The 2016 ACMSF report acknowledges that manufacturing of egg products can result in crosscontamination with a range of bacteria from the shells, and it can include the use of eggs that have been downgraded, such as cracked eggs or eggs contaminated with *Salmonella* (but not S. Enteritidis). Pasteurisation of liquid egg is an important step in preventing foodborne illness, as is effective refrigeration of pooled eggs.

The ACMSF also identified *Salmonella* outbreaks listed with egg products including – mayonnaise,(3 outbreaks, one of which states the mayonnaise was made with raw shell eggs), egg fried rice (5 outbreaks) and various egg dishes such as desserts, cress sandwiches, Yorkshire pudding, egg noodles and pasteurised liquid egg white. This association of salmonellosis with eggs products has continued (see Section 4.5).

When preparing mayonnaise, pH and temperature are the key factors in controlling *Salmonella*. A pH of 4.6 and a temperature of 37°C was the most effective at reducing S. Typhimurium (lost viability after 24 hours), and 23°C was less effective but still more effective than 4°C where S. Typhimurium survived for more than 10 days. There is a risk that if the pH is not accurately measured and controlled, then storage above 4°C could allow growth. The survivability in acidified environments at 4°C is also important in terms of cross-contamination within the kitchen environment (Keerthirathne et al., 2019). Homemade preparations are potentially more risky with consumers less likely to have easy access to pH meters and recipes may follow taste preference rather than a safe ratio of acid to egg (Cardoso et al., 2021).

3.6 Consumption of raw or undercooked eggs

Based on industry data, the consumption of eggs per person per year in 2021 was 202. This has increased from 171 eggs per person per year in 2004 and is a small increase from 195 in 2016 when the ACMSF risk assessment was written. The majority of UK eggs are sold through retail. The percentage sold at retail in 2021 was estimated at 65%, an increase from 48% in 2004. There was particularly strong growth between 2008 and 2021 where retail sale of eggs grew by 50% (British Lion Eggs, 2022). The ACMSF risk assessment did not report specifically on consumption of raw or LTTC eggs by vulnerable groups.

Data from the national diet and nutrition survey (NDNS) (DHSC, 2013, PHE and FSA, 2014, 2016, 2018, 2020) on the consumption of raw and poached (used as a LTTC equivalent and include those used in recipes) by vulnerable groups (under 5s and over 65s) are presented in Table 6. No specific information on immunocompromising conditions are given in the survey, therefore this group has not been fully represented in this data (uncertainty).

In the data from NDNS, over 40% of foods made with raw eggs contain less than 5% eggs in proportion of the whole food product. This indicates that in these cases, raw eggs are used in small quantities as an ingredient in a recipe containing other ingredients. The quantity of both raw and poached eggs consumed by under 5s is lower than that consumed by the over 65s.

Table 6: Data of amount of raw and poached eggs (with recipes) consumed by vulnerable groups from the NDNS data 2015-2020. For reference, a raw egg (without shell) weighs around 55 g.

Consumed including in recipes	Number of consumers: 4 to 18 months (2683 respondents)	1.5 to 4 years (1157 respondents)	65+ years (1538 respondents)	Mean acute consumption (g/person/day)*: 4 to 18 months	1.5 years to 4 years	65+ years
Raw eggs	21	39	74	1.3	4.3	3.2
Poached eggs	17	32	140	54	61	78
Both raw and poached eggs	38	71	205	26	31	52

* Rounded to 2 significant figures

Self-reported rates of consumption of eggs eaten raw, LTTC and thoroughly cooked indicated that, of the respondents that consume eggs at home, only a small percentage eat raw eggs once a week or more. A higher percentage eat LTTC eggs and thoroughly cooked eggs (wave 2 of Food and You 2 (FSA, 2021)). 61% of respondents never eat raw eggs, 22% never eat LTTC eggs and 8% never eat thoroughly cooked eggs (Table 7).

Table 7: Taken from Food and You 2 survey on the eating habits of UK consumers (FSA,2021).

Frequency	raw (eggs that are uncooked for example, in homemade mayonnaise or homemade desserts like mousse or soft meringues)	LTTC (eggs that have a runny yolk for example, soft boiled)	cooked thoroughly (eggs that have firm yolk for example, hard boiled)
Every day	1%	1%	2%
Most days	2%	4%	5%
2 to 3 times a week	4%	12%	15%
About once a week	7%	24%	27%
2 to 3 times a month	4%	15%	16%
About once a month	4%	11%	15%
Less than once a month	14%	9%	11%
Never	61%	22%	8%

Salmonella risk profile of UK-produced hen shell eggs: Hazard characterisation

4.1 Disease characterisation

The symptoms of *Salmonella* infection can range from asymptomatic carriage to severe diarrhoea and septicaemia. The incubation period of non-typhoidal *Salmonella* infection is usually from 12 to 96 h, although periods of 96 to 144 h (4 to 6 days) are not unusual and incubations of 7 to 9 days and occasionally longer also occur (Eikmeier, Medus and Smith, 2018). The principal symptoms of mild fever, nausea and vomiting, abdominal pain, and diarrhoea last for a few days but can persist for a week or more.

Whilst the illness is usually self-limiting, it can be more severe in vulnerable groups, including the elderly, pregnant women, the young and immunocompromised, leading to systemic infection and death. Currently there is no human vaccine available for non-typhoidal *Salmonella*. After

symptoms have subsided, carriage and shedding of the organism can occur for a few weeks, up to months.

The median infective dose required to infect 50% of the population (ID50) for non-typhoidal *Salmonella* is generally high, estimated from volunteer feeding studies to be 104 CFU, but varies between serovar and food vehicle, for instance the ID50 in eggs is suggested to be as low as 1.09 x 101 CFU (Teunis et al., 2010).

4.2 Survival and growth

Salmonella growth has been observed between 5 and 47 °C with an optimum growth temperature of 37 °C. Salmonella are readily destroyed by pasteurisation temperatures and the standard 70 °C for two minutes cooking advice is normally sufficient (FSA, 2018). This is affected by the food composition, however, for example in low water activity foods, such as peanut butter, the survival of Salmonella at 70 °C

is increased (Beuchat et al., 2013). Curing and fermentation are also generally effective at reducing bacterial loads (Mandal and Kwon, 2017), but are unlikely to be utilised in the production of eggs or egg based products or recipes (uncertainty).

The minimum water activity that permits growth of *Salmonella* is 0.94, however, cells are able to survive in dried foods for extended periods of time (Beuchat et al., 2013). Cells exposed to desiccation are also more tolerant to heat, UV, and chemical treatments. It has been reported that *Salmonella* can grow at pH 3.8-9.5 although the optimal pH for growth is 7.

The age of the egg also has an impact on the viability of *Salmonella*, as the pH of an egg white increases, from neutral (pH 7) to alkaline (pH 9 – 9.5), through the loss of carbon dioxide via the eggshell pores. The optimum pH for Salmonella growth is pH 7, although it can grow between pH 3.8-9.5. Therefore, a pH above 9 would reduce the viability of *Salmonella*. An additional confounding factor in raw egg-based sources and dressings is the fat content. High fat content decreases the water activity which increases the survival of *Salmonella* during thermal treatment (Szpinak, Ganz and Yaron, 2022) and lowers the dose required for infection. Fat free mayonnaise was found to have a faster decrease in *Salmonella* levels compared to full-fat mayonnaise at the same pH (Keerthirathne et al., 2019). The addition of other flavouring compounds such as garlic and salt may also have an impact of the survivability of *Salmonella* as these compounds are often used as antibacterial agents.

Chlorine and ozone-based treatments have been shown to reduce *Salmonella* counts on a variety of foods, although are unlikely to be utilised in the egg supply chain (uncertainty). In addition, UV treatment has been shown to reduce bacterial counts in foods (Mandal and Kwon, 2017), and is often utilised in the egg supply chain (British Lion Eggs, 2021), but it is unknown how effective this process is (uncertainty) and will not be effective for contamination internalised within the egg.

Cold temperatures have been shown to decrease the growth of *Salmonella* in the egg yolk and albumen but also increase the survival of *Salmonella* on the egg surface (Khan et al., 2021). A study of eggs contaminated with *Salmonella* from New Zealand found that the viability of Salmonella declined over time on the shell surface of the egg at 15C and at 22C. They also found that the bacterium survived better on visibly clean eggshells at 15C than at 22C. Survival on the eggshell was enhanced by the presence of faecal contamination and the study found no contamination within the egg contents at either temperature (Kingsbury, Thom and Soboleva, 2019).

4.3 Antimicrobial resistance

The ACMSF report, (2016), observed that resistance profiles of *Salmonella* strains can reflect the country of origin – for instance nalidixic acid resistance is typically found in isolates from countries where fluoroquinolones are inexpensive and routinely used. An FSA survey of *Salmonella* contamination of non-UK eggs on retail sale between March 2005 and July 2006 found that the majority of isolates (83.2%) were resistant to one or more antimicrobial drugs of which most were resistant to nalidixic acid with reduced susceptibility to ciprofloxacin (78.6%) (ACMSF, 2016). Ten *Salmonella* strains found in UK eggs were fully sensitive to 10 antimicrobial compounds (Food Standards Agency, 2004).

The *Salmonella* in animals and feed report from APHA, for 2021, found that most isolates from chickens were fully susceptible to a panel of 16 antimicrobials (74.3%).

Resistance to sulphonamide compounds (15.7% of isolates), tetracycline (10.8%) and streptomycin (10.5%) were the most common in chickens. Cefotaxime, ceftazidime or ciprofloxacin resistance was not detected in S. Enteritidis in 2021 (APHA, 2022). A study on *Salmonella* isolates received at Public Health England's Gastrointestinal Bacteria Reference Unit between 2014 and 2015 found full susceptibility in 63% of isolates. Resistance to tetracycline was most commonly observed (26%), followed by sulphonamide resistance (24%) and ampicillin resistance (21%) (Neuert et al., 2018)

4.4 Salmonella cases in the UK

Prior to the 1980s, S. Enteritidis was rarely responsible for disease. During the late 1980s, it was the cause of a foodborne epidemic infecting more than 500,000 people in England and Wales, traced back to contaminated chicken but especially shell eggs (ACMSF, 2016). Infections started to decline with the introduction of risk management actions such as vaccination and flock hygiene programmes (see Figure 1).

Levels of *Salmonella* over the last 10 years have been relatively steady (Figure 3 and Figure 4). In 2020 and 2021, the COVID-19 pandemic has resulted in a decrease of reported gastrointestinal illness, including *Salmonella* levels (Love et al., 2022). This is likely due to a combination of factors, including increased underreporting of gastrointestinal illness, reduced foreign travel, UK lockdowns and implementation of enhanced hygiene measures. It is uncertain whether this downward trend in *Salmonella* cases will persist.

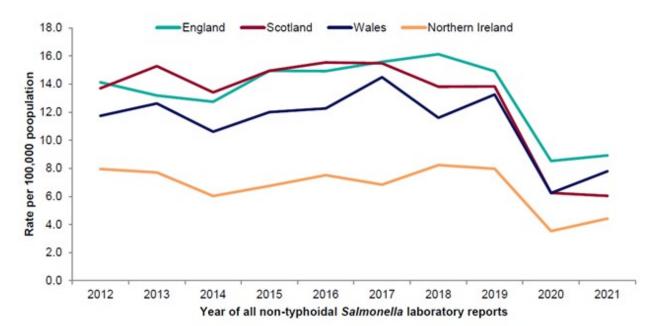
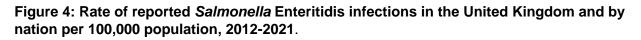
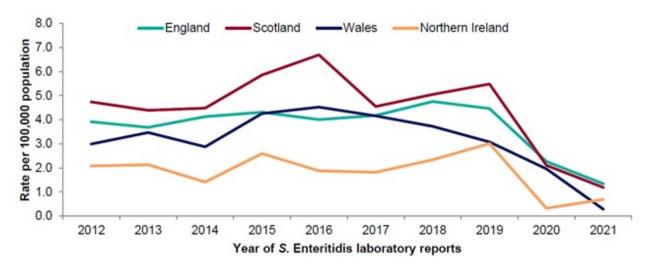


Figure 3: Rate of reported non-typhoidal *Salmonella* infections by country per 100,000 population for 2012-2021.

Source: (EFIG 2022 (UKHSA, PHS, PHW, PHA, 2021)) The number of reported cases is shown for the four nations, England, Scotland, Wales and N. Ireland, with year of laboratory report against rate per 100,000 of population. Please note that data for 2020 onwards is provisional.





Source: (EFIG 2022 (UKHSA, PHS, PHW, PHA, 2021)). Please note that data for 2020 onwards is provisional.

4.5 Summary of foodborne outbreaks associated with *Salmonella* and eggs

The 2016 ACMSF report includes descriptions of 26 salmonellosis outbreaks with confirmed or putative links to eggs or egg products over the period 2009 – 2014, with a total of 1176 cases. Most of these outbreaks (22/26) reported fewer than 40 cases. The largest outbreaks, in 2009 (489 cases), 2011 (263 cases) and 2014 (100 cases), were linked to imported eggs (ACMSF, 2016). A further outbreak of S. Typhimurium in 2010 with 81 cases was linked to consumption of UK-produced duck eggs. Four small general outbreaks were linked to UK produced eggs, of which only one outbreak of 33 cases in 2009 was linked to Lion Code eggs.

The data collected from UKHSA non-typhoidal *Salmonella* outbreaks is shown in Table 8. This data is provisional and incomplete and does not indicate whether the eggs associated with the outbreaks were Lion Code or imported and which of the outbreaks are recurring with the data only available up to 2019. Additional data has been requested from UKHSA. Therefore, this presents a key uncertainty. Between 2015 and 2019, a total of 954 confirmed cases of salmonellosis were investigated as part of outbreak investigations and determined to be associated with consumption of eggs and/or egg products, in a total of 15 reported investigations. The majority of outbreaks (10/15) reported fewer than 45 cases. The largest outbreaks occurred in 2016 (158 cases), 2017 (162 cases) and 2018 (259 cases).

Table 8: Foodborne outbreaks of non-typhoidal Salmonella reported in England and Wales associated with eggs and/or egg products (UKHSA, 2016, 2018, 2021a, 2021b, 2021c.)

Year	Agent	Total affected	Laboratory confirmed	Hospitalised	Deaths	Setting	Food description
2015	Salmonella Enteritidis	3	3	0	0	Hotel	eggs and egg products

Year	Agent	Total affected	Laboratory confirmed	Hospitalised	Deaths	Setting	Food description
2015	Salmonella Enteritidis	2	2	1	0	Residential institution	eggs and egg products
2016	Salmonella Enteritidis	90	116	2	0	National: multiple exposure settings	eggs
2016	Salmonella Enteritidis	95	95	0	0	National: multiple exposure settings	eggs
2016	Salmonella Enteritidis	116	158	14	0	National: multiple exposure settings	eggs
2016	Salmonella Enteritidis	21	13	1	0	Household	Tiramisu
2017	Salmonella Enteritidis	162	162	0	0	National: multiple exposure settings	eggs
2017	Salmonella Enteritidis	27	27	1	0	National: multiple exposure settings	eggs
2018	Salmonella Enteritidis	26	26	0	0	Multiple places of exposure	eggs
2018	Salmonella Enteritidis	259	259	0	0	Multiple places of exposure	eggs
2019	Salmonella Enteritidis	44	44	11	0	Multiple places of exposure	eggs
2019	Salmonella Enteritidis	2	2	1	1	Multiple places of exposure	eggs
2019	Salmonella Enteritidis	22	22	0	0	Multiple places of exposure	eggs
2019	Salmonella Enteritidis	22	22	0	0	Multiple places of exposure	eggs
2019	Salmonella Enteritidis	3	3	1	0	Multiple places of exposure	eggs

*this data is provisional a full dataset has been requested from UKHSA.

Not all outbreaks are microbiologically linked to an implicated food vehicle as food vehicles are not always identified or available for microbiological testing, and the level of evidence derived through epidemiological and microbiological investigations varies with some outbreaks having stronger epidemiological evidence in support of a link between the implicated food product and the outbreak than other outbreaks (uncertainty).

Additionally, for some outbreaks, not all individuals linked to the outbreak will have laboratory confirmation of illness. The number of hospitalisations reported is only known for cases which received public health follow-up, for example via interviews. Where individuals are reported to have died, it is usually not known whether the cause of death was directly related to the outbreak (uncertainty).

Some outbreaks highlighted in Table 8 were recurring, intermittently persistent, cluster events, which occurred across multiple years with epidemiological evidence for some of these explained below. While published UKHSA data is not available for 2020- 2022, FSA records show no new outbreaks have occurred. Additional cases were recorded for ongoing outbreaks, as described below.

A recurring cluster of S. Enteritidis t5:175 and t5:360 was re-opened and investigated after first being identified in 2016. In 2016 the outbreak was linked to eggs imported from Poland. There have been 533 cases in this outbreak since June 2014; 251 cases in 2018 to date, with 77 alone in September 2018 (the highest number of cases reported in any month in this outbreak). A cluster of these cases was linked to liquid egg product which was produced in a factory linked to contamination with the outbreak strain t5:360. WGS was carried out on the liquid egg product with the result indistinguishable from that of an isolate found in an egg from Poland during the 2016 outbreak.

A recurring cluster of S. Enteritidis t5:2669 has had 117 cases to date from April 2016 – March 2020. Epidemiological investigations indicated that four farms associated with this cluster in Nottinghamshire, Northamptonshire, Dorset and Kent were all part of the BEIC Lion Code egg assurance scheme. All farms used the same packing centre. There is the potential that cross contamination occurred at the packing centre with eggs from non-positive farms as the outbreak isolate was found in the packing centre environment. It was not clear if infection was moved from laying farms to packing centre or vice versa (uncertainty) (Internal FSA communication, 2022).

A recurring cluster in 2019 identified samples of S. Enteritidis from two farms and environmental samples from an egg packing centre. It linked to a t5:180 UK lineage sub-cluster which was first identified in early 2019 and has been intermittently persistent in 2019, 2020 and early 2021. To date isolates have been linked to four farms and one packing centre. Epidemiological investigations indicated most cases with onset of illness between June and October 2020 had purchased and consumed Lion Code eggs predominantly from two retailers. As of February 2022, 109 cases of S. Enteritidis t5:180 UK lineage cases have been identified using whole genome sequencing (WGS)

4.6 Detection

Since the ACMSF report in 2016 whole genome sequencing has been implemented as a routine method for further characterisation of *Salmonella*. WGS can be used to perform genome characterisations of isolates as well as identify antimicrobial resistance genes.

Across the EU WGS is already in use in different sectors including public health and food safety (EFSA et al., 2018). The UKHSA have been routinely sequencing all presumptive *Salmonella* isolates since 2014 (Chattaway et al., 2019). The epidemiological precision offered by WGS for outbreak investigation and attribution is much more valuable than previous systems that were used (ACMSF, 2016) and can link cases of illness to outbreaks that would have been deemed unrelated by previous methods such as PCR, gene probes and enzyme-linked immunosorbent assay (ELISA)(ACMSF, 2016).

The high resolution WGS typing of isolates for pathogen strain discrimination has enhanced the detection of outbreaks and enables 'sensitive and specific' case definitions to be applied, improving case ascertainment, focussing outbreak investigations and increasing the strength of association in analytical studies to identify the implicated food vehicles. Where possible, integration of the microbiological, genomic and epidemiological data derived from analysis of the human disease data with that from animal samples, environmental sampling or the food chain, has significantly improved the ability to identify the source of the outbreak and better understand transmission of contamination through food supply chains. The use of WGS has also resulted in an enhanced ability to detect re-emergence of outbreaks and trace them back to the same source of contamination as previously identified when control measures have not been fully effective in eliminating contamination (Kathie Grant et al., 2018).

Implementation of WGS has enabled the consolidation of multiple local/regional outbreaks into single national level outbreaks based on the WGS and epidemiological information obtained during the investigations. This has resulted in a higher proportion of outbreaks being identified to be national rather than local/regional outbreaks with an associated increase in case numbers. Therefore, while consideration of total numbers of outbreaks reported is useful, these data are affected by whether WGS is used or not.

Both the re-emergence of cases associated with outbreak clusters and the consolidation of multiple outbreaks into large national outbreaks of long duration has meant that comparison of number of foodborne outbreaks and number of associated cases pre and post the implementation of WGS should be undertaken with caution, and the foodborne outbreak surveillance data reported for the years prior to implementation of WGS (pre-2014 for *Salmonella*) is not directly

comparable to the data held for subsequent years. Therefore, the size of the outbreak and number of individuals affected should be considered together with the information given on the overall numbers of outbreaks.

Salmonella risk profile of UK-produced hen shell eggs: Conclusions

This report reviewed recent evidence, including scientific peer-reviewed publication, and UK surveillance data, to determine whether the assumptions underpinning the ACMSF 2016 risk assessment on UK shell eggs remain appropriate and consider whether an updated risk assessment is necessary.

There is limited new evidence in the scientific literature with regards to *Salmonella* in eggs and the egg production environment since the risk assessment undertaken by the ACMSF in 2016.

Surveillance since 2016 shows there has been an increase in prevalence of *Salmonella* Enteritidis and regulated *Salmonella* serovars in general (but within the 2% NCP requirements) in laying flocks. The prevalence levels have not returned to the high levels seen in 80s and 90s, before vaccination was introduced.

Since 2016, the oldest flocks have increased from 60/70 weeks to 80/90 weeks old, which could lead to an increase in *Salmonella* prevalence in older flocks, although further research is needed to support this.

A ban on the use of formaldehyde-based products in animal feed was implemented in 2018. This has also been suggested as a possible contributor to the increased isolations of *Salmonella* in the broiler sector from 2018, although additional research is required.

A number of packing centres have made a change from single use cardboard trays to transport eggs, to re-usable plastic trays, which should be disinfected between use. This could lead to cross-contamination between eggs if the plastic trays are not washed adequately and could be linked to outbreaks that have occurred at packing centres in recent years, however more research is required to confirm this.

There is a lack of detail in data provided for outbreaks and additional data has been requested from UKHSA.

The total number of outbreaks and cases linked to consumption of eggs and egg products per year has not changed significantly since the 2016 ACMSF risk assessment. However, the ACMSF identified only one small outbreak in 2009 due to Lion Code eggs, whereas our data show at least two outbreaks with over 100 cases per outbreak associated with UK Lion Code eggs since 2016.

Overall, analysis of *Salmonella* in UK-produced hen shell eggs does not indicate a need for a risk assessment at this time.

5.1 Uncertainties and evidence gaps

5.1.1 Uncertainties

The key uncertainties of this risk profile are presented below in order of importance based on their effect on the risk levels:

- the number of outbreaks/cases associated with UK eggs and egg products, including eggs produced in assurance schemes; further information on the specific egg products involved in outbreaks (UKHSA data pending).
- the effect of the COVID-19 pandemic on the prevalence of *Salmonella* in eggs, underreporting of *Salmonella* cases, changes in the egg supply chain, including consumption behaviours, and whether site visits and audits done through assurance schemes have returned to usual pre-COVID levels or the affect this will have on the quality assurance of these schemes.
- lack of data on the transference of *Salmonella* during processing whether *Salmonella* from contaminated eggs / eggshells could be transferred to the shells of other eggs during packing.
- investigation of Salmonella levels in eggs and egg products at retail
- the effects of flock types, such as free-range vs caged, and older flocks on the prevalence of *Salmonella*.
- the effect of the prolonged 2021-2022 Avian Influenza Prevention Zone on *Salmonella* levels in flocks.
- the effect of the formaldehyde ban in feed on Salmonella levels in flocks.
- the effectiveness of UV treatments applied in the eggs supply chain on *Salmonella* prevalence.
- how effectively *Salmonella* isolates currently circulating within the egg supply chain can proliferate within the egg albumen.
- the prevalence or presence of novel serovars of *Salmonella* and whether these are a significantly higher risk compared to current strains when found in the egg supply chain in terms of virulence and antimicrobial resistance.
- whether internal contamination with *Salmonella* will lead to spoilage characteristics of eggs perceptible to consumers leading to a reduction in the consumption of these eggs.
- data and information on consumption of raw eggs, for example in cookie dough or as a protein source, without any cooking or mitigation processes.

5.1.2 Evidence gaps

These evidence gaps should be considered prior to undertaking a full risk assessment or commissioning research around *Salmonella* in eggs:

- UKHSA data for outbreaks 2016-2022
- change from cardboard to plastic trays at many packing centres & the effect on *Salmonella* cross-contamination
- older flocks and effects on prevalence of Salmonella
- impact of removing formaldehyde-based products from feed on prevalence of Salmonella
- literature on virulence of new serovars of Salmonella since 2016
- the effect of UV treatments applied in the eggs supply chain on Salmonella prevalence

Salmonella risk profile of UK-produced hen shell eggs: References

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Salmonella risk profile of UK-produced hen shell eggs: Appendix

7.1 UK assurance schemes

The Lion Code of Practice states:

- all Lion hens and eggs guaranteed British
- hens vaccinated against Salmonella Enteritidis and Salmonella Typhimurium
- registration and a unique 'passport' system, ensuring complete traceability of hens, eggs and feed
- increased hygiene controls and Salmonella testing of all flocks in the integrated egg production chain, in excess of the National Control Programme, including turnaround swabbing of breeding, pullet rearing and laying flocks; and packing centre hygiene swabbing
- regular egg testing (not included in National Control Programme)
- stringent feed controls, including production of feed to Universal Feed Assurance Scheme (UFAS) standards
- Lion Quality eggs stamped on farm with the farm code and production method
- best-before date and Lion logo printed on the shell of Lion Quality eggs as well as on the egg box
- regular independent auditing, including unannounced audits, of all producers and packers in the Lion scheme, in accordance with the ISO 17065 standard.

The Laid in Britain code of practice was updated in August 2021 and states:

- prior to joining the Laid in Britain assurance scheme, all producers must be audited to this Code and compliance assessed. Any non-compliances must be rectified before acceptance to the scheme.
- all producers will be robustly audited on an annual basis by an independent and suitably trained auditor. Any critical non-compliance will result in immediate suspension from the scheme, until rectified and verified. Any non-critical non-compliance must be rectified in a timescale agreed with the scheme.
- in addition to the critical non-compliances, any producer who fails to have a robust protocol for traceability and recall will be immediately suspended from the scheme.