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.

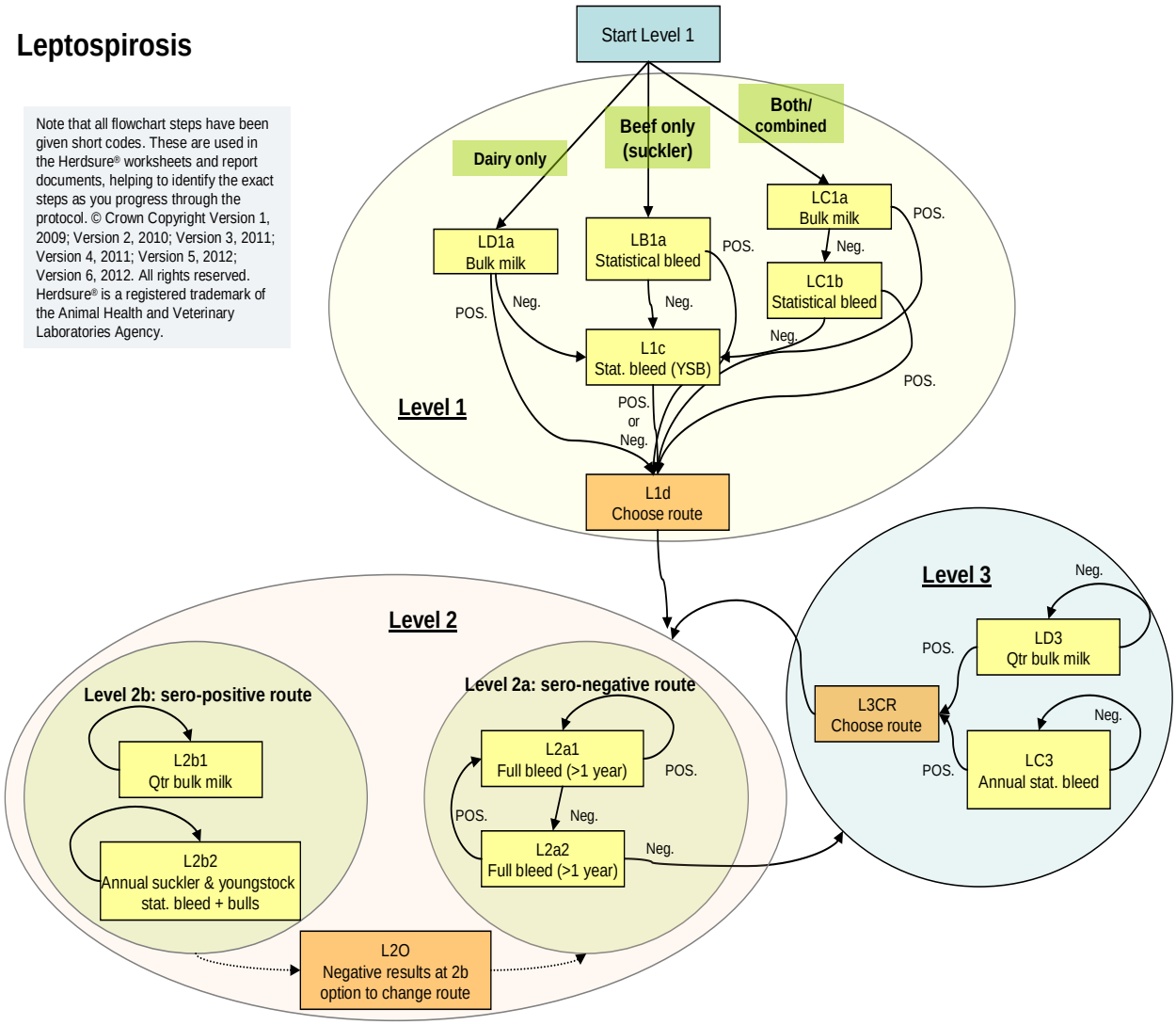
CHeCS accreditation

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



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.