

Theileria *Veterinary Handbook*



Ministry for Primary Industries
Manatū Ahu Matua



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Introduction

Theileria Veterinary Handbook, June 2014

By editors Andy McFadden (MPI) and Roger Marchant (NZVA)

Theileria associated bovine anaemia (TABA) will continue to have an impact on cattle farming for some years to come. This *Theileria* veterinary handbook is designed as a 'ready-to-use' reference for veterinarians advising clients on management of TABA and contains a number of articles and resources that have been published over the last 12 months.

The number of outbreaks of TABA in cattle herds has steadily increased since Ikeda was first identified in New Zealand in August 2012 (identified indirectly in spring 2011 from historically stored serum). There are several streams of data suggesting that the number of outbreaks may be greater this coming spring (2014) than previous ones (2012 and 2013). These data relate to temporal trends in the epidemic that showed an increase in the number of cases during autumn 2014 in comparison with the same time last year. The forecast for greater numbers in the spring 2014 is supported by predictive modelling using data from the epidemic.

There is also likely to be some variation in the regional impact of TABA. Surveillance data

from the first part of 2012 showed that some regions (East Cape and the Bay of Plenty) have high tick activity; but to date have had herds with relatively low levels of exposure to Ikeda. The impact in these regions may be greater in the future. Outbreaks in Auckland and Waikato are also likely to continue to be significant. Naïve livestock moved into endemic areas such as Northland are also likely to be at risk depending on when the movement occurs and the physiological status of the animal.

Future research and understanding of the disease may mean that the management practices detailed here are superseded. However, the information provided is what we know now.

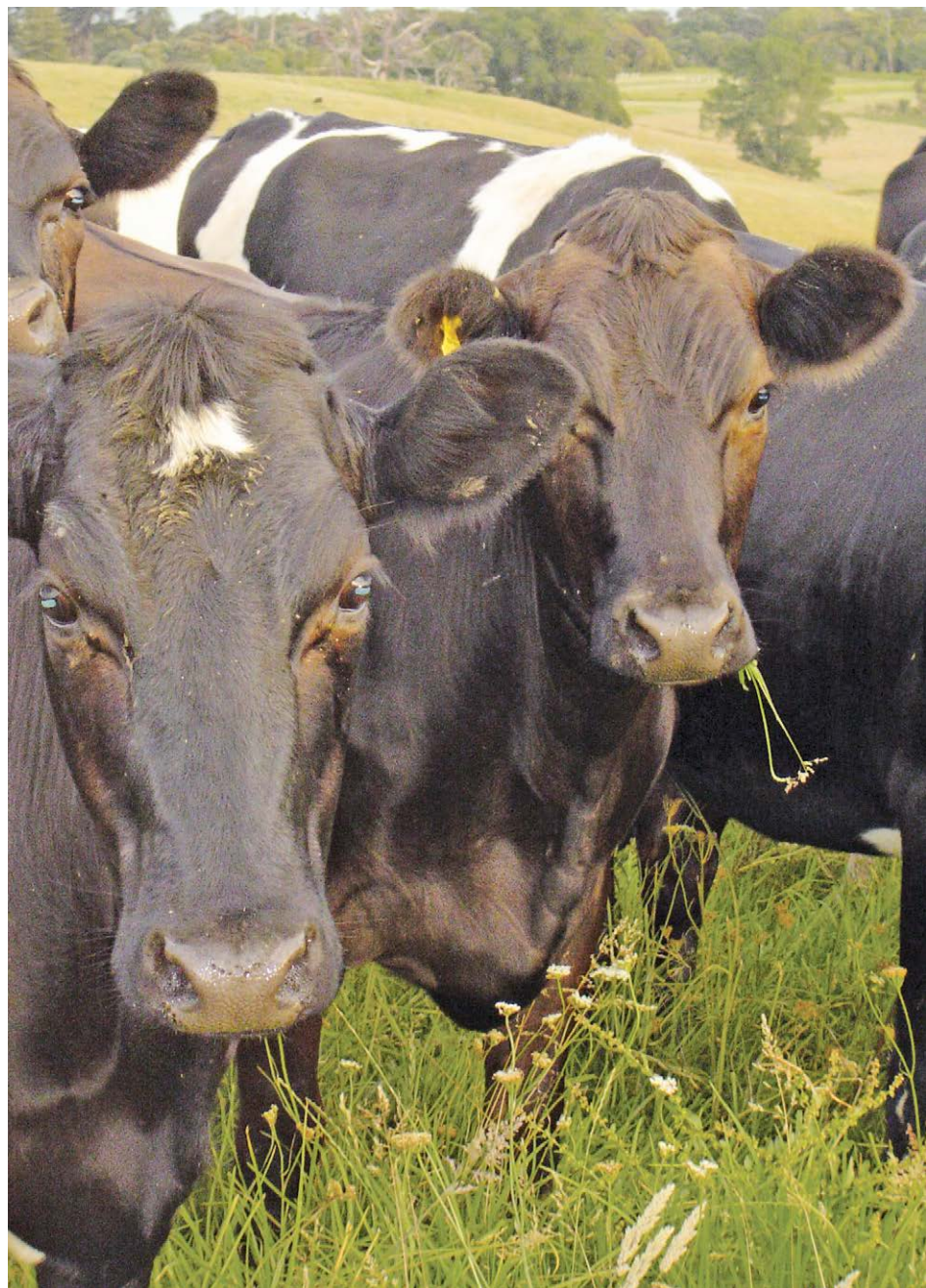
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Contents

Introduction: Theileria Veterinary Handbook, June 2014	1
Industry-level strategies to mitigate effects of <i>Theileria</i>-associated bovine anaemia	4
Assumptions	4
Management practices	4
Acknowledgements	9
References	9
Diagnostic tree for anaemia in cattle	10
Farm-level management of <i>Theileria orientalis</i> Ikeda outbreaks in dairy cattle	11
Diagnosis and treatment of the individual animal	11
Management of the herd	15
Management of diseased animals and the herd	16
Discussion	17
Useful resources	18
References	19
<i>Theileria orientalis</i> (Ikeda) associated bovine anaemia: The epidemic to date	20
References	23
Tick biology drives infestation	25
Managing ticks within the farm system	27
Pasture management to reduce tick numbers	28
Further reading	28

Industry-level strategies to mitigate effects of *Theileria*-associated bovine anaemia

By Andy McFadden (Ministry for Primary Industries), Bill Pomroy (Massey University), Roger Marchant (NZVA), Allen Heath (AgResearch), Caleb King (Keinzley Agvet), Kevin Lawrence (Massey University) and Neil MacPherson (NZVA Society of Dairy Cattle Veterinarians)

New Zealand cattle herds have been affected over the past few years from disease associated with introduction of the *Theileria orientalis* Ikeda strain. Difficulty in managing the disease has occurred as a result of our incomplete knowledge of the epidemiology of this new agent and its vector, the cattle tick (*Haemaphysalis longicornis*).

The disease, known as *Theileria*-associated bovine anaemia (TABA), is likely to continue to have an impact on cattle herds for years to come. Management systems will need to adapt to deal with TABA within affected regions where the cattle tick is active and also with the risk of disease resulting from movement of cattle into and out of these areas.

Assumptions

This article summarises management practices to deal with important issues raised by farmers and veterinarians. The suggestions are far from comprehensive and reflect the short period we have had to observe and research the disease in this country. In addition, the management practices are based on several important assumptions that may or may not be refuted in the future. These are as follows.

- Transtadial infection in the cattle tick occurs.
 - Infection does not get transferred by the female tick to her eggs and hence the unfed larval stage will be uninfected, but piroplasms can be transmitted between other stages after they have fed and moulted.
 - Female ticks die once they lay eggs.

- Nymphs and sometimes adult ticks have a behavioural diapause, which may last from April to mid-July in most areas, during which they do not quest and feed.
- Once cattle are infected with Ikeda they are infected for life; relapse with clinical disease generally does not occur.
- Only one tick is necessary to transmit infection of Ikeda to cattle.
 - It is possible that there is a dose-related effect from Ikeda on cattle; however, we have insufficient evidence to state this.
- The incubation period of infected cattle is four-to-six weeks.
- Previous exposure to the Chitose strain is not protective for disease associated with the Ikeda strain.

Management practices

As knowledge broadens, management of the disease will need to adapt and change accordingly. Future industry-led research may clarify the deficiencies in our current knowledge base. The practices proposed in this article are based on what we know now and are an attempt to provide steps to mitigate the on-farm impacts from TABA.

1 Control programme for an outbreak of TABA on a dairy property

- Maintain awareness for potential outbreaks, particularly if outbreaks have occurred on nearby properties.
 - Observe the herd for indications of anaemia such as lethargic cows, pale udders, decreased milk production or unresponsive metabolic cases.
- Confirm the diagnosis of TABA by excluding other possible causes of regenerative anaemia. (The developing epidemic means that, in most cases, only limited ancillary testing and investigation may be necessary; however, the incidental finding of Ikeda in cattle during outbreaks of facial eczema and zinc toxicity have resulted in false positive diagnoses of TABA during autumn 2014 (see McFadden et al (2013) for case definition; refer to page 10 for the diagnostic tree and Vink et al (2013) for management of an affected herd)).
- Identify affected animals by using the Field Anaemia Nearest Indicator (FANI) card (check the colostrum and springer mobs daily at first, then less frequently later through lactation). Determine the haematocrit (HCT)/packed cell volume (PCV) on suspect cases (Vink et al, 2013).
- Determine the scale of the problem (based on the prevalence of cases within an affected herd, geographic location in relation to current tick distribution map and origin of affected cattle) and whether a whole-herd approach is necessary versus treating a small number of individually affected animals.
- Treatments to consider for individual cattle:
 - blood transfusions (see Vink et al (2013) and O'Driscoll (2013) for cow blood transfusion tips)
 - buparvaquone (the long withholding periods, time associated with ear tagging and recording and so on may mean this treatment is not cost effective or warranted – see (3) below).
- Where a whole-herd approach is necessary, work with the client to develop a plan. Seek to reduce stress within the herd. Reduction of stress may be dealt with by:
 - improved nutrition (buy in feed if necessary and avoid feeding brassicas)
 - once-a-day milking and reduction of walking time to the shed for affected animals
 - minimise handling
 - ancillary treatments where appropriate (acaricides, anthelmintics and so on).
- It is not known if acaricides (tickicides) will reduce impact from the outbreak.
 - Individual animals showing clinical signs of disease may have been exposed four-to-six weeks before signs are observed. Transmission from these initial clinical cases will require the ticks to moult to the next stage and this takes an average of 2 to 5 weeks depending upon temperature to occur.
 - Parasitaemia can precede overt clinical disease by two-to-three weeks, so some ticks that have fed on these affected cattle in the previous few weeks are likely to be already infected.
 - If the outbreak has occurred in a marginal tick area there may be more benefit in using an acaricide in the face of an outbreak than in an area where there are high tick numbers, particularly to move the timing of appearance of the disease to when cattle are under less stress.

2 Elements of a control programme for an outbreak of TABA in beef calves (generally occurring at six-to-eight weeks of age)

- Confirm the diagnosis of TABA (as indicated in (1) above).
- The epidemiology of disease in beef calves is not fully understood.
 - Disease in six- to eight-week-old calves implies exposure to Ikeda soon after birth.
 - The tick cycle in Northland (where most clinical cases in young beef calves have been reported to date) is accelerated with both nymphs and adult ticks likely to be present at the time of calving.
- Acaricides are of limited value in controlling outbreaks in beef calves.
- Management of outbreaks in beef calves may be problematic and the stress of catching animals for treatments (both blood transfusion and use of acaricides) means that, in most cases, individual treatments are not a practical option.
- At present sufficient information is not available to provide a clear management plan for this scenario and further research is necessary.

3 Management strategies available to limit the future impact in adult dairy cattle in a herd that has already experienced an outbreak of TABA

- To understand risk for future outbreaks it is important to know the percentage of the herd that remains naïve to Ikeda.
 - Determine the prevalence of exposure (and thus the percentage naïve) by carrying out polymerase chain reaction (PCR) analysis on blood from individual cows (rather than using pooled blood samples, which

will inform only on the presence or absence of Ikeda). The accuracy of the prevalence measure will increase with the more animals that are tested; however, a sample of 10 will be sufficient to gain an idea of whether a large number of naïve animals still remain in the herd.

- If more than one-third of the herd remain naïve (as determined from the prevalence study), acaricides may be considered pre-calving using the provisos discussed (see (9) below).
- Despite an outbreak having occurred, naïve cattle may continue to enter the herd, either through introductions of purchased animals or through young stock entering the herd as replacements. These naïve animals are likely to be exposed at some point. The small relative number of naïve animals may mean that herd disease does not become obvious (see (4) below).

4 Management strategies to limit the future impact in dairy young stock (R1 and R2) in cattle herds that have already experienced an outbreak of TABA

- Most young stock are reared off-farm, and the risk of exposure at these locations is often unknown.
- In regions where this disease is at moderate-to-high prevalence, young stock should be visited weekly and monitored for signs of anaemia (ill thrift, lethargy and so on).
- Exposure in summer and autumn in this class of stock is better than exposure during spring calving (due to the physiological stress experienced at this time).
 - If the previous outbreak has been significant (more than just sporadic individual animals affected clinically

and subclinically), natural exposure through grazing young stock in endemic areas with high numbers of ticks infected with Ikeda may be one management method of ensuring that first exposure does not occur at calving during spring.

- There is an ethical responsibility to consider the potential impact of sending infected (parasitaemic) cattle from a property having previously experienced an outbreak to properties where naïve animals may be present and where there is a viable tick population.
 - Movement of these cattle may also result in risk to neighbouring properties.
- The use of acaricides will not nullify risk.

5 Risk of TABA in naïve cattle moved to another location (either into an endemic or marginal area)

- Grazing naïve cattle in endemic regions (or in herds where Ikeda and ticks are present) carries risk. It may be difficult to understand risk because the status of the areas cattle are moved to is not clear, for example, a marginal tick area. A careful assessment of that level of risk should be undertaken before advising on movements of cattle.
- The risk will vary with the physiological status of the cattle moved. For example, pregnant cattle sent off for winter grazing (generally from 1 June) are more at risk than dry stock (although outbreaks in dry stock have occurred).
- Cattle born and raised in the South Island and lower North Island will be naïve and the risk of disease will be high.
- If the status of cattle to be moved is unknown, the risk of disease can be

elucidated by a small prevalence study involving testing 10 randomly selected animals by PCR (10 PCR tests rather than one pooled test; see (3) above).

- Risk will also depend on the status of the destination farm that cattle are being moved to. Understanding risk on the destination farm can be determined by carrying out a pooled PCR for Ikeda (10 animals). A pooled test should indicate the presence or absence of Ikeda on that farm.
- *For a naïve herd it is strongly advisable to send cattle to a farm of similar status.*

6 Mitigating the risk of TABA in naïve cattle that are to be moved into an endemic area

- No steps can be applied to fully mitigate disease risk.
- Acaricides may delay but not truly prevent transmission.
- See (1) above for the important elements of an outbreak response programme.

7 Risk of causing TABA by sending cattle back from an endemic area to the home herd (destination herd) naïve to Ikeda (for example, returning young stock from grazing)

- If ticks are present in the home herd (destination herd), moving these cattle will result in future risk of disease either through import of parasitaemic cattle or through introduction of infected ticks.
- Where ticks are absent from the home herd, other theoretical pathways are available for transmission, such as sucking lice, needles or stable flies; however, these are unlikely to be of significance.
- If the environment is suitable for ticks but they are not present, then there is a risk that the vector may establish.

- Movement of other classes of stock to a naïve farm, for example, sheep or deer (carrying infected larvae or nymphs), may present a risk for transfer of infected ticks to cattle.
- Acaricides may prevent transfer of ticks but will not prevent future transmission if ticks are present on the home farm.

8 Role of buparvaquone (BPQ) in the treatment of TABA

- BPQ has been found to be efficacious against other *Theileria* species; however, when animals are severely affected, it is not a substitute for blood transfusion.
- The long withholding periods (for both meat and milk), time involved in recording and tagging animals, traceability and so on, mean that BPQ may have limited value as a treatment in future.
- The most effective management for dealing with outbreaks of TABA is good husbandry and stock care, and reducing stress in cattle (see (1) above).

9 Value of regular acaricide treatments in cattle as a preventative for TABA and timing of applications

- We do not know the value of using acaricides on cattle in reducing the overall impact of TABA. Strategic and frequent use of acaricides will reduce the relative abundance of ticks, but even then we do not know if this is cost-effective.
 - It is unreasonable to assume that acaricide treatment will be 100 percent efficacious in preventing disease. However, it may reduce risk where the numbers of ticks are low.

- In endemic areas, acaricides are unlikely to be an effective control measure to prevent infections of Ikeda in cattle.
- If acaricides are to be used, a protective treatment around parturition or early lactation is more likely to reduce impact than would a “preventative approach” (treatment in summer and autumn to reduce larval numbers; see “Assumptions” above).
 - The most valuable time to apply acaricides to adult cattle is from July through the calving period with at least two treatments at three-to-four weeks apart. (Treatments in June are unlikely to catch nymphs coming out of diapause.) The principle aim of these treatments is to prevent infection of cattle during the vulnerable period of calving and early lactation.
- Given our assumption of only one infected tick being necessary to induce infection, other management practices to reduce ticks may be very limited in the dairy context.
- Alternatives are available in the beef context, for instance:
 - cross-species grazing (for example, with or without organophosphate-dipped sheep)
 - set stock calving cows or cows with very young calves in to areas where there is less tick habitat, for example, hill paddocks at altitude should be safer than the flats with rushes. Gullies and warmer paddocks are likely to present a higher risk.
- These management options are mainly around preventing blood loss by reducing tick numbers rather than from preventing

exposure to Ikeda. At this point, we have no evidence that these will be successful in preventing disease impact from Ikeda. Further research is necessary in this area.

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Diagnostic tree for anaemia in cattle

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Anaemia present and clinical signs including:

lethargy/depression, pale/white/jaundiced mucous membranes,
fever, possible haemoglobinuria

Regenerative anaemia

1. Is Theileria present on a blood smear or detected by PCR?

2. Is there evidence of haemorrhage?

Trauma or haemorrhage into the body cavities and lungs?

3. Are cattle acutely affected with signs of haemolytic anaemia?

Cu toxicity (Kidney copper levels most diagnostic or check serum/liver levels), post-parturient haemoglobinuria (Low PO₄; check levels), Check ration (Onions? Brassicas? Heinz bodies present on the blood smear?); Leptospirosis (vaccinated? High titres or rising titres?; Pomona mostly affects calves rather than adults).

Acute facial eczema (some animals may have haemoglobinuria; check liver enzymes).

Bacillary haemoglobinuria (Germination of *Clostridium hemolyticum* spores secondary to liver damage, e.g. from migrating liver fluke; very rare).

4. Is more than one animal affected?

If only one animal, Autoimmune haemolytic anaemia? (drugs administered recently?; very rare).

5. Is there a heavy burden of ticks present ?

Particularly in calves check around the neck on white patches.

6. Are there other agents or conditions or a history suggestive of secondary theileriosis?

BVD Ag test on affected animal (+/-Pooled herd/mob antibody test), Management stress through weaning or calving etc, Movement of cattle to or from Northland.

Non regenerative anaemia

1. Is Theileria present on a blood smear or detected by PCR?

2. Are cattle in good condition?

(Cu, B12, Se deficiency; Check levels), Renal disease, Liver disease (Is it a facial eczema or liver fluke area; serum biochemistry/faecal egg test), Chronic disease (Johnes, parasitism).

3. Are only young animals affected?

Genetic disorders (e.g. congenital dyserythropoiesis in Polled Herefords and Murray Grey calves; often apparent from six weeks of age).
Coccidiosis (mild to moderate diarrhoea and weight loss; oocysts in faeces).

4. Are any metabolic disorders present?

(Mg deficiency "Taranaki anaemia").

Farm-level management of *Theileria orientalis* Ikeda outbreaks in dairy cattle

By Daan Vink (Ministry for Primary Industries), Ashley O'Driscoll (Hauraki Veterinary Services), Sarah Briggs (Franklin Vets), David Moors (Franklin Vets) and Kevin Lawrence (Ministry for Primary Industries)

The epidemic outbreaks of *Theileria orientalis* Ikeda in cattle over 2012–13 have received substantial recent media attention, including coverage in *Vetscript* (Watts et al, 2013; Lawrence, 2013).

Because the protozoan agent is transmitted by the cattle tick *Haemaphysalis longicornis*, the affected farms are mostly within the known distribution areas of the tick: the northern and central North Island. On the farm level, the disease causes haemolytic anaemia in animals of all ages, which varies from mild to fatal. The median farm-level prevalence of animals with clinically severe anaemia to date has been about 1.0 percent, with a mortality of 0.25 percent. Peak incidence has coincided with periods of stress in the host cattle (spring and autumn) and/or following periods of high activity of the vector; the spring 2013 epidemic was still escalating at the time of writing.

This article provides a practitioner's farm-level perspective on the disease. It discusses the diagnosis and treatment of the individual patient with severe haemolytic anaemia, presents a screening method to detect other clinically affected animals in the herd and provides recommendations for management of the disease.

Diagnosis and treatment of the individual animal

Clinical diagnosis

The expression of clinical disease is variable and likely depends on factors such as the

health and stress level of the animal, tick burden and infectious dose. The diagnosis is made on the basis of history and clinical signs, which include lethargy and depression, inappetence and reduced milk production; abortion may occur (Eamens et al, 2013). The most characteristic clinical sign is the development of haemolytic anaemia, resulting in pale or icteric mucous membranes (marked jaundice becomes clearly apparent when the packed cell volume (PCV) drops below 12 percent). The vulval or conjunctival mucosae can be used to assess mucosal colour; a colour chart (Figure 3) may be used to estimate the PCV.

After initial examination, a presumptive diagnosis and treatment plan can be made based on clinical signs and an in-house PCV. The diagnosis is confirmed by taking EDTA and serum blood samples; the EDTA sample should be used to perform a blood smear that can be evaluated by the laboratory. Full haematology should be performed, particularly if this is the first case on a particular farm, to confirm that the anaemia is regenerative. Subsequently, if there is any doubt, the serum sample can be used to analyse other parameters of disease.

After an individual diagnosis has been

confirmed on a farm, the incidence of severe haemolytic anaemia should be considered pathognomonic: these animals will urgently require a blood transfusion and should be triaged. It is advisable to arrange a herd screening to identify additional animals meeting the case definition and develop a management plan.

Blood transfusion and treatment

Cows with severe anaemia (PCVs of less than 10 percent) are likely to die unless a blood transfusion is given. A transfusion is recommended for animals with a PCV of less than 15 percent. The effects of a transfusion are direct and gratifying. Although the lifespan of the red cells in the transfused blood is probably only two-to-three days (Soldan, 1999), these animals are in a strong regenerative phase and should rapidly raise their PCV. Transfusion will drastically reduce recovery time and increase the chance of keeping the cow in milk; animals should recover in two-to-three weeks, although weight loss and stress may have protracted impacts.

Three general elements are involved in performing a blood transfusion:

- selecting a donor
- collecting blood
- transfusing the blood into the recipient.

The donor cow should be young (less than eight years old, but ideally four-to-five years old), healthy and as far away from calving as possible (that is, dry and more than a month pre-calving or in milk and more than a month post-calving). The larger and quieter the cow, the better; body condition score should be 4.5 or higher. Take as much volume as possible (four-to-five litres): healthy cows can donate up to eight litres without adverse

effects. It is recommended to do a PCV on the potential donor to ensure they have a normal PCV, because cows can have a PCV as low as 15 percent before looking sick.

Soldan (1999) and Malmo et al (2010) provide excellent instructions for blood collection and transfusion, essential reading for any practitioner performing bovine blood transfusions. Several techniques are described, and a variety of commercial and home-made collection bags and giving systems can be used.

It is the individual practitioner's responsibility to consider which technique is the most appropriate. Here, we present collection (Figure 1) using the jugular cut technique, and transfusion using a pressure sprayer. We choose to do this because it complements the information in the aforementioned references and substantial field experience has shown that this method is quick, practical and efficient when large numbers of transfusions are performed. Whichever approach is chosen, the additional input of an assistant (who may be the farmer) will be required.

While you are preparing the donor, have your assistant administer antihistamine (10mL Antimine) intramuscularly to the recipient. Place the donor into the head bail and restrain tightly with a rope halter, tying the head back at cow shoulder height to expose the jugular vein. Use a small amount of sedation (0.5–1.0mL Xylazine intravenously) if restraint is poor. Clip an exposed area of jugular and surgically prepare the site. Inject about 10mL of lidocaine subcutaneously along an 8cm line directly over the jugular. Give the donor cow a single injection of Engemycin (oxytetracycline), and potentially an iron supplement (for example, Hemo15). Dissolve sodium citrate in sterile saline and place it in

your collection container. Place the funnel on top of the container and give it to your assistant. Make a 6cm incision perpendicular to the jugular groove through the skin and subcutaneous tissue; you should be able to feel the jugular as a distinct tube under your finger. Hold the scalpel half-way up the blade; place the scalpel perpendicular to the jugular and make a stab incision into the vessel. Extend the longitudinal cut (not horizontal) in the jugular

by 1–2cm (anything less is likely to result in clotting during collection): this ensures that blood will run directly out of the incision instead of running down the side of the neck. Hold off the jugular. Your assistant should continuously swirl the blood as it streams into the collection container to prevent clotting. Collection takes about five minutes. Then, release the back pressure on the jugular. Instruct your assistant to close the container

Figure 1: Collection kit



Notes to Figure 1:

Items needed for collecting blood:

- rope halter
- Xylazine 2%
- anticoagulant: 15g sodium citrate and 400mL sterile saline (not Hartmanns)*
- collection container (5L) and a funnel
- lidocaine
- scalpel blades (incise)
- clippers and prep solution
- needle, skin suture, needle holders, scissors, two-to-three swabs

- non-sterile latex gloves
- oxytetracycline (for example, 40mL Engemycin).

Items needed for giving blood:

- 10–12mL antihistamine (Antimine)
- giving set attached to lid of collection container
- 14 gauge needle
- clippers and prep solution
- oxytetracycline (for example, 40 mL Engemycin)
- 5–10mL adrenalalin solution (1mg/mL) in case of transfusion reactions.

* Precise quantities vary slightly between references – this is a practical average.

and continue to gently agitate for one minute. Best practice indicates that this blood will be suitable for use over a 24-hour period.

Depending on the position of the head (low or high) and the pressure of the head bail on the jugular, you may have minimal or significant bleeding from the wound. If you have significant bleeding, have your assistant hold the skin closed while you suture. Alternatively, place a swab in the incision to stop the bleeding while you close (removing the swab as you finish closure). Advise the farmer to monitor the wound for abscesses, and remove the skin sutures two weeks later.

Figure 2: Transfusing a down cow



Note: A closed system is used here, rather than a pressure sprayer.

To give the blood, place the recipient into the head bail and restrain tightly with the halter, exposing the jugular. If necessary, transfusion can be done in the paddock on a down cow restrained with a halter tied to her leg (Figure 2).

Clip an exposed area of the jugular groove and surgically prepare the site. Try to minimise back pressure on the jugular due to the head bail or restraint technique, as this will slow transfusion. Seat the 14 gauge needle securely in the jugular.

Prime the pump, taking care not to over-pressurise. Let blood run through the giving set, then attach the set to the needle. Soldan (1999) recommends running the blood in relatively slowly for the first few minutes: while there is a low risk of transfusion reactions (manifested as respiratory distress, tachycardia, hiccupping, thrashing or sweating), the recipient should be continuously monitored. In case of reactions, inject 5–10mL adrenalin intramuscularly (or 0.2–0.5mL intravenously in case of emergency). The remainder of the blood can be run in over five-to-10 minutes.

Subsequently, remove the needle and give the animal a single injection of penicillin. If there are no concurrent diseases or conditions, she should look noticeably brighter within an hour after transfusion. Rinse the giving set and container thoroughly with very dilute antiseptic solution, and air dry. Restock your transfusion kit.

Buparvaquone treatment is recommended for all transfused animals because it greatly aids recovery and increases the likelihood of the cow staying in milk. This should be administered while the animal is well restrained; it acts almost immediately and there are almost no parasites in the

blood after four days (Carter, 2011). The withholding periods and controls associated with buparvaquone treatment should be discussed with the farmer before treatment. If buparvaquone is used, the animal must be tagged with the blue THL tag and its National Animal Identification and Tracing (NAIT) number read at the time of treatment. Treatment details and the NAIT number must be entered into the Ministry for Primary Industries database within two working days of the animal being treated.

Management of the herd

Herd screening to detect affected animals

If one or multiple cases of theileriosis have been identified, it is advisable to screen the herd. This will give an insight into the prevalence and impact of disease at the time of screening, can serve as a baseline for monitoring and help determine a farm management plan. Such screening can be carried out by farm personnel, provided they have received some training in disease recognition; in addition, these people will often have intimate knowledge of their

animals' behaviour, temperament and performance. Screening is best performed during milking, where cows can be marked and drafted. Young stock can be inspected separately. To record the observations made on a large number of animals, a suggested record sheet can be downloaded from <http://tinyurl.com/ksrmwu5>

Visual inspection of the colour of the vulval mucosa is a convenient and rapid screening test to assess the presence and degree of anaemia. A colour chart for the estimation of the degree of clinical anaemia has been previously developed and validated for use in small ruminants (Van Wyk & Bath, 2002); it has been applied extensively worldwide. Based on this, the Field Anaemia Nearest Indicator (FANI) card has been developed as a practical tool to assist herd screening. A copy of this card can be ordered (see "Useful resources" below). Four categories span the range from healthy to severely anaemic, as well as a category differentiating anaemia due to blood loss (Figure 3). This system was designed to provide sufficient representation of the full range of disease signs but be simple enough to facilitate classification. It enables a cow-side interpretation and decision to be made on the basis of each observation.

To estimate an animal's PCV, hold the vulva open with one hand by grasping the top of the vulva between your thumb and forefinger, and spreading open to inspect. Hold the card against the vulva with the other hand. Evaluate the colour of the mucosa halfway down the vulva (this avoids blanching near your fingers). Note the colour of the mucosa adjacent to the pigmented vulva, as well as the colour gradient from the centre of the vulva to the periphery. Detailed recommendations for the interpretation are given on the card.

Figure 3: Assessment categories of the FANI card (figures are packed cell volume %)



Animals with a PCV of 25 percent to >35 percent do not require follow up, although PCVs should be run for animals showing clinical signs of theileriosis. Some overlap is likely between this category and the mildly anaemic category (20–24 percent PCV). This falls within the range of variation and may have a physiological origin (for example, early lactation (Nazifi et al, 2008)) or a cause unrelated to theileriosis. However, this may also suggest mild theileriosis, so it is recommended to monitor these animals for clinical signs as early as possible to detect any progression. PCVs of 15–19 percent are abnormally low and a confirmatory PCV is strongly advised. These animals may be stable or deteriorate; if they show clinical signs of theileriosis, they will benefit from treatment. Animals with PCVs of less than 15 percent require immediate intervention, as described above.

Management of diseased animals and the herd

Communication with the farmer is essential to minimise the impact of the disease. The management strategy on the herd level will consist of a combination of measures, such as the following.

- Identifying and managing affected animals:
 - frequency of screening for theileriosis cases, plus confirmatory PCV if needed
 - treatment decisions, particularly for more mildly diseased animals
 - stratification of supportive care by the severity of disease
 - monitoring and management of disease in calves.
- Reducing stress and optimising herd health:
 - good management and husbandry,

particularly at key times (dry cow management, calving and so on)

- good nutrition, including mineral supplementation, avoiding feed stress and so on.
- General biosecurity measures:
 - strategic tick control, including pasture management and application of acaricide treatments such as Bayticol
 - limiting animal movements and introductions from *Theileria*-affected farms;
 - monitoring or quarantine of livestock bought in or grazed off the property.

Supportive care for affected animals is of high importance to reduce stress and accelerate recovery. This includes providing cow covers or shelter, situating patients close to the shed, providing a paddock near the shed with adequate good-quality feed and reducing them to once-a-day (OAD) milking. Minimise handling and avoid exertion. Rapid weight loss in clinical cases has expressed itself as clinical ketosis, and Ketol/StarterPlus drenches to affected animals may be advisable in some instances to prevent this.

The farmer should check sick cows every day and suspect or mildly diseased cows at least weekly. If the farm personnel are motivated and competent at performing the screening, veterinary intervention can be limited to confirming the diagnosis and treatment. On farms where whole-herd screening is not feasible, targeted screening of at-risk cows can be performed. This includes cows or heifers still to calve, naïve or bought-in animals, those approaching peak lactation (less than six weeks post-calving) and cows with a body condition score of less than 4. Separating these cows and applying pre-emptive measures, such as OAD

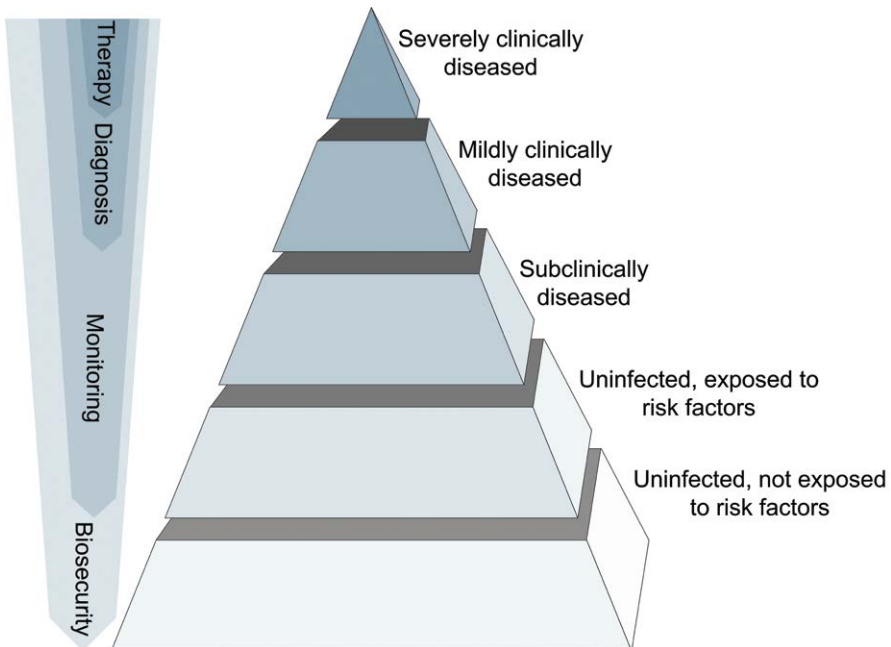
milking and preferential feeding, may reduce the severity of clinical disease and, hence, the need for treatment or further intervention.

Discussion

The clinical manifestation of theileriosis is likely to encompass a spectrum of disease. In analogy with the model of an iceberg or pyramid (Bhopal, 2000; Figure 4), a variable but relatively small proportion of animals becomes moribund and requires immediate treatment. Some may show milder signs, such as decreased milk production and lethargy. Other animals may be infected but not show signs of disease; the remaining animals are uninfected. A screening tool such as the FANI card may facilitate diagnosis and be applied

for monitoring. Monitoring will ensure that mildly diseased, subclinically infected animals and uninfected animals that are exposed to infection and risk factors (for example, the tick vector) are identified as soon as possible, should they develop progressive disease. In addition to monitoring, general biosecurity measures (for example, limiting animal introductions and movements, tick prophylaxis, quarantine and vigilance) are aimed at reducing the exposure to infection and risk factors, so that the course of disease experienced by animals is mild. The combined effect of these measures is to minimise the impact of the disease at farm level, from an animal health as well as a production perspective.

Figure 4: The pyramid of disease, with corresponding management actions at each level



Source: Modified from Bhopal (2000).

The FANI screening tool was developed under field conditions. The system of categorisation was designed to be of practical value in terms of decision making. While the mucosal colours were validated by performing photography and confirmatory PCV tests on about 200 animals, the diagnostic reliability of the tool has not yet been assessed, that is, the repeatability (agreement between repeated observations made on the same animal by the same observer) and reproducibility (agreement between observations made on the same animal by different observers) (Dohoo et al, 2009). Such work can yet be undertaken. However, given the current impact of the disease and the need for screening, it was a priority to make this tool available as soon as possible for field use. The authors would gladly receive feedback on its performance, utility and other constructive suggestions.

At the time of writing, the epidemic appears to be still propagating. It is important for the veterinary profession to provide appropriate advice and recommendations to farmers to minimise the impact, whether farms are newly infected or suffer recidivating disease. Current indications are that peak incidence of clinical disease occurs over two periods:

- autumn, when the parasite burden is likely to be high (vector activity is highest over summer and parasite burden peaks six-to-eight weeks post-infection)
- spring, when the physiological stress of the host animals is highest.

It seems likely that the disease will become endemically stable in areas overlapping with the known tick distribution. Over time, this may mean that the farm-level pattern of disease

will shift: once adult cattle have been exposed and developed immunity, susceptible young stock (particularly calves) will be at relatively higher risk of developing disease. In addition to vigilance during high-risk periods, it is possible that the management strategy will evolve, for instance, to expose calves to controlled natural infection at an early age so they experience mild disease and develop protective immunity, or to expose animals at a time where they are under the least amount of stress.

Useful resources

- Related to this article, the following resources are available from:
<http://tinyurl.com/ksrmwu5>
 - for FANI cards contact **daan.vink@mpi.govt.nz**
 - screening record sheet
 - image gallery and video of collecting and transfusing blood
 - blood transfusion volume calculator.

Briggs S (2014). *Blood transfusion in cattle*. Proceedings of The Society of Dairy Cattle Veterinarians of the NZVA Annual Conference, 2014, 3.16.1-3, 2014: see video demonstration of blood transfusion technique at: **www.nzva.org.nz/theileria**

- Web-based information on *Theileria* and anaemia in cattle:
 - Ministry for Primary Industries: **<http://tinyurl.com/ksrmwu5>**
 - DairyNZ: **<http://tinyurl.com/k796mlf>**
 - NZVA: **<http://tinyurl.com/mmtd7p>**

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Theileria orientalis (Ikeda) associated bovine anaemia: The epidemic to date

By Kevin Lawrence, Andrew McFadden and David Pulford (Ministry for Primary Industries)

Theileria orientalis is a vector-borne protozoan of cattle that can cause anaemia, jaundice, abortion and death in naive or stressed cattle (Shimizu et al, 1992).

In New Zealand, the main vector is the cattle tick *Haemaphysalis longicornis*, although biting flies, sucking lice and iatrogenic transmission could also be involved in local spread (Heath, 2013). Until recently, anaemia associated with *T. orientalis* in New Zealand had been considered to be a sporadic event that only affected cattle debilitated by another condition (Thompson, 1998; McFadden et al, 2011).

T. orientalis was first identified in New Zealand in 1982 in a dairy farm near Wellsford (James, 1985). The associated epidemic covered the period 1982 to 1986 with cases and outbreaks peaking at 60 in 1985. It was believed that *T. orientalis* had been imported into New Zealand by subclinically infected cattle (Thompson, 1991). More recently, from 2000 to 2012, there have been only 12 notifications to the Ministry for Primary Industries (MPI) of *Theileria*-associated bovine anaemia – on average one report a year and for most of these historic reports, there was generally only one animal affected.

Since late August 2012, there have been more than 700 notifications of *Theileria*-associated anaemia, and the mortality and morbidity rates experienced by many of the affected herds have been far greater than ever before. Molecular strain typing has established that a previously unrecorded strain of *T. orientalis*, *T. orientalis* (Ikeda) is significantly associated

with these outbreaks, OR = 15 (95 percent CI = 3–131).

The clinical signs seen with *T. orientalis* (Ikeda) infection are mostly a consequence of the degree of anaemia experienced by the affected animal and include lethargy, inappetence, pale udders (observed at milking), exercise intolerance, pale gums and mucous membranes, jaundice, abortion and stillbirth. Deaths are more commonly seen around times of stress such as calving or early lactation.

Haemoglobinuria is not a common finding – in an investigation of five dairy herds in Waikato presenting with haemoglobinuria, only one herd was positive for *T. orientalis* (Ikeda). The average number of recorded deaths in a survey of 122 affected farms was 0.8 percent with a range of zero to 19.2 percent. The average period prevalence of clinical disease for the duration of the outbreak was 2.75 percent with a range of 0.0 percent to 33.4 percent.

The average prevalence of anaemia, as a measure of subclinical disease, was 33 percent with a range of 10 percent to 88 percent. In one severely affected herd followed longitudinally over time, the anaemia persisted for eight months. For this herd, the sub-optimal performance associated with prolonged anaemia was devastating, with a 25 percent

empty rate, conception rate of only 30 percent and a milk solid production of 80kg less per cow per year. This is expected to be the area of greatest financial cost in other affected herds. The disease outbreaks often follow the same pattern as described by McFadden et al (2011). That is, the disease occurs in presumed naive cattle, subsequent to their mixing with infected animals or moving to an area with infected ticks.

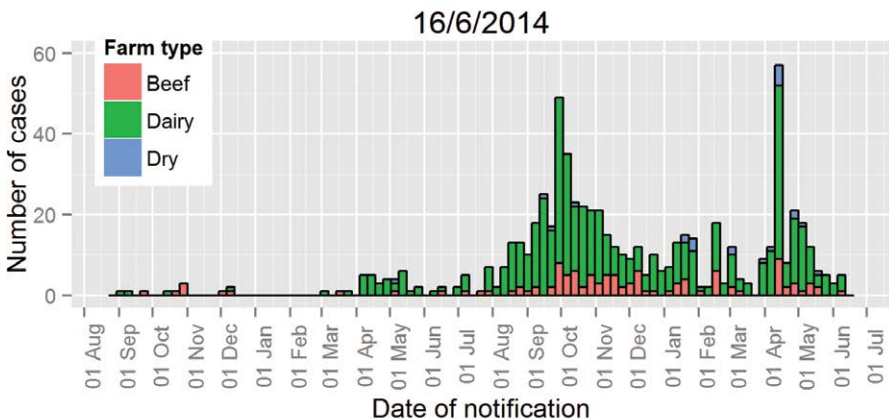
However, there is one important difference that impacts on the timeline leading up to clinical disease. The populations of naive ticks in an area have to first be infected themselves before they can in turn infect naive cattle. This has occurred primarily through the movement of parasitaemic cattle, initially from Northland to the Auckland and Waikato regions and latterly to the rest of New Zealand. For each of two herds in Rangiora and Eketahuna, an individual cow became infected after the movement of parasitaemic animals into the herd. However, the impact of the disease

both within these herds and on neighbouring properties was minimal, which we believe reflects the absence of a tick vector in these areas.

The epidemic curve in Figure 1 shows three distinct phases: spring 2012, autumn 2013 and spring 2013. The first phase of the outbreak in spring 2012 mostly involved young calves (two to four months old) on beef farms in the Northland region. The second phase, in autumn 2013, almost exclusively involved mixed-age dairy cows, often in split or autumn calving herds in the Auckland and Waikato regions. The third phase, which is ongoing, involves the spread of disease to dairy farms in the Waipa, Bay of Plenty, Manawatu and South Taranaki regions. A small number of beef farms continue to be affected but, at present, the greatest impact of *T. orientalis* (Ikeda) is with the dairy industry.

The cases of *Theileria*-associated anaemia that occurred in beef calves born onto Northland properties in the spring of 2012 were the first

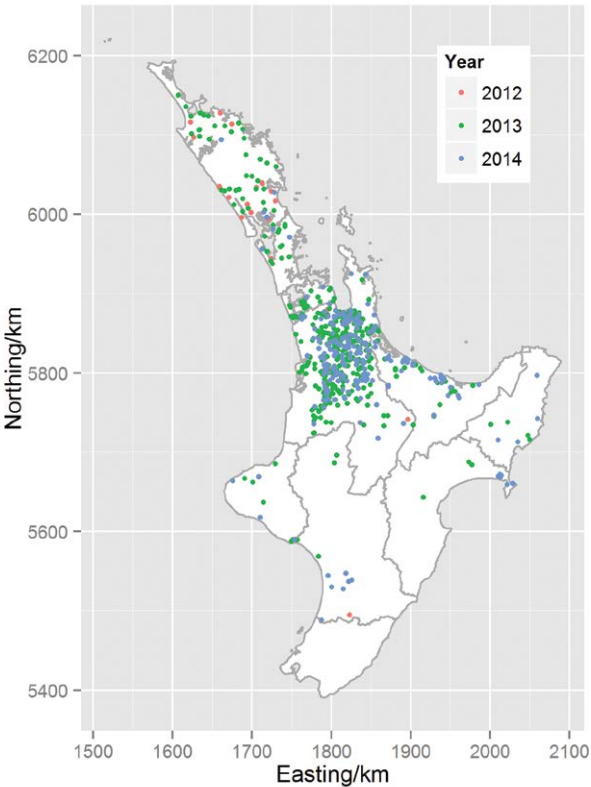
Figure 1: Epidemic curve for cases of *Theileria orientalis* (Ikeda) associated bovine anaemia by type of farm enterprise affected



cases of anaemia associated with *Theileria* reported in calves in New Zealand. Spatial temporal modelling has shown that there is significant local spread of infection for up to 15km and within 30 days of a farm being infected. Methods of local spread are currently being investigated but could possibly involve stable flies, *Stomoxys calcitrans*, mechanical transference of infection or rabbits and hares physically transporting infected ticks. Local spread is suggested as the cause of outbreaks on farms where there has been no recorded cattle movement onto the affected properties. As a result of our investigations into the Ikeda strain, we have increased our understanding of

the epidemiology of *Theileria* in general. Figure 1 clearly illustrates the change in affected farm type from beef to dairy farms as the epidemic progressed. The change in spatial distribution is shown in Figure 2. *T. orientalis* (Ikeda) associated bovine anaemia was first reported in New South Wales, Australia, in November 2006 (Izzo et al, 2010) and has since spread by cattle movements down the coast to southeast Victoria and overland into Western Australia. This is the same strain of *T. orientalis* (Kamau et al, 2011; Eamens et al, 2013) as associated with the New Zealand disease outbreak.

Figure 2: Distribution of cases by time showing migration of cases of *Theileria orientalis* (Ikeda) associated bovine anaemia from Northland in 2012 into the Auckland and Waikato regions in 2013



Until recently, suggested anti-microbial treatments of diseased animals have either been 20mg/kg of long-acting oxytetracycline given every second day for three treatments or 10mg/kg of short-acting oxytetracycline given daily for up to five days. Anecdotal evidence from practitioners is of mixed success using either of these treatment regimes.

For severely anaemic animals (PCV<0.12), blood transfusions have proved to be highly effective, and many practices are now routinely carrying these out on severely anaemic or recumbent dairy cows. Unfortunately, blood transfusions are less likely to be a feasible economic option for beef farmers.

The recent importation of buparvaquone on a special licence now offers some practitioners a genuine anti-*Theileria* pharmaceutical, however, extended milk and meat withdrawal periods mean that its use has to be carefully monitored. Even so, excellent responses to treatment have been reported by day four.

The strategic use of pour-on tickicides to manage the exposure of naive cattle to infected ticks, especially around calving, is also an important tool. Ideally, tick treatments should be given twice, three-to-four weeks apart, with the first treatment given three weeks before calving. For split calving herds, the autumn calving herd may also need treating before calving in the autumn. Ideally, the decision to use tickicides should form part of a whole-farm plan that would also include pasture management to limit tick habitats and culling of feral deer to remove alternative tick hosts. Where the climatic conditions are most favourable for high tick abundance, such as in Northland, then endemic stability for the disease will be rapidly achieved and most disease outbreaks will be restricted to those

naive herds that relocate into these areas.

Practitioners should work with these clients to manage the exposure of the naive animals to infected ticks such that the peak exposure does not coincide with the most stressful periods such as calving or weaning. However, for areas where tick abundance is variable or patchy, such as lower Waikato, then areas of endemic instability could develop (Bill Pomroy, pers. comm.).

Disease scenarios in these areas will not only involve moved naive herds but also disease breakdowns in herds that have remained unchanged in the locale and by default have been assumed to be immune. In the latter case, a decline in tick abundance, triggered by weather or pasture improvements, could cause the herd immunity to fluctuate with tick numbers. In the endemically unstable areas, practitioners will need to be constantly aware of the potential for the disease to re-emerge. For veterinarians in non-tick areas, there will only be sporadic individual cases post-movement and careful history taking should avoid missing these cases. However, it is highly likely that the few sick individual cows will need transfusing so these practices need to be prepared.

The emergence of *T. orientalis* (Ikeda) represents a severe challenge to the New Zealand cattle industry, and there is an urgent need to develop tools for monitoring herd immune and infection status, especially for the endemically unstable areas.

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Tick biology drives infestation

By Nita Harding (DairyNZ animal husbandry team leader), Eric Hillerton (DairyNZ chief scientist) and Allen Heath (AgResearch senior scientist)

Only one tick affects livestock in New Zealand and this is the New Zealand cattle tick, *Haemaphysalis longicornis*. This tick originates from eastern Asia and was introduced into New Zealand more than a century ago, most probably on imported cattle.

The tick will feed on all ruminants and has also been reported on other mammals and birds. As with all ectoparasites (parasites that live on the surface of the host), a heavy infestation can cause anaemia, considerable local skin irritation, some loss of body condition and, very occasionally, death (particularly in young animals).

In dairy cows, heavy tick infestations have been suggested to cause a reduction in milk production. Sheep with heavy tick infestations will rub due to irritation from ticks, reducing the quantity and quality of the wool clip. Heavily infested sheep may also lose weight.

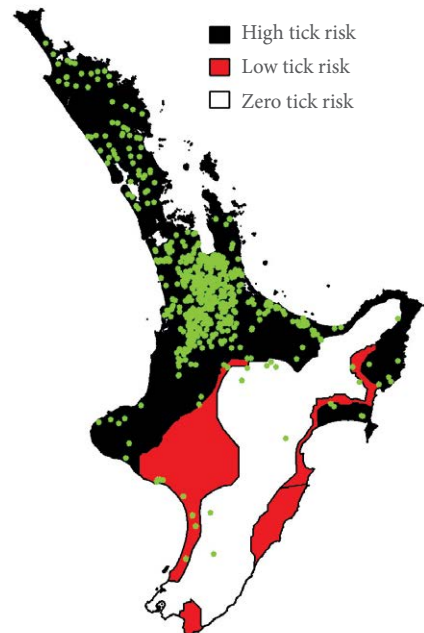
The tick is distributed in much of the North Island of New Zealand, but is more prevalent in warmer, northern regions. It has been recorded in Northland, Auckland, much of the Waikato, Bay of Plenty, Gisborne and Taranaki, but also occurs in Hawkes Bay, Wairarapa and down through the Manawatu. Distribution in the South Island is limited, with the tick present in Marlborough, Nelson and Takaka.

Reports of tick occurrence are, however, limited by how close observations have been. The juvenile ticks, especially the larvae, are very small and therefore not obvious especially where their dark colour camouflages them against a dark skin surface. Ticks feed from lesions they produce in the skin and imbibe fluids that pool there.

Tick distribution

Although ticks occur in the regions shown in the map (Figure 1) they can be transferred on animals to tick-free areas of New Zealand. All cattle in areas where ticks are present are at

Figure 1: Map of distribution of the cattle tick with *Theileria orientalis* Ikeda cases (green dots). This shows a correlation between tick distribution and clinical cases of bovine anaemia caused by *Theileria orientalis* Ikeda.



risk of being infected with *Theileria* (Figure 1).

Tick activity

The cattle tick has four developmental stages – egg, six-legged larva, eight-legged nymph and eight-legged adult. Generally, the tick completes one life-cycle (generation) per year. However, in warm and moist conditions more than one generation may occur.

The New Zealand strain is parthenogenetic (reproduces asexually), so only adult female ticks are present.

Tick populations show a distinct seasonal pattern. Eggs are usually laid from early October to late January. These hatch into larvae after about 70–100 days depending upon temperature. The larvae are active from early January to late March and once fed larvae drop into the sward base and moult to become nymphs.

Nymphs first start to feed in early July. They derive from fed larvae that moulted in late summer/early autumn and go into behavioural diapause after that; which means they cannot be induced to feed at that time. Nymphs are most common through to late October. Adults first appear in early October, with their peak period of activity in late November to early January.

The tick spends most its life at the bottom of vegetation. Each stage, apart from the egg,

must feed on blood from an animal host, but only once, before development into the next stage. The period of time spent on the host animal is short – larvae feed for up to nine days, nymphs for 3–8 days and adults for 7–14 days. After feeding, the tick falls to the ground and develops (moult) into the next stage around 3–6 weeks later, or it can be longer, if temperatures are low. Unfed ticks can survive for at least 12 months in New Zealand and nymphs have been found alive in Japan after 2 years in soil.

When hardened-off after moulting and hungry, the larva, nymph or adult climbs up a plant stem to wait for a passing host; although grazing stock will put their muzzles in amongst ticks on pasture as well. The tick senses the presence of a host by warmth, movement, change in light and carbon dioxide gradients. This is called “questing”. They grasp onto the host, using their front legs. Once on the host, they will feed anywhere on the skin surface but the softer-skinned and better protected areas such as around the udder, under the tail and in the ears seem to be preferred. Tick behaviour is broadly controlled by hunger, weather and day length. This tick does not like overly-hot, dry conditions, being most active when the temperature is between 10–30°C and humidity is above 60 percent. Longevity is controlled by fat reserves and desiccation (water balance).

Figure 2: Life-cycle of the cattle tick. Left is the nymph (1.5 mm), centre is the unfed adult tick (3 mm) and right is the fully fed adult tick (6 mm)



In hot, dry conditions this tick and especially larvae have a short life if they do not encounter a host.

In cool and moist conditions, a tick will survive many months waiting for a host. Thus this tick is found in cold and snowy winter environments such as Hokkaido, Japan's large northern island and home of their dairy industry, where winter snowfall can easily be a metre deep.

Larvae are red-brown, small and not easily seen on cattle. Measuring up to 1.5 mm, nymphs and females are bigger and also red-brown in colour. Unfed adult ticks are about 3 mm long and fully-fed adults can be up to 6 mm in diameter. Large numbers on cattle are obvious.

Managing ticks within the farm system

Heavy tick burdens can have a severe impact on young animals. Deaths of fawns within a few days of birth, as a result of anaemia, were a particular problem for the deer industry in the 1980s and still occur at times. Cattle have also died from overwhelming tick infestations.

Because ticks spend most of their life off animals, controlling them by treating hosts has a limited impact on the total population of ticks on a farm.

It may be important to deal with large infestations on animals and a new and immediate threat by chemical control. Understanding the conditions that favour ticks, and managing pastures and grazing to reduce tick survival in the pasture, will be more effective long-term.

Control of ticks on animals

Inspecting animals for ticks is recommended as part of general biosecurity precautions when animals arrive on a farm. Even if no ticks are

found, all animals should be treated and then quarantined for 7–28 days, depending on the disease threat. If ticks are present, the risk of theileriosis can be assessed and precautions taken to protect the resident herd, if necessary.

Currently, two tick treatments are licenced in New Zealand. When used according to the manufacturers' recommendations, especially frequency and quantity of use, the products have a nil milk and meat withholding period.

As synthetic pyrethroids are the only ectoparasiticide active ingredients registered for use on deer and cattle, overuse is discouraged, as this could hasten a resistance to the chemicals and leave farmers with no suitable products for treating lactating dairy cows.

Control of ticks by pasture management

Ticks require adequate cover for survival. Long, rank pasture, fern, scrub, rushes and sheltered areas along hedges favour tick survival. Newly developed or closely grazed pastures are unlikely to offer much protection for the various stages of the tick life-cycle.

Well-managed dairy pastures may have little in the way of suitable tick habitat. Pasture at runoffs may be of more concern, as it may not be grazed as intensively and may have rougher areas that are difficult to graze closely.

The longer grass around the paddock's edge after a crop has been harvested (silage, hay or maize) can be an area where favourable tick habitats exist. After not being grazed for some time, this may contain large numbers of ticks awaiting hosts.

Warm, moist conditions enhance tick survival and development. When seasons are unusually warm (winter 2013) or when paddock conditions remain moist, more ticks

will survive. Farmers in areas of the country where ticks occur may find it useful to evaluate the tick risk of their paddocks, based on how favourable conditions at the base of the pasture are for ticks.

Longer grass and sheltered paddocks are likely to be higher risk and the first grazing of these paddocks may be best done by older stock or sheep, rather than more susceptible younger stock such as calves.

Estimating tick abundance and activity is best done by a blanket drag.

A white woollen blanket or piece of corduroy, about 50 cm wide, stiffened by a pole or cane, and up to 1 metre long, is dragged by a string along a series of 5 metre strips of vegetation until ticks are found.

Adult and nymphal ticks that attach to the blanket can be counted. Larvae are best counted and removed at the end of the series of drags because they are not easily dislodged from the blanket during sampling, and are easily overlooked at the end of each sample drag because of their size.

Blanket dragging over different paddocks will give an indication of the numbers of ticks and relative risk of different parts of the farm.

Once ticks are established in an area, it is unlikely they can be eradicated, unless extremes of temperature and drought are experienced, and even then some may survive. Though ticks do not move far once they fall from a host to the base of the pasture, other animals, including birds, but more likely hares and perhaps feral deer, dogs and horses can transfer ticks from one property to another. It is considered that local spread of up to 5 km or more can occur this way.

Pasture management to reduce tick numbers

1. Plan ahead.
2. Prepare pastures for susceptible stock one year in advance, by grazing to low residuals during December to March. This can be done sequentially so that not too much land is out of production at one time.
3. Have paddocks/breaks a suitable size for the mobs of animals, so they are grazed evenly and to the desired residual (1500–1600 kg DM/ha).
4. Avoid putting young stock in paddocks where tick populations are likely to be high (rougher, longer pasture or pasture that has not been grazed for some time).

Further reading

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