



Livestock & Wildlife Disease Diagnosis at APHA Guidance on sample and test selection

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Introduction

Background

This booklet is intended to assist veterinary practitioners with sample and test selection for diagnosis of common clinical presentations in livestock and wildlife. It has been compiled by Animal and Plant Health Agency (APHA) Species Expert Group members and other veterinary colleagues.

The main remit of Government-funded scanning surveillance at APHA is the detection of new and (re)emerging livestock and wildlife disease threats. The types of threat include novel pathogens, novel diseases, novel presentations of known diseases, occurrence of diseases in novel species, strains or serotypes which are new or exotic to Great Britain; marked changes in endemic disease trends; rare, newly emerging or concerning antimicrobial resistances and threats to food safety or public health. Scanning surveillance findings are reported monthly in the Veterinary Record and quarterly online: http://apha.defra.gov.uk/vet-gateway/surveillance/reports.htm.

APHA funded provision of post-mortem examination (PME) within the APHA scanning surveillance network in England and Wales includes the APHA Veterinary Investigation Centres (VIC), APHA Lasswade (poultry only) and non-APHA partner post mortem providers (SRUC VS, University of Bristol, Royal Veterinary College, University of Surrey and Wales Veterinary Science Centre, Aberystwyth). Further details are available at this link: http://apha.defra.gov.uk/postcode/pme.asp.

In order to maximise the surveillance value of this service, for which Defra provides a significant level of financial support, APHA requires every submission to be accompanied by a fully completed submission form available from this link (species-specific forms are available): http://apha.defra.gov.uk/vet-gateway/surveillance/forms.htm.

Please fully complete the submission form and send with the submitted samples or animals. Scanning or emailing the submission form when sending animals for post mortem examination is also acceptable, once there has been discussion with a Veterinary Investigation Officer (VIO), and the animals are accepted for post mortem examination. Similar information is needed by non-APHA partner post mortem providers.

All the diagnostic tests for each submission should be sent together to a single location. We recommend choosing a laboratory that performs at least one of the tests required as indicated in the following guide:

http://apha.defra.gov.uk/documents/surveillance/diag-test-samples-locations.pdf

For a list of available tests, their prices and other details, please refer to the current Disease Surveillance Price List: https://science.vla.gov.uk/Tests/Default.aspx?SiteName=DST

Veterinary staff at the APHA VICs can give advice where the details in this guidance do not provide sufficient information, or where assistance is needed to investigate complex, unusual or problematic outbreaks of disease, even if you do not send samples to APHA. Veterinary staff at APHA VICs and Lasswade should also be contacted directly to discuss submissions for post mortem examination. VIC Contact Details:

http://apha.defra.gov.uk/vet-gateway/surveillance/diagnostic/national-network.htm

Veterinary Investigation Diagnosis Analysis (VIDA)

The surveillance information you provide on the submission form is recorded in the VIDA database, together with any diagnosis reached. VIDA is a national database, providing analysis of all diagnostic submissions to APHA, SRUC Veterinary Services Disease Surveillance Centres and the network of non-APHA partner PME providers. Diagnoses follow strict criteria. VIDA allows monitoring of diagnoses, clinical syndromes and disease trends and epidemiological features associated with these. Submissions in which a diagnosis is not reached (DNR) after reasonable testing are also scrutinised to determine whether they provide any evidence of a new or re-emerging threat. The Species Expert Groups analyse and report these VIDA data.

- VIDA annual reports are available here:
 http://apha.defra.gov.uk/vet-gateway/surveillance/scanning/vida.htm
- Interactive disease surveillance dashboards are available here: http://apha.defra.gov.uk/vet-gateway/surveillance/scanning/disease-dashboards.htm

Submission of animals for post mortem examination

Submission of animals for post mortem examination (PME) remains a key part of surveillance. The Government has reduced funding over recent years, so it is very important that the types and numbers of animals submitted are representative and of suitable quality. This is why, if considering submission of animals for diagnostic PME, we encourage dialogue with your designated PME provider. At times, post mortem examination will not be the best way of investigating disease and a discussion with your PME provider will quickly clarify the best course of action.

To find out who your local PME provider is and whether the animals to be submitted are located at a premises eligible for free carcase collection, please use the link to the online postcode search tool: http://apha.defra.gov.uk/postcode/pme.asp. We will need to know the clinical history of the case with the date and estimated time of death of animal(s) (if there is mortality) to be submitted.

Selection criteria for post mortem examination

- Animals dead for more than 24 hours are likely to be of less diagnostic value and will
 usually only be accepted after careful consideration of factors likely to influence
 diagnostic value including storage temperature since death.
- Animals dead for more than 48 hours will not be accepted.
- Carcases which have been frozen may not be accepted since this limits their diagnostic value.
- In outbreaks, a maximum of three mammalian and 5-10 bird carcases of the same species may be submitted together from a single disease incident on each farm.
- Sometimes the clinical history indicates that diagnosis is best undertaken in the first instance by submission of samples rather than animals for PME.

Information in this guidance document provides general and species-specific guidelines on diagnosis of common disease presentations in livestock and wildlife and selection of the most appropriate carcase or non-carcase material for different disease conditions.

Submission of live animals for post mortem examination

Occasionally it is advisable to submit live animals for clinical and post mortem examination, for example, investigations of enteric or nervous disease. Submission of a combination of live affected and freshly dead birds is often appropriate for flock health investigations in poultry and game birds. However, submission of a live animal or bird must be discussed with veterinary staff at the relevant PME provider within the APHA network at the same time that the PME is agreed, and is not possible where an APHA-funded carcase collection service is being used as they are not able to transport live animals. The decision must take account of the current welfare in transport legislation in England, Scotland and Wales. To ensure the wellbeing of any live animals, the private veterinary surgeon should agree to its transport and ensure supervision for the journey in line with the current legislation which covers, inter alia, the transport of animals for non-commercial purposes:

- England: http://www.legislation.gov.uk/uksi/2006/3260/article/4/made
- Wales: https://gov.wales/welfare-animals-during-transport

Notifiable diseases

It is important that all those involved with livestock health and production remain vigilant for signs of any notifiable disease. The Animal Health Act 1981 requires that anyone having in their charge an animal affected or suspected of having certain diseases must notify that fact to the veterinary authorities. The diseases covered by this legal requirement are known as notifiable diseases. If you suspect the presence of notifiable disease, you must immediately call APHA:

- In England via the Defra Rural Services helpline: 03000 200 301.
- In Wales on 0300 303 8268.
- In Scotland via the local APHA field office for further contact details see <u>www.gov.uk/government/organisations/animal-and-plant-health-agency/about/access-and-opening</u>

There is information on notifiable diseases of farmed livestock available on the following links, including information on their clinical signs and pathology:

- https://www.gov.uk/government/collections/notifiable-diseases-in-animals.
- WOAH World Organisation for Animal Health

Abbreviations

- Ab Antibody
- Ag Antigen
- AGIDT Agar Gel Immunodiffusion Test
- AT Agglutination Test
- BAL Bronchoalveolar lavage
- BVD Bovine Viral Diarrhoea
- CCN Cerebrocortical Necrosis
- CEL Chicken Embryo Liver
- CIT Citrate
- CFT Complement Fixation Test
- CIE Counter Immuno Electrophoresis
- CNF Cytotoxic Necrotising Factor
- CTM Charcoal Transport Medium
- DAT Direct Agglutination Test
- DEL Duck Embryo Liver
- EDTA Ethylene Diamine Tetra-acetic Acid
- ELISA Enzyme-Linked Immunosorbent Assay
- EM Electron Microscopy
- FAT Fluorescent Antibody Test
- FAVN Fluorescent Antibody Virus Neutralisation
- FPT Four Plate Test
- HAT Haemagglutination Test
- HAIT Haemagglutination Inhibition Test
- Hb Haemoglobin
- HEP Heparin
- IBR Infectious Bovine Rhinotracheitis
- ID Identification
- IFAT Indirect Fluorescent Antibody Test
- IHC Immunohistochemistry
- IPMA Immunoperoxidase Monolayer Assay

- ISH In Situ Hybridisation
- LAT Latex Agglutination Test
- LC Large Colony Variant
- MAT Microscopic Agglutination Test
- MIC Minimum Inhibitory Concentration
- MRT Milk Ring Test
- MZN Modified Ziehl-Neelsen Stain
- NEFA Non-Esterified Fatty Acids
- NPLA Neutralising Peroxide Linked Assay
- OCD Osteochondritis dissecans
- OXF Oxalate Fluoride
- PAGE Polyacrylamide Gel Electrophoresis
- PBS Phosphate Buffered Saline
- PCR Polymerase Chain Reaction
- PCV Packed Cell Volume
- PDNS Porcine Dermatitis and Nephropathy Syndrome
- PED Porcine Epidemic Diarrhoea
- PGE Parasitic Gastro-enteritis
- PI3 Parainfluenza 3
- PME Post Mortem Examination
- PMWS Postweaning Multisystemic Wasting Syndrome
- PNP Porcine Necrotising Pneumonia
- PoA Price on Application
- PRCV Porcine Respiratory Coronavirus
- PRRS Porcine Reproductive and Respiratory Syndrome
- RBC Red Blood Cells
- RBT Rose Bengal Test
- RIA Radio Immuno Assay
- RSA Rapid Slide Agglutination
- RSV Respiratory Syncytial Virus
- SAF Scrapie Associated Fibrils
- SAT Serum Agglutination Test
- SC Small Colony Variant
- SNT Serum Neutralisation Test
- SPF Specific Pathogen Free
- TGE Transmissible Gastro-enteritis
- VMAT Vaginal Mucus Agglutination Test
- VTEC VeroToxic Escherichia coli
- VTM Virus Transport Medium
- VI Virus Isolation
- VIC Veterinary Investigation Centre/s (formerly Regional Laboratories)
- WBC White Blood Cells

Colour codes for blood tubes

Stopper Colour	Anticoagulant
Red	None (for serum samples)
Green/orange	HEP
Purple	EDTA
Grey/yellow	OXF
Blue	CIT

Test locations and where to send samples

Please refer to the test locations document to confirm where to send samples:

http://apha.defra.gov.uk/documents/surveillance/diag-test-samples-locations.pdf

Histology

Sampling

- Tissue samples should not be more than 1 cm thick
- Samples should be fully representative of the basic organ structure and include the junction between gross lesions and normal tissue
- Samples should be immersed in 10-20 times their volume of fixative as soon as possible
- Samples should be sent in an appropriately sized container with a wide opening
- Brain is best fixed whole allowing the pathologist to select appropriate sites
- Collect intestinal samples, as soon after death as possible (ideally within 30 minutes), from several sites of small and large intestine. Immersion fixation of gut tubes 1-2 cm in length is satisfactory, but avoid crushing with forceps. Gentle agitation of the sample in the fixative will help displace food material and allow fixative to enter the lumen

If the above guidelines are followed, primary fixation of most samples should take 24-48 hours – this time period will be extended if the fixative is cold (below 5°C). However, whole brains will take longer - please discuss with your VIC.

Packing and sending

Material must be properly packaged. Packaging must conform to the postal regulations for packaging of pathological material:

www.royalmail.com/sites/default/files/Guidance-Document-Infectious-Substances-171012.pdf

Urgent cases can be sent immediately if the container is filled with fixative so that primary fixation occurs in transit. If non urgent, tissue can be initially fixed for 48 hours then sent in a reduced volume of fixative. This method is particularly appropriate for brain.

The recommended fixative for most cases is 10% neutral buffered formalin.

Bacteriology

Types of samples

- Portions of fresh tissue in clean containers are suitable if they are not autolysed or contaminated, are submitted same day or by overnight post and are kept cool during transport
- Purulent material is preferable to swabs
- Faeces samples (not just swabs) are essential if tests other than basic bacteriology are required

- For anaerobic culture, fill container to brim or wrap tissues in cling film to exclude air
- Charcoal swabs are suitable for aerobic and anaerobic culture but where anaerobes are the target organism, commercial transport media are available that are aimed specifically at anaerobe preservation. Please discuss with the VIC
- Plain swabs required for fluorescent antibody test (FAT) e.g. for Streptococcus suis 2
- Plain swabs with wire or plastic stems for PCR tests (not wooden stems)

Sampling for aerobic bacteriology (request test code TC0101)

- Tissues to be sampled should be as fresh as possible
- Sear the surface of organs with a flame or heated scalpel blade prior to incision with a sterile scalpel and swab the incised surface
- In cases of serositis (pleurisy, pericarditis, arthritis etc), rub the swab on the lesioned serosal surface and avoid just dipping the swab in fluid exudate

Sampling for anaerobic bacteriology

- Follow similar guidelines to the above for aerobic bacteriology but, for anaerobic culture, request test code TC0528
- For diagnosis of clostridial enterotoxaemia, send a minimum of 1 ml of small or large intestinal contents. Do not add any preservative. Submit for *Clostridium perfringens* toxin ELISA (TC0035)
- For clostridial myositis or black disease in cattle or sheep, or Clostridium novyi infection (hepatitis) in pigs, take four impression smears from the cut surface of affected muscle or liver, air-dry, fix with Acetone/Methanol (ratio of 75.25) and send in slide box for clostridial FAT (TC0032), or submit a portion of whole lesioned tissue in a sealed airtight container.

Bacteriology

- Initial isolation of most bacterial pathogens in cultures occurs after 24 hours incubation following initiation of cultures at the laboratory
- Some exceptions are:
 - o Glaesserella parasuis minimum of 2 days
 - Salmonella spp. by enrichment minimum 2 days
 - Campylobacter spp up to 7 days
 - o Brucella spp minimum of 4 days
 - Mycobacterium avium paratuberculosis (Johnes) up to 16 weeks
 - Mycobacterium species (TB) 6 to 12 weeks
 - Avibacterium spp and other avian Pasteurellaceae minimum of 2 or 3 days
- Full identification of bacterial pathogens can take from one day to a week, and occasionally longer depending on the nature of the particular pathogen and the degree of contamination.

• Fastidious organisms exist such as *Mycoplasma*, *Brachyspira* and *Campylobacter* species and *Leptospira* serovars may require specialist techniques. Please contact your usual APHA VIC to discuss testing for these pathogens.

Antimicrobial sensitivity

- Antimicrobial sensitivity will be initiated, if requested, once the pathogen has been obtained in pure growth which may require subculture
- Disc diffusion antimicrobial sensitivity testing (TC0401) takes 24 hours in most cases
- Minimum inhibitory concentrations (MICs) can be undertaken for some pathogens for selected antimicrobials. Please contact your usual APHA VIC for more information.

Mastitis examinations (TC0544)

Misleading results are obtained if milk samples are contaminated.

Follow this procedure to avoid contamination:

- 1. Wash and dry your hands thoroughly
- 2. Wash teat to be sampled only if obviously dirty; dry immediately
- 3. Discard first two draws of milk
- 4. Clean end of teat:
 - **a.** Use small piece of cotton wool, dampen with surgical spirit (80% spirit/20% water)
 - **b.** Rub end of teat with "swab" until visibly clean
 - **c.** Repeat using a second swab, make sure swab appears clean after use. If not, repeat using another clean swab; last swab should be spotless after wiping
- **5.** To take sample:
 - a. Open sterile sample bottle Keep lid clean, never place open-side down and preferably hold it facing downwards in crook of little finger, do not allow lid to touch teat
 - **b.** Hold sample bottle at an angle to teat
 - c. Discard a further draw of milk
 - d. Collect 1-3 streams of milk to fill sample bottle at most half-full
 - e. Immediately replace lid carefully
- 6. Label sample bottle include cow's number, quarter, date, name of farm and farmer.

Mycology

- Request fungal culture (TC0101 Sabouraud's medium) on the submission form
- Fluids (e.g. foetal stomach contents) can also be examined for fungal hyphae by direct microscopy (TC0580)
- For ringworm (dermatophyte) culture (TC0080), submit hair plucks
- Dermatophyte cultures take up to four weeks.

Serology

General

Serology is used to detect whether animals have been exposed to a particular pathogen. It is used in the diagnosis of disease and for monitoring the pathogen status of a group or herd/flock.

If animals are vaccinated against the pathogen in question, serology is of debatable value as antibody produced to vaccine cannot usually be distinguished from that produced to field infection. Only a few DIVA (Differentiation of Infected from Vaccinated Animals) vaccines are available for veterinary use (e.g. gE-deleted IBR vaccine) which allow a distinction to be made.

The possibility of maternally derived antibody being detected must be borne in mind in young animals; these may persist up to eight months of age (calves). Maternal antibodies interfere with interpretation and cannot be distinguished from antibody produced in response to active infection of the animal.

Diagnostic serology

- Sample several affected animals
- Single serology
 - o presence of antibody only indicates exposure to the pathogen
 - does not indicate how recently the exposure occurred
 - o if negative, rules out involvement of some pathogens
 - presence of antibody may be useful diagnostically if the animal(s) were supposed to be free from the pathogen
- Paired serology
 - sera tested from the same animals during acute and convalescent periods
 - o detects seroconversion (seronegative to seropositive) or a significant rise in titre
 - o establishes a temporal association of seroconversion and disease
 - acute samples must be collected within the three to four days of clinical signs occurring or animals will already have seroconverted
 - o sampling interval can vary but should not be less than two weeks
 - is not usually useful in reproductive disease investigation as maternal seroconversion has usually already occurred by the time disease manifests
- Cohort serology
 - used where conventional paired serology problematic e.g. in pigs or poultry with no individual identification
 - sera collected from groups of pigs or poultry at different ages within one management system
 - assists in assessing the timing of exposure.

Monitoring serology

Healthy animals may be tested for antibody to a pathogen to establish the status of a group or herd/flock with respect to that pathogen, so long as the animals are not vaccinated (unless a DIVA vaccine is used). The aim of the monitoring needs to be clear and is usually either:

- a. to detect presence of pathogen here the detection of a single seropositive animal is sufficient
- b. to estimate prevalence of pathogen here an estimate of the proportion of animals exposed to infection is needed and this usually involves testing a larger number of animals.

In both situations, the numbers of animals tested from an epidemiological group depends on several factors including the suspected prevalence of infection, the degree of confidence needed in the results and the number of animals in the group. Epidemiological sample size calculation tables exist which assist in establishing the numbers of animals that should be sampled and tested for given group sizes, confidences and prevalence.

The number of animals which are tested is also influenced by the logistics of sampling, cost of the tests and the sensitivity and specificity of serological tests available.

Parasitology

Fresh faeces

- Submit in a wide mouthed, screw capped container sealed with insulation tape
- Submit 3g minimum for individual faecal egg count (TC0060), 40g for fluke egg examination (TC0061), and 50g for lungworm larvae examination (TC0062)
- Monitoring faecal egg counts in sheep (the composite faecal egg count, TC0668): submit 10 x 3g (minimum) as separate faecal samples from each group; these will be pooled at the laboratory
- Monitoring fluke egg counts in cattle and sheep (TC0689): submit 10 x 5g (minimum) as separate faecal samples from each group; these will be pooled at the laboratory.

Blood

For blood parasites (TC0256) submit 2ml whole blood in EDTA tube.

Skin

 For skin parasites (TC0081) send multiple deep scrapings (i.e. firm enough to draw blood) and scabs, with hair/feathers (for mange/feather mites) or plucked underlying hair (for ringworm). Send fresh undamaged specimens of ticks, lice and fleas. All samples should be submitted in screw-capped containers; please do not submit the scalpel blade.

Virology

Ruminant respiratory viruses

Polymerase Chain Reaction (PCR) tests permit rapid identification of IBR, PI3 and RSV in both affected live animals and carcases. If collecting tissue samples only, submit both fresh and formalin-fixed samples that can be subject to microbiology testing and histopathology respectively.

Animal selection

- Select recently affected animals
- · Animals with mucopurulent nasal discharge are less likely to yield virus
- Broncho-alveolar washings (BAL) and guarded intranasal brush swabs are the preferred samples
- Nasal or ocular swabs are suitable for IBR, but are unlikely to detect PI3 or RSV
- Plain swabs must be used, but do not use swabs with a wooden stem
- Samples must be submitted as soon as possible after collection, certainly no longer than the day after collection.

Carcases

- Submit intact fresh carcases, pluck or portions of lung tissue
- For the latter collect two or three blocks of lung tissue (2cm cubes) from the junction between healthy and affected tissue
- Tracheal and/or bronchial swabs may also be collected
- Tissues or swabs should be forwarded APHA within 24 hours.

Isolation of viruses from field cases is not routinely undertaken, is time-consuming and often difficult as some mammalian respiratory viruses survive poorly in transport. When virus isolation is required, it may be necessary for the samples to be submitted in virus transport medium (VTM). Consult APHA before submission.

Other mammalian viral diseases

- Sample as advised under the specific species sections
- Where swabs are submitted, avoid use of swabs with wooden stems
- For enteric viruses send intestinal contents or faeces, without VTM, rather than swabs
- For viral skin diseases send deep scrapings, fresh biopsies or aspirated fluid (if available) in screw-topped containers.

Sampling by species and disease conditions

Avian

Poultry & game birds

Please refer to the Sampling by Discipline sections on pages 7 – 12 for general sampling information.

Please feel free to discuss avian investigations with a VIO or avian pathologist prior to submission and supply a fully completed avian submission form - http://apha.defra.gov.uk/vet-gateway/surveillance/forms.htm - that includes a good clinical history with affected bird age, morbidity and mortality patterns, information on medication and the vaccination programme and the type of husbandry system.

For post mortem examination, consider submitting a batch of birds that comprises some that have recently died and live, affected birds (if they are fit to travel and their welfare is not compromised). Individual birds may be all that is available from small flocks.

Recommended batch sizes for post mortem submissions of birds are:

- Up to 10 birds if they are less than 2 weeks old
- Up to 5 birds if they are more than 2 weeks old

Acutely affected and untreated birds are ideal candidates for post mortem examination. Fresh carcases should be submitted as post mortem autolysis occurs rapidly, particularly with chicks. Carcases should not be frozen as this leads to tissue damage and renders histopathology of very limited value.

For brain histology, fix one half of a sagittal section of the head with the brain in situ and retain the other half of the brain fresh (unfixed) for other tests.

For bacteriology, specialist avian pathogens may require a minimum of 2-3 days to achieve satisfactory growth, followed by identification. Definitive identification of some pathogens (such as *Avibacterium paragallinarum*) may require additional molecular testing such as 16S rRNA sequencing.

Parasitic infections caused by motile protozoa (*Spironucleus* – formerly *Hexamita*, and *Trichomonas/Tetratrichomonas*) are particularly prevalent in game birds. It is essential that live birds are submitted for an accurate diagnosis to be made of intestinal motile protozoan infections.

Serology can often be a useful diagnostic tool on a flock basis to help establish a diagnosis, particularly in some viral infections where virus isolation is difficult. This can

include paired (cohort) serology. Information about available serology tests and packages, including test and sample types and costs is detailed under the 'Avian' section of the 'Disease Surveillance Tests' price list:

http://science.vla.gov.uk/Tests/Default.aspx?SiteName=DST

Note that for some serology tests (*Mycoplasma gallisepticum*, *M. meleagridis* and *M. synoviae* RSA) only fresh serum samples can be used; frozen or haemolysed sera are not suitable.

Provision of an accurate flock vaccination history is essential to aid interpretation of serological tests.

Avian virology

Specific information about available tests, including test and sample types and costs is detailed under the 'Avian' section of the price list on the APHA website: https://science.vla.gov.uk/Tests/Default.aspx?SiteName=DST.

Please also feel free to discuss investigations with a VIO or avian pathologist prior to submission.

Pools of tissue from each bird (brain and trachea in one pool; liver, lung, kidney and spleen in a second; intestinal tract in a separate pool) are useful for general virus isolation tests. Lymphoid tissue may also be required as a separate pool (thymus, spleen and/or bursa) depending on the investigation.

If swabbing (for both virus isolation and/or PCR testing) plastic or wire-stemmed swabs are required. Do not use wooden stemmed swabs as these contain substances that can interfere with PCR tests. PCR tests are available for IBV, aMPV and some other avian viruses.

Some viruses target specific organs that may be useful to sample, for example:

- Trachea, caecal tonsils, kidney, reproductive tract for Infectious Bronchitis virus (IBV) infections
- Spleen is required for Haemorrhagic Enteritis Virus detection in turkeys and Marble Spleen Disease in pheasants
- Bursae are useful for the diagnosis of Infectious Bursal Disease (Gumboro)
- Brain for viruses causing encephalitis
- Intestinal contents may be examined by electron microscopy (EM) when looking for enteric viruses or PAGE for rotavirus
- Skin/oropharyngeal samples for pox viruses

Birds (poultry, game birds and waterfowl)

In many cases, investigation requires the submission of birds for post mortem examination as described on page 15. The following is a brief guide to particular situations encountered in the field where the submission of samples and carcases is of diagnostic value. The list is not comprehensive; please contact a VIO or avian pathologist to discuss individual cases.

NB: Please rule out notifiable disease. If suspected, please use the contact details on page 5.

NB: Please discuss all cases of suspected or confirmed poisoning in food animals with a VIO, as voluntary measures to control contamination of the food chain may be requested. In rare circumstances statutory controls imposed under the Food & Environmental Protection Act (FEPA) may be required.

Diseases in poultry and game birds

Increased mortality

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages NB rule out notifiable disease	Consider bacterial, viral, fungal, metabolic/ toxic causes	Carcases	Post mortem examination	
All ages NB rule out notifiable disease	Systemic bacterial infections/ septicaemias	Carcases	Post mortem examination farmed poultry and game birds over two weeks; up to 5 birds (TC0001) Post mortem examination farmed poultry and game birds up to two weeks; up to 10 birds (TC0021)	For systemic infections, aseptic cultures from spleen, liver and heart valves (if abnormal) are often useful.
All ages NB rule out notifiable disease	Systemic bacterial infections/ septicaemias	Fixed tissues	Histopathology TC0008/ TC0010	For systemic infections, aseptic cultures from spleen, liver and heart valves (if abnormal) are often useful.

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages NB rule out notifiable disease	Systemic bacterial infections/ septicaemias	Fresh tissues or swabs	Primary bacterial culture (TC0101) Anaerobic bacterial culture (TC0528)	For systemic infections, aseptic cultures from spleen, liver and heart valves (if abnormal) are often useful.
All ages NB rule out notifiable disease	Systemic bacterial infections/ septicaemias	Fresh tissues or swabs	Antibiotic sensitivity test - aerobe (TC0401)	For systemic infections, aseptic cultures from spleen, liver and heart valves (if abnormal) are often useful.
All ages NB rule out notifiable disease	Metabolic toxic causes	Blood	Example: glucose	Glucose requires OxF anticoagulant. Consult lab of choice for advice on individual lab test and sample requirements.
All ages NB rule out notifiable disease	Metabolic toxic causes	Tissues	Example: lead	Lead testing requires kidney (preferably). Consult lab of choice for advice on individual lab test and sample requirements.
Pheasants	Consider coronavirus nephritis	Carcases, fixed kidney tissue	Post mortem examination Histopathology	
Pheasants	Consider coronavirus nephritis	Fresh caecal tonsil or swab	Infectious bronchitis virus/coronavirus IBV RT PCR (single swab or sample, TC0787, pooled swabs, TC0887)	Kidney tissue can also be used.
Pheasants	Consider Marble Spleen Disease	Carcases, fixed spleen	Post mortem examination Histopathology	
Pheasants	Consider Marble Spleen Disease	Fresh spleen	Virus detection HEV/MSD AGIDT (TC0910)	

Avian
Wet litter, abnormal faeces and caecal cores

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages	Consider bacterial, parasitic, viral, toxic/nutritional causes; trauma	Carcases	Post mortem examination	Material must be examined very fresh. Live or freshly dead birds should be submitted. Fixed intestine can also be of value for histopathology.
All ages	Consider bacterial, parasitic, viral, toxic/nutritional causes	Fixed intestine	Histopathology TC0008/TC0010	Please fix the material as soon as possible to reduce autolysis.
All ages	Consider bacterial, parasitic, viral, toxic/nutritional causes	Faeces, caecal contents	Bacteriology: routine -Primary culture (TC0101) Clostridium spp - Anaerobic bacterial culture (TC0528) Brachyspira (avian intestinal spirochaetosis) - Brachyspira culture (TC0331 and TC0332)	Samples for <i>Brachyspira</i> culture should be sent if possible so that anaerobic conditions are maintained (for example in a full, screw cap sample pot. The pot should be thoroughly sealed with waterproof tape). Samples should preferably be received within 24 hours of being collected.
All ages	Salmonellosis (statutory sampling)	Faeces, boot swabs or other samples as stipulated in the National Control Programme (NCP) guides	Statutory Salmonella culture -Salmonella isolation from routine CSPO submissions (TC0699S)	The current guidelines for sampling are given in the documents for the UK National Control Programme for Salmonella available on GOV.UK.
All ages	Salmonellosis (diagnostic sampling)	Faeces; internal organs (spleen liver, caecum) for pullorum disease and fowl typhoid	Salmonella culture (TC0025)	S. Pullorum and S. Gallinarum differ from other salmonellas in that they are best isolated from tissues. Serological tests are also available for S. Pullorum and Gallinarum.
All ages	Viral enteritis	Fresh intestine	Virus isolation in tissue culture (TC0819)	Please discuss with an avian pathologist at APHA Lasswade before submitting.

Category	Condition/cause	Sample type	Recommended tests	Further information
Chicks/poults	Viral enteritis	Faeces, caecal contents	Rotavirus PAGE - Rotavirus antigen (TC0582)	
Young birds	Viral enteritis	Faeces, caecal contents	EM for virus detection - Electron microscopy (TC0317)	
All ages	Haemorrhagic Enteritis Virus of turkeys	Fixed spleen Fresh spleen	Histopathology TC0008/TC0010 Virus detection - HEV/MSD AGIDT (TC0910)	
All ages	Intestinal parasitism	Faeces or caecal contents	Worm egg/coccidial oocyst count (TC0060)	
All ages	Intestinal parasitism	Intestinal contents	Worm presence; worm identification - Microscopy (TC0580), parasite identification (TC0616)	
All ages	Motile protozoan infection in game birds	Intestinal contents from freshly dead bird	Microscopy for protozoa - (TC0580)	Material must be examined very fresh. Live or freshly dead birds should be submitted. Fixed intestine can also be of value for histopathology.

Lameness/recumbency

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages	Consider viral, bacterial, deficiencies, toxic/nutritional, trauma See also under 'swollen head and upper respiratory disease' (for Mycoplasma testing) and 'nervous disease' below	Carcases, tissue (tendon, muscle, tibiotarsus, femoral head, skin); swabs of lesioned areas for bacteriology	Post mortem examination Histopathology TC0008/TC0010	
All ages	Consider viral, bacterial, deficiencies, toxic/nutritional, trauma See also under 'swollen head and upper respiratory disease' (for Mycoplasma testing) and 'nervous disease' below	Tissue (tendon, muscle, tibiotarsus, femoral head, skin); swabs of lesioned areas for bacteriology	Bacteriology Primary bacterial culture (TC0101) Antibiotic sensitivity – aerobe (TC0401)	Tissues and swabs must be taken aseptically
All ages	Consider viral, bacterial, deficiencies, toxic/nutritional, trauma See also under 'swollen head and upper respiratory disease' (for Mycoplasma testing) and 'nervous disease' below	Tissue (tendon, muscle, tibiotarsus, femoral head, skin); swabs of lesioned areas for bacteriology	Virus isolation in tissue culture (TC0819)	

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages		Tissue (tendon, muscle, tibiotarsus, femoral head, skin); swabs of lesioned areas for bacteriology	<i>Mycoplasma</i> spp - DGGE/PCR (TC0672)	Samples for <i>Mycoplasma</i> detection should preferably be sent in <i>Mycoplasma</i> transport broth.
All ages	Rickets and other skeletal disorders	Affected bones	Histopathology TC0008/TC0010	For rickets, histopathology of growth plates required (such as proximal tibia).
All ages	Lameness, swellings over joints	Clotted blood (serum)	Mycoplasma serology - M gallisepticum and M synoviae Rapid Slide Agglutination (RSA, TC0306, TC0308); RSA flock screen (PC0932); or Western Immunoblotting (TC0749)	Minimum of 10 birds for flock screen. For RSA the serum must be freshly taken and not haemolysed or frozen. The RSA is not recommended for use in game birds.

Uneveness/poor condition in poultry

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages	Consider enteric disorders (see also under wet litter), other systemic infections, nutrition	Carcases	Post mortem examination and other testing as indicated by findings and flock background	

Avian

Loss of production in layers (ie egg drop and reduced egg quality

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages	Consider systemic infections and other causes	Serum	Mycoplasma serology (as above), Infectious Bronchitis (see under respiratory disease below), Egg Drop Syndrome '76 HAIT (TC0908)	

Swollen head and upper respiratory disease

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages	Consider bacterial, viral, trauma	Carcases, fixed tissues, sinus swabs, bloods	Post mortem examination and tests as indicated below	
All ages	Infectious coryza and other bacterial causes	Sinus swabs (or choanal cleft swabs in live birds)	Bacteriology Primary culture (TC0101)	Take sinus swabs as aseptically as possible. Preferably use plain plastic or wire stemmed swabs pre-moistened in sterile distilled water, or alternatively charcoal transport swabs.
All ages	Mycoplasmosis	Conjunctival and tracheal swabs	Mycoplasma detection by DGE/PCR - TC0672	Swabs should preferably be sent in <i>Mycoplasma</i> transport broth. It is preferable to sample several birds. Costs can be reduced by inoculating pooled swabs into transport broth. The RSA is not recommended in
				game birds.
All ages	Mycoplasmosis	Clotted blood or serum	Mycoplasma serology (as above)	Swabs should preferably be sent in <i>Mycoplasma</i> transport broth. It is preferable to sample several birds. Costs can be reduced by inoculating pooled swabs into transport broth. The RSA is not recommended in game birds.

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages	Avian meta- pneumovirus (aMPV, ART, TRT)	Oropharyngeal swab	RT PCR - aMPV PCR for single swabs (TC0786), or for a pool of up to 5 swabs (TC0397)	Plastic or wire stemmed swabs must be used.
All ages	Avian meta- pneumovirus (aMPV, ART, TRT)	Clotted blood or serum	ELISA - (TC0940)	
All ages	Infectious bronchitis and IBV-like gamma coronaviruses	Oropharyngeal and cloacal swabs	RT PCR - IBV PCR for single swabs (TC0787), or for a pool of up to 10 swabs (TC0887)	Plastic or wire stemmed swabs must be used. Positive results are followed up by sequencing of S1 gene to identify strain.
All ages	Infectious bronchitis and IBV-like gamma coronaviruses	Clotted blood or serum	HAIT for a specified single strain (TC0912), or for 3 specified strains (TC0640)	Interpretation of IBV serology requires knowledge of the IBV vaccination history. Paired serology is recommended.
All ages	Respiratory cryptosporidiosis	Carcases or fresh or fixed heads	Histopathology TC0008/TC0010	C. parvum recognised in red grouse and occasionally other species.

Respiratory disease

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages	Consider bacterial, viral, fungal, parasitic and non-infectious causes. There is often a mixed aetiology. See under swollen head and upper respiratory infections	Carcases, fixed tissues, swabs, bloods	Post mortem examination, histopathology and tests as indicated below and under swollen head/upper respiratory disease as above	
All ages	Tracheitis: Infectious laryngo- tracheitis (ILT)	Fixed trachea	Histopathology TC0008/TC0010	In the chronic stages of the disease, confirmatory diagnosis of ILT may be difficult. May also cause upper respiratory tract disease.

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages	Tracheitis: Infectious laryngo- tracheitis (ILT)	Fresh trachea	Virus isolation in tissue culture (TC0819)	In the chronic stages of the disease, confirmatory diagnosis of ILT may be difficult. May also cause upper respiratory tract disease.
All ages	Tracheitis: Infectious laryngo- tracheitis (ILT)	Blood from recovered birds	Serology by SNT - ILT SNT (TC0812)	In the chronic stages of the disease, confirmatory diagnosis of ILT may be difficult. May also cause upper respiratory tract disease.
All ages	Gapeworm (syngamosis)	Fresh or fixed tissue	Gross examination, histopathology TC0008/TC0010	There may be pathology in lung tissue. Some <i>Syngamus</i> -like species in waterfowl and other species parasitise the bronchi and sometimes other sites.
All ages	Bacterial and fungal pneumonia	Carcases, fixed and fresh tissues	Histopathology TC0008/TC0010	A variety of infectious agents may be implicated including <i>E. coli</i> , <i>Ornithobacterium rhinotracheale</i> (ORT) and other <i>Pasteurella</i> –like organisms.
All ages	Bacterial and fungal pneumonia	Carcases, fixed and fresh tissues	Primary bacterial culture, fungal culture - TC0101	A variety of infectious agents may be implicated including <i>E. coli</i> , <i>Ornithobacterium rhinotracheale</i> (ORT) and other <i>Pasteurella</i> –like organisms.

Non-specific finding: immune-suppression

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages	Consider viral causes	Fixed lymphoid tissues	Histopathology TC0008/TC0010	Multiple bursas can be examined per slide.

Nervous disease

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages	Consider viral, bacterial, nutritional deficiencies / toxicities		Post mortem examination	
All ages	Differentials include bacterial encephalitis in chicks (Enterococcus species), listeriosis, Marek's disease, Avian Encephalomyelitis, crazy chick disease (Vitamin E deficiency)	Fixed brain or peripheral nerve	Histopathology TC0008/TC0010	Nerve: examination of nerves can be useful even if no gross lesions are observed. Brain: fix one half of a sagittal section of the head with the brain in situ and retain the other half of the brain fresh (unfixed) for other tests.
All ages	Bacterial encephalitis	Swab of fresh brain	Bacterial culture - Primary culture (TC0101), <i>Listeria</i> culture (TC0663)	Aseptic sampling is essential.

Neoplasia

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages	Consider viral aetiology (Marek's disease; rarely Avian Leukosis Virus or other oncogenic viruses)	Fixed tumour, liver, spleen and nerve Fresh tumour tissue	Post mortem examination Histopathology TC0008 / TC0010	Retain fresh tissues frozen in case required for molecular testing, especially if unusual tumour distribution.

Skin and feathers

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages	Consider ectoparasites (mites, lice)	Collect parasites in water for identification	Ectoparasites- microscopic examination (TC0081)	Correct identification of mite species is important in planning control measures.
All ages	Poxvirus (also sometimes see lesions in oral cavity)	Fixed lesion tissue	Histopathology TC0008/TC0010	'Dry' pox form refers to lesions on skin, 'wet' pox to lesions in oral cavity.
All ages	Poxvirus (also sometimes see lesions in oral cavity)	Fresh tissue	Electron microscopy-avian (TC0317); virus isolation for pox virus (TC0817)	'Dry' pox form refers to lesions on skin, 'wet' pox to lesions in oral cavity.

Unusual conditions

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages	Any variations of the above – cause unknown	Carcases	Post mortem examination	Please contact one of our avian pathologists at APHA Lasswade for advice: Also keep fresh frozen material at -70°C for possible future analysis.
All ages	Any variations of the above – cause unknown	Histopathology on fixed affected tissue	Histopathology TC0008/TC0010	Please contact one of our avian pathologists at APHA Lasswade for advice: Also keep fresh frozen material at -70°C for possible future analysis.

Avian
Selected diseases of waterfowl (otherwise see above)

Category	Condition/cause	Sample type	Recommended test	Further information
Water fowl	Duck Viral Enteritis, Duck Viral Hepatitis, Goose Parvovirus	Carcases	Post mortem examination	Please contact the laboratory to discuss tissues of choice.
Water fowl	Duck Viral Enteritis, Duck Viral Hepatitis, Goose Parvovirus	Fixed tissues	Histopathology TC0008/ TC0010	Please contact the laboratory to discuss tissues of choice.
Water fowl	Duck Viral Enteritis, Duck Viral Hepatitis, Goose Parvovirus	Fresh tissues	Virus isolation in duck/goose eggs (TC0820)	Please contact the laboratory to discuss tissues of choice.
Water fowl	Duck Viral Enteritis, Duck Viral Hepatitis, Goose Parvovirus	Clotted blood (serum)	DVE SNT (TC0906), DVH SNT (TC0907) GPV AGIDT (TC0302)	Please contact the laboratory to discuss tissues of choice.
Water fowl	Duck septicaemia (Riemerella anatipestifer)	Carcases	Post mortem examination	
Water fowl	Duck septicaemia (Riemerella anatipestifer)	Swabs	Bacteriology Primary culture (TC0101)	Aseptic swabs of brain tissue are particularly useful.

Cattle

Abortion and stillbirth

- Whole fetus, placenta and maternal serum are the submission of choice. There is a statutory requirement to report all bovine abortion cases:
 - o In England via the Defra Rural Services helpline: 03000 200 301
 - o In Wales on 0300 303 8268
 - o In Scotland via the local APHA field office for further contact details see

www.gov.uk/government/organisations/animal-and-plant-health-agency/about/access-and-opening

Category	Condition/ cause	Sample type	Recommended test	Further information
Adult	Most bacterial causes and mycotic abortion	Fetal stomach contents and placenta	Culture, stained smears and wet preparation (TC0101 for routine culture, TC0026 for Campylobacter, TC0580 for wet preparation for fungi)	Can be combined with PCRs for Neospora caninum and BVD virus (see below for sample requirements) under TC0015. If a full range of samples is submitted, testing will be carried out in a stepwise fashion. If there are no significant findings from step 1 (bacteriology/mycology), then the PCRs will be carried out as step 2. Tests additional to steps 1 and 2 (e.g. PCR for Leptospira sp.) can be commissioned, but will require additional samples. Please discuss with a VIO.
Adult	Neospora caninum	Fetal brain	PCR on fresh brain (TC0852)	A positive PCR result confirms Neospora infection of the fetus, but does not confirm that neosporosis was the cause of abortion. Confirmation of Neospora abortion can be achieved through histopathology on fixed tissue.
Adult	Neospora caninum	Fixed fetal brain; Fixed fetal heart	Histopathology (PC0006)	
Adult	BVD virus	Fetal spleen or thymus	PCR (TC0655)	

Category	Condition/ cause	Sample type	Recommended test	Further information
Adult	IBR/BVD/ L.hardjo /N.caninum (Maternal Serology)	Blood - clotted	Bovine abortion/stillbirth serology package A (PC0387) (L.hardjo/ N.caninum) Bovine abortion/stillbirth serology package B (PC0405) (IBR /BVD/ L.hardjo/ N.caninum)	Paired samples are of limited diagnostic value. Single samples are useful in maintaining disease surveillance and can rule out neosporosis.
Adult	lodine deficiency	Fetal thyroid	lodine assay	Consider in stillbirths and fetal death in last week of gestation.
Adult	lodine deficiency	Fixed fetal thyroid	Histopathology (PC0006)	Consider in stillbirths and fetal death in last week of gestation.

Investigation of cattle herd infertility requires a systematic approach. Laboratory investigations can be an important component of this approach. Please discuss individual herd problems and the potential for laboratory testing to inform an investigation with a VIO.

When investigating suspected cases of bovine venereal campylobacteriosis, please note that *Campylobacter* culture and identification from sheath washings and vaginal mucus samples requires a specific sampling kit and submission form. Samples for this test (TC0098) should be sent to APHA Starcross VIC, ensuring arrival within 24 hours of sampling. Please consult a VIO before submitting these samples.

Enteric disorders

Category	Condition/cause	Sample type	Recommended test	Further information
Calves 1 – 5 days	E. coli (K99 +ve), Salmonella, cryptosporidia, rotavirus, coronavirus	Faeces (5g)	Enteric package for 1 – 5 day old calves (PC0069)	Individual components may be selected.
Calves 6 - 21 days	Salmonella, cryptosporidia, rotavirus, coronavirus	Faeces (5g)	Enteric package for 6 – 21 day old calves (PC0070)	Individual components may be selected.

Category	Condition/cause	Sample type	Recommended test	Further information
Calves from 22 days	Salmonella, coccidiosis, PGE	Faeces (10g)	Enteric package for young ruminants (PC0071)	Individual components may be selected: Salmonella culture (TC0025) and worm egg/coccidial oocyst count (TC0060).
Adult	Salmonella, fasciolosis, Johne's disease	Faeces (40g) and Blood - clotted	Enteric package for adult cattle (PC0073)	Johne's disease testing is carried out by serology (ELISA). Additional tests such as PCR on faeces may be carried out for an additional charge but require additional faeces. Individual components may be selected: Salmonella culture (TC0025), fluke egg examination (TC0061) and Johne's disease antibody ELISA (TC0366).
All ages	Persistent BVD infection and Mucosal Disease	Blood – heparin or clotted	ELISA antigen (TC0772) and antibody tests (TC0390)	In calves ≤ 30 days old, consider use of BVD PCR (TC0655).
All ages	Acute BVD infection	Blood – clotted	Paired ELISA antibody test (TC0390) PCR (serum) (TC0655) on acute sample	Diagnosis of acute infection by antibody ELISA requires paired acute and convalescent sera with an interval of 3 weeks.
Adult	Winter dysentery (Coronavirus- associated diarrhoea)	Blood – paired clotted	Paired ELISA antibody test (TC0176)	Usually seen in housed adult dairy cattle. Usually characterised by high morbidity but low mortality, with spontaneous recovery in a few days.

III thrift

Category	Condition/cause	Sample type	Recommended test	Further information
Adult, pre- and post- weaned	Endoparasitism	Faeces (50g)	Worm egg count and examination for fluke eggs (PC0064)	Individual components may be selected: Worm egg count (TC0060) and fluke egg examination (TC0061). A composite worm egg count (TC0688) is available, but is usually used for monitoring rather than diagnostic purposes. A composite fluke egg examination (TC0689) is available.
All ages	Trace element deficiency	Consult testing laboratory	Copper and GSH- Px (for selenium)	Sample at least 6 animals. Individual components may be selected. Liver copper assay may also provide useful information.
All ages	Persistent BVD infection and Mucosal Disease	Blood - clotted	ELISA antigen (TC0772) and antibody tests (TC0390).	In calves ≤ 30 days old, consider use of BVD PCR (TC0655).
Adult	Adults: Johne's disease	Blood - clotted	ELISA antibody test (TC0366)	Additional tests such as PCR on faeces may be carried out for an additional charge.

Other syndromes

Category	Syndrome	Condition/cause	Sample type	Recommended test	Further information
All ages	Locomotor disorders	Nutritional myopathy	Consult testing laboratory		Consult lab of choice for advice on individual lab test and sample requirements.
Adult	Downer cows	Downer cow	Consult testing laboratory	Downer cow profile: Calcium, magnesium, phosphate, CPK, BHB and urea suggested	Always consider the other 'non-metabolic' causes of recumbency.
Adult	Mastitis	Common mastitis pathogens	Milk	Culture (TC0544)	Aseptic technique in sample collection is essential. See page 9.
Adult	Mastitis	Mycoplasma mastitis	Milk	DGGE/PCR (TC0672)	

Category	Syndrome	Condition/cause	Sample type	Recommended test	Further information
Adult	Herd level milk drop	drop in dairy herd approach. Labora approach. Please	s. Investigatio tory investiga discuss indiv	on of problem herds tions can be an imp	ous causes of herd level milk requires a systematic ortant component of this and the potential for VIO.

Respiratory disease

Category	Condition/cause	Sample type	Recommended test	Further information
Neonatal, pre- and post- weaned	Most bacterial causes of calf pneumonia	Guarded long nasotracheal swabs or BAL samples	Routine culture (TC0101)	
All ages	Mycoplasmosis	Guarded long naso tracheal swabs or BAL samples	DGGE/PCR (TC0672)	Do not use wooden stemmed swabs.
All ages	IBR	Nasopharyngeal or ocular swabs	Multiplex Respiratory virus PCR (TC0019)	Swabs must be plain, not charcoal transport medium. Avoid sampling in chronic phase. Do not use wooden stemmed swabs.
All ages	RSV and PI3	BAL samples	Multiplex Respiratory virus PCR (TC0019)	Avoid sampling in chronic phase.
All ages	IBR, RSV, P13, BVD, Mycoplasma bovis and Histophilus somni (Serology)	Blood - paired clotted	Bovine respiratory disease serology package A (PC0384) (IBR, RSV, P13, BVD) Bovine respiratory disease serology package B (PC0385) (IBR, RSV, P13, BVD, Mycoplasma bovis and Histophilus somni)	Paired acute and convalescent sera collected with an interval of 2 -3 weeks. Ensure fully filled blood tubes are submitted.

Category	Condition/cause	Sample type	Recommended test	Further information
All ages	Dictyocaulosis (lungworm)	Faeces (50g)	Baermann examination (TC0062)	Positive Baermann result indicates patent infestation. Baermann examination will be negative in the pre-patent phase. Positive antibody ELISA result indicates exposure in
All ages	Dictyocaulosis (lungworm)	Blood - clotted	ELISA antibody test (TC0507)	current/recent grazing season, but not necessarily current patent infestation. Haematology (EDTA blood) can provide useful supportive evidence of relative or absolute eosinophilia.
All ages	Malignant Catarrhal Fever	Blood - heparin	PCR for OvHV-2 (TC0747)	

Sudden death

Always consider anthrax. Any suspicion of disease call APHA:

- In England via the Defra Rural Services helpline: 03000 200 301.
- In Wales on 0300 303 8268.
- In Scotland via the local APHA field office for further details see

www.gov.uk/government/organisations/animal-and-plant-health-agency/about/access-and-opening

Category	Condition/cause	Sample type	Recommended test	Further information
Adult	Hypomagnesaemia (cows)	Eye fluid (preferably vitreous humour)	Consult lab of choice for advice on individual lab test and sample requirements	Blood samples from at least six cows in same cohort are useful to screen for blood magnesium concentrations.

Category	Condition/cause	Sample type	Recommended test	Further information
Under two years	Blackleg (Clostridium chauvoei)	Four airdried impression smears of cut surface of muscle lesion (or muscle lesion in a full sealed container to exclude air)	FAT (TC0032)	Submission of a carcase for post mortem examination is the preferred diagnostic approach.
Under two years	Blackleg (Clostridium chauvoei)	Fixed muscle lesion	Histopathology (PC0006)	
All ages	Lead poisoning	Kidney	Tissue lead (TC0246)	Please discuss all cases of suspected lead poisoning with a VIO. Please see footnote below.

Fixed = Tissue has been immersed in a suitable fixative such as 10% neutral buffered formalin.

Nervous disease

Investigation of fatal cases of nervous disease often requires examination of the whole brain. Submission of a carcase for post mortem examination is probably the preferred diagnostic approach in fatal cases.

Category	Condition/cause	Sample type	Recommended test	Further information
All ages	Hypomagnesaemia			Consult lab of choice for advice on individual lab test and sample requirements.
All ages	Nervous acetonaemia			Consult lab of choice for advice on individual lab test and sample requirements.
All ages	Lead poisoning	Kidney (from carcase)		Please discuss all cases of suspected lead poisoning with a VIO. Please see footnote below.
All ages	Skin disease	Skin scrape/scab material	Microscopic examination (TC0081)	Histological examination of a fixed skin punch biopsy can be a useful diagnostic approach in more complex skin disease cases

NB: Please discuss all cases of suspected or confirmed poisoning in food animals with a VIO, as voluntary measures to control contamination of the food chain may be requested. In rare circumstances statutory controls imposed under the Food & Environmental Protection Act (FEPA) may be required.

Small ruminants (sheep, goats)

Abortion and stillbirth

We would like to encourage vets to take samples from aborted fetuses in the first instance of an abortion. However if abortions continue submission of a fetus plus placenta with maternal serum is recommended. If there is maternal illness, then the submission of a ewe/ewes may be advisable. VIOs at all investigation centres are happy to offer advice on cases and sample collection.

A summary of the samples most suitable for undertaking a complete abortion investigation is shown below. The tests are added sequentially, first looking for EAE, then undertaking bacteriology and toxoplasma testing.

Sample	Test	Cause of abortion tested for
Placenta including multiple cotyledons and	Gross examination for placentitis and stained smear	Chlamydia (EAE), Coxiella (Q'fever) and Brucella
intercotyledonary membrane	PCR	Toxoplasma gondii
Foetal Stomach Contents collected aseptically via a syringe and needle or with a Vacutainer	Bacteriology	Bacterial causes including Campylobacter species and Salmonella
Foetal Fluid from thoracic or abdominal cavity	Antibody iFAT	Toxoplasma gondii Cannot be used for goats
Spleen (Fresh)	PCR	Border disease
Liver (Fresh)	Additional Bacteriology	Bacterial causes including Campylobacter species
Brain (Fresh)	PCR	SBV
Maternal blood	Ab Elisa	Chlamydia abortus
Serum (red top)	LAT	Toxoplasma gondii
	Ab Elisa PCR	Border disease

Abortion and stillbirth continued: Primary tests and test codes

Category	Condition/cause	Sample type	Recommended tests	Further information
Adult	Enzootic abortion (Chlamydophila abortus)	Placenta	Gross examination, stained smears (TC0033)	A positive result on a stained smear confirms the diagnosis. Positive maternal serology indicates exposure.
Adult	Enzootic abortion (Chlamydophila abortus)	Maternal Blood – single	Ab ELISA (TC0801)	Maternal serology can be used to rule out exposure to EAE.
Adult	Toxoplasmosis	Placenta	Gross examination PCR (TC0858)	PCR on placenta is the test of choice, if placenta is not available carry out IFAT on fetal fluid.
Adult	Toxoplasmosis	Thoracic fluid	IFAT on fetal fluid (TC0628)	Cannot be used for goats.
Adult	Toxoplasmosis	Maternal blood – single	LAT on maternal blood (TC0383)	Negative maternal serology can be used to rule out exposure. Positive maternal serology indicates exposure.
Adult	Campylobacter and other bacterial infections	Fetal stomach contents and liver	Culture and stained smears. (TC0101 for routine culture, TC0026 for campylobacter).	

Send whole fetus and placenta if samples do not resolve a diagnosis.

Downer cases

Category	Condition/cause	Sample type	Recommended tests	Further information
Adult	Downer / recumbent ewes Hypocalcaemia, pregnancy Toxaemia, hypo- magnesaemia	blood (see *) or aqueous humour (if necropsy samples)	Biochemistry at non-APHA laboratory (see *)	NB – listeriosis should also be considered - diagnosed on necropsy. *Consult lab of choice for advice on clinical chemistry lab test and sample requirements.

Enteric disorders

Category	Condition/cause	Sample type	Recommended tests	Further information
Small ruminants 1-5 days	Bacteria (<i>E.coli</i> , <i>Salmonella</i>), rotavirus, Cryptosporidia	Faeces	Enteric package 1-5 day old small ruminants including goats (PC0059): 5g faeces required	Individual components; Bacterial culture with identification of isolates (TC101) and sentest as appropriate. Salmonella culture (TC0025). rotavirus PAGE. Cryptosporidia smear (TC0033).
Small ruminants 6-21 days	Salmonella, rotavirus, Cryptosporidia	Faeces	Enteric package 6- 21 day old small ruminants including goats (PC0066). 5g of faeces required	See above for individual components.
Small ruminants over 3 weeks	Coccidia & PGE, Salmonella	Faeces	Enteric package young ruminants (PC0071) 10g of faeces required	Individual components may be selected: Salmonella culture (TC0025), Worm egg and coccidial oocyst counts (TC0060).
Adult	Johne's disease, Salmonella, fluke and PGE	Faeces	Enteric package adult sheep and goats (PC0075): (40g faeces).	Note that fasciolosis and Johne's disease do not always present with diarrhoea and individual components of the enteric package may be selected: Johne's disease smear (TC0776). Johnes PCR (TC0693) pool of up to 5 samples (5g). Fluke eggs (TC0061). Worm egg count (TC0060), Salmonella culture (TC0025). Adult sheep usually acquire immunity to enteric parasites.
Adult	Johne's disease, Salmonella, fluke and PGE	Blood	Blood (Johne's disease): ELISA (TC0366)	Diagnosis of Johne's disease in sheep may require post mortem examination as ELISA and faecal microscopy are less useful in this species compared to in cattle.

III thrift

Category	Condition/cause	Sample type	Recommended tests	Further information
Young animals	Parasitism	Faeces	Worm egg count (TC0060)	Further information on investigation of anthelmintic resistance can be found at www.scops.org.uk
Young animals	Border disease	Blood – clotted	ELISA for antibodies (TC0292) PCR for virus (TC0755)	Often there is a history of abortions/hairy shaker lambs earlier in the year.
Young animals	Copper deficiency	Blood (see *)		Suggest sampling 5-6 animals not receiving concentrates. * Consult lab of choice for advice on test and sample requirements.
Young animals	Vitamin B ₁₂ (cobalt) deficiency	Blood (see *)		Suggest sampling 5-6 animals; animals should not be yarded for more than 6 hours prior to sampling as this may falsely elevate serum B ₁₂ levels. * Consult lab of choice for advice on test and sample requirements.
Adult	Parasitism	Faeces	Worm egg count (TC0060) Fluke egg examination (TC0061) 40g faeces Parasite disease: Worm egg count and fluke egg examination (PC0064) Fluke egg examination (herd screen) (TC0689) Composite – 10 x 5g faeces	VIOs happy to discuss cases of suspected anthelminthic resistance? Post mortem may be indicated if persistently high worm egg counts, or wasting /scour with low worm egg counts. Further information on investigation of anthelmintic resistance can be found at www.scops.org.uk
Adult	Parasitism	Serum Faeces	Ab ELISA (TC0678) *Fluke Coproantigen test	*Consult laboratory of choice for advice on sample requirements

Category	Condition/cause	Sample type	Recommended tests	Further information
Adult	Johne's disease	Faeces	Johne's disease smear (TC0776)	Diagnosis of Johne's disease in sheep may require post mortem
Adult	Johne's disease	Blood - clotted	Ab ELISA (TC0366)	examination as ELISA and faecal microscopy are less useful in this species compared to in cattle.
Adult	Copper deficiency	Blood (see *)		Suggest sampling 5-6 animals not receiving concentrates. * Consult lab of choice for advice on test and sample requirements.
Adult	Chronic illnesses such as Maedi Visna and OPA	Blood- clotted Carcase	AGIDT (TC0373) Ab ELISA (TC1529) Post mortem examination	Serology works well for Maedi Visna, but need post mortem examination for Ovine Pulmonary Adenocarcinoma (OPA).

Locomotor disorders

Category	Condition/cause	Sample type	Recommended tests	Further information
Young animals	Nutritional myopathy	Blood (see *)	GSH-PX and Vit E CK and AST	Consult lab of choice for advice on test and sample requirements.
Young animals	Arthritis due to Streptococcus dysgalactiae	Joint aspirate	Culture (TC0101)	Joint ill in lambs most commonly affects the carpal and tarsal joints.
Young animals/adults	Arthritis due to Erysipelothrix spp.	Blood – Clotted	SAT (TC0361)	
Young animals/adults	Arthritis due to Erysipelothrix spp.	Joint fluid aspirate	Culture (TC0101)	
Adult	Maedi Visna	Blood – Clotted	AGIDT (TC0373) Ab ELISA (TC1529)	Sporadic cases in affected flocks. Nervous form of disease may present as in-coordinated gait.
Suckled lambs	Chronic lead exposure	Carcase	Post mortem examination	Chronic low level exposure in lead mining areas can cause osteoporosis and brittle bones, manifesting as stiffness and pathological fractures.

Skin disease

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages	Orf	Scab from lesion	Electron microscopy (TC0082)	
All ages	Sheep scab, lice	Skin scrape/wool pluck from margin of lesion	Microscopic examination (TC0081)	Please do not use liquid paraffin.
All ages	Sheep scab, lice	*Blood - Clotted	Sheep Scab ELISA	Can be used to investigate exposure. *Consult lab of choice for advice on test and sample requirements.
All ages	Dermatophilus	Wool pluck from abnormal wool	Microscopy (TC0033) culture (TC0101)	
All ages	Bacterial	Swab	Culture TC0101	

Respiratory disorders

Category	Condition/cause	Sample type	Recommended tests	Further information
Adult	Maedi Visna	Blood – clotted	AGID test (TC0373) Ab ELISA (TC1529)	Sheep affected with maedi-visna may have histological changes in other tissues including brain. Submission of a whole carcase may be required.
Adult	Maedi Visna	Lung	Lung histopathology (PC0006)	
All ages	Pasteurellosis,	Lung	Pasteurella – culture (TC0101)	Pasteurella and Mannhemia species most commonly.
Adult	Ovine pulmonary adenocarcinoma	Lung	OPA – histopathology (PC0006)	

Nervous disorders

Scrapie is a notifiable disease and suspect cases should be reported call APHA:

- In England via the Defra Rural Services helpline: 03000 200 301.
- In Wales on 0300 303 8268.
- In Scotland via the local APHA field office for further contact details see

www.gov.uk/government/organisations/animal-and-plant-health-agency/about/access-and-opening

Category	Condition/cause	Sample type	Recommended tests	Further information
Adult	Hypocalcaemia Pregnancy toxaemia Hypo- magnesaemia			NB – listeriosis should also be considered. Consult laboratory of choice for advice on test and sample requirements.
All ages	CCN, listeriosis	Carcase	If deaths occur post mortem examination is indicated	NB – a blood test is not available to confirm disease in the live animal.
All ages	Cause uncertain	Carcase	If deaths occur post mortem examination will be more likely to yield a diagnosis - possibly at less cost	
All ages	Louping ill	Serum	HAIT (TC0391)	Note Louping ill is a zoonotic disease.
All ages	Louping ill	Carcase	If deaths occur post mortem examination is indicated.	Note Louping ill is a zoonotic disease.
All ages	Lead	Blood or tissue biochemistry		Blindness, convulsions and head pressing can be seen. Consult laboratory of choice for advice on test and sample requirements.
Pre-/post- weaned	Drunken Lamb Syndrome / Nephrosis	Blood, but preferably a carcase	If deaths occur post mortem examination will be more likely to yield a diagnosis - possibly at less cost	Two distinct age groups affected: 10 days to 4 week old and 2-4 months old. Latest studies suggest the initial clinical signs are mainly due to metabolic acidosis in the younger age group not due to kidney failure/nephrosis.

Sudden death

Always consider Anthrax call APHA:

- In England via the Defra Rural Services helpline: 03000 200 301.
- In Wales on 0300 303 8268.
- In Scotland via the local APHA field office for further contact details see

www.gov.uk/government/organisations/animal-and-plant-health-agency/about/access-and-opening

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages	Clostridial enterotoxaemia/ pulpy kidney	Small intestinal contents – fresh (do not add preservative)	Clostridum perfringens toxin test ELISA (TC0035)	ELISA on contents from affected portion of bowel - if not obviously affected send ileal contents bulked from at least 3 sites. Please remove contents from intestine prior to sending.
All ages	Pasteurellosis	Lung and liver	Culture (TC0101)	Culture of multiple organs advised as systemic distribution.
All ages	Other clostridial disease Fasciolosis / haemonchosis etc	Carcase	Post mortem examination of a fresh carcase at the VIC is more likely to yield a diagnosis and may be cheaper than submission of viscera	
All ages	Toxins and plant poisoning	Carcase and/or suspected substance/ plant	Post mortem examination of a fresh carcase at the VIC is more likely to yield a diagnosis and may be cheaper than submission of viscera	History may suggest the possibility of poisoning.

NB: Please discuss all cases of suspected or confirmed poisoning in food animals with a VIO, as voluntary measures to control contamination of the food chain may be requested. In rare circumstances statutory controls imposed under the Food & Environmental Protection Act (FEPA) may be required.

Mastitis

Category	Condition/cause	Sample type	Recommended tests	Further information
Adult	Staphylococci, pasteurellae, coliforms etc	Milk	Culture (TC0101)	Consider inadequate nutrition of the ewes as underlying cause

Pigs

Pigs

General

Please feel free to discuss diagnostic investigations with a VIO prior to submission of samples and you must speak to a VIO or vet at non-APHA partner PME provider site before submitting pigs. Please provide a fully completed porcine submission form that includes a full clinical history and information on medication and vaccination. http://apha.defra.gov.uk/vet-gateway/surveillance/forms.htm

For post mortem examination, consider submitting a batch of up to three pigs, typical of the clinical problem being investigated. It is sometimes appropriate to submit pigs which have been euthanased or which are live (if they are fit to travel and their welfare is not compromised). This should be discussed with the VIO or vet at non-APHA partner PME provider site before submission. Individual pigs or samples may be all that it is possible to submit from small herds.

Disease and other information relating to pig surveillance can be found on the Vet Gateway: http://apha.defra.gov.uk/vet-gateway/surveillance/seg/pig.htm.

Note that if plain swabs are used, plastic or wire-stemmed swabs are required. Do not use wooden stemmed swabs as these contain substances that can interfere with PCR tests.

NB: Please discuss all cases of suspected or confirmed poisoning in food animals with a VIO, as voluntary measures to control contamination of the food chain may be requested. In rare circumstances statutory controls imposed under the Food & Environmental Protection Act (FEPA) may be required.

Reproductive disease: with fetopathy e.g. abortion, stillbirth, weak piglets at birth

Category	Condition/cause	Sample type	Recommended tests	Further information
Adult	Suspected infectious cause	All aborted and stillborn piglets in litter (and euthanased weak where appropriate) including placentas	Abortion/stillbirth investigation (TC0011 one sow, TC0012 two sows)	Diagnostic post-mortem examination and stepwise diagnostic testing at discretion of VIO to include PRRS, leptospirosis and bacterial/fungal causes, and heart histopathology from the outset where the material submitted is suitable

Pigs

Category	Condition/cause	Sample type	Recommended tests	Further information	
				Fetal tissue preferable but sometimes virus only detected in serum from aborting sow	
Adult	Porcine Reproductive and Respiratory Syndrome (PRRS)	Fetal thymus, spleen or lung	PRRSV PCR (TC0718)	Detection of PRRS in abortions in vaccinated sows may need multiple samples and sows should be sampled at the time of abortion	
	(FIXIXO)			PRRSV serology useful in sows if not vaccinated but only diagnostic if paired	
Adult	Porcine Reproductive and Respiratory Syndrome (PRRS)	Maternal serum	PRRSV PCR (TC0718)	Note that pooled PRRSV PCR (TC0918) is validated for growing pigs, not adult pigs	
Adult	Porcine parvovirus PPV	Fetal heart	PPV PCR only available as part of TC0011 or TC0012 stillbirth investigations	Hearts of mummified fetuses can also be tested by PCR. Fetal serology by HAIT only useful if fetuses at ≥ 70 days gestation	
Adult	Porcine parvovirus PPV	Fetal fluid (e.g. pleural)	PPV HAIT TC0375		
Adult	Leptospirosis	Fetal kidney	Pathogenic Leptospira PCR (TC0856)	Zoonotic. Autolysis interferes with the PCR test	

Pigs

Category	Condition/cause	Sample type	Recommended tests	Further information
Adult	Leptospirosis	Maternal sera as follow-up to positive PCR	Leptospira MAT 6 pools (see below), 19 serovars (TC0399) Pool 1: Canicola, Copenhageni, Ballum, Icterohaemorrhagiae Pool 2: Pomona, Mozdok, Tarassovi, Grippotyphosa Pool 3: Australis, Bratislava, Autumnalis Pool 4: Hebdomadis, Mini, Sejroe Pool 5: Javanica, Bataviae, Zanoni Pool 6: Hardjo prajitno, Hardjo	Positive PCR results should be followed up with serology in recovered pigs or in contact pigs in the affected cohort to identify the infecting serovar Histopathology (liver, kidney) can provide supporting evidence
Adult	Swine influenza	Nasal swabs (plain) from sows	Swine influenza PCR (TC0771)	Consider if sows are showing transient pyrexia, malaise and/or
Adult	Swine influenza	Maternal paired sera	Swine influenza HAIT serology (TC0160 four strains)	respiratory signs. Pigs sampled for virus must be in the first few days of infection. Testing for virus* is free of charge (not serology)
Adult	Erysipelas (and other bacterial causes including fungi)	Fetal stomach contents (liver is second choice)	Bacterial culture including fungal (TC0101)	Collect from stomach using a plain vacutainer to limit contamination. Do not pool from different fetuses Histopathology on placenta also useful for confirming fungal placentitis
Adult	PCV2 or PCV3- associated foetopathy	Fetal heart – fresh and fixed	Histopathology (PC0006) and PCV- 2 IHC or PCV3 ISH if necessary	If myocarditis detected by histopathology, PCV-2 IHC and PCV3 ISH will be progressed

 $[\]frac{\text{*http://apha.defra.gov.uk/documents/surveillance/diseases/Swine\%20influenza\%20surveillance\%20vets}{\%20Dec2020.pdf}$

Pigs
Reproductive disease: infertility (no fetopathy)

Category	Condition/cause	Sample type	Recommended tests	Further information
Adult	Common infectious causes	Clotted sow bloods	Serology PRRSV ELISA (TC0412) PPV ELISA (TC0669) Erysipelas SAT (TC0361) Swine influenza HAIT TC0160 Leptospira Bratislava (TC0451) or All 19 Lepto serovars (TC0399)	Serology is unlikely to achieve a diagnosis but can rule out involvement of a pathogen. Serology is not advised for pathogens for which sampled sows are vaccinated. Paired serology is rarely possible as, by the time infertility manifests, the sow has seroconverted, it may be achievable by bleeding cohorts of sows or gilts at service and rebleeding Many regular returns-to-service are physiological or managemental
Adult	PRRS	Clotted sow bloods	PRRSV PCR (TC0718)	Testing in weeks after vaccination may detect live vaccine virus - refer to data sheets

Enteric disease

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages	Unknown or suspected cause for differential diagnosis	Ideally affected untreated pigs early in the course of disease	Post mortem examination (TC0017 (neonatal) or TC0002, and tests at VIO discretion)	Batch of up to three pigs ideal. Live if welfare allows (assists diagnosis if pre-weaned) and only if preagreed with vet at PME site, otherwise freshly dead. Where possible, submit untreated pigs. If treatment is failing, it may be appropriate to submit treated pigs also
1 to 5-day- old pigs	Bacterial culture with <i>E. coli</i> adhesin test, rotavirus.	Faeces or intestinal contents	Neonatal enteric package (PC0067) Note: TGE/PEDV PCR currently at no charge; sensitivity testing provided if appropriate.	Request clostridial toxins at extra charge, need minimum 0.5ml Further <i>E. coli</i> typing at extra charge: <i>E. coli</i> virulence gene PCR (TC0330) or serotyping (TC0040)

Pigs

Category	Condition/cause	Sample type	Recommended tests	Further information
6-day to 6- week-old pigs	Bacterial culture with <i>E. coli</i> adhesin test, <i>Salmonella</i> culture, rotavirus.	Faeces or intestinal contents	Pre- and peri- weaned enteric package (PC0068) Note: TGE/PEDV PCR currently at no charge; sensitivity testing provided if appropriate.	Coccidial oocyst count at extra charge, need minimum 3g faeces. Further <i>E. coli</i> typing at extra charge: <i>E. coli</i> virulence gene PCR (TC0330) or serotyping (TC0040)
Pigs over 6- weeks-old to adult	Salmonella culture, Brachyspira culture and Brachyspira PCR.	Faeces or intestinal contents	Grower-Adult enteric package (PC0072) Note: TGE/PEDV PCR currently at no charge; sensitivity testing provided if appropriate.	Representative Salmonella isolates are fully identified under Zoonoses Order funding. B. hyodysenteriae isolates from pigs in Great Britain are sent for antimicrobial sensitivity testing and whole genome sequencing at no charge
Neonatal and pre- weaned	Clostridial enterotoxaemia	Intestinal contents (faeces second choice)	Clostridium perfringens toxin ELISA (TC0035)	Need 0.5ml intestinal contents per pig, do not pool. Types A (milder) and C are mainly diagnosed in pigs. Confirming a diagnosis of type A should include histopathology for supportive evidence
Neonatal and pre- weaned	Colibacillosis (Enterotoxigenic <i>E.coli</i>)	Intestinal contents or faeces (charcoal swabs second choice)	Bacterial culture (TC0101) and (TC0829) <i>E.coli</i> fimbrex (K88) test typing	Further <i>E. coli</i> typing at extra charge: <i>E. coli</i> virulence gene PCR (TC0330) or serotyping (TC0040)
Neonatal and pre- weaned	Non-ETEC <i>E.coli</i> e.g. attaching and effacing <i>E.coli</i> (AEEC), Enterohaemorrhagic <i>E.coli</i> (EHEC)	Live pigs	Post mortem examination (TC0017 (neonatal) or TC0002)	Culture results with histopathology essential for diagnosis and intestines must be fixed within minutes of death Further <i>E. coli</i> typing possible at extra charge: <i>E. coli</i> virulence gene

Pigs

Category	Condition/cause	Sample type	Recommended tests	Further information	
Neonatal and pre- weaned	Non-ETEC <i>E.coli</i> e.g. attaching and effacing <i>E.coli</i> (AEEC), Enterohaemorrhagic <i>E.coli</i> (EHEC)	Intestinal contents, faeces or charcoal swabs and fixed intestines	Bacterial culture (TC0101 and TC0829 <i>E.coli</i> fimbrex test typing and histopathology PC0006)	PCR (TC0330) or serotyping (TC0040)	
Neonatal and pre- weaned	Rotaviral enteritis	Intestinal contents or faeces	Rotavirus PAGE (TC0582)		
Neonatal and pre- weaned	Cryptosporidiosis	Intestinal contents or faeces	Cryptosporidium smear and ID (TC0492)	Rarely diagnosed in pigs, zoonotic.	
Neonatal and pre- weaned	Coccidiosis	Intestinal contents or faeces (min. 3g)	Coccidial oocyst count (TC0702). Speciation possible at extra cost (TC0648)	Absence of oocysts does not rule out a diagnosis of coccidiosis. Intestinal histopathology may be required to achieve a diagnosis and needs live affected pigs to be submitted	
Neonatal and pre- weaned	Salmonellosis	Intestinal contents or faeces (charcoal swabs second choice)	Salmonella culture (TC0025)	Rare in neonates, uncommon preweaning. Zoonotic	
Neonatal and pre- weaned	Transmissible gastro-enteritis	Intestinal contents or faeces	TGE/PED PCR (TC0398)	PCR is method of choice for diagnosis and provided at no	
Neonatal and pre- weaned	Transmissible gastro-enteritis	Paired sera	TGEv ELISA (TC0488)	charge on diagnostic submissions Last VIDA diagnosis in GB in 1999	
Neonatal and pre- weaned	Porcine epidemic diarrhoea	Intestinal contents or faeces	TGE/PEDV PCR (TC0398)	Note PED is notifiable in England and Scotland* PCR is method of choice for	
Neonatal and pre- weaned	Porcine epidemic diarrhoea	Paired sera	TGE/PED ELISA (TC0377)	diagnosis and provided at no charge on diagnostic submissions Last VIDA diagnosis in GB in 2002, virulent strain has not been detected in UK	

Pigs

Category	Condition/cause	Sample type	Recommended tests	Further information
Neonatal and pre- weaned	Hypogamma- globulinaemia (poor colostral antibody uptake)	Clotted bloods from piglets up to one-week- old	Gammaglobulin estimation (ZST) Consult lab of choice for advice on test and sample requirements	Useful to determine if poor colostral antibody transfer is predisposing to enteric disease in neonatal piglets Sample piglets less than 7-daysold. Biochemistry not available within APHA
Weaners to about six weeks old (two weeks post- weaning))	Bowel oedema (verocytotoxic <i>E.coli</i>)	Distal small intestinal contents or charcoal swab or faeces	Bacterial culture (TC0101), E. coli virulence gene PCR (TC0330) or serotyping (TC0040)	Isolation of causative <i>E. coli</i> more likely from small intestine of untreated severe cases. Brain histopathology can provide supportive evidence
Growers to adults	Brachyspira hyodysenteriae (swine dysentery)	Intestine, intestinal contents or faeces	Brachyspira culture (TC00331) Brachyspira PCR (TC0495)	Fill container with freshly voided faeces or LI contents to below brim (anaerobic organism) Culture and PCR is recommended. B. hyodysenteriae isolates from pigs in Great Britain are currently sent for antimicrobial sensitivity testing and whole genome sequencing at no charge
Growers to adults	Brachyspira pilosicoli colitis	Intestine, intestinal contents or faeces	Brachyspira culture (TC0031) Brachyspira PCR (TC0495)	Fill container to below brim (anaerobic organism) Same methods for other uncommon <i>Brachyspira</i> species e.g, <i>B. suanatina, hampsonii</i>
Growers to adults	Lawsonia intracellularis	Lesioned ileum and (on occasion) lesioned large intestine fresh and fixed	MZN smear (TC0033) Histopathology (PC0006) and silver stains	Several forms exist (necrotic ileitis, proliferative enteropathy, haemorrhagic enteropathy). MZN smears needs very fresh material and can be insensitive. PCR alone is not diagnostic
Growers to adults	Lawsonia intracellularis	Intestinal contents or faeces	Lawsonia PCR (TC0657)	j

Pigs

Category	Condition/cause	Sample type	Recommended tests	Further information
Growers to adults	Trichuris colitis or other nematodes (Hyostrongylus, Ascaris spp)	Faeces	Worm egg count (TC0060)	Egg output can be low. Hyostrongylus species associated with anaemia and ill thrift
Growers to adults	Gastric ulceration, intestinal torsion	Dead pigs	Post mortem examination (TC0017 (neonatal) or TC0002)	

 $[\]hbox{* \underline{https://www.gov.uk/guidance/porcine-epidemic-diarrhoea-how-to-spot-and-report-the-disease} \\$

Pigs
Respiratory disease

Category	Condition/cause	Sample type	Recommended tests	Further information	
All ages	Unknown or suspected cause for differential diagnosis	Ideally affected untreated pigs or plucks	Post mortem examination (TC0017 (neonatal) or TC0002, and tests at VIO discretion)	Batch of up to three pigs/plucks ideal. Where possible, submit untreated pigs. If treatment is failing, it may be appropriate to submit treated pigs also. Severe cases early in the course of disease are ideal. Porcine respiratory disease commonly involves multiple pathogens	
All ages, especially post- weaning	PRRS	Fresh lung, (spleen, serum or lymph node)	PRRSV PCR (TC0718) Pooled PRRSV PCR on up to five sera (rearing pigs only) (TC0918)	Vaccinated pigs may become viraemic when undergoing challenge and positive PCR results require further investigation. PRRS	
All ages, especially post- weaning	PRRS	Positive PCR tissue or serum	PRRSV ORF5 sequencing (TC0118)	vaccination history with timing of vaccination should be stated on submission	
All ages, especially post- weaning	PRRS	Paired sera	Paired serology PRRSV ELISA (TC0412)	Sequencing differentiates vaccine-like virus strains from others and can assist investigations Serology useful if not vaccinated	
All ages, especially post- weaning	PRRS	Fixed lung – multiple sections	Histopathology (PC0006) and PRRSV IHC (TC1402) if necessary	PRRSV IHC progressed at no charge on subsidised diagnostic submissions if APHA pathologist considers appropriate. Otherwise charged.	
All ages, especially post- weaning	PCV2-associated respiratory disease	Lymph node and fixed lung	Histopathology (PC0006 and PCV-2 IHC if necessary)	If lymphoid lesions seen with viral inclusions, PCV-2 IHC is not progressed by VIO as not necessary. Inguinal and	

Pigs

Category	Condition/cause	Sample type	Recommended tests	Further information
				mesenteric lymph nodes commonly examined.
All ages, especially post- weaning	Swine influenza	Pooled tonsil, trachea and lung (max 3 pigs) or plain nasal swabs (max 12 pigs)	Swine influenza PCR (TC0771) For nasal swabs use plastic or wire stems, not wooden	Do not pool tissues from different pigs. Pigs sampled for virus must be in the first few days of infection. Testing for virus** is free of charge (not serology)
All ages, especially post- weaning	Swine influenza	Paired sera	Swine influenza HAIT (TC0160) four strains	(not serology)
All ages, especially post- weaning	Enzootic pneumonia (<i>Mycoplasma</i> <i>hyopneumoniae</i>)	Fresh lung	Mycoplasma spp DGGE PCR (TC0672)	Sample lung from cranioventral region at
All ages, especially post- weaning	Enzootic pneumonia (<i>Mycoplasma</i> <i>hyopneumoniae</i>)	Fixed lung	Histopathology (PC0006) not diagnostic but allows interpretation of significance when <i>Mycoplasma</i> species detected by DGGE-PCR, especially in vaccinated pigs	interface between consolidated and non-consolidated lung. DGGE-PCR also detects M. hyorhinis which can cause EP-like lesions
All ages, especially post- weaning	Enzootic pneumonia (<i>Mycoplasma</i> <i>hyopneumoniae</i>)	Paired sera	Paired serology TC0546	Serology useful if not vaccinated. <i>M. hyopneumoniae</i> vaccination history should be provided
All ages, especially post- weaning	Bacterial respiratory pathogens - Pasteurella multocida, Strep suis, Bordetella bronchiseptica, Actinobacillus pleuropneumoniae, Glaesserella parasuis	Fresh lesioned lung (large portion) or charcoal swab from cut surface of lesioned lung	Bacterial culture (TC0101)	Avoid contamination when sampling, sear surface of lung before incising and swabbing if possible, or use sterile scalpel to incise

Pigs

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages, especially post- weaning	Glässer's disease (<i>Glaesserella parasuis,</i> Hps)	Charcoal swabs from up to 4 sites	Bacterial culture for Glaesserella parasuis only (TC0103)	TC0103 allows up to four lesioned sites to be cultured for just Gps under one test code charge (e.g. lung, pleura, pericardium, joint). Where possible, sample untreated freshly dead pigs
All ages, especially post- weaning	Progressive atrophic rhinitis	Charcoal swab from lesioned nasal chamber	Pasteurella multocida – toxigenic strain ELISA (TC0623)	Uncommon nowadays in GB pigs

^{**}http://apha.defra.gov.uk/documents/surveillance/diseases/Swine%20influenza%20surveillance% 20vets%20Dec2020.pdf

Systemic disease

Category	Condition/cause	Sample type	Recommended tests	Further information
All ages	Unknown or suspected cause for differential diagnosis	Affected pigs ideally untreated	Post mortem examination (TC0017 (neonatal) or TC0002, and tests at VIO discretion)	If pigs are euthanased for submission, it is useful to obtain clotted and EDTA bloods from them first
All ages	PRRS	See details under respiratory disease section – spleen/serum for PCR	See details under respiratory disease section – spleen/serum for PCR	Consider testing in upsurges of other endemic disease, gastric ulceration, non-specific malaise
Neonatal	Hypogamma- globulinaemia	Clotted bloods for serum	Gammaglobulin (ZST) Consult lab of choice for advice on test and sample requirements	Sample piglets less than 7-days-old. Biochemistry not available within APHA

Pigs

Category	Condition/cause	Sample type	Recommended tests	Further information
Mainly pre- weaned and early post- weaned	Iron deficiency anaemia	Typically affected pigs, batch of 3 ideal, euthanased or freshly dead	Post mortem examination (TC0017 (neonatal) or TC0002, and tests at VIO discretion)	Full haematology confirms anaemia supporting diagnosis Liver iron estimation if no serum
Mainly pre- weaned and early post- weaned	Iron deficiency anaemia	EDTA and clotted bloods	Full haematology and serum iron. Consult lab of choice for advice on test and sample requirements	available. Biochemistry not available within APHA
Pre and postweaned pigs	Systemic leptospirosis	Fresh kidney	Pathogenic Leptospira PCR (TC0856)	Pigs may be jaundiced. Leptospires
Pre and postweaned pigs	Systemic leptospirosis	Paired serology sera (also as follow-up to positive PCR)	Leptospira MAT 6 pools, 19 serovars (TC0399)	are zoonotic Histopathology (liver, kidney) can provide supporting evidence
Mainly post- weaned pigs from six weeks old, occasionally younger preweaned pigs or young adults	Porcine circovirus 2- associated disease including PMWS/PDNS	Typically affected pigs, batch of 3 ideal, euthanased or freshly dead	Post mortem examination (TC0002 and tests at VIO discretion)	Post mortem investigation best to identify concurrent disease and, if not PCVAD, to establish diagnosis. PCV-2 vaccination history should be provided. If lymphoid lesions seen with viral inclusions, PCV-2 IHC is not progressed by VIO as not necessary.

Pigs

Category	Condition/cause	Sample type	Recommended tests	Further information
Mainly post- weaned pigs from six weeks old, occasionally younger preweaned pigs or young adults	Porcine circovirus 2- associated disease including PMWS/PDNS	Formalin- fixed lymph nodes (inguinal, mesenteric) and other fixed tissues if lesioned	Histopathology (PC0006 and PCV-2 IHC if necessary to establish diagnosis)	PCVAD can have wider clinical presentation than illthrift, including mainly enteric or respiratory disease
Post- weaned pigs mainly from around 10-weeks- old	Porcine dermatitis and nephropathy syndrome (PDNS)	Typically affected pigs – euthanased or freshly dead	Post mortem examination (TC0002 and tests at VIO discretion)	Lesions of PDNS can resemble those of swine fever and, if in doubt, cases should be notified to APHA field
Post- weaned pigs mainly from around 10-weeks- old	Porcine dermatitis and nephropathy syndrome (PDNS)	Formalin- fixed lymph nodes (inguinal, mesenteric), skin and kidney	Histopathology (PC0006)	should be notified to APHA field services as suspect swine fever, especially if multiple cases are occurring with mortality
Any age but especially pre and post- weaned pigs	Bacterial septicaemia (e.g. erysipelas, streptococcal, pasteurellosis, Klebsiella pneumoniae and Actinobacillus suis and colisepticaemia in preweaned piglets)	Typically affected pigs – freshly dead	Post mortem examination (TC0017 or TC0002 and tests at VIO discretion)	Sudden or rapid death. Prior antimicrobial treatment may affect culture results

Pigs

Category	Condition/cause	Sample type	Recommended tests	Further information
Any age but especially pre and post- weaned pigs	Bacterial septicaemia (e.g. erysipelas, streptococcal, pasteurellosis, Klebsiella pneumoniae and Actinobacillus suis and colisepticaemia in preweaned piglets)	Fresh tissues or charcoal swabs from liver, lung, meninges	Bacterial culture (TC0101)	Sudden or rapid death. Prior antimicrobial treatment may affect culture results
Any age but especially pre and post- weaned pigs	Glaesserella parasuis (Gps) septicaemia and/or polyserositis (Glässer's disease)	Charcoal swabs from up to 4 sites	Bacterial culture for <i>Glaesserella</i> parasuis only (TC0103)	TC0103 allows up to four sites to be cultured for just Gps under one test code charge (sites could include lung, meninges, liver, pleura)
Any age but especially pre and post- weaned pigs	Polyserositis due to <i>Mycoplasma</i> <i>hyorhinis</i>	Plain swabs	Mycoplasma spp. DGGE_PCR (TC0672) — multiple swabs from same pig can be pooled (not swabs with wooden stems)	Polyserositis can include arthritis. Advisable to submit charcoal swabs for routine bacteriology and plain for <i>Mycoplasma</i> testing if no bacterial pathogen isolated
Any age	Toxicity	Affected pigs	Post mortem examination (TC0017 (neonatal) or TC0002, and tests at VIO discretion)	The two toxicities most frequently diagnosed in pigs at APHA are coal tar (liver necrosis) and bracken (cardiomyopathy and acute heart failure). Information on these are found on the Pig Expert Group page on the Vet Gateway
Any age	Neoplasia	Lesioned tissues	Histopathology (PC0006)	Individual pigs. Lymphoma most common neoplasia in pigs

Pigs **Circulatory disease**

Category	Condition/cause	Sample type	Recommended tests	Further information
Any age	Unknown or suspected cause for differential diagnosis	Typically affected pigs – live if welfare allows or freshly dead	Post mortem examination (TC0017 or TC0002 and tests at VIO discretion)	Signs often sudden death or non- specific meriting submission of pigs
Pre-weaned pigs	Thrombo- cytopaenic purpura (isoimmune disease – colostral antibodies to foetal thrombocyte antigens)	Typically affected pigs – live if welfare allows or freshly dead	Post mortem examination (TC0017 and tests at VIO discretion including haematology and bone marrow histopathology if submission allows)	Signs include weakness, anaemia, skin haemorrhages and death of non-pyrexic pigs up to about 30 days old. Gilt litters not affected. Lesions can resemble the swine fevers; a good clinical history and examination is usually sufficient to rule out concern, if in doubt, report cases to APHA field services
Pre and post-weaned pigs	Mycoplasma suis (eperythro- zoonosis)	Blood (EDTA)	Mycoplasma DGGE/PCR (TC0672) Blood parasite detection. Consult lab of choice for advice on test and sample requirements	EDTA bloods from three pigs can be tested under one TC0672 for <i>Mycoplasma suis</i> detection Full haematology detects anaemia and assists interpretation. Haematology not available within APHA. Bone marrow and other histopathology assists interpretation
Any age	Endocarditis	Valve lesions or charcoal swab	Bacterial culture (TC0101)	Erysipelas and Streptococcus suis are the two main causes
Post-		Typically affected pigs – freshly dead	Post mortem examination (TC0002) and histopathology	Usually presents as sudden death. If diagnosed, consider submitting bloods for vitamin E and selenium status from affected cohort. Consult
weaned pigs occasionally pre-weaned	Mulberry heart disease (MHD) and Hepatosis dietetica (HD)	Fixed heart and lung (for MHD) Fixed liver (for HD)	Histopathology (PC0006)	lab of choice for advice on test and sample requirements. Main differentials for MHD: acute pulmonary oedema form of PCVAD, bracken poisoning, fumonisin toxicity. Main differentials for HD: coal tar toxicity, PCV2-associated hepatitis

Pigs

Musculoskeletal disease

Category	Condition/cause	Sample type	Recommended tests	Further information
Any age	Unknown or suspected cause for differential diagnosis	Typically affected pigs –freshly dead/ euthanased	Post mortem examination (TC0017 or TC0002 and tests at VIO discretion)	Videos of affected pigs are a helpful diagnostic aid Collect bloods prior to euthanasia where possible Identify the affected limb(s) and indicate severity of lameness
Any age but mainly post-weaned	Bacterial arthritis including erysipelas, Streptococcus suis, Glaesserella parasuis	Typically affected pigs – freshly dead	Post mortem examination (TC0017 or TC0002 and tests at VIO discretion)	PME allows diagnosis of osteochondrosis (OCD), an important differential in older pigs
Any age but mainly post-weaned	Bacterial arthritis including erysipelas, Streptococcus suis, Glaesserella parasuis	Charcoal joint swabs or fluid aspirates	Bacterial culture (TC0101)	
Any age but mainly post-weaned	Bacterial arthritis including erysipelas, Streptococcus suis, Glaesserella parasuis	Paired sera	Erysipelothrix sp. SAT (TC0361)	It can be problematic isolating Erysipelothrix species from joints
Any age but mainly post-weaned	Bacterial arthritis including erysipelas, Streptococcus suis, Glaesserella parasuis	Charcoal joint swabs or fluid aspirates	Bacterial culture for Glaesserella parasuis only (TC0103)	Fibrinous polyarthritis may be part of Glässers disease. TC0103 allows up to four lesioned sites to be cultured for just Gps under one test code charge
Any age but mainly post- weaned	Infectious synovitis due to Mycoplasma spp e.g. M. hyosynoviae or hyorhinis	Plain joint swabs or fluid aspirates	Mycoplasma spp. DGGE PCR (TC0672) – multiple swabs from same pig can be pooled	M. hyorhinis usually younger postweaned pigs and may be part of polyserositis. M. hyosynoviae mainly older postweaned pigs or young breeding replacements. Histopathology on synovium provides supportive evidence

Pigs

Category	Condition/cause	Sample type	Recommended tests	Further information
Post- weaned and young breeding pigs	Osteochondrosis dissecans, femoral epiphyseolysis, fractures	Typically affected pigs – freshly euthanased	Post mortem examination (TC0002 and tests at VIO discretion)	Gross pathology alone is often diagnostic. Bone analysis and histopathology would be considered if findings suggest an osteodystrophy
Post- weaned pigs	Osteodystrophy	Typically affected pigs – euthanased	Post mortem examination (TC0002 and tests at VIO discretion)	Full PME recommended
Post- weaned pigs	Osteodystrophy	Fresh bones (mid shaft rib and long bone)	Bone analysis (not available within APHA)	Discuss with APHA VIO Fresh bone must be stripped of all soft tissue Feed analysis outside APHA
Post- weaned pigs	Osteodystrophy	Growth plates	Histopathology (PC0006)	,
Breeding sows	Osteoporosis (lactational)	Typically affected pigs – freshly euthanased	Post mortem examination (TC0002 and tests at VIO discretion)	Usually young sows with fractures periweaning
Any age	Spinal abscess/ discospondylitis	Typically affected pigs – freshly euthanased	Post mortem examination (TC0002 and tests at VIO discretion)	Histopathology would also confirm
Any age	Spinal abscess/ discospondylitis	Charcoal swabs from lesions	Bacterial culture (TC0101)	Consider anaerobic cultures also

Nervous disease

Category	Condition/cause	Sample type	Recommended tests	Further information
Any age	Unknown or suspected cause for differential diagnosis	Typically affected pigs – live if welfare allows or freshly dead	Post mortem examination (TC0017 or TC0002 and tests at VIO discretion)	Videos of typically affected pigs are a helpful diagnostic aid. Where pigs are euthanased for submission, trauma to the brain due to the method of euthanasia limits full diagnostic investigation Veterinary administration of barbiturate is ideal. Collect bloods prior to euthanasia where possible.

Pigs

Category	Condition/cause	Sample type	Recommended tests	Further information
Any age	Salt poisoning (water deprivation)	Typically affected pigs – live if welfare allows or freshly dead	Post mortem examination (TC0017 or TC0002 and brain histopathology)	Pigs are particularly sensitive to water deprivation-induced salt poisoning. Submission of heads of affected pigs would allow diagnosis of salt poisoning
Neonatal	Congenital tremor (CT)	Typically affected pigs – live if welfare allows or freshly dead Fresh brain or spinal cord (1 cm)	Post mortem examination (TC0017 and histopathology on brain and spinal cord) Atypical porcine pestivirus (APPV) PCR (discuss with an APHA VIO, no test code)	Clinical history and histopathology identify the type of CT. Classical swine fever is a cause of congenital tremor type A1 and, in CT outbreaks, the submitting vet must consider whether there are grounds to report as suspect swine fever to APHA field services APPV is a recently recognised cause of CT type A2
Pre and post-weaned mainly	Bacterial meningitis	Typically affected pigs – freshly dead	Post mortem examination (TC0017 or TC0002)	Streptococcus suis and Glaesserella
Pre and post-weaned mainly	Bacterial meningitis	Meningeal swabs (charcoal)	Bacterial culture (TC0101)	parasuis are the main bacterial causes. Cultures must include chocolate agar plates for <i>G. parasuis</i> isolation. Submission of heads of affected pigs would also allow
Pre and post-weaned mainly	Bacterial meningitis	Meningeal swabs (plain)	Streptococcus suis 2 FAT (TC0032)	diagnosis
Postweaned pigs mainly	Middle/inner ear disease	Typically affected pigs – freshly dead	Post mortem examination (TC0017 or TC0002)	
Postweaned pigs mainly	Middle/inner ear disease	Swabs from lesions (charcoal)	Bacterial culture (TC0101)	Sectioning of cranium likely to be needed to reveal lesions Can follow meningitis, or swine
Postweaned pigs mainly	Middle/inner ear disease	Swabs from lesions (plain, no wooden stems)	<i>Mycoplasma</i> spp. DGGE PCR (TC0672)	influenza

Pigs

Category	Condition/cause	Sample type	Recommended tests	Further information
Post- weaned pigs	Bowel oedema	Typically affected pigs – live if welfare allows or freshly dead	Post mortem examination (TC0017 or TC0002)	Verocytotoxigenic <i>E. coli</i> types involved. Histopathology on brain provide supportive evidence and
Post- weaned pigs	Bowel oedema	Intestinal contents or faeces	Bacterial culture (TC0101) and <i>E. coli</i> typing: virulence gene PCR (TC0330) or serotyping (TC0040)	may be indicated where clinical signs/gross lesions not typical. Isolation of causative <i>E. coli</i> more likely from untreated severe cases
Any age	Viral polioencephalo-myelitis (e.g. porcine sapelovirus)	Typically affected pigs – live if welfare allows or freshly dead	Post mortem examination (TC0017 or TC0002)	Post mortem examination best option as uncommon and need to rule out differentials. Aujeszky's disease and Teschen disease are causes of viral encephalitis and are
Any age	Viral polioencephalo-myelitis (e.g. porcine sapelovirus)	Fixed brain and spinal cord	Histopathology (PC0006 suspect lesions confirmed by IHC)	notifiable diseases. Submitting vet must consider whether there are grounds to notify severe outbreaks of nervous disease to APHA field services, especially if unresponsive to antimicrobial treatment
Any age	Viral polioencephalo- myelitis (e.g. porcine sapelovirus)	Fresh spinal cord (1 cm) and fresh brain (small portion of frontal cortex)	Sapelovirus PCR possible but not available as commercial test	Sapelovirus, astrovirus and teschovirus PCRs can be done at discretion of VIO if histopathology and other findings suggestive of involvement of neurotropic virus

Urogenital disease

Category	Condition/cause	Sample type	Recommended tests	Further information
Breeding females most likely	Cystitis and pyelonephritis	Typically affected pigs – freshly dead	Post mortem examination (TC0002)	Anaerobic culture essential to isolate Actinobaculum suis Urine samples need collection as aseptically as possible

Pigs

Breeding females most likely	Cystitis and pyelonephritis	Fresh kidney or urine	Bacterial culture aerobic (TC0101) and anaerobic (TC0528)	Histopathology allows diagnosis but not identification of causative organism
Breeding females most likely	Cystitis and pyelonephritis	Fixed kidney	Histopathology (PC0006)	

Pigs **Skin disease**

Category	Condition/cause	Sample type	Recommended tests	Further information
Any age		Affected pigs	Discuss with APHA VIC	
Any age	Many conditions, e.g. exudative epidermitis, mange, ringworm, swine pox, parakeratosis, congenital conditions	Charcoal swabs	Bacterial culture (TC0101) for Staph. hyicus	Note that vesicular lesions on feet or snouts/mouths of pigs should be reported to APHA field service without delay – more details on
Any age		Hair plucks	Ringworm culture (TC0080) – this takes several weeks	reporting here: https://www.gov.uk/government/collections/notifiable-diseases-in-animals
Any age		Skin biopsies	Electron microscopy for pig pox virus (TC0082)	It is often helpful for images of the skin condition to be sent to the APHA
Any age		Skin/ear wax scrapings	Ectoparasite examination (TC0081)	VIC

Miscellaneous and exotic farmed species

Please note the lists of differential diagnoses below concentrate on the more commonly seen conditions mainly in New World Camelids (NWC) and are not intended to be exhaustive. Other conditions may be involved.

Tests for other farmed species such as deer, bison, buffalo should use the tables for cattle or small ruminants as appropriate.

Abortion and stillbirth

*Whole fetus/stillborn, placenta and maternal clotted blood would be the submission of choice.

Category	Condition/cause	Sample type	Recommended tests	Further information
See above (*)	Bacterial causes (brucella, salmonella, campylobacter etc) and mycotic abortion	Fetal stomach contents or liver	Routine culture (TC0101) Campylobacter culture (TC0026) and wet preparation for fungi (TC0580)	Can be combined with PCRs for Neospora caninum and BVD virus (see below for sample requirements) under TC0015. If a full range of samples is submitted, testing will be carried out in a stepwise fashion. If there are no significant findings from step 1 (bacteriology/mycology), then the PCRs will be carried out as step 2. Tests additional to steps 1 and 2 (e.g. PCR for Leptospira sp.) can be commissioned, but will require additional samples and will attract an additional charge. Please discuss with a VIO
See above (*)	Chlamydia abortus, Coxiella burnetti	Fresh placenta	Stained smear exam (TC0033) PCR for <i>Chlamydia</i> (TC0417) PCR for Q fever ¹ (TC0791)	Stained smear for <i>Chlamydia</i> abortus is routinely carried out as part of TC0015
See above (*)	Chlamydia abortus, Coxiella burnetti	Formalin fixed placenta	Histology (PC0006)	Stained smear for <i>Chlamydia</i> abortus is routinely carried out as part of TC0015.
See above (*)	Neospora caninum	Formalin fixed heart and whole brain.	Histology (PC0006)	

Category	Condition/cause	Sample type	Recommended tests	Further information
See above (*)	Neospora caninum	Fresh brain stem	Neospora PCR ¹ (TC0852)	A positive PCR result confirms Neospora infection of the fetus, but does not confirm that neosporosis was the cause of abortion. Confirmation of Neospora abortion can be achieved through histopathology on fixed tissue (usually brain and heart).
See above (*)	Leptospirosis	Fresh kidney	PCR for pathogenic leptospira ¹ (TC0856)	
See above (*)	BVD	Fresh foetal spleen or thymus.	PCR for BVD antigen ¹ (TC0655)	
See above (*)	Toxoplasma gondii	Placenta	PCR ¹ (TC0858)	Rare cause of abortion. Cross reaction can occur between Toxoplasma and Neospora when using the IFAT test.
See above (*)	Toxoplasma gondii	Fixed tissues including brain	Histopathology (PC0006)	

¹Tests not validated for use in camelids

Enteric disorders

Category	Condition/cause	Sample type	Recommended tests	Further information
< 7 days of age	Bacteria (<i>E.coli,</i> salmonella), rotavirus, cryptosporidia	Faeces (5g)	Enteric package (PC0059)	Individual components – routine culture and identification of isolates (TC0101) and sentest as appropriate, salmonella (TC0025), rotavirus PAGE* (TC0582), cryptosporidia (TC0492).
7-21 days of age	Salmonella, rotavirus, cryptosporidia	Faeces (5g)	Enteric package (PC0066)	As above but no routine culture or identification of isolates. Note cryptosporidiosis has been recorded as the cause of diarrhoea and death in animals up to 8 months of age.

Category	Condition/cause	Sample type	Recommended tests	Further information
> 7days of age	Giardia	Faeces (5g)	Flotation (TC0491)	
				Camelids only. Coccidiosis is more common between 3 weeks and 3 months of age.
> 3 weeks of age	parasitic	Faeces (10g)	Worm and coccidia count (TC0860)	Nematodirosis can be a problem in young animals from 2 months of age.
				Haemonchonsis often presents as severe anaemia.
Adults	Liver fluke, coccidia, parasitic gastroenteritis	Faeces (40g + 10g)	Parasitology package (PC0864): Fluke egg count (TC0061) plus worm and coccidia count (TC0860)	Camelids only

III thrift/ weight loss

(Consider TB which is zoonotic in all age groups)

Category	Condition/ cause	Sample type	Recommended tests	Further information
Young animals	Darasitic	Faeces (10g)	Worm and coccidia count (TC0860)	Camelids only. Coccidiosis is more common between 3 weeks and 3 months of age. Nematodirosis can be a problem in young animals from 2 months of
ariiriais	gastroenteritis		Count (10000)	age.
				Haemonchonsis often presents as severe anaemia.
Any age	BVD	Heparin blood or fresh lymphoid tissue (eg.spleen, thymus,lymph node).	PCR for BVD antigen ¹ (TC0655)	
Any age	BVD	Clotted blood	SNT for BVD antibodies ¹ (TC1165)	

Category	Condition/ cause	Sample type	Recommended tests	Further information
Any age	Mycoplasma haemolamae	EDTA blood	<i>Mycoplasma</i> DGGE/PCR (TC0672)	Can cause anaemia, vague signs or no overt disease. Note the causative organism can sometimes be detected on examination of a fresh blood smear (TC0256).
Adults	Liver fluke, coccidia, parasitic gastroenteritis	Faeces (40g + 10g)	Parasitology package (PC0864) Fluke egg count (TC0061) plus worm and coccidia count (TC0860)	Camelids only
Usually > 1 year of age	Johne's disease	Faeces (5g)	Detection of acid alcohol fast bacteria (TC0776)	Weight loss is the primary sign with diarrhoea only occurring terminally.
Usually > 1 year of age	Johne's disease	Faeces (5g)	Pool of up to 5 samples for PCR for <i>Map</i> ¹ (TC0693)	Weight loss is the primary sign with diarrhoea only occurring terminally.
Usually > 1 year of age	Johne's disease	Faeces (5g)	Liquid culture (TC0713)	Can take up to 12 weeks for result.
Usually > 1 year of age	Johne's disease	Fixed ileo- caecal junction and associated lymph node.	Histology (PC0006)	Useful to differentiate from intestinal neoplasia and TB.
Usually > 1 year of age	Johne's disease	Clotted blood	ELISA for Johnes disease antibodies ¹ (TC0366)	Confirm positive ELISA results by PCR ¹ (TC0693) or culture (TC0713)
Young animals/ adults	Trace element deficiency (eg B12/cobalt, GSH-Px/ selenium, copper)			Generally rare. Consult lab of choice for advice on individual lab test and sample requirements
Deer	Lungworm	Faeces (50g)	Baermann (TC0062)	Generally presents as weight loss rather than respiratory signs. Rarely recorded in NWC in the UK.

¹Tests not validated for use in camelids

Other conditions

Category	Condition/cause	Sample type	Recommended tests	Further information
Any age	CCN/ encephalopathy	Carcase	Post mortem examination at a VIC	
Any age	Meningitis/ encephalitis eg listeriosis	Carcase	Post mortem examination at a VIC	Often bacterial cause in young crias.
Young adult/adults	Lead poisoning	Carcase	Post mortem examination at a VIC	Consult lab of choice for advice on individual lab test and sample requirements May present as a sudden death Please discuss all cases of suspected lead poisoning in food producing animals with a VIO. Please see footnote below.
Any age	Unknown cause	Carcase (plus clotted, EDTA and heparin bloods if available)	Post mortem examination at a VIC	Blood samples can be useful particularly when an assessment of hepatic or renal function is required.
Any age	Pneumonia	BAL	Culture (TC0101)	Viral infections of camelid
Any age	Pneumonia	Carcase	Post mortem examination at a VIC	respiratory system are rare. Consider TB which is zoonotic in all age groups
Any age	Skin parasites (Chorioptes, Psoroptes, Sarcoptes, Demodex, harvest mites, lice)	Superficial and deep skin scrapes and hair plucks.	Microscopic exam (TC0081)	Suggest sampling of interdigital and margins of affected areas for mites. Sarcoptic mange represents a significant zoonotic risk. Demodex – rare. Prevalence of lice in NWC in the UK is unknown – likely to be low.
Any age	Bacteria including CLA	Deep swab or tissue sample by biopsy.	Culture (TC0101)	CLA and S. aureus are rare in NWC – more common in Old World Camelids.

Category	Condition/cause	Sample type	Recommended tests	Further information
Any age	Dermatophilus	Fresh moist scale and crust with hair pluck.	Stained smear exam (TC0033) Culture (TC0101)	Rare in NWC – more common in Old World Camelids.
Any age	Dermatophytosis/ ringworm	Hair pluck.	Wet preparation (TC0580)	Rare to uncommon and tends to be self-limiting in NWC.
Any age	Dermatophytosis/ ringworm	Skin scale and scrape	Selective culture (TC0080)	Rare to uncommon and tends to be self limiting in NWC.
Any age	Orf	Fresh skin tissue.	EM (TC0082)	Uncommon to rare.
Any age	Orf	Formalin fixed tissue	Histology (PC0006)	
Young adult/adults	Neoplasia	Formalin fixed skin tissue	Histology (PC0006)	Papillomas, fibropapillomas, squamous cell carcinoma, melanocytoma and mast cell tumours.
Any age	Unknown skin disease	Formalin fixed tissue/ biopsy	Histology (PC0006)	Useful in assessing skin disease if other tests have failed to establish a diagnosis.
Sudden death Anthrax should be considered. TB can present as sudden death in camelids and should be included as a differential.	Clostridial enterotoxaemia	Small intestinal contents.	Clostridial toxin ELISA (TC0035)	
Sudden death Anthrax should be considered. TB can present as sudden death in camelids and should be included as a differential.	Clostridial enterotoxaemia	Carcase	Post mortem examination at a VIC	

Category	Condition/cause	Sample type	Recommended tests	Further information
Sudden death Anthrax should be considered. TB can present as sudden death in camelids and should be included as a differential.	Other clostridial diseases	Carcase	Post mortem examination at a VIC	
Sudden death Anthrax should be considered. TB can present as sudden death in camelids and should be included as a differential.	Toxicities, poisoning	Carcase	Post mortem examination at a VIC	Copper and lead are the most common heavy metals involved in toxicities. See also lead poisoning in nervous disease section above. Please discuss all cases of suspected toxicities/poisoning in food producing animals with a VIO.
Sudden death Anthrax should be considered. TB can present as sudden death in camelids and should be included as a differential.	(Nutritional) myopathy	Carcase	Post mortem examination at a VIC	Consult lab of choice for advice on individual lab test and sample requirements Can be associated with cardiac myonecrosis.
Sudden death Anthrax should be considered. TB can present as sudden death in camelids and should be included as a differential.	Unknown cause	Carcase	Post mortem examination at a VIC	Hepatic lipidosis and abdominal disorders such as C3 ulceration and peritonitis can present as sudden death. A fresh carcase is more likely to yield a definitive diagnosis in these cases.

Category	Condition/cause	Sample type	Recommended tests	Further information
Musculo- skeletal disease (Note recumbency can result from nervous, parasitic, digestive, urinary, cardiovascular, metabolic or septic/toxic conditions)	(Nutritional) myopathy	Carcase	Post mortem examination at a VIC	Consult lab of choice for advice on individual lab test and sample requirements

NB: Please discuss all cases of suspected or confirmed poisoning in food animals with a VIO, as voluntary measures to control contamination of the food chain may be requested. In rare circumstances statutory controls imposed under the Food & Environmental Protection Act (FEPA) may be required.

Wildlife

Unusual and mass mortality of wildlife (terrestrial vertebrate species and seals) can be examined at Veterinary Investigation Centres (VICs) under the Diseases of Wildlife Scheme (DoWS).

Carcases for post mortem examination or samples for laboratory testing can be submitted by veterinary surgeons, wildlife charities and members of the public, after consultation with your local VIC. Cases will be triaged to ensure they are appropriate for the scheme; carcases with obvious trauma e.g. road traffic accident or animals which have been in a rehabilitation facility for over 48 hours will generally be screened out. The provision of additional information including any clinical history and the map reference where the animal was found, if known, can be very useful. Those submissions accepted under the scheme will be examined free of charge and we can provide the person submitting the carcase with a written report of our post mortem examination findings and subsequent laboratory testing results. If you wish to receive these reports, please ensure you add your email address on the submission form.

It is always advisable to wear appropriate personal protective equipment when handling wildlife carcases or collecting samples.

New and emerging disease, zoonoses (in particular, mass mortalities of wild birds that may be caused by Avian Influenza Virus, West Nile Virus and Usutu Virus) and disease threats to livestock and biodiversity are the priorities in this Defra supported surveillance. When discussing cases with your local VIC, we may refer you to other projects that might be more suitable. These include:

- Avian influenza Wild Bird Survey: You should call the Defra helpline (03459 33 55 77) if you find:
 - o one or more dead bird of prey or owl
 - 3 or more dead gulls or wild waterfowl (swans, geese and ducks)
 - 5 or more dead birds of any species
- <u>UK Cetacean Strandings Investigation Programme</u> for cetaceans (porpoises, dolphins, whales) and sea turtles
- Garden Wildlife Health for hedgehogs, garden birds, amphibians and reptiles
- <u>Wildlife Incident Investigation Scheme</u> for any suspect malicious or accidental poisoning or misuse of pesticides or agrochemicals affecting wildlife
- Cases of suspected wildlife crime should be reported to the police.

The table below provides basic guidance for frequently occurring incidents involving wildlife; it is not intended to be exhaustive. Please ring to discuss cases with your local VIO for diseases not mentioned below.

Wild birds

Category	Condition/cause	Sample type	Recommended tests	Further information
Nervous disease in wild birds	Consider avian influenza virus, West Nile virus, Usutu virus, paramyxovirus, avian botulism, louping ill (grouse) and poisoning	Carcase	PME (TC0004)	Clinical history essential. Virology may take several weeks. For animals in captivity e.g. rehabilitation facilities, the suspicion of any notifiable disease should be report (refer to information on page 3)
Enteric disease	Consider oral trichomonosis, salmonellosis, Duck Viral Enteritis (DVE)	Carcase, intestinal contents, faeces, fixed tissues	PME (TC0004), microscopy (TC0580), routine bacterial cultures (specify if yersinia and/or listeria suspected) (TC0101), Salmonella cultures (TC0025), Cryptospordium (TC0492), histopathology (PC0006)	

Wild mammals

Category	Condition/cause	Sample type	Recommended tests	Further information
Nervous disease	Listeriosis, , poisoning, botulism consider rabies and avian influenza	Carcase	PME (TC0004)	For animals in captivity e.g. rehabilitation facilities, the suspicion of any notifiable disease should be report (refer to information on page 3)

Category	Condition/cause	Sample type	Recommended tests	Further information
Respiratory disease	Consider parasites, bacterial and viral infections	Carcase, tracheal/lung swabs, lung/pluck, fixed tissues	PME (TC0004), routine bacteriology (specify if fungal cultures needed) (TC0101), histopathology (PC0006)	
Enteric disease	Consider parasites, bacteria (including salmonellosis, yersiniasis and listeriosis) and viral disease	Carcase, intestinal contents, faecal sample, gralloch, fixed tissues	PME (TC0004), faecal worm egg count (TC0060), coccidial oocyst speciation (TC0408), routine bacterial cultures (specify if yersinia and/or listeria suspected) (TC0101), Salmonella cultures (TC0025), Cryptospordium (TC0492), histopathology (PC0006)	
Mortality in wild bats	EBLV, bacterial disease, parasites, trauma, cat predation, poisoning, whitenose syndrome	Carcase	PME (TC0004)	Bats submitted will be screened for EBLV by APHA prior to further testing, please allow an additional 7-14 days for laboratory results to account for this. Do not directly handle carcases unless appropriate PPE is worn and submitted in a "Bat pack" which can be obtained from the Bat Conservation Trust
Mortality in wild squirrels	Red squirrel adenovirus, squirrel pox	Carcase, skin lesions (pox virus) or intestinal contents (adenovirus), fixed tissues	PME (TC0004), electron microscopy (TC0082), histopathology (PC0006)	

Category	Condition/cause	Sample type	Recommended tests	Further information
Mortality in wild lagomorphs	Myxomatosis, Rabbit Haemorrhagic Disease (RHD), European Brown Hare Syndrome (EBHS), leporine dysautonomia	Carcase, skin lesion (Myxomatosis), liver (RHD), fixed tissues	PME (TC0004), electron microscopy (TC0082), RHDV PCR (TC0159) histopathology (PC0006)	



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