Herdsure® Cattle Health Improvement Service

Handbook for Veterinary Practitioners

AHVLA
Animal Health and Veterinary Laboratories Agency

Herdsure®

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Acknowledgements

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Quick Guide
to the Herdsure® Cattle Health Improvement Service

Introduction

This is a quick guide to the Herdsure®[1] Cattle Health Improvement Service and how you access it. The main body of the handbook describes the service fully.

Cattle health management is vital to maximise the productive potential of herds and to reduce the cost of disease to the farm business. It is also an essential part of animal health and welfare. 'Proactive' cattle health management involves assessing the impact of disease on the farm and implementing a strategy to reduce the impact of that disease.

The Animal Health & Veterinary Laboratories Agency (AHVLA) aims to act as the **facilitator** of health management via the service, offering a means of assessing and improving the health status of cattle.

Sampling, testing and management protocols are available for bovine viral diarrhoea (BVD), Johne's disease, liver fluke infection, infectious bovine rhinotracheitis (IBR), leptospirosis and neosporosis.

Protocols provide step-by-step guidance on how to approach the control or eradication of each disease, depending on the local circumstances of each farm. They help the veterinary practitioner to develop a robust plan for testing, treatment and monitoring of the herd. The protocols are supported by high quality laboratory tests, many of which are subject to third party accreditation by the United Kingdom Accreditation Service (UKAS) operating to the internationally recognised ISO/IEC 17025 standard for laboratory competence. All tests carried out under the Herdsure® service are accredited to this standard unless marked with '†' on reports.

Advice on biosecurity and other herd management factors is provided within the protocols.

The service is supported by the Herdsure® Management System (HMS), a specially developed IT system used by AHVLA, designed to manage customer records, monitor each farm and prompt when sampling is due.

[1] The name Herdsure® has been introduced and registered as a trademark for use with AHVLA's new cattle health services. This, and associated names, may be applied to future services as well as the Cattle Health Improvement Service – for example other new offerings originating from the wide range of services currently delivered for the overall health of livestock and support of AHVLA's customers.

What are the main benefits for the veterinary practitioner?

- Herdsure® offers an additional service to provide to your farmer clients in order to support health management.
- Health management strategies and planned and structured approaches to sampling and testing, developed by AHVLA experts, ensure optimal quality of service.
- The unique IT support system in use by AHVLA, HMS, provides prompts and the identification of animals to be sampled to both you and your farmer client (as appropriate), thus reducing the management role of your busy veterinary practice.
- Guidance and advice are provided in this handbook, plus further telephone consultancy is available if required.

What are the main benefits for your farmer clients?

- Improved herd health through the control of up to six major cattle diseases.
- Better knowledge of cattle health status leading to improved production and less financial loss.
- User-friendly and straightforward to implement.
- Minimal record keeping owing to automatic tracking of progress and prompting when tests are due.
- Minimal paperwork, for example by informing farmers (and their veterinary practitioner) when tests are due.

Main features of the service

- Sampling, testing and management protocols suitable for both beef and dairy herds, commercial and pedigree, developed by AHVLA disease experts.
- Detailed guidance and advice for veterinary practitioners in this handbook and further consultancy when required.
- Access to a telephone helpline for technical enquiries and consultancy related to farms enrolled into the service for the veterinary practitioner.
- Flexible/tiered subscription based on the size of the herd and the diseases chosen.
- A separate handbook for your subscribing farmer clients, which explains the main features of the service and their role within it.
- Submission forms with pre-printed ear tag numbers of animals to be sampled, where appropriate.
- Provision of sampling containers and packaging for farmers to collect relevant samples. These can then be returned using convenient postage-paid Business Reply labels.

- Proactive monitoring of each registered farm, including reminders to both veterinary practitioners and farmer when tests are due.
- Generation of reports in ways that best suit your practice.
- Issue of annual herd progress reports.

Key to symbols used throughout the protocols

	Use Herdsure® postage-paid Business Reply labels.
	Indicates there will be reminders sent when tests are due.
	This symbol indicates a biosecurity issue.

The three levels of health status within the protocols

Protocols describe a programme of testing, treatment and continued monitoring that assist both the veterinary practitioner and the farmer to control and reduce disease and its detrimental impact on the farm and the herd.

- **Level 1** establishes the health status of the herd with regard to the diseases included in the Herdsure® protocols.
- **Level 2** aims to improve the health status of the herd with regard to the diseases included in the Herdsure® protocols.
- **Level 3** monitors and aims to maintain the improved (or established as satisfactory at Level 1) health status of the herd with regard to the diseases included in the Herdsure® protocols.

An **annual herd progress report** will be issued to Herdsure® members. This progress report will detail the level achieved for each protocol for which the herd is enrolled on the date of issue.

What are the roles of AHVLA and the veterinary practitioner?

AHVLA provides you with:

- Detailed sampling, testing, interpretation and management protocols for each disease and for each of the three levels within Herdsure®.
- Testing of samples according to the relevant testing protocol.
- Advice on management and biosecurity relevant to the control of each disease within the Herdsure® service.
- Prompts for testing and, where appropriate, the identification of animals to be tested.
- Access to a national helpline if further advice is needed.

Your role will be to:

- Identify clients who wish to enrol in Herdsure® and complete a registration form for each.
- Identify management groups within the herd, in relation to the Herdsure® disease protocols.
- Collect samples according to the relevant testing protocol and following reminders by the Herdsure® Management System.
- Advise your client on how to implement necessary management actions, in accordance with the guidance in the Herdsure® protocols, following the reporting of test results by AHVLA.
- Help your clients to identify and implement changes in biosecurity procedures, in accordance with guidance in the Herdsure® protocols.

Getting started

Registration of the veterinary practice

Your practice will have registered (using Form A in the 'Forms' section of this handbook) in order to receive this handbook. You need only register once, no matter how many farms you enrol.

What does it cost my practice?

There is no charge for veterinary practices to register for the service.

Enrolling farms

Please enrol farms using Registration Form B included in the 'Forms' section of this handbook. A separate registration should be photocopied and completed for each farm.

Herds participating in other health programmes may be eligible to join a Herdsure® health programme at Level 2 or 3. Requests for consideration of these cases should be made to a Herdsure® consultant, via the Herdsure® helpline, when an appropriate decision will be made based on veterinary criteria.

Some herds may be considering Cattle Health Certification Standards (CHeCS) accreditation; for enrolling these refer to the CHeCS section below.

What does it cost the farmer?

A subscription is payable by farms to enrol, renewed annually. It is based on the size of herd and the number of diseases to which they subscribe. Please refer to the section on 'Accessing the service – definition of a registered herd' for the definition of a 'herd' for this purpose. All communications (including invoicing) will be with the registered veterinary practice.

Tests are competitively priced at special rates. For details see www.ahvlascientific.com.

What happens next?

Depending on which disease protocol(s) the farmer has chosen to subscribe to, the relevant protocol(s) will be triggered. Sample requests will be sent to your practice and on occasion, e.g. milk, also to the farmer.

Important: No samples should be submitted before receiving the first notification.

Test results, in the form of a report, will be sent to your practice and will include notification of the next action (which will depend on test results). You will be sent detailed sampling instructions when this action is due.

An **annual herd progress report** will be issued to Herdsure® members. The progress report will detail the level achieved for each protocol for which the herd is enrolled on the date of issue.

The report relates to the herd rather than to an individual animal.

Cattle Health Certification Standards – (CHeCS)

Herdsure® is licensed by CHeCS. Farmers enrolling in Herdsure® have the option to become CHeCS-accredited as being free of disease for BVD, IBR, Johne's disease and Leptospirosis.

Your client will be eligible to apply for CHeCS accreditation and awarded certificates of health status provided they comply with strict CHeCS biosecurity rules. These can be found in the CHeCS technical document (www.checs.co.uk).

Completion of Form D following the Appendices should be completed at registration, along with Form B, and should also accompany each sample submission.

Pen Cards

One of the benefits of the Herdsure® Management System is that results are stored for individual animals against their official ear tag numbers. This enables Herdsure® members to request (for a small fee) Pen Cards, a summary of test results for individual animals, prior to sale. Please also note that although animals may be part of a tested herd it does not necessarily follow that they have been tested individually. Should your client wish to apply for these please call the helpline and we will send you an application form. Please note that Herdsure® requires the following notice:

- 6 weeks notice when testing is required
- 2 weeks notice if testing has already been completed

otherwise we cannot guarantee they will be sent on time.

Service Guidance
for the Herdsure® Cattle Health Improvement Service

Background to cattle health management

Farm health planning

Farm health planning is supported by Defra and is central to the delivery of the Animal Health and Welfare Strategy in England (2004). Defra has funded 27 projects in the beef and dairy sectors in England to promote the use of active farm health planning and has highlighted five challenges that had to be met for the selection and implementation of projects:

- Overcoming initial barriers to widespread adoption of farm health planning.
- Achieving higher standards of animal health and welfare.
- Contributing to a more profitable and sustainable cattle industry.
- Generating a support network to promote and advise on farm health planning.
- Promoting voluntary adoption of farm health planning techniques so they become an automatic part of livestock keeping.

'Proactive' cattle health management

'Proactive' cattle health management involves assessing the impact of disease on the farm and implementing a strategy to reduce the impact of that disease.

It involves a dynamic partnership between farmer, veterinary practitioner and relevant consultants (such as nutritionists, housing or milking plant consultants and consultants in the control of infectious disease). It is not simply about producing the documentation necessary for assurance schemes or audit trails. Cattle health management issues on farms may be diverse and wide ranging but they need not all be tackled at once.

Often the starting point will be clear. For example, a farm may be experiencing a high incidence of Johne's disease, leading to deaths or an increase in the number of adult cattle culled. This may be recognised as the major cattle health issue on the farm without looking at the records.

For others, keeping records of infectious disease, mastitis, fertility, abortion, sudden deaths, traumatic injuries, lameness and production will be the key to the start of 'health improvement planning'. Record keeping and identifying areas for improvement are important parts of 'farmer-veterinary practitioner' partnerships.

Quantifying the financial losses can be difficult, especially when the consequences are diverse and variable in different herds. Where the result is tangible, the financial impact to a farm business can be measured; for example, a reduction in the abortion rate in a dairy herd will produce a financial benefit. Other losses, e.g. those associated with the immunosuppressive effect on cattle exposed to BVD virus, are more difficult to quantify and are often overlooked. A study carried out for the Milk Development Council by the Scottish Agricultural College[2] concluded that 'BVD is of great economic importance and highlights reproductive and immunosuppressive losses that are frequently ignored'.

[2] SAC Final Report for the Milk Development Council, *Estimating the Economic Losses associated with BVD infection in the UK Dairy Herd,* Nov. 2000.

AHVLA's approach to supporting cattle health management

Facilitating health management

For many commercial herds, attaining an accreditation of 'freedom from disease' is not necessarily the priority. Instead, the priority is control of disease and the reduction of its impact on animal health and welfare, as well as its impact on the economics of the farm. AHVLA, via the Herdsure® Cattle Health Improvement Service, aims to facilitate a partnership between the veterinary practitioner and the farmer to achieve this.

Use of the protocols

The testing and management protocols used in Herdsure® provide the veterinary practitioner with the tools to implement effective management of disease on farms.

Their implementation is very much in the hands of the veterinary practitioner; his or her partnership with the farmer client will ensure that all the appropriate procedures are successfully completed.

Each protocol provides a step-by-step guide to how to approach the control or eradication of a particular disease, providing scientifically robust and supported testing approaches coupled with advice on biosecurity and other herd management factors that need to be considered.

Descriptions of the levels of 'health status' within the protocols

Within the protocols there are three levels of health status. The current status of the herd and the date, and type of the next test due, will be included when reporting Herdsure® results.

In addition, an **annual herd progress report** will be issued to Herdsure® members. The progress report will detail the level achieved for each protocol for which the herd is enrolled on the date of issue. The report relates to the herd rather than to an individual animal.

For a small fee, an updated progress report can be produced. The updated progress report, like the annual report, will detail the level achieved for each protocol for which the herd is enrolled on the date of issue.

Level 1: Establishes the health status of the herd

Level 1 acknowledges that sufficient testing has been carried out in the herd, in accordance with the relevant disease protocol, to establish the disease status of the herd for that disease. It does not infer absence of disease or that an active programme to control or eradicate disease is in progress.

Where there is documented evidence of a particular disease currently in the herd, testing at Level 1 will be considered superfluous. In these cases herds will be advised to start the protocol at Level 2.

Level 2: Aims to improve the health status of the herd

Level 2 acknowledges implementation of a planned intervention that is aimed to improve the infectious disease status of the herd. This level is facilitated through the veterinary practitioner using the relevant disease protocol and the implementation of biosecurity measures and management actions.

This level recognises that, in some cases, it will be unrealistic to eliminate a particular infectious disease from the herd quickly; culling of a significant number of potentially infected animals over a short period of time may not be economically viable for the herd. However, the disease may be controlled by taking specific management actions, including vaccination in some instances. The success of that control can be monitored using the testing approaches described in the relevant disease protocol until the level of disease is sufficiently reduced to make its elimination a realistic goal.

In other cases, Level 1 results may indicate that a more proactive approach through the removal of a limited number of infected animals may be successful in quickly eliminating active disease.

Level 3: Monitors and aims to maintain the improved (or established as satisfactory at Level 1) health status of the herd

Herds will be able to move to Level 3 in two ways:

- Directly from Level 1, when the results of testing in the Level 1 protocol show no evidence of active disease.
- Via Level 2, when sampling and testing carried out at Level 2 has resulted in the removal of active disease.

An **annual herd progress report** will be issued to Herdsure® members. The progress report will detail the level achieved for each protocol for which the herd is enrolled on the date of issue.

The annual progress report will only remain valid until the next scheduled testing is complete. Where this testing is not conducted, no further progress reports will be issued and the holding will be deemed to have lost its status.

The importance of biosecurity to the success of cattle health improvement

Control of infectious disease cannot be achieved without addressing biosecurity on the farm. In recent years, the pressure on farms to maximise the economy of scale has resulted in a need to increase cattle numbers more rapidly than the breeding programme will allow. Furthermore, this effect is exacerbated by losses from culling as a result of disease. Faced with these realities, many units have been forced to abandon the 'closed herd' policy and 'buying in' replacement cattle has become a regular practice on the majority of farms.

Once a herd has embarked on a programme to eradicate a disease, or indeed has achieved eradication and wishes to remain disease-free, it is important that a number of biosecurity principles are adhered to. Disease-specific biosecurity principles are included in the relevant sections in this handbook and the protocols themselves include further information.

It is acknowledged that, for some herds, purchase of cattle will necessarily continue and, for this reason, the protocols for each disease identify the procedures and testing necessary to guard against introducing disease through 'bought-in' cattle.

Procedures for added animals

The risks involved in importing disease are highlighted in the relevant disease sections. The Herdsure® service recommends that all added animals are isolated and tested before joining the herd. Testing and isolation of added animals is mandatory for herds seeking CHeCS accreditation. The only exception to this is where animals are sourced from CHeCS disease-free certificated herds. Refer to the CHeCS technical document for the rules that apply as these may differ for the diseases covered.

Participating in the Herdsure® Cattle Health Improvement Service

Service instructions

Registering the veterinary practice

A simple registration process is in place for registering the veterinary practice onto the service, and it is most likely that this process will have been completed already if you have been issued with this handbook. However, Registration Form A in the 'Forms' section of this handbook is available to register a veterinary practice, should you need to do so.

This is a one-off process. There is no charge for a veterinary practice to register.

The forms included in the 'Forms' section may be photocopied , or you may request a copy by post or electronically by telephoning the Herdsure® helpline on 0300 303 1556. The forms may also be printed from the Herdsure® website at www.ahvlascientific.com. If posting completed forms, these should be sent to the Herdsure® address below:

AHVLA Herdsure® Cattle Health Improvement Service
Animal Health and Veterinary Laboratories Agency
PO Box 653, Newcastle upon Tyne, NE12 2ET

Receiving the registration pack

Upon receipt of completed registration forms for the practice, the following will become available:

- access to the handbook via the Herdsure® pages of the AHVLA Scientific website
- an information pack, which includes an information sheet about the service for you to give to your farmer client prior to enrolling the farm.

Enrolling farms

Registration Form B in the 'Forms' section of this handbook should be completed for each farm and then returned to the address given above. The information provided on this form will allow AHVLA to understand the type of livestock held on a farm, its location and which elements of the Herdsure® service would be of most benefit.

Each enrolled farm will receive a Herdsure® 'Handbook for Farmers', sent to your practice for distribution.

Herds participating in other health programmes may be eligible to join a Herdsure® health programme at Level 2 or 3. Requests for consideration of these cases should be made to a Herdsure® consultant, via the Herdsure® helpline, when an appropriate decision will be made based on veterinary criteria.

Some herds may be considering Cattle Health Certification Standards (CHeCS) accreditation; for enrolling these refer to the CHeCS section.

Farmer subscription

A subscription is payable by farms to enrol, renewed annually, and is based on the size

of herds (<200 and 200 and above) and the number of diseases for which they wish to sign up. Current prices may be viewed by visiting the Herdsure® pages at www.ahvlascientific.com.

Definition of a registered herd

For the purposes of determining subscriptions, and also for herd management within protocols, a registered farm needs to be a single holding with a single County Parish Holding number (CPH). One registration covers all the animals registered on that holding, whether dairy or beef and whether managed within a single herd or multiple herds or groups. The selected protocol(s) for a disease (or number of diseases) will be applied to all the cattle kept under one holding number.

If a herd (that is cattle under one management system) is kept on two separate holdings with separate CPH numbers, both holdings should be registered at the same time. In order that cattle can continue to move between the two (or more) holdings, both holdings should follow the selected disease protocol(s) at the same time. Where two or more herds are under the same ownership but under different management systems and have different CPH numbers, they will need to be registered for Herdsure® separately and will be tracked separately through the disease protocols.

Definitions of cattle classes according to age and type

For the purpose of the Herdsure® service, the following definitions of cattle classes will apply.

Milking cow or dairy cow

An animal that has given birth to at least one calf and is used for the production of milk.

Beef cow, beef suckler cow or suckler cow

An animal that has given birth to at least one calf and is used to suckle the calf or other calves.

Breeding bull

An entire male animal which is over a year old and is, or is intended to be, used for breeding purposes.

Youngstock

All male and all female cattle (up to the birth of their first calf) from birth, regardless of breed or intended purpose. The only exception is breeding bulls over 1 year of age.

Sampling and testing[3]

Sample requests, pre-printed lists of ear tag numbers (see below), pre-printed sampling submission worksheets (see Appendix 1 for an example) and postage-paid Business Reply labels will be issued to correspond with testing regimes.

Important: No samples should be submitted before receiving the first notification.

Submitting samples

Samples should be submitted according to the particular disease protocol and, in most cases, these will be requested by AHVLA (as identified by the symbol in the protocols).

Specific sampling instructions are given in the protocols and in Appendix 3. In addition, sampling kits and detailed sampling instructions will be issued for collection of bulk milk samples. Where possible, these will be sent directly to the holding, allowing your farmer client to collect samples and submit them directly to the laboratory using the Business Reply labels supplied with the kit. The farmer's veterinary practitioner will be notified if this happens.

Sampling requests that require veterinary input, such as blood collection, will be sent to the veterinary practitioner. These sampling requests will include information on what needs to be sampled and, where appropriate, will be accompanied by a pre-printed ear tag list (used to identify animals eligible for testing, based on age and breed).

In every case, the dedicated Herdsure® sampling submission forms, as shown in Appendix 1 and which differ from those issued by AHVLA for other purposes, should be used.

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[3] Tests are carried out under **AHVLA 'Standard Terms and Conditions for Supply of Services'**. These are available on request from your local AHVLA Regional Laboratory, from AHVLA Weybridge or on the AHVLA website at www.defra.gov.uk/ahvla. By submitting a sample to any AHVLA laboratory, customers are deemed to accept these arrangements. Customers will be notified if samples are unsuitable for testing.

No disease or disease-free status *per se* of any animal is conferred, nor should be implied, by any single test result. No value is conveyed, nor should be implied, to any animal(s) to which results relate. Maximum liability to the customer for loss or damage arising from an inaccurate report based on a single test is limited to £1,000. It should be borne in mind that all tests have intrinsic inaccuracies.

In circumstances where a test result may be used as part of an assessment of disease status, a number of repeated tests or test samplings is advisable to minimise the chance of conferring a misleading status to any animal. If any test result is used to establish the disease status of any animal(s), due consideration should be given to seeking independent insurance to cover any implied value or cost that this status or change of status may convey.

Statistical sampling

The Herdsure® protocols sometimes require statistically significant sampling of a subset of animals in each management group. This is done to ensure that the protocols are cost-effective for the farmer. The number of animals to be sampled is based on statistically valid sampling approaches that ensure 95% confidence in the results. Confidence is important in any disease control programme, especially if no positive animals are detected. If too few animals have been sampled and a negative herd result is obtained, a diseased herd could be wrongly classified as negative.

All Herdsure® protocols provide a 95% confidence that the disease is not present in the herd at a level that warrants intervention. The relevant detection level for each disease in each management group of animals was determined by AHVLA disease experts according to their experience and scientific knowledge of each disease. Test experts were consulted on the sensitivity of each test and, based on this information, the sample sizes were calculated by a veterinary epidemiologist for each level in each protocol.

Where statistical sampling is needed, the protocol includes a table that shows exactly how many animals in each separately managed youngstock group should be blood sampled to obtain 95% confidence that at least one sero-positive animal is detected.

How everything works

The supporting system

The service is supported by the in-house Herdsure® Management System (HMS). This is a specially developed IT system, used by AHVLA, which will manage customer records and monitor each farm to ensure that the protocols are followed in a systematic way. This means that veterinary practitioners will have very little record keeping to manage.

About 2 weeks before testing is due, the system will trigger the dispatch of ‘sampling submission worksheets’ (see example in Appendix 1).  These will be sent out with any appropriate consumables and will act as a reminder that the protocol requires specific testing to be carried out. Where appropriate (i.e. except for sampling from small numbers of cattle), these sampling worksheets will be accompanied by Cattle Tracing Scheme (CTS) ear tag lists (see Appendix 2) that identify the eligible cattle for each test, based on age and breed. Any added animals that qualify for testing but are not listed must be appended to the list, for example recently purchased animals. It should be noted that the ear tag lists provided in this way are for assistance only. The accuracy of this information depends on timely provision of herd movements, births and deaths to the British Cattle Movement Service (BCMS).

Identifying and recording cattle against sample numbers

It is important for the practice to use the sampling worksheets provided for sample/animal identification. This will not only assist the practice in selecting appropriate animals but allows AHVLA to print the ear tags in barcode format on the sampling forms against a space for vacutainer labels or other sample identifiers. As handwritten ear tags are not so easily handled at the data entry stage, AHVLA encourages use of its barcoded lists wherever possible to help ensure error-free data entry and subsequent reporting.

Additional testing of cattle from Herdsure® herds

Occasionally, veterinary practitioners responsible for subscribed herds may wish to take samples for testing ***in addition to those specified in the Herdsure® protocols***. Guidance follows for two categories of samples.

1. Sampling for diagnostic purposes from cattle with clinical signs of disease in Herdsure®-subscribed herds

Investigation, sampling and testing for diagnostic purposes is not included within the Herdsure® service. Such samples should be sent to your local AHVLA laboratory, using the 'General Submission Form' (VLA3) available from the AHVLA website at www.defra.gov.uk/ahvla.

NB: In the event that disease is confirmed by this route, please **notify the Herdsure® helpline on 0300 303 1556 without delay**.

2. Unscheduled (mostly unprompted but relevant to the protocol) sampling of cattle from Herdsure®-subscribed herds

Additional sampling (non-triggered) may be required and can be managed and delivered within Herdsure®. Form C in the 'Forms' section of this handbook should be used, and is also available from the website at www.defra.gov.uk/ahvla. Tests requested in this way must be listed on this form and included in the Herdsure® protocol being subscribed to for the animal being tested. Samples under this category should be from animals showing no signs of clinical disease (see point 1 above).

The reason for submission must be identified by ticking the appropriate box on Form C and must fit into one of the following categories:

- testing of a bought-in animal or any animal returning to the farm following a visit to a show or sale or a period of hire (test according to protocol).
- testing of young and new-born calves in the BVD protocol
- retest of a BVD antigen positive animal to confirm persistent infection
- cattle (10-4 weeks before calving) or new calves (<14 days old) requiring a qualifying test in the Neospora protocol

Other categories of submission must be sent to your local AHVLA laboratory using the General Submission Form (VLA 3).

For unscheduled samples, the samples and the completed paperwork should be sent to:

AHVLA Herdsure® Cattle Health Improvement Service
Animal Health and Veterinary Laboratories Agency
PO Box 653, Newcastle upon Tyne, NE12 2ET

As most of the scenarios when you would submit unscheduled samples will not have been triggered by the service, postage-paid Business Reply labels will not have been issued. In this case, please add postage when returning samples. Alternatively, you may request labels for this purpose from the Herdsure® helpline on 0300 303 1556. Please note that postage-paid Business Reply labels should be used for Herdsure® samples only.

Reporting of Herdsure® test results

The results of tests will be returned to the submitting farmer's veterinary practitioner in a report signed by a responsible officer at the AHVLA laboratory, incorporating a standard text for interpretation of results.

At registration, it will be confirmed with the veterinary practitioner by which preferred method reports should be sent – electronically or paper.

If at any time a full history of a farm's test results is required, this may be requested by contacting the Herdsure® helpline on 0300 303 1556.

Telephone advice as part of the service

The testing regimes have been designed to use up-to-date testing technologies and to operate with minimum consultancy. Also, this handbook has been designed so that minimal additional advice should be needed. However, when required, additional advice is available to registered veterinary practices. It is most likely that the type of consultancy required will be specific to a herd/farm and closely related to the protocols.

You may contact AHVLA for veterinary advice relating to Herdsure®-enrolled farms by using the dedicated Herdsure® helpline on 0300 303 1556. The name of the practice, along with the CPH of the participating farm, should be quoted. In this way, the AHVLA veterinary consultant will be able to locate and view the records of the farm/herd concerned and be able to advise appropriately.

Bespoke consultancy

Bespoke consultancy for specific scenarios related to cattle health is also available for an additional cost that would be agreed between a farm/practice and AHVLA. An example of this would be an 'on-farm' consultation. Please make an initial enquiry to the Herdsure® helpline on 0300 303 1556 if you are interested in this service.

Invoicing for Herdsure® services

A monthly invoice for Herdsure® tests and annual subscriptions together with farmer identification for each will be issued to your practice. This is separate to the usual invoice received from AHVLA.

Herdsure® protocol for bovine viral diarrhoea (BVD) in cattle herds

Herdsure® Chapter 1

AHVLA
Animal Health and Veterinary Laboratories Agency

Herdsure®

Contents

Introduction

This protocol describes the process used to establish the disease status for bovine viral diarrhoea (BVD) in cattle and for the control and subsequent monitoring of BVD in those herds.

This protocol is suitable for both dairy and suckler herds.

The three principal elements of this Herdsure® protocol for BVD are:

- sampling and testing to determine status
- sampling and testing to identify and remove persistently infected cattle and, therefore, active infection from the herd
- advice on appropriate measures to reduce the risk of re-introducing BVD virus (BVDV) infection together with sampling and testing to monitor the improved BVD health status of the herd.

BVD is a widespread and significant disease of cattle and other species, manifesting itself in a wide range of clinical signs and syndromes. This protocol does not address the management and treatment of clinical BVD but addresses the control of active BVDV infection.

Since acute infection with BVDV is accompanied by a period of immunosuppression in infected cattle, effective control of BVD is also likely to reduce the risk of other diseases such as viral pneumonia and neonatal diarrhoea.

This protocol refers only to the control of BVDV infection in cattle. If other susceptible animals are present on the farm, especially if they share grazing with cattle, BVD monitoring and, if necessary, control measures should also be applied to these animals. Your AHVLA Regional Laboratory is able to provide appropriate testing and advisory services for this purpose.

The BVD protocol comprises three levels of 'health status':

Level 1	Establishes the BVD status of the herd.
Level 2	Aims to improve the health status of the herd for BVD.
Level 3	Monitors and aims to maintain the improved (or established as satisfactory at Level 1) health status of the herd for BVD.

- All blood samples for Levels 1, 2 and 3 of this BVD protocol should be clearly identified with barcoded labels and referenced to the ear tag number of the animal of origin.
- All samples for BVD testing should be collected in heparinised (green top) blood tubes.
- Tissue samples from calves for BVD virus testing should be collected using appropriate ear tags and sample pots. Please note: BVD tissue samples must be

sent to Herdsure for testing. We are unable to import results from another lab onto the Herdsure Management System.

- Definitions of age and type of cattle referred to in this protocol are explained in this handbook, as is the definition of the 'herd' for registration purposes.

The disease

Introduction

Bovine viral diarrhoea (BVD) is the name used to describe disease caused by infection with bovine viral diarrhoea virus (BVDV), a member of the *pestivirus* group of viruses. The disease is manifested by a number of different syndromes determined by the stage of pregnancy of the infected animal and immune status.

The economic losses resulting from BVDV infection of cattle are considerable but are difficult to quantify because of the complexity and diversity of the consequences of BVDV infections. It is only relatively recently that the escalation of research in the field of *pestivirus* infections has clarified the pathogenesis and epidemiology of the associated diseases.

Impact of the disease

BVDV intrauterine infection is central to the maintenance of disease in cattle herds and avoidance of foetal infection is therefore crucial to the control of disease. If cattle are infected just before breeding, conception rates are reduced. Infection at the time of breeding may lead to early embryonic death; abortion can be the result of intrauterine infection at any stage of pregnancy. However, intrauterine infection during the first third of pregnancy is critical because at this stage the immune system of the foetus has not yet achieved the ability to remove the virus. Furthermore, because of the capability of the virus to evade the developing foetal immune system, it will never achieve the ability to remove the virus. As a result of this the foetus is eventually born with what is termed 'persistent infection'.

It is these persistently infected (PI) cattle, which do not produce antibody against BVDV, that maintain the cycle of active infection. They produce large amounts of virus throughout their lives and, although they may appear to be normal, they often succumb to a fatal systemic disease before reaching maturity. This is known as mucosal disease. However, significant numbers of PI cattle do survive well into maturity. On occasion, particularly where BVDV infection takes place towards the end of the first third of pregnancy or at the beginning of the middle third of pregnancy, calves may show a range of congenital defects at birth. These include cataracts and cerebellar hypoplasia, resulting in locomotor incoordination, as well as growth defects such as domed heads, shortened tails or limbs and changes to the texture of the coat.

PI cattle maintain the cycle of infection when they come into contact with pregnant cattle and the opportunity for the creation of new PI foetuses arises once again.

When non-pregnant cattle of any age are infected by BVDV, the consequences are usually less devastating. A short period of viraemia results and this is termed 'transient' or 'acute' infection. This may go unnoticed but, during this short phase of transient infection, a variable period of leucopaenia results in immuno-compromise of the transiently infected animal. During this period it will have an increased susceptibility to other diseases such as enteric infections or virus pneumonia. Transient infection of bulls may result in reduced fertility with excretion of virus in the semen for a variable period of time. Very occasionally, when young bulls are transiently infected, excretion of BVDV is maintained for life.

Testing for the disease

Detection and removal of PI calves are key to the control and eradication of BVD. AHVLA utilises tests that identify BVD antigen in blood samples. In addition, PCR[4] tests, which identify the genetic material of BVDV in blood, milk and tissue samples, have been developed by AHVLA. Finally, antibody tests are used not only to screen cattle to assess their exposure to BVDV but also as indicators of the possibility of the presence of antibody-negative PI calves. Antibody to BVDV can be measured in both blood and milk samples.

The following tests are used for the Herdsure® BVD protocol:

TC0123	Bulk milk antibody ELISA
TC0390	Individual blood antibody ELISA
TC0772	Blood antigen detection ELISA (Erns)
TC0709	Viral RNA in milk PCR (bulk milk,individual or in pools of up to 10 milks)
TC0655	Viral RNA in blood PCR
TC0758	Viral RNA in pools of up to 10 bloods
TC0872	Erns antigen ELISA in tissue

All blood samples should be taken into heparinised (green top) blood tubes. Collection bottles for bulk milk samples should contain preservative (e.g. bronopol). Tissue samples should be collected using appropriate ear tags and sample pots. Please note: BVD tissue samples must be sent to Herdsure. We are unable to import results from another lab onto the Herdsure Management System.

See Appendix 4 for details of the collection of individual milk samples for PCR examination in pools of 10.

[4] Sometimes called 'molecular photocopying', the polymerase chain reaction (PCR) is an inexpensive and rapid technique used to make many copies of small, specific target areas of DNA or RNA. A single strand of DNA or RNA can be amplified into many millions of strands during a PCR run. Real-time PCR (RT-PCR) allows the accumulated PCR product to be detected in 'real-time' through the use of fluorescent dyes. RT-PCR has the advantage of being highly sensitive and extremely specific and rapid, with the added bonus of the ability to detect more than one organism, strain or type in a single tube.

Biosecurity

Avoiding the introduction of the virus into a 'clean herd'

Control and eradication of BVD is also dependent on meticulous attention to biosecurity principles. The virus can be introduced by various means including via the clothing of personnel or shared machinery. To minimise this risk, herd managers may decide that vaccination of all the breeding cattle on the farm is a necessary measure.

Added cattle are a potentially dangerous source of new infection. The introduction of a PI animal into a herd can have disastrous consequences. Furthermore, a pregnant animal might carry a foetus that proves to be a PI calf when it is tested at birth. The programme outlined in this handbook includes a strategy to allow for the discovery of infected cattle before they are able to infect contact stock.

Principles of biosecurity

- The boundaries of the farm premises must prevent cattle from straying off or onto the farm and must prevent nose-to-nose contact over fences or walls. For herds wishing to undergo or maintain CHeCS accreditation, installation of double fencing with a 3 metre gap between scheme cattle and neighbouring cattle is required.
- Isolation facilities must be used for all added animals and must prevent contact with other stock. A dedicated building separate from other cattle buildings is ideal but a separate paddock that prevents contact with other stock may suffice. No air space, drainage or dung storage may be shared with other cattle. Dung may only be removed from the dedicated storage area, to be spread or added to the main dung store, when all animals in the isolation facility have passed the required health tests. If any of the animals are confirmed as infected, dung from the isolation facility must not be disposed of onto pasture that is to be grazed by cattle within 2 months. Where paddocks have been used for the quarantine of infected animals, other cattle must not be allowed to graze there for at least 2 months.
- A defined isolation period must be observed for all additions to the herd whose health status is known and appropriate testing carried out as specified in the protocol. It is only when both the isolation period and the requisite tests have been completed, with results indicating freedom from infection, that those animals can enter the herd. Additional rules apply to the purchase of bulls and are found in the added animals section.
- Ideally, cattle, sheep and camelids should not graze together unless the disease status of the co-grazers is known.
- Staff who work on other livestock enterprises must use farm-dedicated protective clothing and footwear. Care should be taken to prevent disease transfer via fomites and personnel.
- Colostrum from non-health scheme herds, or from health scheme herds of a lower status, must not be used.
- Delivery and pick-up points should be at a site isolated from other cattle on the farm. Where possible, the driver should remain in the cab and should certainly never assist in removing cattle from pens unless using farm-dedicated protective clothing and footwear.

- Equipment such as drenching guns, surgical instruments and hypodermic needles must not be shared with cattle from another herd. Veterinary surgical instruments must be sterilised before use in the herd.
- Equipment, machinery, livestock trailers and handling facilities that are shared between Herdsure® cattle and other livestock must be cleaned and disinfected before use.
- Where embryo transfer is used, it is important to ensure that embryos are sourced from herds of equivalent or higher health status than the recipient herd.
- Known health status is specific to BVD. Where two or more herds are accredited for different diseases, the rules for movement and contact between herds are those which apply in relation to herds of unknown health status.
- Accredited cattle must not be grazed on pasture previously grazed by non-accredited cattle until a period of 2 months has elapsed. The same grazing restrictions apply to accredited cattle if slurry or manure collected from non-accredited cattle has been used on the pasture. Where herds are seeking CHeCS accreditation the veterinary practitioner will be asked to confirm that the appropriate biosecurity and management measures have been implemented on the farm.

The point at which herds are eligible for CHeCS accreditation is identified in the protocol.

Herd owners and managers who are participating in the Herdsure® BVD service must inform the supervising veterinary practitioner of any changes that could affect herd biosecurity.

Keeping track of your herd's progress in Herdsure®

An **annual herd progress report** will be issued to Herdsure® members. The progress report will detail the level achieved for each protocol for which the herd is enrolled on the date of issue.

For a small fee, an updated progress report can be produced. The updated progress report, like the annual report, will detail the level achieved for each protocol for which the herd is enrolled on the date of issue.

The protocol

Level 1

Level 1: Objective of Herdsure® testing

- Level 1 testing aims to establish whether or not there is evidence of active BVD in a herd.
- If active infection is present it is assumed that 25% of the adult cattle and 10% of the youngstock will have sero-converted. This level will also utilise any herd history and history of relevant test results already held by the veterinary practitioner or by AHVLA.

Level 1: Sampling and testing protocol

Also refer to the flowchart summary of the protocol at the end of Level 3.

1.1	If there is evidence of active BVDV infection in the previous 12 months herds should join at Level 2. Otherwise sampling should proceed as below.
1.2	In dairy herds, a 25ml milk sample should be collected from each bulk milk tank for BVD antibody ELISA testing (TC0123). The collection bottle(s) should contain preservative (e.g. bronopol). Where this result is positive, herds progress to Level 2. Where the result is negative, sampling of youngstock should be carried out as at paragraph 1.5.
1.3	In suckler herds, 10 cows and all the breeding bulls should be blood sampled for BVD antibody (TC390). If any of the samples are positive, herds move to Level 2. Where all the samples are negative, youngstock are sampled as at paragraph 1.5.
1.4	The veterinary practitioner identifies all the management groups of cattle on the farm (excluding the milking cows and the suckler cows) and the number of cattle in each group.*

*A single management group is considered to be a group of cattle grazing the same piece of land, or a group of cattle housed in the same building in such a way as to allow nose-to-nose contact between animals in the group. It should be noted that, where animals share the same air space but where nose-to-nose contact is not possible, the cattle should be considered to be in separate management groups.

1.5	A statistically valid number of blood samples are collected from animals in each management group of 9-18 months of age for BVD antibody ELISA testing (TC0390). The number sampled per group is determined by referring to Table 1 below. The animals sampled should be home reared and non- vaccinated.
1.6	Milk and blood samples are submitted directly to the AHVLA laboratory for testing.

Table 1: Statistical sampling of youngstock according to group size

To be 95% confident that at least one animal will test positive, if at least 10% of the sampled group is carrying antibodies, the following number of cattle should be tested:

Group size	Number of youngstock to be sampled
<10	all
11-12	11
13	12
14-15	13
16-17	14
18-19	15
20–24	16
25–34	20
35–44	21
45–69	23
70–89	24
90–110	25
>110	26

Level 1: Interpretation of results

1.7	Milking herds will be designated as 'no evidence of active BVDV infection' following a negative result for the bulk milk BVD ELISA, together with negative results for BVD antibody of youngstock. Herds in this category have the option to proceed to Level 3 directly; other herds proceed to Level 2. Suckler herds will be designated 'no evidence of BVDV infection' only when all of the blood samples collected from suckler cows, bulls and youngstock show results that are negative for BVD antibody. Herds in this category have the option to proceed to Level 3 directly; other herds should proceed to Level 2. Where herds consist of both milking cows and beef suckler cows, all milk and blood samples should show negative BVD antibody levels to allow these herds to proceed to Level 3; other herds should proceed to Level 2. If only one animal from all the samples tests positive for antibody then a retest may be carried out after 4 weeks. This option should be discussed with a Herdsure® consultant.

Level 2

Level 2: Objective of Herdsure® testing

- Level 2 testing aims to reduce the detrimental influence of active BVDV infection in the herd by identifying the PI cattle that are responsible for the maintenance of the infection. Once identified, these cattle must be removed.

Level 2: Sampling, testing and interpretation protocol

The sampling and testing protocol at Level 2 follows a sequence determined by the results of each test. For this reason, guidance on interpretation of the test results is included within the sampling and testing section below.

Also refer to the flowchart summary of the protocol at the end of Level 3.

2.1	In dairy herds, a bulk milk sample from lactating cows is collected and submitted for PCR testing for BVDV (TC0709). A record of the identities of those cows contributing to this sample should be kept on the herd records. **NB:** As the bulk milk test is validated for a maximum of 300 contributors, care should be taken to ensure that no greater than this number have contributed to the tank from which the sample is taken. Where more than 300 cows have contributed to the tank sample, a Herdsure® consultant will be able to advise on a sampling strategy. **Suckler herds will enter Level 2 sampling at the blood sampling stage see paragraph 2.5 below.**
2.2	Where a positive result is obtained, further individual milk or blood testing of lactating cows will be carried out. Individual samples will be submitted directly to the AHVLA laboratory where batches of 10 samples will be pooled for PCR testing (TC0709 (milk) or TC0758 (blood)) for BVD virus (see Appendix 4 on the collection of individual milk samples). Ear tag numbers should be recorded on the worksheet against individual samples A positive result confirms the presence of viral RNA, indicating at least one BVDV positive animal (PI or acute) among the milking cows. The test does not differentiate between acute and persistent infection.
2.3	On receipt of results for batch milk testing, blood samples should be collected from the individual animals contributing to any positive pools and submitted for BVD antigen ELISA (TC0772). All samples should be collected in heparinised (green top) blood tubes. Where blood testing is carried out as an alternative to batch milk testing individual samples contributing to any positive pools will be forwarded for antigen ELISA (TC0772). If testing fails to identify any positive pools a second bulk milk sample can be submitted for PCR testing(TC0709). This may occur if there is acute infection circulating within the milking herd rather than the presence of a PI cow.

2.4	Animals testing positive for antigen may be considered to be viraemic and culled. However, the veterinary practitioner may wish to retest these animals after 4 weeks to confirm that they are persistently infected. These animals should be isolated pending the confirmatory test. Any animals confirmed to be Persistently Infected must be culled. Where inconclusive antigen results are obtained, samples will be re-tested using TC0655, PCR examination for BVDV in individual blood samples.
2.5	**Animals from suckler herds enter Level 2 at this stage.** All dry cows and cows not contributing to the bulk milk sample, suckler cows, bulls and youngstock over 30 days of age should now be blood-sampled and the samples submitted to the AHVLA laboratory. These samples will be tested in pools of 10 by PCR (TC0758) for BVDV. Individual samples will be retained at the laboratory and the constituent samples from any positive pools will be examined for antigen using the Erns ELISA test (TC0772). All samples should be taken into heparinised (green top) tubes. This test can reliably detect viral antigen in cattle and calves of 30 days of age or older despite the presence of maternally derived immunity. The veterinary practitioner may wish to use the option to retest antigen positive animals to confirm persistent infection. All cattle deemed to be PI must be culled.
2.6	Calves under 30 days of age at the time of total herd testing will not have been tested. These calves may be sampled on reaching 30 days of age to ascertain their BVD status using the antigen ELISA (TC0772). Alternatively the samples can be collected in batches of 10, these samples will be pooled in the laboratory and examined for virus by the PCR test (TC0758). Constituent samples of any PCR positive pools must be re-examined by the antigen ELISA (TC0772) to identify viraemic animals in the group. All calves giving positive results may be considered to be persistently infected and culled. However, before this action is carried out, the option to resample after 4 weeks to confirm persistent infection can be utilised here, with animals confirmed as Persistently Infected being culled. Calves awaiting sampling may be infectious to other susceptible animals and should be isolated where possible. Calves reaching 30 days of age should continue to be monitored (as above) or alternatively tissue samples may be taken from newborn calves and submitted for antigen Erns ELISA testing (TC0872). Calves should be tagged according to the standard animal identification rules. Tissue samples allow calves to be tested from birth thus eliminating the risk of PI calves infecting pregnant cattle whilst awaiting a blood test. Once calves have been tested for a period of 12 months following the removal of any PI cattle from the herd, the herd can proceed to Level 3.

Level 3

Level 3: Objective of Herdsure® testing

- Level 3 testing aims to monitor and maintain the improved health status of the herd. The sampling and testing is designed to alert the veterinary practitioner to the re-introduction of active infection into the herd.

Level 3: Sampling and testing protocol

Also refer to the flowchart summary of the protocol at the end of Level 3.

3.1	Monitoring of continued freedom from active infection should continue after active virus has been removed from the premises. Monitoring of dairy herds will consist of two components: 3-monthly bulk milk antibody testing and annual testing of a statistically significant number of cattle from each management group between 9 and 18 months of age. This test should be carried out 12 months after entry into level 3. The number of animals for sampling is ascertained according to group size as shown in Table 1 at Level 1. Monitoring of suckler herds consists of annual testing of a number of cattle from each management group between 9 and 18 months of age as above.
3.2	In dairy herds, a bulk milk sample should be collected every 3 months and submitted to the AHVLA laboratory for examination for BVD antibody. The first sample is due immediately after the herd enters Level 3. A significant rise* in the antibody titre to BVD in subsequent samples will trigger immediate youngstock monitoring even if the annual sampling is not yet due, see paragraph 3.3 below. In this event, a bulk milk sample from the milking herd should be submitted for BVDV PCR testing (TC0709). If this is positive, sampling and testing as at Level 2 should be carried out. There is no requirement to test cattle previously identified as non-PIs, either by milk or blood testing, provided the results have been recorded on the Herdsure® Management System.
3.3	In all suckler and dairy herds, blood samples from each separately managed group of youngstock between the ages of 9 and 18 months should be collected. Sample sizes for testing are calculated as before. These should be submitted for BVD antibody testing. The first of these tests will be due 12 months after the completion of Level 2 testing. If BVD vaccination is used on the farm, the samples should be collected before these youngstock receive their first dose of vaccine. If any positive samples are identified, sampling and testing as at Level 2 should be carried out. Again, there is no requirement to test cattle previously identified and recorded as non-PIs. Identification numbers of cattle fulfilling these criteria will be provided on the Herdsure® sampling worksheet. This procedure should be repeated annually.

Level 3: Interpretation of results

3.4	Milking herds will be designated as 'no evidence of active BVDV infection' provided there is no significant rise* in bulk milk antibody titre together with negative results for the BVD antibody test of youngstock. Suckler herds will be designated 'no evidence of BVDV infection' only when all of the blood samples collected from youngstock are negative. Where herds consist of milking cows and beef cows, they will be designated as 'no evidence of BVDV infection' only when all of the milk and blood samples fulfil the same criteria as above.

***** *A significant rise is defined as a rise which results in the milk antibody status of the herd being elevated from negative to low/mid or high positive from low positive to mid or high positive or from mid positive to high positive.*

CHeCS Accreditation

Herds entering Level 3 direct from Level 1 will be eligible to apply for BVD CHeCS accreditation following completion of 12 months testing at Level 3, provided all the results are negative.

Herds entering Level 3 following a PI search at Level 2 will be eligible for BVD accreditation status after 24 months of testing at Level 3, provided all results are negative.

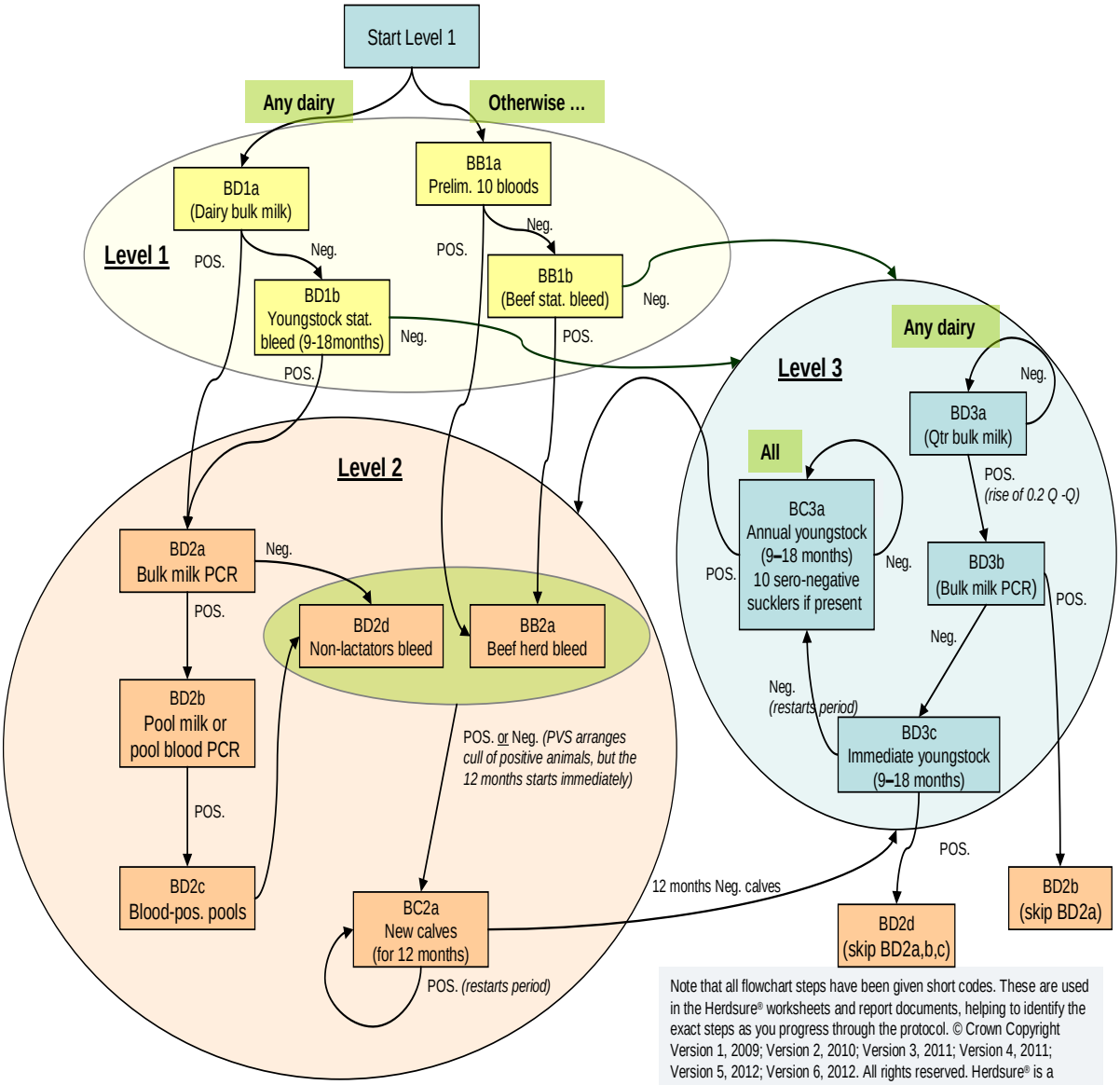
Please note there are additional biosecurity rules for herds undergoing accreditation and stringent requirements for farmer and vet declarations. More information is available on the CHeCS website (www.checs.co.uk).

Key to flowchart summary of the Herdsure® protocol for BVD

Step	Step name	
BD1a	Dairy bulk milk antibody	In dairy herds Level 1 starts with examination of a bulk milk for BVD antibodies (TC0123).
BD1b	Dairy youngstock statistical bleed - 9-18months of age	This step follows a negative result for the bulk milk antibody test. It samples sufficient youngstock to detect BVD (with 95% confidence) if it is on the farm.
BB1a	Preliminary beef bleed	In suckler herds 10 cows and all breeding bulls are sampled to give an initial indication of whether BVD is on the farm.
BB1b	Beef statistical bleed	This step follows a negative result for BB1a. Sufficient youngstock are be sampled to allow BVD to be detected (with 95% confidence) if it is on the farm.
BD2a	Bulk milk PCR	A bulk milk will be tested to determine if there is BVD virus.
BD2b	PCR pooled milk or pooled blood samples	If the bulk milk PCR is positive then it is followed up by individual milk or blood samples. These are pooled at VLA and tested by TC0709 for milk or TC758 for blood.
BD2c	Blood from positive milk pools	Blood samples are requested from animals in positive pools. These will be tested for BVD antigen via TC0772.
BD2d	Non-lactators bleed	All animals over 30 days of age that were not tested via milk testing are now bled and pool-tested via TC0758. Samples in positive pools are individually tested by TC0772.
BB2a	Beef herd bleed	Blood samples from all animals over 30 days of age are tested in pools by PCR TC758; samples from positive pools are individually tested for antigen by TC0772.
BC2a	12-month calf monitoring	Once PI animals are removed from the herd, the farm must send in blood or tissue samples from all new calves for testing for BVD virus. The herd qualifies for Level 3 when a period of 12 months without the disclosure of any PI calves has been achieved.
BD3a	Quarterly bulk milk monitoring	Bulk milks are submitted quarterly for TC0123. A significant increase in antibody levels over time will trigger a further sample for PCR.
BD3b	Bulk milk PCR	This bulk milk PCR is undertaken following positive results from quarterly antibody monitoring.
BD3c	Immediate youngstock testing following bulk milk PCR	This step is triggered by a negative bulk milk PCR at BD3b. It happens immediately, and resets the annual timetable for routine annual youngstock testing (9-18months of age). Presence of PI cows in the milking herd has now been discounted. This step uses sentinel animals to monitor the introduction of virus to the youngstock.
BC3a	Annual testing of youngstock.	This is routine annual statistically significant sampling of animals between 9 and 18 months of age.

Flowchart summary of the Herdsure® protocol for BVD

Bovine viral diarrhoea (BVD)



Note that all flowchart steps have been given short codes. These are used in the Herdsure® worksheets and report documents, helping to identify the exact steps as you progress through the protocol.

Procedures for added animals

Continued freedom from infection will depend on certain biosecurity measures and establishment of the BVD status of any added cattle before they come into contact with the herd.

The Herdsure® service recommends that all added animals are isolated and tested before joining the herd. Testing and isolation of added animals is mandatory for herds seeking CHeCS accreditation. The only exception to this is where animals are sourced from CHeCS disease-free certificated herds. Refer to the CHeCS technical document for the rules that apply.

All classes of cattle should be tested for BVD antigen, ideally prior to leaving the farm of origin or, if not, on entry into isolation. Any antigen positive cattle should not be brought onto the farm.

Non-pregnant cattle

Following a 4 week period of isolation, non-pregnant cattle should be tested for BVD antibody and the results entered onto the Herdsure® database.

If significant numbers of sero-positive milking cows are brought into a herd, a significant rise in the bulk milk antibody readings for the herd will result. This is important in assessing the significance of rising BVD antibody levels in herds that have achieved Level 3 status.

Pregnant cattle

Pregnant cattle that are antigen negative may be brought onto the farm but should be isolated for a period of 4 weeks. Prior to release from isolation, a blood sample for BVD antibody should be collected and the results recorded. If the sample is negative no further action is required. If the antibody results are positive then these cows should remain isolated from the rest of the herd until they calve. A blood sample should be taken from the calf as soon as possible after birth and tested for BVDV by individual PCR (TC0655). Calves showing positive results are PIs and should be removed from the farm immediately.

Breeding Bulls

Bulls that have been transiently infected and, as a result, are antibody positive may excrete virus in semen. This is a rare occurrence but, for this reason, they should not be used for breeding until they have been in on farm for a minimum period of 9 weeks. They should remain in isolation for the first 4 weeks and be blood tested for BVD antibody after this time. There is a documented incidence of prolonged excretion of BVD in semen following transient exposure. If herds wish to exclude this risk, a Herdsure® consultant will be able to advise the practitioner on screening semen for BVDV.

Establishment of a new herd from accredited stock

Where it is intended to establish a BVD-free accredited herd by acquiring cattle accredited free of BVD, the premises must be inspected by the veterinary practitioner before the new stock is introduced in order to ascertain that the biosecurity of the premises and farm boundaries meet the requirements of CHeCS. Accreditation testing for BVD must be carried out no sooner than three months after establishing the herd. Once testing has been completed, with satisfactory results, the herd can be recognised as having achieved accredited status.

Shows and sales

Contact with other stock puts the status of the herd at risk. CHeCS-accredited cattle attending CHeCS-accredited sections may return to their herds of origin without isolation and testing. All other cattle will lose their Herdsure® health status. Consequently, on returning to their herd of origin, they will be subject to the testing and isolation requirements described above. For CHeCS-accredited herds, animals moving off the owner's holding for preparation for sale will lose accredited status if the CHeCS biosecurity rules are not adhered to on the premises where preparation is taking place.

Herdsure® protocol for Johne's disease in cattle herds

Herdsure® Chapter 2

AHVLA
Animal Health and
Veterinary Laboratories
Agency

Contents

Introduction

This protocol describes the process used to establish the disease status for Johne's disease in cattle and for control and monitoring Johne's disease in those herds.

This protocol is suitable for both dairy and beef herds.

The three principal elements of this Herdsure® protocol for Johne's disease are:

- sampling and testing to determine status
- sampling and testing to identify infected cattle with advice on appropriate strategies to reduce new infection rates and so improve the health status of the herd
- sampling and testing to monitor the improved health status of the herd together with advice on appropriate measures to reduce the risk of re-introducing infection.

Johne's disease is an endemic disease of cattle that causes well-documented clinical signs. This protocol does not address the management and treatment of clinical Johne's disease in individual animals but addresses the control of the disease in the herd.

This protocol refers only to the control of Johne's disease in cattle. If other susceptible animals are present on the farm, especially if they share grazing with cattle, Johne's disease monitoring and, if necessary, control measures should also be applied to these animals. Your local AHVLA Regional Laboratory is able to provide appropriate testing and advisory services for this purpose.

The Johne's disease protocol comprises a testing regime to establish herd health status at three different levels:

Level 1	Establishes the Johne's disease status of the herd.
Level 2	Aims to improve the health status of the herd for Johne's disease.
Level 3	Monitors and aims to maintain the improved (or established as satisfactory at Level 1) health status of the herd for Johne's disease.

The disease

Introduction

Johne's disease, or paratuberculosis, is a chronic illness causing progressive loss of condition, diarrhoea and loss of milk production. It is a bacterial infection caused by *Mycobacterium avium* subspecies *paratuberculosis* (MAP). This organism is present in the faeces of infected animals and it can survive for over a year in the environment. It can also be present in colostrum and milk from both clinically and sub-clinically infected animals.

Impact of the disease

Calves can be infected *in utero* and may therefore be born already infected. Young calves are most susceptible to infection and the bacterium is faeco-orally transmitted. Increasing age is associated with increasing natural immunity and so infection during adulthood is considered uncommon. The disease has a long incubation period (years) and animals usually show clinical signs around 3 to 5 years of age. The basic pathology of the disease is progressive change in the intestinal mucosa leading to a protein-losing enteropathy. Infection is most likely to enter a herd by the purchase of clinically normal but infected cattle. Uncontrolled infection in the herd can lead to increasing losses owing to reduced longevity, poor quality cull cows and reduced milk production.

Control of the disease on the farm involves making changes to management practices around calving and the rearing of calves and youngstock. A list of the procedures to prevent new infections appears in the Biosecurity section below. Herdsure® offers a testing protocol to identify infected animals, which can be removed; this will eventually reduce general environmental contamination. If the infection rate in a herd is high, consideration may need to be given to the use of vaccines. This will not remove infection from the herd but may reduce clinical cases to improve longevity and productivity. Vaccines are generally administered to calves less than a month old. Because it is not possible to differentiate between vaccinated and infected animals, vaccinated herds will have to leave the Herdsure® programme.

Sero-positive and infected animals and their offspring should be culled as soon as possible. However, a staged removal may be adopted if a large proportion of the herd is infected. This would involve culling those animals that pose the largest risk to others first. At the same time, any offspring of these animals should be identified; there is a high risk that these animals are infected and therefore may become sero-positive later in life. For this reason it may be wise to remove them from the herd before they reach 2 years of age.

Cattle that have high antibody titres should also be prioritised for culling. Other factors, such as reduced milk production, declining body condition and persistent diarrhoea, as well as the availability of replacements, may also influence prioritisation. Calves born to cattle with high antibody titres should not be kept as replacements but could be reared for beef or culled.

It may be preferable to test individual animals more frequently than recommended in this protocol, in addition to the annual herd test. This would provide information for management of the individual animals at calving and allow farmers to adjust the management of their calves according to the disease status of the dam. Please discuss any additional testing with a Herdsure® consultant via the Herdsure® Helpline. Additional samples should be submitted using Unscheduled Submission Form C.

Testing for the disease

Infection of an animal by MAP produces an antibody response that can be detected in a blood or milk sample. Alternatively, the presence of the organism can be detected in faeces or samples of intestine and associated lymph nodes.

Detection of antibody is by an ELISA test. The sensitivity of this test is highest in an animal displaying clinical signs. Sensitivity is lower when animals are sub-clinically infected.

Following a tuberculin test, animals may produce antibody that may cross-react in the Johne's antibody test to give 'false positives'. To avoid this, it is recommended that blood/milk testing is not carried out within 3 months of a tuberculin test.

When milk samples from cows that have calved in the last 10 days are tested, there is a chance that the high levels of antibodies present could lead to a false positive result. It is therefore recommended that cows up to 10 days post calving are not tested using the milk ELISA.

The organism can be detected in faeces or tissues by culture, which may take up to 8 weeks to give a positive result.

MAP nucleic acid may be detected in the same samples by gene detection using polymerase chain reaction (PCR) technology. This PCR test can produce a result within a few days.

Whole herd faeces screen

Although it is possible to screen a herd by taking individual rectal faeces samples from all animals >2 years old and screening by culture or PCR this is not part of the Herdsure® protocol. If a whole herd faeces screen is being considered, this should be discussed with a Herdsure® consultant.

The following tests are used for the Herdsure® Johne's disease protocol:

TC0366	Serology (serum) – red top tubes
TC0866	Serology (individual milk)
TC0693	PCR (faeces)
TC0713	Liquid culture (faeces)

All blood samples must be collected in red top (plain) blood tubes.

All individual milk samples must consist of at least 10ml of milk collected direct from the cow's teat into an identifiable sample pot containing preservative e.g. bronopol.

All faeces samples must be at least 5g of faeces collected in an identifiable sample pot.

Biosecurity

Biosecurity is the prevention of disease-causing agents entering or leaving any place where farm animals are present (or have been present recently). It is the responsibility of the herd owner and manager in consultation with their veterinary practitioner to ensure good biosecurity of Herdsure® herds. Owners or farm managers must have facilities available for disinfecting vehicles, footwear and clothing.

The following points need to be considered to make the entry of MAP onto a farm less likely.

- It is important that colostrum from a non-Herdsure® herd or a herd of lower status is not used on a Herdsure® farm.
- Natural water sources are a risk for Johne's disease. It is preferable that if non-Herdsure® cattle, sheep or South American Camelids (SACs) have access upstream or the water has passed through another farm then Herdsure® cattle should not have access to it. Piped mains water should be used rather than natural water sources whenever possible.
- Co-grazing with sheep and other ruminants or SACs: Johne's disease also affects other ruminants such as sheep, deer, goats and SACs and the possibility of interspecies transmission is at present unresolved.
- Accredited cattle must not be grazed on pasture previously grazed by non-accredited cattle until a period of 12 months has elapsed. The same grazing restrictions apply to accredited cattle if slurry or manure collected from non-accredited cattle has been used on the pasture.
- Movements of people, vehicles or equipment into areas where the cattle are kept, including fields, farm buildings and other holding areas, should be kept to a minimum.
- People entering the premises to handle the cattle or their products should wear protective clothing and footwear. This must be clean and must be disinfected before and after contact with the cattle; alternatively, disposable protective clothing can be used. Other visitors to the farm should be kept away from direct contact with the cattle.
- Equipment, machinery, livestock trailers and handling facilities that are shared between Herdsure® cattle and other livestock must be cleaned and disinfected before use.
- Other vehicles entering the farm should not come into contact with the areas used by cattle. They should be parked on clean hard-standing away from the cattle. These vehicles must be visibly free of animal excreta, slurry and so on; otherwise entry of the vehicle should be refused.
- Delivery and pick-up points should be at a site isolated from other cattle on the farm. Where possible, the driver should remain in the cab and should certainly never assist in removing cattle from pens unless using farm-dedicated protective clothing and footwear.
- Veterinary equipment such as drenching guns, surgical instruments and hypodermic needles, which may draw blood, must not be shared with cattle from another herd. Veterinary surgical instruments must be sterilised before use in the herd.

- Farm boundaries must prevent cattle from straying off or onto the farm.
- When buying feed and bedding, care must be taken to avoid the risk of introducing infection into the herd. Feed and bedding stores should be protected against access by vermin and wildlife.

Where herds are seeking CHeCS accreditation, the veterinary practitioner will be asked to confirm that the appropriate biosecurity and management measures have been implemented on the farm.

Herd owners and managers who are participating in the Herdsure® service must inform the supervising veterinary practitioner of any changes that could affect herd biosecurity.

You will be sent a risk assessment questionnaire on enrolment, covering biosecurity and the management of all animals, especially the calf and youngstock, to help identify action points to try and reduce the number of new infections.

The following management changes should reduce the occurrence of new infections on infected premises.

- Maintain clean and dry housing for calving with adequate bedding. Ideally, calving enclosures should be disinfected between each calving.
- Ensure udders are clean if suckling takes place.
- Remove calves as soon as possible after birth.
- Do not use pooled colostrum or waste milk for breeding calves. Ideally, feed only milk replacer or pasteurised milk to female and breeding male calves.
- Avoid faecal contamination of milk and colostrum if fed to calves.
- Ensure youngstock do not graze on manure-fertilised pasture.
- Do not mix youngstock with adult stock until they are >12 months old.
- Isolate any cows with possible Johne's disease and test to establish disease status.
- Cull infected animals, see advice above.

Keeping track of progress in Herdsure®

Each registered holding with be issued an **annual herd progress report**. The progress report will detail the level achieved for each protocol for which the herd is enrolled on the date of issue.

For a small fee an additional progress report can be produced. This progress report, like the annual report, will detail the level achieved for each protocol for which the herd is enrolled on the date of issue.

The protocol

Introduction

This service for Johne's disease in dairy and beef herds covers a laboratory testing programme to establish disease status and progression to monitored freedom from disease. Progress involves regular screening and, where appropriate, the managed removal of test-positive animals.

Beef and dairy herds will be tested using the same protocol; milk samples are an alternative to blood samples for dairy cows. On premises where there are both dairy and beef herds, both must join the service; where the protocol refers to 'all animals', this will include both beef and dairy animals.

Summary of objectives

This protocol aims to establish the infection status of the herd for Johne's disease and to monitor with increasing confidence the absence of infection in the herd.

If evidence of infection is found in the herd, Herdsure® offers management advice to reduce the likelihood of new infections and a testing programme to identify infected animals, which can be removed. If infection is satisfactorily removed and the herd undergoes four clear herd tests, the herd would be eligible for CHeCS accreditation. If CHeCS accreditation is to be sought in the future, Form D must be completed at the time of registration.

Level 1

Level 1: Objective of Herdsure® testing

- To establish whether Johne's disease infected animals are present in the herd.

Level 1: Sampling and testing protocol

Also refer to the flowchart summary of the protocol at the end of Level 3.

1.1	Herds with a laboratory-confirmed history of disease e.g. Johne's disease confirmed by serology, culture or post mortem and histopathology, should progress to Level 2b without testing at Level 1; otherwise herds should proceed as below.
1.2	In herds with no confirmed history of Johne's disease, blood samples (red top) should be collected from all breeding male and female cattle over 2 years of age. Individual milk samples (with preservative e.g. bronopol) are an alternative for dairy cows. It is acceptable to submit a combination of milk and blood samples from a single herd. It is assumed that, where fattening cattle over 2 years of age are present, they will be kept entirely separately from the dairy or suckler herds. Where this is not the case please contact a Herdsure® consultant before commencing sampling.
1.3	Blood and/or individual milk samples are submitted to AHVLA (using the Herdsure® Business Reply labels) for testing using TC0366 or TC0866. Results will be interpreted and reported as either negative or positive.

Level 1: Interpretation of results

1.4	There are two outcomes from Level 1 testing: **(a)** All tested animals give a negative test result. These herds can then progress directly to Level 2a. **(b)** The herd test detects one or more sero-positive animals. Optionally, these animals can be further investigated (see below). If not, the herd progresses by testing at Level 2b. *Where small numbers of positives (<2% of the cattle eligible for testing) are identified in homebred animals in herds with no previous evidence of infection you may want to confirm these results by faecal PCR to eliminate the possibility of cross-reaction. This should be discussed with a Herdsure® consultant.* *While awaiting confirmation of test results, sero-positive animals should be isolated from the herd.*

Level 2

Level 2: Objective of Herdsure® testing

- To reduce the detrimental influence of Johne's disease in the herd.

Level 2: Sampling, testing and interpretation protocol

Herds enter Level 2 by two routes:

- 'clear test at Level 1'

or

- 'known history of Johne's disease' or 'sero-positive animals at Level 1'.

Also refer to the flowchart summary of the protocol at the end of Level 3.

Level 2a: Clear test at Level 1	
2.1.1	These herds will confirm their Johne's disease status by a 'final qualifying test' at Level 2a, 12 months after the clear test at Level 1.
2.1.2	Blood and/or individual milk samples from all adult animals are submitted directly to AHVLA laboratory (using the Herdsure® Business Reply labels) for testing using TC0366 or TC0866.
2.1.3	There are two outcomes from Level 2a testing: **(a)** All tested animals give a negative test result. These herds will progress to Level 3, 'monitored freedom'. Provided biosecurity measures are met and appropriate monitoring is carried out at Level 3, herds reaching this point qualify for CHeCS-accredited status. **(b)** The herd test discloses one or more sero-positive animals. These animals must be kept isolated until further testing has been carried out: **(i)** A sample of faeces will be examined for MAP by culture/PCR. **(ii)** If the animal is slaughtered, samples of ileo-caecal junction and associated lymph node should be examined for MAP by culture/PCR or histology. If an animal gives a positive result to any of these tests, this confirms the presence of MAP/Johne's disease in the herd. Further progression is by testing at Level 2b. If an animal gives a negative result, the case must be discussed with a Herdsure® consultant via the Helpline. If the animal in question is homebred, has had no previous positive serology results and there is no history of Johne's disease in the herd, it may be possible for the herd to progress to Level 3. If this further testing is not carried out, animals giving positive results will be considered to be reactors by default and further progression through the Johne's protocol will be via the level 2b route.

Level 2b: Known history of Johne's disease or 'sero-positive animals at Level 1'	
2.2.1	The second route of entry to Level 2 is for those herds with a known history of Johne's disease on the farm and those herds with positive test results at Level 1. These herds enter at Level 2b, the 'disease reduction' phase. Owing to the risk of younger infected animals sero-converting later in life, these herds must have three consecutive negative annual full adult herd tests before entering Level 3.
2.2.2	At each annual test there will be two possible outcomes: **(a)** All the results are negative; these herds remain at Level 2b until they have achieved three consecutive clear herd tests 12 months apart before progressing to Level 3. Provided biosecurity measures are met and appropriate monitoring is carried out at Level 3, herds achieving three consecutive clear tests qualify for CHeCS-accredited status. **(b)** If any of the samples tested are positive, then the herd returns to a repeat annual test at the Level 2b 'disease reduction' phase. As these herds have prior evidence of Johne's disease on the premises there is no requirement or eligibility to confirm infection using a confirmatory test.
2.2.3	Animals giving a positive blood or individual milk result are identified as sero-positive and infected and should be culled as soon as possible. If the animal is pregnant, a decision should be made about the timing of its removal. If the animal is allowed to calve before removal, the calf should be removed from the herd and not reared as a replacement. For options for managing infected animals, see the section on 'Biosecurity'.
2.2.4	In herds with few sero-positive animals (<5%), a test and cull policy may be the most cost-effective way to achieve freedom from disease. However, this is only effective if management practices are also changed to eliminate risks of transmission and re-introduction of disease (as outlined in the section on 'Biosecurity'). Although the Level 2 testing protocol uses annual adult herd blood or milk sampling, additional tests can be submitted at any time to clarify the status of an individual animal. These samples should be submitted using Unscheduled Submission Form C. For herds with a high sero-prevalence, where immediate culling of all the positive animals is not economically feasible, a 'staged' removal of sero-positive cattle and their offspring (as described in the section on 'Impact of the disease') may be employed. In some instances, vaccination may be considered. Because it is not possible to differentiate between vaccinated and infected animals, vaccinated herds will have to leave the Herdsure® programme. The veterinary practitioner and farmer client should be aware that it may take a number of years to successfully eliminate Johne's disease from infected herds.

2.2.5	**Clinical disease** Any animals over 6 months of age showing clinical signs that may suggest the presence of Johne's disease (i.e. weight loss, diarrhoea or milk drop) should be isolated and examined by the attending veterinary practitioner. If Johne's disease cannot be ruled out, samples of blood and faeces should be taken and submitted to the nearest AHVLA Regional Laboratory as routine diagnostic samples. Animals that die before blood or faeces samples are taken should be examined *post mortem* and samples of ileo-caecal junction and associated lymph node should be examined for MAP by culture/PCR or histology. Positive results from cases of possible clinical disease (diagnostic testing) must be reported to the Herdsure® administrator by the veterinary practitioner.

Level 3

Level 3: Objective of Herdsure® testing

- To monitor and maintain the improved health status of the herd.
- Infected animals can start to shed MAP without showing signs of Johne's disease. To increase confidence, the disease-free status the herd must be monitored for a number of years at Level 3. Every year a herd is 'monitored negative' increases the degree of confidence that infection has been controlled. The Herdsure® Management System will maintain records to reflect progress.

Level 3: Sampling, testing and interpretation protocol

Also refer to the flowchart summary of the protocol at the end of Level 3.

3.1	The Year 1 test at Level 3 is due 12 months after the qualifying test at Level 2.
3.2	Blood and/or individual milk samples from all adult animals are submitted directly to the AHVLA laboratory (using the Herdsure® Business Reply labels) for testing by TC0366 or TC0866. This testing protocol will be repeated in Year 3 of Level 3.
3.3	There are two outcomes from Level 3 Year 1 testing: **(a) All tested animals give a negative test result**. Provided biosecurity measures are met and appropriate monitoring is carried out, herds qualify for CHeCS-accredited status. **(b) The herd test finds one or more sero-positive animals**. Positive animals must be kept isolated until further testing has been carried out on them: **(i)** A faeces sample examined for MAP by culture/PCR. **(ii)** If the animal is sent for slaughter, samples of ileo-caecal junction and associated lymph node should be examined for MAP by culture/PCR or histology. If an animal gives a positive result to any of these tests, the herd status reverts to Level 2b. If an animal gives a negative result by culture/PCR the case must be discussed with a Herdsure® consultant via the Helpline. If the animal in question is homebred, has had no previous serology results and there is no history of Johne's disease in the herd, the herd may be able to remain at Level 3.

3.4 All purchased incoming animals should be blood or individually milk sampled annually and samples tested by TC0366 or TC0866 until four clear tests have been achieved.

3.5	All animals over 2 years of age will be retested every 12 months. Any animal with positive results must be kept isolated until further testing (see paragraph **3.3** above) has been carried out. If the annual herd test reveals Johne's positive animals then the herd will move to Herdsure® Level 2 and lose its accreditation status.
3.6	Once a herd has reached Level 3 and had two further consecutive, clear annual herd tests there is an option to test homebred cattle biennially, rather than annually.

CHeCS accreditation

Accredited free status: A herd is Accredited Free if **three** clear qualifying tests (full herd tests) at an interval of 12 months have been achieved without any reactor being detected. Please note that under CHeCS rules, milk samples submitted towards CHeCS accreditation must be collected by the milk recorder, veterinary practitioner or other practice employee e.g. nurse or paraprofessional. A herd will be eligible for accreditation either following a full herd test after 12 months at Level 3 via Level 2a or upon entry at Level 3 via Level 2b. The date the herd first achieved Accredited Free status will be included on the Certificate of Accreditation. Should a herd lose status and then regain it having met the subsequent testing requirements the date on the certificate will be when accreditation was regained. More information is available on the CHeCS website (www.checs.co.uk).

CHeCS Health Plan – new for 2012/2013

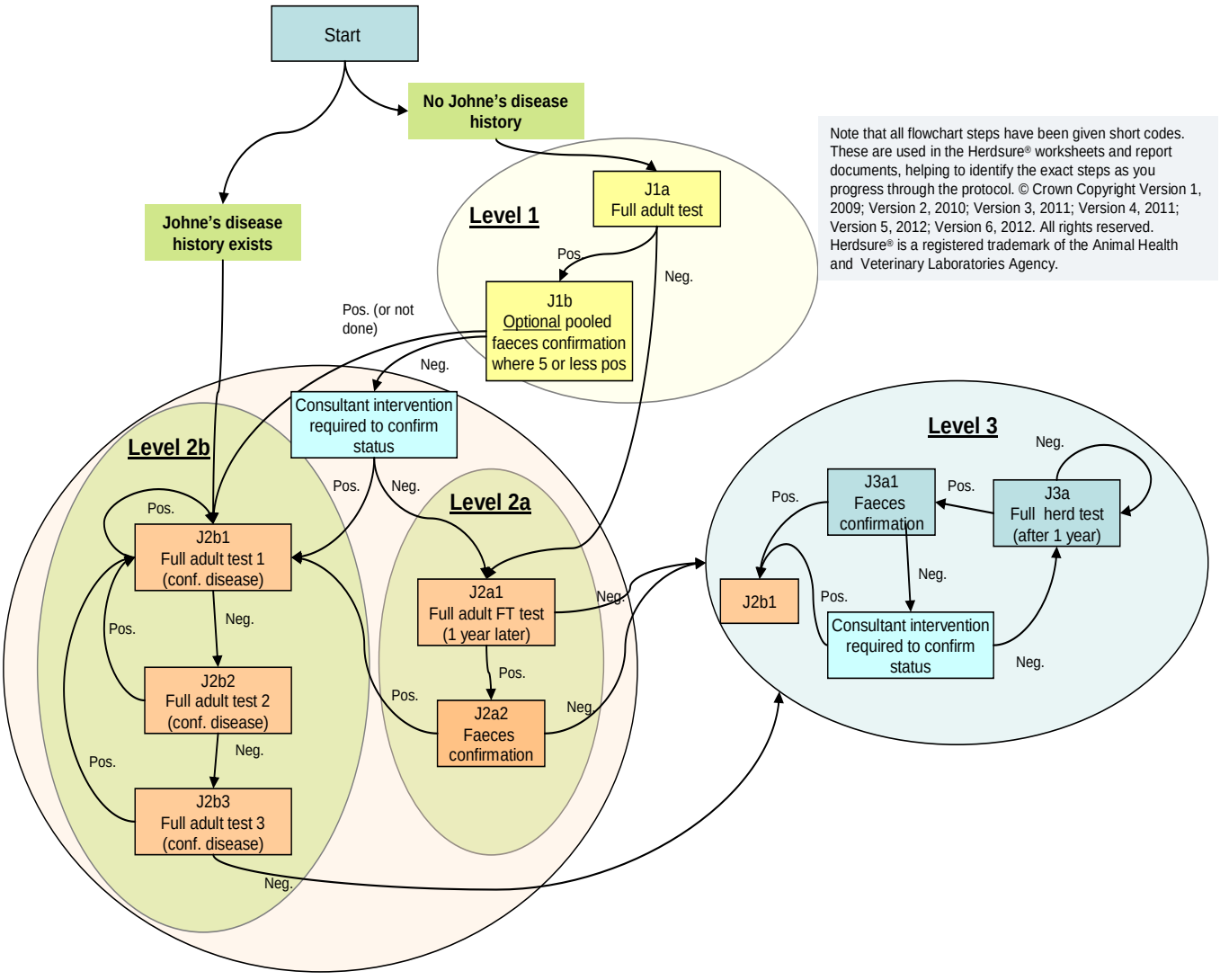
For members working towards CHeCS accreditation a health plan covering Johne's disease must be in place. It must be updated annually and signed off by both the vet and herd owner/manager. On joining the Accreditation Programme the health plan must be submitted to Herdsure® within 2 months of carrying out the herd test. Thereafter the health plan (covering the last year) must be submitted with the herd test. The health plan must cover the three mandatory control elements detailed in the CHeCS Technical Document: 3.1 Antibody positive animals, 3.2 Cull all reactors and 3.3 Offspring of female test positive animals. The veterinary practitioner must detail in writing why any particular guideline has not been followed. For further information, visit www.checs.co.uk

Key to flowchart summary of the Herdsure® protocol for Johne's disease

Step	Step name	
J1a	Full adult test	All cows/bulls >2 years of age are sampled and tested using TC0366 or TC0866. Ear tag details are provided.
J1b	Optional faeces confirmation	Where five or fewer positives are disclosed at the full herd test these may be confirmed via faeces using PCR TC0693.
J2a1	Full adult test (fast-track)	This full test can qualify the farm for entry to Level 3. All cattle >2 years of age are tested using TC0366 or TC0866. Ear tag details are provided.
J2a2	Faeces confirmation	Positives from the fast-track test can be retested using faeces. Samples are tested with PCR TC0693 in pools of up to 5. The ear tag details of positive animals will be provided.
J2b1	Full adult test No.1	The first annual test in Level 2b. All cattle >2 years are tested using TC0366 or TC0866. Ear tag details are provided.
J2b2	Full adult test No.2	The second annual test in Level 2b. All cattle >2 years are tested using TC0366 or TC0866. Ear tag details are provided.
J2b3	Full adult test No.3	The third annual test in Level 2b. All cattle >2 years are tested using TC0366 or TC0866. Ear tag details are provided.
J3a	Full adult test No.1 at Level 3	The first annual test in Level 3. All cattle >2 years are tested using TC0366 or TC0866. Ear tag details are provided. This is carried out 12 months after completion of Level 2 testing and entry into Level 3.
J3a1	Faeces confirmation (test No.1)	Positives from a full test are tested using faeces. Samples are tested with PCR TC0693 in pools of up to 5. The ear tag details of positive animals will be provided.

Flowchart summary of the Herdsure® protocol for Johne's disease

Johne's disease



Note that all flowchart steps have been given short codes. These are used in the Herdsure® worksheets and report documents, helping to identify the exact steps as you progress through the protocol.

Procedures for added animals

The Herdsure® service recommends that all added animals are isolated and tested before joining the herd. Testing and isolation of added animals is mandatory for herds seeking CHeCS accreditation. The only exception to this is where animals are sourced from CHeCS disease-free certificated herds. Refer to the CHeCS technical document for the rules that apply.

Sourcing new animals

New animals could be infected but still test negative and thereby introduce infection into the herd. This risk can be reduced by purchasing from a herd of similar or higher herd health status.

If introducing animals of any age from a herd with unknown health status is necessary, the animals should be tested by blood and faeces sampling on the farm of origin. If an animal or source herd is found to be positive, animals must not enter the herd. If testing at the farm of origin is not possible, new animals should be held in isolation[5] on entry to the herd and tested by both blood and faeces sampling. The animals should not enter the herd unless both tests are negative. If groups of animals from different herds of origin are tested and one or more from any group tests positive, none of that group can enter the herd without loss of the herd's status. Such animals should be held in isolation until the results of all tests are known. If infection is confirmed, the building(s) should be thoroughly cleansed and disinfected and the manure held for at least 12 months before it is spread on to pasture. If the infected animals were at pasture, the field(s) should not be grazed for at least 12 months.

On entry to the herd, added animals should be tested by TC0366 (blood) or TC0866 (individual milk) annually on reaching 2 years of age until four consecutive clear tests have been achieved. This may include the pre-entry test if the animal was 2 or more years of age at the time of sampling.

Establishment of a new herd from accredited stock

Where it is intended to establish a Johne's-free accredited herd by acquiring cattle accredited free of Johne's, the premises must be inspected by the veterinary practitioner before the new stock is introduced in order to ascertain that the biosecurity of the premises and farm boundaries meet the requirements of CHeCS. Accreditation testing for Johne's must be carried out no sooner than three months after establishing the herd. Once testing has been completed, with satisfactory results, the herd can be recognised as having achieved accredited status.

Shows and sales

Due to the absence of a test with potential for detecting infection soon after possible exposure to MAP, no testing is required when show animals return to the herd. The risk of infection is considered highest in calves under 6 months of age and so these animals

[5] An isolation facility that prevents contact with other stock must be provided for all added animals. A dedicated building separate from other cattle buildings is ideal, but a separate paddock that prevents contact with other stock may suffice. No air space, drainage or dung storage may be shared with other cattle. Dung may only be removed from the dedicated storage area, to be spread on land or added to the main dung store, when all cattle in the isolation facility have passed all the required health tests and have been added to the herd.

must be tested for four consecutive years once they are 2 years of age.

For CHeCS-accredited herds, the CHeCS rules must be followed:

If cattle from an accredited Johne's disease-free herd leave their home premises for fewer than 7 days and are prevented from direct contact with other animals' faeces and soiled bedding, these can rejoin the herd without the need for isolation and testing.

Herdsure® protocol for liver fluke infection in cattle herds

Herdsure® Chapter 3

AHVLA
Animal Health and Veterinary Laboratories Agency

Herdsure®

Contents

Introduction

This protocol describes the processes used to determine the liver fluke status of a cattle herd and for control and subsequent monitoring of liver fluke status in the herd.

This protocol is suitable for both dairy and beef herds.

The three principal elements of this Herdsure® protocol for liver fluke are:

- sampling and testing to determine status
- advice on appropriate treatments, if any, following test results
- advice on appropriate management measures, if any, to reduce the risk of liver fluke infection following test results.

The protocol addresses only the measurement and control of *sub-clinical* liver fluke infection in cattle herds. It does ***not*** address any aspect of *clinical* liver fluke infection (fasciolosis). Fasciolosis is a significant disease of cattle and other animals which requires veterinary intervention, not provided by Herdsure®, for diagnosis and treatment. Effective control of *sub-clinical* liver fluke infection in cattle herds is likely to reduce the risk of clinical fasciolosis.

This protocol refers only to the control of liver fluke infection in cattle. If other susceptible animals are present on the farm, especially if they share grazing with cattle, liver fluke monitoring and, if necessary, control measures should also be applied to these animals. Your AHVLA Regional Laboratory is able to provide appropriate testing and advisory services for this purpose.

The liver fluke protocol comprises three levels of 'health status':

Level 1	Establishes the liver fluke infection status of the herd.
Level 2	Aims to reduce the burden of liver fluke in the herd to a level at which it does not significantly affect cattle production or welfare.
Level 3	Monitors and aims to maintain the improved (or established as satisfactory at Level 1) health status of the herd for liver fluke infection.

The disease

Introduction

The liver fluke *Fasciola hepatica* is a trematode parasite that infects grazing animals including cattle, sheep, deer, camelid species, rabbits and, occasionally, humans. The parasite causes liver damage by migration of immature larvae and blood loss by the feeding activities of the adult parasite. The term for diseases caused by the liver fluke is 'fasciolosis'.

The parasite has been endemic in the wetter western half of the UK for centuries. Over the past 15 years there has been significant spread to other parts of the UK, including the east of Scotland and East Anglia (Mitchell, 2002; Pritchard *et al*, 2005). This spread has been made possible by:

- the greater movement of infected animals, particularly sheep, to farms that operate poor biosecurity
- changing weather patterns, with increased rainfall and milder winters throughout all of the UK, which benefit the lifecycle of the parasite and its intermediate snail host.

The liver fluke is now considered to be endemic in all regions of the UK.

Eradication of the liver fluke is difficult because wildlife reservoirs can maintain infection on many farms. The objective, therefore, is to reduce infection to a manageable level in order that:

- the economic benefits of control significantly outweigh the cost of control measures
- animal welfare is safeguarded.

Impact of the disease

The parasite can cause significant economic loss, with one estimate in 2001 for the economic loss worldwide put at $200 million (Mulcahy and Dalton, 2001). Another study of the financial loss in Switzerland in 2005 was estimated as €299 per infected animal (Schweitzer *et al*, 2005). A more recent study involving bulk tank milk samples estimated the loss of production in dairy cows as a decrease in annual average milk yield of 0.7 kg per cow per day and an increase in the mean inter-calving interval of 4.7 days (Charlier *et al*, 2007).

Pathogenesis and clinical signs

Lesions in the definitive host are seen in the liver parenchyma caused by migration of immature fluke and in the bile ducts caused by adult flukes. The parasite releases cathepsin protease enzymes, which cause tissue destruction that facilitates migration and feeding.

In addition, damage to the host's liver and bile ducts is caused by the spines attached to the outer layer of the parasite – the tegument.

Acute fasciolosis occurs when migration of large numbers of immature fluke cause tissue destruction, haemorrhage and inflammation to the liver that can lead to the death of the animal. It is estimated that 10,000 metacercariae ingested over a short period of time are required to cause acute fasciolosis. The symptoms of acute fasciolosis are seen more commonly in sheep and only rarely in cattle because the bovine liver is larger and more able to withstand this challenge.

Chronic fasciolosis is more common in cattle and occurs when small numbers of metacercariae are ingested over several weeks (trickle infection). The lesions are of hepatic and bile duct inflammation and necrosis and, later, liver and bile duct fibrosis. The fibrotic reaction is more pronounced in cattle compared to sheep. The adult fluke's ability to feed can be limited by this fibrosis.

Clinical signs in cattle are associated with blood feeding of the adult parasite and associated hypoalbuminaemia and manifest as weight loss and reduction in milk yield. In severe cases there is anaemia, subcutaneous oedema ('bottle jaw') and recumbency. Chronic fasciolosis is commonly associated with diarrhoea in cattle.

Metabolic disease associated with chronic fasciolosis has also been described (Mason, 2004).

Subclinical liver fluke infection results in poorer growth rates in growing cattle and lowered milk yields in dairy cows, without any other obvious clinical signs.

Testing for the disease

The gold standard for diagnosis of liver fluke infection is necropsy examination and finding the parasite in the liver.

(1) The detection of liver fluke eggs in faeces

A positive result indicates *current* liver fluke infection. The test has a sensitivity of 60%. There are a number of reasons for this. Faecal egg detection cannot confirm immature fluke infection where no eggs are produced. Also, in chronic infections due to non-specific immunity, it is possible that very small numbers of eggs are produced. Finally, liver fluke eggs can accumulate in the gall bladder and may be excreted intermittently.

The composite fluke egg detection test (TC0689) is used in the Herdsure® liver fluke protocol. It is suitable for screening groups of animals for liver fluke infection and requires the collection and subsequent laboratory pooling and testing of 10 faeces samples.

Testing of four such composite samples, requiring the collection of 40 individual faeces samples, is specified in this Herdsure® protocol. This provides 95% confidence of detecting liver fluke eggs in at least one pool if 20% or more of the cattle in the herd are infected and excreting fluke eggs, assuming a test sensitivity of 60%.

Interpretation

The detection of liver fluke eggs in one or more of the four composites is interpreted as indicating significant liver fluke infection in the group.

(2) The detection of antibody to liver fluke in blood

An ELISA (TC0678) is used to detect antibody to liver fluke in blood. This is a means of detecting past exposure to liver fluke infection.

Measurable antibody develops 3 weeks after ingestion of metacercariae and persists for ***at least*** 3 months following infection.

This test is offered within Herdsure® as an alternative to faecal egg detection, as collection of clotted blood samples by a veterinary practitioner may, under certain circumstances, be more convenient than collection of floor faeces samples by the farmer.

Sampling is by collection of blood samples from a random selection of a calculated number of cattle that will provide 95% confidence of a positive result if 20% or more of the cattle in the herd are infected and possess antibody (see Table 2 following the Level 1 protocol).

Interpretation

Results will be presented as positive, negative or inconclusive.

- A result is positive when there is greater than 20% positivity (when compared to a positive control).
- A result is negative when there is less than 15% positivity (when compared to a positive control).
- A result is inconclusive when there is a percentage positivity between 15% and 20% inclusive (when compared to a positive control).

The presence of one or more positive or inconclusive ELISA results is interpreted as indicating significant liver fluke infection in the group.

(3) The detection of antibody to liver fluke in bulk tank milk (dairy herds only)

An ELISA (TC0692) is used to detect antibody to liver fluke in bulk tank milk.

The test result provides an estimate of the likely percentage of cows (that contributed milk to the bulk tank) that possess antibody to liver fluke. For this to be an accurate reflection of the status of the entire milking cow herd, at least 75% of the cows that are currently milking should have contributed to the bulk tank from which the sample was taken.

If a herd has more than one bulk milk tank, each should be sampled and each sample should be submitted for separate testing.

Interpretation

Results will be presented as positive, negative or inconclusive.

- A result is positive when there is greater than 40% positivity (when compared to a positive control).
- A result is negative when there is less than 27% positivity (when compared to a positive control).
- A result is inconclusive when there is a percentage positivity between 27% and 40% inclusive (when compared to a positive control).

A positive or inconclusive result is interpreted as indicating that 25% or more of the cows contributing to the bulk tank possess antibody to liver fluke.

A negative result is interpreted as indicating that less than 25% of the cows contributing to the bulk tank possess antibody to liver fluke.

Biosecurity

Effective control of liver fluke infection requires appropriate biosecurity measures to prevent the introduction, or re-introduction, of fluke infection onto the farm and into the herd.

Ideally, animals that are to be introduced onto the farm from elsewhere should be **either**:

- sampled and tested for antibody to liver fluke, with a 'negative' result required before entry to the farm of destination or release from quarantine (note that a false negative result could arise from ingestion of liver fluke metacercariae by a naïve animal in the 2 weeks before sampling)

or

- treated with a flukicide (that kills both adults and immature liver fluke) 4 weeks before they are moved onto the pastures of the farm of destination. (This allows time for any fluke eggs in the intestinal tract to be expelled before arrival.)

If this is not possible, then animals should be quarantined on arrival on the farm of destination. They should not have access to pastures for 4 weeks after being treated with a flukicide (that kills both adults and immature liver fluke). Faeces should not be put directly onto grazing land because of the risk of seeding pasture with fluke eggs.

Both pre-movement and quarantine treatments may need to be repeated if bought-in animals have been very recently infected.

The risk of introducing infected animals that have fluke that are resistant to triclabendazole should be considered. If there is evidence of lack of efficacy of triclabendazole on the farm of origin, added animals should be given another flukicide before quarantine. The risk of introducing resistant fluke is reduced by not sourcing animals from farms with a history of non-efficacy of triclabendazole or any other flukicide.

Treatment and control

If liver fluke infection is detected by Herdsure® liver fluke Level 1, Level 2 or Level 3 testing, then treatment is indicated.

Treatment strategies should be reviewed by the veterinary practitioner and modified, if necessary, to address local risks and farm management practices. In particular, rainfall and temperature have a significant effect on the risk of animals acquiring liver fluke infection. The risk of liver fluke infection is likely to increase significantly if there is high rainfall in summer and this may warrant further treatments in addition to those recommended below.

As stated above, fasciolosis is potentially a very severe disease and it is the duty of the person responsible for the care of the cattle on the farm to obtain and implement veterinary advice, beyond the scope of this protocol.

Which animals should be treated?

All animals that are considered to have been at risk of infection from the grazing of metacercariae at pasture should be treated.

When should treatments be given?

For non-lactating cattle

The minimum number and timing of recommended treatments are:

- Treatment 1: in winter, as soon as possible after the receipt of a positive Herdsure® liver fluke result.
- Treatment 2: in the subsequent May, following the receipt of a positive Herdsure® liver fluke result.
- Treatment 3: in the subsequent autumn or early winter (possibly at housing) following the receipt of a positive Herdsure® liver fluke result.

In years in which weather (rainfall and temperature) and possibly other local factors indicate a high risk of infection, additional treatments may have to be given.

For lactating cattle

In most dairy herds it is likely that treatment will be given at the time of ‘drying-off’ in order to comply with manufacturers’ instructions and to avoid lengthy periods of milk withdrawal. A local assessment of risk should be made to determine if more immediate treatment of milking cows is necessary.

What type of flukicide should be used?

- Treatment 1: a flukicide effective against both adult and immature liver fluke.
- Treatment 2: for cattle that are housed over the winter, a flukicide effective against adult liver fluke; for cattle that graze pastures that may be contaminated with liver fluke metacercariae, a flukicide effective against both adult and immature liver fluke.
- Treatment 3: a flukicide effective against both adult and immature liver fluke.

See the following websites for further information about flukicide treatments:
www.noahcompendium.co.uk
www.norbrook.co.uk

Mixed grazing

On mixed cattle and sheep farms where fluke infection has been diagnosed, treatment of sheep is advised. Please contact your AHVLA Regional Laboratory if you require advice on the treatment or control of liver fluke infection in sheep. Other susceptible animals on the farm, such as goats and camelids, should also be treated.

Other control measures

It is advisable to keep lactating cattle on the drier, less infective pastures, particularly in the autumn because of difficulties in treating the dairy herd. Wetter areas of the farm, or around ponds and streams, can be fenced off. Drainage of land is largely discouraged now, particularly in regions that are designated environmentally sensitive areas. For the same environmental considerations, molluscicides are not a realistic means of control.

Resistance to triclabendazole

Resistance to triclabendazole has been reported in liver fluke in sheep in the UK, the Netherlands and Australia. This has implications for the treatment of cattle.

Forecasting fasciolosis

The following factors increase the risk of infection:

- warmer and wetter weather
- encouragement of wetland habitats and other environmentally sensitive schemes
- increased movement of sheep across the UK that introduce infection to farms, if biosecurity measures are not implemented on arrival
- greater survival of metacercariae on pasture from the previous year because of milder winters, and so the metacercariae would be present to infect grazing animals newly turned out to grass
- greater survival of fluke eggs over the winter so that they are present to infect snails in the spring, thus beginning the lifecycle
- preferential grazing of cattle in wet habitats favoured by the intermediate snail hosts.

The most important of these factors are increased rainfall and warmer temperatures (Ollerenshaw, 1974).

Monthly and quarterly disease surveillance reports produced by AHVLA will highlight periods of high risk of liver fluke infection. See http://www.defra.gov.uk/vla/reports/rep_surv.htm.

Keeping track of your herd's progress in Herdsure®

An **annual herd progress report** will be issued to Herdsure® members. The progress report will detail the level achieved for each protocol for which the herd is enrolled on the date of issue.

For a small fee, an updated progress report can be produced. The updated progress report, like the annual report, will detail the level achieved for each protocol for which the herd is enrolled on the date of issue.

References

Charlier, J., Duchateau, L., Claerebout, E., Williams, D. and Vercruysse, J. (2007) Associations between anti-fasciola hepatica antibody levels in bulk tank milk samples and production parameters in dairy herds. *Preventive Veterinary Medicine,* 78, 57–66.

Mason, C. (2004) Fasciolosis associated with metabolic disease in a dairy herd, and its effects on health and productivity. *Cattle Practice,* 12, 7–13.

Mitchell, G. (2002) Update on fasciolosis in cattle and sheep. *In Practice,* 24, 378–385.

Mulcahy, G. and Dalton, J.P. (2001) Cathepsin L proteinases as vaccines against infection with *Fasciola hepatica* (liver fluke) in ruminants. *Research in Veterinary Science,* 70, 83–86.

Ollerenshaw, C.B. (1974) Forecasting liver fluke disease. *British Society of Parasitology,* 42, 32–52.

Pritchard, G.C., Forbes, A.B., Williams, D.J.L., Salimi-Bejestani, M.R. and Daniel, R.G. (2005) Emergence of fasciolosis in cattle in East Anglia. *Veterinary Record*, Nov. 2005, 157, 19, 578–582.

Schweitzer, G., Braun, U., Deplazes, P. and Torgerson, P. (2005) Estimating the financial losses due to bovine fasciolosis in Switzerland. *Veterinary Record*, Aug. 2005, 157, 188–193.

The protocol

Level 1

Level 1: Objective of Herdsure® testing

- Level 1 establishes the liver fluke infection status of the herd.

Level 1: Sampling protocol

Also refer to the flowchart summary of the protocol at the end of Level 3.

1.1	**When to sample:** ▪ in November, December or January **and, for dairy herds:** ▪ when at least 75% of the milking cows are contributing milk to the bulk tank. (If fewer than 75% of cows are contributing milk to the bulk tank in November, December or January, contact the Herdsure® helpline for guidance.)
1.2	**What to sample:** ▪ the main cow herd: the lactating cows in a dairy herd and the main group of suckler cows in a beef herd **and** ▪ the other 'at risk' cattle (see paragraph 1.3).
1.3	**Definition of 'at risk' cattle group** **The 'at risk' groups for sampling should *include*:** ▪ cattle that have grazed pastures of the subscribed farm **and** experienced an interval of at least 12 weeks between the earliest exposure to pasture (in the preceding grazing season) and sampling. **The 'at risk' group for sampling should *exclude*:** ▪ the main cow herd (sampled separately) ▪ cattle that have been treated for liver fluke infection within the 3 months before sampling ▪ bought-in cattle, wherever possible (contact the Herdsure® helpline if further guidance is necessary) cattle with a significantly low risk of having acquired liver fluke infection for any other reason (for example, animals that have grazed pastures of likely low risk of harbouring fluke-infected snails)

	▪ cattle that have co-grazed with the main cow herd and that are thought to have the same or lower risk of having acquired liver fluke as the adult cows (for example, suckler calves). **In a *dairy* herd,** the 'at risk' group could, therefore, include dry cows and other cows not contributing to the bulk tank, replacement heifers, grazing youngstock and calves and bulls that have had access to pasture. **In a *beef* herd,** the 'at risk' group could, therefore, include cows managed separately from the main suckler herd, heifer replacements, beef youngstock and weaned calves and bulls that have had access to pasture.
1.4	**Farms with a dairy component** **(1)** A milk sample should be collected from the bulk tank (see Appendix 2). **(2)** In addition, the 'at risk' group (see paragraph 1.3) should be sampled by **either**: ▪ collection of 40 floor faeces samples (see Appendix 2) **or** ▪ collection of clotted blood samples (red top) from a statistically valid number of cattle (see Table 2 below). Additionally, if the cattle on a single subscribed farm are subdivided so that they exclusively graze two or more distinct and separate areas of land then they should be sampled as if they are two separate herds. For example, if a farm keeps a group of cattle on one set of pastures and an entirely different group of cattle on an entirely different set of pastures, each group should be sampled separately.
1.5	**Farms with no dairy component** **(1)** If there is no dairy component in the herd, then the main herd of suckler cows should be sampled by **either**: ▪ collection of 40 floor faeces samples (see Appendix 2) **or** ▪ collection of clotted blood samples (red top) from a statistically valid number of cattle (see Table 2 below). **(2)** In addition, if there is also an 'at risk' group (see paragraph 1.3), it should be sampled by **either**: ▪ collection of 40 floor faeces samples (see Appendix 2) **or** ▪ collection of clotted blood samples (red top) from a representative number of cattle (see Table 2 below). Additionally, if the cattle on a single subscribed farm are subdivided so that they exclusively graze two or more distinct and separate areas of land then they should be sampled as if they are two separate herds. For example, if a farm keeps a group of cattle on one set of pastures and an entirely different group of cattle on an entirely different set of pastures, each

	group should be sampled separately.

1.6	All samples should be submitted in the same parcel with the paperwork provided (in advance) to AHVLA using the Herdsure® Business Reply labels.

Table 2: Statistical-based sampling according to group size

Number of cows in group	Number of cows to test from group
1–10	All
11–15	10
16–30	12
31–50	13
51–190	14
>190	15

Assumptions

- It is expected that, if liver fluke is present in the herd, at least 20% of cattle in a group will possess antibody.
- Cattle that are at a significantly lower risk of having acquired liver fluke on the farm (e.g. animals that have not grazed or that have been treated) are excluded from the group to be sampled.
- All animals for testing should be selected at random.
- Sero-positivity is relatively independent of age, stage in lactation cycle and seasonality.

Level 1: Testing protocol and interpretation of results

1.7	Clotted blood samples will be tested by ELISA (TC0678) for antibody to liver fluke. Results will be presented as positive, negative or inconclusive. ***Interpretation*** ▪ A serum sample with percentage positivity of greater than 20% (of a positive control) is regarded as positive. ▪ A serum sample with percentage positivity of between 15% and 20% (of a positive control) is regarded as inconclusive. ▪ If one or more of the samples from the group are recorded as positive or inconclusive for antibody, the herd will be classed as significantly infected.
1.8	Faeces samples will be pooled into four composites of 10 samples each. The composite faeces fluke egg detection test (TC0689) will be used to determine the presence of fluke eggs in each of four composite samples. Results will be presented as positive or negative. ***Interpretation*** ▪ Detection of one or more fluke eggs in a composite sample (of 10 individual samples) is regarded as positive. ▪ If one or more of the four composites are found to be positive for fluke eggs, the herd will be classed as significantly infected.
1.9	Bulk milk will be tested by ELISA (TC0692) for antibody to liver fluke. Results will be presented as positive, negative or inconclusive. ***Interpretation*** ▪ A bulk milk sample with percentage positivity greater than 40% (of a positive control) is regarded as positive. ▪ A bulk milk sample with percentage positivity between 27% and 40% (of a positive control) is regarded as inconclusive. ▪ A positive or inconclusive result indicates that 25% or more of the cows contributing to the bulk tank sample possess antibody to liver fluke.

What happens when Herdsure® Level 1 testing has been completed?

This depends on the results of testing.

- If one or more of the test results for the farm are positive or inconclusive, the herd enters Level 2 for a 12-month period.
- If all of the test results for the farm are negative, the herd enters Level 3.

Level 2

Level 2: Objective of Herdsure® testing

- Level 2 aims to reduce the burden of liver fluke in the herd to a level at which it does not significantly affect cattle production or welfare.

Level 2: Treatment and continued monitoring

Entry to Level 2 is by:

- detection of liver fluke infection in the herd by the Herdsure® Level 1 protocol

or

- demonstration of fasciolosis by another means (such as clinical diagnostic testing, necropsy or by meat inspection).

Level 2 of the Herdsure® liver fluke protocol spans a period of 12 months.

The veterinary practitioner should use the advice in this handbook and relevant information elsewhere to devise an appropriate 12-month treatment and control programme for liver fluke infection on the farm.

A herd progress report is issued after 12 months and the herd either remains at Level 2 or proceeds to Level 3.

Liver fluke infection in the herd should be treated according to the guidance given above in the section on 'Treatment and control'.

Management actions to reduce the risk of acquisition of liver fluke infection should be implemented according to the guidance given above in the section on 'Treatment and control – other control measures'.

Biosecurity actions to reduce the risk of the introduction of liver fluke infection should be implemented according to guidance given above in the section on 'Biosecurity'.

Level 2: Sampling protocol

Also refer to the flowchart summary of the protocol at the end of Level 3.

2.1	**When to sample** Continued monitoring for liver fluke infection is achieved by sampling: ▪ in November, December or January **and, for dairy herds:** ▪ when at least 75% of the milking cows are contributing milk to the bulk tank. (If fewer than 75% of cows are contributing milk to the bulk tank in November, December and January, contact the Herdsure® helpline for guidance.)

2.2 **What to sample**

Sample collection is the same as in Level 1, namely:

Farms with a dairy component

(1) A milk sample should be collected from the bulk tank (see Appendix 2).

(2) In addition, the 'at risk' group (some or all of the remainder of the herd: dry cows, suckler cows, bulls and young-stock (see paragraph 1.3)) should also be sampled by **either**:

- collection of 40 floor faeces samples (see Appendix 2)

  or

- collection of clotted blood samples (red top) from a representative number of cattle (see Table 2 given in the Level 1 protocol).

Farms with no dairy component

(1) If there is no dairy component in the herd, then the main herd of suckler cows should be sampled by **either**:

- collection of 40 floor faeces samples (see Appendix 2)

  or

- collection of clotted blood samples (red top) from a representative number of cattle (see Table 2 given in the Level 1 protocol).

(2) In addition, the 'at risk' group (some or all of the remainder of the herd: dry cows, suckler cows, bulls and young-stock (see paragraph 1.3)) should also be sampled by **either**:

- collection of 40 floor faeces samples (see Appendix 2)

  or

- collection of clotted blood samples (red top) from a representative number of cattle (see Table 2 given in the Level 1 protocol).

Additionally, if the cattle on a single subscribed farm are subdivided so that they exclusively graze two or more distinct and separate areas of land then they should be sampled as if they are two separate herds. For example, if a farm keeps a group of cattle on one set of pastures and an entirely different group of cattle on an entirely different set of pastures, each group should be sampled separately.

2.3 

Sampling reminders will be sent by AHVLA each November.

What happens after the herd has been within the Herdsure® Level 2 testing, treatment and management protocol for 12 months?

This depends on the results of testing:

- If one or more of the test results for the farm are positive or inconclusive (see paragraphs 1.7, 1.8 and 1.9), the herd re-enters Level 2 for a further 12-month period. The liver fluke treatment and control programme is reviewed by the veterinary practitioner and the farmer and improved, if necessary.
- If all of the test results for the farm are negative, the herd enters Level 3.

Level 3

Level 3: Objective of Herdsure® testing

- Level 3 monitors and maintains the improved, or otherwise satisfactory, liver fluke status of the herd.

Level 3: Continued monitoring

Entry to Level 3 is by:

- all test results being negative following sampling according to the Herdsure® liver fluke Level 1 protocol

or

- all test results being negative following treatment, management and sampling according to the Herdsure® liver fluke Level 2 protocol.

Level 3 of the Herdsure® liver fluke protocol spans a period of 12 months.

A herd progress report is issued after 12 months and the herd either remains at Level 3 or reverts to Level 2.

Management actions to reduce the risk of the introduction of liver fluke infection should be implemented according to the guidance given above in the section on 'Treatment and control – other control measures'.

Level 3: Sampling protocol

Also refer to the flowchart summary of the protocol at the end of Level 3.

3.1	**When to sample** Continued monitoring for liver fluke infection is achieved by sampling: ▪ in November, December or January **and, for dairy herds:** ▪ when at least 75% of the milking cows are contributing milk to the bulk tank. (If fewer than 75% of cows are contributing milk to the bulk tank in November, December and January, contact the Herdsure® helpline for guidance.)

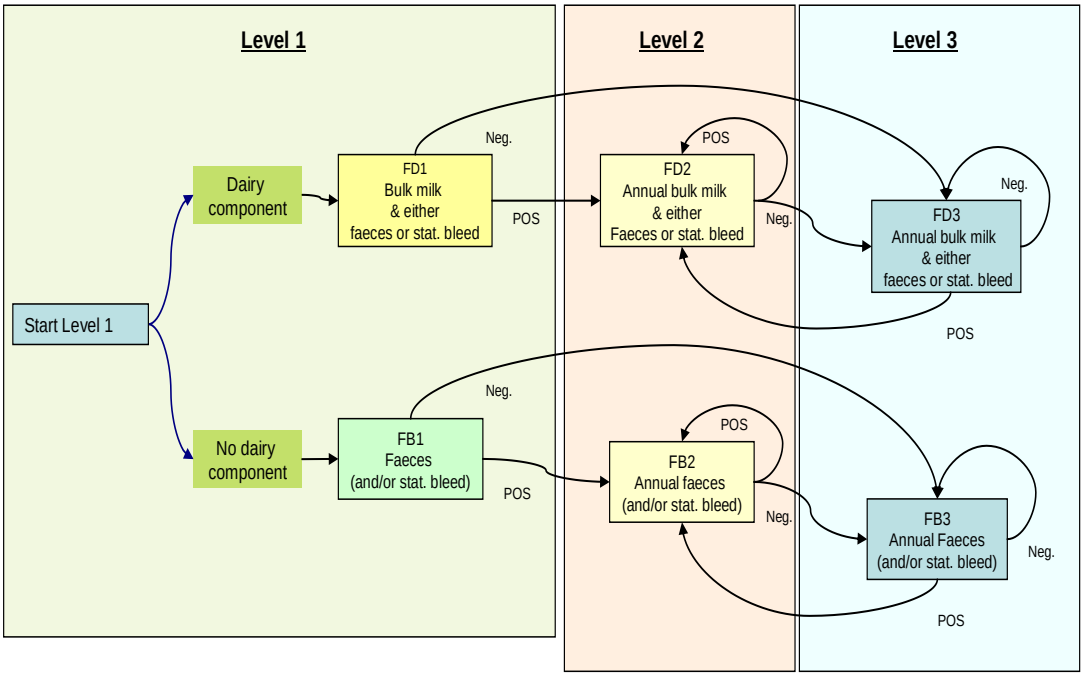
3.2	**What to sample** Sample collection is the same as in Level 1, namely: **Farms with a dairy component** **(1)** A milk sample should be collected from the bulk tank (see Appendix 2). **(2)** In addition, the 'at risk' group (some or all of the remainder of the herd: dry cows, suckler cows, bulls and youngstock (see paragraph 1.3)) should also be sampled by **either**: ■ collection of 40 floor faeces samples (see Appendix 2) **or** ■ collection of clotted blood samples (red top) from a representative number of cattle (see Table 2 given in the Level 1 protocol). **Farms with no dairy component** **(1)** If there is no dairy component in the herd, then the main herd of suckler cows should be sampled by **either**: ■ collection of 40 floor faeces samples (see Appendix 2) **or** ■ collection of clotted blood samples (red top) from a representative number of cattle (see Table 2 given in the Level 1 protocol). **(2)** In addition, the 'at risk' group (some or all of the remainder of the herd: dry cows, suckler cows, bulls and youngstock (see paragraph 1.3)) should also be sampled by **either**: ■ collection of 40 floor faeces samples (see Appendix 2) **or** ■ collection of clotted blood samples (red top) from a representative number of cattle (see Table 2 given in the Level 1 protocol). Additionally, if the cattle on a single subscribed farm are subdivided so that they exclusively graze two or more distinct and separate areas of land then they should be sampled as if they are two separate herds. For example, if a farm keeps a group of cattle on one set of pastures and an entirely different group of cattle on an entirely different set of pastures, each group should be sampled separately.
3.3	Sampling reminders will be sent by AHVLA each November.

Key to flowchart summary of the Herdsure® protocol for liver fluke infection

Step	Step name	
FD1	Initial sampling for herds with a dairy component	Bulk milk sample for liver fluke antibody ELISA (TC0692) for dairy herds together with a representative number of blood samples for fluke ELISA antibody testing (TC0678), or 40 faeces samples for fluke egg identification (TC0689) from 'at risk' cattle.
FB1	Initial sampling for beef herds	Blood samples from representative cattle for fluke ELISA antibody testing (TC0678), or 40 faeces samples for fluke egg identification (TC0689) from suckler cows and 'at risk' cattle. Blood samples from one group and faeces samples from the other, or the same sample type for each group are acceptable.
FD2	Annual sampling for herds with a dairy component	Bulk milk sample for liver fluke antibody ELISA (TC0692) for dairy herds together with a representative number of blood samples for fluke ELISA antibody testing (TC0678), or 40 faeces samples for fluke egg identification (TC0689) from 'at risk' cattle.
FB2	Annual sampling for beef herds	Blood samples from representative cattle for fluke ELISA antibody testing (TC0678), or 40 faeces samples for fluke egg identification (TC0689) from suckler cows and 'at risk' cattle. Blood samples from one group and faeces samples from the other, or the same sample type for each group are acceptable.
FD3	Annual sampling for herds with a dairy component	Bulk milk sample for liver fluke antibody ELISA (TC0692) for dairy herds together with a representative number of blood samples for fluke ELISA antibody testing (TC0678), or 40 faeces samples for fluke egg identification (TC0689) from 'at risk' cattle.
FB3	Annual sampling for beef herds	Blood samples from representative cattle for fluke ELISA antibody testing (TC0678), or 40 faeces samples for fluke egg identification (TC0689) from suckler cows and 'at risk' cattle. Blood samples from one group and faeces samples from the other, or the same sample type for each group are acceptable.

Flowchart summary of the Herdsure® protocol for liver fluke infection

Liver fluke



Note that all flowchart steps have been given short codes. These are used in the Herdsure® worksheets and report documents, helping to identify the exact steps as you progress through the protocol.

Herdsure® protocol for Leptospirosis in cattle herds

Herdsure® Chapter 4

AHVLA
Animal Health and Veterinary Laboratories Agency

Herdsure®

Contents

Introduction

This protocol describes the process used to establish the disease status for leptospirosis in cattle and for the control and subsequent monitoring of leptospirosis in those herds.

The protocol is suitable for both dairy and beef herds.

Leptospira Hardjo is the cause of a significant disease of cattle (and to a lesser extent, sheep) resulting in loss of milk yield/milk drop, infertility and abortion. It is also an important zoonosis. Cattle and other animal species can also acquire infection with *L.* Icterohaemorrhagiae (also zoonotic) resulting in systemic illness, including abortions. *L.* Icterohaemorrhagiae infection is not included in the Herdsure® protocol so any references to leptospirosis hereafter relate only to *L.* Hardjo in cattle. Your AHVLA Regional Laboratory is able to provide appropriate advice on the significance of *L.* Hardjo infection in other species if necessary.

The leptospirosis protocol comprises three levels of 'health status':

Level 1	Establishes the leptospirosis status of the herd.
Level 2	Aims to improve the health status of the herd for leptospirosis.
Level 3	Monitors and aims to maintain the improved (or established as satisfactory at Level 1) health status of the herd for leptospirosis.

The disease

Introduction

In Great Britain, leptospirosis in cattle invariably refers to serovar Hardjo as conventionally identified by serology (MAT or ELISA). With recent advances in molecular technology, Hardjo has been shown to comprise two distinct genomospecies, namely *Leptospira interrogans* serovar Hardjo (serovar Hardjo prajitno) and *Leptospira borgpetersenii* serovar Hardjo (serovar Hardjo bovis). It is currently unclear which of these predominates in Great Britain since serological tests are insufficiently sensitive to definitively distinguish between them. They both cause similar disease and the means to control them are essentially the same. Hence, for the purposes of this handbook, both will be referred to generically as Hardjo.

Impact of the disease

Leptospirosis in cattle causes infertility, milk drop, abortion and the birth of weak and unviable calves. Leptospirosis is an important zoonosis: infection in man occurs through the skin and mucous membranes and results in pyrexia and 'flu-like' symptoms. The disease can be severe and occasionally fatal.

Following infection in cattle the organism localises in the kidneys and the urinary tract. Urine is the main source of infection for other cattle but the organism can also localise in the reproductive tract (leading to venereal transmission) and be excreted in semen, uterine discharges and the products of abortion. Infected animals can excrete the organism for long periods (months to years) and the organism may continue to be excreted after infected cattle have become sero-negative. Sheep may act as asymptomatic carriers and have been identified as a potential source of infection for cattle.

Sources of infection

Cattle are the principal host and, unlike other serovars such as Icterohaemorrhagiae (which is carried by rats), wildlife are not involved in the transmission cycle of Hardjo.

The major sources of infection for cattle are:

- infected urine – infection can be introduced into herds through purchased cattle or hired/shared bulls
- contaminated/shared drinking water courses
- sheep.

Disease syndromes associated with *L.* Hardjo infection

Abortion

This usually occurs in the second half of pregnancy. Infection in late gestation may result in the birth of weak or unviable calves.

When infection is endemic in a herd, the only sign of disease may be abortion in newly introduced unvaccinated or naïve heifers.

Milk drop

This may affect a large proportion of the herd simultaneously. Some affected cows are pyrexic, some may show a 'flabby' udder affecting all four quarters. There can be an associated high somatic milk cell count.

The classical acute milk drop syndrome is now uncommon in Great Britain. However, reduced herd milk yield may occur as a result of sub-clinical cases in naïve animals in endemically infected herds.

Infertility

Infertility can be significant in the first year following infection and is characterised by a low conception rate to first service, an increased calving to first service interval and an increase in the number of services for each successful pregnancy.

It may also typically be seen in heifers in infected herds that stop vaccinating.

Diagnosis

Serology is used to provide evidence of exposure to *L*. Hardjo infection.

Several serological tests are available but, for monitoring purposes within Herdsure®, an ELISA is used to detect antibody in serum and in milk samples:

TC0125	ELISA test for *L.* Hardjo antibodies in bulk milk samples
TC0638	ELISA test for *L.* Hardjo antibodies in individual serum samples

ELISA measures IgG; titres are not detectable until 3 or 4 weeks post infection and persist for 2 or 3 years.

The sensitivity of the ELISA is approximately 90%.

Control of leptospirosis

Whole-herd vaccination according to the manufacturer's instructions is the main control and preventative measure for *L.* Hardjo in cattle. Vaccination in young calves can prevent reproductive tract colonisation and later infertility, while vaccinating heifers before first calving prevents abortion.

Owing to the zoonotic potential of leptospirosis, vaccination of the herd should be seriously considered if infection is demonstrated.

Vaccination of infected cattle does not prevent shedding of *L*. Hardjo. The use of antibiotics has been used to clear infection from sero-positive animals.

Attempting to eliminate infection from endemically infected herds in cattle-dense areas is not usually possible unless all known risk factors can be rigorously controlled. In herds in which there is a very low (<5%) sero-prevalence, elimination of infection may be considered especially if there are likely to be benefits that offset the costs (e.g. trading status). Removing sero-positives ('test and cull' approach) might be considered if there are apparent reasons for some cattle being sero-positive, such as a history of part-herd vaccination or exposure of a particular age cohort to a known risk factor (e.g. away-grazed heifers). Where this strategy is adopted, strict adherence to biosecurity measures and monitoring of status must be undertaken to ensure infection does not re-enter the herd.

Further details of monitoring and control of leptospirosis are described within the protocol.

Testing for the disease

- All blood samples for Levels 1, 2 and 3 of the leptospirosis protocol should be clearly identified and referenced to the ear tag number of the animal of origin.
- All blood samples for *L.* Hardjo antibody testing should be collected in plain (red top) blood tubes.
- Definitions of age and type of cattle referred to in this protocol are explained in the introductory section of this handbook along with the definition of the 'herd' for registration purposes.

When cattle in the herd are vaccinated against *L.* Hardjo they produce an antibody response, which cannot be distinguished from the antibody produced in response to field infection. Therefore, for the purposes of this protocol, vaccinated cattle cannot be considered free of infection.

Bulk milk *Leptospira* antibody test – TC0125

The bulk milk *L.* Hardjo ELISA test is very sensitive and detects antibody to both *L.* Hardjo-bovis (the main pathogenic species) and the closely related serovars *L.* Hardjo-prajitno and *L.* Saxkoebing. However, this is not usually significant when used as a screening test.

The test result is expressed as an OD ratio against a positive control serum.

Interpretation

Category	OD ratio	Interpretation – bulk milk serology
Negative	<0.07	Naïve/unvaccinated
Low positive	0.07–0.40	A few sero-positive cows but **active infection unlikely**
Mid positive	0.40–0.70	Situation less clear.
High positive	>0.70	Heavily infected/vaccinated

Blood *Leptospira* antibody test – TC0638

The *L*. Hardjo ELISA is highly sensitive and the test result is expressed as an OD ratio against a positive control serum.

Interpretation

For the Herdsure® leptospirosis protocol, where the ELISA OD ratio is equal to or greater than 0.20, the result is positive.

Where the ELISA OD ratio is less than 0.20, the result is negative.

Keeping track of progress in Herdsure®

Each registered holding will be issued an **annual herd progress report**. The progress report will detail the level achieved for each protocol for which the herd is enrolled on the date of issue.

For a small fee an additional progress report can be produced. This progress report, like the annual report, will detail the level achieved for each protocol for which the herd is enrolled on the date of issue.

Biosecurity

It is the responsibility of the herd manager, in consultation with their veterinary practitioner, to ensure good biosecurity in Herdsure® herds.

The following potential means of introduction of leptospirosis into herds should be addressed and the risk kept to a minimum.

- **Movements of people, vehicles or equipment** into areas where the cattle are kept, including fields, farm buildings and other holding areas, should be kept to a minimum.
- **Persons entering premises** to handle the cattle (or their products) should wear protective clothing and footwear. This must be clean and disinfected before and after contact with the cattle. Alternatively, disposable protective clothing can be used. Other visitors to the farm should be kept away from direct contact with the cattle.
- **Shared farm equipment:** Equipment, machinery, livestock trailers and handling facilities that are used on herds of unknown leptospirosis status must be cleaned and disinfected before use by herds subscribed to Levels 2 and 3 of Herdsure® leptospirosis.
- **Other vehicles** entering the farm should not come into contact with the areas used by cattle unless they have been thoroughly cleaned and disinfected.
- **Delivery and pick-up points** should be at a site isolated from other cattle on the farm. Drivers should remain in their cabs and should not assist in removing cattle from pens unless using farm-dedicated protective clothing and footwear.
- **Veterinary equipment** such as drenching guns, surgical instruments and hypodermic needles, which may draw blood, must not be shared with cattle from another herd. Veterinary surgical instruments must be sterilised before use in the herd.
- **Farm boundaries** must prevent cattle straying off the farm and neighbouring cattle straying onto the farm.
- **Contact with cattle of different health status:** Cattle herds subscribed to Levels 2 and 3 of Herdsure® leptospirosis must not come into contact with cattle from herds that are not of an equal or higher Herdsure® leptospirosis status, otherwise they will lose their status. To re-introduce them to the herd, they must be regarded as added animals (see requirements for added animals at the end of the protocol section).
- **Added cattle:** See the section on ‘added cattle’ at the end of the protocol.

- **Isolation facility:** An isolation facility that prevents contact with other stock must be provided for all added animals. A dedicated building separate from other cattle buildings is required, although a separate paddock that prevents contact with other livestock may suffice. The drainage or dung storage area should not be shared with other cattle. Dung should only be spread on land or added to the main dung store when all cattle in the isolation facility have passed all the required health tests and have been added to the herd. Where cattle are confirmed as reactors, dung must not be disposed of onto pasture that is to be grazed by cattle within 12 months.

- **Isolation period:** All cattle entering the herd must be isolated for 28 days. Appropriate testing should be carried out (see section on buying in cattle at the end of the protocol). It is only when both the isolation period and the requisite tests have been completed, with negative results, that those cattle can enter the herd.

- **Co-grazing with sheep or other domestic ruminants or camelids:** Although not a mandatory requirement, it is strongly recommended that, wherever possible, cattle and sheep do not graze together. It is also recommended that cattle do not co-graze with other domestic ruminants and camelids.

- **CHeCS accreditation**: there must be a 2 month interval before accredited cattle follow non-accredited cattle, sheep or other potentially infected animals (other domestic ruminants or camelids) onto pasture The same grazing restrictions apply to accredited cattle if slurry or manure collected from non-accredited cattle has been used on the pasture.

- **Feed and bedding:** When buying feed and bedding, care must be taken to avoid the risk of introducing infection into the herd. Feed and bedding stores should be protected against access by vermin and wildlife.

- **Water:** Piped mains water should be used rather than natural water sources whenever possible. Water sources which arise from neighbouring land could be contaminated by infected urine and therefore should not be used.

- **Notification:** Herd owners and managers who are participating in Herdsure® must inform the supervising veterinary practitioner of any changes that could affect herd biosecurity.

Where herds are seeking CHeCS accreditation the veterinary practitioner will be asked to confirm that the appropriate biosecurity and management measures have been implemented on the farm.

The protocol

Level 1

Level 1: Objective of Herdsure® testing

- Level 1 testing aims to establish whether or not there is evidence of *L.* Hardjo infection in a herd. Level 1 also utilises any herd history and relevant test results already held by the veterinary practitioner or by AHVLA.

If infection is present, it is assumed that at least 10% of the cattle will have sero-converted to *L*. Hardjo.

Level 1: Sampling, testing and interpretation protocol

Also refer to the flowchart summary of the protocol at the end of Level 3.

1.1	Where laboratory test results indicate evidence of leptospirosis in the herd in the previous 12 months then the herd should enter at Level 2.
1.2	Differentiation between vaccinal antibody and field infection antibody is not possible so vaccinated herds are addressed in the same way as infected herds for the purposes of this protocol and will therefore start at Level 2.
1.3	For herds with no vaccination history or where relevant test results are not available, samples will be collected according to the instructions described below.
1.4	Herds with milking cows A bulk milk tank sample should be submitted for antibody testing under test code TC0125. Avoid sampling when fewer than 75% of the cows are lactating. The collection bottle should contain preservative (e.g. Bronopol). Results will be reported as either negative or positive with the OD ratio included in the report (refer to the table in the disease section above for additional information on interpretation). ▪ Herds receiving a positive result proceed directly to Level 2. ▪ Herds receiving a negative result proceed to paragraph 1.5 if they contain suckler cows or paragraph 1.6 if they do not.

1.5	Herds with any suckler cows or suckler cows only Blood samples will be collected from a statistically significant sample of the suckler cows and submitted for testing by ELISA. Please refer to Table 3 below to calculate the number of cows to be tested. Sample reference numbers should be recorded against ear tag numbers for the cows submitted. Results will be reported as negative or positive. ■ For positive results the herd proceeds directly to Level 2. ■ For negative results, regardless of herd composition, the next step is to carry out blood sampling of a statistically significant number of youngstock as at paragraph 1.6.
1.6	All herds that are negative to this point need to have a statistically significant blood sampling of all youngstock over 1 year of age that are intended for introduction to the adult herd. If youngstock are present in more than one management group (see below for definition) then each group must be sampled separately. At this time, individual blood samples are also required from all breeding bulls. All samples will be tested by ELISA. The number of animals to be sampled is determined by referring to Table 3 below. Results will be reported as negative or positive.
1.7	When the tests defined in paragraphs 1.4, 1.5 and 1.6 have been carried out, the results will enable a decision to be made as to whether the herd follows route **2a** or route **2b** at Level 2. ■ A herd will be designated as **sero-positive** at Level 1 if any bulk milk ELISA or blood sample is positive. Bulk milk interpretation is shown earlier in the disease section above. ■ Herds will be designated as **sero-negative** if all samples tested are negative.

Statistically significant sampling of management groups

A single management group is considered to be a group of cattle grazing the same piece of land.

If the cattle are housed then each separate group should be considered as a management group even if the separate groups share the same air space.

Further guidance, if required, is available from a Herdsure® consultant.

The number of animals to test is dependent on the group size and ensures a 95% confidence in finding at least one sero-positive animal with a test sensitivity of 90%, if 10% of the animals are positive for antibody.

Table 3: Statistical-based sampling according to group size

Number of suckler cows/youngstock in management group	Number of cows to test in group
<10	10
20	19
30	24
40	28
50	31
70	34
100	38
150	40
200	42
300	43
500	45
800	45

*For values that fall between those in the table use the next highest figure in the table or all the animals in the group, whichever is lower.

Level 2

Level 2: Objective of Herdsure® testing

- Level 2 testing aims to improve the health status of the herd for leptospirosis by reducing the detrimental influence of *L.* Hardjo infection in the herd. This is done by identifying the cattle that are responsible for the maintenance of the infection. Once identified, these cattle may be removed or they may be retained and a disease reduction strategy applied.

Level 2: Sampling, testing and interpretation protocol

The sampling and testing protocol at Level 2 follows a sequence determined by the results of each test. For this reason, guidance on interpretation of the test results is included within the sampling and testing section below.

Also refer to the flowchart summary of the protocol at the end of Level 3.

2.1	There are two routes available through Level 2: ▪ **Level 2a** aims to establish an entirely serologically negative herd – the 'sero-negative route'. ▪ **Level 2b** aims to maintain and monitor the immunity of the herd – the 'sero-positive' or 'maintained immunity route'. The farmer and veterinary practitioner should decide which route is the most appropriate for the herd and inform the Herdsure® helpline so that appropriate reminders can be offered as Level 2 progresses.
2.2	A comprehensive biosecurity policy should be adopted, based on advice provided in the disease section, and reviewed by the farmer and veterinary practitioner annually.
Level 2a: sero-negative route	
2.3	All breeding animals aged 1 year or older must be blood sampled and the samples examined by ELISA. Serologically positive animals must be removed from the herd. Further blood sampling at intervals of no less than 6 months and no more than 12 months should be carried out until no sero-positive animals are detected.

2.4	The herd will remain at Level 2a until two consecutive clear herd tests are recorded. The herd can then proceed to Level 3. The herd also qualifies for CHeCS accreditation on reaching this point.
2.5	Any animal that aborts or shows ill health, where Leptospirosis cannot be excluded on clinical grounds, should be isolated. Blood samples should be collected on first examination, and again 28 days later, and tested for antibody using TC0638. Any confirmed or suspected cases of leptospirosis should be discussed with a Herdsure® consultant. (Diagnostic testing is not done within Herdsure® – see the introductory section on 'Additional testing of cattle from Herdsure® herds'.)
2.6	Leptospirosis is a serious zoonotic disease so introduction of infection into a naïve or sero-negative herd is of particular concern. Therefore, in addition to the action at paragraph 2.5, the revelation of further sero-positive cows in an unvaccinated herd during the course of testing should be regarded as an indication of active infection. In this event a decision should be made whether to proceed with the sero-negative route of Level 2a or to change to Level 2b, i.e. vaccinate and monitor the herd. Any requirement to change from Level 2a to Level 2b or vice versa should be notified to the Herdsure® consultant by telephoning the Herdsure® helpline.
Level 2b: sero-positive or maintained immunity route	
2.7	Level 2b requires implementation of a control and monitoring policy for leptospirosis. Advice on control of the disease has been given earlier in the disease section of this protocol.
2.8	Any animal that aborts or shows ill health, where Leptospirosis cannot be excluded on clinical grounds, should be isolated. Blood samples should be collected on first examination, and again 28 days later, and tested for antibody using TC0638. Any confirmed or suspected cases of leptospirosis should be discussed with a Herdsure® consultant. (Diagnostic testing is not done within Herdsure® – see the introductory section on 'Additional testing of cattle from Herdsure® herds'.)
2.9	**Herds with milking cows** A bulk milk sample should be tested every 3 months using the bulk milk ELISA.

2.10	**Herds with any suckler cows or suckler cows only** A statistically significant sampling of the adult suckler cows should be carried out annually. Table 3 given in Level 1 of the protocol should be used to identify the number of cows that will require sampling. **All herds** Annual statistically significant blood sampling should be carried out. Sampling should include each management group containing youngstock between 8 and 11 months of age. All breeding bulls must be sampled individually. Please refer to Table 3 provided at Level 1 of this protocol for the number of cattle in each management group to be sampled. Samples will be tested by ELISA.
2.11	Unless the herd is using a vaccination policy, there should be a reduction over time in the bulk milk antibody level and in the number of sero-positive cattle. Indeed, the herd may eventually become serologically negative. At any stage the client and veterinary practitioner may wish to consider moving to the sero-negative route at Level 2a. The Level 2a route is required for herds seeking CHeCS accreditation. If an increase in milk antibody or an increase in the proportion of sero-positive cattle is seen, this may indicate the introduction of active infection. As leptospirosis is a serious zoonotic disease, introduction of infection or evidence of active infection is of serious concern. Evidence may be revealed as: ▪ a diagnosis of leptospirosis through investigating clinical disease consistent with leptospirosis ▪ the disclosure of an increase in bulk milk antibody ratio by more than 0.2 OD units between consecutive quarterly samples ▪ the disclosure of an increase of 10% or more in the number of suckler cows showing positive results at the annual statistically significant sampling visit. In these instances you will be contacted by a Herdsure® consultant. If the herd in question is unvaccinated, vaccination may be considered. Any change in vaccinal status should be notified to the Herdsure® helpline as this will affect the future bulk milk and serum antibody levels and their interpretation.
2.12	Demonstration of effective disease reduction strategies must be shown by keeping up-to-date records of investigations of suspected clinical cases of disease and by keeping records of the use of vaccine if applicable.

Level 3

Level 3: Objective of Herdsure® testing

- Level 3 aims to monitor and maintain the improved health status of the herd. The sampling and testing is designed to provide assurance that leptospirosis is not present in the herd and to alert the veterinary practitioner if *L.* Hardjo is re-introduced into the herd.

Level 3: Sampling, testing and interpretation protocol

Also refer to the flowchart summary of the protocol at the end of Level 3.

3.1	Any animal that aborts or shows ill health, where Leptospirosis cannot be excluded on clinical grounds, should be isolated. Blood samples should be collected on first examination, and again 28 days later, and tested for antibody using TC0638. Any confirmed or suspected cases of leptospirosis should be discussed with a Herdsure® consultant. (Diagnostic testing is not done within Herdsure® – see the introductory section on 'Additional testing of cattle from Herdsure® herds'.)
3.2	A comprehensive biosecurity policy, based on advice provided in this handbook and reviewed by the farmer on an annual basis, should be maintained.
3.3	**Herds with milking cows** Quarterly monitoring of bulk milk samples should be carried out. Bulk milk samples will be tested by ELISA. This sampling should begin 3 months after entry into Level 3.
3.4	**Herds with any suckler cows or suckler cows only** Annual statistically significant blood sampling of suckler cows should be carried out, starting 1 year after successful completion of Level 2. Please refer to Table 3 provided at Level 1 of this protocol for the number of cattle in each management group to be sampled. Samples will be tested by ELISA.

3.5	**All herds** Annual statistically significant blood sampling should be carried out, starting 1 year after successful completion of Level 2. Sampling should include each management group containing youngstock intended for introduction into the milking or suckler herds that are 12 months or older. All breeding bulls must be sampled individually. Please refer to Table 3 provided at Level 1 of this protocol for the number of cattle in each management group to be sampled. Samples will be tested by ELISA. For CHeCS-accredited herds this testing, if negative, will maintain the herd status.
3.6	Where any positive samples are revealed as a result of testing of any milk or blood sampling at Level 3, the herd will revert to Level 2a or 2b depending on the decision of the veterinary practitioner and farmer. Where a herd reverts to Level 2 the Herdsure® consultant must be informed, by telephoning the Herdsure® helpline, whether the herd will enter at Level 2a or 2b.

CHeCS accreditation

The Level 2a route is required for CHeCS accreditation. Herds are eligible for CHeCS accreditation at Level 3 following 2 consecutive clear herd tests at Level 2a. More information is available on the CHeCS website (www.checs.co.uk).

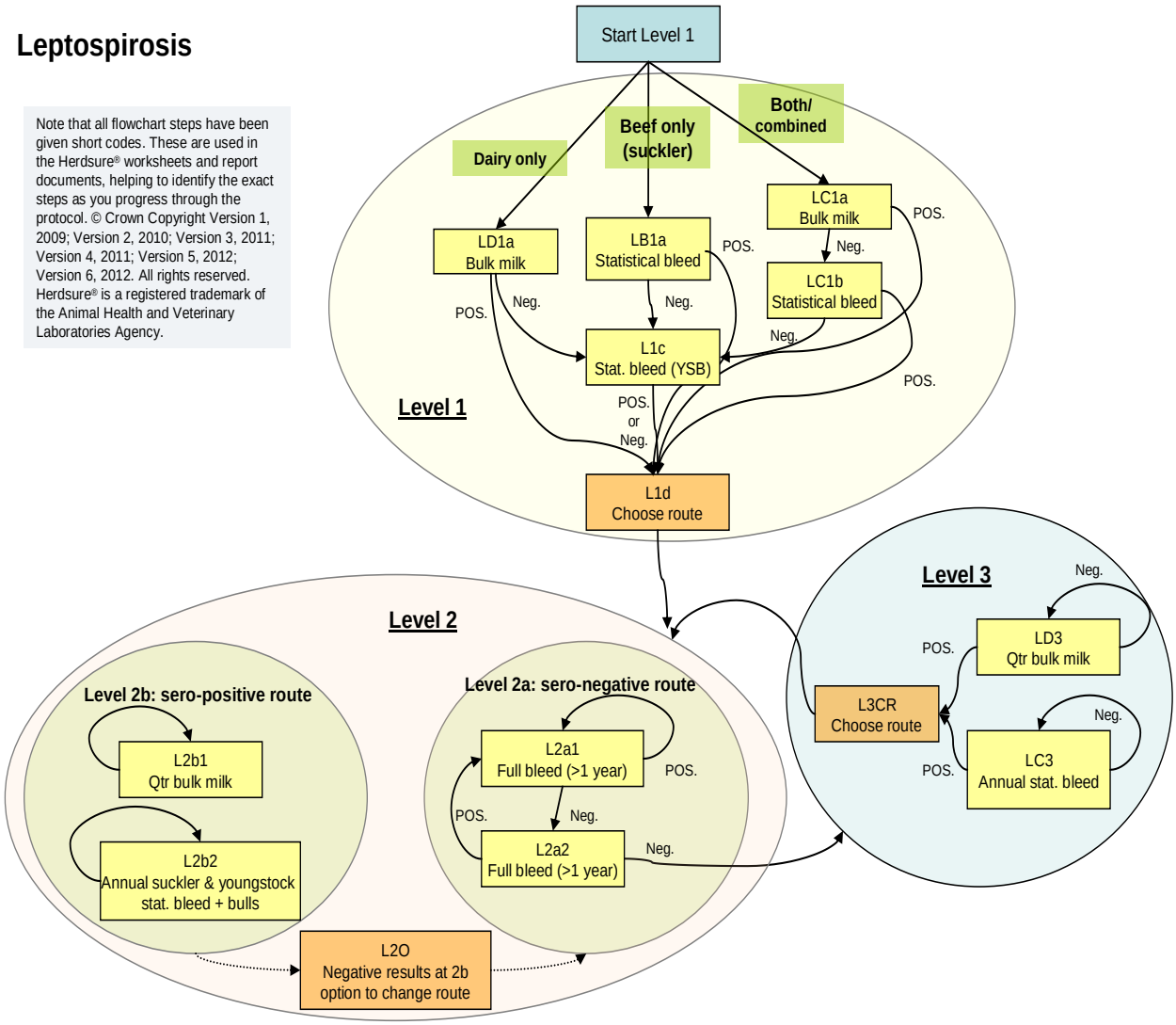
Key to flowchart summary of the Herdsure® protocol for leptospirosis

Step	Step name	
LB1a	Suckler cow statistical blood sampling	This is a statistically significant bleed of suckler cows, with testing using TC0638. Ear tag details of animals over 2 years of age will be provided.
LD1a	Dairy bulk milk	This is a bulk milk test for antibody, using TC0125.
LC1a	Combined bulk milk	For combined dairy/beef farms, this bulk milk test (TC0125) is designed to be followed (if negative) by a statistically significant bleed of suckler cows (LC1b).
LC1b	Suckler cow statistical blood sampling	This statistically significant bleed of suckler cows, using TC0638, is designed to follow on from the bulk milk test in combined beef/dairy herds. Ear tag details are provided.
L1c	Statistical sampling of youngstock and individual sampling of breeding bulls	A statistically significant bleed of all management groups of youngstock over 12 months of age, together with individual blood samples from all breeding bulls using TC0638. A list of ear tags of animals over 12 months of age is provided.
L1d	Choose route	The customer chooses the route that best reflects their requirements.
L2a1	Full bleed of breeding cattle over 12 months	This is the first of two blood samplings of all animals over 12 months of age, for testing by TC0638.
L2a2	Full bleed of breeding cattle over 12 months following a negative full bleed	This is the second blood sampling of animals over 12 months of age, and follows a negative result for the earlier bleed. It can occur between 6 months and 12 months after the first bleed.
L2b1	Qtr bulk milk	A bulk milk sample is taken for TC0125.
L2b2	Suckler cows, youngstock and breeding bulls statistical bleed	Statistically significant blood sampling of suckler cows and management groups of youngstock between 8 and 11 months of age for testing by TC0638. Also individual samples for all breeding bulls. Ear tag details are provided. This step is repeated annually until the herd progresses to a suitable stage to enter Level 2a.
L2O	Level 2 option to change route	Herds in Level 2b that are reported negative will be given the option to change to Level 2a. Changing the route at Level 2 can also be requested due to other circumstances, for example a breakdown in Level 2a.
LD3	Level 3 bulk milk monitoring (quarterly)	Bulk milk antibody testing by TC0125.
LC3	Annual statistical bleed to include bought-in but not dairy cows	Statistically significant blood sampling of each management group over 12 months of age to include suckler cows (but excluding dairy cows) for testing by TC0638. Breeding bulls should be sampled individually, as should any animals purchased since the first full herd blood sampling at Level 2. A list of ear tags is provided.
L3CR	Choose route	If a positive result occurs in Level 3, the farm reverts to one of the two routes through Level 2. The customer chooses the route that best reflects their requirements.

Flowchart summary of the Herdsure® protocol for leptospirosis

Leptospirosis

Note that all flowchart steps have been given short codes. These are used in the Herdsure® worksheets and report documents, helping to identify the exact steps as you progress through the protocol.



Procedures for added animals

Added cattle are one of the most likely ways to introduce leptospirosis into a herd. Minimising the number of cattle added to the herd from other sources reduces the risk of introducing infection. The leptospirosis history of the herd of origin of cattle intended for purchase should be ascertained in order to avoid adding cattle from a herd with active infection.

It is preferable to test cattle intended to be added to the herd while still at the herd of origin, so that ELISA antibody positive animals may be identified and rejected. The 'unscheduled submission form' Form C, in the 'Forms' section at the back of this handbook) should be used for submission of samples for ELISA (TC0638) testing.

The Herdsure® service recommends that all added animals are isolated and tested before joining the herd. Testing and isolation of added animals is mandatory for herds seeking CHeCS accreditation. The only exception to this is where animals are sourced from CHeCS disease-free certificated herds. Refer to the CHeCS technical document for the rules that apply.

Adding cattle of unknown or lower health status

All added cattle must be moved directly into an isolation facility (see the biosecurity section of this protocol for the definition of an isolation facility). They should be tested immediately after movement into the isolation facility by the ELISA test TC0638, to allow rapid identification and removal of any positive cattle, thus reducing the period in isolation. The 'unscheduled submission form' Form C should be used.

After a period of 28 days of isolation the added cattle must be tested for antibody to *L.* Hardjo by the ELISA test TC0638. Cattle showing negative results may be introduced into the herd. If cattle are bought in consignments of more than one animal and any positive animals are disclosed at ELISA testing, they must be removed and the remaining negative animals must go through another period of 28 days of isolation starting after the removal of the positive cattle. After the period of 28 days of isolation the animals should be re-tested using the ELISA test TC0638. The 'unscheduled submission form' Form C should be used.

Although the risk is very low, added cattle may show negative ELISA results but may still be infected. Therefore all added cattle that are 12 months of age or over must also be tested by ELISA 12 months after introduction to the herd. Cattle added under the age of 12 months will require negative ELISA results for the samples collected at isolation only. These tests are in addition to the requirements for other sampling carried out as part of the protocols.

Note: it is possible for infected carrier cattle to give negative antibody results, particularly when infection happened some years previously. It is therefore recommended that sero-negative cattle from non-accredited sources should receive treatment with an appropriate antibiotic as advised by your veterinary practitioner.

If cattle show positive ELISA results while in isolation

Any cattle testing positive while in isolation should be removed from the holding without delay. Bedding and waste from the isolation facility must not be disposed of onto pasture that will be grazed by cattle within 12 months. Isolation facilities should be thoroughly cleaned and disinfected with a disinfectant effective against leptospires. Care should be taken to ensure water used in washing isolation facilities is not allowed to contact the cattle in the herd.

Establishment of a new herd from accredited stock

Where it is intended to establish a leptospirosis-free accredited herd by acquiring cattle accredited free of leptospirosis, the premises must be inspected by the veterinary practitioner before the new stock is introduced in order to ascertain that the biosecurity of the premises and farm boundaries meet the requirements of CHeCS. Accreditation testing for leptospirosis must be carried out no sooner than three months after establishing the herd. Once testing has been completed, with satisfactory results, the herd can be recognised as having achieved accredited status.

Shows and sales

Contact with other stock puts the status of the herd at risk. CHeCS-accredited cattle attending CHeCS-accredited sections may return to their herds of origin without isolation and testing. All other cattle will lose their Herdsure® health status. Consequently, on returning to their herd of origin, they will be subject to the testing and isolation requirements described above. For CHeCS-accredited herds, animals moving off the owner's holding for preparation for sale will lose accredited status if the CHeCS biosecurity rules are not adhered to on the premises where preparation is taking place.

Herdsure® protocol for infectious bovine rhinotracheitis (IBR) in cattle herds

Herdsure® Chapter 5

AHVLA
Animal Health and
Veterinary Laboratories
Agency

Contents

Introduction

This protocol describes the process used to establish the disease status for infectious bovine rhinotracheitis (IBR) in cattle and for the subsequent monitoring of IBR in those herds.

This protocol is suitable for both dairy and beef herds.

IBR is a widespread and significant disease of cattle, manifesting itself in a wide range of clinical signs and syndromes. This protocol does not address the management and treatment of individual cases of clinical IBR; rather, it addresses the control of active IBR infection which leads to the expression of those syndromes.

The IBR protocol comprises three levels of 'health status':

Level 1	Establishes the IBR status of the herd.
Level 2	Aims to improve the health status of the herd for IBR.
Level 3	Monitors and aims to maintain the improved (or established as satisfactory at Level 1) health status of the herd for IBR.

The disease

Introduction

Infectious bovine rhinotracheitis (IBR) is caused by bovine herpes virus 1. The virus primarily causes respiratory disease and infected cattle can harbour the virus for life.

Surveys suggest that more than half of UK herds are infected with IBR. Since several European countries have achieved national IBR-free status for their cattle through national control plans, IBR infection is a potential barrier to international trade.

Impact of the disease

IBR infection can result in a spectrum of clinical signs from severe, fatal disease to mild or even sub-clinical infection. Although outbreaks of disease can occur in cattle of all ages, IBR is the most common cause of respiratory disease in cattle of 12 to 18 months of age. The disease is also commonly seen in young adult dairy cattle after introduction to the milking herd.

Sources of infection

IBR is spread by close contact between cattle either through nose-to-nose contact or by natural mating in the case of Infectious Pustular Vulvo-Vaginitis and Infectious Pustular Balano-Posthitis (IPVV/IPB). Cattle with clinical signs are the most infectious but IBR has the ability to survive in cattle long after they have recovered from the initial infection. These cattle are called ‘latently infected’; they may become infectious again and spread disease after periods of stress even while not showing any clinical signs. Latently infected cattle nearly always have antibody to IBR so they can be detected using serological tests. Latent infection is the primary way by which IBR persists in a herd.

Spread between herds is frequently as a result of the purchase of latently infected cattle and often outbreaks of disease are seen shortly after the introduction of cattle. Disease can be spread by nose-to-nose contact with infected stock in adjoining fields or by straying of cattle. Disease can also be spread by the use of shared equipment or personnel moving between farms, although the virus can only survive a few days in the environment and it is susceptible to disinfectants. Theoretically, infection could also be introduced to a herd by the purchase of semen for artificial insemination (AI), but the stringent controls and tests on AI studs makes this route of infection extremely unlikely.

Disease syndromes associated with IBR infection

Respiratory disease

IBR most frequently affects the upper respiratory tract and causes a raised temperature, increased respiratory rate and discharges from the eyes and nostrils that are initially clear but become cloudy or white. There may be a painful conjunctivitis and obvious reddening of the lining of the nostrils, which has led to the term 'red nose' being used to describe outbreaks in the USA. If there is extensive damage to the tracheal mucosa, it can cause severe breathing difficulty and lead to asphyxia or fatal pneumonia from secondary bacterial infection.

Reproductive disease

IBR can also cause reproductive disease. It can cause abortion at any stage of gestation and this may follow an outbreak of IBR with clinical signs or where infection was not apparent. Abortion can occur shortly, or up to 3 months, after infection. This manifestation of the disease is not common in Great Britain.

When IBR is spread by natural service, it results in inflammation of the penis or vulva and formation of pustules on the mucosa. There may be an odourless discharge and it may be painful. The infection of cows is known as Infectious Pustular Vulvo-Vaginits (IPVV) and of bulls as Infectious Pustular Balano-Posthitis (IPB). These are uncommon manifestations of IBR infection in herds in the UK.

Rapidly fatal disease in young calves

Rarely, IBR can cause severe and rapidly fatal disease in very young calves.

Production loss

IBR contributes to loss of production by causing death and longer finishing times in growing cattle and by causing decreased milk production, abortion and deaths in adult cattle.

Infection without clinical signs

Sometimes spread of IBR has been detected where there have been minimal clinical signs: The absence of obvious clinical signs does not mean that IBR infection is not present in a herd.

Diagnosis

Diagnosis is by competitive gB ELISA of individual sera (TC0675) and indirect ELISA of bulk milk (TC0124), to detect antibody to IBR. These ELISA tests detect antibody to both field and vaccinal strains of the IBR virus.

The gE ELISA (TC0680, for individual sera only) detects exposure to field strains of the virus only.

Control of IBR

Introduction of disease to a herd can be prevented by the application of appropriate biosecurity measures.

Cattle with antibodies to IBR are likely to be latently infected and able to spread infection to other cattle. IBR infection can be eliminated from a herd by repeated testing and removal of all the antibody-positive cattle.

This 'test and cull' route has been successful in many countries. However, one complication has become apparent during the application of IBR control programmes: A small proportion of latently infected cattle do not have antibody to IBR. These latently infected cattle are not detectable by serological tests so are not removed and may potentially spread disease. This can delay the eradication of infection from a herd but, despite this delay, the test and cull approach has been shown to result in eventual eradication of IBR from the herd.

Vaccination

If a large proportion of the herd is infected, the test and cull route is often not a practical option, but it is possible to limit or stop the spread of infection by vaccination. The antibodies produced by conventional vaccines do not allow vaccinated animals to be differentiated from those infected by IBR. 'Marker vaccines' have been developed allowing a specific antibody test to be used to differentiate between marker-vaccinated cattle and those infected with IBR.

Vaccination of a herd using a marker vaccine stops or severely limits the spread of disease in a herd while still allowing the detection of IBR-infected cattle. This is one of the options in Herdsure® and has been used in control programmes in several countries.

Testing for the disease

- All blood samples for Levels 1, 2 and 3 of the IBR protocol should be clearly identified and referenced to the ear tag number of the animal of origin.
- All blood samples for IBR testing should be collected in plain (red top) blood tubes.
- Definitions of age and type of cattle referred to in this protocol are explained in the section on service instructions on page 18 of this handbook, as is the definition of the 'herd' for registration purposes.

Infection of an animal by IBR virus produces an antibody response that can be detected in a blood or milk sample. Detection of antibody is by the ELISA test. The sensitivity of this test is extremely high. When cattle in the herd are vaccinated with IBR marker vaccine, an ELISA test that does not detect antibody produced in response to vaccination with marker vaccine will be used.

The following tests are used in this protocol:

TC0124	Indirect ELISA test for IBR antibodies in bulk milk samples
TC0675	Competitive ELISA test for IBR gB antibodies in individual serum samples
TC0680	Blocking ELISA test for IBR gE antibodies in individual serum samples for animals vaccinated with gE-deleted marker vaccine

Bulk milk IBR antibody test – TC0124

This is an indirect ELISA test for the detection of IBR antibody in milk.

Interpretation

Category	OD ratio	IBR interpretation/action: brief guide
Negative	<0.10	**Naïve/unvaccinated**
Low positive	0.10–0.40	Some sero-positive cows but **extensive active infection unlikely**
Mid positive	>0.40–0.70	Situation less clear
High positive	>0.70	**Heavily infected/vaccinated.** Significant virus circulation likely due to reactivation and re-excretion of latent infection. Clinical recrudescence may occur sporadically in unvaccinated herds

Blood IBR antibody test – TC0675

This is a competitive ELISA (cELISA) for gB antibodies that is used as a screening test to detect the serological status of an animal. This test is used primarily to ascertain the presence or absence of antibody rather than to quantify the level of antibody.

Interpretation

The cELISA is the preferred test for individual sera to distinguish between sero-positive and sero-negative status. The cELISA is the most suitable test for monitoring IBR status. Results are reported as either positive or negative.

Blood IBR antibody test (marker vaccine) – TC0680

The IBR gE ELISA test is used to test individual animals that have been vaccinated using an IBR gE-deleted marker vaccine to determine if the animals have been exposed to field virus.

Interpretation

For animals vaccinated with an IBR gE-deleted marker vaccine:

- those that **have not** been exposed to field strains of IBR or vaccinated with a non-marker vaccine will be detected as **negative** on the IBR gE ELISA
- those that **have** been exposed to field strains of IBR and/or vaccinated with a non-marker vaccine will be detected as **positive** on the IBR gE ELISA.

Biosecurity

Avoiding the introduction of the virus into a 'clean herd'

It is the responsibility of the herd owner or manager, in consultation with their veterinary practitioner, to ensure good biosecurity in Herdsure® herds. The following potential means of introduction of IBR into herds should be addressed and kept to a minimum.

- **Movements of people, vehicles or equipment** into areas where the cattle are kept, including fields, farm buildings and other holding areas, should be kept to a minimum.
- **Persons entering premises** to handle the cattle (or their products) should wear protective clothing and footwear. These must be clean and disinfected before and after the contact with the cattle. Alternatively, disposable protective clothing should be used. Other visitors to the farm should be kept away from direct contact with the cattle.
- **Shared farm equipment:** Equipment, machinery, livestock trailers and handling facilities that are used on herds of unknown IBR status must be cleaned and disinfected before use by herds subscribed to Levels 2 and 3 of Herdsure® IBR.
- **Other vehicles** entering the farm should not come into contact with the areas used by cattle unless they have been thoroughly cleaned and disinfected.
- **Delivery and pick-up points** should be at a site isolated from other cattle on the farm. The driver should remain in the vehicle cab and should not assist in removing cattle from pens unless using farm-dedicated protective clothing and footwear.
- **Veterinary equipment** such as drenching guns, surgical instruments and hypodermic needles, which may draw blood, must not be shared with cattle from another herd. Veterinary surgical instruments must be sterile before use.
- **Farm boundaries** must prevent cattle from straying off or onto the farm and must prevent nose-to-nose contact over fences or walls. Installation of double fencing, with a gap of 3 metres, between the farm's cattle and any neighbouring cattle is essential.

- **Contact with cattle of different health status:** Cattle herds subscribed to Levels 2 and 3 of Herdsure® IBR must not come into contact with cattle from herds that are not of an equal or higher Herdsure® IBR status, otherwise they will lose their status. To re-introduce them to the herd, they must be regarded as added cattle. (See procedures for buying in cattle at the end of the protocol section.)

- **Added animals** are particularly high risk sources of new infection - see section on 'Added animals' below.

- **Isolation period:** All cattle entering the herd should be isolated for 4 weeks and appropriate testing carried out (see section on buying in cattle at the end of the protocol). It is only when both the isolation period and the requisite tests have been completed, with results indicating freedom from infection, that these cattle can enter the herd.

- **Isolation facilities** that prevent contact with other stock must be provided for all bought-in cattle. A dedicated building separate from other cattle buildings is required. The air space, drainage or dung storage area should not be shared with other cattle. Dung should only be spread on land or added to the main dung store after all cattle in the isolation facility have passed all the required tests and have been added to the herd. If bought-in cattle are positive, dung must not be disposed of onto pasture that is to be grazed by cattle within 12 months.

- **Notification:** Herd owners and managers who are participating in Herdsure® must inform the supervising veterinary practitioner of any changes that could affect herd biosecurity.

- CHeCS accreditation: there must be a 2 month interval before accredited cattle follow non-accredited cattle onto pasture. The same grazing restrictions apply to accredited cattle if slurry or manure collected from non-accredited cattle has been used on the pasture.

Where herds are seeking CHeCS accreditation the veterinary practitioner will be asked to confirm that the appropriate biosecurity and management measures have been implemented on the farm.

Keeping track of your herd's progress in Herdsure®

An **annual herd progress report** will be issued to Herdsure® members. The progress report will detail the level achieved for each protocol for which the herd is enrolled on the date of issue.

For a small fee, an updated progress report can be produced. The updated progress report, like the annual report, will detail the level achieved for each protocol for which the herd is enrolled on the date of issue.

The protocol

Level 1

Level 1: Objective of Herdsure® testing

- Level 1 testing aims to establish whether or not there is evidence of IBR infection in a herd. Level 1 also utilises any herd history and history of relevant test results already held by the veterinary practitioner or by AHVLA.

If active infection is present it is assumed that 10% of the adult cattle and 10% of the youngstock will have sero-converted to IBR.

Level 1: Sampling, testing and interpretation protocol

Also refer to the flowchart summary of the protocol at the end of Level 3.

1.1	Where appropriate laboratory test results indicate evidence of IBR in the herd from the previous 12 months, the herd should enter at Level 2.
1.2	Before commencing on Level 1, the veterinary practitioner must inform the Herdsure® helpline whether marker vaccine is in use in the herd. This will enable the team to set the herd up on the correct route through the protocol, with the appropriate choice of tests at each stage.
1.3	Differentiation between non-marker vaccine antibody and field infection antibody is not possible, so herds vaccinated with non-marker vaccine are addressed in the same way as infected herds for the purposes of this protocol and will join the programme at Level 2.
1.4	For herds with no vaccination history, and for which previous test results are not available, samples will be collected according to the instructions provided below.
1.5 	**Dairy herds that have not used marker vaccine** A bulk milk tank sample should be submitted and will be examined for antibody. Avoid sampling when less than 75% of the cows are lactating. The collection bottle will contain preservative (e.g. Bronopol). Results will be reported as either negative or positive with the OD ratio included in the report. ▪ Herds receiving a positive result proceed directly to Level 2. ▪ Herds receiving a negative result proceed to paragraph 1.7 below if they contain suckler cows or to paragraph 1.8 if they do not.

1.6	**Dairy herds that have used marker vaccine** These herds should have the IBR status of their milking herd determined by blood sampling a statistically significant number of the milking cows. This number can be determined from Table 4 below. The samples will be tested with the gE ELISA to identify antibody produced in response to challenge with a field strain of virus or a conventional vaccine, as opposed to antibody produced in response to a gE-deleted marker vaccine. Results will be reported as either negative or positive. ■ Herds receiving a positive result proceed directly to Level 2. ■ Herds receiving a negative result proceed to paragraph 1.7 if they contain suckler cows or to paragraph 1.8 if they do not.
1.7	**Beef suckler herds** Where beef suckler cows and dairy cows are present, the dairy cows should be sampled as in paragraphs 1.5 and 1.6 above and the suckler cows as described here. Blood samples should be collected from a statistically significant sample of the adult suckler cows for IBR antibody (by gB ELISA (TC0675) if they have not been vaccinated or gE ELISA (TC0680) if they have been vaccinated with a marker vaccine). Refer to Table 4 below to calculate the number of animals to sample according to the group size. Herds vaccinated with marker vaccine should be identified as such on the submission form. Results will be reported as either negative or positive. ■ Herds receiving a positive result proceed to Level 2. ■ Herds receiving a negative result proceed to paragraph 1.8.
1.8	**All herds that are negative to this point** will now need to carry out statistically significant blood sampling of all distinct management groups of youngstock over 12 months of age. See the section below for a definition of a management group and Table 4 showing the number of animals to sample. Individual blood samples from all breeding bulls are also required. Results will be reported as either negative or positive.
1.9	When Level 1 has been completed, the results will enable a decision to be made as to whether the herd follows route **2a** or route **2b** at Level 2. A herd will be designated as **sero-positive** at Level 1 if any bulk milk ELISA or blood sample is positive. Herds will be designated as **sero-negative** if all samples tested are negative.

Statistically significant sampling of management groups

A single management group is considered to be a group of cattle grazing the same piece of land.

If the cattle are housed, then each group should be considered as a separate management group unless there is contact between them that would result in the spread of IBR between the groups. In this case, the groups sharing the same air space should be considered as a single management group.

The number of animals shown in Table 4 should be tested.

Table 4: Statistical-based sampling according to group size

Group size	Animals to sample
10	10
20	19
30	24
40	28
50	31
70	34
100	38
150	40
200	42
300	43
500	45
800	45

*For values that fall between those in the table use the next highest figure in the table or all the animals in the group, whichever is lower.

Level 2

Level 2: Objective of Herdsure® testing

- Level 2 testing aims to improve the health status of the herd by reducing the detrimental influence of IBR infection in the herd. This is done by identifying the cattle that are responsible for the maintenance of infection. Once identified, these cattle may be removed or they may be retained and a disease reduction strategy applied.

Level 2: Sampling, testing and interpretation protocol

The sampling and testing protocol at Level 2 follows a sequence determined by the results of each test.

Please also refer to the flowchart summary of the protocol at the end of Level 3.

2.1	Two routes are available through Level 2: ▪ Level 2a aims to establish an entirely serologically negative herd – the 'sero-negative' route. ▪ Level 2b aims to maintain and monitor the immunity of the herd – the 'sero-positive' or 'maintained immunity' route. The farmer and veterinary practitioner should decide which route is the most appropriate for the herd and inform the Herdsure® helpline. This will enable AHVLA to offer the appropriate reminders as Level 2 progresses. Herds seeking CHeCS accreditation must ultimately follow Level 2a, although they may start with Level 2b and change at a later date, if appropriate.
2.2	Level 2 testing is also influenced by the use of marker vaccine. For herds that entered at Level 1, information on whether this is used will already have been supplied and thus the choice of Level 2 tests will be automatic. For herds that join at Level 2, this information will need to be supplied now, to ensure the correct testing is allocated by the Herdsure® Management System.
2.3	A comprehensive biosecurity policy, based on advice provided in the 'Biosecurity' section above, should be adopted and reviewed by the farmer and veterinary practitioner on an annual basis.
Level 2a: sero-negative route	
2.4	All animals aged over 12 months old must be blood sampled together with any younger animals which are not home-bred. Samples will be examined for IBR antibody (by gB ELISA if they have not been vaccinated or gE ELISA if they have been vaccinated with a marker vaccine). Serologically positive animals must be removed from the herd. Further blood sampling at intervals of no less than 1 month and no more

	than 12 months should be carried out until no more sero-positive animals are detected.
2.5	The herd will remain at Level 2a until two clear herd tests, no less than 1 month and no more than 12 months apart, are achieved. The herd can then move to Level 3. The herd also qualifies for CHeCS accreditation on reaching this point.
2.6	All incidents of disease in which clinical signs consistent with IBR infection, as detailed, must be investigated. Appropriate samples should be sent to the local AHVLA laboratory for diagnostic testing, using the General Submission Form (VLA 3). Diagnostic testing is not done within Herdsure® – see section on 'Additional testing of cattle from Herdsure® herds' on page 21. Any confirmed or suspected cases of IBR should be discussed with a Herdsure® consultant.
2.7	In addition to the action at paragraph 2.6, the revelation of further sero-positive cows in an unvaccinated herd or a marker-vaccinated herd during the course of testing should be regarded as an indication of active infection. In this event, a decision should be made as to whether to proceed with the sero-negative route of Level 2a or to change to Level 2b (i.e. vaccinate and monitor the herd). Any requirement to change from Level 2a to Level 2b or vice versa should be notified to the Herdsure® consultant by telephoning the Herdsure® helpline.
Level 2b – sero-positive or maintained immunity route	
2.8	Level 2b requires implementation of a control and monitoring policy for IBR. Advice on control of the disease has been given earlier in the 'disease' section of this protocol. Investigations must be carried out whenever there is an episode of disease with clinical signs consistent with possible IBR infection (see 'disease' section).
2.9	**Herds with milking cows** Where marker vaccine **is not** used, the milking cows will be tested for antibody by 3-monthly testing of bulk milk samples by ELISA. Where marker vaccine **is used** in the herd, statistically significant blood sampling of milking cows (see Table 4 at Level 1 for sampling numbers) to monitor serological status of the herd will be carried out **annually**. Samples will be tested by gE ELISA.

2.10	**Beef suckler herds** Where beef suckler cows and dairy cows are present, the dairy cows should be sampled as in paragraph 2.9 above and the suckler cows as described here. Statistically significant blood sampling of adult suckler cows should be carried out **annually** to monitor the serological status of the herd. (See Table 4 in Level 1 for the number to be sampled.)
2.11	**For all herds** Annual statistically significant sampling should be carried out for each management group of youngstock between 8 and 11 months of age. (Refer to Table 4 in Level 1 for the sample size.) For marker-vaccinated herds the gE ELISA will be used; otherwise the gB ELISA will be used. At this time all breeding bulls should be sampled.
2.12	Over a period of time, the antibody levels disclosed at the 3-monthly bulk milk tests and the number of sero-positive cattle disclosed at the annual test should decrease. Indeed the herd may eventually become serologically negative to field virus. At any stage the client and veterinary practitioner may wish to consider moving to the sero-negative route (Level 2a). The Level 2a route is required for herds seeking CHeCS accreditation.
2.13	If an increase in milk antibody or an increase in the proportion of sero-positive cattle is seen, this may indicate the introduction of active infection. Evidence may be revealed as: ▪ a diagnosis of IBR through investigating clinical disease consistent with IBR ▪ the disclosure of an increase in bulk milk antibody ratio by more than 0.2 OD units between consecutive quarterly samples ▪ the disclosure of an increase of 10% or greater in the number of cattle showing positive results at the annual statistically significant sampling visit ▪ the disclosure of an increase of 10% or greater in the number of cattle showing positive results at the annual statistically significant sampling visit of the 8- to 11-month-old management groups. If the herd in question is unvaccinated, vaccination may be considered. Any change in vaccination status should be notified to the Herdsure® helpline as this will affect the allocation of future tests.
2.14	Implementation of effective disease reduction strategies must be demonstrated by up-to-date records of investigations of suspect clinical cases, as outlined by the veterinary practitioner.

Level 3

Level 3: Objective of Herdsure® testing

- Level 3 testing aims to monitor and maintain the improved health status of the herd. The sampling and testing is designed to provide assurance that IBR is not present in the herd and to alert the veterinary practitioner if IBR is re-introduced into the herd.

Level 3: Sampling, testing and interpretation protocol

Please also refer to the flowchart summary of the protocol at the end of Level 3.

3.1	All episodes of disease in which clinical signs are consistent with IBR infection, as detailed by the veterinary practitioner, must be investigated.
3.2	A comprehensive biosecurity policy should be maintained, based on advice provided in this handbook. This should be reviewed by the farmer and veterinary practitioner on an annual basis.
3.3	**Herds with milking cows** Quarterly monitoring of bulk milk samples should be carried out, starting 3 months after entry into Level 3. Bulk milk samples will be tested by ELISA. For marker-vaccinated herds, annual statistically significant blood sampling of the milking herd should be carried out. (Refer to Table 4 in Level 1 for the number of cattle to be sampled.) For marker-vaccinated herds with CHeCS accreditation, annual blood sampling of **all** marker-vaccinated animals is required. This sampling should begin 1 year after entry into Level 3; samples will be examined by the gE ELISA.
3.4	**Beef suckler herds** Where beef suckler cows and dairy cows are present, the dairy cows should be sampled as in paragraph 3.3 above and the suckler cows as described here. Annual statistically significant blood sampling, starting 1 year after successful completion of Level 2, of suckler cows should be carried out. (Refer to Table 4 in Level 1 for the number of cattle to be sampled.) For marker-vaccinated herds with CHeCS accreditation, annual blood sampling of **all** marker-vaccinated animals is required. Samples will be examined for IBR antibody (by indirect ELISA if they have not been vaccinated or gE ELISA if they have been vaccinated with a marker vaccine).

3.5	**All herds** **Annual** statistically significant blood sampling must be carried out, starting 1 year after successful completion of Level 2. Sampling needs to include each management group containing animals that are 12 months or older. (Refer to Table 4 in Level 1 for the number of cattle in each management group to be sampled.) All breeding bulls, bought-in animals from non-accredited herds and all marker-vaccinated animals must be sampled individually. Where any positive samples are revealed as a result of testing of any milk or blood sampling at Level 3, the herd will revert to Level 2a or 2b depending on the decision of the veterinary practitioner and farmer. Where a herd reverts to Level 2, the Herdsure® consultant must be informed (via the Herdsure® helpline) whether the herd will re-enter at Level 2a or 2b.

CHeCS accreditation

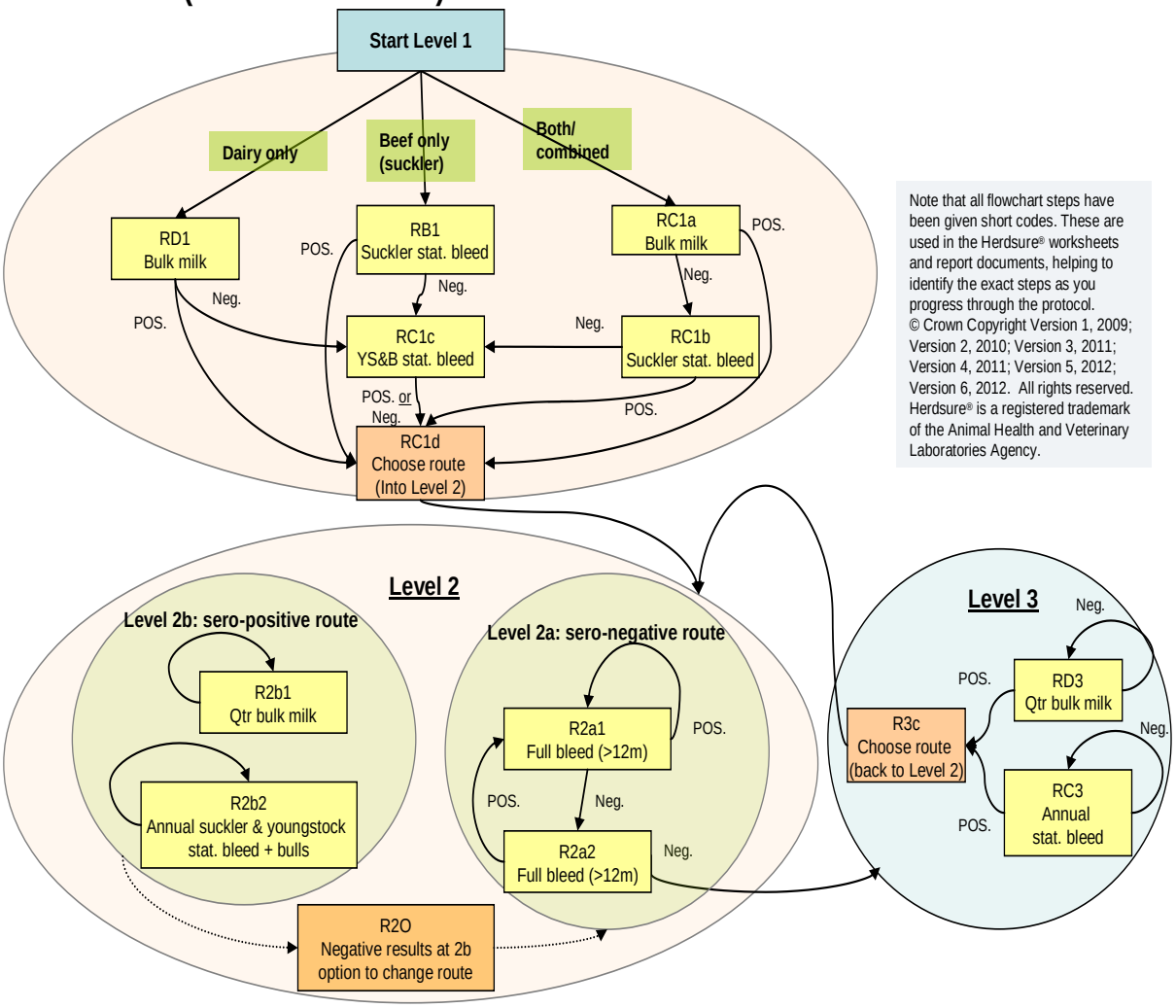
The Level 2a route is required for CHeCS accreditation. Herds are eligible for CHeCS accreditation at Level 3 following 2 consecutive clear herd tests at Level 2a. More information is available on the CHeCS website (www.checs.co.uk).

Key to flowchart summary of the Herdsure® protocol for IBR – standard version

Step	Step name	
RB1	Suckler cow statistical blood sampling	This is a statistically significant bleed of suckler cows, with testing using TC0675. Ear tag details for animals over 2 years of age will be provided.
RD1	Dairy bulk milk	This is a bulk milk test for antibody, using TC0124.
RC1a	Combined bulk milk	For combined dairy/beef farms, this bulk milk test (TC0124) is designed to be followed (if negative) by a statistically significant bleed of suckler cows.
RC1b	Suckler cow statistical blood sampling	This statistically significant bleed of suckler cows, using TC0675, is designed to follow on from the bulk milk test in combined beef/dairy herds. Ear tag details are provided.
RC1c	Statistical sampling of youngstock and individual sampling of breeding bulls	A statistically significant bleed of all management groups of youngstock over 12 months of age, together with individual blood samples from all breeding bulls using TC0675. A list of ear tags of animals over 12 months of age is provided.
RC1d	Choose route	The customer chooses the route that best reflects their requirements.
R2a1	Full bleed over 12 months	This is the first of two blood samplings of all animals over 12 months of age, for testing by TC0675.
R2a2	Full bleed following a negative full bleed (over 12 months)	This is the second blood sampling of animals over 12 months of age, and follows a negative result for the earlier bleed. It can occur between 1 and 12 months following the first bleed.
R2b1	Quarterly bulk milk	A bulk milk sample is taken for TC0124.
R2b2	Suckler cows, youngstock and breeding bulls statistical bleed	Statistically significant blood sampling of suckler cows and management groups of youngstock between 8 and 11 months of age. Also individual samples for all breeding bulls. Ear tag details are provided. This step is repeated annually until the herd progresses to a suitable stage to enter Level 2a.
R2O	Level 2 option to change route	Herds in Level 2b that are reported negative will be given the option to change to Level 2a. Changing the route at Level 2 can also be requested due to other circumstances, for example a breakdown in Level 2a.
RC3	Annual statistical bleed to include bought-in animals but not dairy cows.	Statistically significant blood sampling of each management group over 12 months of age to include suckler cows but excluding dairy cows. Breeding bulls should be sampled individually, as should all bought-in animals from non-accredited herds. A list of ear tags is provided.
RD3	Level 3 bulk milk monitoring	This step is bulk milk antibody testing by TC0124.
R3c	Choose route	If a positive result occurs in Level 3, the herd reverts to one of the two routes through Level 2. The customer chooses the route that best reflects their requirements.

Flowchart summary of the Herdsure® protocol for IBR – standard version

Infectious bovine rhinotracheitis (IBR) – standard version (no marker vaccine)

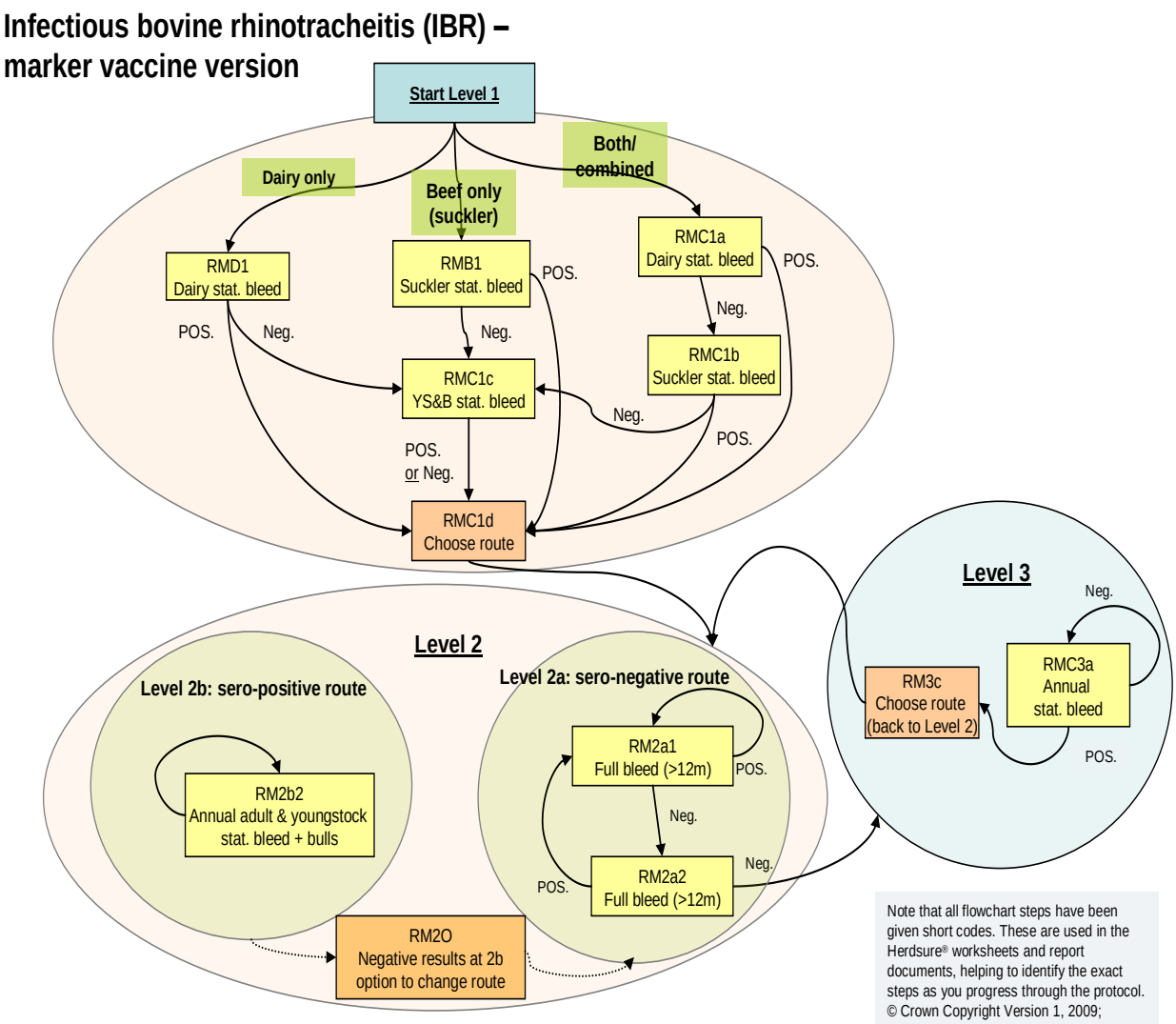


Note that all flowchart steps have been given short codes. These are used in the Herdsure® worksheets and report documents, helping to identify the exact steps as you progress through the protocol.

Key to flowchart summary of the Herdsure® protocol for IBR – marker vaccine version

Step	Step name	
RMB1	Suckler cow statistical blood sampling	A statistically significant bleed (for TC0680), designed for a solely suckler herd. Ear tag details are provided.
RMD1	Dairy cow statistical blood sampling	A statistically significant bleed (for TC0680) of the dairy herd. Ear tag details are provided.
RMC1a	Combined dairy statistical bleed	For combined dairy/beef farms. This statistically significant bleed (for TC0680) of dairy animals is designed to be followed (if negative) by a suckler bleed. Ear tag details are provided.
RMC1b	Suckler cow statistical blood sampling	For combined dairy/beef farms. This statistically significant bleed (for TC0680) follows a negative dairy bleed. Ear tag details are provided.
RMC1c	Statistical sampling of youngstock and individual sampling of breeding bulls	A statistically significant bleed of all management groups of youngstock over 12 months of age, together with individual blood samples from all breeding bulls using TC0680. A list of ear tags of animals over 12 months of age is provided.
RMC1d	Choose route	The customer chooses the route that best reflects their requirements.
RM2a1	Full bleed over 12 months	This is the first of two blood samplings of all animals over 12 months, for testing by TC0680.
RM2a2	Full bleed following a negative full bleed (over 12 months)	This is the second blood sampling of animals over 12 months, and follows a negative result for the earlier bleed. It can occur between 1 and 12 months after the first bleed.
RM2b2	Milking, suckler cows, youngstock and breeding bulls statistical bleed	Statistically significant blood sampling of all milking cows, suckler cows and management groups of youngstock between 8 and 11 months of age. Also individual samples for all breeding bulls using TC0680. Ear tag details are provided. This step is repeated annually until the herd progresses to a suitable stage to enter Level 2a.
RM2O	Level 2 option to change route	Herds in Level 2b that are reported negative will be given the option to change to Level 2a. Changing the route at Level 2 can also be requested due to other circumstances, for example a breakdown in Level 2a.
RMC3	Annual statistical bleed to include bought in animals	Statistically significant blood sampling of each management group over 12 months of age to include suckler cows and dairy cows. Breeding bulls should be sampled individually, as should any bought-in animals from non-accredited herds. For CHeCS-accredited herds all marker-vaccinated animals must be sampled. A list of ear tags is provided.
RM3c	Choose route	If a positive result occurs in Level 3, the herd reverts to one of the two routes through Level 2. The customer chooses the route that best reflects their requirements.

Flowchart summary of the Herdsure® protocol for IBR – marker vaccine version



Note that all flowchart steps have been given short codes. These are used in the Herdsure® worksheets and report documents, helping to identify the exact steps as you progress through the protocol.

Procedures for added animals

Added cattle are one of the more likely ways to introduce IBR into a herd. Minimising the number of cattle added to the herd from other sources reduces the risk of introducing infection.

It is wise to establish the IBR history of the herd of origin of cattle intended for purchase in order to avoid buying cattle from a herd with IBR infection.

It is preferable to test cattle intended to be added to the herd while still within the herd of origin so that antibody positive animals may be identified and rejected. The ELISA antibody test TC0675 for unvaccinated cattle or TC0680 for marker-vaccinated cattle will be used. Herds vaccinated with marker vaccine should be identified as such on the submission form. The 'unscheduled submission form' Form C, in the 'Forms' section at the back of this handbook) should be used.

The Herdsure® service recommends that all added animals are isolated and tested before joining the herd. Testing and isolation of added animals is mandatory for herds seeking CHeCS accreditation. The only exception to this is where animals are sourced from CHeCS disease-free certificated herds. Refer to the CHeCS technical document for the rules that apply.

Adding cattle of unknown or lower health status

All added cattle must be moved directly into an isolation facility. (See the biosecurity section at the beginning of this protocol for the definition of an isolation facility.) They should be tested immediately after movement into isolation to allow rapid identification and removal of any positive cattle, thus reducing the period in isolation. The ELISA antibody test TC0675 for unvaccinated cattle or TC0680 for vaccinated cattle will be used. Herds vaccinated with marker vaccine should be identified as such on the submission form. Use the 'unscheduled submission form' Form C.

After a period of 28 days of isolation, the added cattle must be tested for antibody to IBR by the appropriate ELISA test. Cattle showing negative results may be introduced into the herd. If cattle are bought in consignments of more than one animal and any positive animals are disclosed at ELISA testing, the positive animal(s) must be removed and the remaining negative animals must go through another period of 28 days of isolation starting after the removal of the positive cattle. After the period of 28 days of isolation the animals should be re-tested using the ELISA test. Herds vaccinated with marker vaccine should be identified as such on the submission form. The 'unscheduled submission form' Form C should be used.

Very occasionally, cattle with latent infection may show negative ELISA serum antibody results but still be infected. Therefore, all added cattle that are 12 months of age or over must also be tested by the appropriate ELISA test (TC0675 or TC680) 12 months and 24 months after their introduction into the herd. Cattle added under the age of 12 months will require negative ELISA results for the samples collected at isolation only. These tests are in addition to the requirements for other sampling carried out as part of the protocols.

Where cattle show positive ELISA results while in isolation

Any cattle testing positive should be removed from the holding without delay. Bedding and waste from the isolation facility must not be disposed of onto pasture that will be grazed by cattle within 12 months. Isolation facilities should be thoroughly cleaned and disinfected.

Establishment of a new herd from accredited stock

Where it is intended to establish an IBR-free accredited herd by acquiring cattle accredited free of IBR, the premises must be inspected by the veterinary practitioner before the new stock is introduced in order to ascertain that the biosecurity of the premises and farm boundaries meet the requirements of CHeCS. Accreditation testing for IBR must be carried out no sooner than three months after establishing the herd. Once testing has been completed, with satisfactory results, the herd can be recognised as having achieved accredited status.

Shows and sales

Contact with other stock puts the status of the herd at risk. CHeCS-accredited cattle attending CHeCS-accredited sections may return to their herds of origin without isolation and testing. All other cattle will lose their Herdsure® health status. Consequently, on returning to their herd of origin, they will be subject to the testing and isolation requirements described above. For CHeCS-accredited herds, animals moving off the owner's holding for preparation for sale will lose accredited status if the CHeCS biosecurity rules are not adhered to on the premises where preparation is taking place.

Herdsure® protocol for neosporosis in cattle

Herdsure® Chapter 6

AHVLA
Animal Health and Veterinary Laboratories Agency

Contents

Introduction

This protocol describes the process for establishing the neosporosis disease status of cattle herds and for controlling and monitoring the disease where it is found to be present. It is suitable for both dairy and beef suckler herds.

The protocol does not describe procedures for the diagnosis of abortions that may be due to *Neospora caninum*, but it does provide the means to measure and reduce the amount of *N. caninum* infection in the herd.

The neosporosis protocol comprises a sampling, testing and management regime to establish herd health status at three different levels:

Level 1	Provides an indication of the herd's neosporosis status based on a snapshot bleed or historical data.
Level 2	Aims to improve the health status of the herd for neosporosis.
Level 3	Monitors the herd's improved neosporosis health status.

The disease

Introduction

Neosporosis is the disease in cattle caused by *Neospora caninum*, an apicomplexan protozoal parasite with worldwide distribution. The definitive host is the dog. Cattle are an intermediate host of the parasite and infection can result in abortion.

Impact of the disease

Although rare cases of neurological disease in congenitally affected calves have been described, by far the most common sign of neosporosis in cattle is abortion. Neosporosis is the most frequently diagnosed cause of abortion in cattle in the UK. The cost of abortion, which involves not just the loss of the calf but also reduced milk production and additional breeding costs, is well documented. Therefore, the cost of neosporosis to the UK cattle industry is significant.

The organism causes infection of cattle by one of two routes:

- endogenous ('vertical') infection in which the organism passes transplacentally from an infected dam to its calf, or
- exogenous ('horizontal') infection in which the pregnant cow ingests oocysts from dog faeces contaminating the environment, e.g. feed or pasture grass.

Cattle are most commonly infected via the endogenous route.

Both these routes give rise to infection of the foetus. Infection may, in turn, lead to abortion, usually between 3 and 8 months gestation (median 6 months). However, not all cases of intra-uterine infection lead to abortion; some infected calves are clinically normal. It is not currently possible to distinguish between the two routes using available diagnostic tests.

Infection can enter a naïve herd either via infective dog faeces contaminating feed or pastures or by the purchase of congenitally infected animals. It is perpetuated by vertical transmission and dogs may be infected by the ingestion of dead calves or placentae. Therefore, control of the disease is by a combination of biosecurity measures and the identification of infected animals and their removal from the breeding herd.

Testing for the disease

An ELISA (TC0175), which detects antibodies to *N. caninum*, is used for screening in the Herdsure® protocol. Sero-positive animals are approximately six to seven times more likely to abort compared to sero-negative animals. The sensitivity of this test is highest in animals that are in the late stage of pregnancy or that have recently aborted. All blood samples taken during the course of this protocol should be collected in plain (red top) vacutainer tubes without anticoagulant.

It is important for the success of the neosporosis protocol that all incidents of abortion are appropriately investigated. For further advice please contact your local AHVLA Regional Laboratory. Relevant samples should be submitted to your local AHVLA Regional Laboratory for diagnostic investigation; all such investigations should be carried out as AHVLA diagnostic submissions outside Herdsure®. (See the section entitled 'Additional testing of cattle from Herdsure® herds' on page 21 of this handbook.)

Antibody to *N. caninum* fluctuates throughout the life of an infected animal. Confirmation that an animal is not infected requires sampling at a specific time:

- during an animal's pregnancy (10 to 4 weeks before the due calving date), ***or***
- at the time of abortion, provided this occurs within the period of 10 to 4 weeks before the due calving date.

Interpretation of results

N. caninum ELISA test results are expressed as 'percentage positivity'. These results will be interpreted by the AHVLA's laboratory management system as negative or positive. Interpretation may change if AHVLA changes the ELISA, but it is currently:

- sero-negative – samples showing a positive inhibition of <30%
- sero-positive – samples showing a positive inhibition of ≥30%.

Keeping track of progress in Herdsure®

Each registered holding with be issued an **annual herd progress report**. The progress report will detail the level achieved for each protocol for which the herd is enrolled on the date of issue.

For a small fee an additional progress report can be produced. This progress report, like the annual report, will detail the level achieved for each protocol for which the herd is enrolled on the date of issue.

Biosecurity

It is the responsibility of the herd owner or manager in consultation with their veterinary practitioner to ensure good biosecurity in Herdsure® herds. Below is a list of biosecurity measures that are specific to the control of neosporosis. It is important that herds participating in Herdsure® Levels 2 and 3 conform to these.

- All dogs must be prevented from having access to the calving areas and any carcase material or placentae. This includes farm dogs, visitors' dogs, dogs belonging to members of the public and fox hounds.
- Placentae, stillborn calves and carcases should be removed from the calving accommodation/paddocks as soon as possible to a secure location ready for removal by fallen stock contractors. This location should be inaccessible to vermin such as foxes, badgers, rats and mice. Dogs must also be prevented from gaining access to potentially infected material.
- The public and dogs must not have access to paddocks used for calving.
- Feed storage facilities, both 'straights' and forage, must be dog-proof to prevent contamination with dog faeces. They should also be vermin-proof to prevent contamination by foxes, badgers, rats and mice.
- Suppliers of feed should provide assurance that measures are in place to prevent contamination of feed by faecal material from dogs.
- Access of dogs to pasture used for cattle grazing or for the production of cattle forage should be kept to a minimum because it could increase the risk of cattle becoming infected with *N. caninum*.
- Added cattle: see the section on added cattle at the end of the protocol.

The protocol

Level 1

Level 1: Objective of Herdsure® testing

- The objective of Level 1 is to determine the neosporosis status of the herd. This is done by carrying out a **snapshot bleed**.

The snapshot bleed involves testing a specified proportion of the female breeding herd (using TC0175) 10 to 4 weeks before their expected calving date.

When determining the size of the breeding herd, all the cattle on the farm that are over 2 years of age and are or will be part of the breeding herd should be counted (see Table 5 below). If both milking and suckler cows are present on the same farm they should be considered as one group.

It is recommended that the snapshot bleed is carried out at a time when as many cattle as possible are between 10 and 4 weeks before calving. It is preferable that these are tested at one visit.

The snapshot bleed will provide an early indication of whether neosporosis is on the farm. However, it is not possible to conclusively prove the absence of infection by a snapshot bleed. Where tested cattle give negative ELISA results, these will count as one of the two negative tests results needed to classify them as neosporosis-free in Level 2.

Level 1: Sampling, testing and interpretation protocol

Choosing the animals to bleed for the snapshot bleed	
1.1	If the AHVLA laboratory reports for the previous year show a clear indication of sero-positive cows or confirmation of *N. caninum* in aborted foetuses by foetal serology or foetal histology, it is unnecessary to carry out the Level 1 snapshot bleed; the herd should enter the neosporosis protocol at Level 2. For any herd to complete a valid snapshot bleed, there must be enough cattle within the period 10 to 4 weeks before calving to make up a valid sample size as calculated from Table 5. The veterinary practitioner and farmer should decide if a valid snapshot bleed can be performed. If so, blood samples should be collected as instructed below and submitted to AHVLA Luddington for ELISA testing.
1.2	**What to do if there are insufficient suitable animals for the snapshot bleed** Herds with protracted calving periods may not have an adequate number of cows in the 10- to 4-week window to satisfy the required sample size. When review of the expected calving dates indicates that a valid snapshot bleed cannot be performed at a single visit, it is advised that herds should enter the protocol directly at Level 2.
Results interpretation	

1.3	The ELISA test results will be reported as either negative or positive.
1.4	It is important that all incidents of abortion are investigated. Blood samples from aborted cows should be submitted to your local AHVLA Regional Laboratory for ELISA antibody testing outside Herdsure®. (See section on 'Additional testing of cattle from Herdsure® herds' on page 21.) All other relevant samples of aborted material should also be submitted to your local AHVLA Regional Laboratory for diagnostic and, if appropriate, statutory investigation with testing outside Herdsure®. Where blood samples prove positive or where *N. caninum* is confirmed as the cause of abortion in diagnostic samples the Herdsure® consultant must be informed by telephoning the Herdsure® helpline.
1.5	The presence of any infected animals is evidence of 'infected herd' status. The absence of any sero-positive animals does not confirm absence of infection but it is certainly a result that should be viewed optimistically before progressing to Level 2.

Rationale and sample size calculations for the snapshot bleed

Sampling should be carried out when as many cows as possible are in the 10 to 4 weeks pre-calving period.

It is assumed that, if *N. caninum* infection is present, at least 5% of adult cows will have measurable antibody between 10 and 4 weeks before calving. The test sensitivity in this period is 90%. To be 95% confident that at least one positive animal is detected, ***at least*** the number of in-calf cows (between 10 and 4 weeks before calving) shown in Table 5 should be sampled. Animals for sampling should be selected randomly.

If more than the minimum number of qualifying cows are present, all of these may be tested as the results will be of use during Level 2.

Table 5: Statistical-based sampling according to group size

Group size	Cows to sample
<23	all
23–25	22
26–30	26
31–35	31
36–40	35
41–50	36
51–55	39
56–60	43
61–80	47
81–100	51
101–150	53
>151	61

Level 2

Level 2: Objective of Herdsure® testing

- The aim of Level 2 is to improve the neosporosis status of the herd by identifying and removing sero-positive cows. Successful completion of Level 2 will result in all the cows in the herd achieving neosporosis-clear status. The Level 2 testing is known as the progressive bleed.

An animal will be confirmed free of *N. caninum* infection when it has achieved two clear ELISA antibody tests on samples collected as described in the section 'Testing for the disease'. Any negative ELISA results gained in a Level 1 snapshot bleed will be credited to the individuals now entering Level 2.

Animals with positive ELISA test results should be removed from the breeding herd.

Once an animal is confirmed free of *N. caninum* infection, all the calves born to her are considered free of *N. caninum* and will not require antibody testing. Thus the amount of required testing of cows will reduce over time until the entire breeding herd is free. At that point all home-bred animals can be considered free of *N. caninum* infection.

Only when all the breeding animals have been confirmed free of *N. caninum* can the herd be classified as neosporosis-free and move to Level 3.

Level 2: Sampling, testing and interpretation protocol

2.1	**Identification of breeding females** The veterinary practitioner will be sent a list of all adult females on the farm. Those tested during Level 1 will be annotated with their snapshot bleed test result. This list should be photocopied and a copy returned to the Herdsure® consultant. A column is included to indicate those animals that are not in the breeding herd by annotating them 'NB'. This information is needed to produce the '*Neospora* Status Reports'. (See the section below on 'Tracking the Level 2 progressive bleed.)
2.2	**Sampling of breeding females** In Level 2, each breeding cow should be tested between 10 and 4 weeks before calving. This is a ***progressive bleed*** with sampling and testing repeated until each cow has two negative ELISA results. All the samples taken for the progressive bleed should be submitted to AHVLA Luddington. The eligible animals should be batched in groups of those fulfilling the 10- to 4-week pre-calving window to help economy of veterinary visits. This table shows possible combinations of sampling times which will confirm

freedom from neosporosis in an individual animal (born to a dam of unknown status).

Period for sampling	Option 1		Option 2	
	First sample*	Second sample	First sample*	Second sample
10–4 weeks pre-calving	X		X	
10–4 weeks pre next calving		X		
Abortion, if 10–4 weeks pre-calving				X

*The ELISA result of the first sample must be negative in order to proceed to the second.

To assist with management of this process, the HMS will provide a list of all female cattle known to be in the breeding herd, showing the ELISA results of the individuals where appropriate. This list will be updated regularly (see section on 'Tracking the Level 2 progressive bleed' below).

2.3 Dealing with sero-positive cattle

Sero-positive animals should be removed from the herd.

Sero-positive animals should not be used for breeding. If herds have a high prevalence of positive animals they can be removed from the herd over a period of time to reduce the financial impact of their loss to the herd, provided they are not then used for further breeding. If these animals are inseminated, served or used as recipients for embryos they are at risk of either aborting or producing an infected calf. They may be retained and used for embryo donation or for fattening before they are removed from the herd or culled.

2.4 Progress through Level 2

The progressive blood sampling of female breeding cattle in the herd should continue until they have all reached an *N. caninum* infection-free status with two negative blood tests.

Calves born to cows with *N. caninum* infection-free status will be deemed to have inherited that free status and will not require any further testing.

2.5	**Investigation of abortions** Any abortions that occur within the herd must be investigated to determine whether or not *N. caninum* has played a role. The minimum requirement for herds that are in Level 2 of the Herdsure® protocol for neosporosis is the submission of a blood sample from the cow that has aborted for the *N. caninum* ELISA test. However, a definitive diagnosis of neosporosis as the cause of abortion can be made only by the submission of an aborted foetus. The relevant samples should be submitted to your local AHVLA Regional Laboratory for diagnostic investigation. Advice on these can be obtained from the local AHVLA Regional Laboratory. All tests on maternal blood samples and aborted foetuses will be carried out according to procedures for diagnostic submissions to AHVLA and not as part of Herdsure®. (See section on 'Additional testing of cattle from Herdsure® herds' on page 21.) All positive *N. caninum* ELISA results must be reported to the Herdsure® consultant by telephoning the Herdsure® helpline. All such cattle should be dealt with as in paragraph 2.3.

Tracking the Level 2 progressive bleed: *Neospora* Status Reports

At Level 2, the HMS helps to manage the progressive bleed by storing results from each Herdsure® neosporosis submission. *Neospora* Status Reports (see example below) are used to summarise the status of individual animals. The *Neospora* Status Report has four main functions:

- It gives a picture of progress of the herd.
- It defines the status of all the breeding animals in the herd.
- It provides a means of recording calving dates for planning tests of in-calf animals.
- It can be used to identify which animals are to be used for breeding. When a *Neospora* Status Report is received for the herd, any cattle included in it that will ***not*** enter the female breeding herd should be annotated 'NB'. A photocopy of the amended report should be sent to the Herdsure® consultant who will ensure that these cattle are removed from the database list of female breeding cattle.

Neospora Status Reports are available at 1-, 2- or 6-monthly intervals but will be sent to the veterinary practitioner 6-monthly unless otherwise requested.

The figure below shows the layout for *Neospora* Status Reports. In this example the farm is halfway through the second year of a progressive bleed; most animals have been tested once and around half have been tested twice. Each of the target animals is listed by ear tag and date of birth (from CTS). Customers can use the first column to note expected calving dates in order to help with planning of sampling visits.

The status column indicates the current status of each animal. The status codes are explained in the key. The remaining columns show first and second test results for each animal where these have been carried out.

Neospora Status Report

Date **01 January 2012**
Practice **Gridlington Vets**
Farm **Home Farm, Gridlington**
CPH **12/123/1234**

Key	
NN	Two negative results – confirmed sero-negative
N	One negative result – potentially sero-negative
P	POSITIVE (should be removed from herd)
DN	Dam Negative (NN so no test needed for offspring)
U	As yet Untested

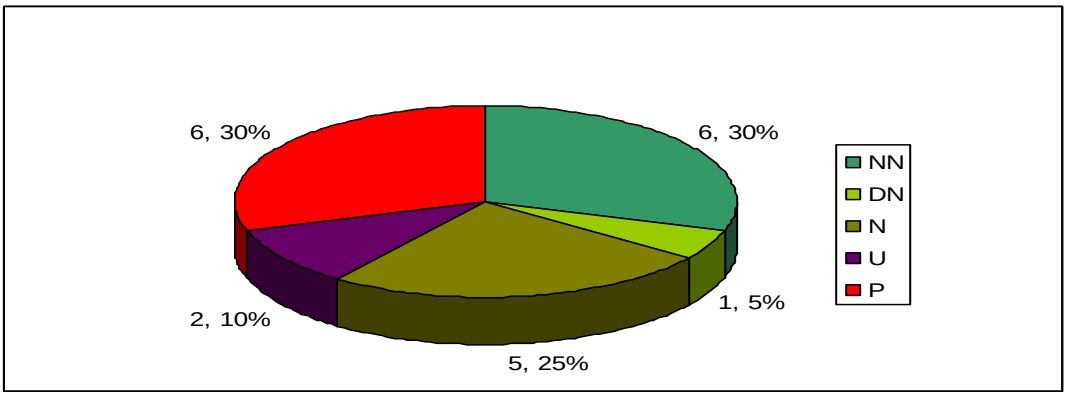
				Neospora test results			
				First test		Second test	
Due to calve?	Ear tag	Born	Status	Date	Result	Date	Result
	UK281526700065	01/02/08	NN	01/11/10	Negative	20/11/11	Negative
	UK602440100006	02/02/08	NN	30/11/10	Negative	20/11/11	Negative
	UK602440100024	06/02/08	P	30/11/10	Negative	20/11/11	**POSITIVE**
	UK602440100061	07/02/08	NN	01/11/10	Negative	20/11/11	Negative
	UK602440100066	08/02/08	NN	01/11/10	Negative	20/11/11	Negative
	UK602440100076	09/02/08	P	01/11/10	**POSITIVE**		-
	UK602440100122	10/02/08	P	30/11/10	**POSITIVE**		-
	UK602440100146	12/02/08	NN	12/12/10	Negative	20/11/11	Negative
	UK602440100150	13/02/08	U				
	UK602440100157	14/02/08	NN	01/11/10	Negative	20/11/11	Negative
	UK602440100164	15/02/08	P	30/11/10	Negative	20/11/11	**POSITIVE**
	UK602440100178	16/02/08	N	12/12/10	Negative		
	UK602440200011	17/02/08	N	20/12/10	Negative		
	UK602440200018	18/02/08	N	01/11/10	Negative		
	UK602440200019	14/02/10	DN				
	UK602440200025	20/02/08	P	01/11/10	**POSITIVE**		-

Current herd status

Veterinary practitioners can choose the frequency of these reports for each of their registered farms.

Pie chart as visual guide to the herd's neosporosis progress

This pie chart gives a visual impression of the herd's current status with respect to neosporosis. (See above for key.)



Level 3

Level 3: Objective of Herdsure® testing

- Level 3 testing aims to monitor and maintain the improved neosporosis health status of the herd.

Monitoring the continued freedom from infection is carried out by ELISA testing of all aborting cows.

Level 3: Sampling and testing protocol

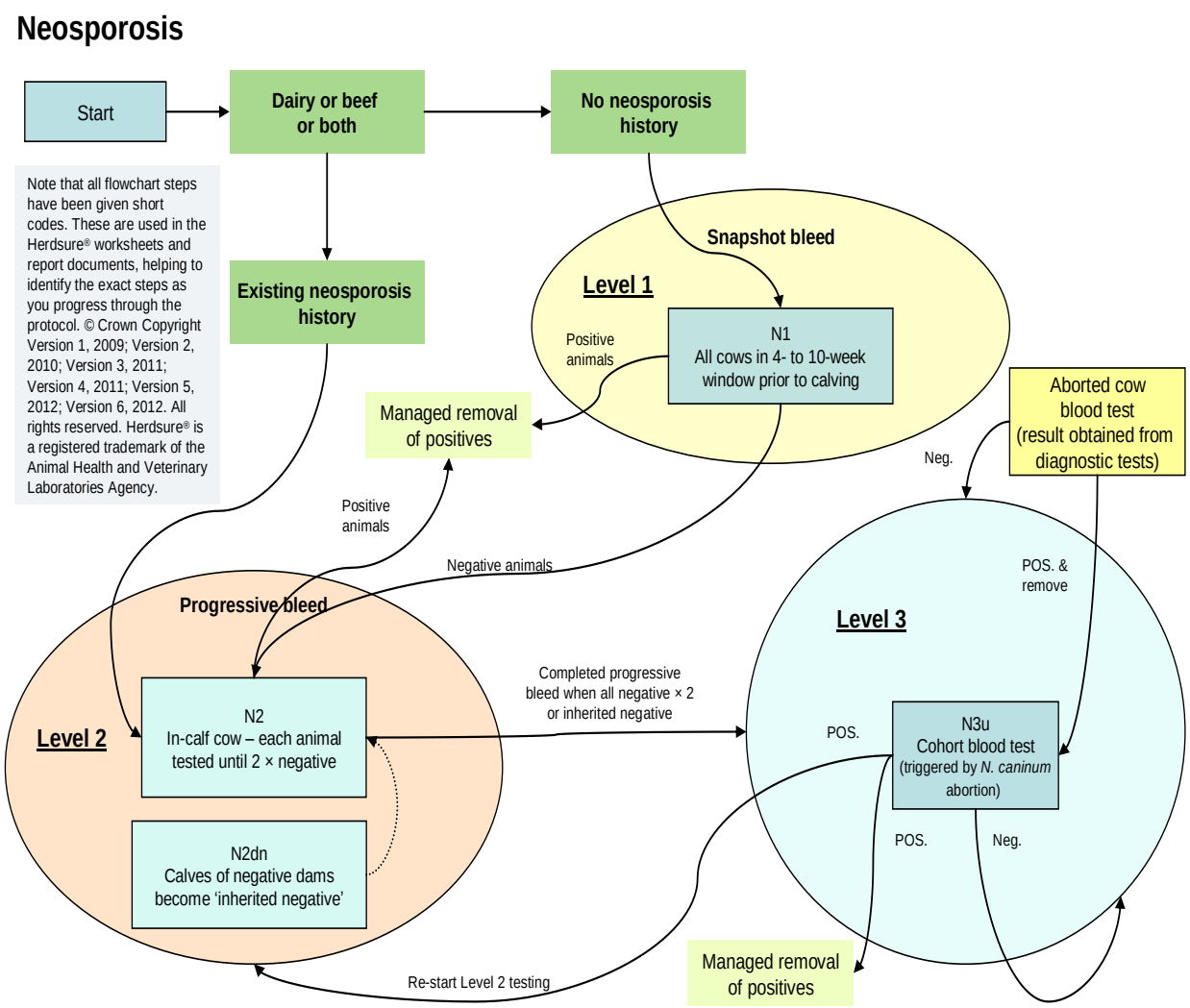
3.1	Strict biosecurity and adherence to the added animal policies must be maintained. (See the section on biosecurity at the end of this protocol.)
3.2	**Investigation of abortions** Any abortions that occur must be investigated to confirm whether *N. caninum* has played a role. The minimum requirement for herds that are in Level 3 of the Herdsure® neosporosis protocol is the submission of a blood sample from each aborting cow for the *N. caninum* ELISA test. However, a definitive diagnosis of neosporosis as the cause of abortion can be made only by the submission of an aborted foetus. The relevant samples should be submitted to your AHVLA Regional Laboratory for diagnostic investigation. Advice on these can be obtained from your AHVLA Regional Laboratory. All tests carried out on maternal blood samples and aborted foetuses will be carried out according to procedures for diagnostic submissions to AHVLA and not as part of Herdsure®. (See section on 'Additional testing of cattle from Herdsure® herds' on page 15.) All positive *N. caninum* ELISA results and all results confirming *N. caninum* as the cause of abortion following examination and testing of abortion material must be reported to the Herdsure® consultant by telephoning the Herdsure® helpline. All such cattle should be dealt with as in paragraph 2.3.
3.3	**Action if neosporosis is diagnosed as a cause of abortion** If *N. caninum* infection is found to have played a part in the abortion, then the herd will revert to Level 2 while a herd investigation is carried out in the following way. Immediate blood sampling for ELISA testing of all adult animals in the same cohort as the aborting cow should be carried out. This cohort is defined as all animals which are due to calve or have calved from 1 month before to 1 month after the calving date of the aborted cow. If all of the cows and heifers in the cohort are sero-negative, the herd may return to Level 3. Sero-positive results in one or more animals in the cohort suggest an external

	source of infection and means the herd restarts Level 2 and all the breeding animals must requalify for Level 3. This will mean that the neosporosis-free status of all the breeding female cattle in the herd will be lost and the herd will begin sampling and testing for Level 2 again. Farm biosecurity should be reviewed and, if necessary, improved following the breakdown in order to identify potential risks and routes of entry of *N. caninum* infection.

Key to flowchart summary of the Herdsure® protocol for neosporosis

Step	Step name	
N1	Snapshot bleed	Where the calving pattern allows for a significant proportion of the breeding herd to be sampled 10 to 4 weeks before calving, all of these cows should be tested using ELISA test TC0175. The herd includes both dairy and suckler cows where present. For the minimum number to be sampled refer to Table 5 in Level 1. A list of ear tags of breeding age cattle will be provided to assist with the sampling process. Positive animals are removed from the herd.
N2	In-calf blood sampling	A sampling worksheet is provided for submission of blood samples from all in-calf cows, 10 to 4 weeks before calving. Samples are submitted for the ELISA test TC0175. Positive animals are removed from the herd; negative animals are repeat tested in the window 10 to 4 weeks before calving until they are classified negative (2 × negative tests). The Herdsure® consultant may be able to assist with planning optimum sampling visits.
N2dn	Inherited negative – no testing	No sampling is needed here. Calves of dams that are classified negative (two negative Herdsure® tests) inherit their negative status. When this occurs the *Neospora* Status Report will be updated automatically.
	Abortion – 'diagnostic submission only'	Although not part of the Herdsure® sampling protocol, any abortion must be investigated by submitting samples to your local AHVLA laboratory. As part of this investigation the minimum required is a TC0175 of a maternal blood sample. Positive results must be notified to the Herdsure® consultant.
N3u	Cohort blood sampling	Abortions involving neosporosis trigger immediate blood sampling of all in-calf animals in the same cohort. Samples will be tested by the ELISA for antibody to *N. caninum* (TC0175). This cohort is defined as all cows due to calve or that have calved from 1 month before to 1 month after the date of the abortion. Samples should be submitted using the unscheduled sample submission form (Form C in the 'Forms' section at the back of this handbook).

Flowchart summary of the Herdsure® protocol for neosporosis



Procedures for added animals

Added animals are one of the most likely ways to introduce infection to a herd. Minimising the number of added animals will keep the risk of introducing infection low. You may wish to consider a closed herd policy.

If possible only buy animals that have been confirmed negative according to the Herdsure® protocol. These animals can be added without testing.

Cattle of 'unknown health status' can be introduced, but they should be subject to testing as described below before they join the remainder of the herd. Blood samples from all added animals should be submitted to AHVLA Herdsure accompanied by Form C (Unscheduled Submissions) for TC0175.

Added animals should preferably be blood sampled at the farm of origin and sero-positive animals should not be purchased.

If this is not possible, added animals should be tested on arrival and, if sero-positive to *N. caninum,* should be removed from the herd as soon as possible. Under no circumstances should any offspring be retained from sero-positive added animals to introduce into the breeding herd.

Animals that give negative results cannot be regarded as free of infection on the basis of one blood test. Sero-negative animals should be regarded as *potentially* infected until they have achieved two negative blood tests taken between 10 and 4 weeks before two calvings. This could include the pre-purchase blood sample if taken in this period of gestation. Although sero-negative cattle can enter the herd, they will be recorded on the HMS as only having attained one clear test and thus Level 3 herds will revert to Level 2 status. Only when all breeding cattle in the herd have achieved two clear blood tests at the appropriate stage of pregnancy can the Level 3 status of the herd be reinstated.

Embryos must only be implanted into sero-negative recipients.

APPENDICES

APPENDIX 1: Example of a sampling submission worksheet

AHVLA Animal Health and Veterinary Laboratories Agency

Herdsure®

J Bloggs and Son
Any Veterinary Hospital
252 Any Road
Anytown
RB12 7JB
Contact: John Smith

Date: 10, January 2011

Herdsure® Sampling Worksheet
Johne's Level 1 Step J1a – Full Adult Bleed

Note: This form MUST be submitted with the samples

Request Reference	HS0010-01-11
Farm & CPH	A. B. Taylor
Sample Required	Blood
For Test	TC0366 – Mycobacterium Paratuberculosis Ab ELISA
Required by	07/02/2011
Enclosed	
Instructions	Please collect blood samples (red top) from all breeding animals over 2 years of age. An ear tag list of cattle over 2 years of age is provided.

HS0010-01-11

Data Protection Act 1998: In addition to recording the information on this form onto the Herdsure Management System, we may also use the data provided for other purposes. Please see the data protection statement on our website www.vlascientific.com.

This slip can be removed and retained for your records

Herdsure Sampling Worksheet

Activity – Johne's Level 1 Step J1a – Full Adult Bleed	
Request Reference	**HS0010-01-11**
Farm & CPH	**A. B. Taylor**
Sample Required	**Blood**
For Test	**TC0366**
Required by	**07/02/2011**
Enclosed	

APPENDIX 2: Example of a Sampling Worksheet – Ear Tag Annex

AHVLA Animal Health and Veterinary Laboratories Agency

Herdsure®

Herdsure Sampling Worksheet – Ear Tags Annex
Johne's Level 1 Step J1a – Full Adult Bleed

It is important that samples submitted to the laboratory are annotated in such a way as to make them clearly traceable to an animal or management group. The following options can be used:

(1) Write the animal ID on the vacutainer tube and annotate the ear tag list accordingly.

(2) Write the vacutainer tube number onto the ear tag list (or a partial vacutainer number if appropriate). Please ensure when using non-sequentially numbered vacutainer tubes that no duplication of numbers occurs.

For option (2), enter first Vacutainer Number

Ear tag barcode	Ear tag	CTS age	Sample identifier
UK226257300001	UK226257300001	6y 10m	
UK226257500003	UK226257500003	6y 9m	
UK226257600004	UK226257600004	5y 8m	
UK226257300001	UK226257300001	5y 8m	
UK226257100006	UK226257100006	4y 9m	
UK226257200007	UK226257200007	4y 6m	
UK226257300008	UK226257300008	3y 7m	
UK226257600011	UK226257600011	3y 5m	
UK226257200012	UK226257200012	5y 1m	
UK226257100120	UK226257100120	5y 6m	
UK226257500200	UK226257500200	5y 9m	

.

APPENDIX 3: Sampling instructions

Bulk milk collection

A pre-labelled sample collection pot, preservative and packaging have been provided to allow for the collection of a bulk milk sample.

- Collect a bulk milk sample of 25 ml (a full pot) from the bulk milk tank in the sampling pot provided. **NB:** Do ***not*** dip the sampling pot with preservative into the bulk tank. See the separate safety guidelines for preservative that will be issued with the pots.
- The pot contains a small amount of preservative which will make the milk turn slightly blue; this is normal and will not affect the performance of the test.
- Firmly replace the lid (it is advised to seal the lid with insulation tape in order to avoid leaks).
- Place the sample pot inside the plastic transport container provided and snap shut.
- Place the sample and submission request form back into the envelope and seal with tape. Affix the postage-paid Business Reply label - this covers the full postage cost.
- Post the package back to the testing laboratory.
- The test results will be returned to you via your nominated veterinary practice.

(**NB:** The safety advice that accompanies the bottles should be adhered to.)

Pooled faeces collection (applicable to liver fluke)

- Collect 5 g of faeces (2 teaspoonfuls) of faeces from either pens or field from at-risk cattle on farm.
- Place each faeces sample into a sampling pot and firmly replace the lid.
- Repeat this sampling until all 40 pots contain a 5 g sample. (Select different sites around the pens/field in order to cover a range of at-risk animals.)
- It is advised to seal the lid with insulation tape in order to avoid leaks.
- Place the pots in a box, along with the submission request form.
- Seal the box with tape and affix the postage-paid Business Reply label - this covers the full postage cost.
- Post the package back to the testing laboratory.
- The test results will be returned to you via your nominated veterinary practice.

Individual milk collection (applicable to BVD – for pooling at the laboratory)

This procedure will be used following a positive PCR bulk milk sample. The purpose is to identify smaller groups of cows that may include the cow, or cows, that are responsible for excretion of virus in their milk and therefore into the bulk milk sample. By doing this it is possible to reduce the number of cows for BVD antigen testing.

SEE APPENDIX 4 FOR FULL INSTRUCTIONS.

Appendix 4: Collection of individual milk samples for PCR examination in pools of 10

This procedure will be used following a positive PCR bulk milk sample. The purpose is to identify smaller groups of cows that may include the cow, or cows, that are responsible for excretion of virus in their milk and therefore into the bulk milk sample. By doing this it is possible to reduce the number of cows for BVD antigen testing.

For this test, milk should be collected individually from each cow in the group by expressing approximately 5–10 ml of milk directly from the teat, taking care to avoid cross-contamination.

All the samples collected in this way will be pooled in the testing laboratory. Cross-contamination of samples will not result in the inaccurate identification of viraemic cows as the animals in positive pools will be individually blood sampled for BVD antigen. It may, however, result in additional false positive pools with additional antigen testing becoming necessary as a result.

Where two separate bulk milk tanks are in use on a farm, it will be possible to provide two separate bulk milk samples in the first instance. By doing this it may be possible to exclude one part of the herd from individual milk sampling. If this option is chosen, a Herdsure® consultant should be informed before submitting the samples. It is important that the animals contributing to each bulk milk sample are accurately identified and that their identities are annotated on the worksheet accompanying the samples.

Follow this procedure to avoid contamination:

1. Wash and dry your hands thoroughly.
2. Only wash the teat to be sampled if it is obviously dirty and then dry it immediately.
3. Discard the first four to five draws of milk.
4. **Clean the end of the teat even if you have just washed and dried it:**
   a) Use a small piece of cotton wool soaked in surgical spirit (80% spirit / 20% water).
   b) Wipe the end of the teat with the cotton wool until it is completely clean.
5. **Take the sample:**
   a) Open the sample bottle – **keep the lid clean** – never place the lid open-side down and preferably hold it in the crook of your little finger.
   b) Hold the sample bottle at an angle to the teat.
   c) Discard a further draw of milk.
   d) Fill the sample bottle to at least half-full.
   e) Replace the lid carefully. Seal the universal with tape if necessary.
6. **Label the sample bottle** with the cow's ear tag or other unique identifier. Make sure that the ID corresponds with the Herdsure® Sampling Worksheet.
7. Send the samples using the pre-paid address label to AHVLA Herdsure® Cattle Health Improvement Service, Animal Health and Veterinary Laboratories Agency, PO Box 653, Newcastle upon Tyne, NE12 2ET
   (**NB:** If preservative is added safety advice should be adhered to)

APPENDIX 5: Example - Herd Progress Report

Herdsure® Herd Progress Report

Animal Health and Veterinary Laboratories Agency, PO Box 653, Newcastle upon Tyne, NE12 2ET. Email:Herdsure@ahvla.gsi.gov.uk

The Herd Owned by: _____

Kept at: _____

Holding no: _____

Signed on behalf of the Animal Health and Veterinary Laboratories Agency annually. All samples have been tested in accordance with Herdsure® protocols:

Protocol	Level	Protocol	Level
BVD		Johne's disease	
Leptospirosis		IBR	
Liver fluke		Neosporosis	

Signed: _____

Name: _____

Date: _____

With regard to diseases specified above:
Level 1 – establishes the disease status of the herd
Level 2 - improves the health status of the herd
Level 3 - monitors and aims to maintain the improved health status

To be signed optionally by the person responsible for management of the herd.

The biosecurity recommendations and management actions for the control of the enrolled Herdsure® protocols, listed above as described in the Herdsure® handbook, have been implemented in this herd.

Signed: _____

Name: _____

Date: _____

Please retain for your records. Not to be used as a sales certificate.
Additional copies of this report may be requested at any time for a small fee

Appendix 6: Example of a Results Report

AHVLA
Animal Health and
Veterinary Laboratories
Agency

Herdsure®

Any Veterinary Hospital
252 Any Road
Anytown
RB12 7JB
Contact: Joe Swift

Herdsure® Report

BVD Level 2 Step BD2a – Bulk milk PCR

Request reference	HS00003-09-09
Farm & CPH	Home Farm 01/123/1234
Sample/Tests	Bulk milk / TC0709 – Viral RNA in milk PCR
Verdict	**Negative**

Verdict Interpretation

Step BD2a (Negative)

PCR on bulk milk has proved negative therefore blood testing of all cattle over 30 days old on the holding can begin, Step BD2d. A sampling worksheet is attached.

TC0709 (†) Results

Sample	Sample Ref	BVD PCR Bulk Milk Result
HS11-13699	Bulk Milk	Negative

John Williams
Responsible Officer
07, April 2011

‡ - Test subcontracted; opinions given and interpretations of the result are outside the scope of UKAS accreditation.
† - Not UKAS accredited; opinions given and interpretations of the result are outside the scope of UKAS accreditation.
For further details of the test methods used, and other terms and conditions, please refer to the VLA Website.

UKAS TESTING 1769

Appendix 7: Sampling Rules

HERDSURE has been designed to cost effectively manage the testing of many herds for multiple diseases over a long time frame. In order to do this we ask veterinary practitioners to assist us by testing the correct animals at the correct time.

SAMPLING RULES

1. Please do not carry out any sampling until you receive sampling submission forms with instructions.

2. Follow Herdsure® protocols as specified on the sample submission form. The Herdsure Management System follows a stepped testing process and skipping steps will cause delays for your client.

3. Use the correct blood tubes and fill to a minimum of half way, to prevent having to revisit the farm and resample animals.

   BVD - Green top (Heparinised)
   All other diseases - Red top (Plain)

4. If animals are not sampled, please record on the submission form the reason why.

5. Bulk milk sample pots should contain preservative. (These are normally supplied by Herdsure with clear instructions)

6. When taking youngstock blood samples please ensure the animals are in the correct age group for sampling.

7. **All samples must correspond with full ear tag numbers. Otherwise testing will be delayed until we receive this information. Freeze brands and part numbers are not acceptable.**

8. Ensure all paperwork is included with the samples - front page and ear tag list with corresponding blood tube number. Ear tag lists from TB or brucellosis worksheets are acceptable. Neospora Level 2 does not have an ear tag list so please use Form C (unscheduled submission form).

9. Please tape together multiple boxes of bloods to avoid boxes being separated in the post.

10. If you are unsure of testing requirements please call the Herdsure helpline prior to arriving on farm.

    Information at www.vlascientific.com (click on Health Improvement Services)