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Bacterial Tick-Borne Diseases of Livestock Animals

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Additional information is available at the end of the chapter

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Abstract

Bacterial tick-borne diseases (BTBDs) are very significant in practical one health medicine. In contrast to the restrictions related to diagnostic and clinical application, the control and prevention of bacterial tick-borne diseases are difficult because they require the disruption of a complicated transmission chain, involving vertebrate hosts and ticks, which interact in a constantly changing environment. Q fever, rickettsiosis, borreliosis, ehrlichiosis, anaplasmosis and tularemia are BTBDs, which are discussed in this chapter. Epidemiology, clinical symptoms, diagnosis and prevention subtopics are planning to be prepared under main topics. This chapter presents a brief background of key livestock BTBDs and ticks and reviews the general aspects of BTBDs to identify topics in knowledge and understanding of these diseases, propose areas for future research and draw attention to the need for improved tools for the diagnosis and control of BTBDs.

Keywords: tick, bacterial zoonoses, Q fever, rickettsiosis, borreliosis, ehrlichiosis, anaplasmosis, tularemia

1. Introduction

Bacterial tick-borne diseases (BTBDs) affect the productivity of livestock animals in various regions of the world, leading to a significant adverse impact on the production of resource-poor farming communities. Hence, the livestock industry has become an integral part of world economy, and the large number of dairy cattle is being imported between continents in order to meet an increasing demand of meat and dairy products, it is essential to review current status of bovine BTBDs and to identify diagnosis and prevention in the knowledge of BTBDs and their prevention. Although there has been a recent increase in the number of studies of BTBDs in

various geographical regions, information on their prevalence, distribution, tick vectors and control is limited.

2. Bacterial tick-borne diseases of livestock animals

2.1. Q fever

Q fever is a zoonosis associated with *Coxiella burnetii* that is an obligate intracellular parasite classified within the family Rickettsiaceae and which can be divided into six genomic groups based on restriction fragment length polymorphism. Unlike the other members of Rickettsiae, *C. burnetii* is quite resistant to environmental influences and is not dependent upon arthropod vectors for transmission. *C. burnetii* exhibits two antigenic phases: phase I and phase II (**Figure 1**). Phase I organisms are more infectious. The organism has worldwide distribution, although a large serological survey argues that it is not present in New Zealand [1].

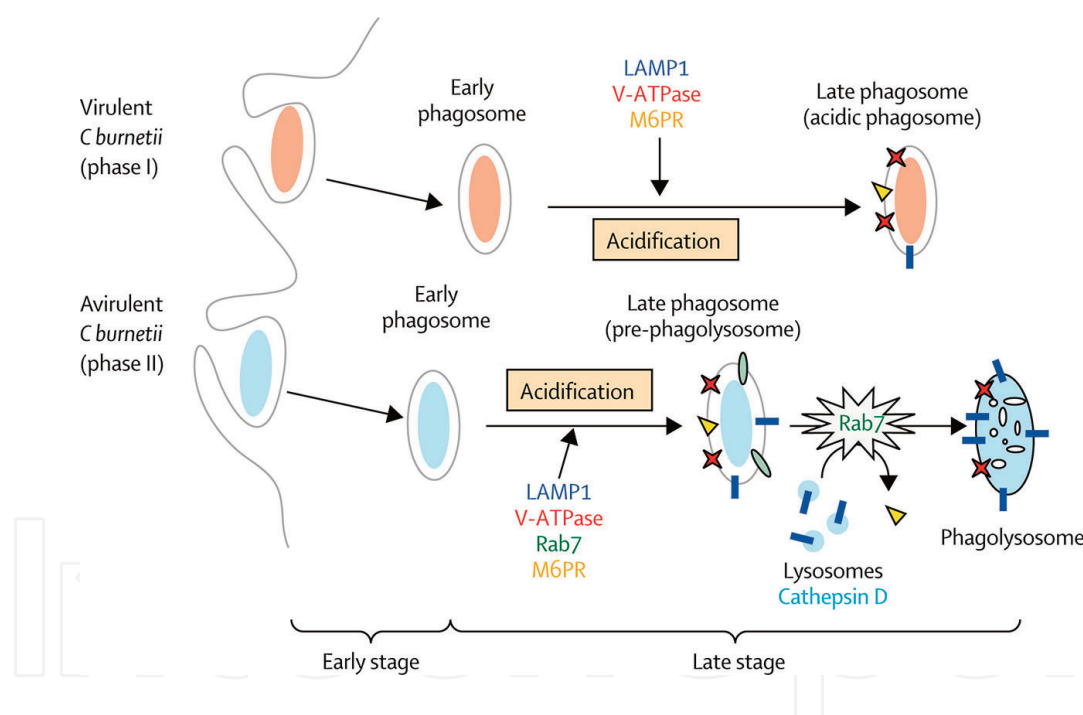


Figure 1. *Coxiella burnetii* mobilization in macrophages [2].

C. burnetii cycles in a wide variety of wildlife species and their ectoparasites. The infection also cycles in domestic animals. Rates of infection in farm animals vary considerably between locations, between countries and with time as there appears to be cycles of infection within regions [3].

In cattle, prevalence figures range from 6 to 82% of cattle and 23 to 96% of herds seropositive depending upon location and country. Seropositivity rates in sheep and goats are similar but also vary according to year and region. There is little information on management or other

factors that might influence this variation in prevalence but one study found a significantly higher prevalence in housed cattle compared to cattle kept at pasture. The transmission of infection is spread by direct contact and inhalation. Infection of non-pregnant animals is clinically silent and is followed by latent infection until pregnancy when there is recrudescence with infection in the intestine, uterus, placenta and udder and excretion from these sites at parturition. The organism is present in high concentration in the placenta and foetal fluids, and subsequent vaginal fluids are also excreted in urine and are present in the faeces of sheep from 11 to 18 days post-partum [4, 5]. Infection can result in abortion, stillbirths or poorly viable lambs but commonly the neonates of infected, excreting, ewes are born clinically normal. Abortion usually does not occur at successive pregnancies but there can be recrudescence of infection and excretion at these pregnancies, especially the one immediately following [6].

Goats also excrete the organism in vaginal discharges for up to 2 weeks, and it is present in goat milk for up to 52 days after kidding and also in faeces. Maximum shedding in cattle also occurs at parturition and for the following 2 weeks but cattle excretes the organism in the milk for at least several months and up to 2 years and infection is common in bulk tank milk [7–10].

There is strain variation in the organism and differences in plasmid sequence types have been correlated with differences in the type of disease occurring in humans. The organism is highly infectious, and it is estimated that the infective dose for humans approximates one organism. Zoonotic implications in human infection is primarily by inhalation. Sources of infection include such diverse materials such as soil, air-borne dust, wool, bedding and other materials contaminated by urine, faeces or birth products of animals. The potential for human infection from these sources is substantial; for example, ovine manure used as a garden fertilizer has been incriminated as a source. Sheep have traditionally been incriminated as the major reservoir of infection for humans, but the trend for urban populations to locate in close proximity to large dairy herds suggests that cattle could become an increasingly significant reservoir [11–13].

The organism is found in the milk of infected livestock. A significant proportion of seropositive cattle excrete the organism in milk and periods and duration of excretion are variable but may persist at least 2 years. Rates of seropositivity in humans vary markedly between surveys, but there is a higher rate of seropositivity in people (farm workers, veterinarians, livestock dealers, dairy plant and slaughter house workers, shearers, etc.) that are associated with domestic animals and their products and with farm environments [14, 15]. Several incidents of infection in humans have been linked to exposure to parturient sheep and goats [16].

Infection of ruminants can occur at any age and is usually clinically unapparent. In the experimental disease in cattle, anorexia is the only consistent clinical finding. Abortion occurs during the latter part of the lambing period in the flock and in the latter period of pregnancy in individual ewes. The dam shows no signs of impending abortion. As with sheep, infection in goats can be accompanied by abortion, but abortion in cattle is rare although it is recorded. Correlations between herd level seroprevalence and herd fertility are equivocal. There are a number of serological tests available including complement fixation, microagglutination, enzyme-linked immunosorbent assay (ELISA) and indirect immunofluorescence (IF). The IF assay is used as the sero-reference test for the serodiagnosis of Q fever. It can detect antibody

to phase variants and can provide epidemiological information as phase I antibody is associated with recent and acute infections and phase II antibody with chronic infections [17].

There are seldom gross lesions in aborted fetuses, but foci of necrosis and inflammation are occasionally seen in the liver, lung and kidney microscopically. The placenta from aborting animals is usually thickened and a purulent exudates or large, red-brown foci of necrosis are typically seen in the thickened intercotyledonary areas. Microscopically, large numbers of necrotic neutrophils are usually visible on the chorionic surface and swollen trophoblasts filled with the organisms can also be found in well-preserved specimens. Examination of placental impression smears stained with Gimenez, Koster's, or other appropriate techniques provides a means of rapid diagnosis. However, care must be taken to avoid confusing *Coxiella*-infected trophoblasts with cells containing *Chlamydophila* organisms. Coxiellosis can be confirmed fluorescent antibody staining of fresh tissue or immunohistochemical staining of formalin-fixed samples. In most laboratories, culture is not attempted due to the zoonotic potential of this agent. Polymerase chain reaction (PCR) is the most accurate tool for the diagnosis of infectious abortions. In a previous study, six (4.3%) samples were detected PCR positive out of 138 samples [18]. In another research, *C. burnetii* gene was detected in 34.66% of the samples taken from 200 cattle, 200 sheep and 200 goats in the Aegean region of Turkey [19]. In a multidisciplinary research made with veterinarians, farm workers and butchers, among 92 people, 32 (34.8%) and 9 (9.8%) people were positive and equivocal by ELISA and immunoglobulin G (IgG), respectively. The ELISA positive and equivocal sera were studied further by the immunofluorescence antibody (IFA) test, and seven (7.6%) cases were confirmed with immunoglobulin M (IgM), 39 (42.4%) cases were confirmed with IgG. There was no significant difference for Coxiellosis seropositivity among the profession groups ($p > 0.05$). Only four (4.3%) cases were confirmed with PCR positive [20].

Aborting animals should be isolated for 3 weeks and aborted and placental contaminated material burnt. Ideally, manure should be composted for 6 months before application to fields. Feed areas should be increased to keep them free from contamination with faeces and urine. While Q fever has significant implications for human health, it is not significantly important enough to have generated national or regional control strategies based on control in the animal population [21–23].

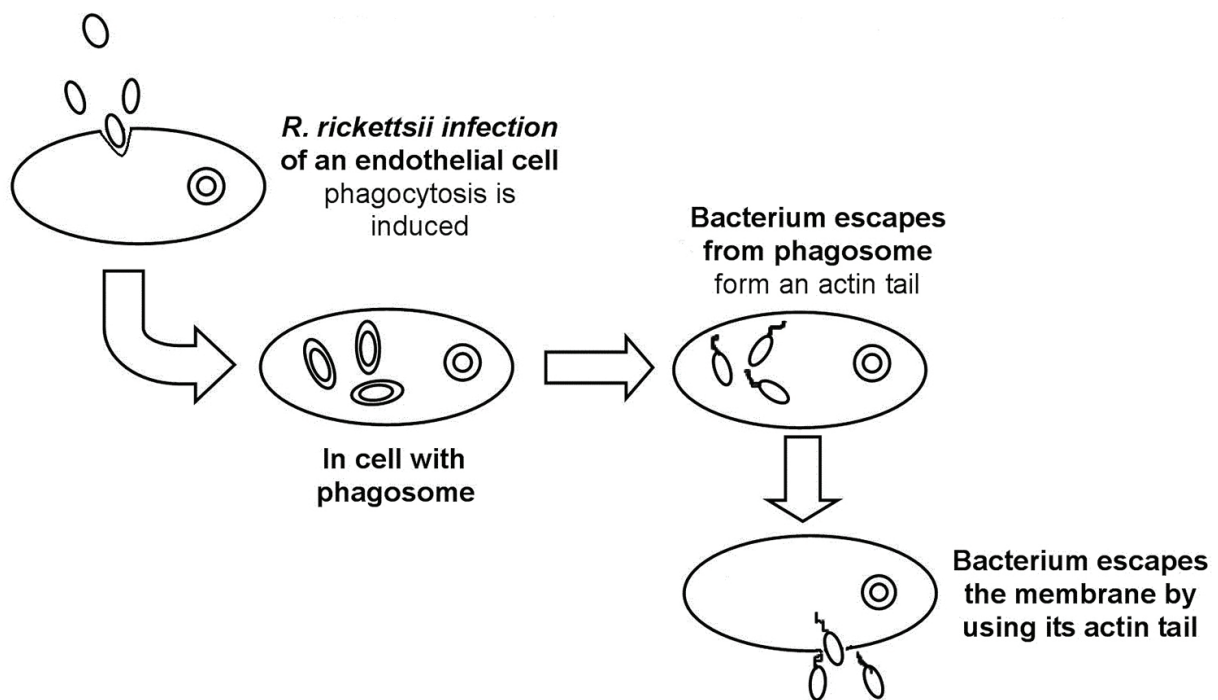
Milk and milk products should be pasteurized. Veterinarians dealing with herds that provide raw milk should ensure that these herds are seronegative for *C. burnetii*. Vaccine trials with killed vaccines in animals show a good and persistent antibody response and suggest that vaccination can limit the excretion of the organism. However, there is little economic incentive for a vaccination programme involving livestock, and livestock vaccines are not available in most countries [24].

2.2. Rickettsiosis

The members of the family Rickettsiaceae have cell walls similar to those of other Gram-negative bacteria. Ultra structural studies have shown that the Anaplasmataceae family have outer membranes but lack an obvious peptidoglycan layer [25]. Organisms in the family Rickettsiaceae, referred to as rickettsiae, generally target endothelial cells. Although several

new species of rickettsiae have recently been identified in domestic animals using molecular techniques, their pathogenicity is uncertain and currently the only species of veterinary importance in the family Rickettsiaceae is *Rickettsia rickettsii*, the causative agent of Rocky Mountain spotted fever. Many *Rickettsia* species including the causal agents of typhus (*R. prowazekii*), murine typhus (*R. typhi*) and scrub typhus (*R. tsutsugamushi*) are primarily human pathogens. These highly pathogenic organisms have a predilection for the endothelial cells of small blood vessels, resulting in vasculitis and thrombosis in many organs. *Rickettsia* species produce phospholipase that damages the membranes of phagosomes allowing the organisms to escape into the cytoplasm (**Figure 2**). *R. rickettsii* replicates in both the cytoplasm and the nucleus of host cells, inducing cytotoxic effects [26].

Definitive classification of the members of the Rickettsiales is based on 16S ribosomal ribonucleic acid (RNA) sequencing, lipopolysaccharide content and metabolic requirements. In diagnostic laboratories, identification of these organisms is based on the species affected, cell predilection, microscopic appearance and molecular techniques. Some members of the Rickettsiales can be cultured in embryonated eggs or tissue culture cells. These difficult procedures are usually performed only in laboratories engaged in research or vaccine production [28].



Made by C. Gibson

Figure 2. Infection diagram of *R. rickettsii* [27].

R. rickettsii affects mainly humans and dogs. *Rhipicephalus sanguineus* and *Amblyomma cajenense* are the main vectors in Central and South America. Ticks acquire the pathogen while feeding on infected small wild mammals [29].

An infected tick must remain attached for up to 20 hours before salivary transmission to the host occurs. The organisms, which replicate in endothelial cells of infected dogs, produce vasculitis, increased vascular permeability and haemorrhage. Rocky Mountain spotted fever should be considered in dogs with systemic diseases, which have been exposed to ticks in endemic areas. Indirect fluorescent antibody test (FAT) or ELISA demonstrating an increasing antibody titre to *R. rickettsii* is diagnostic. Antibodies are not demonstrable until at least 10 days after infection. A marked thrombocytopenia and leucopenia may be present during the acute phase of the disease. The disease must be differentiated from acute canine monocytic ehrlichiosis. PCR detection in tick tissues has been described by a number of workers. Tetracycline therapy, which usually produces clinical improvement within 24 hours, must be continued for 2 weeks. Supportive therapy is necessary for severely debilitated dogs. Frequent removal of ticks is recommended. Because the disease is zoonotic, gloves should be worn during this procedure or a forceps should be used [30].

Ticks acquire the pathogen while feeding on infected small wild mammals. *R. rickettsii* is maintained in the tick population by transovarial and transstadial transmission and thus the tick acts as both a reservoir and a vector of the organism. An infected tick must remain attached for up to 20 hours before salivary transmission to the host occurs. The incubation period of the disease is 2–10 days and the course is usually less than 2 weeks. Clinical signs include fever, depression, conjunctivitis, retinal haemorrhages, muscle and joint pain, coughing, dyspnoea and oedema of the extremities [31].

2.3. Borreliosis

Borreliae, which are longer and wider than other spirochaetes, have a similar helical shape. In addition to a linear chromosome, which is unique among bacteria, borreliae possess linear and circular plasmids, some of which appear to be essential for growth and survival of the organism. Although these spirochaetes can cause disease in animals and humans, subclinical infections are also common. Borreliae are transmitted by arthropod vectors. Arthropod vectors are responsible for transmission of *Borrelia* species in animals. Borreliae are obligate parasites in a variety of vertebrate hosts. Although these organisms persist in the environment for short periods, they depend on vertebrate reservoir hosts and arthropod vectors for long-term survival. Associations of certain *Borrelia* species with particular arthropod vectors and reservoir hosts are important in determining the epidemiology of infections with *Borrelia* species. After entering the bloodstream of a susceptible host, borreliae multiply and are disseminated throughout the body (**Figure 3**). Organisms may be demonstrated in joints, brain, nerves, eyes and heart. Whether disease is caused by active infection or by host immune responses to the organism is unclear. Persistent infection leading to the induction of cytokines may contribute to the development of lesions [32]. There may be an association between different genotypes of *Borrelia burgdorferi* and particular clinical syndromes in humans; *B. burgdorferi* sensu stricto (s.s.) is frequently associated with arthritis, *B. garinii* with neurological disease and *B. afzelii* with skin disease [33, 34].

Chickens have been infected experimentally, and it was found that these animals quickly became immune to *B. burgdorferi* s.s. and did not show any clinical symptoms [36]. More recent

studies have shown that pheasants can function as reservoir hosts of *B. garinii* and *B. valaisiana* in the United Kingdom (UK) [37], but no symptoms of disease in infected birds have been reported.

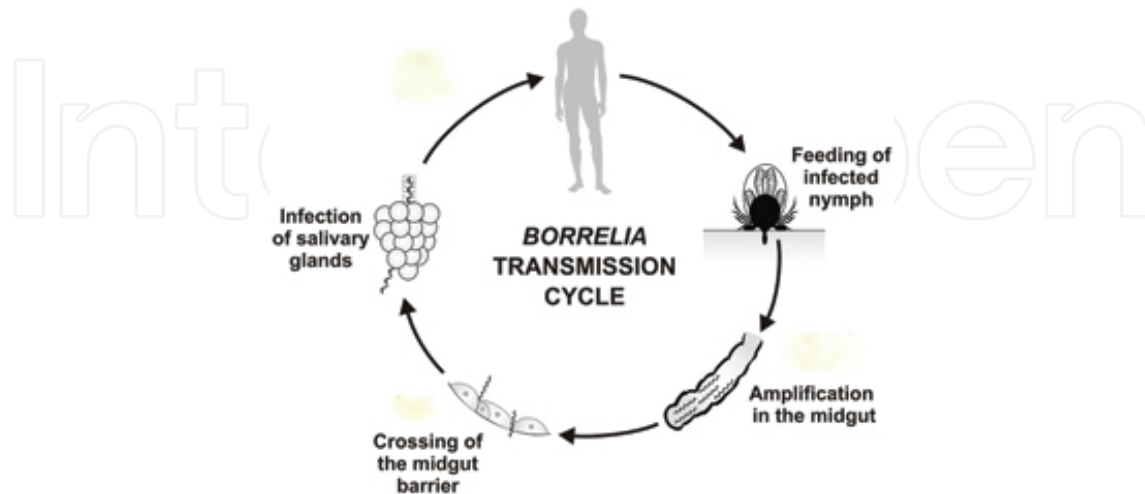


Figure 3. Life cycle of *Borrelia* spp. [35].

Most infections are subclinical. Serological surveys demonstrate that exposure is common in both animal and human populations in endemic areas. The clinical manifestations of Lyme disease are mainly related to the sites of localization of the organisms. Clinical disease is reported frequently in dogs. Symptoms include fever, lethargy, arthritis and evidence of cardiac, renal or neurological disturbance. In the United States of America (USA), arthritis is a common finding whereas neurological disturbance is the most frequent clinical feature in Europe and Japan. The clinical signs in horses are similar to those in dogs and include lameness, uveitis, nephritis, hepatitis and encephalitis. However, some authors observe that definitive evidence of clinical Lyme disease in horses is lacking [38]. Lameness in cattle and sheep associated with *B. burgdorferi sensu lato* infection has been reported.

Laboratory confirmation of Lyme disease may prove difficult because the spirochetes may be present in low numbers in specimens from clinically affected animals. In addition, the organism is fastidious in its cultural requirements. A history of exposure to tick infestation in an endemic area in association with characteristic clinical signs may suggest Lyme disease. Increasing antibody titres to *B. burgdorferi sensu lato* along with typical clinical signs are indicative of disease. Because subclinical infections are common in endemic areas, high titres alone are not confirmatory. The ELISA is extensively used for antibody detection; western immunoblotting is sometimes used for confirmation of ELISA results. It has been shown that ELISA techniques based on this antigen may be able to differentiate naturally infected and vaccinated animals [39]. Immunofluorescence assays may also be used but the results of these methods may be difficult to interpret. Culture of borreliae from clinically affected animals is confirmatory. Cultures in Barbour-Stoenner-Kelly medium should be incubated for 6 weeks under microaerophilic conditions and should be carried out in specialized laboratories. Low numbers of borreliae can be detected in samples by PCR techniques.

Acute Lyme disease responds to treatment with amoxicillin and oxytetracycline. In chronic disease, prolonged or repeated courses of treatment may be required. Acaricidal sprays, baths or dips should be used to control tick infestation. Where feasible, tick habitats such as rough brush and scrub should be cleared. Prompt removal of ticks from companion animals may prevent infection. However, because some tick species can transmit spirochetes shortly after attachment, it cannot be assumed that daily removal of ticks will prevent infection [40].

A number of vaccines, including whole cell bacterins and recombinant subunit vaccines, are commercially available for use in some countries. An outer surface protein A (OspA) recombinant vaccine stimulates the production of antibodies, which are able to kill the borreliae in the gut of the tick and thus prevent infection of the host. However, the benefit of vaccinating animals with currently available vaccines is disputed [41].

2.4. Ehrlichiosis and anaplasmosis

Ehrlichia (Cowdria) ruminantium is a Gram negative, intracellular rickettsial organism in the genus *Ehrlichia*. It occurs in colonies or morulae with a predilection for the vascular endothelium and stains blue with Giemsa stain. The organism is coccoid, 0.2–0.5 μ in diameter. It can now be cultivated *in vitro*, and it can also grow in mice. Cyclical development is believed to take place in intestinal and salivary epithelia of ticks. Although strain differences exist, all isolates possess a major antigenic protein 1 (MAP 1) that is used for diagnosis. However, the antigen cross-reacts with other *Ehrlichia* spp., including *Ehrlichia equi*, the cause of equine granulocytic ehrlichiosis. *Anaplasma* spp. is obligate intraerythrocytic parasites belonging to the order Rickettsiales and infecting ruminants. Infection occurs more sporadically in temperate climate areas. In the USA and other countries, the disease has occurred beyond the boundaries of tick-infested areas and the area distribution in Europe has been advancing northward in recent years with sporadic cases in France, Switzerland, the Netherlands, Hungary and Austria. Anaplasmosis of sheep and goats has a distribution similar to that of cattle. Disease occurs sporadically in the northern states and Canada. In Australia, infection is closely related to the distribution of *Boophilus microplus*, which is restricted to the northern areas. Differences in enzootic and epizootic areas in South America and South Africa are also largely related to tick distribution and climate [42].

Heartwater is limited in its occurrence to sub-Saharan Africa, Madagascar and three Caribbean islands of Guadeloupe, Marie Galante and Antigua. It is one of the main causes of death in imported breeds of cattle, sheep and goats in endemic areas. Heartwater has been diagnosed recently in the island of Mayotte in the Indian Ocean. Measures of disease occurrence in endemic areas, morbidity and mortality rates are low, but the percentage of sera positive titres for heartwater could be as high as 100% in adults, depending on the abundance of tick vectors [43]. Case mortality can be as high as 100% in peracute cases in sheep and goats and as low as 0–10% in cattle. The disease is less severe in indigenous breeds and related game animals reared in enzootic areas, some of which may become symptomless carriers. The N'Dama breed in West Africa is said to be well adapted to heartwater, partly because it can resist tick burdens under the traditional farming system. The method of transmission in the Caribbean, cattle egrets are suspected to spread *Amblyomma variegatum* between islands. Consequently, heart-

water is considered threats to the American mainland where potential vectors are present but do not harbour the disease or where the vector may be introduced and become established. Infection in ticks is transmitted transstadially and possibly transovarially. Vertical transmission to calves in colostral milk has also been reported. Several wild ruminants can be infected and become subclinical carriers and reservoirs. Ticks feeding on them can transmit the disease to domestic ruminants. The organism does not infect humans. Cattle are infected with *Amblyomma marginale* and *Amblyomma centrale* and sheep with *Amblyomma avis*. *A. marginale* will establish in sheep by experimental infection but *A. avis* will not infect cattle. A variety of species of wild ruminants in both North America and Africa can be infected and may have significance as reservoirs for *A. marginale*. In the United States, the black-tail deer in the West Coast region is believed a reservoir and a number of species of antelope play a similar role in South Africa. The prevalence of infection in cattle in endemic areas is very high with seropositivity rates exceeding 60% and often approaching 90%. Seropositivity is much lower in regions that interface between endemic and non-endemic regions. Source and methods of transmission recovery from acute infection result in persistent infection characterized by repetitive cycles of rickettsemia. Persistent carriers are the reservoir for herd infection. The level of parasitemia is often too low for detection by microscopy but can be detected by nucleic acid probe analysis. Transmission occurs biologically by ticks [44].

Heartwater is the most important rickettsial infection of ruminants in Africa and it is regarded as the most important disease of ruminants. In general, heartwater is a more serious problem where *Amblyomma habraeum* is the vector. In countries or regions where there is endemic stability, losses from heartwater are minimal until new animals are introduced. On the other hand, since most losses are in exotic animals, heartwater is a major constraint to livestock improvement in sub-Saharan Africa. Furthermore, it has the potential to spread from the Caribbean to the American mainland. Heartwater requires the vector tick to get established in any community. Therefore, there is concern about possible illegal importation of infected animals or ticks to southern United States where potential vectors exist. In ewes intra-uterine infection appears to occur with ease in experimental cases provided the ewe is exposed during the latter two-thirds of pregnancy. In sheep and goats, infection is usually subclinical but in some cases, particularly in goats, a severe anaemia may occur and a clinical picture similar to that found in cattle may be seen. Severe reactions of this type in goats are most frequent when the animals are suffering from concurrent disease. Goats may show hyper excitability and may bite at inanimate objects. The experimental disease in lambs includes fever, constipation or diarrhoea, pale, icteric conjunctivae and severe anaemia 15–20 days after inoculation. The anaemia is not completely resolved in 3–4 months. *A. avis* are usually situated at the periphery of erythrocytes but as many as 40% of infested cells may show sub-marginal protozoa [45].

The incubation period is 1–3 weeks after transmission in tick saliva. Depending on the susceptibility of individual animals and the virulence of the infecting organism, the resulting disease may be peracute, acute, subacute or mild and unapparent. Peracute cases show only high fever and death with terminal convulsions in 1–2 days. Acute cases are more common and have a course of about 6 days. A sudden febrile reaction is followed by inappetence and rapid breathing followed by the classical nervous syndrome that is characteristic of heartwater.

It comprises ataxia, chewing movements. Profuse, fetid diarrhoea is frequent. Subacute cases are less severe but may terminate in death in 2 weeks or the animal may gradually recover. The mild form is often subclinical and is seen mainly in indigenous animals and wild ruminants with high natural or induced resistance. The case mortality rate in peracute cases is 100%, in acute cases 50–90% and in calves below 4 weeks of age it is 5–10%, most animals recover in mild cases [46].

Haematological changes in heartwater are not specific but there may be thrombocytopenia, neutropenia, eosinopenia and lymphocytosis. Confirmatory diagnosis is based on identifying the *Rickettsia* in capillary endothelial cells using a Giemsa stained squash preparation of brain tissue at post-mortem. The rickettsiae occur as blue to reddish-purple colonies or morulae of five to several hundred coccoid organisms (0.2–0.5 μ in diameter) in the cytoplasm of the cells. An immunohistochemical staining technique has also been described [47]. Injection of blood into sheep may also be used as a diagnostic procedure. The available serological test is an indirect fluorescent antibody test used for surveys but the close antigenic relationship with other *Ehrlichia* spp. often leads to false positives. An ELISA based on recombinant MAP 1 protein of *C. ruminantium* was reported to be more sensitive. In general, clinical detection of heartwater is not always easy because all serological assays so far available have poor sensitivity or specificity. Diff-Quik staining of blood smears is as accurate as Giemsa in the detection of *A. marginale* and can be completed in 15 seconds as compared to nearly an hour for Giemsa. There are no diagnostic clinical chemistry findings. A rapid card agglutination test, which tests serum or plasma for antibodies against *A. marginale*, is cheap and quick, and sufficiently accurate to be used as a herd test. Currently, in most countries, the card agglutination and complement fixation (CF) tests are routinely available. It is also an accurate test for selecting recently affected animals. A dot-ELISA with high sensitivity, specificity and predictive value is also described and could be particularly applicable to field examinations. A competitive inhibition ELISA test, with high sensitivity and specificity, has been developed that detects antibody to a major surface protein that is conserved among *Anaplasma* species; this test can be used to detect cattle persistently infected for as long as 6 years. Vaccinated animals may react to all of the serological tests for periods of over 1 year. Nucleic probe analysis can be used to detect low levels of parasitaemia. Transmission to splenectomised animals has been used to detect carriers but is expensive and is now replaced by PCR in countries where this technology is available [48]. A polymerase chain reaction assay has therefore been suggested as the method of choice for detection of *E. ruminantium* infection [49].

Field cases of heartwater are difficult to treat successfully because available drugs are effective only in early febrile stages before neurological signs develop. In the early stages, short-acting tetracyclines at 10–20 mg/kg body weight (BW) and long-acting forms at reduced doses are effective. Sulphonamides can also be used in the early stages but are less effective. Hyperimmune serum is said to be of no curative value. Supportive therapy to reduce either the pulmonary oedema or the neurologic signs or to stabilize membranes in general is being investigated but with little success. Chemoprophylaxis involves administration of tetracyclines or subcutaneous implantation of doxycycline in susceptible animals when they are introduced into an endemic area. Results are not always predictable. Anaplasmosis treatment is with

tetracyclines. Treatment of clinical disease can be with oxytetracycline, 6–10 mg/kg BW daily for 3 days, or a one dose application of long-lasting 20 mg/kg oxytetracycline intramuscularly. The convalescent period is long. Concurrent administration of estradiol cypionate (14.3 mg/kg BW intramuscularly) appears to improve the rate of recovery by promoting parasitemia during treatment. Tetracycline treatment will not eliminate infection and immunity will persist. Blood transfusions are indicated in animals with a packed cell volume (PCV) less than 15%. Rough handling must be avoided. Imidocarb (3 mg/kg BW) is also an effective treatment for clinical cases and does not interfere with the development of acquired immunity to *A. marginale*. The risk for infection in the rest of the herd should be assessed and, if necessary, temporary or prolonged protection should be provided. Protection can be provided by tetracyclines, or by vaccination [50].

Past efforts to control heartwater were based on intensive acaricide treatment in endemic areas. It involved frequent use of acaricides (plunge dipping) up to 52 times a year. This has now been shown to be environmentally unfriendly, economically unsustainable, and would invariably lead to animals that remained always susceptible. For example, it was observed in Zimbabwe that large farms applying acaricides very frequently (more than 30 times per annum) had higher morbidity and mortality than farms applying acaricides less frequently. Vaccination is based on infection and treatment regimen that was first developed more than 50 years ago. It involves an intravenous injection of virulent organisms in cryopreserved sheep blood, followed by treatment with tetracyclines at the first indication of fever. Most control programmes in enzootic areas are based on increasing the resistance of the population by immunization. In any vaccination programme, particular attention should be paid to the animals at high risk, particularly animals brought in from non-enzootic areas, those in surrounding similar areas to which infection may be spread by expansion of the vector population under the influence of suitable climatic conditions, and animals within the area are likely to be exposed to climatic or nutritional stress [51].

Vaccination may lead to some deaths, the immunity may wane in the absence of reinfection, and animals may become carriers. More recently, cattle were successfully immunized for up to 10 months with a killed vaccine from a lysate of *E. ruminantium* formulated in Freund's adjuvant. In another study, the use of inactivated vaccines from cell-cultured *E. ruminantium* combined with an adjuvant led to a reduction in mortality from heartwater in cattle, sheep and goats exposed to field challenges in Botswana, Zambia, Zimbabwe, and South Africa. Experimental studies using deoxyribonucleic acid (DNA) recombinant vaccines so far have met with only limited success. Killed *A. marginale* are usually in an adjuvant vehicle. The vaccine requires two doses, 4 weeks apart, the last dose given at least 2 weeks before the vector season. However, there is a risk for neonatal isoerythrolysis. This can be reduced by vaccinating only empty cows and avoiding unnecessary booster injections. When this vaccine is used in the face of an outbreak, tetracyclines can also be given to provide temporary protection during the period of development of immunity; tetracyclines do not interfere with the development of this immunity. Preliminary reports of the efficacy of DNA vaccines are not encouraging. A living *A. centrale* vaccine is used extensively in Australia, Africa, Israel and Latin America, but not in the USA and there is some reluctance to introduce it into areas where *A. centrale* does not

already occur. A single vaccination is used in endemic areas and the immunity is reinforced by continuous challenge and considered to persist for life in tick areas. Vaccine administration is limited to the relatively resistant age group below 1 year of age, to the winter months when vectors are sufficiently rare to avoid the chance of spread to other age groups, and to circumstances where animals that react severely can be restrained and treated adequately. The method has the serious disadvantage of creating a large population of carrier animals which may subsequently spread the disease. Attenuated vaccines have been attempted by irradiation of strains and passage of the organism through sheep or deer and the use of naturally low virulence isolates [52, 53].

For tick control, flumethrin 1% pour on at 45 days interval was found to provide effective protection of Friesian/Zebu crossbred cattle against important ticks, but it must be applied correctly at the recommended dose. Pure Zebu and N'Dama cattle would probably require less frequent applications, Flumethrin pour-on is gradually replacing plunge dipping for the control of ticks and tick-borne diseases in general. Other than routine surveillance, there are no special biosecurity concerns with heartwater, since transmission requires the presence of the vector [54].

2.5. Tularemia

The disease causes acute septicaemia, with localization and granulomatous lesions and the organs (particularly the liver and spleen). Signs are very non-specific, as expected with bacteraemia, and include fever, anorexia, lethargy, and in some cases cough, rapid respiration or diarrhoea. Stiffness and oedema of the limbs may be seen. The incubation period of the disease is usually 2–14 days in companion animals [55].

Tularemia is a highly contagious disease occurring principally in wild animals but it may transmit to farm animals, causing septicaemia and high mortality. *Francisella tularensis* is the causative organism [56].

Tularemia is primarily restricted in its occurrence to countries in the northern hemisphere and occurs in most of them. In North America, the disease is most prevalent in farm animals in the north-western states of the USA and the adjoining areas of Canada, although in these areas it is rare and the majority of reports in livestock are historical. *F. tularensis* has a wide host range and is recorded in over 100 species of bird and wild and domestic animal. Disease is recorded among farm animals, most commonly in sheep and pigs and to a lesser extent in calves, which appear more resistant but can be infected in association with heavy tick infestation [57]. Sheep and pigs of all ages are susceptible but most losses occur in lambs, and in pigs clinical illness occurs only in piglets. There is a sharp seasonal incidence, the bulk of cases occurring during the spring months. The morbidity rate in affected flocks of sheep is usually about 20% but may be as high as 40%, and the mortality rate may reach 50%, especially in young animals. With sheep, transmission occurs chiefly by the bites of the wood tick, *Dermacentor andersoni*, and from *Haemaphysalis otophila*, the ticks becoming infected in the early part of their life cycle when they feed on rodents. In Europe *Ixodes ricinus* and *Dermacentor reticulatus* are vectors [58]. Transstadial and transovarial transmission occurs in the tick. The adult ticks infest sheep, and pastures bearing low shrubs and brush are particularly favourable to infestation. The ticks are

found in greatest numbers on the sheep around the base of the ears, the top of the neck, the throat, axillae and udder. It is assumed that sheep are relatively resistant to tularemia but become clinically affected when the infection is massive and continuous. Transmission to pigs and horses is thought to occur chiefly by tick bites but mechanical transmission to laboratory animals does occur with tabanid and blackflies. Tularemia is an acute septicaemia but localization occurs, mainly in the parenchymatous organs, with the production of granulomatous lesions [59].

In the sheep, the incubation period has not been determined. A heavy tick infestation is usually evident. The onset of the disease is slow with a gradually increasing stiffness of gait, dorsiflexion of the head and a hunching of the hindquarters; affected animals lag behind the group. The pulse and respiratory rates are increased, the temperature is elevated up to 42°C (107°F), and a cough may develop. There is diarrhoea, the faeces being dark and fetid, and urination occurs frequently with the passage of small amounts of urine. Body weight is lost rapidly, and progressive weakness and recumbency develop after several days, but there is no evidence of paralysis, the animal continuing to struggle while down [60]. Death occurs usually within a few days but a fatal course may be as long as 2 weeks. Animals that recover commonly shed part or the entire fleece but are solidly immune for long periods. In pigs, the disease is latent in adult pigs but young piglets show fever up to 42°C, accompanied by depression, profuse sweating and dyspnoea. The incubation period of the disease is about 7–10 days. In horses, fever (up to 42°C) and stiffness and oedema of the limbs occur. Foals are more seriously affected and may show dyspnoea and incoordination in addition to the above signs [61]. Necropsy usually reveals ticks on the carcass. Often, reddened or necrotic areas appear in and under the skin at the site of the infected bites. Regional lymph nodes may be swollen and congested. Congestion and oedema of the lungs are common [62].

An agglutination test is available for the diagnosis of tularemia, a titre of 1:50 being regarded as a positive test in pigs. Serum from pigs affected with brucellosis does not agglutinate tularemia antigen, but serum from pigs affected with tularemia agglutinates brucellosis antigen. Cross-agglutination between *F. tularensis* and *Brucella abortus* is less common in sheep and an accurate diagnosis can be made on serological grounds because of the much greater agglutination that occurs with the homologous organism. Titres of agglutinins in affected sheep range from 1:640 to 1:5000 and may persist at levels of 1:320 for up to 7 months. A titre of 1:200 is considered as positive in sheep. In horses the titres revert to normal levels in 14–21 days. An intradermal sensitivity test using 'tularin' has been suggested as being more reliable as a diagnostic aid in pigs than the agglutination test, but is unreliable in sheep. In sheep, large numbers of ticks may be present on the hides of fresh carcasses. In animals that have been dead for some time, dark red subcutaneous areas of congestion up to 3 cm in diameter are found and may be accompanied by local swelling or necrosis of tissues [63]. These lesions mark the attachment sites of ticks. Enlargement and congestion of the lymph nodes draining the sites of heaviest tick infestation are often noted. Pulmonary oedema, congestion or consolidations are inconstant findings. In pigs, the characteristic lesions are pleuritis, pneumonia and abscessation of submaxillary and parotid lymph nodes. The organisms can be isolated from the lymph nodes and spleen, and from infected ticks. Isolation

can also be effected by experimental transmission to guinea pigs. Techniques such as immunoperoxidase staining of fixed specimens and PCR of fresh tissues can circumvent the need for culture of this zoonotic agent. Samples for confirmation of diagnosis are based on

- Bacteriology: lung, lymph node, spleen (CULT—requires cysteine-enriched media, PCR).
- Histology: above tissues plus liver, fixed in formalin [64].

Treatment early in the course of infection is effective. Aminoglycosides, tetracyclines or cephalosporins all are probably beneficial initially, until results of antimicrobial susceptibility testing are available. Streptomycin, gentamicin, the tetracyclines and chloramphenicol are effective treatments in humans and companion animals. Oxytetracycline (6–10 mg/kg BW) has been highly effective in the treatment of lambs and much more effective than penicillin and streptomycin. Insecticide removal of ticks from affected animals and herdmates is important. An outbreak of tularemia in sheep can be rapidly halted by spraying or dipping with insecticide to kill the vector ticks. In areas where ticks are enzootic, sheep should be kept away from shrubby, infested pasture or sprayed regularly during the months when the tick population is greatest. An experimental live attenuated vaccine has been developed, but there is no routine vaccination of livestock [65].

3. Conclusion

Given that the livestock industry has become an integral part of world economy and a large number of dairy cattle are being imported between countries, in order to meet an increasing demand of meat and dairy products, it is essential to review current status of bovine BTBDs and to identify diagnosis and prevention in the knowledge of BTBDs and their control. Although there has been a recent increase in the number of studies of BTBDs in various regions and facilities, information on their prevalence, distribution, tick vectors and control is limited. This chapter provides a brief background on key bovine BTBDs and ticks and reviews the general aspects of bovine BTBDs to identify gaps in knowledge and understanding of these diseases, propose areas for future research and draw attention to the need for improved tools for the diagnosis and control of BTBDs.

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