REFERENCES


FURTHER PUBLICATIONS OF INTEREST


http://dx.doi.org/10.1371/journal.pone.0023713.


Kirchner M, Marier E, Miller A, Snow L, Mclaren I, Davies RH, Clifton-Hadley FA and Cook AJC (2011) Application of variable number of tandem repeat analysis to track Salmonella enterica ssp enterica serovar Typhimurium infection of pigs reared on three British farms
through the production cycle to the abattoir. Journal of Applied Microbiology 111 (4), pp 960-970.


region, Salmonella enterica phage type and bird species. Veterinary Record 166 (14), pp 419-421.


Quality Statement

SECTION A

1. Coherence
Reports are obtained by various routes: direct submissions to AHVLA Regional Laboratories, reports of *Salmonella* isolations by private laboratories and Scottish submissions to Scottish Agricultural Colleges.

AHVLA is responsible for collation of data. Submissions result from cases of clinical disease in livestock, monitoring of healthy livestock and investigations of possible links with a human *Salmonella* outbreak.

All private laboratories submitting reports of *Salmonella* isolates to AHVLA do so using the standard AHVLA submission & supplementary forms or customised forms developed for them by AHVLA. Scottish submissions use the SRUC submission form & supplementary forms which are compatible with the AHVLA system and interpreted in the same way. All use the same definitions and essential categorisation.

All submissions are included in this report including those still under investigation with pending results.

An incident comprises the first isolation and all subsequent isolations of the same serovar or serovar and phage/definitive type combination of a particular *Salmonella* from an animal, group of animals or their environment on a single premises, within a defined time period (usually 30 days).

An antimicrobial susceptibility test is performed for surveillance purposes against an extended panel of 16 antimicrobials on *Salmonella* isolates sent for serotyping to AHVLA Weybridge and AHVLA Lasswade.

<table>
<thead>
<tr>
<th>Antimicrobial</th>
<th>Concentration (µg per ml)</th>
<th>Code</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 Nalidixic acid</td>
<td>30</td>
<td>NA</td>
</tr>
<tr>
<td>2 Tetracycline</td>
<td>10</td>
<td>T</td>
</tr>
<tr>
<td>3 Neomycin</td>
<td>10</td>
<td>N</td>
</tr>
<tr>
<td>4 Ampicillin</td>
<td>10</td>
<td>AM</td>
</tr>
<tr>
<td>5 Furazolidone</td>
<td>15</td>
<td>FR</td>
</tr>
<tr>
<td>6 Ceftazidime</td>
<td>30</td>
<td>CAZ</td>
</tr>
<tr>
<td>7 Sulphamethoxazole/trimethoprim</td>
<td>25</td>
<td>TM</td>
</tr>
<tr>
<td>8 Chloramphenicol</td>
<td>30</td>
<td>C</td>
</tr>
<tr>
<td>9 Amikacin</td>
<td>30</td>
<td>AK</td>
</tr>
<tr>
<td>10 Amoxicillin/clavulanic acid</td>
<td>30</td>
<td>AMC</td>
</tr>
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### Table: Antimicrobial Concentrations and Codes

<table>
<thead>
<tr>
<th>Antimicrobial</th>
<th>Concentration (µg per ml)</th>
<th>Code</th>
</tr>
</thead>
<tbody>
<tr>
<td>11 Gentamicin</td>
<td>10</td>
<td>CN</td>
</tr>
<tr>
<td>12 Streptomycin</td>
<td>10</td>
<td>S</td>
</tr>
<tr>
<td>13 Sulphonamide compounds</td>
<td>300</td>
<td>SU</td>
</tr>
<tr>
<td>14 Cefotaxime</td>
<td>30</td>
<td>CTX</td>
</tr>
<tr>
<td>15 Apramycin</td>
<td>15</td>
<td>APR</td>
</tr>
<tr>
<td>16 Ciprofloxacin</td>
<td>1</td>
<td>CIP</td>
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</table>

This panel is updated when there is a clear need to detect new or emergent types of resistance or to replace outdated antimicrobials. On specific occasions (e.g. detection of *Salmonella* vaccine strains, characterisation of 3rd generation cephalosporins resistance) more than 16 antimicrobials are used for susceptibility testing.

From 1st January 2007 some of the breakpoints used in assessing antimicrobial resistance, which were previously set at less than or equal to 13mm, have changed. These new breakpoints were set at: Ceftazidime (CAZ) less than or equal to 27mm, Amikacin (AK) less than or equal to 18mm, Ciprofloxacin (CIP) less than or equal to 19mm and Cefotaxime (CTX) less than or equal to 29mm. This may result in an increased number of isolates resistant to these antimicrobials in 2007 in comparison with previous years. The breakpoint for all other antimicrobials used remains at less than or equal to 13mm.

In 2008, the disc concentrations for streptomycin and chloramphenicol were changed to adopt the disc concentrations recommended by BSAC. In the case of streptomycin, the disc concentration was reduced from 25µg to 10µg. The zone size remained unchanged, so this change would be expected to increase the detection of isolates with lower level streptomycin resistance. Work done at AHVLA has shown that the 10µg disc provides much better discrimination between resistant and sensitive isolates (defined using the gold standard measure of MIC determination) than the 25µg disc.

The only other change made to the breakpoints and disc concentrations used over the period 2008-2013 related to the ceftazidime disc where the zone size was reduced from 29 to 26mm in 2012, in line with BSAC recommendations.

Some of the *Salmonella* serovars are recorded and reported in AHVLA under the old nomenclature. The nomenclature for these serovars under the Kauffmann-White scheme is clarified in the table below:
AHVLA Serovar | White-Kauffmann-Le Minor Serovar
Pullorum          | Gallinarum (biovar Pullorum)
Java              | Paratyphi B var. Java
Newington         | Anatum var. 15+

The Salmonella serovars S. Binza and S. Thomasville which were previously recorded by the AHVLA under their old nomenclature are now recorded using the White-Kauffmann-Le Minor notation as Salmonella Orion var. 15+ and Salmonella Orion var. 15+, 34+ respectively; however for 2009 both these variants are shown in the tables as S. Orion only. This change was implemented during 2008.

2. Accuracy and precision

Sampling error: Isolations of Salmonella from statutory species are required to be reported, however the level of testing (for species without a NCP) depends on submission of samples for laboratory investigation by private vets as well as on economic factors e.g. distance to laboratories etc.

A susceptibility test is often performed on representative Salmonella isolates before the allocation of an automatic incidence reference by the computer system. It is important for the Regional Laboratories to provide information to the testing laboratory on whether the submitted isolates are considered to comprise new incidents. As some companies perform extensive testing for Salmonella, this could skew the overall antimicrobial resistance data leading to the patterns obtained, at least in part, reflecting the intensity of sampling procedure. Also, limited resources may prevent susceptibility testing of all isolates.

Coverage error: The reasons for sample submissions (particularly for non-NCP samples) need to be considered, as sources of error can be dependent on this factor. Also the ability to isolate Salmonella needs to be considered (dependent on sample type taken, age of sample, storage and transport, culture method used, laboratory staff technical expertise etc).

Non-response error: Although all Salmonella isolations from statutory species are required to be reported, not all data items requested are mandatory under the Zoonoses Order. Different categories of submissions may have different non-response rates for different data items.

Measurement error: Different Salmonella culture methods vary in their sensitivity, which varies according to sample type, type of Salmonella present and profile of competitive flora in the sample. Data on the
AHVLA and SRUC forms are subject to individual interpretation by the person submitting the information, despite the guidance to authorised personnel.

The requirement of this report is to include as much data as is available. However only approved submissions are included, although efforts are made to ensure that all submissions are approved before the data is extracted. Data are scrutinised to correct errors in results for strategically important isolates (e.g. resistant to 3rd generation cephalosporins, resistant to ACSSuT pattern). It is not expected to routinely see resistance to amikacin, ciprofloxacin, ceftazidime or cefotaxime in any isolate. If any appears, it is followed up at the time of detection and the isolate would normally be re-tested.

Both laboratories at AHVLA Lasswade and AHVLA Weybridge that perform the expanded susceptibility testing have third party accreditation to ISO17025 provided by UKAS.

**Data processing error:** It is often difficult to obtain the required information from the sample submitted for non-mandatory data. It is the responsibility of the Nominated Officer to ensure that the data are accurate and complete. A validation exercise is carried out on a weekly basis at the AHVLA Regional Laboratories and by DoES, and on a quarterly basis for NCP submissions.

As a result of refinements to the method of defining incidents, it may not always be possible to reproduce isolation figures in previously published reports.

3. Timeliness and punctuality
The report includes provisional data (with the exception of the flock-level data for the chicken and turkey NCPs) which are subject to change. The AHVLA *Salmonella* warehouse is updated every night.

4. Accessibility and clarity
*Salmonella* data (AHVLA) have a related metadata profile (see section B).

5. Comparability
*Salmonella* cases in animals are reported both as isolations and incidents. An incident is defined as the first and all subsequent isolations of the same serovar or serovar and phage type combination of a particular *Salmonella* from an animal, group of animals or their
environment on a holding within a defined time period, which is usually 30 days. An incident report is a herd/flock (which is the epidemiological group of interest) level outcome.

Changes in the number of *Salmonella* isolations from poultry and pigs over time may reflect changes in the monitoring activity conducted by the livestock industry and not necessarily changes in incidence in *Salmonella* infection. The number of tests carried out by authorised laboratories is collated by Defra.

Sampling error, coverage error and measurement error is minimised for submissions from NCP samples as they follow a robust, harmonised protocol and test method.

Chicken and turkey data are not directly comparable before and after implementation of the NCPs. Therefore, data are only presented from 2009 onwards for chickens and from 2010 onwards for turkeys, when the respective NCPs were in place. Comparisons are more valid for years in which the NCPs have run for a full year previously. For example, before 2010 the turkey NCP was not in operation so all turkey submissions were voluntary whereas from the beginning of 2010, most turkey submissions were from statutory monitoring.

The data on positive findings of *Salmonella* in laying, breeding and broiler chicken flocks, and in turkey flocks is reported as the number of positive flocks, as required by the legislation, as well as the number of positive isolations detected during the year. The number of reported isolations of *Salmonella* detected in chickens and turkeys does not equate directly to the overall number of positive flocks that are detected during the year. A flock is counted as positive only once, irrespective of the number of isolations occurring and the number of serovars identified.

**Hatchery isolations not associated with a specific flock.**
Starting with samples collected from 1st January 2006, any hatchery isolates where there are no supply flock details available are treated as isolations only and not incidents as they cannot be traced back to a specific flock.

**SRUC and other isolations/reports without cultures submitted.**
Submissions received from the Scotland's Rural College (SRUC), and any submissions received without a sample are allocated an incident reference whereas previously these were not allocated such references.
These reports appear in the quarterly reports. This improvement was put in place for all reports on the database in 2008.

Not all isolates of *S*. Typhimurium from bovine animals received from SRUC are phage typed. As the system does not allocate an incident reference number to a report of *S*. Typhimurium until the phage type result is received, this means that some isolates of *S*. Typhimurium from SRUC will not be allocated an incident reference and therefore the actual number of incidents of *S*. Typhimurium may be higher than the number recorded on the database.

AHVLA Quality Assurance Statement
The policy of the Animal Health and Veterinary Laboratories Agency (AHVLA) is to maintain a high standard of quality in all aspects of its operation and to continually satisfy our customers in respect of all the services offered.

The laboratory facilities are UKAS accredited to BS EN ISO 17025:2000 (Lab Nos. 0941, 1769 and 2112) for an extensive range of tests supported by proficiency testing accredited to ISO/IEC Guide 43-1 1997 (Lab No. 0004). AHVLA is certificated to BS EN ISO 9001:2000 for ‘the provision of a range of specialist veterinary scientific services to the Government and other interested parties worldwide’ (Certificate Nos. LRQ 4000436, 4001071, 0962413 and 4001392).

Additionally, AHVLA holds Good Laboratory Practice and Good Manufacturing Practice approval and complies with the Joint Code of Practice for Research projects and Good Clinical Veterinary Practice quality standards.

AHVLA Weybridge is accredited to BS EN ISO 14001:2004 for environmental management system.
## SECTION B

### METADATA ELEMENTS

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### ADDITIONAL REPORT METADATA ELEMENTS

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**Acknowledgements**

Isolations were reported by Nominated Officers of the Animal Health and Veterinary Laboratories Agency for England and Wales and Divisional Veterinary Managers for Scotland and, through them, by private laboratories.

Regional Veterinary Leads/Divisional Veterinary Managers of the Animal Health and Veterinary Laboratories Agency are responsible for the collection of samples of processed animal protein.

Staff of the Animal Health and Veterinary Laboratories Agency processed the data.

The following reference laboratories made or confirmed the majority of isolations:

- PHE Laboratory of Enteric Pathogens, Colindale.
- Scottish *Salmonella* Reference Laboratory, Glasgow.

This report was compiled by:

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AHVLA is an Executive Agency of the Department for Environment, Food and Rural Affairs.