

Chapter 5.2 Antimicrobial Resistance (AMR) Monitoring Programme

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1. Overview

1.1 Introduction

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1.3 FSA role

1.1 Introduction

1.1.1 Survey overview

The Veterinary Medicines Directorate (VMD) oversees the monitoring programme of antimicrobial resistance (AMR) in *Campylobacter jejuni* and commensal *Escherichia coli* isolated from broilers, fattening turkeys and fattening pigs at slaughter.

VMD also oversees the monitoring programme of AMR in *Salmonella* isolates from fattening pigs and poultry at slaughter and poultry both at slaughter. For details of this process please refer to Chapter 4.3 Verifying Operator's Own Checks, Section 3 Verification of Microbiological Criteria.

1.1.2 Co-ordination and collection

The Antimicrobial Resistance Team at the VMD is responsible for the co-ordination of the AMR monitoring programme:

Email: m.bos@vmd.defra.gsi.gov.uk

Telephone number: 01932 338448

The contractors for the monitoring programme are APHA. APHA will be responsible for the testing of samples submitted and determining the total number of samples required from selected abattoirs. APHA will send sampling schedules and kits to participating abattoirs.

FSA OV's will undertake the collection of samples from approved abattoirs participating in the monitoring programme.

The survey requires the collection of samples from a number of different slaughtered batches / producers for each species.

Samples will be collected on the following annual basis:

Species	Year
Broiler/Turkeys	2014
Pigs	2015
Broiler/Turkeys	2016
Pigs	2017
Broiler/Turkeys	2018
Pigs	2019
Broiler/Turkeys	2020

1.2 Legislation

1.2.1 Applicable legislation

The sampling programme is designed to meet EU requirements on the monitoring and reporting of antimicrobial resistance in zoonotic and commensal bacteria as outlined in Commission Implementing Decision 2013/652/EU.

The Decision requires the monitoring and reporting of AMR in zoonotic and commensal indicator organisms, isolated from pig, broiler and turkey caecal samples at slaughter, on a biennial basis for each species.

1.3 FSA role

1.3.1 Target population

Broilers, fattening turkeys and fattening pigs.

1.3.2 OV requirements

The OV must:

- ensure continuity of evidence when samples are collected, prepared, labelled, stored and despatched, and
- always obtain evidence for the origin of the samples collected

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- ensure the data collection forms, AMR1 (for Turkeys), AMR2 (for Pigs) and AMR3 (for Broilers) are fully completed, and three copies are taken (see annexes 1, 2 and 3 for examples of the forms)
- ensure one copy of the AMR form is sent with the samples, the second copy is given to the named FBO contact (which will be supplied by SLA and Contracts Team) and the third copy is retained by the FSA team.

1.3.3 Relevant establishments

These instructions apply to FSA staff at plants participating in the monitoring programme on AMR in *Campylobacter jejuni* in broilers and turkeys, and *E.coli* in broilers, turkeys and pigs.

1.3.4 Time coding

All work undertaken as part of this survey in the collection, storage, packaging and despatch of samples is to be coded to GVLA.

2. Sampling

- 2.1 Sampling programme
- 2.2 Sampling equipment
- 2.3 Broilers and Turkeys: collecting samples
- 2.4 Pigs: collecting samples
- 2.5 Minimising the risk of sample contamination
- 2.6 Completing the AMR form
- 2.7 Storage, packing and despatch of samples

2.1 Sampling programme

2.1.1 Sampling requests

FSA OVs in plant will receive a sampling schedule prepared by APHA, from the SLA and Contracts Team, which will list the number of batches that need to be sampled during the sampling period (the schedule will be sent in advance either monthly or quarterly, as appropriate).

The schedule will provide details on the date of sampling, the number of batches that need to be sampled on a given day and the ID of the batch to sample.

Please note that as the sampling schedule is weighted according to plant throughput, larger processing plants will sample more regularly than smaller processing plants.

Note: The ID batch number refers to:

- the sequence of slaughter batches going through the abattoir on the day of sampling (for broilers and turkeys)
- the sequence of slaughtered animals (for pigs)

For example, ID batch 2 would be the second batch of turkeys slaughtered on the given sampling day or the second pig killed on the slaughter line on the given sampling day.

2.1.2 Monitoring definitions

A 'slaughter batch' is defined as, a quantity of broilers/turkeys which have been raised on the same farm premises, in the same house, and delivered to the abattoir in the same vehicle.

Pig Kill Number is the actual pig kill number of the sampled pig at the start of the slaughter line on that particular day of sampling.

2.1.3 Exclusion criteria

Slaughter batches / loads from more than one house or from more than one farm (mixed batches) are to be excluded from the monitoring programme.

2.1.4 Selection of slaughter batches

To avoid bias, slaughter batches must be randomly selected for sampling.

When collecting **broiler or turkey** samples, beside each allocated sampling day on the schedule there are three numbers per sampling batch labelled 'ID of batch to sample', 'ID batch (1st reserve)' and 'ID batch (2nd reserve)'. These are random numbers generated using the average number of batches processed during the abattoir's working day, and represent the particular batch that must be identified and sampled.

Batches of broilers or turkeys from mixed houses, or from more than one farm, must be excluded. Therefore, if the selected batch is from a mixed house or from more than one farm, then the reserve batch should be sampled if that is not a mixed batch. The ID of the batch sampled should be marked clearly on the data collection form, AMR1 or AMR3.

Example:

Allocated sampling days	ID of batch to sample	ID batch (1 st reserve)	ID batch (2 nd reserve)
08/11/18	5	7	15
13/11/18	2	6	24
13/11/18	3	9	17
05/12/18	7	2	5

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When collecting **pig** samples, if the selected animal on the schedule is from a mixed batch / load or from more than one farm, select a pig from a single producer and sample. The ID (kill number) of the pig sampled should be marked clearly on the data collection form AMR2.

Sampling for the surveillance programme will only be carried out Monday to Friday. If broilers, turkeys or pigs are not slaughtered on the specified sampling day, please sample the same ID batch number allocated but on the next processing day.

The revised sampling date and the ID of the batch sampled should be marked clearly on the data collection forms, AMR1, AMR2 or AMR3.

Reference: See annex 1 for a sample copy of form AMR1, annex 2 for a sample copy of form AMR2, annex 3 for a sample copy of form AMR 3 and annex 4 for guidance notes on arranging the delivery to APHA laboratories.

Note: If the samples cannot be despatched on the same day as collection, or if there are any questions on the sampling schedule, contact the SLA and Contracts Team on 01904 232082.

2.1.5 Repeat sampling

When preparing to take a sample, please ensure the 'previously sampled report' from APHA is reviewed to prevent repeated sampling of the same flocks or holdings.

2.1.6 Selection process

The following table outlines the slaughter batch selection process:

Step	Description
1	If the batch identified for sampling is not eligible (it is not from a single house or cannot be sampled) sample the 1st reserve batch for broilers and turkeys or select a suitable animal to sample from for pigs.
2	If the 1st reserve batch is not eligible or cannot be sampled, sample the 2nd reserve batch (Note: for broiler or turkey sampling only).
3	If the 2nd reserve batch is not eligible or cannot be sampled, sample the next available eligible batch on the same processing day (Note: for broilers and turkeys only).
4	Mark the sampled ID batch number on the AMR1, AMR2 or AMR3 forms
5	If there are no more eligible batches processed on the same day, sample the first available eligible batch on the next processing day (and mark the date and batch number clearly on the AMR1, AMR2 or AMR3 forms).

2.2 Sampling equipment

2.2.1 Introduction

APHA will provide the relevant establishments with sampling kits and the data collection forms (AMR1, AMR2 or AMR3). The SLA and Contracts Team will contact FSA staff at the establishments to inform them of delivery arrangements for sampling kits.

2.2.2 Non-delivery of sample kits

Sampling kits and forms should be received at least four days before sampling begins. If the kit and form is not received, or if any of the equipment listed below is missing, contact the SLA and Contracts Team on 01904 232082.

2.2.3 Sampling kit contents

Broiler and Turkey kit:

- 1 x Biotherm shipping box
 - Broilers – Biotherm 3
 - Turkeys – Biotherm 5 and 7
- Sample pots
 - Broilers – 90ml screw cap sampling pot
 - Turkeys – 300 ml screw cap sampling pot
- A4 pathoseal absorbent bags for sample pots
- sterile gloves
- grip-seal bags
- AMR1 and AMR3 forms
- Biochills - these must be kept away from direct contact with the samples using the bubble wrap (for broiler samples) or the polystyrene spacers (for turkey samples)
- bubble wrap (Broilers)
- polystyrene spacer (Turkeys)
- security seal

- UN3373 label

Note: Biochill packs must be completely frozen when packed in the sampling box, ensure that they are placed in a freezer at least 48 hours before sampling.

For turkeys, the biochills are placed on top of the polystyrene separator (which comes in the biotherm 5 and 7 boxes). For broilers, the bubble wrap would act as a separator in the smaller biotherm 3 boxes. See details for packaging in section 2.7.3.

Pig kit: for sampling 1 caeca per fattening pig:

- 1 x Biotherm shipping box
- 1 x 90ml screw cap sampling pot
- disposable scalpal
- A4 pathoseal absorbent bag (with absorbent lining)
- sterile gloves
- grip-seal bags
- AMR2 form
- bubble wrap
- 4x Biochills
- security seal
- UN3373 label

Note: Only 2 of the Biochill packs must be completely frozen when packed in the sampling box, ensuring they are placed at the top of the sampling box.

The Biochills must be placed in a freezer at least 48 hours before sampling.

The remaining two Biochill packs must be chilled and placed either side of the sample within the box.

2.3 Broiler and Turkey: Collecting samples

2.3.1 Caeca samples

One pair of full and intact caeca for broilers and turkeys will be sampled at the evisceration point from one bird per slaughter batch. The pair of caeca will be put into the appropriate sampling pot.

Turkey sampling is to be carried out at the time of evisceration. Birds are to be sampled at random during the selected batch avoiding the first part of the batch.

Depending on the line speed, and facilities available in each establishment, the paired caeca taken from each bird can be separated from the eviscerated intestines either on the slaughter line, or alternatively the whole offal can be removed and carried in a tray or similar receptacle to a separate area before removing the caeca.

Note: It is important that full and intact caeca are collected.

2.3.2 Sample handling

Samples must:

- be packaged according to the instructions in this topic
- be despatched, on the same day of collection (where possible)
- arrive at APHA Weybridge within 48hrs of sampling

Reference: See topic 2.7 on 'Storage, packaging and despatch of samples' in for additional information.

Caution:

- Samples must be kept in a cool dark place until collected by TopSpeed.
- Samples must not be frozen.
- Keep box out of direct sunlight.
- Despatch Monday to Thursday only.

2.4 Pig: Collecting samples

2.4.1 Caeca samples

5g of caecal content will be sampled at the green offal inspection point from one fattening pig. The caeca will be put into a sampling pot.

Pig sampling is to be carried out at the Green Offal Inspection Point. Pigs are to be sampled from the selected kill number identified at the evisceration point. Depending on the facilities available in each establishment, the caeca taken from each pig can be separated from the eviscerated intestines either at the inspection

point on the slaughter line, or alternatively the whole offal can be removed and carried in a tray or similar receptacle to separate area before removing the caeca.

Note: It is important that 5g of caeca are collected.

2.4.2 Sample handling

Samples must:

- be packaged according to the instructions in this topic
- be despatched, on the same day of collection (where possible)
- arrive at APHA Weybridge within 48hrs of sampling

Reference: See topic 2.7 on 'Storage, packaging and despatch of samples' in part 2 for additional information.

Caution:

- Samples must be kept in a cool dark place until collected by Topspeed.
- Samples must not be frozen.
- Keep box out of direct sunlight.
- Despatch Monday to Thursday only.

2.5 Minimising the risk of sampling contamination

2.5.1 Caeca sample contamination

The main objective is to collect the caeca whilst minimising any external contamination.

One pair of full and intact caeca for broilers and turkeys will be sampled at the evisceration point from one bird per slaughter batch. This is best achieved by careful manual traction to the portion of intestine either side of the caeca so that both caeca are removed intact with a short length of intestine.

The sampler needs to verify that the caeca are intact and full. If they are not, the paired caeca should be disregarded and a new bird selected instead.

The caeca will be put into the appropriate sampling pot. Each sampling pot should then be sealed securely and placed into a small pathoseal absorbent bag (one pot per bag).

Note: Caeca from different carcasses or slaughter batches should not be placed in the same pot.

For pig samples, this is best achieved by careful piercing of the caecum so that caecal contents can be collected in the sampling pots.

Caecal content should be collected per fattening pig and put into a labelled pot. Each pot should then be sealed securely and placed into a small pathoseal absorbent bag.

Note: Caeca from different fattening pigs should not be placed in the same pot.

2.6 Completing the AMR forms

2.6.1 Details to record

The following details must be fully recorded on the AMR1, 2 and 3 forms:

- abattoir details
- sampling details including the name of the sampler and the date and time of collection
- confirmation of the type of animal slaughtered, for example, fattening turkey or fattening pig

On the AMR1 and AMR3 forms, the following details must also be fully recorded:

- producer details, for example, farm name, address, CPH number
- batch details including the number of birds in the batch slaughtered, the number of birds in the house, the shed / house number, age of the birds and the average weight of the birds

On the AMR2 form, the following details must also be fully recorded:

- producer details, for example, farm name, address, CPH number
- animal details including the slapmark number and the weight of the carcase

Note: If an error is made when recording any of the above data on the AMR form, or if anything is unclear that might need going over again, cross through the entry and enter the correct details then initial the change. Any necessary amendments must be made before the copies of the AMR form despatch with the sample.

2.7 Storage, packaging and despatch of samples

2.7.1 Chilling

Samples must be kept chilled (not frozen) from the time of sampling until delivery to APHA. Please place the closed sampling kit in a cool area and away from direct heat until the courier arrives. If a cool room is available the entire sampling kit can be stored here until despatch to APHA Weybridge.

Note: Samples must be kept cool by storing them inside the insulated shipping box containing the frozen gel packs.

2.7.2 Specimen collection and handling

Analysis can be affected by the growth of other bacteria. Therefore, care must be taken to ensure that samples are taken appropriately, chilled as described and transported to APHA Weybridge as quickly as possible.

Extreme temperatures must be avoided.

2.7.3 Packing

Packing in line with the following procedures:

Step	Action
1	Ensure that the APHA reference number at the top of the data collection form AMR1 / AMR2/ AMR3 matches the number on the sample sampling pot.
2	Remove biochills from the chiller and/or freezers. Broilers: frozen biochills to be placed either side of the sample (which is wrapped in bubble wrap to avoid direct contact within the biotherm 3 box). Turkeys: samples to be placed at the bottom of the biotherm 5 or 7 box, then place the polystyrene divider on top with the frozen biochill packs on top of the polystyrene divider. Pigs: wrap the sample in bubble wrap to avoid contact with frozen biochills. The two chilled biochill packs are placed either side of the samples (top and bottom) and then the two frozen biochill packs on the top of these, use the polystyrene divider on top of the samples if provided in the kit.
3	Slide the completed form into the plastic document pouch to protect from any leakages that may occur and place into the sampling kit.
4	The sample box must be closed securely without delay. It is important that the pack is not left open (or closed without freezer packs) for any length of time as this may damage the samples.

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Note: All frozen Biochill packs provided in the sampling kit should be used. Care must be taken not to place these in direct contact with the specimen pots.

2.7.4 Labelling cardboard outer cartons

Apply the adhesive address label provided by the carrier to the outer carton across the box flaps.

Apply the UN3373 diamond label and Biological substance category B labels to the outer carton.

Apply the security seal to the carton lid.

2.7.5 Despatching samples

Samples are to be despatched to APHA using the Topspeed next day service:

Step	Action
1	Arrange collection by Topspeed using the process at annex 4.
2	Provide Topspeed with the following information: <ul style="list-style-type: none">• number of items (boxes) in consignment• kill date and time• name of person making the booking
3	Write the barcode nos. as reference for the collection in the plant day book. Topspeed to collect as arranged.

Note: Any problems collecting samples can also be notified to the Lab by emailing the AMR Survey Team at AMRSurvey@apha.gsi.gov.uk as well as calling the SLA and Contract Team.

2.7.6 Despatch of all samples

Samples are to be sent to:

Bacteriology, Building 17
Animal Plant Health Agency
Woodham Lane
New Haw, Addlestone
Surrey
KT15 3NB

2.7.7 Despatch failure

Should despatch fail, you must contact Topspeed and make an attempt to rearrange despatch, then notify APHA Weybridge by email to advise them of the despatch failure: amrsurvey@apha.gsi.gov.uk

2.7.8 Complaints procedure

Should Topspeed fail to collect samples within the agreed timeframe, contact the SLA and Contracts Team on 01904 232082, who will escalate the failure to Topspeed headquarters.

3. Annexes

Annex 1	AMR 1
Annex 2	AMR 2
Annex 3	AMR 3
Annex 4	Sample despatch process