Animal & Plant Health Agency

Diagnostic and monitoring standards recommended to detect positive flocks, assess prevalence and take corrective actions against *S*. Enteritidis and *S*. Typhimurium

Dr. Doris Mueller-Doblies
Animal and Plant Health Agency (APHA)
United Kingdom

Introduction

Salmonellosis is one of the most important food borne illnesses across the European Union, with 82,694 confirmed cases and an EU notification rate of 20.4 cases per 100,000 reported from 27 member states in 2013. The number of human salmonellosis cases has been declining steadily over the past five years, and it is likely that the decrease in human cases was the result of both the successful implementation of the *Salmonella* control programmes across the different chicken and turkey production sectors and of several different control measures which were implemented on farms and along the food chain[i]. This review will give an overview over the history of food borne salmonellosis, describe current EU legislation, which has been instrumental in reducing the *Salmonella* prevalence in poultry production, and discuss other factors which have contributed to the fact that the number of human salmonellosis across the EU is at its lowest level in years.

Human salmonellosis in the past three decades

In the mid to late 1980s, a massive *Salmonella* epidemic began to emerge in the United Kingdom, and data from other countries around the world indicate, that an increase of *S*. Enteritidis-cases took place at the same time at least in North America, South America and several countries in Europe[ii]. It is not exactly clear how and why *S*. Enteritidis could cause such an epidemic, but infected eggs clearly played an important role. *S*. Enteritidis is more skilful than other serovars at colonizing the reproductive tract of laying hens, therefore leading to a higher proportion of eggs being contaminated in the contents compared to infection with other serovars[iii]. Infected breeding eggs were an important source of infection for laying hen flocks, which in turn produced infected table eggs.

In 1990, the WHO surveillance system suggested that *S*. Enteritidis had become a predominant serovar in many countries, although different strains were involved in different countries, with phage type 4 being predominant in the United Kingdom and phage types 8 and 13b being predominant in the United States[iv]. In the United Kingdom, a first drop of human cases was seen after the introduction of a vaccine for breeding chickens in 1994, followed by a more substantial decline after the introduction of a vaccine for laying flocks in 1997[v]. Several control measures have been implemented by the poultry industry over the past two decades, which have undoubtedly contributed to a reduction in prevalence of *Salmonella*-positive chicken flocks. These include vaccination of breeding and laying birds against *S*. Enteritidis and *S*. Typhimurium, improved general hygiene and biosecurity, improved pest control, improved control over feed, improved cleaning and disinfection standards and introduction of an all-in-all-out system where possible. A study performed on British laying farms identified these factors to be associated with a reduced risk of *S*. Enteritidis infection[vi].

Over the past three years, a substantial drop in the number of human salmonellosis cases has been observed throughout the European Union[vii], and figure 1 shows data published by the European Food Safety Authority [EFSA Journal 2015;13(1):3991]. Information on *Salmonella* serovars from cases of human infection was available from 25 member states (MS). As in previous years, the two most commonly reported *Salmonella* serovars in 2013 were *S.* Enteritidis and *S.* Typhimurium, representing 39.5 % and 20.2 %, respectively, of all reported serovars in confirmed human cases (N=73,627). *S.* Enteritidis continued to decrease, with 4,760 fewer cases reported in the EU in 2013 than in 2012 and with a decrease in confirmed cases of 19.3 % compared with 2011. In the two-year period from 2011 to 2013, cases of *S.* Typhimurium decreased by 26.0 %. Cases of monophasic *S.* Typhimurium 1,4,[5],12:i:-, however, increased by 68.8 %, with four additional countries reporting this variant in 2013 compared with 2011 (3rd most common serovar).

Figure 1: Distribution of reported confirmed cases of human salmonellosis in the EU/EEA, 2011-2013, by the 20 most frequent serovars in 2013.

		2011			2012			2013		
	Salmonella Serovar	Cases	Member State	%	Cases	Member State	%	Cases	Member State	%
1	Enteritidis	36.064	27	44.6	33.850	27	41.2	29.090	27	39.5
2	Typhimurium	20.068	27	24.8	18.216	27	22.2	14.852	27	20.2
3	Monophasic Typhimurium	3.739	10	4.6	5.932	12	7.2	6.313	14	8.6
4	Infantis	1.760	25	2.2	2.007	26	2.4	2.226	25	3.0
5	Derby	710	22	0.9	732	21	0.9	818	21	1.1
6	Stanley	516	22	0.6	1.115	20	1.4	813	21	1.1
7	Newport	803	23	1.0	770	21	0.9	714	21	1.0
8	Kentucky	579	22	0.7	647	23	0.8	651	23	0.9
9	Agona	476	21	0.6	470	18	0.6	581	24	0.8
10	Virchow	495	25	0.6	544	20	0.7	571	22	0.8
11	Muenchen	187	18	0.2	253	20	0.3	448	17	0.6
12	Napoli	320	14	0.4	376	16	0.5	434	14	0.6
13	Bovismorbificans	423	19	0.5	421	20	0.5	412	20	0.6
14	Saintpaul	384	18	0.5	372	18	0.5	401	18	0.5
15	Montevideo	375	18	0.5	298	18	0.4	375	18	0.5
16	Panama	259	14	0.3	705	14	0.9	352	16	0.5
17	Brandenburg	272	13	0.3	303	17	0.4	274	15	0.4
18	Oranienburg	371	18	0.5	315	16	0.4	274	15	0.4
19	Hadar	291	18	0.4	307	20	0.4	267	19	0.4
20	Rissen	250	17	0.3	293	19	0.4	266	20	0.4
21	Other	12.690	-	17.7	14.550	-	17,7	13.745	-	18.7
	Total	**80.782**	**27**	**100.0**	**82.183**	**27**	**100.0**	**73.627**	**27**	**100.0**

Source: EFSA Scientific Report: The European Union summary report on trends and sources of zoonoses, zoonotic agents and food-borne outbreaks in 2013 http://www.efsa.europa.eu/en/efsajournal/doc/3991.pdf

EU legislation

Regulation (EC) No. 2160/2003[viii] and Directive 2003/99/EC[ix] formed the basis for the introduction of *Salmonella* legislation across the European Union, and separate regulations for each production sectors were introduced in the years to follow. *Salmonella* legislation is currently only targeted at chicken and turkey production above a certain holding size, i.e. small producers are not included, as are producers of other poultry species, such as ducks, geese and game birds. *Salmonella* surveillance of breeding chickens started in 2007, followed by laying chickens in 2008, broiler chickens in 2009 and turkeys in 2010. Each member state was required to draw up a National Control Programme, and the Competent Authority is responsible for delivering the programme and submitting data to the European Commission on an annual basis. The National Control Programmes are “flock-based”, i.e. the unit to be tested is a flock of poultry. Only the two most prevalent serovars in humans (*S.* Enteritidis and *S.* Typhimurium) are regulated in all production sectors, with the exception of chicken breeding flocks, where *S.* Hadar, *S.* Virchov and *S.* Infantis are also regulated to some extent. The programmes are based on both operator sampling and official sampling, which are overseen by the Competent Authority. Proper collection of samples, testing according to ISO 6579 and reporting of positive isolates are essential pillars of the surveillance system.

Figure 2: Surveillance scheme for breeding and laying flocks in the European Union, based on self-monitoring (operator sampling) and official monitoring.

	Phase	Self-monitoring	Official monitoring	Serovars
Breeders Any flock > 250 birds	**Rearing**	▪ 1 day ▪ 4 weeks of age ▪ 2 weeks before lay	-----	**Top 5** *S.* Enteritidis *S.* Typhimurium *S.* Infantis *S.* Hadar *S.* Virchow
	Production	▪ Every 2 weeks	▪ Onset of lay ▪ Middle of lay ▪ End of lay	
Layers Any flock > 1000 birds	**Rearing**	▪ 1 day ▪ 2 weeks before moving to the laying unit"	-----	**Top 2** *S.* Enteritidis *S.* Typhimurium
	Production	▪ Every 15 weeks	▪ One flock per holding per year	

Source: The author.

Whole food chain approach

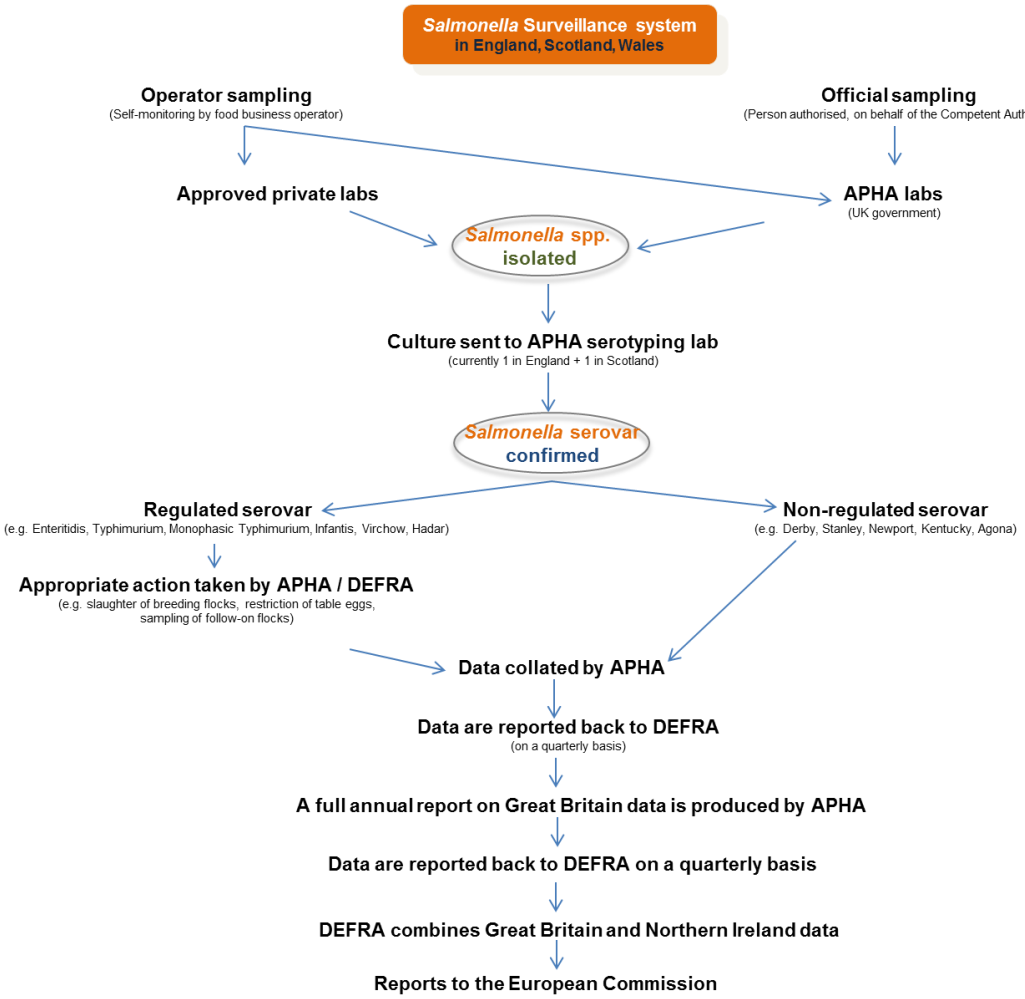
The National Control Programmes are to be seen in conjunction with other pieces of EU legislation, which ensure that food safety is being achieved not only through controls at the farm level, but also in the slaughterhouse. This "whole food chain approach" includes Regulation (EC) 852/2004[x] on the "Hygiene of foodstuffs" and Commission Regulation (EC) No. 2073/2005[xi] on "Microbiological criteria for foodstuffs", which lays down the microbiological criteria for certain micro-organisms and the implementing rules to be complied with by food business operators.

How to implement a *Salmonella* surveillance system

In order to implement a functioning *Salmonella* surveillance system, a network of expertise, comprising the Competent Authority, accredited laboratories and a State Veterinary Service to implement restrictions are necessary in each member state. According to Regulation 2160/2003, each member state must nominate a Competent Authority to oversee the programme. This authority is also responsible for transmitting data to the European Commission on an annual basis and for regular compliance and record checks on farms.

Member states need to establish a National Reference Laboratory; however, other laboratories can also participate in the control programmes, given they have been designated by the Competent Authority, apply quality assurance systems conform to the current ISO standard and regularly participate in collaborative tests. In the United Kingdom, the *Salmonella* surveillance system is the result of a collaborative approach between governmental entities of the four countries: England: Defra (Department for Environment, Food and Rural Affairs) Wales (Welsh Government), Scotland: (Scottish Government) and Northern Ireland (DARD - Department of Agriculture and Rural Development).

Figure 3: Common workflow for *Salmonella* Surveillance system implemented in England, Scotland and Wales.



Source: Animal & Plant Health Agency, United Kingdom

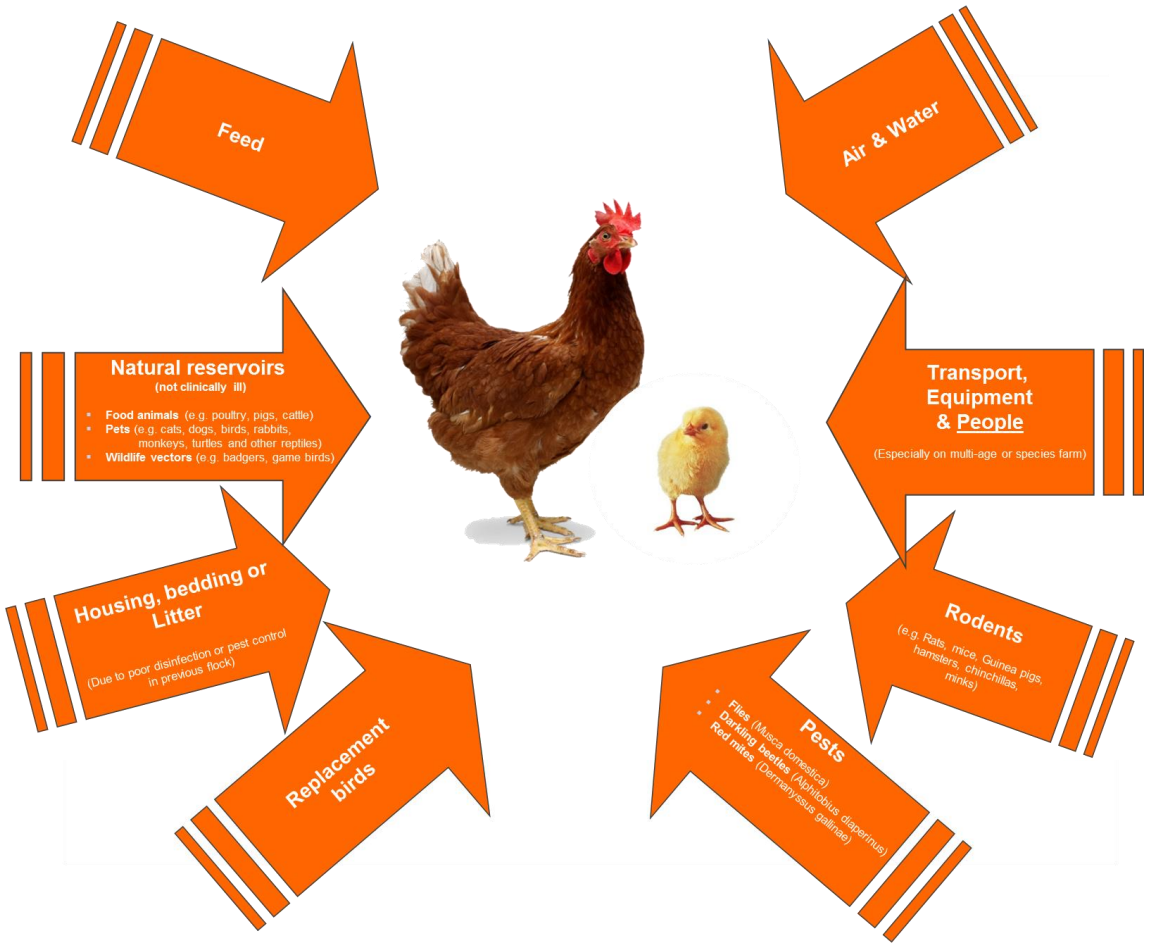
In member states, where the use of live vaccines is permitted, laboratories must be capable of reliably distinguishing *Salmonella* vaccine strains from field strains when testing samples taken under the National Control Programmes. Some of the vaccines currently on the market can be distinguished from field strains via their specific antimicrobial resistance pattern, whereas others contain mutations causing auxotrophism for essential components and can be identified by culturing them on special agar plates.

Food business operators and organisations representing such operators are an important part of the control programmes in many member states, and the Competent Authority needs to satisfy itself that the industry run control programmes comply with the relevant requirements set out in the EU legislation. In the United Kingdom, the British Egg Industry Council (BEIC) plays an instrumental role in delivering the National Control Programme, and members of their Quality Assurance Scheme (The Lion Code) need to adhere to strict rules set out in the Lion Code of Practice[xii]. These rules cover a wide range of activities on farm, including registration and traceability, breeding flock controls, pullet farms, vaccination programmes, laying birds, time and temperature controls on-farm, controls on egg packs, controls on feed, packing centre hygiene, animal welfare and more. Members of the scheme are audited regularly by the scheme to assure compliance.

Main sources of *Salmonella* on poultry farms

Infected replacement birds, contaminated equipment and contaminated feed are some of the most common sources of *Salmonella* infection on a poultry farm. Once the infection gets established inside a poultry house and/or the environment, it can be difficult to eliminate, and a great effort is often needed to get a house *Salmonella*-free after a positive flock. *Salmonella* can become established in rodent populations and other vectors, such as litter beetles or mites. *Salmonella* have been shown to survive for many months both under dry conditions, i.e. in dust in poultry houses, and under wet conditions in the environment. Insufficient cleansing and disinfection are often the reason for carry-over from one crop to the next, as is poor general hygiene and biosecurity[xiiixiv]. Some serovars are also known to be good biofilm-producers, which enables them to survive long term in feed mills, hatcheries or on farms. Elimination of infection from a poultry house or establishment requires a holistic approach involving several aspects of disease control. The main routes of *Salmonella* introduction on farms are shown in fig. 4.

Figure 4: Main sources of *Salmonella* contamination in the poultry farm.



Source: Animal & Plant Health Agency, United Kingdom

Summary and conclusion

Salmonellosis is (still) the second most common food borne illness in the EU, although significant progress has been made by the poultry industry over the past decade to reduce the *Salmonella* prevalence in chicken flocks. EU legislation has been instrumental in implementing control measures at farm level, and both official and operator sampling are necessary to implement a successful control programme.

Vaccination of breeding and laying flocks against *S*. Enteritidis and *S*. Typhimurium has also played an important role in eliminating the two most prevalent *Salmonella* serovars from chicken flocks in many countries and is now a requirement in quality assurance schemes in several member states. Overall, an integrated approach, including vaccination, diagnostic & monitoring, nutritional management, biosecurity, hygiene at all levels of production, pest control and feed control, is necessary to keep the prevalence of *Salmonella*-infection in poultry flocks at a low level.

References

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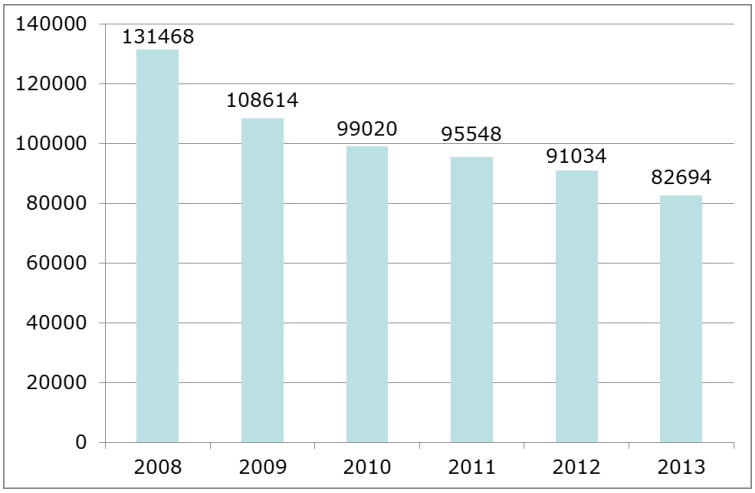

Fig. 1: European data, collated by the European Food Safety Authority (EFSA), show that the prevalence of human salmonellosis cases has fallen by around 37% between 2008 and 2013.