Submission of Samples

to the Veterinary Laboratories Agency

Veterinary Laboratories Agency

Working for public and animal health

VLA is an Executive Agency of the Department for **Environment, Food** & **Rural Affairs**

Issued August 2003

Contents

Submissions of Samples

- **This booklet is intended to assist with the selection and submission of samples for diagnostic testing to VLA Regional Laboratories (RLs).**
- **For a list of available tests and their prices, please refer to the current Disease Surveillance and Export Tests Price List. Most, but not all tests for export purposes are carried out at VLA Weybridge. Again, please see the booklet for details.**
- **Veterinary staff at the RL will be pleased to provide advice on sampling.**

When submitting parcels by post, please take note of postal regulations for the packaging of pathological material.

Sample testing will normally be carried out at the laboratory receiving the submission but in the interests of efficiency tests may also be carried out in other accredited laboratories at VLA.

In some circumstances accredited tests may also be subcontracted to non-VLA laboratories. On these occasions the client's permission will always be obtained first. These occasions will include the breakdown of equipment, staff problems or where there is a particularly large contract.

Non - accredited work may also be subcontracted to non-VLA laboratories.

Where it is necessary to use other laboratories, preference will be given to laboratories holding the appropriate UKAS accreditation. Where this is not possible other laboratories will be used; in such circumstances an assessment of their suitability will be made by VLA Management.

Test Methods

Test methods for which laboratories of VLA hold UKAS accreditation are listed in the Accreditation Schedule for the individual laboratory. Different methods for the same determinate may be used in different situations. Clients wishing to have full details of the methods used should contact the RL receiving the submission for further details of the schedule of accreditation and of the test methods themselves.

Reporting of Results

Where accredited tests have been carried out by a laboratory of VLA other than the receiving laboratory, these will not be indicated on the reports.

Tests subcontracted to non-VLA laboratories will be clearly indicated on reports. Non-accredited tests carried out within VLA will be clearly indicated on reports.

The UKAS accreditation certificate specifies VLA Weybridge as the accredited laboratory. The VLA Regional Laboratories and VLA Lasswade also form part of this accreditation as specified in the UKAS accredited schedule. As a result, reports from members of the Group will have the name and address of the laboratory responsible for the work and not that of the laboratory specified on the certificate.

By submitting a sample to any RL customers are accepting these arrangements.

If a notifiable disease is suspected, please contact the Divisional Veterinary Manager (DVM) before submitting samples to VLA.

Regional Laboratories

VLA Aberystwyth
Y Buarth, Aberystwyth, Ceredigion SY23 1ND
Tel: 01970 612374 Fax: 01970 612424
Email: aberystwyth@vla.defra.gsi.gov.uk

VLA Bury St Edmunds
Rougham Hill, Bury St Edmunds, Suffolk IP33 2RX
Tel: 01284 724499 Fax: 01284 724500
Email: bury-st-edmunds@vla.defra.gsi.gov.uk

VLA Carmarthen
Job's Well Road, Johnstown, Carmarthen
Carmarthenshire SA31 3EZ
Tel: 01267 235244 Fax: 01267 236549
Email: carmarthen@vla.defra.gsi.gov.uk

VLA Langford
Langford House, Langford, Bristol BS40 5DX
Tel: 01934 852421 Fax: 01934 852981
Email: langford@vla.defra.gsi.gov.uk

VLA Lasswade
International Research Centre
Pentlands Science Park, Bush Loan, Penicuik
Midlothian EH26 0PZ
Tel: 0131 445 6169 Fax: 0131 445 6166
Email: lasswade@vla.defra.gsi.gov.uk

VLA Luddington
Luddington, Stratford-upon-Avon, Warwickshire CV37 9SJ
Tel: 01789 750212 Fax: 01789 750281
Email: luddington@vla.defra.gsi.gov.uk

VLA Newcastle
Whitley Road, Longbenton, Newcastle upon Tyne NE12 9SE
Tel: 0191 266 2292 Fax: 0191 266 3605
Email: newcastle@vla.defra.gsi.gov.uk

VLA Penrith
Merrythought, Calthwaite, Penrith, Cumbria CA11 9RR
Tel: 01768 885295 Fax: 01768 885314
Email: penrith@vla.defra.gsi.gov.uk

VLA Preston
Barton Hall, Garstang Road, Barton, Preston, Lancashire PR3 5HE
Tel: 01772 861611 Fax: 01772 862026
Email: preston@vla.defra.gsi.gov.uk

VLA Shrewsbury
Kendal Road, Harlescott, Shrewsbury, Shropshire SY1 4HD
Tel: 01743 467621 Fax: 01743 441060
Email: shrewsbury@vla.defra.gsi.gov.uk

VLA Starcross
Staplake Mount, Starcross, Exeter, Devon EX6 8PE
Tel: 01626 891121 Fax: 01626 891766
Email: starcross@vla.defra.gsi.gov.uk

VLA Sutton Bonington
The Elms, College Road, Sutton Bonington, Loughborough,
Leicestershire LE12 5RB
Tel: 01509 672332 Fax: 01509 674805
Email: sutton-bonington@vla.defra.gsi.gov.uk

VLA Thirsk
West House, Station Road, Thirsk, N. Yorkshire YO7 1PZ
Tel: 01845 522065 Fax: 01845 525224
Email: thirsk@vla.defra.gsi.gov.uk

VLA Truro
Polwhele, Truro, Cornwall TR4 9AD
Tel: 01872 272150 Fax: 01872 223443
Email: truro@vla.defra.gsi.gov.uk

VLA Winchester
Itchen Abbas, Winchester, Hants SO21 1BX
Tel: 01962 779966 Fax: 01962 842492
Email: winchester@vla.defra.gsi.gov.uk

Weybridge

Enquiry points for tests done at Weybridge

	Telephone	Facsimile
General enquiry point (including Avian Chlamydophila)	01932 357335	01932 357838
Mammalian Serology	01932 357335	01932 357838
Determinative Bacteriology	01932 357251	01932 357595
Avian Virology	01932 357708	01932 357856
Primate Tests (for export)	01932 357302	01932 357232
Rabies Tests (PETS travel scheme only)	01932 357345	01932 357838
Rabies Tests (Non-PETS travel scheme)	01932 357645	01932 359406

Address samples to:

VLA Weybridge
The Sample Reception Area, Veterinary Laboratories Agency, New Haw, Addlestone, Surrey KT15 3NB
Tel: 01932 357335 Fax: 01932 357838 Email: lab.testing@vla.defra.gsi.gov.uk

Abbreviations

Ab	Antibody
Ag	Antigen
AGIDT	Agar Gel Immunodiffusion Test
ALT	Alanine Aminotransferase
Alk P	Alkaline Phosphatase
AST	Aspartate Aminotransferase
AT	Agglutination Test
BAL	Bronchoalveolar Lavage
BHB	Beta-hydroxybutyrate
BVD	Bovine Virus Diarrhoea
CCN	Cerebrocortical Necrosis
CEL	Chicken Embryo Liver
CIT	Citrate
CFT	Complement Fixation Test
CIE	Counter Immuno Electrophoresis
CPK	Creatine phospho - kinase
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
GGT	Gamma Glutamyl Transferase
GLDH	Glutamate Dehydrogenase
GSH-Px	Glutathione Peroxidase
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
IPX	Immunoperoxidase Assay
LAT	Latex Agglutination Test
LC	Large Colony Variant
LDH	Lactate Dehydrogenase
MAT	Microscopic Agglutination Test
MCH	Mean Cell Haemoglobin
MCHC	Mean Cell Haemoglobin Concentration
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
PI_3	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 Immune Assay
RL	Regional Laboratories/Laboratory
RSA	Rapid Slide Agglutination
RSV	Respiratory Syncytial Virus
SAF	Scrapie Associated Fibrils
SAT	Serum Agglutination Test
SC	Small Colony Variant
SNT	Serum Neurtralisation Test
SPF	Specific Pathogen Free
TGE	Transmissible Gastro-enteritis
VMAT	Vaginal Mucus Agglutination Test
VTEC	Vero Toxic *Escherichia coli*
VTM	Virus Transport Medium
VI	Virus isolation
WBC	White Blood Cells

Colour Code for Blood Tubes

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

Sampling by Discipline

Biochemistry

All biochemical and haematology tests are carried out at VLA Shrewsbury. Samples should be submitted, together with a VLA Biochemistry submission form, direct to the Clinical Chemistry Unit, VLA Shrewsbury, Kendal Road, Harlescott, Shrewsbury, Shropshire SY1 4HD. All results and reports will be issued by the customer's local Regional Laboratory.

Sampling

- All biochemical tests in the Disease Surveillance and Export Tests Price List can be performed on serum from clotted blood (red top vacutainer) with the exception of:
  - Copper, GSH-PX and lead require a heparinised sample (green top vacutainer).
  - Glucose and inorganic phosphate require an OXF sample (grey top vacutainer).
  - Zinc assays require the use of all plastic collection vessels.

Tissues

- For most biochemical assays 10g of liver is the tissue of choice.
- For magnesium (animals up to nine months old) and fluoride assays, a tail vertebra or rib is required. For fluoride 250 ml urine is an alternative.
- When sampling cases of suspect poisoning submit 10g liver, 10g kidney, stomach contents, fat and muscle.
  For organophosphorus poisoning also submit the brain, frozen (this test is not performed by the Clinical Chemistry Unit).

How many animals to sample?

- Trace element profiles require a minimum of six animals.
- Metabolic profiles in cows require samples from six in early lactation, six in mid-lactation and six dry cows.

Sampling tips

- Take samples from a standard site, e.g. tail vein.
- For profiles avoid sampling within seven days of a feeding change.

Volume

- One 7 ml vacutainer per animal will contain enough blood/serum for most profile packages.

Storage

- Blood should be kept cool before despatch, but clotted samples should not be refrigerated until the clot has formed. It is better to remove serum unless blood can be despatched promptly.
- Haemolysis can interfere with analysis. It is usually caused by poor handling and storage.

Enzyme assay tips

- Select enzymes as specific as possible for the tissue being investigated.
- Enzymes are broken down in the body at varying rates and most need to be assayed soon after sampling, some within a few hours.

Bovine/Ovine Biochemical Profiles

Profile	Sample(s)	Tests
Extended Profile	Serum + Ox/F (Red+Grey)	Albumin, AST, BHB, GLDH, GGT, Glucose, Magnesium, Phosphorus, Total Protein, Urea, Alk P, CPK and Creatinine
General Profile	Serum (Red)	Urea, Creatinine, Albumin, Total Protein, Bilirubin, AST, GGT and Alk P
Basic Production Profile	Serum (Red)	Albumin, Urea and BHB
Ewe Metabolic Profile	Serum (Red)	Ca, Mg and BHB
Fertility Profile	Serum + Ox/F + Heparin (Red+Grey+Green)	Albumin, Urea, BHB, Glucose, P and Cu
Fatty Liver Profile	Serum + Ox/F (Red+Grey)	AST, NEFA, BHB, GLDH and Glucose
Liver Profile	Serum (Red)	Total Protein, Albumin, Urea, Bilirubin, AST, GGT, Alk P and GLDH
Downer Cow Profile	Serum + Ox/F (Red+Grey)	Ca, Mg, P, AST, Urea and CPK
Myopathy Profile	Serum + Heparin (Red+Green)	GSH-Px, Vit E and CPK
Trace Element Profile	Serum + Heparin (Red+Green)	Cu, B12 (Cobalt) and Selenium (GSH-Px)

Haematology

Sampling

- Fresh EDTA blood and fixed 'animal-side' smear for routine haematology sent to the laboratory within 24 hours. Ensure tubes are filled but not overfilled. Mix well but do not shake. Heparin and Ox-F affect red and white blood cells and should be avoided.
- For clotting times and fibrinogen use fresh citrated blood (blue top vacutainer).

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 x 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 (minutes not hours), 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.

If the above guidelines are followed, primary fixation of most samples should take 24-48 hours. However, whole brains will take longer - please discuss with RL.

Packing and Sending

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

- 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% formol saline.

Microbiology

Sampling for Aerobic Bacteriology

- Samples should be as fresh as possible.
- Sear the surface of organs with a flame or heated scalpel blade prior to swabbing.
- Submit swabs in suitable transport medium. Charcoal transport swabs are satisfactory for most purposes but not for a flourescent antibody test (FAT) examination.
- Faeces samples (not just swabs) are essential if tests other than basic bacteriology are required.
- Fastidious organisms such as mycoplasma, campylobacter and leptospira require special attention. Please contact the RL to discuss sampling arrangements.
- Most bacteria will grow on culture medium after 24 hours incubation and antibiotic sensitivity will entail a further 24 hours. Some exceptions are:
  - Salmonella by enrichment - minimum 3 days
  - Campylobacter - up to 5 days
  - *Brucella abortus* - minimum of 4 days
  - *Mycobacterium paratuberculosis* (avian) spp - 6 to 12 weeks
  - Mycoplasma - 10 days to 4 weeks
  - Dermatophytes - 3 weeks.

Microbiology (continued)

Sampling for Anaerobic Bacteriology

- For swabs use anaerobe transport medium.
- For fluids fill the container to very top to exclude any air.
- Wrap tissues tightly in polythene to exclude any air.
  - For clostridial enterotoxaemia diagnosis, send a minimum of 2 ml of small intestinal contents collected from at least three sites in the ileum. Do not use any preservative. (Note: Clostridial toxins cannot be detected in kidney.)
  - For clostridial myositis or black disease in cattle, or *Clostridium novyi* infection in pigs, take four impression smears from the cut surface of affected muscle or liver, air-dry and send in slide box for a FAT, or submit tissue specimen in a sealed air tight container.

Cytology

- Aspirated fluid is best placed in an EDTA container before submission. Direct smears should be made in the same way as blood smears then air-dried and fixed in methanol for a minimum of 5 minutes. Please submit 2-3 smears to allow for different staining methods.

Mastitis examinations

- Contaminated milk samples will give misleading results. To avoid contamination use the following collection method:
  - Brush loose dirt, bedding and hair from teat. Grossly dirty teats should be washed and dried thoroughly.
  - Discard 5 streams of milk from the teat.
  - Pre-dip using an effective pre-dip product allowing 30 seconds contact time.
  - Dry teat thoroughly with a paper towel.
  - Scrub the teat end with cotton wool moistened with 70% alcohol. Use as many cotton wool pieces as necessary to clean the teat end, the last piece should be spotless.
  - Allow the teat to dry.
  - Remove the cap of a sterile sampling tube and hold it facing downwards. When collecting the milk sample, hold the tube at approximately at 45° angle. Do not allow the lip of the tube to touch the teat. Collect 1-3 streams of milk and immediately replace the cap.

Parasitology

Faeces

- Submit at least 10g faeces in a wide-mouthed, screw-capped container (50g for lungworm larvae).

Blood

- For blood parasites send 2 ml whole blood in EDTA tube or two thinly spread films fixed in methanol.

Skin

- For skin parasites send deep scrapings (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 in screw-capped containers. All should be submitted in screw-top containers and not envelopes.

Serology

- Paired sera are valuable in the diagnosis of respiratory and other disease outbreaks but please note the following:
  - If the first sample is taken too late the animals may have already seroconverted and the outbreak may be over by the time the second sample is due.
  - Sample several animals ensuring that they are identified properly for second bloods to be taken.
  - Sampling intervals can vary but as a general rule, they should not be less than two weeks.
  - Antibody levels in single samples are of limited value since they are difficult to interpret. Consult the RL for further advice.

Virology

Respiratory Viruses

FAT permit rapid identification of IBR, P13, RSV and BVD in both live animals and carcases.

Animal selection

- Select recently affected animals.
- Animals with muco-purulent nasal discharge are less likely to yield virus.
- Trachio/bronchial washings are the sample of choice and give the best results. Guarded nasal brush swabs are an alternative.

Carcases

- Submit intact fresh carcases or lung tissue.
  - Two or three blocks of lung tissue (2 cm cubes) from the junction between healthy and affected tissue or tracheal/bronchial swabs.
  - Tissues or swabs should be forwarded to the RL as soon as possible.

Isolation of viruses from field cases is not routinely undertaken, is time-consuming and often difficult as some respiratory viruses survive poorly in transport. When virus isolation is required the samples should be submitted in virus transport medium (VTM). Consult the RL before submisson.

Other Viral Diseases

- Sample as advised under the specific diseases.
  - For enteric viruses send faeces, without VTM, rather than swabs.
  - For viral skin diseases send deep scrapings and aspirated fluid (if available) in screw-topped containers.
  - For viral haemorrhagic disease of rabbits send a fresh carcase or liver sample.

Sampling by Disease Condition

Cattle

Condition/Infection	Specimen	Comments
Abortion, Stillbirth and Infertility		
***Abortion and stillbirth**		
Most bacterial infections and mycotic abortion	Whole fetus	Full examination with samples of choice taken at RL
	Fetal stomach and contents, fetal liver and placenta	Routine, selective and enrichment media cultures, stained smears and wet preparations
Neospora caninum	Fixed fetal heart, fixed fetal brain	In 10% formol saline for histopathology and ICC if appropriate
Fetal Serology BVD/*N.caninum/Leptospira hardjo*	Fetal fluid	Ensure fetus is immune competent- approx 100 days gestation
Maternal Serology IBR/BVD/*L.hardjo/N.caninum*	Blood - clotted	Paired samples are of limited diagnostic value. However, single samples are useful in maintaining disease surveillance and can identify a new infection entering the herd
Iodine deficiency	Fetal thyroid	Iodine assay/histopathology stillbirths - need to know thyroid and fetus weights plus gestation age

* All abortions and still births must be reported to the local Defra Animal Health Divisional Office who may require an official Brucellosis Investigation (BS7) to be undertaken.

Infertility		
Dietary insufficiency: BHB, urea and albumin	Blood - clotted	Sample a cohort of 5-6 cows Group mean BHB value >0.5mmol/L indicates inadequate energy intake
Cu and Selenium (GSH-Px)	Blood - heparin	Sample 5-6 animals not receiving concentrates
Campylobacter fetus venerealis	Vaginal mucus or lavage Sheath washings	Discuss sampling details with RL
Other		
Retained fetal membranes Iodine deficiency	Blood - heparin Blood - heparin	Vit E, and Selenium (GSH-Px) Plasma inorganic iodine
Downer Cows		
Complicated hypocalcaemia (prognostic tests)	Blood - Clotted and OxF	Ca, Mg, P plus AST, CPK and Urea
Cold Cow Syndrome	Blood - Clotted and OxF	Cu, Mg,P plus, AST, CPK and Urea

Cattle

Condition/Infection	Specimen	Comments
Enteric Disorders		
Calves up to 3 weeks		
Rotavirus, Coronavirus *Escherichia coli* K99, Salmonella Cryptosporidia	Faeces - from 3-5 affected calves	Neonatal Calf Scour Package
Calves over 3 weeks		
Salmonella, Coccidia and PGE	Faeces	Young Ruminant Scour Package
Adults		
Salmonella, Johne's disease, Liver Fluke and PGE	Faeces	Adult Ruminant Scour Package
Persistent BVD Infection and Mucosal Disease	Blood - heparin or EDTA	ELISA antigen and antibody tests. State age if less than 6 months (See Disease Surveillance and Export Tests Price List for bulk sampling cost reductions)
Acute BVD Infection	Blood - heparin or clotted	ELISA antibody test paired samples at two weeks interval. ELISA antigen on acute sample
Ill Thrift		
Copper, Cobalt/Vit B12, Selenium (GSH-Px) and Manganese deficiencies	Blood - heparin	Sample 5-6 animals on grass not receiving concentrates
Locomotor Disorders		
Nutritional myopathy	Blood - clotted and heparin	GSH-Px, Vitamin E and CPK

Cattle

Condition/Infection	Specimen	Comments
Mastitis and Milk Yield		
Common mastitis pathogens	Milk	Aseptic technique in sample collection is essential. See page 11. NB Batch charge for up to 10 samples submitted at one time and can be frozen prior to submission
Mycoplasma	Milk - very fresh	Milk samples for Mycoplasma culture should be kept cool and delivered to the RL within a few hours of collection
Reduced Milk Yield		
Dietary insufficiency	Blood - clotted and Ox-F	Sample animals typical of the group whose yields have not already fallen. Do not include sick animals or exceptionally high yielding animals
Milk Drop		
Leptospira hardjo	Blood - clotted	MAT is the test of choice for demonstrating current clinical disease. Paired samples with an interval of two weeks are recommended. Because the antibodies measured by MAT decline fairly rapidly ELISA is the test of choice for determining herd sero-prevalence and long-term evidence of infection in individual animals
IBR	Nasopharyngeal or ocular swabs BAL Blood - clotted	FAT - Swabs must be plain or in VTM not Charcoal Transport Medium. A minimum of four acute cases should be sampled. Paired samples at two week intervals
Tick-borne Fever	Blood - EDTA - clotted	Sample pyrexic animals. Microscopic examination of blood film. Paired samples at minimum of two weeks interval
Nervous Disorders (Always consider BSE)		
Hypomagnisaemia Nervous acetonaemia	Blood - Clotted	Mg, BHB
Listeriosis	Blood - paired sera 2 weeks interval	SAT - single samples have virtually no diagnostic value
Lead poisoning	Blood - Heparin or OxF Liver/kidney/paint or other suspected source	
Cause uncertain	Carcase	If deaths occur post-mortem examination will be more likely to yield a diagnosis

Cattle

Condition/Infection	Specimen	Comments
Respiratory Disease		
IBR Mycoplasmosis	Nasopharyngeal or ocular swabs BAL	FAT - swabs must be plain or in VTM not Charcoal Transport Medium Transport media available from RL.
RSV and PI3	Nasopharyngeal swab or lung sample BAL	FAT - swabs must be plain or in VTM not Charcoal Transport Medium
IBR, RSV, P13, Haemophilus, Mycoplasma & BVD	Blood - paired sera at 2 or more weeks interval nasopharyngeal swab BAL	ELISA/CFT on paired SERA Bacterial culture and FAT
Patent husk	Faeces Blood - Clotted	Fresh samples most useful. Only indicates recent exposure to the parasite ELISA
Re-infection husk	Blood - Clotted & EDTA	ELISA indicates previous antigenic contact. Eosinophilia may be seen if sampled early
Malignant Catarrhal Fever	Blood - clotted	IFAT for ovine herpes virus - 2 antibody. PCR for antigen no longer available
Other causes	Carcase	If deaths are occurring submission of a whole carcase may be the cheapest and most reliable diagnostic approach
Sudden Death (always consider Anthrax)		
Hypomagnesaemia	Bone - rib or tail vertebra aqueous humour	Only of value for calves less than 9 months of age Results must be interpreted with caution
Blackleg	Muscle - fresh, in a full, sealed container to exclude air	For FAT and anaerobic culture
Nitrate poisoning and acorn poisoning	Aqueous humour (must be fresh)	Discuss with RL
Water dropwort, yew, acorn and other plant poisoning	Suspicious material from rumen contents (in sealed container)	Discuss with RL
Arsenic poisoning	Rumen contents, liver and kidney	Discuss with RL
Lead poisoning	Liver, kidney, paint or other suspected source	Discuss with RL

NB: Please discuss all cases of suspected poisoning in food animals with the RL, as voluntary measures to control contamination of the food chain may be requested or statutory controls imposed under the Food & Environmental Protection Act (FEPA). Submission of whole carcase is advisable, where possible, in most incidents.

Screening or Monitoring for Infectious Diseases

IBR, BVD and *L.hardjo*	Milk - Bulk* - Individual	Bronopol preservative required RLs can supply sample tubes.
IBR, BVD, *L.hardjo,* MAP Husk, Fasciolosis, Neospora EBL	Blood - clotted Bulk milk for BVD PCR (contact RL first).	Screening or purchased animals, hired bulls and monitoring of immune status of heifer groups etc.

* Further milk antibody ELISA tests eg. Fasciolosis are currently being developed and will be publicised when these are available.

Sheep and Goats

Condition/Infection	Specimen	Comments
Abortion, Stillbirth and Infertility		
Abortion and stillbirth		
Whole fetus and placenta is the submission of choice		
Enzootic abortion	Placenta Maternal blood	Gross examination, stained smears CFT
Toxoplasmosis	Placenta Thoracic fluid Maternal blood - single	Gross examination, FAT DAT or IFAT LAT
Campylobacter and other bacterial infections	Fetal stomach contents and liver	Culture and wet preparations. Send whole fetus and placenta if samples do not resolve a diagnosis
Infertility, other		
Cause uncertain		Discuss cases with RL
Downer Cases		
Hypocalcaemia, pregnancy toxaemia, hypomagnesaemia	Blood - Clotted	Ca, BHB, Mg. Also of use may be AST, CPK, phosphorus and urea. NB - listeriosis should also be considered
Enteric Disorders		
Lambs up to 3 weeks		
Bacteria, rotavirus, cryptosporidia and coccidia	Faeces	Neonatal scour package reduces cost of tests - see Disease Surveillance and Export Tests Price List culture, PAGE and stained smear
Lambs over 3 weeks		
Coccidia & PGE	Faeces	Worm egg count and coccidial oocyst count on faeces
Johne's disease	Faeces Clotted blood	If Johne's disease is suspected, sample several cases as chance of detection is low. Ideally submit carcase AGID test
Ill Thrift - Young Animals		
Parasitism	Faeces	Faecal egg count
Border disease	Blood - Heparin and Clotted	ELISA for antibodies and culture for virus
Copper deficiency	Blood - Heparin	Sample 5-6 animals **not** receiving concentrates
B12 deficiency	Blood - Clotted	Sample 5-6 animals. Magnetic separation assay

Condition/Infection	Specimen	Comments
Locomotor Disorders		
Nutritional myopathy	Blood - Clotted and Heparin	GSH-PX, CPK, Vit.E
Arthritis due to *Erysipelothrix* spp.	Blood - Clotted Joint fluid aspirate	SAT Culture
Mastitis		
Staphylococci, pasteurellae, coliforms etc	Milk	Culture
Nervous Disorders (scrapie is a notifiable disease and suspect cases should be reported to the DVM)		
Hypocalcaemia, pregnancy toxaemia, hypomagnesaemia	Blood - Clotted	Mg, BHB, Ca
CCN	Carcase	If deaths occur post-mortem examination is indicated
Listeriosis	Blood - paired sera 2 weeks interval essential Carcase	SAT Post-mortem examination
Cause uncertain	Carcase	If deaths occur post-mortem examination will be more likely to yield a diagnosis - possibly at less cost
Respiratory Disease		
Maedi Visna	Blood - clotted Carcase	ELISA or AGID test Post-mortem examination
Pasteurellosis, sheep pulmonary adenomatosis	Carcase	Submission of a whole fresh carcase may be the cheapest and most productive diagnostic approach
Sudden Death (always consider Anthrax)		
Clostridial enterotoxaemia/ pulpy kidney	Intestinal contents - fresh (do not add preservative)	ELISA on contents from affected portion of bowel - if not obviously affected send ileal contents bulked from at least 3 sites
Pasteurellosis	Lung and liver	Culture
Other clostridial disease Fasciolosis/haemonchosis etc	Carcase	Post-mortem examination of a fresh carcase at the RL is more likely to yield a diagnosis and may be cheaper than submission of viscera

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

Pigs

Introduction

- It is important to inspect, examine and sample sufficient animals to identify a problem of significance in a herd. Submission of individual pigs, or samples from individual pigs, is not ideal.
- Where possible, submit animals at the acute stage of the disease process.
- Where possible, sample untreated animals. Take samples for attempted bacterial culture from acute cases and from untreated pigs.
- You may wish to discuss cases with RL staff in advance of submitting samples.
- For certain disease syndromes (e.g. reproductive, enteric, respiratory) packages are available - please refer to the current VLA Disease Surveillance and Export Tests Price List.
- The following notes are guidelines for sampling and are not comprehensive.
- Post-mortem examination of representative animals at the RL is often more cost beneficial than submitting tissue samples from dead animals. Always telephone the RL before submitting carcases. Where possible, and subject to welfare considerations, submission of live animals is preferable to dead.

Condition/Infection	Specimen	Comments
Abortion, Stillbirth and Infertility		
Abortion and Stillbirth		
Most bacterial infections	Fresh fetal stomach contents Liver and cervical/vaginal swabs	Culture
Parvovirus	Lung and liver Thoracic fluid Single maternal bloods	Fetuses less than ~17cm crown-rump length or mummies: ELISA for antigen. Fetuses greater than ~17cm: HAIT for antibody (sows will have seroconverted)
Erysipelas, influenza, Leptospirosis and PRRS	Maternal bloods	Single or paired serology
Cause uncertain	Whole litter of fetuses with fetal membranes and maternal bloods (clotted/heparin)	Discuss with RL
Infertility, other reproductive disorders e.g. vulval discharge	Guarded vaginal/cervical swabs taken via speculum	Culture - misleading results may occur due to urinary infection
Returns-to-service	Group blood samples Paired serology	Serology for evidence of active PRRS, influenza, leptospirosis, erysipelas & parvovirus. Most regular returns-to-service are physiological or managemental

Pigs

Condition/Infection	Specimen	Comments
Enteric Disorders		
Neonates/sucklers		
Diarrhoea or death in all age groups	Carcases or (preferably) live animals	Submit acutely affected pigs. Discuss with RL as necessary, especially if TGE or PED suspected
Enterotoxogenic *E.coli* (ETEC)	Rectal swabs, faeces, carcases	Culture, serotype and antimicrobial susceptibility
Non-ETEC coliforms e.g. attaching and effacing *E.coli* (AEEC), Enterohaemorrhagic *E.coli* (EHEC)	Live pigs	Histology essential for diagnosis
Clostridial necrotic enteritis	Carcases or necrotic bowel with contents	Gross pathology, Gram smear, culture, ELISA for toxins
Salmonella	Faeces (preferably >15 gm) Swabs are less sensitive	Culture. Infection is often subclinical but of public health significance
Coccidiosis (usually 7 - 14 days)	Live untreated acutely scoured piglets	Histology of intestine essential. Faeces usually negative for oocysts during early clinical period. Mucosal smears are of limited diagnostic value
Cryptosporidium spp	Faeces/large intestinal contents. Live pigs	Often subclinical
Rotavirus	Faeces/intestinal contents	PAGE test
TGE	Live pigs Faeces/intestinal contents	Antigen ELISA FAT/serology - check with RL first.
PED	Live pigs Faeces/intestinal contents	Antigen ELISA/serology - check with RL first
Weaners/growers		
E.coli, Salmonella, rotavirus, TGE and PED	as above	
Swine dysentery, spirochaetal and non-specific colitis	Fresh faeces, carcases, colon/caecum, large intestinal contents	PCR†, gross pathology, histology, FAT/anaerobic culture
Proliferative enteropathy/adenomatosis *(Lawsonia intracellularis)*	Carcases/live pigs Faeces Clotted blood	Gross pathology, histology, silver stains, PCR†, MZN smears, serology†
Bowel oedema	Carcases	Gross pathology, culture, serotyping verocytotoxic *E.coli*
PGE	Faeces	Worm egg count
Gastric ulceration	Carcases	Gross pathology

Pigs

Condition/Infection	Specimen	Comments
Adults		
Swine dysentery, PGE, Salmonella, proliferative enteropathy/adenomatosis	Faeces, Carcases	
Iron deficiency anaemia	Blood - EDTA/heparin	Haematology/iron biochemistry
Mastitis	Milk (sampled aseptically)	Culture
Heart diseases		
Endocarditis	Carcases Swab from vegetative lesion	Culture
Mulberry heart disease	Carcases Fixed heart Blood (clotted/heparin)	If Vitamin E/selenium deficiency is suspected, consider blood sampling a representative group
Respiratory Diseases		
Influenza, PRRS/PRCV	Blood - paired at 2-3 weeks interval or group sampling carcases.	Gross examination and histopathology and IHC
PCV-2 pneumonia and PNP	~ 4 acutely affected pigs (usually 10-16 weeks old) or formalin fixed lung and respiratory tract lymph node from at least 3 acutely affected pigs	Gross pathology, histopathology and IHC if appropriate
Progressive atrophic rhinitis (toxigenic *Pasteurella multocida*)	Nasal/tonsil swabs	Culture - NB essential to sample before obvious clinical signs. Submit at least 20 swabs where the level of overt disease is low.
	Snouts collected at slaughter (>=20)	Monitoring of chronic damage to turbinates and nasal septum
Actinobacillus pleuropneumoniae, Pasteurella multocida, Haemophilus parasuis	Lungs	Culture
Enzootic pneumonia *(Mycoplasma hyopneumoniae)*	Lungs	For monitoring of gross lesions, examine at least 20 lungs. PCR for diagnosis†

Tests marked with † are not performed by VLA and may still be awaiting validation data.

Pigs

Condition/Infection	Specimen	Comments
Other Conditions		
PMWS/PDNS	4 to 6 acutely affected pigs (usually 8-12 weeks old) or formalin fixed lymph nodes (inguinal, mesenteric and renal) and kidney from at least 3 acutely affected pigs	Gross pathology, histopathology and IHC if appropriate.
Sudden death (always consider anthrax)	Carcases	Discuss with RL where appropriate
Locomotor Disorders		
Erysipelas and *Mycoplasma hyosynoviae* arthritis	Blood - Clotted, paired samples Joint fluid aspirate Carcases	SAT for *Erysipelothrix* spp., culture for *M.hyosynoviae* and *Erysipelothrix* spp.
OCD, spontaneous fractures, etc	Carcases	
Nervous disorders		
e.g. streptococcal meningitis, salt poisoning, oedema disease, congenital tremors, etc.	Carcases Meningeal swabs for bacteriology	
Skin diseases		
Many conditions, e.g. exudative epidermitis, PDNS, mange, ringworm, swine pox	Carcases Skin scrapings Swabs	Discuss with RL
Urogenital disease		
e.g. cystitis/pyelonephritis	Carcases Urine	
Polyserositis and Glasser's disease	Carcases Serosal surface swabs	Culture
Eperythrozoonosis	Blood (heparin) or fresh smears	Preferred sample is a blood smear prepared from a live pig, air-dried and methanol-fixed on the farm

Birds

Poultry and Game Birds

- In most cases a flock problem is already being investigated. Please supply a good clinical history to include age, morbidity and mortality patterns, information on medication and vaccination policy and the type of husbandry system. A batch of birds (e.g. 3 to 6) should be submitted for post-mortem, dead and live moribund birds should be submitted as appropriate. Individual birds may be all that is available from small flocks. Serology can often be a useful diagnostic tool in flock problems.

- Fresh carcases should be submitted as post-mortem autolysis occurs rapidly particularly with chicks. Carcases should not be frozen as this leads to tissue deterioration and renders histology useless.

- Parasitic infections caused by motile protozoa (Hexamita, Trichomonas, Spironucleus/Hexamita etc) are particularly prevalent in game birds. It is essential that live birds are submitted for an accurate diagnosis to be made.

Psittacines

- For safety reasons post-mortems should only be carried out in a microbiological safety cabinet. If sending carcases through the post, please ensure they are double-wrapped in plastic bags before being boxed. On the outside bag apply an obvious note 'Suspect Psittacosis' so that our staff are not accidentally exposed to infection whilst unwrapping the post.

- All psittacines are treated as potentially infected with psittacosis and will be routinely screened for this condition. If you wish to screen live birds, pooled faeces from an aviary or individual bird samples can be tested by the PCR. Send fresh unpreserved faeces or cloacal swabs but do not use swabs with wooden shafts. If a negative result is found then a repeat sample 7-10 days later gives greater assurance of freedom from infection.

Other

Exotic Farmed Animals and Zoological Collections

The basic guidelines for the principal agricultural livestock species listed above should be used as a guide.

VLA cannot examine primates, insects or most reptiles. When submitting animals from zoological collections the RL must be consulted prior to submission.

Wildlife

VLA does have the facility to examine wildlife casualties but the RL must be consulted before submission of carcases or diagnostic material. Of particular interest to VLA and Defra is mortality in wild birds.

Any suspect malicious or accidental poisoning or misuse of pesticides or agrochemicals affecting wildlife should be reported to the Defra Wildlife Incident Investigation Scheme (WIIS) in the first instance (Freephone 0800 321 600).

The Pet Travel Scheme (PETS)

A VLA submission Form (VLA RAB 1) must accompany any sample submissions before registration can take place. Samples must be sent to VLA Weybridge.

VLA produces a 'Mailshot' package consisting of:

- Plain tube
- Polythene bag (for tube)
- Submission Form
- Pre-paid self-addressed padded jiffy bag.

'Mailshots' are now generally issued upon request.
The current turnaround time to produce results is four weeks from sample receipt (see page 5 for contact details).

What VLA does with the data you supply

Animal disease surveillance is a very important function of the Veterinary Laboratories Agency. In order to carry it out effectively we require high quality information from the field. This data is used in a variety of ways to the benefit of both Defra, who need to monitor endemic diseases and the practising veterinary surgeon who is involved in practical control at the farm level.

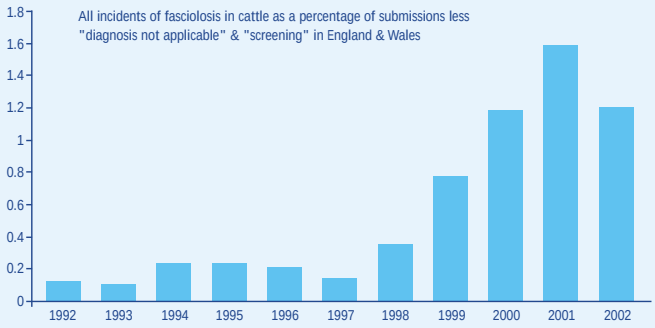

General Submission Form

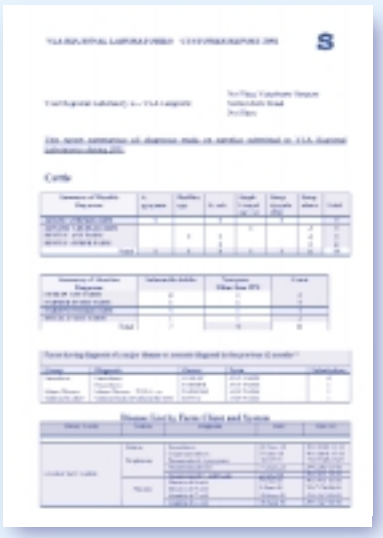
Chronic diarrhoea

Adult ruminant enteric package

Disease Surveillance

The information supplied with the submission form helps VLA to give Defra an accurate assessment of the trends in endemic animal disease and an indication of the emergence of any novel conditions. It also allows us to provide rapid epidemiological analysis of new and emerging diseases and assess their relative risk at an early stage.





Benefits for the Practising Veterinary Surgeon

The powerful database used to store the information supplied on the submission form is used to produce reports for veterinary practices which summarise all the diagnoses made by farm clients for the past year. These reports will be refined in response to feedback from the practices. The reports can currently provide the following;

- A convenient record of disease diagnoses by client for the benefit of all members of the practice.
- A document which can be used as a record of disease diagnosis and a demonstration of the degree of disease monitoring which has been undertaken to support herd and flock health plans.
- A way of identifying farm clients who would benefit from a specific disease control programme such as vaccination.

In the Future

Provision of accurate information on farm holding numbers (CPHH) and addresses will allow us to relate disease incidents to animal populations and geographical location by using GIS (Geographical Information Systems). Using these systems we will be able to detect clusters of disease incidents, relate the numbers of diagnoses to animal populations and track whether we have examined sufficient samples from different regions to provide adequate disease surveillance. In this way we can provide the highest quality information for both Defra and the practising veterinary surgeon.

Where to view disease surveillance information

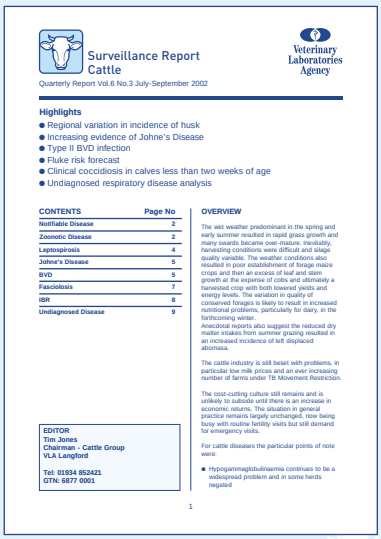
Everyone who has an interest in animal disease surveillance can now access this via the Defra website:

www.defra.gov.uk/animalh/diseases/surveillance_reports

Or simply go to the Defra website www.defra.gov.uk and navigate via Animal Health and Welfare/Disease Surveillance and Control/Endemic Disease Surveillance.

It can also be accessed via the VLA website www.defra.gov.uk/corporate/vla

You can view information on diseases of cattle, sheep, goats, pigs, exotic farmed species, poultry and wildlife compiled by VLA Species Groups on the Endemic Disease Surveillance pages.



Surveillance Report
Cattle

Veterinary Laboratories Agency

Quarterly Report Vol.6 No.3 July-September 2002

Highlights

- Regional variation in incidence of husk
- Increasing evidence of Johne's Disease
- Type II BVD infection
- Fluke risk forecast
- Clinical coccidiosis in calves less than two weeks of age
- Undiagnosed respiratory disease analysis

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OVERVIEW

The wet weather predominant in the spring and early summer resulted in rapid grass growth and many swards became over-mature. Inevitably, harvesting conditions were difficult and silage quality variable. The weather conditions also resulted in poor establishment of forage maize crops and then an excess of leaf and stem growth at the expense of cobs and ultimately a harvested crop with both lowered yields and energy levels. The variation in quality of conserved forages is likely to result in increased nutritional problems, particularly for dairy, in the forthcoming winter.
Anecdotal reports also suggest the reduced dry matter intakes from summer grazing resulted in an increased incidence of left displaced abomasa.

The cattle industry is still beset with problems, in particular low milk prices and an ever increasing number of farms under TB Movement Restriction.

The cost-cutting culture still remains and is unlikely to subside until there is an increase in economic returns. The situation in general practice remains largely unchanged, now being busy with routine fertility visits but still demand for emergency visits.

For cattle diseases the particular points of note were:

- Hypogammaglobulinaemia continues to be a widespread problem and in some herds negated

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Surveillance Report
Small Ruminants

Veterinary Laboratories Agency

Quarterly Report Vol.6 No.3 July-September 2002

Highlights

- Ovine chorioptic mange diagnosed in Devon
- Small ruminant lentivirus infection conference report
- Summary of diagnosis not reached incidents
- *Mycoplasma adleri* in UK sheep

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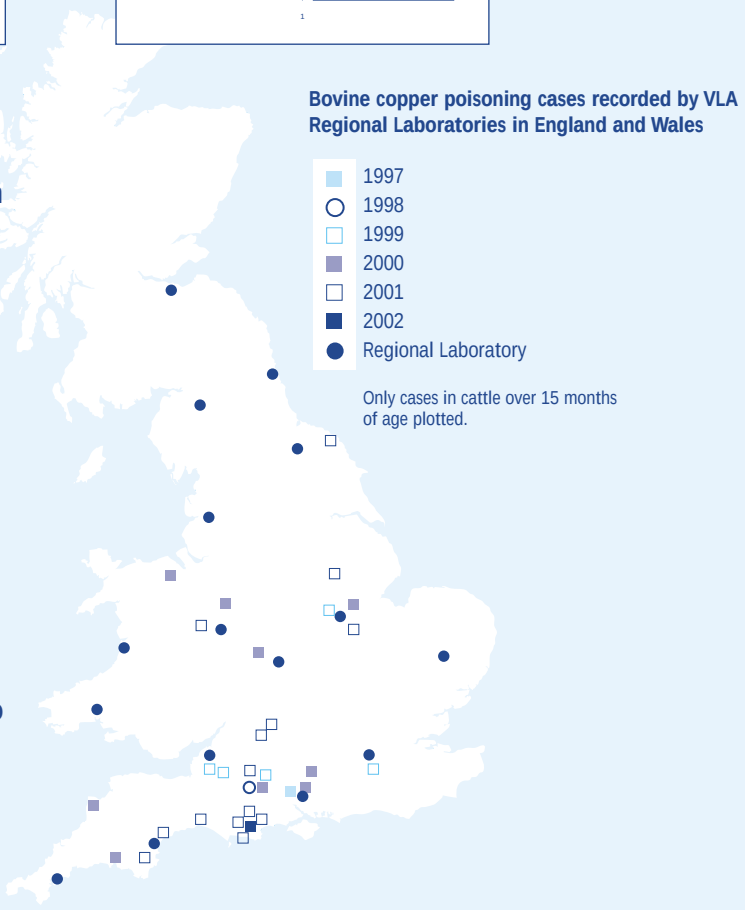
Why accurate farm identification is important

Relating disease outbreaks to individual holdings is an essential part of disease surveillance and monitoring.
It is important for VLA to track the number of submissions to Regional Laboratories to ensure that the Defra Surveillance Division is aware of the degree of national coverage. It is also important in providing denominator data to measure trends in certain diseases.
The map shows how diseases can be tracked geographically. This will eventually result in much better information being available for all involved in controlling disease.

The entry of an accurate farm address and CPHH number on all submissions is essential to this process.

Bovine copper poisoning cases recorded by VLA Regional Laboratories in England and Wales



Only cases in cattle over 15 months of age plotted.

VLA would like to thank all its customers for providing this information and helping us to achieve our surveillance aims.

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Veterinary Laboratories Agency

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defra