

## **Report of the**

## Task Force on Zoonoses Data Collection on the Analysis of the baseline survey on the prevalence of *Salmonella* in broiler flocks of *Gallus gallus*, in the EU, 2005-2006<sup>1</sup>

## Part B: factors related to *Salmonella* flock prevalence, distribution of *Salmonella* serovars, and antimicrobial resistance patterns

(Question N° EFSA-Q-2006-040)

Adopted by The Task Force on 26 October 2007

<sup>&</sup>lt;sup>1</sup> For citation purposes: Report of the Task Force on Zoonoses Data Collection on the Analysis of the baseline survey on the prevalence of *Salmonella* in holdings of broiler flocks of *Gallus gallus*, Part B, *The EFSA Journal* (2007) 101, 1-86.



## Summary

A European Union-wide baseline survey was carried out to determine the prevalence of *Salmonella* in commercial flocks of broilers with at least 5,000 birds in order to provide the scientific basis for setting a Community reduction target for *Salmonella* in broiler flocks of *Gallus gallus*. The sampling of the flocks took place between October 2005 and September 2006. Five faeces samples were taken from the flocks within 3 weeks before leaving for slaughter. A total of 6,325 holdings corresponding to 7,440 flocks with validated results were included in the survey analysis. The analysis of the *Salmonella* prevalence was carried out earlier and was published by the European Food Safety Authority on 30 March 2007 in part A report.

In a further analysis published in this part B report only few factors were found to be associated with *Salmonella* flock prevalence at the Community-level. The Community *Salmonella* flock prevalence varied significantly and importantly between months of sampling. The months found to be associated with higher flock prevalence were not consistently the same for different *Salmonella* serovars.

Flocks with younger broilers were associated with a higher risk of being *S*. Enteritidis positive, whereas broiler houses with a higher number of cycles of flocks per year were associated with higher flock prevalence for serovars other than *S*. Enteritidis and *S*. Infantis. The flock production type and the medication status were found to be associated with *S*. Infantis flock prevalence but since the Community *S*. Infantis prevalence was mostly driven by one Member State, these findings should be interpreted with caution.

*S.* Enteritidis was clearly the most frequently reported serovar in broiler flocks in the EU. *S.* Infantis, *S.* Mbandaka, *S.* Typhimurium, *S.* Hadar, *S.* Agona, *S.* Livingstone, *S.* Senftenberg, *S.* Montevideo, *S.* Tennessee and *S.* Virchow were also reported in between 8 to 12 MSs and should be regarded as important serovars of the broiler flock population.

The diversity of observed serovars differed greatly between MSs from a single serovar reported to more than 20 different serovars reported. Also the distribution of the serovars varied strongly amongst the MSs. Though 17 MSs reported *S*. Entertitidis, a formal spatial analysis identified two MSs as the most likely clusters for this serovar, whereas the most likely clusters for *S*. Typhimurium included three MSs.

The serovar distribution in broiler flocks and those reported in holdings with flocks of laying hen appeared to be similar in the EU. Similarities in *Salmonella* prevalence and serovar distributions were also found between broiler flocks and breeding flocks for broilers within the MSs, indicating that breeding flocks are likely to form an important source of *Salmonella* infections for the broiler flocks. Moreover, there was often a good agreement between the serovar and phage type distribution in human salmonellosis cases and in broiler flocks. These findings suggest that in the EU broiler meat is an important source of *Salmonella* infections in humans, although this importance is likely to differ between the MSs due to the varying *Salmonella* prevalence in broiler flocks.

The antimicrobial susceptibility testing information reported was not representative of the whole of the EU. The proportion of resistant isolates to third generation cephalosporins in the reporting MSs was very low, although three MSs reported the presence of *S*. Paratyphi B var. Java isolates resistant to ceftiofur and to cefotaxime. *S*. Enteritidis isolates were relatively susceptible to the tested antimicrobials, while resistance in *S*. Typhimurium was generally higher.

Since few risk factors were found to be associated with *Salmonella* flock prevalence at the Community-level, MSs are invited to carry out studies to identify the factors that put broiler flocks at risk of becoming infected with *Salmonella* at the national level. MSs are also encouraged to guarantee effective *Salmonella* control in breeding flocks for broilers in order to reduce and



prevent the subsequent contamination of the broiler flocks. It is also further recommended that MSs serotype all *Salmonella* isolates originating from broiler flocks to enable evaluation of the public health importance of the findings.

Making the reporting on antimicrobial resistance and phage typing obligatory in future baseline studies would provide for more representative information.



#### **Table of contents**

Summary	2
1. Introduction	6
2. Objectives	7
3. Materials and methods	8
3.1. Data description	8
3.2. Analysis of factors associated with the EU Salmonella broiler flock preva	alence8
3.2.1. Definition of the outcome variables	8
3.2.2. Choice of factors to be investigated	9
3.2.3. Correlation analysis	9
3.2.4. Identification of possible factors related to EU Salmonella flock prevalence	10
3.3. Analysis of the serovar and phage type distribution	10
4. Results	
4.1. Analysis of factors associated with EU Salmonella broiler flock prevalen	ce11
4.1.1. Univariate description of the flocks sampled	11
4.1.2. Analysis of the correlation amongst covariates	12
4.1.3. EU analyses of factors associated with <i>Salmonella</i> flock prevalence	
4.1.3.1 Salmonella Enteritidis	
4.1.3.3 Serovars other than Salmonella Enteritidis and Salmonella Infantis	
4.2. Analysis of the <i>Salmonella</i> serovar and phage type distribution	
4.2.1 Serovar frequency distribution in the EU	17
4.2.2. Differences in servar distribution between the countries	
4.2.3. Spatial distribution of the estimated number of Salmonella positive broiler flo	ck in the EU21
4.2.4. Comparison between the EU serovar distribution in broiler flocks and in holdi	ngs of laying hens
4.2.5. Comparison between the EU serovar distribution in broiler flocks and in breed broilers	ling flocks for
4.2.6. Comparison between the EU serovar distribution in broiler flocks and in salme humans	onellosis cases in25
4.2.7. Phage type distribution	
4.2.7.1 <i>S</i> . Enteritidis phage types	
4.2.7.2 S. Typhimurium phage types	
4.2.8. Comparison between phage type distribution in oroners, raying nens and num	alls20
4.3. Analysis of antimicrobial resistance testing information	
4.3.1.1 Antimicrobial resistance in S. Enteritidis	
4.3.1.2 Antimicrobial resistance in S. Entertudis phage types	
4.3.1.4 Antimicrobial resistance in <i>S</i> . Typhimurium definitive type isolates	
4.3.1.5 Antimicrobial resistance in selected other <i>Salmonella</i> serovar isolates	
5. Discussion	
5.1. Analysis of factors associated with Salmonella broiler flock prevalence a	t the EU-level37
5.1.1. The analysis results valid for all Salmonella serovars	
5.1.2. Specific results for Salmonella Enteritidis	



5.1.3. Specific results for <i>Salmonella</i> Infantis
5.1.4. Specific results for serovars other than S. Enteritidis and S. Infantis
5.2. Analysis of the serovar and phage type distribution
5.2.1. Differences in serovar distribution between MSs
5.2.2. Comparison of the serovar and phage type distribution in human cases, in broiler flocks and in laying hen holdings
5.3. Analysis of antimicrobial resistance testing information41
5.3.1. General considerations
5.3.2. Resistant to third generation cephalosporins
5.3.3. Resistance in S. Paratyphi B var. Java42
5.3.4. Resistance in S. Enteritidis and S. Typhimurium
6. Conclusions
7. Recommendations
Task Force on Zoonoses Data Collection members    46
Acknowledgements
Abbreviations
List of Tables
List of Figures
List of Annexes
Annexes



## **1. Introduction**

In order to provide the scientific basis for setting the Community target for reduction of the prevalence of *Salmonella* in broiler flocks of *Gallus gallus*, a European Union-wide *Salmonella* baseline survey was carried out between 1 October 2005 and 30 September 2006 in accordance with Regulation (EC) No 2160/2003 on the control of *Salmonella* and other specified zoonotic agents<sup>1</sup>. The survey provides comparable information on the prevalence of *Salmonella* in broiler flocks with at least 5,000 birds in the European Union (EU) Member States (MSs) and Norway.

A report from the Task Force on Zoonoses Data Collection on the Analysis of the baseline survey on the prevalence of *Salmonella* in broiler flocks of *Gallus gallus* in the EU, 2005-2006, part A *Salmonella* prevalence estimates<sup>2</sup> (Part A report) was issued on 30 March 2007. This report includes the analyses of the prevalence of *Salmonella* in broiler flocks, the most frequent *Salmonella* servers reported, and the sampling design.

The Part B report contains the analysis of potential risk factors and in-depth analyses of the serovar distribution, phage types and antimicrobial resistance of *Salmonella* isolates.

The objectives, the sampling frame and the diagnostic testing methods, as well as the collection of data, evaluation, reporting and timelines of the baseline survey are specified in Commission Decision 2005/636/EC concerning a baseline survey on the prevalence of *Salmonella* in broiler flocks of *Gallus gallus*<sup>1</sup>.

<sup>&</sup>lt;sup>1</sup> Regulation (EC) No 2160/2003 of the European Parliament and of the Council of 17 November 2003 on the control of *salmonella* and other specified food-borne zoonotic agents. *Official Journal of the European Union* 2003; L 325/1: 12.12.2003.

<sup>&</sup>lt;sup>2</sup> Report of the Task Force on Zoonoses Data Collection on the Analysis of the baseline survey on the prevalence of Salmonella in broiler flocks of Gallus gallus, Part A, The EFSA Journal (2007) 98, 1-85.



## 2. Objectives

The objectives of the baseline survey are described in detail in the Part A report.

The specific objectives related to the Part B report regarding the European Union-wide *Salmonella* in broiler flocks survey were:

- to investigate the effect of potential risk factors which may be associated with the *Salmonella* flock prevalence,
- to investigate in detail the Salmonella serovar distribution in broiler flocks across the EU,
- to analyse the information submitted by MSs regarding *S*. Enteritidis and *S*. Typhimurium phage types,
- to analyse the information submitted by MSs regarding antimicrobial resistance of *Salmonella* isolates.



## 3. Materials and methods

A description of the sample design and size of the baseline survey and of the bacteriological testing is found in the document of the European Commission DG SANCO entitled "Baseline survey on the prevalence of *Salmonella* in broiler flocks of *Gallus gallus* in the EU: Technical Specifications (SANCO/1688/2005 Rev1)"<sup>1</sup> and in the Part A report.

### 3.1. Data description

A detailed description of the validation and cleaning of the dataset carried out is provided in the Part A report. The final dataset contained data from 6,005 broiler holdings and 7,120 broiler flocks in 23 MSs and no data from Luxembourg and Malta. It also included data from 320 holdings (and flocks) in Norway.

# 3.2. Analysis of factors associated with the EU *Salmonella* broiler flock prevalence

The general assumptions and framework of the statistical analysis carried out are reported in detail in the Part A report. The flock observed prevalence was defined as the proportion of broiler flocks with at least 5,000 birds raised over the one year period of the baseline survey in the MSs which proved positive for *Salmonella*.

The effect of potential risk factors was analyzed on flock prevalence only, using the same modelbased approach as used and described in Part A report. A flock was considered positive if the presence of *Salmonella* or the specific serovar was detected in at least one of the five samples taken, otherwise it was considered negative.

Flocks with positive samples where the isolated *Salmonella* serovar was unspecified (e.g. 'nontypeable', 'other specify', or '*Salmonella* spp.') were excluded from the risk factor analyses.

#### 3.2.1. Definition of the outcome variables

Three outcome variables were defined for the investigation of factors associated with EU *Salmonella* broiler flock prevalence. The choice was based on the frequency of isolation of these serovars, the public health importance of the serovars, and their special epidemiology. This resulted in the independent analysis of the following *Salmonella* serovars or *Salmonella* serovars-groups at the EU level:

- S. Enteritidis,
- S. Infantis, and
- serovars other than *S*. Enteritidis and *S*. Infantis.

<sup>&</sup>lt;sup>1</sup> European Commission DG SANCO. Baseline survey on the prevalence of *Salmonella* in broiler flocks of *Gallus gallus* in the European Union: Technical specifications. SANCO/1688/2005 Rev1. Working document, 15 July 2005. Presented at the meeting of the Standing Committee on the Food Chain and Animal Health on 19 July 2005. (http://ec.europa.eu/food/biosafety/salmonella/tech\_spec\_sanco-1688-2005\_rev1\_en.pdf)



#### 3.2.2. Choice of factors to be investigated

A list of possible factors associated with *Salmonella* flock prevalence was established based on their plausible effect on the flock prevalence from an epidemiological point of view, on their relevance for control purposes, as well as on the availability of data.

The size variables for the holdings and flocks to be investigated in case of collinearity were flock size and number of flocks per holding.

The vaccination status variables were not investigated. This is because the descriptive analysis revealed that only 29 of the 7,108 flocks (0.40%) were reported vaccinated against *Salmonella*.

Moreover, it was decided to remove the variable "Type of sample taken" from the analysis because only Italy and Ireland had reported occasional use of hand swabs – instead of boot swabs. The investigated covariates are listed in Table 1.

## Table 1. Factors investigated for association with the Salmonella flock prevalence in broiler flocks in the EU, 2005-2006.

Explanatory variables investigated	Definition / Description and particularity		
Calendar month of sampling	Month of collection of the samples		
Delay (in days) of bacteriological analysis	-		
Number of flocks in the holding at any given time	Capacity of the holding		
Number of broilers in flock	Approximate number of broilers present at time of sampling		
Flock production type	conventional, free range standard, free range organic and unknown <sup>1</sup>		
Age of broilers at sampling (in days)	Age of the birds at sampling in the flock		
Number of cycles (crop) per year in this flock	Each new cycle (start of cycle within the survey period) is accounted for during a year period.		
Medication status (Yes vs. No)	Have antimicrobials been used during the last two weeks, prior to sampling.		

<sup>1</sup> In a conventional flock types the birds are kept inside the houses. A free-range flock system is a flock production type where the birds have access to outside. An organic flock system is a production type that is similar to the free-range system and that fulfils the requirements set for organic production; birds have access to outside and are registered with a recognised Organic Standard Regulatory Organisation.

#### 3.2.3. Correlation analysis

In order to assess potential additional collinearities between factors, two additional preliminary exploratory analyses were performed, at the EU level; Principal Component Analysis and evaluation of the correlation matrix. These analyses were done using the SAS software (version 9.1.3) using the PRINCOMP and CORR procedures, respectively. A detailed description of the correlation analysis is in Annex I.



3.2.4. Identification of possible factors related to EU Salmonella flock prevalence

The statistical methodology used for factor (covariates) screening was based on the two-step forward-backward approach and is described in Annex II.

A base model (with no covariate) was fit to the data similarly as in Part A Report. Then following the forward-backward model building process, covariates were added into or removed from the base model based on likelihood ratio tests with a significance level of 5%.

The forward (selection) step consisted in testing independently the effect of each single factor on the flock prevalence. At the end of this step, a final model was built, integrating all significant factors.

The backward (elimination) step consisted in testing whether each selected factor could be removed or not from the total model. At the end of this step, a final model was built, which integrates all significant factors.

From the final model fitting, the quantitative evaluation of the selected effects on flock prevalence base can be made at the EU level to investigate the size of effect. It is important to bear in mind that all effects shown are only statistical relationship between potential risk factors and flock prevalence, and it does not prove necessarily any link of causality.

All model fitting and comparisons were done using the NLMIXED procedure of the SAS software (version 9.1).

### 3.3. Analysis of the serovar and phage type distribution

The serotyping of *Salmonella* isolates was mandatory according to the technical specifications of the survey. At least one isolate from each positive sample was to be typed according to the Kaufmann-White Scheme. Results from any flock where the serovar information was not available for any isolate were excluded from the final dataset. The frequency distribution of the *Salmonella* serovars reported is presented in Part A report.

MSs and Norway could submit additional information on *S*. Enteritidis and *S*. Typhimurium phage types. The survey protocol recommended phage typing of at least one isolate of *S*. Enteritidis and *S*. Typhimurium from each positive sample, using the phage typing protocol defined by the Health Protection Agency Colindale, London.

The descriptive analysis of the *Salmonella* serovar and phage type data was performed in SAS Enterprise Guide v3.0 and Microsoft Excel. Maps displaying the estimated flock prevalence for *S*. Typhimurium, *S*. Enteritidis, *S*. Hadar, *S*. Mbandaka and *S*. Infantis were developed using Arc GIS 9. The prevalence of each serovar was divided into 5 categories, using Jenk's optimized natural breaks<sup>1</sup>.

The statistical methodology used for analysing the spatial distribution of reported *Salmonella* serovars is presented in Annex III.

<sup>&</sup>lt;sup>1</sup> Jenks GF, 1967. "The Data Model Concept in Statistical Mapping", International Yearbook of Cartography 7: 186-190.



## 4. Results

# 4.1. Analysis of factors associated with EU Salmonella broiler flock prevalence

The dataset used for the analysis of factors associated with the *Salmonella* flock prevalence (total 7,108 flocks) was derived from the global EU dataset used for the prevalence estimates, with the complementary exclusion of 12 flocks for which an unspecified *Salmonella* serovar was reported.

#### 4.1.1. Univariate description of the flocks sampled

A graphical display of the numbers of flocks sampled at the global EU-level in each month during the survey is presented in Figure 1, whereas analogous MS-specific figures are shown in Figure 8 of Annex IV. Relatively few flocks were sampled during the first month of the survey and the number of flocks sampled each month gradually augmented and was highest in the last month of the survey. Most MSs distributed the sampled flocks fairly equally over the one-year period but some MSs deviated from this survey requirement and were lacking samples for one (Austria, Belgium and Greece), two (Latvia, Lithuania and The Netherlands) or three (Estonia and Ireland) months. Portugal reported only samples for four months (June to September).

MS-specific descriptions of the numbers of flocks sampled in each month, of the numbers of flocks sampled for each age category, and of the numbers of flocks sampled for each category of numbers of cycles per year in the house are presented, respectively, in Figure 8, Figure 9 and Figure 10 of Annex IV. These figures show that there are differences between MSs concerning those investigated factors. Austria, Finland, Greece, Ireland and Slovakia sampled on average flocks with younger broilers, whereas France, Italy, Slovenia and Spain sampled more flocks with older broilers. Cyprus, France, Italy and Lithuania had more flocks sampled with a lower number of cycles, whereas Czech Republic, Denmark, Germany and Sweden sampled on average more flocks with a high number of cycles.

A detailed univariate description of the factors at EU-level and how they relate to the selected *Salmonella* serovars can be found in Annex V.





Figure 1. Number of broiler flocks sampled, per month, EU, Baseline survey in broiler flocks, 2005-2006.

4.1.2. Analysis of the correlation amongst covariates

The results of the analysis of the possible correlation amongst the factors associated with the *Salmonella* flock prevalence are presented in

Annex I. No collinearity was discerned between the factors (covariates). Consequently, grouping of factors (putative explanatory variables) was not relevant.

#### 4.1.3. EU analyses of factors associated with Salmonella flock prevalence

The detailed results regarding the forward-backward model selection are presented in Annex VI.

#### 4.1.3.1 Salmonella Enteritidis

The factors that were statistically significantly associated with the EU S. Enteritidis flock prevalence were:

- the month of sampling, and
- the age of broilers at sampling.

The results of the statistical analysis are detailed in Table 2 in a descending order of importance. If the P-value is smaller than 0.05, the difference in the flock prevalence to the compared basis is considered significant. In such cases the odds ratio (OR) differs significantly from one.



Factor	OR <sup>1</sup>	P-value
Month of the year of sampling (basis for comparison: January 2006)		< 0.001
October 2005	0.10	
November 2005	2.50	
December 2005	1.70	
February 2006	0.82	
March 2006	0.52	
April 2006	0.40	
May 2006	0.33	
June 2006	0.66	
July 2006	0.52	
August 2006	0.64	
September 2006	0.18	
Age of broilers at sampling (in days)	0.32	< 0.001

#### Table 2. Factors associated with the S. Enteritidis EU broiler flock prevalence, 2005-2006.

OR<sup>1</sup>: odds ratio

The variation of flock prevalence between the months was high. At the EU level up to a factor 10 difference could be found between the flock prevalence of *S*. Enteritidis between certain months. A higher *S*. Enteritidis flock prevalence was observed during the period November-February and to June-August, as displayed in Figure 2.

Flocks with younger birds were associated with a higher risk of being *S*. Enteritidis positive. The EU *S*. Enteritidis flock prevalence in flocks with broilers sampled at 20 days of age, and flocks sampled at 40 days of age, was 14% and 8%, respectively.

The factors that were not statistically significantly associated with the *S*. Enteritidis flock prevalence in the EU were:

- the time gap (in days) between the date of sampling and the date of start of bacteriological detection testing in the laboratory (which was limited in the protocol to 48 hours after receipt),
- the flock production type (conventional, free-range standard and free-range organic),
- the number of broilers in flock,
- the number of cycles (crops) per year in the house,
- the number of flocks in the holding at any given time of the year, and
- medication of the flock with antimicrobials within two weeks prior to sampling.



Figure 2. *Salmonella* Enteritidis flock prevalence<sup>1</sup> with CI's, per month, in the EU, Baseline survey in broiler flocks, 2005-2006.



<sup>1</sup>: for the median age of broilers at sampling of 28 days

#### 4.1.3.2 Salmonella Infantis

The factors that were statistically significantly associated with the EU S. Infantis flock prevalence were:

- the month of sampling,
- the medication of the flock with antimicrobials within two weeks prior to sampling, and
- the flock production type (conventional, free-range standard and free-range organic).

The results of the statistical analysis are detailed in Table 3 in a descending order of importance.

Higher S. Infantis flock prevalence was associated with the period November-January and April. Lower prevalence was associated with the months July to October. Flocks medicated with antimicrobials within two weeks prior to sampling were associated with two to three times lower S. Infantis flock prevalence compared to non-medicated flocks (respectively 1% and 2.5% flock prevalence estimates). Non-conventional production type (free range standard or free range organic) was associated with lower S. Infantis flock prevalence compared to conventional production. This difference reached up to a factor 10.



Factor	OR	<b>P-value</b>
Month of the year of sampling (basis for comparison: January 2006)		< 0.001
October 2005	0.29	
November 2005	0.66	
December 2005	1.88	
February 2006	0.45	
March 2006	0.26	
April 2006	1.96	
May 2006	0.39	
June 2006	0.31	
July 2006	0.24	
August 2006	0.16	
September 2006	0.25	
Medication status	0.34	< 0.001
(basis for comparison: non-medicated flocks)		
Flock production type	0.04	< 0.001
(basis for comparison: conventional production)		

 Table 3. Factors associated with the S. Infantis prevalence in the broiler flocks in the EU, 2005-2006.

The factors that were not statistically significantly associated with the EU S. Infantis flock prevalence in the EU were:

- the time gap (in days) between the date of sampling and the date of start of bacteriological detection testing in the laboratory (which was limited in the protocol to 48 hours after receipt),
- the number of broilers in flock,
- age of broilers at sampling,
- the number of cycles (crops) per year in the house, and
- the number of flocks in the holding at any given time of the year.

#### 4.1.3.3 Serovars other than Salmonella Enteritidis and Salmonella Infantis

The factors that were statistically significantly associated with the EU flock prevalence to serovars other than *S*. Enteritidis and *S*. Infantis were:

- the month of sampling, and
- the number of cycles (crops) per year in the house.

The results of the statistical analysis of factors associated with the prevalence of the serovars other than *S*. Enteritidis and *S*. Infantis at the EU level are detailed in Table 4 in a descending order of importance.



Factor	OR	P-value
Month of the year of sampling (basis for comparison: January 2006)		< 0.001
October 2005	1.71	
November 2005	0.86	
December 2005	0.80	
February 2006	0.62	
March 2006	0.55	
April 2006	0.43	
May 2006	0.43	
June 2006	0.34	
July 2006	0.33	
August 2006	0.40	
September 2006	0.37	
Number of cycles (crop) per year in the flock considered	4.2	< 0.001
1		

Table 4.	Factors associated with the flock prevalence of serovars other than S.	Enteritidis
and S. Inf	ıfantis, EU, 2005-2006.	

OR<sup>1</sup>: odds ratio

The variation of flock prevalence between months was high. At the EU level up to a factor 10 difference could be found between two months (in the same year). Higher flock prevalence of serovars other than *Salmonella* Enteritidis *Salmonella* Infantis was associated to the period October to January, as displayed in Figure 3.

Houses with a higher number of cycles per year were associated with higher flock prevalence for serovars other than *S*. Enteritidis and *S*. Infantis. For example, in houses with 4 cycles per year compared to houses with 7 cycles per year, the EU flock prevalence was 10% and 15%, respectively.

The factors that were not statistically significantly associated with the EU flock prevalence to serovars other than S. Enteritidis and Infantis were:

- the age of broilers at sampling,
- the time gap (in days) between the date of sampling and the date of start of bacteriological detection testing in the laboratory (which was limited in the protocol to 48 hours after receipt),
- the flock production type (conventional, free-range standard and free-range organic),
- the number of broilers in flock,
- the number of flocks in the holding at any given time of the year, and
- medication of the flock with antimicrobials within two weeks prior to sampling.



Figure 3. Prevalence<sup>1</sup> of flocks positive to serovars other than *Salmonella* Enteritidis and *Salmonella* Infantis, with CI's, per month, in the EU, Baseline survey in broiler flocks, 2005-2006.



<sup>1</sup>: for the median number of cycles (crops) of 5 per year in the house

### 4.2. Analysis of the Salmonella serovar and phage type distribution

#### 4.2.1. Serovar frequency distribution in the EU

Among the positive samples reported in the survey, 100 different serovars were recorded by 22 MS. The ten most frequently isolated *Salmonella* serovars in the EU, ranked by the percentages of specific *Salmonella* serovar-positive flocks, are listed in Table 5.

As already reported in the part A report, *S*. Enteritidis was clearly the most frequently reported serovar in broiler flocks in the EU, while *S*. Infantis was the second most isolated serovar. Together, these two serovars were isolated from more than half of all *Salmonella* positive flocks in the survey. The next three most frequent serovars were *S*. Mbandaka, *S*. Typhimurium and *S*. Hadar. Though *S*. Virchow was found in only 2.1% of all *Salmonella* positive broiler flocks, it was reported by 11 MSs indicating that it is among the more widely spread serovars throughout the EU.



Serovars (N tot= 4,962)		1	Flocks with se (N <sub>tot</sub> = 1,44	No. of MSs		
	Ν	%	Ν	%	reporting	
S. Enteritidis	1,677	33.8	538	37.1	17	
S. Infantis	1,090	22.0	295	20.4	14	
S. Mbandaka	400	8.1	114	7.9	12	
S. Typhimurium	150	3.0	65	4.6	15	
S. Hadar	186	3.7	59	4.1	8	
S. Kentucky	130	2.6	44	3.0	5	
S. Livingstone	105	2.1	39	2.7	8	
S. Anatum	90	1.8	32	2.2	8	
S. Montevideo	84	1.7	31	2.1	6	
S. Virchow	93	1.9	30	2.1	11	
Other serovars	969	19.5				
Salmonella spp.	26	0.5				

Table 5.	Frequency distribution of isolated Salmonella serovars in the Baseline survey in
broiler flo	ocks, 2005-2006.

Figure 4 shows the maps presenting the estimated broiler flock prevalence for *S*. Enteritidis and *S*. Typhimurium, *S*. Infantis, *S*. Mbandaka and *S*. Hadar, in the MSs and Norway. Strong differences were observed between the MSs.

Figure 4. The distribution on the estimated broiler flock prevalence of *S*. Enteritidis, *S*. Typhimurium, *S*. Enteritidis or *S*. Typhimurium, *S*. Infantis, *S*. Mbandaka and *S*. Hadar in countries participating in Baseline survey in broiler flocks, 2005-2006.







4.2.2. Differences in serovar distribution between the countries

The diversity of observed serovars varied greatly between the countries from a single serovar reported (e.g. Estonia, Lithuania, Finland and Norway) to more than 20 different serovars reported (e.g. Belgium, Estonia, France, Greece and Italy). This is illustrated in Table 6 that presents the number of flocks, the percentage positive flocks and the number of different serovars found in broiler flocks in the various MSs and Norway in this survey.



Table 6.Number of flocks included in the survey, percentage of positive flocks and numberof different serovars found in countries participating in Baseline survey in broiler flocks,2005-2006.

	Number of flocks		No. of different	
Country	in survey	% positive	serovars reported	
Italy	313	30.4	32	
Belgium	373	15.3	27	
France	381	8.9	25	
Spain	388	42.3	22	
Greece	245	27.3	21	
Germany	377	17.2	18	
Cyprus	248	10.9	17	
Hungary	359	65.7	17	
The United Kingdom	382	10.7	16	
The Netherlands	362	10.2	14	
Poland	357	57.7	13	
Portugal	367	42.8	12	
Czech Republic	334	22.5	10	
Ireland	351	27.9	8	
Austria	365	7.7	7	
Denmark	295	3.1	7	
Slovakia	230	8.3	6	
Latvia	121	9.1	2	
Slovenia	326	3.1	2	
Estonia	139	2.2	1	
Lithuania	156	5.1	1	
Finland	360	0.3	1	
Sweden	291	0.0	0	
Total	7,120	20.3	100	
Norway	320	0.3	1	

Not only the diversity, but also the actual distribution of the serovars varied greatly amongst the MSs. Figure 5 shows the frequency distribution of the five most frequently isolated serovars in broiler flocks in the MSs.

*S.* Enteritidis was the dominant serovar in 10 of the 23 MSs. In Estonia and Lithuania, *S.* Enteritidis was the only serovar isolated. Belgium, Denmark, Finland, Ireland, Norway, Sweden and the United Kingdom did not detect *S.* Enteritidis at all from broiler flocks. In Portugal, Poland and Spain, the observed *S.* Enteritidis-flock prevalence was high.

*S.* Infantis and *S.* Mbandaka were into the top 5 list of serovars in the EU mainly due to their dominance in two MSs (see Figure 5 and Annex VII): Hungary reported 71% of the *S.* Infantispositive flocks, whereas 48% of the *S.* Mbandaka-positive flocks were located in Ireland.

S. Typhimurium, S. Agona, S. Livingstone, S. Senftenberg, S. Montevideo, S. Tennessee and S. Virchow were also reported in between 8 to 15 MSs - S. Typhimurium also reported by Norway -



and these serovars should as such also be regarded as important serovars of the broiler flock population.



Figure 5. Frequency distribution of flocks positive to *S*. Enteritidis, *S*. Typhimurium, *S*. Infantis, *S*. Mbandaka and other *Salmonella* serovars in MSs, in the EU, 2005-2006.

4.2.3. Spatial distribution of the estimated number of *Salmonella* positive broiler flock in the EU

To investigate the spatial distribution of the most frequently reported serovars in the EU a formal spatial analysis was performed. Table 7 shows the most likely and secondary spatial clusters with their respective relative risk (RR) and level of significance (P-value).

Though 17 MSs reported *S*. Enteritidis, Portugal and Spain were identified as the most likely cluster for this serovar. A relative risk (RR) of 6.2 suggests that broiler flocks in this area are six times more likely to become infected with *S*. Enteritidis than broiler flocks outside this region. Poland was identified as a secondary cluster with an RR of almost 4 (P<0.001). The most likely cluster for *S*. Typhimurium that was identified included Hungary, Slovakia and Poland (RR=9.6, P<0.001). The cluster for *S*. Enteritidis or *S*. Typhimurium is dominated by the distribution of *S*. Enteritidis. Hungary was identified in the most likely cluster for *S*. Infantis (RR=20.5, P<0.001) and Ireland for *S*. Mbandaka (RR=48.3, P<0.001). The area including Poland was identified as the most likely cluster for *S*. Hadar and secondary cluster for *S*. Enteritidis and *S*. Mbandaka.



Serovar	Cluster Type	MSs included	<b>Relative risk</b>	P-value
S. Enteritidis	Most likely	Portugal and Spain	6.2	0.001
	Secondary	Poland	3.9	0.001
S. Typhimurium	Most likely	Slovakia, Hungary, Poland	9.6	0.001
	Secondary	Belgium	4.3	0.001
S. Enteritidis/Typhimurium	Most likely	Portugal and Spain	5.7	0.001
	Secondary	Poland	3.9	0.001
S. Hadar	Most likely	Poland	5.7	0.001
	Secondary	Spain	4.0	0.001
S. Infantis	Most likely	Slovakia, Hungary, Poland	20.5	0.001
	Secondary			
S. Mbandaka	Most likely	Ireland	48.3	0.001
	Secondary	Hungary, Poland, Greece, Italy	3.4	0.001
Other serovars	Most likely	Greece, Hungary, Slovakia,	3,4	0.001
		Austria, Italy		
	Secondary (1)	The Netherlands, Belgium,	1.4	0.001
		Germany		
	Secondary (2)	Ireland, United Kingdom	1.2	0.001

Table 7. Most frequently reported Salmonella serovars, the most likely and secondaryspatial clusters, and relative risks, in the EU, 2005-2006.

Maps of most likely and secondary clusters are presented in Figure 6.

Figure 6. Distribution of the most frequent reported *Salmonella* serovars, most likely and secondary spatial clusters, relative risks, in the EU, 2005-2006.



















4.2.4. Comparison between the EU serovar distribution in broiler flocks and in holdings of laying hens

A comparison of the serovar distribution in broiler flocks to that in laying hen holdings as reported in the Report of the Task Force on Zoonoses Data Collection on the Analysis of the baseline survey on the prevalence of *Salmonella* in holdings of laying hen flocks of *Gallus gallus*<sup>6</sup> is shown in Table 8.

	Broile	<b>Broiler flocks</b>		Laying hen flocks	
Salmonella serovar	No. Flocks	No. MSs reporting	No. Flocks	No. MSs reporting	
S. Enteritidis	538	17	899	18	
S. Typhimurium	65	15	123	15	
S. Infantis	295	14	171	13	
S. Mbandaka	114	12	101	12	
S. Senftenberg	28	12	30	9	
S. Virchow	30	11	41	8	
S. Agona	16	9	38	10	
S. Anatum	32	8	21	4	
S. Hadar	59	8	53	7	
S. Livingstone	39	8	50	10	
S. Indiana	19	7	11	4	
S. Derby	13	6	14	5	
S. Montevideo	31	6	27	9	
S. Blockley	29	5	4	2	
S. Kentucky	44	5	12	4	
S. Newport	8	5	11	7	
S. Ohio	19	5	35	2	
S. Bredeney	10	4	26	5	
S. Heidelberg	10	4	4	3	
S. Tennessee	5	4	28	9	
Other serovars	-	15	-		

Table 8.Number of flocks and the number of reporting MSs for the 20 most commonlyfound serovars in broiler production in the EU compared with the occurrence and number ofreporting countries for laying hen holdings.

The serovar distribution in broiler flocks and those reported in holdings with flocks of laying hen appears to be very similar in the EU, especially with regard to the most frequently isolated serovars. Broilers and laying hens have *S*. Entertitidis, *S*. Typhimurium, *S*. Infantis, *S*. Mbandaka, *S*. Virchow,

<sup>&</sup>lt;sup>6</sup> Report of the Task Force on Zoonoses Data Collection on the Analysis of the baseline survey on the prevalence of *Salmonella* in holdings of laying hen flocks of *Gallus gallus, The EFSA Journal* (2007) 97.



*S.* Hadar and *S.* Livingstone in common among their ten most frequently isolated serovars. *S.* Senftenberg, *S.* Agona, *S.* Montevideo and *S.* Newport also rank high in both the broiler and layer serovar distribution. Many of the remaining serovars are comparable with regard the number of reporting MSs. In 10 MSs, *S.* Enteritidis was the most prevalent serovar in broiler flocks, but only four MSs (Portugal, Poland, Spain and Czech Republic) had a *S.* Enteritidis prevalence above the EU average (Part A report). The same four MSs also had the highest prevalence of *S.* Enteritidis in the laying hen holdings. In many other MSs the prevalence of *S.* Enteritidis and *S.* Typhimurium was higher in laying hen holdings than in the broiler flocks.

4.2.5. Comparison between the EU serovar distribution in broiler flocks and in breeding flocks for broilers

Throughout the EU, when comparing to the Community Summary Report on Zoonoses 2005<sup>7</sup>, similarities in *Salmonella* prevalence and serovar distributions were found between broiler flocks and breeding flocks for broilers. In general, MSs with a high *Salmonella* prevalence in the broiler flock population in this survey (e.g. Poland, Portugal, Spain, Ireland) also reported a relatively high *Salmonella* prevalence in their breeding flocks for broilers in 2005 (Community Summary Report on Zoonoses 2005). In Ireland, both in the broiler flock population and in the breeding flocks for broilers relatively high *Salmonella* prevalence was reported but in neither population *S*. Enteritidis or *S*. Typhimurium was found. Similarly, in the United Kingdom most *Salmonella* positive breeding flocks for broilers were infected with serovars other than *S*. Enteritidis or *S*. Typhimurium, as found in this survey. In contrast, Italy found a relatively high proportion of broiler flocks *Salmonella* positive in this survey, but reported no positive breeding flocks for broilers in 2005. However, in 2004, Italy reported almost 14% of the breeding flocks for broilers to be *Salmonella* positive.

4.2.6. Comparison between the EU serovar distribution in broiler flocks and in salmonellosis cases in humans

According to the Community Summary Report in  $2005^7$ , of the more than 2,500 known serovars, *S*. Enteritidis caused more than 50% of the reported human *Salmonella* infections in the EU, followed by *S*. Typhimurium (~10%). Other important serovars for humans included *S*. Anatum, *S*. Bovismorbificans, *S*. Derby, *S*. Goldcoast, *S*. Hadar, *S*. Infantis, *S*. Newport and *S*. Virchow.

In Figure 7 a MS-specific comparison of the serovar distribution in salmonellosis cases in humans and broiler flocks is presented. The figure includes the MSs for which sufficient data of humans was available in either 2004 or 2005 (preferred) in the Community Summary Reports on Zoonoses. Only the distribution of the most commonly reported human serovars, besides *S*. Entertitidis and *S*. Typhimurium, is presented. The figure shows that there is a relatively good agreement between the serovar distribution in humans and broiler flocks, particularly for the poultry-associated serovars and for those MSs with high prevalences.

<sup>&</sup>lt;sup>7</sup> The Community Summary Report on Trends and Sources of Zoonoses, Zoonotic Agents, Antimicrobial Resistance and Foodborne Outbreaks in the European Union in 2005, *The EFSA Journal* (2006), 94.



Figure 7. Comparison of the serovar distribution in human cases and broilers in MSs for which sufficient human data was available in either 2004<sup>1</sup> or 2005 (preferred). Only the distribution of the most commonly reported human serovars, besides S. Enteritidis and S. Typhimurium, is presented.



<sup>1</sup> For Austria and the United Kingdom, human serovar data from 2004 was used.

4.2.7. Phage type distribution

#### 4.2.7.1 S. Enteritidis phage types

Data on *S*. Enteritidis phage types was provided by seven MSs (Austria, the Czech Republic, Germany, Italy, Latvia, the Netherlands and Slovakia). Ten MSs with *S*. Enteritidis isolates did not report phage typing information. The other MSs (Belgium, Denmark, Finland, Ireland, Sweden and the United Kingdom) did not detect *S*. Enteritidis at all from broiler flocks.

The MSs that gave information on S. Enteritidis phage types reported a total of 287 S. Enteritidis isolates, out of which 161 isolates (56%) were phage typed. This represented only 9.6% of the total 1,677 S. Enteritidis isolates in the EU. Most frequently reported phage types are presented in Table 9, which also displays the number of MSs and holdings where S. Enteritidis phage types were detected. In this table the ranking is based on the percentages of specific S. Enteritidis phage type-positive holdings in the EU. MS-specific overviews of S. Enteritidis phage types are shown in Annex VIII.



S. Enteritidis (N=	S . Enteritidis (N=161) Phage Type N			Holding phage ( (N=8	s with types (5)	Flocks with phage types (N=93)		
Phage Type	Ν	%		Ν	%	Ν	%	
PT8	41	25.5	4	40	47.1	40	43	
PT4	30	18.6	4	15	17.6	15	16.1	
PT21	30	18.6	4	7	8.2	10	10.8	
PT2	22	13.7	2	6	7.1	6	6.5	
PT1	12	7.5	2	3	3.5	7	7.5	
PT13	7	4.3	1	2	2.4	2	2.2	
PT7	5	3.1	3	4	4.7	4	4.3	
PT23	5	3.1	2	5	5.9	5	5.4	
PT6	2	1.2	2	2	2.4	2	2.2	
PT7a	1	0.6	1	1	1.2	1	1.1	
PT37	1	0.6	1	1	1.2	1	1.1	
PT14B	1	0.6	1	1	1.2	1	1.1	
Non typeable	3	1.9	2	3	3.5	3	3.2	
Non - tested	1	0.6	1	1	1.2	1	1.1	

#### Table 9. Distribution of S. Enteritidis phage types in broiler flocks in the EU, 2005-2006.

*S.* Enteritidis phage type eight (PT8) was the most common reported phage type in the EU followed by PT4 and PT21. Figure 11 in Annex VIII displays for every MS that provided the information the most frequently identified *S*. Enteritidis phage types.

#### 4.2.7.2 S. Typhimurium phage types

Data on *S.* Typhimurium phage types were provided by 7 MSs (Austria, the Czech Republic, Denmark, Germany, Slovakia, The Netherlands and the United Kingdom), whereas 8 MSs with *S.* Typhimurium isolates did not report phage typing information. Norway reported one *S.* Typhimurium isolate that was non typeable.

The MSs that reported information regarding *S*. Typhimurium phage types had 41 *S*. Typhimurium isolates altogether, out of which 41 (100%) were phage typed. This represented 27.3% of the total 150 *S*. Typhimurium isolates in the EU. In Table 10, that presents the most frequently reported phage types, the ranking is based on the percentages of specific *S*. Typhimurium phage type-positive flocks in the EU. MS-specific overviews *S*. Typhimurium phage types are in Annex IX.

The S. Typhimurium phage types seemed to be evenly distributed, although the data were very sparse.



S. Typhimurium	S. Typhimurium phage types (N=41)			Hol	dings	Flocks			
phage type			reporting the phage type	with phage	types (N=14)	with phage	types (N=15)		
	Ν	%		Ν	%	Ν	%		
DT104B low	4	9.8	1	2	14.3	2	13.3		
DT104L	7	17.1	2	2	14.3	2	13.3		
U302	2	4.9	2	2	14.3	2	13.3		
DT012	2	4.9	1	1	7.1	1	6.7		
DT104	3	7.3	1	1	7.1	1	6.7		
DT114	1	2.4	1	1	7.1	1	6.7		
DT15a	3	7.3	1	1	7.1	1	6.7		
DT208	5	12.2	1	1	7.1	1	6.7		
DT85	5	12.2	1	1	7.1	1	6.7		
FT 506 <sup>a</sup>	5	12.2	1	1	7.1	1	6.7		
RDNC <sup>b</sup>	4	9.8	1	2	14.3	2	13.3		
Total	41	100.0		14		15			
Not phage typed	117								

#### Table 10. Distribution of *S*. Typhimurium Phage types in broiler flocks in the EU, 2005-2006.

<sup>a</sup>: FT506 is analogous to DT104

<sup>b</sup>: RDNC = 'Reacts but Does Not Conform' (to a recognised phage lysis pattern)

4.2.8. Comparison between phage type distribution in broilers, laying hens and humans

In order to elucidate the role of *S*. Enteritidis contaminated broiler meat as a source of human salmonellosis, *S*. Enteritidis phage typing results from broilers, laying hens and humans were compared (Table 11). Phage typing distribution in humans is only available from a fraction of the MSs and also only a minor proportion of the MSs applied phage typing on the isolates found in the baseline surveys. Interpretation should consequently be done very cautiously.



Tabla 11	Composition of C	Entopitidic phone	a tumoa igolatad fuar	n humana huailana	and lawing hang
I able 11.	Comparison of 5	. Enternuois dhag	e lydes isolated froi	n numans, proners	s and laving nens.
					· ·····

	Human S. Enteritidis phage types reported in 2004								in	No. of broiler flocks as reported in the EU baseline survey, 2005-2006							No. of laying-hen holdings as reported in the EU baseline survey, 2004-2005								
Phage type	AT	BE	DK	FI	HU	IE	NL	UK	Total	AT	CZ	DE	IT	LV	SK	NL	No. of MSs	AT	CZ	DE	DK	IT	NL	UK	No. of MSs
РТ 4	2,472	210	128	200	-	43	224	2,373	5,650	4	8	-	1	-	2	-	4	16	-	96	-	2	11	15	5
PT 1	250	11	75	173	139	48	85	1,997	2,778	-	1	-	-	1	-	1	3	2	1	4	-	-	2	1	5
PT 6	410	5	34	33	1724	10	63	483	2,762	-	1	-	-	-	1	-	2	1	1	2	-	-	4	7	5
PT 8	1,856	36	114	69	-	10	101	373	2,559	1	33	1	-	-	5	-	4	10	17	16	1	-	3	-	5
PT 21	710	118	62	92	-	18	142	555	1,697	-	-	1	1	1		4	4	5	-	2	-	-	2	-	3
PT 14b	136	45	-	40	-	11	15	1,362	1,609	-	1	-	-	-	-	-	1	-	-	1	-	4	-	1	3
PT 7	18	3	1	-	1125	-	-	10	1,157	-	2	-	-	1	-	1	3	8	2	2	-	-	4	5	5
РТ ба	25	9	-	24	-	11	29	335	433	-	-	-	-	-	-	-	0	-	3	2	-	-	-	1	3
PT 12	15	2	-	-	-	-	-	161	178	-	-	-	-	-	-	-	0	-	1	-	-	-	-	1	2
PT 5c	8	-	-	-	-	-	-	119	127	-	-	-	-	-	-	-	0	-	-	-	-	-	-	1	2
PT 24	-	-	-	-	-	-	-	96	96	-	-	-	-	-	-	-	0	-	-	-	-	-	-	1	1
PT 13a	21	-	-	-	-	1	-	28	50	-	-	-	-	-	-	-	0	-	2	-	-	-	-	-	1
PT 5a	18	-	-	-	-	-	-	31	49	-	-	-	-	-	-	-	0	-	-	1	-	-	-	2	1
PT 2	4	-	-	-	-	1	-	22	27	1	-	-	5	-	-	-	2	-	-	1	-	-	-	-	1
PT 29	16	1	-	-	-	-	-	10	27	-	-	-	-	-	-	-	0	-	-	-	1	-	1	-	2
PT 4b	13	-	-	-	-	3	2	8	26	-	-	-	-	-	-	-	0	-	-	1	-	-	-	-	1
PT 35	-	2	-	-	-	1	-	22	25	-	-	-	-	-	-	-	0	-	-	2	-	-	-	8	2
PT 25	1	-	-	-	-	-	14	1	16	-	-	-	-	-	-	-	0	-	-	1	-	-	1	-	2
PT 13	1	-	-	-	-	-	-	9	10	-	-	2	-	-	-	-	1	-	1	-	-	-	-	-	1
PT 23	7	1	-	-	-	-	-	2	10	-	3	-	-	-	2	-	2	3	8	1	-	-	-	-	3
PT 28	-	5	-	-	-	-	-	2	7	-	-	-	-	-	-	-	0	-	-	-	1	-	-	-	1
PT 21c	5	-	-	-	-	1	-	-	6	-	-	-	-	-	-	-	0	-	-	1	-	-	-	-	1
PT 37	3	-	-	-	-	-	-	1	4	-	-		1				1	-	-	-	-	-	-	-	0
PT 7a	1	-	-	-	-	-	-	2	3	-	1	-	-	-	-	-	1	-	-	1	-	1	-	-	2
PT 4a	1	-	-	-	-	-	-	-	1	-	-	-	-	-	-	-	0	-	-	1	-	-	-	-	1
PT 19	1	-	-	-	-	-	-	-	1	-	-	-	-	-	-	-	0	1	-	1	-	-	-	-	2
PT RDNC	123	-	-	26	-	3	-	44	196	-	-	-	-	-	-	-	0	-	-	6	-	-	-	-	2
Not typable	-	4	-	13	-	-	-	21	38	-	-	-	-	-	2	-	2	2	1	2	1	-	19	7	6
Other	166	27	132	61	2653	10	91	943	4,083	0	0	0	0	0	0	0	0	1	1	1	0	-	0	0	3



Overall, it can be observed for MSs which reported phage type information that all phage types occurring in broilers also occur in laying hens, whereas several phage types occur only in laying hens. The phage types occurring in both sources seem to be the most prevalent ones, not only in broilers and laying hens, but also in humans. In general and at the MS-level (for those few MSs with available data), there is a good agreement between the phage type distribution observed in humans and those reported in broilers and, particularly, in laying hens.

Available data regarding S. Typhimurium phage types were too sparse to justify any further analysis.

### 4.3. Analysis of antimicrobial resistance testing information

MSs and Norway could submit additional information on the antimicrobial resistance of *Salmonella* isolates. The survey protocol recommended using one isolate per serovar per flock for antimicrobial resistance testing. Quantitative methods and the standards for antimicrobial resistance testing given by the Clinical and Laboratory Standards Institute (CLSI, formerly known as the National Committee for Clinical Laboratory Standards NCCLS) for specific antimicrobials were recommended. Six MSs used methods to determine the Minimum Inhibitory Concentration (MIC) whereas seven used the Disc Diffusion Method (Annex X). The antimicrobials tested by the reporting MSs and Norway are presented in Annex XI.

For the purposes of this report isolates reported as intermediately resistant have been included in the resistant category. This was done for three reasons:

- The CLSI breakpoints, which were recommended to be used in the survey, are rather higher than those adopted by many European institutions which set national breakpoints and are also often higher than those set by EUCAST, particularly for compounds such as the third generation cephalosporins<sup>1</sup>. The inclusion of the intermediate with the resistant category tends to offset this trend and therefore provides better comparability with many other European studies.
- 2) Important resistances such as resistance to fluoroquinolones or possession of ESBLs are often best detected by inclusion of the intermediate category with the resistant category.
- 3) In future, epidemiological cut-off values, as defined by the European Committee on Antimicrobial Susceptibility Testing (EUCAST), that is standardising methods in the medical field within Europe, will probably be used in surveys of this type. Inclusion of the intermediate together with the resistant category will provide slightly better comparability with these future studies.

The data on antimicrobial resistance were analysed at the flock level in order to detect differences in resistance occurring in different flocks. The percentage of flocks with fully-susceptible isolates represents the proportion of positive flocks with isolates tested for antimicrobial resistance and in which no resistance was detected. Because different MSs tested different panels of antimicrobials, these figures may not be directly comparable between MSs.

<sup>&</sup>lt;sup>1</sup> Kahlmeter, G., Brown D.F.K., Goldstein F.W., MacGowan A.P., Mouton J.W., Österlund, A., Rodloff, A., Steinbakk, M., Urbaskova, P. and Vatopoulos, A. (2003). European harmonization of MIC breakpoints for antimicrobial susceptibility testing of bacteria. Journal of Antimicrobial Chemotherapy, **52**, 145-148.



Data on the occurrence of antimicrobial resistance in *S*. Enteritidis, *S*. Typhimurium, and/or in serovars other than *S*. Enteritidis or *S*. Typhimurium were provided by 14 MSs and Norway, whilst eight MSs with *Salmonella* isolates did not report the results of antimicrobial resistance testing. The 14 reporting MSs provided susceptibility data for a total of 30 antimicrobials, though the range of antimicrobials tested differed in different MSs.

The following descriptive qualitative analysis focuses on antimicrobial resistance of *S*. Enteritidis, *S*. Typhimurium isolates and other ubiquitous serovars selected for specific interest. Resistance trends to 3<sup>rd</sup> and 4<sup>th</sup> generation cephalosporins, fluoroquinolones and quinolones are highlighted because of their particular importance for public health. Antimicrobial resistance in *Salmonella* spp. and in other *Salmonella* serovars isolated in the survey are presented in detail in Annex XIV and in Annex XV. The findings should be interpreted with caution as the reported antimicrobial resistance data is very sparse for some serotypes, relating to only low numbers of isolates.

Countries may not be mentioned in the analysis if the serovar being analysed was not detected, which is for example the case for *S*. Enteritidis that was not detected in Belgium, Denmark, Finland, Ireland, Norway, Sweden and The United Kingdom.

#### 4.3.1.1 Antimicrobial resistance in S. Enteritidis

The total number of broiler flocks from which *S*. Enteritidis isolates was recovered in the MSs reporting antimicrobial susceptibility testing data was 237. The number of isolates examined for their antimicrobial resistance was 234. The susceptibility data for the total of 30 antimicrobials tested by the MSs is shown in Annex XII, whereas a subset of the results regarding the antimicrobials of most interest is presented in Table 12.

No resistance was detected to the third generation cephalosporins; cefpodoxime, cefotaxime and ceftazidime. Three MSs (Austria, Germany and Poland) tested the antimicrobial resistance of *S*. Entertitidis isolates to the veterinary cephalosporin ceftiofur and no resistance was reported.

Concerning the quinolones and fluoroquinolones, Slovakia was the only reporting MS to test oxolinic acid and no resistance was reported. All reporting MSs tested susceptibility to nalidixic acid. The proportion of *Salmonella* Enteritidis-positive broiler flocks with isolates resistant to nalidixic acid was 21% for all MSs together. The highest resistance to nalidixic acid was reported by Latvia and Poland, respectively 100% and 28%. All reporting MSs except Poland examined isolates for their susceptibility to ciprofloxacin and no resistance was detected. Some MSs examined susceptibility to enrofloxacin and Poland reported a resistance proportion of 10%, but the proportion of full resistance - not intermediately susceptible - to enrofloxacin in *S*. Enteritidis isolates from Poland was only 0.8%.



Country	No. Positive flocks	No. flocks with tested isolates	Percentage Fully Sensitive	Ampicillin	Cefotaxime	Ceftazidime	Chloramphenicol	Ciprofloxacin	Enrofloxacin	Gentamicin	Nalidixic acid	Streptomycin	Sulphonamide	Tetracycline	Trimethoprim
All reporting MSs	237	**	64.3	7	0	0	0.9	0	6	0.4	21	4	7	4	0
Austria	6	6	83.3	17			0	0		0	0	0	0	0	0
<b>Czech Republic</b>	48	$48^{1}$	81.3	2	0		0	0	0	0	13	0	0	0	0
Germany	4	4	100.0	0			0	0		0	0	0	0	0	0
Italy	12	$10^{2}$	80.0	11	0	0	0	0	0	0	11	0		0	
Latvia	9	8 <sup>3</sup>	25.0	50			40	0		0	100	38		60	
Lithuania	8	8 4	75.0	0			0	0	0	0	0	0	25	0	0
Poland	123	123	50.4	9			0		10	0.8	28	5	11	3	0
Slovakia	13	$12^{5}$	91.7	0	0	0	0	0		0	0	0	0	0	0
Slovenia	9	9 <sup>6</sup>	77.8	0	0		0	0	0	0	22	0	0	11	0
The Netherlands	5	5	80.0	0	0	0	0	0		0	20		0	0	0

# Table 12. The proportion (%) of *Salmonella* Enteritidis-positive broiler flocks with resistant (\*) isolates in reporting MSs of the EU, including the percentage of fully-susceptible isolates, in the EU, 2005-2006.

\*The figures include resistant isolates and isolates of intermediate susceptibility.

\*\*Varies for each antimicrobial; can be calculated by referring to figures for individual MSs.

<sup>1</sup>: Czech Republic n=47 for trimethoprim.

<sup>2</sup>: Italy n=8 for ceftazidime and tetracyclines, n=9 for ampicillin, cefotaxime, chloramphenicol,

ciprofloxacin, enrofloxacin, gentamicin, nalidixic acid and streptomycin.

<sup>3</sup>: Latvia n=4 for ciprofloxacin, n=5 for chloramphenicol, gentamicin, nalidixic acid, tetracyclines, n=6 for ampicillin.

<sup>4</sup>: Slovakia n=11 for trimethoprim.

<sup>5</sup>: Slovenia n=8 for trimethoprim.

#### 4.3.1.2 Antimicrobial resistance in S. Enteritidis phage types

Phage types PT 6, PT 13, PT 23 and PT 37 of *S*. Enteritidis were fully-susceptible to the antimicrobials tested by MSs. Detailed results concerning the antimicrobial resistance pattern of phage types of *S*. Enteritidis are presented in Table 13.



Table 13.	The proportion (%) of <i>Salmonella</i> Enteritidis phage type-positive broiler
flocks with	resistant isolates (*) in reporting MSs of the EU, including the percentage of
fully-suscep	tible isolates, in the EU, 2005-2006.

Phage Type	Member States reporting this phage type	No. flocks with isolates tested	Percentage fully sensitive	Ampicillin	Chloramphenicol	Streptomycin	Sulphonamide	Tetracyclines	Nalidixic acid	Trimethoprim + Sulphonamide
PT 1	LV, NL	7 <sup>1</sup>	29	40	33	33	0	33	100	0
PT 2	AT, IT	$6^{2}$	83	0	0	0	0	0	0	20
PT 4	AT, CZ, IT,	$15^{3}$	33	13	0	0	0	0	33	9
PT 7	CZ, LV, NL	$4^{4}$	75	0	0	0	0	0	33	0
PT 8	AT, CZ, DE,	39 <sup>5</sup>	97	0	0	0	0	0	0	0
PT 21	DE, IT, LV,	10 <sup>6</sup>	60	13	10	20	0	20	40	0

The proportion resistant isolates has been rounded to the nearest whole percentage, except where less than 1.

\* The figures include resistant isolates and isolates of intermediate susceptibility.

 $^{1}$  n=1 for sulphonamides, n=2 for trimethoprim+sulphonamide, n= 3 for chloramphenicol, nalidixic acid and tetracyclines, n= 5 for ampicillin, n=6 for streptomycin.

 $^{2}$  n=1 for sulphonamides, n=5 for trimethoprim+sulphonamide.

 $^{3}$  n=14 for sulphonamides n=11 for trimethoprim+sulphonamide.

<sup>4</sup> n=2 for trimethoprim+sulphonamide, n=3 for all other antimicrobials except ampicillin where n=4.

<sup>5</sup> n=36for trimethoprim +sulphonamide.

 $^{6}$  n=5 for streptomycin and sulphonamides n=6 for trimethoprim+sulphonamide, n= 8 for ampicillin.

In general, the highest resistance was observed for *S*. Enteritidis phage type 1.

#### 4.3.1.3 Antimicrobial resistance in S. Typhimurium

The total number of broiler flocks from which *S*. Typhimurium isolates was recovered in the MSs reporting antimicrobial susceptibility testing data was 41. The number of isolates examined for their antimicrobial resistance was 40. The susceptibility data for the total of 30 antimicrobials is shown in Annex XIII, whereas a subset of the results regarding the antimicrobials of most interest is presented in Table 14.

There were no MSs in which all S. Typhimurium isolates tested were fully-susceptible.

No resistance was detected towards the third and fourth generation of cephalosporins. Five MSs (Austria, Belgium, Denmark, Germany and Poland) tested the antimicrobial resistance of S. Typhimurium to the veterinary cephalosporin ceftiofur and resistance was reported in a single isolate from Belgium.

Concerning the quinolones and fluoroquinolones, Slovakia was the only reporting MS to test oxolinic acid and no resistance was reported; all reporting MSs tested susceptibility to nalidixic acid and the proportion of resistance to nalidixic acid for all reporting MSs was 40%. Resistance to nalidixic acid was highest for Poland (87%) and for the United Kingdom (100%, one flock



tested). All MSs except Belgium and Poland examined isolates for their susceptibility to ciprofloxacin and no resistance was detected. Some MSs (Belgium, Czech Republic and Poland) examined susceptibility to enrofloxacin and 11% - overall - of isolates were reported to be resistant or intermediate; these isolates were detected in flocks in Poland where 20% of the tested *S*. Typhimurium isolates were intermediately susceptible to enrofloxacin.

# Table 14. The proportion (%) of *Salmonella* Typhimurium-positive broiler flocks with resistant (\*) isolates in reporting MSs of the EU, including the percentage of fully-susceptible isolates, in the EU, 2005-2006.

Country	No. positive flocks	No. flocks with isolates tested	Percentage Fully Sensitive	Ampicillin	Cefotaxime	Ceftazidime	Chloramphenicol	Ciprofloxacin	Enrofloxacin	Gentamicin	Nalidixic acid	Streptomycin	Sulphonamide	Tetracycline	Trimethoprim
All reporting MSs	41	**	15.0	73	0	0	53	0	11	3	40	69	75	65	14
Austria	2	2	50.0	0			0	0		0	0	50	50	0	0
Belgium	12	$11^{1}$	27.3	55			27		0	0	0	36	55	36	
Czech Republic	2	2	0.0	100	0		0	0	0	50	50	50	50	100	0
Denmark	1	1	0.0	100			0	0		0	0	100	100	0	100
Germany	6	6	33.3	67			50	0		0	17	67	67	67	50
Poland	15	15	0.0	87			87		20	0	87	100	100	93	0
Slovakia	1	1	0.0	100	0	0	0	0		0	0	0	0	0	0
The Netherlands	1	1	0.0	100	0	0	100	0		0	0		100	100	0
The United Kingdom	1	1	0.0	100		0	100	0		0	100	100	100	100	
Norway	1	1	0.0	100	0		100	0		0	0	100	100	0	100

\*The figures include resistant isolates and isolates of intermediate susceptibility.

\*\*Varies for each antimicrobial; can be calculated by referring to figures for individual MSs.

#### 4.3.1.4 Antimicrobial resistance in S. Typhimurium definitive type isolates

Resistance by phage or definitive type in S. Typhimurium was reported by seven MSs. Austria reported a DT 85 isolate which was susceptible to the antimicrobials tested, whilst Germany reported that a DT 104 B low isolate was also fully susceptible. Results are summarised in Table 15. The typical pentavalent resistance pattern of resistance to ampicillin, chloramphenicol, streptomycin, sulphonamides and tetracyclines was seen in some DT 104 and related isolates; florfenicol resistance was also present in some isolates.



Table 15.	The proportion (%) of definitive type S. Typhimurium-positive broiler flocks
with resistant	isolates (*) in reporting MSs of the EU, including the percentage of fully-
susceptible iso	olates, in the EU, 2005-2006

Reporting MS	Phage / Definitive Type	No. flocks with isolates tested	Percentage fully sensitive	Ampicillin	Chloramphenicol	Streptomycin	Sulphonamide	Tetracyclines	Nalidixic acid	Florfenicol	Trimethoprim + Sulphonamide	Trimethoprim
Austria	DT104L	1	0	0	0	0	100	100	0	0		0
	DT 85	1	100	0	0	0	0	0	0	0		0
Creek Denublie	DT 114	1	0	100	0	100	100	100	100		0	0
Czech Republic	U 302	1	0	100	0	0	0	100	0		0	0
Denmark	DT15a	1	0	100	0	100	100	0	0	0		100
	DT 12	1	0	100	100	100	100	100	0	100	0	0
<b>C</b>	DT 104B low	2	1	50	50	50	50	50	0	0	50	50
Germany	DT104L	1	0	100	100	100	100	100	100	100	100	100
	DT 208	1	0	100	0	100	100	100	0	0	100	100
Slovakia	U 302	1	0	100	0	0	0	0	0	0	0	0
The Netherlands	FT 506	1	0	100	100		100	100	0	100		0
The United Kingdom	DT 104	1	0	100	100	100	100	100	100		0	

\*The figures include resistant isolates and isolates of intermediate susceptibility.

#### 4.3.1.5 Antimicrobial resistance in selected other Salmonella serovars

The full susceptibility data for these selected serovars are in Annex XV.

Concerning S. Infantis isolates, an interesting observation is the intermediate resistance / resistance to enrofloxacin (13%) and nalidixic acid (8%) observed in Poland and the fact that the nalidixic acid figure is slightly lower than the enrofloxacin figure.

In *S*. Virchow isolates, the most noteworthy finding in this survey is the high proportion of resistance to nalidixic acid (overall; 85%). Similarly, a high proportion of nalidixic acid resistance in *S*. Hadar isolates (overall; 97%) was observed in this survey.

Three MSs reported susceptibility results for *S*. Paratyphi B var. Java and the overall proportion of nalidixic acid resistance was 63%. Approximately 33% of strains were resistant to third generation cephalosporins. The proportion of resistance to a range of other antimicrobials also tended to be relatively high in *S*. Paratyphi B var. Java in comparison with many other serovars.

Information on resistance in serovars not previously mentioned is also given in Annex XV for serovars to which more than ten isolates were subjected to susceptibility testing. Here the number of isolates tested is an important consideration in interpreting the significance of results for



individual antimicrobials. Resistance or intermediate susceptibility to enrofloxacin in *S*. Thompson from Italy, ciprofloxacin in *S*. Senftenberg from the United Kingdom and to cefotaxime and ceftazidime in *S*. Livingstone from Italy are all of note.

*Salmonella* serovars which were susceptible to all of the antimicrobials tested are listed Annex XVI for all reporting MSs as well as for each reporting MS separately.


## 5. Discussion

The survey in broiler flocks was the second of several baseline surveys to be conducted in the Community. The first part of the analyses of the broiler flock surveys was previously issued on 30 March 2007 in the report on the prevalence of *Salmonella* in broiler flocks of *Gallus gallus* in the EU, 2005-2006, part A *Salmonella* prevalence estimates<sup>1</sup> (Part A report).

The present part B of the report completes the technical analyses, the results of which may provide useful information for MSs, while they are planning their control programmes in broiler flocks. Also the more detailed analysis of the *Salmonella* serovar and phage type distribution and of the antimicrobial resistance in *Salmonella* isolates, clarifies the picture of the epidemiological situation and the relevance of the findings to human health.

# 5.1. Analysis of factors associated with *Salmonella* broiler flock prevalence at the EU-level

To ensure proper understanding of the statistical analysis of the factors associated with *Salmonella* positivity at the Community-level, it should be noted that:

- the factors evaluated are those potentially associated with the *Salmonella* flock prevalence. However, a statistical association between the factor and the *Salmonella* flock prevalence does not necessarily indicate a causal relationship. As a consequence, the analysis can only generate hypotheses for potential risk factors associated with the *Salmonella* flock prevalence,
- the potential risk factors that were evaluated were not comprehensive, and no interaction effects were investigated,
- geographical structure within the MSs and the impact of time on the investigated factors were not accounted for in the analysis.

These limitations are due to the survey design and the nature of the data received. For example, information on biosecurity measures in the broiler holding were not collected even though they are known to be important risk factors. Statistical issues left aside, also the possible biases have to be carefully considered. The MS-specific descriptions of the investigated factors revealed that there were sometimes systematical differences between the MSs.

## 5.1.1. The analysis results valid for all *Salmonella* serovars

The analysis of the broiler flock prevalence suggested an effect of the month of sampling on the prevalence of infection by all the three studied *Salmonella* serovars or serovar-groups. Based on the results, broiler flocks sampled during late autumn and winter of 2005-2006 are generally characterised by a higher risk of *Salmonella* infection at the Community level.

The numbers of sampled flocks were quite balanced and evenly distributed among MSs over the year, supporting the validity of the observed effect of the month. However, as these results derive

<sup>&</sup>lt;sup>1</sup> Report of the Task Force on Zoonoses Data Collection on the Analysis of the baseline survey on the prevalence of Salmonella in broiler flocks of *Gallus gallus*, Part A, The EFSA Journal (2007) 98, 1-85.



only from one sampling year, before making conclusions on possible seasonality, these results should be confirmed by multi-annual observational studies. This is especially the case, as the year 2006 was characterised with an unusual high temperature recorded in most MSs.

Rearing a flock in autumn has been previously reported as associated with an increased risk of *Salmonella* infection in broiler flocks<sup>1</sup>. Interestingly, the months that put flocks at risk of being positive in this survey varied between the servors groups analysed, most importantly *S*. Entertitidis showing prevalence peak also in summer.

Compared to the baseline survey on laying hens, fewer investigated factors were associated with the *Salmonella* prevalence. For example, contrary to the laying hen survey, no significant association was found between *Salmonella* and flock size or the number of flocks in a holding in the broiler survey. The fewer associated factors may be due to the fact that, in this survey, only holding- or flock-level characteristics were considered, while information on biosecurity measures, and *Salmonella* status of feed and day-old chicks were not covered, although their importance for *Salmonella* infection in broilers has been previously recognised in many studies<sup>2,3,4,5,6,7</sup> and also underlined by opinion of the EFSA Scientific Panel on Biological Hazards<sup>8</sup>.

## 5.1.2. Specific results for Salmonella Enteritidis

Only two factors were associated with the *S*. Enteritidis flock prevalence in broiler flocks; the month of sampling and the age of the broilers sampled. In the case of *S*. Enteritidis prevalence, it was the winter and summer months that were characterized by the highest Community-level prevalence. The flocks with younger broilers appeared to be at highest risk of *S*. Enteritidis infection in this survey. The prevalence of intestinal contamination by *Salmonella* in broiler flocks has been reported to decrease with the birds' age in other studies<sup>9</sup>; and resistance of older birds to *Salmonella* spp. infection might be explained by a natural antagonist digestive flora in caecum and colon<sup>10</sup>. However, since there were systematic differences between-MSs in the age of the sampled broilers, this finding should be interpreted with caution at the Community-level.

<sup>&</sup>lt;sup>1</sup> Angen O., Skov M.N., Chriél M., Agger J.F. and Bisgaard M. (1996) A retrospective study on *Salmonella* infection in Danish broiler flocks *Preventive Veterinary Medicine* 26, 223-237.

<sup>&</sup>lt;sup>2</sup> Henken A.M., Frankena K., Goelema J.O., Graat E.A. and Noordhuizen J.P. (1992) Multivariate epidemiological approach to salmonellosis in broiler breeder flocks *Poultry Science* 71, 838-843.

<sup>&</sup>lt;sup>3</sup> Davies R.H. and Wray C. (1996) Persistence of Salmonella Enteritidis in poultry units and poultry feed *British Poultry Science* 37, 589-596.

<sup>&</sup>lt;sup>4</sup> Davies R.H. and Wray C. (1995) Mice as carriers of Salmonella Enteritidis on persistently infected poultry units *Veterinary Record* 137, 337-341;

<sup>&</sup>lt;sup>5</sup> Rose N., Beaudeau F., Drouin P., Toux J.Y., Rose V. and Colin P. (1999) Risk factors for Salmonella enterica subsp. enterica contamination in French broiler-chicken flocks at the end of the rearing period *Preventive Veterinary Medicine* 39, 265-277.

<sup>&</sup>lt;sup>6</sup> Chriel M., Stryhn H. and Dauphin G. (1999) Generalised linear mixed models analysis of risk factors for contamination of Danish broiler flocks with Salmonella Typhimurium *Preventive Veterinary Medicine* 40, 1-17.

<sup>&</sup>lt;sup>7</sup> Gradel K.O. and Rattenborg E. (2003) A questionnaire-based, retrospective field study of persistence of Salmonella Enteritidis and Salmonella Typhimurium in Danish broiler houses *Preventive Veterinary Medicine* 56, 267-284.

<sup>&</sup>lt;sup>8</sup> Opinion of the Scientific Panel on Biological Hazards on a request from the Commission related to the use of antimicrobials for the control of *Salmonella* in poultry. *The EFSA Journal* (2004) 115, 1-76.

<sup>&</sup>lt;sup>9</sup> Bailey J.S. and Cox N.A. (1991) Internal colonisation and external carriage of artificially inoculated Salmonella Typhimurium from floor pen and cage reared chickens *Poultry Science* 70(S1), 142.

<sup>&</sup>lt;sup>10</sup> Nurmi E. and Rantala M. (1973) New aspects of Salmonella infection in broiler production *Nature* 241, 210-211.



It is noteworthy that the *S*. Enteritidis flock prevalence in this survey did not differ between different flock production types (conventional as compared to non-conventional ones).

## 5.1.3. Specific results for Salmonella Infantis

The findings related to factors associated with *S*. Infantis flock prevalence in this survey must be interpreted very cautiously because Hungary accounted for 71% of the *S*. Infantis positive flocks and skewed the overall Community *S*. Infantis prevalence. However, the finding that the *S*. Infantis flock prevalence was highest in conventional flock production types is interesting because the flock production type was not found to have a significant effect on the prevalence of other *Salmonella* serovar groups analysed in this survey.

## 5.1.4. Specific results for serovars other than S. Enteritidis and S. Infantis

For serovars other than *S*. Enteritidis and *S*. Infantis, only two factors were associated with the broiler flock prevalence. Apart from the month of sampling, the number of flocks reared per year in the broiler houses had an effect. The prevalence of the serovars increased with the number of flocks reared. This may be explained by shorter time periods between successive flocks that may limit cleaning and disinfection procedures of the house, resulting in residual contamination. Moreover, the number of thinning and of visits by feed trucks, which are considered as potential contamination routes; typically increase with the number of reared flocks per year.

The role of housing conditions in broiler infections was previously demonstrated by Rose *et al.*<sup>1</sup> and recognised as important by opinion of the EFSA Scientific Panel on Biological Hazards<sup>2</sup>. However, since there were systematic differences between MSs regarding the number of cycles of flocks per year, these findings should also be interpreted with caution at the Community-level. It is interesting to notice that the number of cycles of flocks per year was not associated with prevalence of *S*. Entertitidis in the broiler flocks, probably because vertical transmission plays an important role for this serovar.

As for the *S*. Enteritidis flock prevalence, the flock prevalence for serovars other than *S*. Enteritidis and *S*. Infantis was not different between flock production types (conventional and non-conventional ones).

## 5.2. Analysis of the serovar and phage type distribution

## 5.2.1. Differences in serovar distribution between MSs

The diversity and distribution of the observed serovars in the broiler flocks varied greatly between MSs and there were clusters of MSs with flocks being infected with certain *Salmonella* serovars. For the *S*. Enteritidis and *S*. Typhimurium combination, the Iberian Peninsula was the most important cluster.

<sup>&</sup>lt;sup>1</sup> Rose N., Beaudeau F., Drouin P., Toux J.Y., Rose V., Colin P. (2000) Risk factors for Salmonella persistence after cleansing and disinfection in French broiler-chicken houses. *Preventive Veterinary Medicine* 29; 44 9-20.

<sup>&</sup>lt;sup>2</sup> Opinion of the Scientific Panel on Biological Hazards on a request from the Commission related to the use of antimicrobials for the control of *Salmonella* in poultry. *The EFSA Journal* (2004) 115, 1-76.



The *Salmonella* prevalence and the serovar distributions in broiler flocks seemed to closely mirror that of breeding flocks for broilers within the MSs. This indicates the importance of the breeding flocks as a source of infection. In some MSs the low observed prevelance of *S*. Entertitidis and S. Typhimurium in both the breeding flocks and the broiler flock is likely to be a sign of the positive impact of the successful *Salmonella* control programmes in the breeding flocks.

A large variety of serovars in the broiler flocks observed in some MSs is indicative of a substantial number of *Salmonella* infection sources in these countries, such as contacts with other farmed animal species and wild-life, persistence of infection in broiler houses between successive flocks or *Salmonella* contaminated feed.

5.2.2. Comparison of the serovar and phage type distribution in human cases, in broiler flocks and in laying hen holdings

Eggs are commonly considered the predominant source of human salmonellosis in Europe<sup>3</sup>. This was supported by the results of the *Salmonella* baseline survey in laying hen holdings<sup>1</sup>, where *S*. Enteritidis was demonstrated to occur widely in the laying hen production in many MSs. However, *S*. Enteritis was also the most frequently isolated serovar in the *Salmonella* baseline survey in broilers and *S*. Enteritidis phage types commonly found in human cases were reported from the broiler flocks.

The further comparison of the *Salmonella* serovar and phage typing results from broiler baseline survey, the laying hen baseline survey and that of the reported human cases suggest that although the majority of human *S*. Enteritidis infections is supposed to be caused by contaminated eggs, a certain proportion of human infections occurring from broilers cannot be excluded. This role of broilers and broiler meat as a source of human infections is likely to be more important in countries with a high *S*. Enteritidis prevalence in broiler flocks.

*S.* Typhimurium was the second ranking serovar as a cause of human infections in the EU and responsible for approximately 10% of the reported cases. In the two baseline surveys, *S.* Typhimurium was found in broilers and in laying hens in majority of MSs and mostly the same MSs reported *S.* Typhimurium positive flocks in both the surveys. However, the estimated EU *S.* Typhimurium prevalence were much lower than that of *S.* Enteritidis. *S.* Typhimurium is a serovar, which is often found also in other animal species commonly assumed to form a source of human *Salmonella* infections, such as pigs and cattle. In fact, *S.* Typhimurium is reported to the dominant *Salmonella* serovar in pigs and cattle in the EU<sup>3</sup>. Therefore, it is likely that only a minor part of the human *S.* Typhimurium infections may be attributed to broilers and broiler meat as well as eggs.

For serovars other than *S*. Enteritidis and *S*. Typhimurium, the most commonly reported serovars found in human infections at the EU-level include *S*. Infantis, *S*. Bovismorbificans, *S*. Hadar, *S*. Virchow, *S*. Derby, *S*. Newport, *S*. Anatum and *S*. Goldcoast. While *S*. Bovismorbificans and *S*. Goldcoast were only isolated from a single flock in this survey, *S*. Infantis, *S*. Hadar, *S*. Virchow, *S*. Anatum, and to some extent *S*. Newport were relatively often detected from the broiler flocks. Accordingly, results of this survey suggest that broilers and broiler meat may be a relevant source

<sup>&</sup>lt;sup>1</sup> Report of the Task Force on Zoonoses Data Collection on the Analysis of the baseline study on the prevalence of Salmonella in holdings of laying hen flocks of *Gallus gallus*, The EFSA Journal (2007) 97



of these serovars for humans. However, more detailed knowledge of the serovar distribution in other food animals species are needed to quantify the relative role of broiler meat.

In conclusion, this survey supports the role of broiler meat as an important source of human *Salmonella* infection in the EU, particularly in MSs with a high *Salmonella* prevalence in broilers. Salmonellosis owing to broiler meat is not related to a specific serovar in contrast to those due to eggs or egg products.

## 5.3. Analysis of antimicrobial resistance testing information

## 5.3.1. General considerations

Determining the antimicrobial resistance of *Salmonella* isolates can have a number of useful functions, including:

- describing the epidemiology and tracing the spread of certain resistant serovars, phage types or clones,

- highlighting the emergence of new resistant strains,

- providing an alert as to the existence of strains with particular resistance patterns which have been previously detected and described in other parts of the world, and

- highlighting the development or acquisition of resistance to important therapeutic antimicrobials.

The prevalence of antimicrobial resistance in *Salmonella* can be influenced by a number of factors, including the selective pressure exerted by antimicrobials. However, certain *Salmonella* serovars, phage or definitive types, or clones, are commonly associated with particular patterns of antimicrobial resistance. Furthermore, some serovars of *Salmonella* are commonly fully-susceptible to relevant antimicrobials. The prevalence of these serovars, phage types and clones within the broiler population can, therefore, influence the proportion of resistance that is detected in isolates in a MS. The clonal spread of particular strains can be the result of factors related to husbandry and animal movements. The infection of parent flocks or the infection at the hatchery can strongly influence the proportion of resistant isolates.

This survey aimed to provide baseline data on the antimicrobial resistance of the *Salmonella* isolates from broiler flocks in the EU. It allowed some comparison to be made between resistance patterns in broilers and humans (based on previous studies). However, only 14 MSs out of the 22 MSs that detected *Salmonella* submitted data on the antimicrobial resistance of *Salmonella* isolates. Europewide coverage is important because certain serovars, phage types, or clones might have a limited geographical distribution within Europe, or tend to be associated with certain European regions.

Due to the current lack of complete harmonization of methods and breakpoints and the difference in the range of antimicrobials tested in different MSs, interpretation of the data at the EU level is difficult. Moreover, the sample size of the survey was calculated to estimate the prevalence of *Salmonella* in broiler flocks - not to estimate the prevalence of resistance in *Salmonella* serovars recovered during the survey. For a number of *Salmonella* serovars, either very few isolates were recovered during the survey, or relatively few isolates were subjected to antimicrobial resistance testing. Therefore, the reported antimicrobial resistance figures should be interpreted with caution, and the descriptive analysis of the reported antimicrobial resistance data is only qualitative.



## 5.3.2. Resistant to third generation cephalosporins

The third generation cephalosporins; cefpodoxime, cefotaxime and ceftazidime are commonly used to detect resistance mediated via either extended-spectrum beta-lactamases (ESBLs) or the enzymes AmpC beta-lactamases, both of which are important acquired resistance mechanisms in *Salmonella*, conferring resistance to a wide range of beta-lactam compounds. These types of resistance are currently very rare in farm animals in Europe and monitoring their spread and prevalence is consequently of both public and animal health importance. This is in contrast to some areas outside Europe where the prevalence is higher.

In this survey Italy reported resistance to cefotaxime and ceftazidime in single isolates of *S*. Braenderup and *S*. Livingstone, whilst The Netherlands reported resistance in a single isolate of *S*. Infantis and in two isolates of *S*. Paratyphi B var. Java.

Five MSs tested antimicrobial resistance to the veterinary cephalosporin ceftiofur and resistance was detected by Belgium in *S*. Typhimurium and *S*. Paratyphi B var. Java and by Germany in *S*. Paratyphi B var. Java. The suitability of ceftiofur as an indicator cephalosporin for the detection of ESBLs and other important types of beta-lactamase resistance is unknown. It can therefore be recommended that cefpodoxime or ceftazidime and cefotaxime are included in future surveillance to optimally detect resistance to third generation cephalosporins.

## 5.3.3. Resistance in *S*. Paratyphi B var. Java

The Netherlands, Belgium and Germany all reported the presence of *S*. Paratyphi B var. Java and the strains from Belgium and Germany were resistant to ceftiofur. Nalidixic acid resistance was also a feature of some of these strains and this is significant as fluoroquinolones and third generation cephalosporins are important antimicrobials in the treatment of *Salmonella* infections in humans in many European countries. *S*. Paratyphi B var. Java has been one of the predominant serovars in broilers in The Netherlands since 1998<sup>1</sup>, though the prevalence in the current survey was not high. *S*. Paratyphi B var. Java strains have often tended to show resistance to ampicillin, trimethoprim-sulphonamides, furazolidone and quinolones <sup>1</sup> and this was a feature of the isolates recovered also in this survey. Poultry meat has been strongly implicated as a source of *S*. Paratyphi B var. Java infections for humans in some countries.

## 5.3.4. Resistance in *S*. Enteritidis and *S*. Typhimurium

*S.* Enteritidis isolates were relatively susceptible amongst other *Salmonella* isolates in the reporting MSs. Nalidixic acid resistance in *S.* Enteritidis tends to be associated particularly with certain phage types, particularly phage type 1. There were only 7 *S.* Enteritidis phage type 1 isolates reported in this survey by Latvia and The Netherlands, and all of them were nalidixic acid resistant. Nalidixic acid resistance was also observed in other phage types. Human *S.* Enteritidis phage type 1 isolates which are nalidixic acid resistant have been earlier associated with travel to

<sup>&</sup>lt;sup>1</sup> Van Duijkeren, E., Wannet, W. J.B., Houwers, D.J. and Van Pelt, W. (2003). Antimicrobial susceptibilities of *Salmonella* strains isolated from humans, cattle, pigs and chickens in The Netherlands from 1984 to 2001. Journal of Clinical Microbiology **41**, 3574-3578.



Southern Europe or Asia in previous studies, or with the consumption of poultry products imported from this area<sup>1</sup>.

A high proportion of resistant isolates was detected to nalidixic acid in *S*. Virchow and *S*. Hadar in the current survey. Nalidixic acid resistance in *S*. Hadar has been frequently associated with resistance to amoxicillin, streptomycin and tetracyclines<sup>2</sup>. This phenotype of resistance was also detected by some reporting MSs in the current survey.

Resistance by phage or definitive type in *S*. Typhimurium was reported by seven MSs. Resistance in *S*. Typhimurium was generally higher than that observed in *S*. Enteritidis and that was an expected finding, particularly since some prevalent clones (e.g. DT 104) are associated with particular patterns of multiple antimicrobial resistance. The typical pentavalent resistance pattern of resistance to ampicillin, chloramphenicol, streptomycin, sulphonamides and tetracyclines was indeed seen in some DT 104 and related isolates. Poland and several other MSs reported the typical resistance pattern shown by DT104 and related types, such as U302. Previous studies have reported that *S*. Typhimurium DT 104 is one of the prevalent phage types amongst isolates of *S*. Typhimurium from poultry in Poland<sup>3</sup> and it therefore seems likely that DT 104 was also represented amongst the Polish isolates in this survey.

<sup>&</sup>lt;sup>1</sup> Threlfall, E.J., Fisher I.S.T., Berghold, C., Gerner-Smidt, P., Tschape, H, Cormican, M., Luzzi, I., Schnieder, F., Wannet, W., Machado J. and Edwards, G. (2003) Antimicrobial drug resistance in isolates of *Salmonella enterica* from cases of salmonellosis in Europe in 2003; results of international multi-centre surveillance. Eurosurveillance. **8**, 41-44.

<sup>&</sup>lt;sup>2</sup> Cailhol, J., Lailler, R., Bouvet, P., La Vieille, S., Gauchard, F., Sanders, P., and Brisabois A. (2006) Trends in antimicrobial resistance phenotypes in non-typhoid *Salmonellae* from human and poultry origins in France. Epidemiology and Infection **134**, 171-178.

<sup>&</sup>lt;sup>3</sup> Wasyl, D., Sandvang, D., Skov, M.N. and Baggesen, D. (2006) Epidemiological characteristics of *Salmonella* Typhimurium isolated from animals and feed in Poland. Epidemiology and Infection 134, 179-185.



## 6. Conclusions

- Only few factors were found in this survey to be associated with *Salmonella* prevalence in broiler flock at the EU-level, which is likely to be because some important risk factors for *Salmonella* infections in broiler flocks were not investigated in the survey (such as feed and level of biosecurity). The factors associated with the *Salmonella* flock prevalence may vary between the MSs.
- The observed EU-level *Salmonella* flock prevalence, whether for *S*. Enteritidis, *S*. Infantis or serovars other than *S*. Enteritidis and *S*. Infantis, varied significantly between the months of sampling.
- At the EU-level, flocks with younger broilers had a relatively higher risk of being *S*. Enteritidis positive and the number of cycles of flocks in broiler houses was positively associated with the prevalence of serovars other than *S*. Enteritidis and *S*. Infantis.
- The diversity of observed *Salmonella* serovars in broiler flocks between MSs was high. Also the actual distribution of the serovars varied strongly amongst the MSs.
- S. Infantis, S. Mbandaka, S. Typhimurium, S. Hadar. S. Agona, S. Livingstone, S. Senftenberg, S. Montevideo, S. Tennessee and S. Virchow were frequently isolated serovars from broiler flocks and should be regarded as important in the broiler flock population.
- Clusters of specific serovars were identified in certain MSs. Flocks in Portugal and Spain were the most likely to be positive for *S*. Enteritidis; flocks in Hungary, Slovakia and Poland for *S*. Typhimurium; whereas flocks in Hungary were the most likely to be positive for *S*. Infantis; flocks in Ireland for *S*. Mbandaka and flocks in Poland for *S*. Hadar.
- The serovar distributions in broiler flocks and laying hens appeared to be very similar in the EU, especially with regard to the most frequently isolated serovars.
- Within MSs there are similarities between *Salmonella* prevalence and serovar distribution in broiler flocks and the breeding flocks for broilers, indicating that breeding flocks are an important source of *Salmonella* infections for broiler flocks.
- The results of the serovar and phage types analyses in this survey further suggest that in the EU broiler meat is an important source of *Salmonella* infections in humans. However, the relative importance of broiler meat as a source of infection is likely to differ amongst the MSs due to the varying *Salmonella* prevalence in broiler flocks.
- S. Enteritidis isolates were relatively susceptible to antimicrobials in this survey. Antimicrobial resistance in S. Typhimurium was generally higher than that observed in S. Enteritidis.
- The proportion of resistance of *Salmonella* isolates to third generation cephalosporins in Europe in reporting MSs is very low.
- Three MSs reported *S*. Paratyphi B var. Java isolates resistant to ceftiofur or to cefotaxime, which is of special interest due to the potential public health importance and because 63% of isolates belonging to this serovar were resistant to nalidixic acid.



## 7. Recommendations

- Since only few risk factors were found to be associated with *Salmonella* flock prevalence at the EU-level in this survey, MSs are invited to carry out further studies at the national level to identify the factors that put broiler flocks at risk of becoming infected with *Salmonella*, taking into account their national *Salmonella* prevalence and serovar distribution.
- As the *Salmonella* infection in breeding and in broiler flocks seems to be associated, MSs are encouraged to guarantee effective *Salmonella* control in breeding flocks, especially for the invasive serovars such as *S*. Enteritidis, in order to reduce and prevent the subsequent contamination of the broiler flocks.
- It is recommended that MSs serotype all *Salmonella* isolates originating from broiler flocks to enable the evaluation of the public health importance of the *Salmonella* findings.
- Phage typing and antimicrobial resistance testing should be mandatory in the future baseline surveys to provide a comprehensive picture of the situation in the EU.
- The introduction of common test panels of antimicrobials and use of EUCAST cut-off values would greatly facilitate the analyses of antimicrobial resistance in the future surveys.
- It is recommended that the *Salmonella* isolates showing resistance to the important indicator third generation cephalosporins should be further characterised to identify the responsible resistant mechanism at the genetic level. Identification of the resistance mechanisms involved would allow early identification of the emergence of new resistant clones in Europe.



## **Task Force on Zoonoses Data Collection members**

José Ignacio Arraz Recio, Andrea Ammon, Harry Bailie, Marta Bedriova, Melanie Picherot, Birgitte Borck, Karen Camilleri, Georgi Chobanov, Adriana Costache, Kris De Smet, Matthias Hartung, Birgitte Helwigh, Merete Hofshagen, Vaidotas Kiudulas, Elina Lahti, Peter Much, Lisa O'Connor, Rob A.A. Van Oosterom, Jacek Osek, José Luis Paramio Lucas, Manca Pavšič, Christodoulos Pipis, Saara Raulo, Antonia Ricci, Tatjana Ribakova, Valentina Rizzi, Petr Šatrán, Joseph Schon, Jelena Sõgel, Petra Szabados, Patricia Tavares Santos, Kilian Unger, Luc Vanholme and Dimitris Vourvidis.

## Acknowledgements

The Task Force on Zoonoses Data Collection wishes to acknowledge the contribution of the Working Group that prepared this report: Dirk Berkvens, Vojka Bole-Hribovsek, Rob Davies, Cristina de Frutos-Escobar, Kris De Smet, Tine Hald, Andrzej Hoszowski, Sarolta Idei, Annemarie Käsbohrer, Peter Much, Nicolas Rose, Arjen van de Giessen, Antonia Ricci, Francesca Riolo, Kenneth Mulligan, Billy Amzal, Pierre-Alexandre Beloeil, Pia Mäkelä and Frank Boelaert.

The contributions of Danilo Lo Fo Wong and Chris Teale are also gratefully acknowledged, as well as the implementation of the baseline survey by the Competent Authorities of the Member States and Norway.





## Abbreviations

CI	Confidence Interval
CLSI	Clinical and Laboratory Standards Institute formerly known as
	NCCLS
EEA	European Economic Area
EFSA	European Food Safety Authority
ESBL	Extended Spectrum Beta-Lactamases
EU	European Union
EUCAST	European Committee on Antimicrobial Susceptibility Testing
MIC	Minimum Inhibitory Concentration
MS(s)	Member State(s)
NCCLS	The National Committee for Clinical Laboratory Standards, now CLSI



## List of Tables

Table 1.	Factors investigated for association with the Salmonella flock prevalence in broiler
	flocks in the EU, 2005-2006
Table 2.	Factors associated with the S. Enteritidis EU broiler flock prevalence, 2005-200613
Table 3.	Factors associated with the S. Infantis prevalence in the broiler flocks in the EU,
	2005-2006
Table 4.	Factors associated with the flock prevalence of serovars other than S. Enteritidis and
	S. Infantis, EU, 2005-2006
Table 5.	Frequency distribution of isolated <i>Salmonella</i> serovars in the Baseline survey in broiler flocks 2005-2006
Table 6	Number of flocks included in the survey percentage of positive flocks and number
Table 0.	of different serovars found in countries participating in Baseline survey in broiler
	flocks. 2005-2006
Table 7.	Most frequently reported Salmonella serovars, the most likely and secondary spatial
	clusters, and relative risks, in the EU, 2005-2006.
Table 8.	Number of flocks and the number of reporting MSs for the 20 most commonly found
	serovars in broiler production in the EU compared with the occurrence and number
	of reporting countries for laying hen holdings
Table 9.	Distribution of S. Enteritidis phage types in broiler flocks in the EU, 2005-200627
Table 10.	Distribution of S. Typhimurium Phage types in broiler flocks in the EU, 2005-2006.
Table 11.	Comparison of S. Enteritidis phage types isolated from humans, broilers and laying
	hens
Table 12.	The proportion (%) of Salmonella Enteritidis-positive broiler flocks with resistant
	(*) isolates in reporting MSs of the EU, including the percentage of fully-susceptible
	isolates, in the EU, 2005-2006
Table 13.	The proportion (%) of Salmonella Enteritidis phage type-positive broiler flocks with
	resistant isolates (*) in reporting MSs of the EU, including the percentage of fully-
	susceptible isolates, in the EU, 2005-2006
Table 14.	The proportion (%) of Salmonella Typhimurium-positive broiler flocks with resistant
	(*) isolates in reporting MSs of the EU, including the percentage of fully-susceptible
	isolates, in the EU, 2005-2006
Table 15.	The proportion (%) of definitive phage type <i>S</i> . Typhimurium-positive broiler flocks
	with resistant isolates (*) in reporting MSs of the EU, including the percentage of
	fully-susceptible isolates, in the EU, 2005-2006



## **List of Figures**

Figure 1.	Number of broiler flocks sampled, per month, EU, Baseline survey in broiler flocks, 2005-2006
Figure 2.	Salmonella Enteritidis flock prevalence <sup>1</sup> with CI's, per month, in the EU, Baseline survey in broiler flocks, 2005-200614
Figure 3.	Prevalence <sup>1</sup> of flocks positive to serovars other than <i>Salmonella</i> Enteritidis and <i>Salmonella</i> Infantis, with CI's, per month, in the EU, Baseline survey in broiler
Figure 4.	The distribution on the estimated broiler flock prevalence of <i>S</i> . Enteritidis, <i>S</i> . Typhimurium, <i>S</i> . Enteritidis or <i>S</i> . Typhimurium, <i>S</i> . Infantis, <i>S</i> . Mbandaka and <i>S</i> . Hadar in countries participating in Baseline survey in broiler flocks, 2005-2006 18
Figure 5.	Frequency distribution of flocks positive to <i>S</i> . Enteritidis, <i>S</i> . Typhimurium, <i>S</i> . Infantis, <i>S</i> . Mbandaka and other <i>Salmonella</i> serovars in MSs, in the EU, 2005-2006.
Figure 6.	Distribution of the most frequent reported <i>Salmonella</i> serovars, most likely and secondary spatial clusters, relative risks, in the EU, 2005-2006,
Figure 7.	Comparison of the serovar distribution in human cases and broilers in MSs for which sufficient human data was available in either 2004 <sup>1</sup> or 2005 (preferred). Only the distribution of the most commonly reported human serovars, besides S. Enteritidis and S. Typhimurium, is presented
Figure 8.	Number of broiler flocks sampled per month, per Member State, 2005-2006
Figure 9.	Number of flocks sampled per age category of broilers in the flock, per Member State, 2005-2006
Figure 10.	Number of flocks sampled per number of cycles per year of the flock, per Member State, 2005-2006
Figure 11.	Most frequently identified flocks with <i>S</i> . Enteritidis phage types in the EU broiler survey, 2005-2006
Figure 12.	Most frequently identified flocks with <i>S</i> . Typhimurium phage types in the EU broiler flock baseline survey, 2005-2006



## List of Annexes

Annex I.	Correlation and cluster preliminary analysis	51
Annex II.	Statistical procedure	54
Annex III.	Spatial distribution of the Salmonella serovars at the EU level	57
Annex IV.	Graphical display of the number of sampled flocks, per Member State, 2005-2006.5	58
Annex V.	Univariate description of Salmonella-positive flocks for the investigated factors, at	
	the EU level	51
Annex VI.	Logistic regressions with random effect modelling	52
Annex VII.	Number of broiler flocks reported with the 20 most common Salmonella serovars,	
	per MS, 2005-2006	55
Annex VIII.	Most frequently identified Salmonella Enteritidis phage types in MSs, in the EU	
	broiler flock baseline survey, 2005-2006	56
Annex IX.	Most frequently identified Salmonella Typhimurium phage types in MSs, in the EU	ſ
	broiler flock baseline survey, 2005-2006.	70
Annex X.	Methodology adopted by reporting Member States and Norway to test for antibiotic	
	resistance of <i>Salmonella</i> isolates, in the EU broiler flock baseline survey, 2005-200	6.
	·	74
Annex XI.	Antimicrobials tested by reporting Member States, in the EU broiler flock baseline	
	survey, 2005-2006	75
Annex XII.	The proportion (%)* of Salmonella Enteritidis-positive broiler flocks with resistant	
	isolates in reporting MSs of the EU, including the percentage of fully-susceptible	
	isolates, in the EU broiler flock baseline survey, 2005-2006.	76
Annex XIII.	The proportion (%)* of <i>Salmonella</i> Typhimurium-positive broiler flocks with	
	resistant isolates in reporting MSs of the EU and Norway, including the percentage	
	of fully-susceptible isolates, in the EU broiler flock baseline survey, 2005-20067	77
Annex XIV.	The proportion (%)* of Salmonella spppositive broiler flocks with resistant isolate	es
	in reporting MSs of the EU, including the percentage of fully-susceptible isolates, in	n
	the EU broiler flock baseline survey, 2005-2006.	78
Annex XV.	The proportion (%) of broiler flocks positive to selected <i>Salmonella</i> serovars with	
	resistant isolates in reporting MSs of the EU, including the percentage of fully-	
	susceptible isolates, in the EU broiler flock baseline survey, 2005-2006,	30
Annex XVI.	Salmonella serovars in which no resistance or intermediate resistance was detected	
	in reporting Member States, in the EU broiler flock baseline survey, 2005-20068	36





## Annexes

## Annex I. Correlation and cluster preliminary analysis

## **Correlation matrix**

The correlation matrix between all factors to be investigated for the association with the *Salmonella* broiler flock prevalence was first calculated at the EU level, as shown below.

	Month	Delay	No. of flocks	No. of broilers	Prod type	Age of broilers	No. of cycles	Medication status
Month	1	0.01	-0.05	-0.05	-0.01	0.04	-0.04	-0.03
Delay	0.01	1	-0.04	0.02	-0.03	0.18	-0.08	0.07
No of flocks	-0.05	-0.04	1	0.03	-0.04	-0.08	-0.24	0.19
No of broilers	-0.05	0.02	0.03	1	0.01	-0.23	0.25	0.05
Prod. type	-0.01	-0.03	-0.04	0.01	1	0.26	0.06	-0.12
Age of broilers	0.04	0.18	-0.08	-0.23	0.26	1	-0.31	-0.04
No of cycles	-0.04	-0.08	-0.24	0.25	0.06	-0.31	1	0.04
Medication status	-0.03	0.07	0.19	0.05	-0.12	-0.04	0.04	1

The observed correlations can be considered to be low.

## **Group of countries**

A principal component analysis (PCA) was then performed in order to investigate possible grouping of countries based on their overall characteristics. In order to compare countries with each other, the MSs' mean was derived for each factor to proceed with PCA. Such analysis was performed with SAS version 9.1 PROC PRINCOMP, the outputs are reported below.

From Table AIII.2. showing eigen values from the PCA analysis, one can conclude that the selected variables are relatively uncorrelated between each other. The four first principal components are necessary to cumulate 80% of inertia. Countries are then scattered along the 2 first principal components (Figure AIII.1.) and then along the  $3^{rd}$  and  $4^{th}$  principal components (Figure AIII.2.), in order to visually assess whether such components could discriminate a sub group of countries. The most obvious conclusion is that Latvia, Estonia and Lithuania are clustered aside from the other countries by the first component. This is explained by the fact that the principal component strongly reflects the number of flocks per holding, which is much higher in those two countries than for the others. Components 3 and 4 also discriminate some countries from the others.





## Figure AIII.1. Scatter plot of countries on the 2 first principal components

Figure AIII.2. Scatter plot of countries on the 3<sup>rd</sup> and 4<sup>th</sup> principal components





## Table AIII.1. PCA: Eigen vectors

## The PRINCOMP Procedure

Eigenvectors

		Prin1	Prin2	Prin3	Prin4
month	the mean, month	510711	128621	066553	206964
nf	the mean, delay	0.567772	139512	229911	0.259347
nb prod	the mean, nb the mean, prod	0.210574 136018	0.524889 0.392090	0.178234 0.186881	0.138987 0.691337
age	the mean, age	293567	397467	0.434484	0.342672
nc med	the mean, nc the mean, med	087531 0.489936	0.546397 164185	0.316492 0.283276	460656 243958
		Firenet			

#### Eigenvectors

	Prin5	Prin6	Prin7	Prin8
month	0.579687	0.315855	0.480178	0.094838
delay	332271	0.016920	0.510407	0.217059
nf	0.315681	0.122114	0.056959	0.648260
nb	009596	0.766572	003336	203295
prod	0.266459	416750	0.252537	080231
age	0.182412	0.218764	596816	0.110981
nc	0.197521	216738	288312	0.460765
med	0.557245	169973	050694	501261

## Table AIII.2. PCA: Eigen values

Eigenvalues of the Correlation Matrix

	Eigenvalue	Difference	Proportion	Cumulative
1	2.17709623	0.38126332	0.2721	0.2721
2	1.79583291	0.40021429	0.2245	0.4966
3	1.39561862	0.41890771	0.1745	0.6711
4	0.97671090	0.28680884	0.1221	0.7932
5	0.68990207	0.10968110	0.0862	0.8794
6	0.58022097	0.30378339	0.0725	0.9519
7	0.27643757	0.16825683	0.0346	0.9865
8	0.10818074		0.0135	1.0000



## Annex II. Statistical procedure

### Statistical model

A logistic regression model with random effect on holdings was built, similarly to the ones used in Part A analysis to estimate flock prevalence. Again, the epidemiological unit of interest called "flock" was defined as a physical flock for a given holding in a given cycle. As a consequence, one physical flock over a year of 6 cycles for example corresponds to 6 different flocks in that sense. The random effect was taken as Gaussian on the probit scale (as usual, the probit function is defined as the inverse of the Cumulative Distribution Function of the normalized Gaussian distribution). The choice of probit versus any other link function (e.g. logit) was based on the convergence, but numerical results were not sensitive to that choice. For any holding *j* in a given country *i*, the number of positive flocks  $Y_{i,j}$  was assumed to follow a binomial distribution:

$$Y_{i,j} \sim \mathcal{B}in(p_{ij}, n_{ij})$$

Where  $p_{i,j}$  is the flock prevalence for a given outcome variable in this holding and  $n_{i,j}$  is the number of sampled flocks in this holding.  $p_{i,j}$  was defined via a holding (random) effect on the probit scale, namely:

probit
$$(p_{i,j}) = \eta_{ij}$$
  
And  $\eta_{ij} \sim N(\eta_i, s^2)$  for every holding j

 $\eta_i$  then represents the flock prevalence for a typical holding in the country *i* on the probit scale, prior to any covariate inclusion. In order to estimate the inter-holding variance  $s^2$ , this was assumed to be the same for all countries, following the same approach as developed for the part A analysis. To be consistent with the previous analysis, the variance was defined as growing with  $\eta_i$  for Salmonella Enteritidis and Salmonella Infantis, to account for the fact that countries with high prevalence may show larger inter-holding variability than those with smaller prevalence. In those cases, *s* was then specific to the country *i* (therefore denoted by  $s_i$ ) and defined as:

$$s_i = \alpha + \beta \eta_i$$

with  $\alpha$  and  $\beta$  being 2 positive regression parameters. For servors other then *S*. Enteritidis and or *S*. Infantis,  $s^2$  was chosen as not dependent on  $\eta_i$ .

### Forward-backward procedure

Based on the statistical model described above, the list of predefined potential factors or covariates was investigated to test for any statistical relationship with flock prevalence, for the three outcome variables (*Salmonella* Enteritidis, *Salmonella* Infantis, serovars other then *S*. Enteritidis and or *S*. Infantis), at the EU level. Prior to any investigation, a base model (with no



covariate) was first fitted to the data. The statistical methodology used for covariates screening was based on the standard two-step forward-backward approach in which covariates were added into or removed from the base model based on likelihood ratio tests with a significance level of 5%. More specifically:

• The forward (selection) step consisted in testing, for each single factor independently, the effect on the flock prevalence. Each continuous factor *X* (e.g. age of broilers, number of broilers per flock, etc..) was added to the regression according to a linear pattern on the probit scale:

$$\operatorname{probit}(p_{i,j}) = \eta_{ij} + \theta_X X_k$$

where  $X_k$  corresponds to the value of factor X on the flock k, and  $\theta_k$  is a parameter measuring the size of effect. Each categorical factor Y (e.g. month of sampling, vaccination status, etc.) taking values in the set  $\{y_1, ..., y_p\}$  was added to the regression on the probit scale as:

$$probit(p_{i,j}) = \eta_{ij} + \theta_{y_2} \mathbf{1}_{\{Y_k = y_2\}} + ... + \theta_{y_p} \mathbf{1}_{\{Y_k = y_p\}}$$

where  $Y_k$  corresponds to the value of factor Y on the flock k, and  $\theta_y$  are parameters measuring sizes of effect.

The statistical significance of covariates individually added one-by-one to the base model was then determined by comparing adequacy of the base model with the model with covariate. At the end of this selection process, a full model could be built, and aggregated all selected factors.

• The backward (elimination) step consisted in testing one-by-one whether each selected factor could be removed or not from the full model. At the end of this elimination process, a final model could be built, which integrated all significant factors.

All model comparisons were performed according to a likelihood ratio test with a level of significance of 5% for both forward selection and backward elimination. Since all models tested were nested, and their comparisons could be based on their objective function (-2\*Log Likelihood) as their difference asymptotically follows a chi-square distribution with degree of freedom equals to the difference in number of model parameters. The degree of freedom as well as the p-value computed from this test were reported in the tables of results for both forward and backward selections. All fits were performed using SAS version 9.1 with PROC NLMIXED, which also provided the evaluation of the objective function for each model. Note that in the case of *Salmonella* Infantis, the fit was not always possible for the backward selection due to the number of parameters and the low number of positive flocks. Therefore, it was decided in this case not to estimate the variance parameters ( $\alpha$  and  $\beta$ ), which were fixed according to estimates from the base model (i.e. respectively 1.36 and 0.6).



### **Evaluation of size of effects and odd ratios**

For each outcome variable, the final models (which include all remaining factors after the forward-backward selection) were fitted in order to derive parameter estimates. Then, an order of magnitude of each effect was evaluated based on those estimates, the range of variation of the corresponding factors and the EU-level flock prevalence estimates.

In addition, odd ratios were calculated as it is a common usage. However, those odd ratios were not direct outputs from the SAS procedures as a probit link was used in the models, not a logit one. Therefore, predictions of odd ratios by flock were derived in the PROC NLMIXED using the PREDICT statement where possible. Then, those predictions were averaged at the EU level, and the standard deviations derived.

### Additional modelling

Additional modelling was performed:

- On the "Month" variable: instead of categorical modelling, various Fourrier decompositions were attempted.
- On the "Age of broilers" variable: instead of linear modelling, quadratic modelling was attempted in order to investigate a possible age at maximum risk.



### Annex III. Spatial distribution of the Salmonella serovars at the EU level

As the location (coordinates) of the individual flocks participating in the survey was not known, the analysis of the serovar distribution could only be done at country level. The SaTScan method developed by Kulldorff was applied to detect spatial clusters of serovars in the EU MSs. This software estimates the probability that the frequency of events per trial at each vertex surpasses the expected frequency by chance. SaTScan uses circles and a non-parametric test statistic. It takes into account the observed number of cases inside and outside the circle when calculating the highest likelihood for each circle. SaTScan tests the null hypothesis against the alternative hypothesis that there is an elevated rate of cases within the windows as compared to the outside. The method uses the likelihood ratio  $\lambda$  as the test statistic. The significance of the test statistic  $\lambda$  is determined by a large number of replications of the data set generated under the null hypothesis in a Monte Carlo simulation. The likelihood ratio  $\lambda$  for each replica is computed, and the result is significant at the 0.05 level if the  $\lambda$  value of the real data set is among the top 5% of all the values, including the replicas. Secondary clusters with lower significance can also be identified.

SaTScan uses a circular window of different sizes to scan the survey area until a certain percent of the total population is included. This circle is the most probable cluster, and has a rate that is the least likely to happen by chance alone. SaTScan also accounts for multiple testing through the calculation of the highest likelihood of occurrence for all possible cluster locations and sizes. The Poisson model was chosen, which requires information about the number of estimated positive flocks over the one-year survey-period in each MS and broiler flock population data. The estimated number of positive cases for each serovar was calculated from the estimated prevalence. All estimated positive flocks were geocoded to the centroid of its respective country. The maximum window size was defined here as 50% of the cases and 999 replications were performed. Each serovar was analyzed independently. Only the most likely cluster and non-overlapping significant secondary clusters are displayed in this analysis. For the analysis, the SaTScan output was imported into Arc GIS 9 to create cluster maps to visually examine and compare the identified clusters. Additionally, SaTScan was also used to detect areas of significantly low rate of cases of each serovar.



The EFSA Journal (2007) 101, 1-86

Annex IV. Graphical display of the number of sampled flocks, per Member State, 2005-2006.

Figure 8. Number of broiler flocks sampled per month, per Member State, 2005-2006.



© European Food Safety Authority, 2007



The EFSA Journal (2007) 101, 1-86







The EFSA Journal (2007) 101, 1-86







# Annex V. Univariate description of *Salmonella*-positive flocks for the investigated factors, at the EU level

Factor	Category level	N obs	% pos			
			S. Enteritidis	S. Infantis	Serovars other than SE-SI	
Overall		7,108	7.6	4.2	9.9	
Flock-level						
Flock production type	conventional	6,156	7.6	4.6	10.8	
	non-conventional (free range standard or free range organic)	949	7.1	1.1	4.2	
	unknown	3	0.0	0.0	0.0	
Number of broilers in flock	100 - 11000	1,819	8.3	5.6	9.3	
	> 11000 - 17500	1,809	8.7	5.6	11.2	
	> 17500 - 26095	1,703	8.1	3.0	9.3	
	> 26095 - 234000	1,777	5.1	2.3	9.7	
Age of broilers at sampling (days)	4 - 21	1,924	3.3	1.1	10.1	
	22 - 28	1,957	7.9	4.7	10.6	
	29 - 35	1,652	11.1	5.3	9.2	
	36 - 125	1,575	8.6	6.0	9.5	
					0.0	
Number of cycles per year in the floc	x 0 - 4	1,306	6.7	1.6	10.9	
	5	2,036	13.5	6.4	11.1	
	6	2,119	6.2	5.8	10.2	
	7 - 13	1647	2.6	1.2	7.1	
Madiantian status	No	5 026	76	4.4	0.0	
Medication status	NO Ves	1 272	7.0	4.4	9.9	
Holding_level	105	1,272	1.5	5.2	10.1	
Number of houses	1	3 209	10.2	3.4	10.9	
Number of nouses	2	1 556	5 3	3.9	10.5	
	3	1 343	5.4	63	86	
	> 3	1,000	5.4	4.2	7.9	
Camanal						
<b>General</b> Days to bacteriological analysis	0	1 375	10.6	3.6	10.2	
,	1	2,980	6.3	4.9	8.8	
	2	1.352	4.1	6.8	12.1	
	>= 3	1,401	10.5	0.6	9.9	
Marth of annuling	T	462	7.6	2.2	10.2	
Monul of sampling	January February	402	7.0	3.2	12.5	
	March	491	7.1	5.5	11.8	
	April	466	J.8 4 9	4.0	9.9 10.1	
	May	720	4.7	2.5	8 5	
	June	72)	8.2	3.9	7.0	
	July	798	9.0	1.8	8.1	
	August	785	8.5	2.0	9.3	
	September	855	7.8	8.0	10.9	
	October	153	9.2	3.9	16.3	
	November	561	8.6	5.9	10.5	
	December	433	8.8	8.5	11.8	



## Annex VI. Logistic regressions with random effect modelling

Hereafter are reported tables of results corresponding to the forward-backward factors selection. Inclusion/elimination of factors was based on a likelihood ratio test at level 5%. The corresponding p-values are reported in the last column for each evaluated model. All models showing p-values below 5% are highlighted in italics.

## 1. S. Enteritidis model

### Forward Selection

Model	-2*LL	Degree of	p-value
		freedom	
Base	2599.0	NA	NA
Month	2568.4	11	0.0013
Days delay to lab	2596.4	1	0.1069
analysis			
Number of flocks	2592.4	1	0.0102
(houses) in the			
holding (Nf)			
Log (number of	2596.7	1	0.1294
broilers in the flock			
Nb)			
Flock production	2596.8	1	0.1380
type			
Age the broilers	2594.3	1	0.0302
Number of cycles in	2453.8	1	<0.0001
the flock (Nc)			
Medication status	2596	1	0.0833

### **Backward Selection**

model	-2*LL	Degree of freedom	p-value
Total (Month+Nf+Age+Nc)	2487.8	NA	NA
Total - Month	2521.6	11	0.0004
Total - nf	2490.8	1	0.0833
Total - age	2499.4	1	0.0007
Total - nc	2487.9	1	0.7518

## Final Model

The final model for Salmonella Enteritidis includes the following factors, from the most to the least significant:

• the month of sampling, and



• the age of broilers. The final objective function (-2\*LL) was 2489.9.

## Additional modelling

Additional modelling was performed:

- of the "Month" variable: instead of categorical modelling, various Fourrier decompositions were attempted but resulted in bad fits of data. Consequently the available data did not allow concluding a more precise effect.
- of the "Age of broilers" variable: instead of linear modelling, quadratic modelling was attempted in order to investigate a possible age at maximum risk. However, the fit was then driven by high ages (above 55 days) for which the prevalence was increasing with age. No categorical modelling was done as a country effect could then be confounded with the age effect. In conclusion, a more precise age effect could not be determined.

### 2. S. Infantis model

### Forward Selection

model	-2*LL	Degree of	p-value
		freedom	
base	1211.0	NA	NA
Month	1186.9	11	0.0123
Delay	1210.2	1	0.3711
Nf	1207	1	0.0455
Log(nb)	1177.7	1	<0.0001
Prod type	1201.1	1	0.0017
Age	1208.5	1	0.1138
Nc	1202.2	1	0.0030
Medication status	1198.5	1	0.0004

### **Backward Selection**

model	-2*LL	Degree of freedom	p-value
Total	1170.1	NA	NA
(Month+Nf+Nb+prod+Nc+med)			
Total-Month	1190.7	11	0.0378
Total-nf	1173.3	1	0.0736
Total-nb	1170.2	1	0.7518
Total-prod	1178.8	1	0.0032
Total-nc	1171.1	1	0.3173
Total -med	1174.3	1	0.0404



## Final Model

The final model for Salmonella Infantis includes the following factors, from the most to the least significant:

- Production type
- Month of sampling
- Medication status

The final objective function (-2\*LL) was 1175.3.

## **3.** Serovars other than *S*. Enteritidis and *S*. Infantis model

### Forward Selection

model	-2*LL	Additional No of parameters	p-value
base	3722.0	NA	NA
Month	3675.9	11	<0.0001
Delay	3722	1	1
Nf	3721.4	1	0.4386
Log(nb)	3721.4	1	0.4386
Prod type	3720.2	1	0.1797
Age	3718.5	1	0.0614
Nc	3709.2	1	0.0003
Medication status	3721.5	1	0.4795

## **Backward Selection**

Model	-2*LL	Degree of	p-value
		freedom	
Total (Month+Nc+Age)	3663.4	NA	NA
Month+Nc	3664.3	1	0.3428
Month+Age	3672.2	1	0.003
Age+Nc	3708.2	11	<0.0001

## Final Model

The final model for serovars other than *S*. Enteritidis and *S*. Infantis corresponds then to the "total model" obtained after the forward selection; it includes the following factors, from the most to the least significant:

- Month of sampling
- Number of cycles

There may also be an age effect but not detectable with a purely statistical approach.



						No. of	f Salm	onella	ı posi	tive b	roiler	flocks	s for t	he To	p 20 s	erova	rs per	MSs						
MS	AT	BE	CY	CZ	DE	DK	EE	ES	FI	FR	GR	HU	IE	IT	LT	LV	NL	PL	РТ	SE	SI	SK	UK	Total
No. flocks in	365	373	248	334	377	295	139	388	360	381	245	359	351	313	156	121	362	357	367	291	326	230	382	7120
survey																								
Salmonella serovar																								*
S. Enteritidis	6	0	7	48	4	0	3	124	0	2	9	18	0	12	8	9	5	123	138	0	9	13	0	538
S. Typhimurium	2	13	0	2	6	1	0	3	0	1	5	12	0	1	0	0	1	15	1	0	0	1	1	65
S. Infantis	2	1	0	10	6	2	0	3	0	2	0	209	0	1	0	0	8	38	11	0	1	1	0	295
S. Mbandaka	0	4	0	0	6	0	0	2	0	2	2	11	55	11	0	0	2	14	2	0	0	0	3	114
S. Senftenberg	1	8	2	0	0	0	0	1	0	1	3	1	2	1	0	0	0	2	1	0	0	0	5	28
S. Virchow	1	3	1	0	2	0	0	4	0	3	1	0	0	2	0	0	5	7	0	0	0	0	1	30
S. Agona	0	2	1	1	0	1	0	1	0	0	0	0	3	4	0	0	2	0	1	0	0	0	0	16
S. Anatum	0	4	0	0	13	0	0	1	0	3	1	1	0	0	0	0	1	0	8	0	0	0	0	32
S. Hadar	0	1	0	1	0	0	0	20	0	3	2	0	0	8	0	0	0	23	0	0	0	1	0	59
S. Livingstone	0	0	0	0	1	0	0	0	1	1	3	0	1	22	0	0	5	0	0	0	0	0	5	39
S. Indiana	0	2	0	0	2	0	0	0	0	2	5	0	5	0	0	0	1	2	0	0	0	0	0	19
S. Derby	0	0	0	1	0	2	0	0	0	0	0	0	0	1	0	2	1	6	0	0	0	0	0	13
S. Montevideo	15	0	0	7	0	0	0	0	0	3	1	0	0	4	0	0	0	0	0	0	0	0	1	31
S. Blockley	0	1	1	0	0	0	0	0	0	0	23	1	0	3	0	0	0	0	0	0	0	0	0	29
S. Kentucky	0	1	0	3	0	1	0	0	0	0	0	0	35	0	0	0	0	0	0	0	0	4	0	44
S. Newport	0	0	4	1	0	0	0	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0	1	8
S. Ohio	0	0	0	1	3	0	0	5	0	1	0	0	0	0	0	0	0	0	0	0	0	0	9	19
S. Bredeney	0	0	0	0	0	0	0	0	0	0	2	3	1	4	0	0	0	0	0	0	0	0	0	10
S. Heidelberg	0	0	0	0	2	0	0	0	0	1	0	0	0	4	0	0	0	0	3	0	0	0	0	10
S. Tennessee	1	0	0	0	1	0	0	0	0	0	0	0	0	2	0	0	0	0	1	0	0	0	0	5
Other serovars	0	30	12	0	33	2	0	13	0	11	15	20	1	39	0	0	10	5	4	0	0	1	17	213

## Annex VII. Number of broiler flocks reported with the 20 most common *Salmonella* serovars, per MS, 2005-2006.

\* Due to flocks infected with more than one serovar, the number of flocks with a specific serovar adds up to 1,617 and not 1,448



stria S. Enteritidis	phage types	(N=21)	Holdings with phag	e types (N=6)	Flocks with pl	nage types (N=6)
	Ν	%	Ν	%	Ν	%
PT4	15	71.43	4	66.67	4	66.67
PT8	1	4.76	1	16.67	1	16.67
PT2	5	23.81	1	16.67	1	16.67
Other phage typ	es 0	0.00				
Non typeable	0	0.00				
Total	21	100.00				

Annex VIII. Most frequently identified Salmonella Enteritidis phage types in MSs, in the EU broiler flock baseline survey, 2005-2006.

Czech Republic	S. Enteritidis phag	e types	(N=49)	Holdings with phage	types (N=48)	Flocks with ph	age types (N=48)
		Ν	%	Ν	%	Ν	%
	PT8	33	67.35	33	68.75	33	68.75
	PT4	8	16.33	8	16.67	8	16.67
	PT23	3	6.12	3	6.25	3	6.25
	PT7	2	4.08	2	4.17	2	4.17
	PT7a	1	2.04	1	2.08	1	2.08
	PT6	1	2.04	1	2.08	1	2.08
	PT14B	1	2.04	1	2.08	1	2.08
	Other phage types	0	0.00				
	Non typeable	0	0.00				
	Total	49	100.00				



Germany	S. Enteritidis phag	e types	(N=10)	Holdings with phag	e types (N=4)	Flocks with phage types (N=4)			
		Ν	%	Ν	%	Ν	%		
	PT13	7	70.00	2	50.00	2	50.00		
	PT8	2	20.00	1	25.00	1	25.00		
	PT21	1	10.00	1	25.00	1	25.00		
	Other phage types	0	0.00						
	Non typeable	0	0.00						
	Total	10	100.00						

aly	S. Enteritidis phag	e types	(N=25)	Holdings with phag	ge types (N=8)	Flocks with pl	nage types (N=8)
		Ν	%	Ν	%	Ν	%
	PT2	17	68.00	5	62.50	5	62.50
	PT4	5	20.00	1	12.50	1	12.50
	PT37	1	4.00	1	12.50	1	12.50
	PT21	1	4.00	1	12.50	1	12.50
	Other phage types	0	0.00				
	Non typeable	1	4.00	1			
	Total	25	100.00				

Latvia	S. Enteritidis phag	e types	(N=31)	Holdings with phag	ge types (N=1)	Flocks with pl	nage types (N=9)
		Ν	%	Ν	%	Ν	%
	PT1	11	35.48	1	100.00	6	66.67
	PT21	18	58.06	1	100.00	4	44.44
	PT7	2	6.45	1	100.00	1	11.11
	Other phage types	0	0.00				
	Non typeable	0	0.00				
	Total	31	100.00				



Slovakia	S. Enteritidis phag	ge types	(N=13)	Holdings with phage	types (N=13)	Flocks with ph	age types (N=13)
		Ν	%	Ν	%	Ν	%
	PT8	5	41.67	5	38.46	5	38.46
	PT4	2	16.67	2	15.38	2	15.38
	PT23	2	16.67	2	15.38	2	15.38
	PT6	1	8.33	1	7.69	1	7.69
	Non typeable	2	16.67	2			
	Total	12	100.00				
	Not tested	1	8.33	1	7.69	1	7.69
	Other phage types	0	0.00				

The Netherlands	S. Enteritidis phag	ge types	(N=12)	Holdings with phag	e types (N=5)	Flocks with pl	nage types (N=5)
		Ν	%	Ν	%	Ν	%
	PT21	10	83.33	4	80.00	4	80.00
	PT7	1	8.33	1	20.00	1	20.00
	PT1	1	8.33	1	20.00	1	20.00
	Other phage types	0	0.00				
	Non typeable	0	0.00				
	Total	12	100.00				



Figure 11. Most frequently identified flocks with *S*. Enteritidis phage types in the EU broiler survey, 2005-2006.





Annex IX. Most frequently identified Salmonella Typhimurium phage types in MSs<sup>1</sup>, in the EU broiler flock baseline survey, 2005-2006.

Austria	S. Typhimurium phage	types	(N=8)	Holdings with phage	e types (N=2) Fl	ocks with phage	e types (N=2)
		Ν	%	Ν	%	Ν	%
Ľ	DT85	5	62.50	1	50.00	1	50.00
Ľ	DT104L	3	37.50	1	50.00	1	50.00
C	Other phage types	0	0.00				
Ν	Non typeable	0	0.00				
Т	Total	8	100.00				
Czech Repub	olic S. Typhimurium	phage	types (N=	<b>2)</b> Holdings with	phage types (N=2)	Flocks with	phage types (N=2)
		<u> </u>	J	% N	0/0	N	0/2
			•	/0 11	/0	11	/0
	DTU302		1 50	0.00 1	50.00	1	50.00
	DTU302 DT114	- -	1 50 1 50	0.00 1 0.00 1	50.00 50.00	1	50.00 50.00
	DTU302 DT114 Other phage types		1 50 1 50 0 0	0.00 1 0.00 1 0.00 1	50.00 50.00	1	50.00 50.00
	DTU302 DT114 Other phage types Non typeable		1 50 1 50 0 0	0.00 1 0.00 1 0.00 1 0.00	50.00 50.00	1	50.00 50.00

© European Food Safety Authority, 2007

<sup>&</sup>lt;sup>1</sup> Norway reported one *S*. Typhimurium isolate that was non typeable.



Denmark	S. Typhimurium phage types (N=3)			Holdings with phag	ge types (N=1)	Flocks with phage types (N=1)	
		N	%	N	%	N	%
	DT15a	3	100.00	1	100.00	1	100.00
	Other phage types	0	0.00				
	Non typeable	0	0.00				
	Total	3	100.00				
Germany	S. Typhimurium phage types (N=19)			Holdings with pha	ge types (N=6)	Flocks with phage types (N=6)	
		Ν	%	Ν	%	Ν	%
	RDNC	4	21.05	2	33.33	2	33.33
	DT104B low	4	21.05	2	33.33	2	33.33
	DT208	5	26.32	1	16.67	1	16.67
	DT104L	4	21.05	1	16.67	1	16.67
	DT012	2	10.53	1	16.67	1	16.67
	Other phage types	0	0.00				
	Non typeable	0	0.00				
	Total	19	100.00				
Slovakia	S. Typhimurium phage types (N=1)			Ioldings with phage types (N=1)		Flocks with phage types (N=1)	
		Ν	%	Ν	%	Ν	%
	U302	1	100.00	1	100.00	1	100.00
	Other phage types	0	0.00				
	Non typeable	0	0.00				
	Total	1	100.00				



The Netherlands	S. Typhimurium p	hage types	s (N=5)	Holdings with phage types (N=1)		Flocks with phage types (N=1)	
		Ν	%	Ν	%	Ν	%
	FT 506	5	100.00	1	100.00	1	100.00
	Other phage types	0	0.00				
	Non typeable	0	0.00				
	Total	5	100.00				
The United King	dom <i>S</i> . Typhimuriur	n phage ty	pes (N=3)	Holdings with pha	ge types (N=1)	Flocks with <b>p</b>	phage types (N=1)
The United King	dom <i>S</i> . Typhimuriur	n phage ty N	pes (N=3) %	Holdings with pha N	ge types (N=1) %	Flocks with <b>p</b> N	bhage types (N=1) %
The United King	dom <i>S</i> . Typhimuriur DT104	n phage ty N 3	pes (N=3) % 100.00	Holdings with pha N 1	ge types (N=1) % 100.00	Flocks with J N 1	<b>bhage types (N=1)</b> % 100.00
The United King	dom S. Typhimuriur DT104 Other phage types	n phage ty N 3 0	<b>pes (N=3)</b> % 100.00 0.00	Holdings with pha N 1	<b>ge types (N=1)</b> % 100.00	Flocks with J N 1	<b>bhage types (N=1)</b> % 100.00
The United King	dom S. Typhimuriur DT104 Other phage types Non typeable	n phage ty N 3 0 0	<b>pes (N=3)</b> % 100.00 0.00 0.00	Holdings with pha N 1	<b>ge types (N=1)</b> % 100.00	Flocks with I N 1	<b>bhage types (N=1)</b> % 100.00


Figure 12. Most frequently identified flocks with *S*. Typhimurium phage types in the EU broiler flock baseline survey, 2005-2006.





Annex X. Methodology adopted by reporting Member States and Norway to test for antibiotic resistance of *Salmonella* isolates, in the EU broiler flock baseline survey, 2005-2006.

Member State	Antimicrobial resistance Method
Austria	MIC <sup>a</sup> determination method <sup>b</sup>
Belgium	Disc diffusion test <sup>c</sup>
Czech Republic	Disc diffusion test
Denmark	MIC determination method
Finland	Disc diffusion test
Germany	MIC determination method
Italy	Disc diffusion test
Latvia	Disc diffusion test
Lithuania	Disc diffusion test
Norway	MIC determination method
Poland	MIC determination method
Slovakia	MIC determination method
Slovenia	Disc diffusion test
The Netherlands	MIC determination method
The United Kingdom	Disc diffusion test

a: The minimum inhibitory concentration (MIC) is a basic quantitative measure of the *in vitro* activity of antibiotics. The MIC is the lowest concentration of the antibiotic that results in inhibition of visible growth (*i.e.* colonies on a plate or turbidity in broth culture) under standard conditions.

b: agar or broth dilution.

c: The disc diffusion susceptibility method is based on the principle of a filter paper disc impregnated with an antimicrobial placed on agar plate. The antimicrobial will diffuse from the disc into the agar. This diffusion will place the antimicrobial in the agar only around the disc. The solubility of the chemical and its molecular size will determine the size of the area of chemical infiltration around the disc. If an organism is placed on the agar it will not grow in the area around the disc if it is susceptible to the chemical. This area of no growth around the disc is known as a "zone of inhibition". The zone of inhibition in the disk diffusion test is inversely related to the MIC. This method is well documented and standard zones of inhibition have been determined for susceptible and resistant values. For some antimicrobials, there may also be a zone of intermediate resistance indicating that some inhibition occurs using this antimicrobial but it may not be sufficient inhibition to eradicate the organism from the body.





Country	Amikacin	Amoxicillin	Amoxicillin/ clavulanate	Ampicillin	Ampicillin/ Sulbactam	Apramycin	Cefazolin	Cefotaxime	Cefpodoxime	Ceftazidime	Ceftiofur	Cefuroxime	Cephalothin	Chloramphenicol	Ciprofloxacin	Colistin	Enrofloxacin	Florfenicol	Furazolidone	Gentamicin	Kanamycin	Nalidixic acid	Neomycin	Oxolinic acid	Spectinomycin	Streptomycin	Sulphonamide	Tetracycline	Trimethoprim	Trimethoprim/ Sulphonamide
Austria																														
Belgium				•							•						•						-							
Czech Republic																														
Denmark																														
Finland				•													•												•	
Germany																							-							
Italy																							-							
Latvia																														
Lithuania																														
Poland																														
Slovakia																														-
Slovenia		-															-						-							-
The Netherlands																														
United Kingdom	-																		-											-
Norway																													-	

## Annex XI. Antimicrobials tested by reporting Member States, in the EU broiler flock baseline survey, 2005-2006.



Annex XII. The proportion (%)\* of *Salmonella* Enteritidis-positive broiler flocks subjected to antimicrobial susceptibility testing subjected to antimicrobial susceptibility testing with resistant isolates in reporting MSs of the EU, including the percentage of fully-susceptible isolates, in the EU broiler flock baseline survey, 2005-2006.

Country	No. positive flocks	No. flocks with isolates tested	Percentage Fully Sensitive	Amikacin	Amoxicillin	Amoxicillin/ clavulanate	Ampicillin	Ampicillin/ sulbactam	Apramycin	Cefazolin	Cefotaxime	Ceftazidime	Ceftiofur	Cefuroxime	Cephalothin	Chloramphenicol	Ciprofloxacin	Colistin	Enrofloxacin	Florfenicol	Gentamicin	Kanamycin	Nalidixic acid	Neomycin	Oxolinic acid	Spectinomycin	Streptomycin	Sulphonamide	Tetracycline	Trimethoprim	Trimethoprim + Sulphonamide
All reporting MSs	237	**	64.3	0	0	1	7	0	0	0	0	0	0	4	5	0.9	0	3	6	0	0.4	2	21	0	0 (	).7	4	7	4	0	2
Austria	6	6	83.3		0		17		0				0		0	0	0	0		0	0		0	0		0	0	0	0	0	
Czech Republic	48	$48^{1}$	81.3			2	2		0		0				2	0	0	0	0		0	0	13	0			0	0	0	0	0
Germany	4	4	100			0	0						0			0	0	0		0	0	0	0	0		0	0	0	0	0	0
Italy	12	$10^{2}$	80	0		0	11			0	0	0			0	0	0	0	0		0	0	11	0		0	0		0		20
Latvia	9	8 <sup>3</sup>	25		0		50									40	0				0	13	100				38		60		0
Lithuania	8	8 4	75				0									0	0	0	0		0	0	0	0			0	25	0	0	0
Poland	123	123	50.4				9		0				0	4	7	0		5	10	0	0.8		28	0	(	).8	5	11	3	0	
Slovakia	13	12 5	91.7				0	0			0	0			0	0	0			0	0	8	0		0		0	0	0	0	0
Slovenia	9	9 <sup>6</sup>	77.8		0	0	0				0					0	0		0	0	0	0	22	0		0	0	0	11	0	0
The Netherlands	5	5	80				0				0	0				0	0			0	0		20	0				0	0	0	

The proportion of flocks with resistant isolates has been rounded to the nearest whole percentage, except where less than 1.

\*The figures include resistant isolates and isolates of intermediate susceptibility.

\*\*Varies for each antimicrobial; can be calculated by referring to figures for individual MSs.

<sup>1</sup>Czech Republic n=47 for trimethoprim.

<sup>4</sup>Italy n=1 for amikacin, cefazolin, neomycin, spectinomycin, n=8 for ceftazidime tetracyclines n=9 for amoxicillin/ clavulanate, ampicillin, cefotaxime, cephalothin, chloramphenicol, ciporofloxacin, enrofloxacin, gentamicin, kanamycin, nalidixic acid and streptomycin.

<sup>5</sup>Latvia n=4 for amoxicillin and ciprofloxacin, n=5 for chloramphenicol, gentamicin, nalidixic acid, tetracyclines and trimethoprim/ sulphonamides n=6 for ampicillin. <sup>4</sup>Lithuania n=5 for colistin.

<sup>5</sup>Slovakia n=11 for trimethoprim.

<sup>6</sup>Slovenia n=7 for spectinomycin; n=8 for trimethoprim.



Annex XIII. The proportion (%)\* of *Salmonella* Typhimurium-positive broiler flocks subjected to antimicrobial susceptibility testing subjected to antimicrobial susceptibility testing with resistant isolates in reporting MSs of the EU and Norway, including the percentage of fully-susceptible isolates, in the EU broiler flock baseline survey, 2005-2006.

Country	No. flocks positive	No. flocks with isolates tested	Percentage Fully Sensitive	Amikacın Amoxicillin Amoxicillin/ clavulanate	Ampicillin	Ampicillin/ sulbactam Apramvcin	Cefotaxime	Ceftazidime	Ceftiofur	Cefuroxime	Cephalothin	Chloramphenicol	Ciprofloxacin	Colistin	Enrofloxacin	Florfenicol	Furazolidone	Gentamicin	Kanamycin	Nalidixic acid	Neomycin	Oxolinic acid	Spectinomycin	Streptomycin	Sulphonamide	Tetracycline	Trimethoprim	Trimethoprim + Sulphonamide
All reporting MSs	41	**	15	0 0 40	73	0 (	) ()	0	3	6	0	53	0	0	11	51	0	3	22	40	5	0	75	69	75	65	14	20
Austria	2	2	50	0	0	(	)		0		0	0	0	0		0		0		0	0		50	50	50	0	0	
Belgium	12	$11^{-1}$	27.3		55				9			27			0	27		0		0	0			36	55	36		10
Czech Republic	2	2	0	0	100	(	) 0				0	0	0	0	0		4	50	50	50	50			50	50	100	0	0
Denmark	1	1	0	0	100	(	)		0			0	0	0		0		0		0	0		100	100	100	0	100	
Germany	6	6	33.3	67	67				0			50	0	0		50		0	17	17	17		50	67	67	67	50	50
Poland	15	15	0		87	(	)		0	7	0	87		0	20	80		0		87	0		87	100	100	93	0	
Slovakia	1	1	0		100	0	0	0			0	0	0			0		0	0	0		0		0	0	0	0	0
The Netherlands	1	1	0		100		0	0				100	0			100		0		0	0				100	100	0	
The United Kingdom	1	1	0	0 0	100	(	)	0		0		100	0				0	0		100	0			100	100	100		0
Norway	1	1	0		100		0		0			100	0			0		0	0	0				100	100	0	100	

The proportion of resistant isolates has been rounded to the nearest whole percentage, except where less than 1.

\*The figures include resistant isolates and isolates of intermediate susceptibility.

\*\*Varies for each antimicrobial; can be calculated by referring to figures for individual MSs.

<sup>1</sup>Belgium n=10 for trimethoprim/ sulphonamide.



Annex XIV. The proportion (%)\* of *Salmonella* spp.-positive broiler flocks subjected to antimicrobial susceptibility testing with resistant isolates in reporting MSs of the EU and Norway, including the percentage of fully-susceptible isolates, in the EU broiler flock baseline survey, 2005-2006.

Country	No. flocks positive	No. flocks with isolates tested	Percentage Fully Sensitive	Amikacin	Amoxicillin	Amoxicillin/ clavulanate	Ampicillin	Ampicillin/ sulbactam	Apramycin	Cefazolin	Cefotaxime	Cefpodoxime	Ceftazidime	Ceftiofur	Cefuroxime	Cephalothin	Chloramphenicol	Ciprofloxacin	Colistin	Enrofloxacin	Florfenicol	Furazolidone	Gentamicin	Kanamycin	Nalidixic acid	Neomycin	Oxolinic acid	Spectinomycin	Streptomycin	Sulphonamide	Tetracycline	Trimethoprim	Trimethoprim + Sulphonamide
All reporting MSs	656	**	50.3	0	0	6	19	0	0	33	3	0	4	1	3	7	7	2	2	9	5	3	1	5	24	3	0	18	27	19	16	7	13
Austria	28	27	85.2		0		11		0					0		0	0	0	0		0		0		0	0		4	4	7	0	0	
Belgium	52	49 <sup>1</sup>	61.2				27							8	0		6			0	6		0		18	0			14	25	14		15
Czech Republic	75	75 <sup>2</sup>	78.7			1	7		0		0					3	1	0	0	0			1	1	13	1			3	3	4	0	0
Denmark	9	9 <sup>3</sup>	44.4			0	33		0			0		0		0	0	0	0		0		0		0	0		22	56	22	11	22	
Finland	1	1	0				0				0						0			0					0					0	100	0	
Germany	65	65	41.5			12	31							2			9	9	0		9		0	5	12	5		20	51	29	15	28	23
Italy	94	37 <sup>4</sup>	58.5	0		11	33			33	5		7		0	19	0	0	0	14			5	16	35	30		0	24	88	32	25	15
Latvia	11	7 <sup>5</sup>	28.6		0		50										29	0					0	10	71				40		57		0
Lithuania	8	8 <sup>6</sup>	75				0										0	0	0	0			0	0	0	0			0	25	0	0	0
Poland	206	206	31.6				18		0					0	4	7	10		3	14	6		1		37	0.5		21	39	19	18	0.5	
Slovakia	19	19 <sup>7</sup>	78.9				11	0			0		0			0	0	0			0		0	5	5		0		0	0	0	0	0
Slovenia	10	10 8	80		0	0	0				0						0	0		0	0		0	0	20	0		0	0	0	10	0	0
The Netherlands	37	37	54.1				30				8		8				3	0			3		3		27	3				24	14	22	
The United Kingdom	41	36	30.6	0		0	6		0				0		0		19	3				3	0		17	14			14	36	31		33
Norway	1	1	0				100				0			0			100	0			0		0	0	0				100	100	0	100	

The proportion of flocks with resistant isolates has been rounded to the nearest whole percentage, except where less than 1.

\*The figures include resistant isolates and isolates of intermediate susceptibility.

\*\*Varies for each antimicrobial; can be calculated by referring to figures for individual MSs.

<sup>1</sup>Belgium number of isolates tested (n) =1 for cefuroxime, n=48 for trimethoprim/ sulphonamides.

<sup>2</sup>Czech Republic n=74 for trimethoprim.

<sup>3</sup>Denmark n=8 for cefpodoxime and cephalothin.



The EFSA Journal (2007) 101, 1-86

<sup>4</sup>Italy n=1 for cefuroxime, n=4 for trimethoprim, n=8 for sulphonamides, n=9 for amikacin, cefazolin, spectinomycin, n=10 for neomycin, n=28 for ceftazidime, n=38 for gentamicin, n=39 for ampicillin, n=41 for trimethoprim/ sulphonamides. <sup>5</sup>Latvia n=4 for amoxicillin and ciprofloxacin, n=5 for gentamicin, n=6 for ampicillin, n=10 for kanamycin and streptomycin.

<sup>6</sup>Lithuania n=5 for colistin.

<sup>7</sup>Slovakia n=18 for trimethoprim.
<sup>8</sup>Slovenia n=8 for spectinomycin; n=9 for trimethoprim.



The EFSA Journal (2007) 101, 1-86

Annex XV. The proportion (%) of broiler flocks positive to selected *Salmonella* serovars subjected to antimicrobial susceptibility testing with resistant isolates in reporting MSs of the EU, including the percentage of fully-susceptible isolates, in the EU broiler flock baseline survey, 2005-2006.

Antimicrobial resistance (\*) in Salmonella Infantis in broiler flocks.

Country	No. flocks positive	No. flocks with isolates tested	Percentage Fully Sensitive	Amoxicillin	Amoxicillin/ clavulanate	Ampicillin	Ampicillin/ sulbactam	Apramycin	Cefotaxime	Cefpodoxime	Cefta zidime	Ceftiofur	Cefuroxime	Cephalothin	Chloramphenicol	Ciprofloxacin	Colistin	Enrofloxacin	Florfenicol	Gentamicin	Kanamycin	Nalidixic acid	Neomycin	Oxolinic acid	Spectinomycin	Streptomycin	Sulphonamide	Tetracycline	Trimethoprim	Trimethoprim + Sulphonamide
All reporting MSs	70	69	54.4	0	0	12	0	0	5	0	11	0	0	4	6	0	2	10	0	0	0	7	0	0	13	33	10	0	4	11
Austria	2	1	100	0		0		0				0		0	0	0	0		0	0		0	0		0	0	0	0	0	
Belgium	1	1	100			0						0			0			0	0	0		0	0			0	0	0		0
Czech Republic	10	10	70		0	20		0	0					10	10	0	0	0		0	0	20	0			0	0	0	0	0
Denmark	2	2	100		0	0		0		0		0		0	0	0	0		0	0		0	0		0	0	0	0	0	
Germany	6	6	33.3		0	33						0			0	0	0		0	0	0	0	0		0	67	33	0	33	33
Italy	1	1	0																										100	
Poland	38	38	39.5			8		0				0	0	3	8		3	13	0	0		8	0		16	42	11	0	0	
Slovakia	1	1	100			0	0		0		0			0	0	0			0	0	0	0		0		0	0	0	0	0
Slovenia	1	1	100	0	0	0			0						0	0		0	0	0	0	0	0		0	0	0	0	0	0
The Netherlands	8	8	87.5			13			13		13				0	0			0	0		0	0				0	0	0	

The proportion of flocks with resistant isolates has been rounded to the nearest whole percentage, except where less than 1. \*The figures include resistant isolates and isolates of intermediate susceptibility.



## Antimicrobial resistance (\*) in *Salmonella* Virchow in broiler flocks.

Country	No. flocks positive	No. flocks with isolates tested	Percentage Fully Sensitive	Amikacin	Amoxicillin	Amoxicillin/ clavulanate	Ampicillin	Apramycin	Cefazolin	Cefotaxime	Ceftazidime	Ceftiofur	Cefuroxime	Cephalothin	Chloramphenicol	Ciprofloxacin	Colistin	Enrofloxacin	Florfenicol	Furazolidone	Gentamicin	Kanamycin	Nalidixic acid	Neomycin	Spectinomycin	Streptomycin	Sulphonamide	Tetracycline	Trimethoprim	Trimethoprim + Sulphonamid
All reporting MSs	21	20	15	0	0	0	15	0	0	0	0	0	0	0	10	0	0	27	0	0	0	33	85	5	18	27	15	20	13	0
Austria	1	1	100		0		0	0				0		0	0	0	0		0		0		0	0	0	0	0	0	0	
Belgium	3	3	0				0					0			0			0	0		0		100	0		0	0	0		0
Germany	2	2	50			0	0					0			0	0	0		0		0	50	50	50	0	0	0	50	0	0
Italy	2	1	0	0		0	0		0	0				0	0	0	0	0			0	0	100	0	0	0	100	100		0
Poland	7	7	0				14	0				0	0	0	29		0	43	0		0		100	0	29	57	0	0	0	
										0	0				0	0			0		0		100	0			10	10	10	
The Netherlands	5	5	0				40			0	0				0	0			0		0		100	0			40	40	40	

The proportion of flocks with resistant isolates has been rounded to the nearest whole percentage, except where less than 1. \*The figures include resistant isolates and isolates of intermediate susceptibility.



Antimicrobial resistance (\*) in Salmonella Hadar in broiler flocks.

Country	No. flocks positive	No. flocks with isolates tested	Percentage Fully Sensitive	Amikacin	Amoxicillin/ clavulanate	Ampicillin	Ampicillin/ sulbactam	Apramycin	Cefazolin	Cefotaxime	Ceftazidime	Ceftiofur	Cefuroxime	Cephalothin	Chloramphenicol	Ciprofloxacin	Colistin	Enrofloxacin	Florfenicol	Gentamicin	Kanamycin	Nalidixic acid	Neomycin	Oxolinic acid	Spectinomycin	Streptomycin	Sulphonamide	Tetracycline	Trimethoprim	Trimethoprim + Sulphonamide
All reporting MSs	34	**	0	0	25	35	0	0	100	0	0	0	4	29	0	0	0	32	0	0	20	97	4	0	36	86	14	76	0	0
Belgium	1	1	0			0						0			0			0	0	0		100	0			0	0	0		0
Czech Republic	1	1	0		0	0		0		0				0	0	0	0	0		0	0	100	0			0	0	0	0	0
Italy	8	3	0	0	33	100			100	0	0			67	0	0	0	100		0	33	100	50		0	100	100	100		0
Poland	23	23	0			30		0				0	4	26	0		0	26	0	0		96	0		39	96	8	83	0	
Slovakia	1	1	0			0	0			0	0			0	0	0			0	0	0	100		0		0	0	0	0	0

The proportion of flocks with resistant isolates has been rounded to the nearest whole percentage, except where less than 1.

\*The figures include resistant isolates and isolates of intermediate susceptibility. \*\*Varies for each antimicrobial; can be calculated by referring to figures for individual MSs. Italy n=2 for amikacin, cefazolin, colistin, neomycin, spectinomycin, sulphonamides

n=1 for ceftazidime



Antimicrobial resistance (\*) in Salmonella Paratyphi B var. Java in broiler flocks.

Country	No. flocks positive	No. flocks with isolates tested	Percentage Fully Sensitive	Amoxicillin/ clavulanate	Ampicillin	Cefotaxime	Ceftazidime	Ceftiofur	Chloramphenicol	Ciprofloxacin	Colistin	Enrofloxacin	Florfenicol	Gentamicin	Kanamycin	Nalidixic acid	Neomycin	Spectinomycin	Streptomycin	Sulphonamide	Tetracycline	Trimethoprim	Trimethoprim + Sulphonamide
All reporting MSs	19	19	0	57	79	29	29	33	5	43	0	0	5	0	0	63	0	100	83	63	26	93	75
Delatore	5	5	0		100			60	0			0	0	0		80	0		60	80	40		100
Beigium	5	5	0																				
Germany	7	7	0	57	57			14	14	86	0		14	0	0	86	0	100	100	57	43	100	57

The proportion of flocks with resistant isolates has been rounded to the nearest whole percentage, except where less than 1. \*The figures include resistant isolates and isolates of intermediate susceptibility.



## Antimicrobial resistance (\*) in Salmonella serovars in broiler flocks.

Salmonella serovar	No. flocks positive	No. flocks with isolates tested $^{**}$	<b>Percentage Fully Sensitive</b>	Amikacin	Amoxicillin	Amoxicillin/ clavulanate	Ampicillin	Apramycin	Cefazolin	Cefotaxime	Cefpodoxime	Ceftazidime	Ceftiofur	Cefuroxime	Cefuroxime	Cephalothin	Chloramphenicol	Ciprofloxacin	Colistin	Enrofloxacin	Florfenicol	Furazolidone	Gentamicin	Kanamycin	Nalidixic acid	Neomycin	Spectinomycin	Streptomycin	Sulphonamide	Tetracycline	Trimethoprim	Trimethoprim + Sulphonamide
Anatum	18	18	50			0	28			0		0	0				11	0	0	0	11		0	8	0	6	15	41	28	6	14	12
Derby	12	12	42			0	10	0		0	0	0	0	16		0	0	0	0	0	0		10	0	0	10	13	55	20	33	10	0
Livingstone	35	28	74	0		5	7	0	0	5		4	0	0		7	0	0	0	0	0	0	4	0	7	0	0	10	0	11	0	10
Mbandaka	40	29	28	0		11	10	0	100	0		0	0	0		6	10	0	0	0	0	0	0	13	7	4	43	59	7	7	0	0
Montevideo	27	24	83	0	0	0	8	0		0		0	0	0		0	0	0	0	0	0	0	0	0	8	0	0	4	9	4	0	0
Ohio	13	13	23	0		0	8	0		0		0	0	0		0	0	0	0	0	0	0	0	0	0	39	0	8	69	46	25	69
Senftenberg	17	16	56	0	0	0	0	0		0		0	0	0	0	0	25	14	0	0	0	0	0	0	25	0	0	13	0	0	0	0
Thompson	10	5	0	0		0	60	0	0	0		0		0	0	25	0	0	0	25		0	0	75	60	40	0	40	100	60		20

The proportion of flocks with resistant isolates has been rounded to the nearest whole percentage, except where less than 1.

\*The figures include resistant isolates and isolates of intermediate susceptibility.

\*\*Varies for each antimicrobial; can be calculated by referring to the figures below.

Numbers of Isolates Tested.

Anatum n=17 for ceftiofur, trimethoprim/ sulphonamides n=14 for trimethoprim, sulphonamides, ciprofloxacin, n=13 for amoxicillin/ clavulanic acid, colistin, kanamycin, spectinomycin, n=4 enrofloxacin, n=1 cefotaxime, ceftazidime

Derby n=11 for streptomycin, n=10 for ampicillin, gentamicin, neomycin, trimethoprim, n=9 for apramycin, cephalothin, colistin, florfenicol, n=8 spectinomycin, n=7 enrofloxacin, n=6 cefuroxime, n=4 ciprofloxacin, n=3 amoxicillin/ clavulanate, kanamycin triemthprim/ sulphonamides n=2 cefotaxime, cefpodoxime, n=1 ceftazidime, Livingstone n=26 for ciprofloxacin, gentamicin, n=24 for ceftazidime, n=23 for sulphonamides, n=21 for amoxicillin/ clavulanate, cefotaxime, streptomycin, trimethoprim/ sulphonamide n=16 for colistin, enrofloxacin, kanamycin, n=15 cephalothin, n=12 neomycin, n=7 for trimethoprim, n=6 for amikacin, florfenicol, n=5 for apramycin, cefuroxime, furazolidone, n=2 spectinomycin, n=1 ceftazidire.



The EFSA Journal (2007) 101, 1-86

Mbandaka n= 2 amikacin, n=9 amoxicillin/ clavulanate, n=29 ampicillin, chloramphenicol, gentamicin, nalidixic acid, tetracyclines, n=15 apramycin, cefuroxime, n=1 cefazolin, furazolidone, n=13 trimethoprim/ sulphonamides n=4 cefotaxime, ceftazidime, n=24 ceftiofur, n=16 cephalothin, n=8 kanamycin, n=11 ciprofloxacin, n=22 colistin, trimethoprim, n=20 enrofloxacin, n=21 spectinomycin, n=26 florfenicol, n=27 streptomycin, n=28 neomycin, sulphonamides.

Montevideo n=23 for apramycin, cephalothin, colistin, neomycin, trimethoprim, sulphonamides, n=1 for amikacin, cefuroxime, furazolidone, n=10 for trimethoprim/ sulphonamide, n=9 for amoxicillin/ clavulanate, n=15 for amoxicillin, ceftiofur, florfenicol, spectinomycin, n=8 for cefotaxime, enrofloxacin, kanamycin, Ohio n=0 for amikacin, ceftorizing activation n=10 for amoxicillin ceftorizing activation n=10 for a state n=10 f

Ohio n=9 for amikacin, ceftazidime, cefuroxime, furazolidone, n=10 for apramycin, n=1 for cefotaxime, cephalothin, enrofloxacin, n=3 for ceftiofur, florfenicol, spectinomycin, n=4 for colistin, kanamycin, trimethoprim

Senftenberg n=5 for amikacin, furazolidone, n=1 for amoxicillin, cefotaxime, cefuroxime, kanamycin, n=6 amoxicillin/ clavulanate, ceftazidime, n=16 for ampicillin, chloramphenicol, gentamicin, nalidixic acid, streptomycin, tetracyclines n=8 for apramycin, n=13 for trimethoprim/ sulphonamides n= 10 for ceftiofur, enrofloxacin, florfenicol, n=7 cefuroxime, ciprofloxacin, n=4 cephalothin, colistin, n=15 neomycin, sulphonamides n=3 spectinomyin, trimethoprim,

Thompson n=4 for cephalothin, enrofloxacin, kanamycin, n=3 for cefazolin, cefotaxime, colistin, cefuroxime, spectinomycin, n=1 for apramycin, ceftazidime, furazolidone Excludes serovars listed in Table Q which were susceptible to the panels of antimicrobials tested in MSs.



Annex XVI. *Salmonella* servors in which no resistance or intermediate resistance was detected in reporting Member States, in the EU broiler flock baseline survey, 2005-2006.

Member State	Salmonella serovars in which no antimicrobial resistance
	was detected to the panel of antimicrobials tested
	(number of isolates of that serovar examined).
All Member States	Banana (1), Cerro (1), Coeln (1), Cubana (2), Goldcoast (1),
reporting about	Havana (1), Idikan (1), Lille (1), London (1), Nagoya (1),
antimicrobial	Newport (2), Veneziana (1), Worthington (1), Yoruba (1)
susceptibility	
testing (13)	
Austria	Senftenberg (1), Tennessee (1), Virchow (1)
Belgium	Agona (2), Anatum (4), Banana (1), Blockley (1), Branderup
	(1), Cerro (1), Cubana (2), Havana (1), Indiana (1), Infantis
	(1), Kentucky (1), Mbandaka (4), Nagoya (1), Worthington
	(1)
Czech Republic	Agona (1), Derby (1), Kentucky (3), Newport (1), Ohio (1)
Denmark	Infantis (2), Kentucky (1)
Germany	Enteritidis (4), Heidelberg (2)
Italy	Coeln (1), Isangi (1), Orion (1), Senftenberg (1), Veneziana
	(1)
Slovakia	Infantis (1), Lille (1)
Slovenia	Infantis (1)
The Netherlands	Agona (2), Anatum (1), Goldcoast (1), Indiana (1), Yoruba
	(1)
The United Kingdom	Idikan (1), London (1), Newport (1), Virchow (1)