BIOLOGIJA
2024. Vol. 70. No. 2–3. P. 130–146
ISSN 1392-0146 eISSN 2029-0578
DOI: https://doi.org/10.6001/biologija.2024.70.2–3.5

Brucella canis and *Chlamydia* spp.: insights into canine infections, diagnostics, and potential tick-borne transmission

Viktorija Petrauskaitė*,

Gintaras Zamokas,

Inga Stadalienė,

Birutė Karvelienė

Faculty of Veterinary Medicine, Lithuanian University of Health Sciences, Tilžės St. 18, Kaunas 47181, Lithuania

Brucella and *Chlamydia* spp. are zoonotic pathogens with significant implications for both human and animal health. *Brucella* species, which is responsible for brucellosis, manifest in a range of clinical symptoms and present substantial zoonotic risks. Though less commonly reported, canine infections with *Brucella* raise concerns about the potential for transmission to humans and highlight the need for continued vigilance in veterinary settings. *Chlamydia* spp. is notable for its ability to cause diverse diseases in animals, from mild infections to severe systemic illnesses. In dogs, *Chlamydia* infections can lead to symptoms such as conjunctivitis, respiratory disorders, and reproductive issues, including infertility and abortion. The zoonotic potential of *Chlamydia*, particularly *Chlamydia felis*, emphasises the need for thorough monitoring and control measures in both domestic animals and humans. Ticks play a significant role in the transmission of these pathogens. Research has identified *Chlamydia* spp. in ticks; however, the exact epidemiological implications remain unclear. Similarly, *Brucella* has been detected in ticks, but conclusive evidence of tick-borne transmission to humans or between animals is still lacking. Improved diagnostic tools and control strategies are essential for managing the risks associated with *Brucella* and *Chlamydia* infections in both animals and humans, with a particular focus on the role of ticks as potential vectors.

Keywords: *Brucella canis*, canine brucellosis, *Chlamydia*, zoonosis, tick, transmission

INTRODUCTION

Major zoonotic infections that can affect domestic animals, wildlife, and even humans include chlamydia and brucellosis (Sprague et al., 2009; Jamil et al., 2022). Various *Brucella* species cause brucellosis, an infectious disease also referred to by a few other names, including remitting fever, undulant fever, Mediterranean fever, Malta or Maltese fever, Gibraltar fever, Crimean fever, goat fever, and Bang disease (Kurmanov et al., 2022; Nowar et al., 2024). Typically, host restriction has been used to identify the species of Brucella. As of now, 13 species have been identified, including

* Corresponding author. Email: viktorija.petrauskaite@lsmu.lt

the recently described *B. pseudogrignonensis*, *B. abortus*, *B. melitensis*, *B. suis*, *B. ovis*, *B. canis*, *B. neotonae*, *B. pinnipedialis*, *B. ceti*, *B. inopinata*, *B. microti*, *B. papionis*, and *B. vulpis* (Jamil et al., 2022; Ma et al., 2024; Santos et al., 2021). Some species of *Brucella* have a high virulence factor, making them capable of infecting humans. *B. melitensis* is the most pathogenic species for humans; exposure to 1–10 CFU (colony forming units) is sufficient to cause infection, while *B. suis* and *B. abortus* have intermediate zoonotic potential. Of the traditional *Brucella* species, *B. canis* has the least potential for zoonotic infection (Ma et al., 2024; Santos et al., 2021).

Brucellosis is a common disease that affects animals in more than 170 countries on all six continents. Brucellosis has been successfully eradicated in Europe, Australia, and Canada, but there is still concern in highly endemic areas of Africa, parts of Asia, and Latin America (Ma et al., 2024). However, livestock brucellosis has been eliminated in many European Union (EU) member states from farm animals, although positive tests are still observed (Jamil et al., 2022). Conversely, there was a noticeable rise in the cases of canine brucellosis resulting from *B. canis* infection, particularly in Italy and the United Kingdom (Jamil et al., 2022). Currently, the majority of *B. canis* infection reports in the EU are based on the occurrence of clinical symptoms in either humans or dogs. No cross-sectional or systematic study has been carried out in any EU country to assess this disease. The current state of data and surveillance programmes makes it impossible to pinpoint the precise number of the countries in which the disease may be considered endemic (Djokic et al., 2023). In several countries, including Switzerland, Ukraine, the Netherlands, the United Kingdom, and Turkey, an increasing number of isolated cases, clusters, or outbreaks have been documented since 2017. From 2017 to 2023, the majority of *B. canis* positive tests are reported in these countries: Spain, Portugal, France, Hungary, and Poland. This data should be interpreted cautiously because formal prevalence estimations cannot be made using the results due to the non-systematic sample submission and highly uneven representation of various countries. None of the EU nations have a mandatory programme for testing for *B. canis*, whether through non-specific bacterial culture investigations or specific *B. canis* serological testing (Santos et al., 2021; Djokic et al., 2023; van Dijk et al., 2021; De Massis et al., 2022; Buhmann et al., 2019). As we can see, the prevalence of brucellosis in the world remains high in both productive animals and other animals. In Europe, productive animals are monitored and vaccinated against brucellosis, but as we can see, each year there is an increase in *B. canis* infections among domestic animals like dogs, whose control and spread within the European Union are neither monitored nor recorded. The spread of these diseases poses a risk to human health.

Dogs infected with *B. canis* may show some symptoms of reproductive failure or be subclinically ill. Chronic inflammation of the testicles and epididymis in male dogs can lead to unilateral or bilateral testicular atrophy, infertility (due to abnormal sperm) as well as epididymitis, prostatitis, and orchitis (Rovid, 2018; Hensel et al., 2018). In females, the condition typically results in stillbirths and abortions. Although normal pregnancies can occur in infested bitches, there is a very high risk of perinatal mortality in pups that may be born (Rovid, 2018). Discospondylitis is another well-known manifestation of *B. canis* infection; symptoms include back pain, lameness, muscular weakness, and, less frequently, neurological deficits. Anterior uveitis, panuveitis, endophthalmitis, retinal detachment, and intraocular haemorrhage are less frequent symptoms (Woods, 2024).

Chlamydia is one of the other bacteria that can spread zoonotic diseases (Borel et al., 2018). Throughout the world, *Chlamydia abortus*, *Chlamydia trachomatis*, and *Chlamydia psittaci* are well-known species that cause a variety of diseases in both humans and animals (Sprague et al., 2009; Borel et al., 2018). *Chlamydophila pneumoniae*, a species that was commonly isolated from humans, has recently been discovered in a variety of species, including

horses, koalas, and different amphibians and reptiles (Sprague et al., 2009). There is evidence to suggest that a few other animal pathogenic chlamydial species, such as *C. felis*, *C. caviae*, and *C. suis*, can occasionally infect humans and present with different clinical presentations (Borel et al., 2018).

There has been little research done globally on the prevalence of this bacterium in animals and wildlife. Few epidemiological studies on canine chlamydia infection have been carried out globally; the majority of these studies were published in regional journals (Tian et al., 2014). In studies conducted in China, seropositivity was determined in 17.6% out of 591 examined healthy dogs. However, in Lithuania, compared to the studies conducted in Europe, seropositivity was found in as many as 50% of healthy animals (Tian et al., 2014; Liutkeviciene et al., 2009). Seropositivity was measured in Slovakia at 5.5%, in Germany at 20%, and in Poland at 0.8%. It was not found in Sweden, but these studies usually involve small groups of subjects (Domrazek & Jurka, 2024; Holst et al., 2010). The prevalence of chlamydiosis is poorly studied both globally and within the European population, making it difficult to assess the spread of this disease, particularly among dogs.

Pneumonia, rhinitis, arthritis/polyarthritis, pericarditis, polyserositis, encephalomyelitis, and other urogenital tract-related diseases are among the syndromes caused by *chlamydia* infection of eukaryotic tissues. Infertility issues in female animals, including abortion, perinatal and embryonic death, also complications that result in conjunctivitis, enteritis, and head and ear malformations in newborns are documented (Pagliarani et al., 2020).

The transmission pathways of these two zoonoses are not fully understood. These zoonoses have been found in ticks over the last five years, but it hasn't been established that ticks can actually carry these illnesses. In 2024, there were 16 distinct species of ticks known to harbour brucellosis, with an approximate prevalence of 33.87% in ticks overall. *Brucella* has been detected in ticks at different stages of development: 40.9% in larvae, 4.6% in female ticks, and some positive eggs were discovered (Ma et al., 2024). Only one species of tick, *I. ricinus*, was found to carry chlamydia, with prevalences of 6.4% in some locations and 28.1% in others (Chisu et al., 2020; Croxatto et al., 2014; Pilloux et al., 2015). In the conducted studies, the pathogen was often identified, but not the specific species of the pathogen. Due to the limited number of studies and their specificity, the possibility of ticks as carriers of these zoonoses has not been confirmed (Pilloux et al., 2015; Ma et al., 2024).

The aim of this article is to analyse the methods and types of research samples used to best identify *Brucella* and *Chlamidia* infection diseases and to discuss the characteristics of the pathogen and its transmission routes. It aims to determine the methods that would help standardise the detection of these diseases among asymptomatic domestic dogs.

METHOD

In this review of the literature, we looked at the most recent laboratory diagnostic tests for *Brucella canis* and *Chlamydia* in dogs, the pathological materials that are most likely to show signs of this zoonosis and human infection risk. Thirty-four articles published between 2009 and 2024 were selected using the following keywords: brucella, brucella canis, chlamydia in canines, vector-borne diseases, diagnosis of brucellosis, diagnosis of chlamydia. These articles explain the ways by which humans can become infected, as well as the laboratory diagnostic tests that can be used to detect infections, their sensitivity, and specificity. One article, which explains the emergence of these bacteria, was published before 2009 was described in other articles in the literature. The selected articles did not contain clinical case reports.

BRUCELLOSIS

The genus *Brucella* is a member of the family Brucellaceae, order Rhizobiales, class Alphaproteobacteria, and phylum Proteobacteria. The serious febrile illness known as brucellosis

is caused by the *Brucella* genus (Głowacka et al., 2018). David Bruce discovered this genus in 1887 (Głowacka et al., 2018). The most frequent cause of canine brucellosis is now known to be *Brucella canis*, which was initially identified in the late 1960s (da Silva et al., 2020). *Brucella* is classified as a small coccobacillus, with a measurement of approximately 0.6 to 1.5 μm (Alton et al., 1996). *Brucella* are Gram-negative, small, aerobic rods, non-spore-forming, non-mobile organisms that only rarely form pairs or chains when present in single form. The intracellular pathogen *Brucella* multiplies within macrophages during an infection, adapting to the low oxygen content, low nutrient levels, and acidic pH. Each Gram-negative bacterial cell uses lipopolysaccharide (LPS) to build its structure; the genus *Brucella* produces two different types of LPS. Polysaccharide O-chain is absent from the rough phenotype, whereas the smooth form displays whole LPS in the outer membrane (Głowacka et al., 2018; Sánchez-Jiménez et al., 2020). Only two species – *B. ovis* and *B. canis* – are known to have a rough phenotype. Smooth phenotypes are found in other known species (Kurmanov et al., 2022). Interestingly, naturally occurring rough strains of *Brucella*, like *B. canis*, have a lower survivability within host cells in culture or *in vivo*, but they tend to invade host cells more efficiently than smooth strains because they lack the O-polysaccharide chains of LPS (Santos et al., 2021). The majority of these infectious agents have the ability to hydrolyse urea and produce catalase and cytochrome oxidase. Classical pathogenic factors like exotoxins, cytolysins, exoenzymes, exoproteins, capsules, plasmids, fimbria, and drug-resistant forms are not produced by *Brucella* (Głowacka et al., 2018). At one time, based on the genetic and immunological evidence that all members of this genus are closely related, *Brucella* was once reclassified as the single species *B. melitensis*. The different *Brucella* species were regarded as biovars in this system (Rovid, 2018). These species are separated into 15 biovars (bv; biotypes) in contemporary *Brucella* systematics: *B. abortus* bv. 1–6 and 9, *B. mellitensis* bv. 1–3, and *S. suis* bv. 1–5. Divided bv is absent from five species of *Brucella*: *B. canis*, *B. ceti*, *B. inopinata*, *B. pinnipedialis*, and *B. neotomae* (Kurmanov et al., 2022).

Although the exact pathogenesis of *B. canis* infections is unknown, it most likely follows the general patterns of *Brucella* infections seen in other animal species. Therefore, unless otherwise noted, the mechanisms discussed here are generic to the genus and not unique to *B. canis* (De Massis et al., 2022; Santos et al., 2021). *Brucella* strains can coexist and proliferate in cells that are both phagocytic and non-phagocytic. This bacterium primarily targets trophoblast cells, dendritic cells, and macrophages. *Brucella*, however, can also proliferate within other cell types, such as murine fibroblasts or epithelioid cells. Up to 72 h after infection, *Brucella* can persist within non-phagocytic cells. The pathogen uses cellular tropism to multiply and spread to other tissues in macrophages by evading the host immune response. Bacteria can migrate to the desired tissues in the reproductive tract after entering a host through the lymph nodes (Głowacka et al., 2018). As previously indicated, *B. canis* is not included in the majority of research on *Brucella* pathogenesis; however, several specificities have been reported. For instance, even in its preferred host, *B. canis* infection results in a poor pro-inflammatory response. However, in experimental settings, this species is much less likely to induce inflammation than the smooth pathogenic *Brucella* species, leading to much lower induction of IFNγ production and inflammatory lesions (Santos et al., 2021).

Canine brucellosis caused by *B. canis* is particularly prevalent in shelters, commercial breeding facilities, and environments where dogs reside in large groups (De Massis et al., 2022). Contact with infected secretions can spread canine brucellosis either orally or through venereal infection (Santos et al., 2021). Foetuses and foetal membranes, stillbirths and vaginal secretions from infected bitches can spread brucella; for males, semen can contain high concentrations of *B. canis*; small amounts of bacteria have also been found in urine, nasal and ocular secretions, saliva, and faeces; some

articles even suggest that transmission through broken skin could occur (Rovid, 2018). Males may continue to spread infection even after castration because the bacteria can live in the lymphatic tissues and the prostate. The minimum infectious dose in dogs through oral means is approximately 2×10^6 CFU; the minimum infectious dose through other routes is unknown but is thought to be somewhat lower. By the conjunctival route, the infectious dose ranges from 10^4 to 10^5 CFU. Additionally, starting two to three weeks after infection, it is possible to detect the presence of even more bacteria in the blood than 10^3/ml (De Massis et al., 2022). There are no known specific lesions associated with *B. canis* infection; however, both infected adults and surviving puppies can exhibit splenomegaly and lymph node hypertrophy. There are also inflammatory lesions, both acute and chronic, in the genital system. Plasma cells and macrophages carrying phagocytised bacteria have been observed infiltrating lymph nodes and the splenic sinuses in the cases of chronic bacteraemia. A pervasive submucosal lymphocytic infiltration, primarily affecting the prostate, epididymis, the renal pelvis, and the uterus, is seen in all organs of the urogenital system. Additional kidney lesions could be present, accompanied by weak cell infiltration and thickening of the glomerulus basal membrane. Meningoencephalitis, myocarditis, and focal hepatic necrosis are other changes that have been reported. Granulomatous iridocyclitis and exudative retinitis are conditions that affect the eyes and are characterised by a broad infiltration of neutrophils, plasma cells, and lymphocytes. Even so, *B. canis* has been found in a variety of tissues from neonates who were naturally infected, including the kidney, intestines, stomach, lungs, the central nervous system, the umbilicus, liver, lymph nodes, and the spleen (Santos et al., 2021; De Massis et al., 2022). Besides, the *B. canis* bacterium has a different structure compared to other *Brucella* species (rough phenotype), which can complicate its detection in dogs. Research indicates that this bacterium can localise in various organs or secretions, but the quantity of the bacteria found can vary, making it challenging to detect in animals. The reproductive system, the urinary system, and lymph nodes are the most commonly identified organs where *B. canis* bacteria are detected.

Most often, *B. abortus*, *B. suis*, and *B. melitensis* cause brucellosis, which is a zoonotic disease. Although a study on 306 asymptomatic adults at risk of occupational exposure showed a seroprevalence of 3.6% for *B. canis*, it is possible that *B. canis* is underdiagnosed in human patients (Li et al., 2023; Santos et al., 2021). Both human-to-human and animal-to-human transmission are possible for brucellosis. Direct contact with the fluids or tissues of infected dogs, particularly through genital secretions, materials from aborted and parturiated pregnancies, urine, or blood, can result in the infection of humans with *B. canis*. Rare human-to-human transmission cases include aerosol use, blood transfusions, breastfeeding, sexual transmission, and transplacental transmission (Li et al., 2023; Djokic et al., 2023). Clinical symptoms include chills, fever, sweats, arthritis, appetite loss, weight loss, exhaustion, headaches, muscle soreness, and joint pain, but they are not particularly specific. It can develop into endocarditis, aneurysm, peritonitis, osteomyelitis, and spondylitis in severe cases. It can also affect the reproductive organs in men, causing orchitis and epididymitis; in women, it can cause endometrial, ovarian, and tubal infections, which can result in miscarriage and infertility (Li et al., 2023; Santos et al., 2021). Because of the limitations of clinical or laboratory diagnosis, it is underdiagnosed and its significance for public health is largely overlooked, even though it may be a risk factor for humans.

Diagnosing canine brucellosis can be challenging at times, but using multiple techniques increases the likelihood of a successful diagnosis. If brucellae are found through microscopic analysis of strained smears from the placenta, reproductive discharges, or the contents of the foetal stomach using modified Ziehl-Neelsen straining, this disease may be suspected. Although *Brucella* species stain red, they are not actually acid-fat; instead, they are resistant to

being decoloured by weak acids. Certain organisms can mimic *Brucella*, including *Chlamydia* spp. and *Coxiella burnetii* (Rovid, 2018). The bacterium isolation serves as the sole indicator of a *B. canis* infection. The type of sample, the infection stage, the manner of sample handling, and the bacteriological techniques applied all affect the sensitivity of this method to diagnosis. It is possible to isolate *B. canis* from fresh samples (vaginal discharges, placental and foetal tissues, urine, semen, milk), necropsy samples (lymph modes, spleen, prostate, epididymis, uterus, bone marrow, eye, and intervertebral discs), and blood. When there are clinical symptoms, a fresh sample is more sensitive. However, there are situations when this sample is unavailable, in which case bacteriology on a blood sample is the only workable solution. To prevent clotting and demonstrate the inhibition of bacterial growth, blood samples should be collected into sterile lithium heparin or sodium citrate. Within 24 h, the sample must be submitted to the laboratory at refrigeration temperature (not frozen). This sensitivity of the method can be increased by using liquid culture with sporadic sub-culturing onto solid media, Ferrell's medium, or a modified Thayer-Martin medium. Automated culture methods, like the VITEK-2, can also detect *Brucella* species, but further methods should be used to confirm the results. Regretfully, a negative result does not completely rule out the infection because *B. canis* frequently experiences recurrent bacteraemia, which is partly due to the temporary absence of the bacteria from the cultured sample. Since it can take up to nine days for the culture to grow, laboratory staff are more likely to be exposed. Therefore, other methods are needed to confirm the pathogen (Djokic et al., 2023; De Massis et al., 2022; Hensel et al., 2018). Using the polymerase chain reaction (PCR), one can also obtain a direct diagnosis by identifying the genomic DNA of *B. canis* in a biological sample. In addition to being faster than culture, this method is unaffected by sample contamination or bacterial viability. For PCR, whole blood is the preferred sample; serum can also be used, but the sensitivity is reduced (Santos et al., 2021). Conventional PCR assays can be created to identify multiple targets in a single test (multiplex PCR) or just one target (uniplex PCR). The two most popular PCR types for identifying *Brucella* are duplex and uniplex. Targets for these assays are most frequently the sequences encoding 16S and 23SrRNAs, the BCSP31 protein, and IS711 transposase (Kurmanov et al., 2022). Differentiating *B. canis* using genetic techniques is especially challenging. A small number of papers on *B. canis*-specific PCRs have been published, but the majority of PCR tests only identify *Brucella* to the genus level. Multiplex PCR assays can identify several Brucella species. There have been reports of other tests that can be used to identify species, including matrix-assisted laser desorption/ionisation time-of-flight mass spectrometry (MALDI-TOF MS) and single nucleotide polymorphism (SNP) typing (Rovid, 2018). Using PCR from a blood sample with a negative culture result, one publication found as many as 13.6% positive *B. canis* samples using the IS771 marge gene (da Silva et al., 2020). While some labs may use PCR tests on clinical samples directly, the primary application of these tests for *Brucella* is the identification of organisms in culture. *B. canis*-specific PCR tests for the evaluation of the canine population are still in their early stages (Rovid, 2018). More tests in the canine population must be conducted before using this method to determine its sensitivity and specificity (De Massis et al., 2022).

An innovative technique for quick and extremely specific DNA amplification in an isothermal environment was introduced in 2000 and is called loop-mediated isothermal amplification (LAMP). The use of specific primers (FIP and BIP) that contain sequences from sense and antisense strands of the target DNA is the key component of the method. Two sets (P-1 and P-2) of six LAMP primers were created in order to identify the *Brucella* bcsp31 gene sequence (Kurmanov et al., 2022). Using the LAMP primer set designed by Song et al., the amplified gene sequences obtained through conventional PCR matched those of *B. abortus*, but not those of any other bacteria in GenBank,

suggesting that the omp25 primers could be used to detect Brucella species. Furthermore, the 29 non-*Brucella* species could not be successfully identified using the LAMP primers and conditions, but four species from the *Brucella* genus – *B. abortus*, *B. ovis*, *B. suis*, and *B. melitensis* – were identified. More inclusive and exclusive research involving a wider range of *Brucella* species and strains, such as *B. canis*, as well as non-*Brucella* bacteria, is required for this methodology (Song et al., 2012).

Other methods are employed exclusively for research purposes: they are not regularly used for diagnosis or epidemiological investigations of outbreaks. Real-time PCR, also known as quantitative polymerase chain reaction, or qPCR, which does not require a post-amplification gel analysis, was a step forward from traditional PCR. By using a variety of DNA-intercalating or probe-attached fluorescent dyes, including SYBR Green, fluorescein, cyanine, and many others, the technique allows one to directly observe the accumulation of PCR products as they are amplified. It is a quick, perceptive, and targeted technique. The first report to show how useful this technology is for characterizing the *Brucella* genus is that by Winchell et al. They revealed the creation of seven distinct real-time PCR assays intended to identify different *Brucella* markers specific to genus and species. Five target markers were identified: Bcan, Bmar, Bmel, Bneo, and Boa contained SNP loci that required a more complex allelic discrimination technique. Two of the target markers, Bspp (specific for all *Brucella* spp.) and Bsui (specific for *B. suis*), represented unique genomic sequences, detectable using standard real-time PCR. Even though the six-marker speciation panel worked well for accurately determining the species, we still decided to include the Bspp marker. All members of the *Brucella* genus are detected by this marker, which also acts as an initial identifying indicator of inclusion. This method could enhance the promptness with which results are reported, offer a way to better characterise isolates, support epidemiological studies, and help create a more thorough typing system for this genus (Kurmanov et al., 2022; Winchell et al., 2010). There are additional genetic tests, such as restriction fragment length polymorphism (RFLP), Multi Locus sequence analysis/typing (MLSA/MLST), ligase chain reaction (LCR), and multiple locus variable number tandem repeat (VNTR) analysis (MLVA), but not enough research has been conducted on *B. canis* and other *Brucella* species despite research being done on more virulent species (Kurmanov et al., 2022).

Serologic testing is the first diagnostic procedure and screening tool for suspected cases of brucellosis. Antibody response to cell wall antigens of *Brucella* spp. is assessed by serologic testing. According to their structure, brucella species exhibit two distinct morphologic appearances of their cell walls: smooth and rough. These distinctions are significant because serologic tests intended to identify infections caused by smooth *Brucella* spp. are unable to identify infections caused by *B. canis* (Hensel et al., 2018). The most popular tests in the field in countries where the disease is prevalent are the Rapid Slide Agglutination Test (RSAT), the 2-Mercaptoethanol-Rapid Slide Agglutination Test (2ME-RSAT), and the Tube Agglutination Test (TAT and 2ME-TAT). The Agar Gel Immuno-Diffusion test (AGID), a confirmatory test, uses cytoplasmic antigens ($AGID_{cpa}$) or cell-wall antigens ($AGID_{cwa}$). Additional testing options include the Complement Fixation Test (CFT), the Enzyme-Linked Immunosorbent Assay (ELISA), and the Indirect Fluorescent Antibody Assay (IFA). But so far, they have only been applied in research projects (De Massis et al., 2022). Heat-inactivated *B. ovis* or *B. canis* coloured with Rose Bengal is used in the original RSAT technique developed by George and Carmichael in 1978. The commercially available RSAT test is quick, simple to administer, and easy to read. Test sensitivity, or the likelihood that the test will not result in false negative reactions, is 99%. On the other hand, specificity, which is the likelihood that the test will not result in false positive reactions, is rather low, with false positive rates typically ranging from 20% to even 50%. False positive results are

reportedly caused by cross-reactions between the antigen used and particular antibodies that may be present in the tested serum. These bacteria include *Pseudomonas* spp., *Bordetella* spp., *Streptococcus* spp., and more broadly, some *Enterobacteriaceae*. More recent research shows that specificity can reach 83.34% while sensitivity drops to 70.58% (Santos et al., 2021; De Massis et al., 2022; Hensel et al., 2018). The RSAT test was then altered by adding 2-mercaptoethanol (2ME-RSAT) to the test serum prior to mixing with the antigen in order to lower the frequency of false positives. However, as a result, the specificity of the test increased to 100% while its sensitivity decreased to 31.76% (De Massis et al., 2022). Antibodies *B. canis* can be found by the TAT in dogs that test positive for RSAT or 2ME-RSAT. The test can produce false positive results because it is sensitive but not very specific. 2-mercaptoethanol (2ME-TAT) is added to RSAT in order to decrease false positive reactions. No publicly available data is available to estimate the values of DSn and DSp. The TAT is used as a confirmatory test for RSAT or 2ME-RSAT and is regarded as a semi-qualitative test (Santos et al., 2021; Djokic et al., 2023; De Massis et al., 2022). AGID has the ability to utilise two distinct forms of antigens: the *B. canis* cell-wall antigen ($AGID_{cwa}$) and antigenic proteins that are isolated from the cytoplasm of *B. canis* or other *Brucella* species ($AGID_{cpa}$), specifically *B. abortus* (De Massis et al., 2022). AGID is able to identify precipitins between five and ten weeks post-infection, using surface proteins as antigens. Dogs with chronic infections can have antibodies detected up to three years after infection, even in the absence of bacteraemia, using cytoplastic antigens produced by sonicating *B. canis* (Santos et al., 2021). $AGID_{cwa}$ sensitivity ranges from 60.5% to 87%, while $AGID_{cpa}$ sensitivity is 96%. Both have 100% specificity (Djokic et al., 2023). In kennels where *B. canis* is present, the most effective method is the $AGID_{cpa}$ test, which can be used as a confirmatory test for sera that produce a positive result on the agglutination test, as long as the final diagnosis of *B. canis* infection always needs determination by blood culture (De Massis et al., 2022). An alternative test to consider in cases where RSAT and TAT are not available is the Indirect Immunofluorescence Assay (IFA) test. It is challenging to assess their performance and make recommendations because they lack descriptive or validation data (Djokic et al., 2023; De Massis et al., 2022). The diagnosis of canine brucellosis has not been routinely performed using CFT, despite the test having high specificity (65–100%) and sensitivity (77–100%) and being regarded as confirmatory evidence in campaigns to eradicate bovine and ovine brucellosis in several countries. In the study of Mol et al., the CFT test yielded a positive result in 15.3% of dogs; however, there was no significant agreement with the other tests in the study. The lack of standardisation of CFT as a test technique presents the biggest challenge in the detection of canine brucellosis. In addition to these, this method has additional drawbacks including high cost, equipment requirements, and the need for trained personnel (Mol et al., 2019).

Using the cytoplasm of *B. abortus* or the cell wall of *B. canis* as antigens, immunoenzymatic tests (ELISA) have been developed. The advantage of the cytoplasmic antigens shared by all strains in the genus *Brucella* is that they do not exhibit cross-reactivity with bacteria from other genera. The phase M cell-wall antigens of *Brucella* strains are used in iELISA, which is very specific but not very sensitive (De Massis et al., 2022). Cortina et al. examined the performance of a *B. canis* diagnostic method using iELISA. The rate of true-positive (Se) and false-positive (1-Sp) results for each of the potential reactivity values of the assay was determined using ROC analysis. The findings indicate that the iELISA has a 98.6% sensitivity and a 99.5% specificity (Cortina et al., 2017). Yet, the iELISA study by Sànchez-Jiménez et al. found that the sensitivity was 75% and the specificity 64%. The iELISA results, blood culture, and PCR results were also compared in Sánchez-Jiménez et al. (2020; Table). All these results point to the difficulty caused by the lack of highly sensitive and specific diagnostic assays for *B. canis* (Sánchez-Jiménez et al., 2020; Winchell et al., 2010). Diagnosing canine brucellosis involves

Table. **Comparison of iELISA results, blood culture, and PCR: sensitivity (Se%), specificity (Sp%), and the kappa coefficient in dogs**

	iELISA vs PCR			iELISA vs 2ME-RSAT			iELISA vs Blood culture			PCR vs 2ME-RSAT	2ME-RSAT vs Blood culture	PCR vs Blood culture
	pd-Btuf	tuf	pdhB	pd-Btuf	tuf	pdhB	pd-Btuf	tuf	pdhB			
Se %	75	75	65	73	24	24	72	75	72	78	67	92
Sp %	64	64	61	60	92	92	59	59	59	86	93	86
Kappa/ Concordance	0.306/ fair	0.306/ fair	0.193/ poor	0.179/ poor	0.171/ poor	0.171/ poor	0.149/ poor	0.173/ poor	0.173/ poor	0.514/ moderate	0.569/ moderate	0.591/ moderate

several methods, each with its strengths and limitations. Microscopic analysis of stained smears from reproductive tissues or foetal stomach contents and culture techniques are traditional methods, but they can be hampered by the intermittent presence of bacteria and the complexity of differentiating it from other organisms. PCR offers a more rapid and sensitive alternative by detecting the genomic DNA of *B. canis*, although it often requires further confirmation due to its inability to distinguish between species effectively. Novel techniques like Loop-Mediated Isothermal Amplification (LAMP) and Real-Time PCR (qPCR) provide faster and more specific detection but require extensive validation. Serologic tests, including the Rapid Slide Agglutination Test (RSAT) and Agar Gel Immuno-Diffusion (AGID), are commonly used but may suffer from issues with specificity and sensitivity. These methods highlight the ongoing need for accurate, reliable diagnostic tools to manage and control canine brucellosis effectively.

Socorro Ruiz-Palma et al. conducted proteomic research on several *Brucella* species, including *B. suis*, *B. ovis*, *B. canis*, and *B. neotomae*. Classifying the cargo of outer membrane vesicles (OMVs) into clusters of orthologous protein recognised by anti-*Brucella* antibodies was made possible by the analysis of the pan-proteome of *Brucella* vesicles, which also aids in the development of brucellosis vaccines. This study provided fresh perspectives on the content of OMVs from previously unreported *Brucella* species. Iron-binding proteins in OMVs may play a role in the uptake of nutrients under adverse conditions, which is particularly beneficial for the intracellular lifestyle of *Brucella* species. The presence of protection-inducing proteins in the OMVs of these *Brucella* species, along with orthologous proteins that have been previously identified as immunogenic, make these nanostructures highly appealing for the development of an acellular vaccine that may induce immune cross-protection (Socorro Ruiz-Palma et al., 2021). Their findings highlight the significance of OMVs as a source of immunogenic proteins that could be leveraged to create an effective acellular vaccine with cross-protective potential against brucellosis.

CHLAMYDIOSIS

Worldwide important human and animal pathogens, *Chlamydiae* bacteria can cause both acute and chronic illnesses in their hosts and asymptomatic infections (Bommana & Polkinghorne, 2019). The term 'Chlamydozoa' was first used in 1909 to describe swabs taken from cows with transmissible vaginitis and from healthy cows on multiple farms that contained forms similar to the dreaded 'trachoma-bodies'. There were at least seven attempts to characterise and name what is now known as *Chlamydiae* prior to Pagès' 1966 proposal of a taxonomy for *Chlamydiaceae* (Borel et al., 2018). This bacterium was originally discovered in dogs more than 50 years ago (Liutkeviciene et al., 2009).

In recent years, there has been a constant shift in the nomenclature for chlamydial infections. Along the addition of new families, a proposal was made to divide the genus *Chlamydia* into two genera, *Chlamydia* and *Chlamydophila* because of 16 S and 23 S rRNA gene analysis. The order Chlamydiales is divided into nine families. The family *Chlamydiaceae* contains a single genus, *Chlamydia*, which includes 11 species: *C. abortus*, *C. avium*, *C. caviae*, *C. felis*, *C. gallinacean*, *C. muridarum*, *C. pecorum*, *C. pneumoniae*, *C. psittaci*, *C. suis*, and *C. trachomatis*, as well as three *Candidatus* (Ca) species: *Ca. C. ibidis*, *Ca. C. sanzinia*, and *Ca. C. corallus* (Borel et al., 2018).

Fascinating Gram-negative, obligatory intracellular bacteria that share a special biphasic developmental cycle with their eukaryotic hosts are found in the Chlamydiales order.

A chlamydial elementary body (EB) attaches itself to its host cell, becomes internalised, and forms a membrane-bound cytoplasmic inclusion, which is the start of the cycle. Infectious elementary body that infect host mucosal epithelial cells have a size of 0.2–0.3 μm. EBs develop inside a membrane-bound endocytic vacuole known as an inclusion after adhering to and entering the host cell. Once inside, they differentiate into large, 0.8 μm, metabolically active but noninfectious reticulate bodies (RBs). After maturing into infectious EBs, subsequent populations of RBs are released to infect nearby cells following host cell lysis. Chlamydial RBs may enter a non-replicative, non-infective state in response to suboptimal growth conditions, antibiotic treatment, or viral co-infection. Nevertheless, they will continue to be viable until optimal growth conditions are restored (Borel et al., 2018; Bommana & Polkinghorne, 2019).

Additionally, these investigations into new hosts have revealed both new chlamydial species such as those found in reptiles and birds, as well as extended host ranges for already-existing species. Altogether, the Chlamydiales order of bacteria has a much wider range of animal hosts than previously known, with over 400 host species reported worldwide, the majority of which are wild animals. Depending on the host species afflicted and the chlamydial species involved, chlamydiosis in animals can range from mild infections to serious illnesses that are potentially fatal (Borel et al., 2018). Because *C. abortus* and *C. psittaci* can cause abortion and psittacosis in animals, birds, and humans, respectively, they are particularly important. *C. felis* is a significant agent that may have zoonotic effects. It primarily affects the eyes and upper respiratory tract of cats, and the infection spreads through airborne particles and nasal and ocular secretions from infected cats. Dogs can have the same clinical symptoms as cats, including keratitis, encephalitis, pneumonia, and conjunctivitis (Wu et al., 2013). Bacteria are responsible for the pathogenesis of abortion and possibly the primary cause of infertility in female dogs carrying *C. psittaci* genotype C, which manifests as recurrent keratoconjunctivitis, respiratory disorders, and a decrease in the number of puppies (Domrazek & Jurka, 2024). A small study group did not show any signs of infection, despite studies in Sweden suggesting that these infections can cause ocular diseases and fertility issues in dogs (Holst et al., 2010). Bacteria in male dogs disrupt spermatogenesis, cause apoptosis, and fragment DNA in Leyding and Sertoli cells. After receiving a direct inoculation of *C. trachomatis* in the prostate glands, study participants experienced benign prostatic hyperplasia, which indicates a potential cause of infertility (Domrazek & Jurka, 2024). Depending on the disease of the species, Chlamydial infections can cause a variety of symptoms, such as endometritis/metritis, orchitis/epididymitis/urethritis, infertility, pneumonia, mastitis, rhinitis, arthritis/polyarthritis, pericarditis, polyserositis, encephalomyelitis, and placentitis leading to abortion, stillbirth, or weak neonates. This pathogen causes a wide range of non-specific symptoms in different organs. So far, it is not precisely known in which organs this pathogen can be detected, but based on the symptoms it causes, it can be inferred that it may be found in eye structures, lungs, reproductive organs, the urinary system, and others. The significance of these findings needs

to be clarified, but certain *Chlamidia* species were discovered in the peritoneum, kidney, liver, and spleen (Borel et al., 2018). The expanded understanding of Chlamydiales hosts reveals a broader range of animal species affected by chlamydiosis than previously known, with the potential for both mild and severe disease outcomes. This includes significant zoonotic implications, such as the potential for *C. felis* to cause ocular and respiratory infections in cats and similar symptoms in dogs, and the role of *C. psittaci* in reproductive issues. The diverse clinical manifestations and the wide host range underscore the need for further research to fully elucidate the pathogen's presence in various organs and its broader epidemiological impact.

Human-transmittable *Chlamydia* species pose a serious threat to public health because they can cause atherosclerosis, pneumonia, coronary heart disease, and a host of other illnesses. Depending on the species, the pathogen can cause different symptoms, which can lead to severe pathologies in humans. Some species can be transmitted from person to person (Wu et al., 2013; Stein & Thompson, 2023). *C. psittaci*, *C. pneumoniae*, and *C. trachomatis* are the three main human pathogenic representatives. The only natural host of *Chlamydia trachomatis* is humans, and it is parasitic on the genital and conjunctival epithelia. The infection can spread to the upper genital tract, where it can result in endometritis, salpingitis, tubal factor infertility, ectopic pregnancies, miscarriages, and pelvic inflammatory disease. The bacteria can also cause trachoma, an ocular infection that is the most common infectious cause of blindness in the world, pneumonia, and inclusion conjunctivitis in adults and neonates. After spreading from zoonotic animals to humans, *Chlamydia pneumoniae* became adapted. The majority of *C. pneumoniae*-related respiratory infections are mild or asymptomatic. Along with upper and lower respiratory infections like community-acquired pneumonia, pharyngitis, and bronchitis, as well as ocular infections like follicular conjunctivitis, *C. pneumoniae* has also been linked to lung cancer, asthma, arthritis, and chronic neurological disorders like Alzheimer's disease. It is also the most frequently implicated infectious agent in the pathophysiology of atherosclerotic cardiovascular disease. Psittacosis, also known as ornithosis, is a human systemic zoonotic disease that is primarily caused by the avian pathogen *C. psittaci*, which is widely distributed throughout the world. Human disease can range from asymptomatic to severe respiratory failure and multi-organ failure, and it typically manifests as a flu-like illness or community-acquired pneumonia. Some of the mentioned pathogens have also been found in dogs. However, it has not been confirmed that they can act as carriers (Sprague et al., 2009; Stein & Thompson, 2023). *C. felis* has not been detected in humans, but, as carriers of this zoonosis, cats and dogs can infect humans. Due to the limited number of studies conducted on domestic animals such as dogs, it is difficult to assess the prevalence of this disease, its potential transmission to humans, and to diagnose this pathology in humans (Wu et al., 2013). Overall, human-transmittable *Chlamydia* species represent a significant public health concern due to their potential to cause a range of serious illnesses, including respiratory diseases, cardiovascular conditions, and ocular infections. The diverse symptoms and severe pathologies associated with *C. psittaci*, *C. pneumoniae*, and *C. trachomatis* highlight the need for vigilant monitoring and effective management strategies. Although *Chlamydia* species have been identified in domestic animals, their role as carriers and the risk of zoonotic transmission to humans remain understudied. Improved research and surveillance are essential to better understand these risks and enhance diagnostic and preventative measures to protect public health.

Diagnosing chlamydia in farm animals and birds is usually straightforward when considering the disease history, visible symptoms, and pathologies. However, in domestic animals, this disease often has non-specific symptoms and is rarely diagnosed. Some animals may be asymptomatic, so the only way to confirm the disease is through laboratory tests (Sachse et al., 2009; Domrazek & Jurka, 2024). *Chlamydiae*

are obligate that must be isolated and propagated using tissue culture methods. Diagnosing chlamydial infections in birds and mammals essentially involves two methods. In the first method, the agent is directly found in tissue or swab samples; in the second, blood samples are serologically screened for the presence of anti-chlamydial antibodies. In the end, the test that is performed depends on the kinds of samples that are brought to the diagnostic laboratory for examination (Sachse et al., 2009). Traditionally, the most reliable and sensitive technique of detection for determining the presence of a chlamydial infection has been the pathogen isolation. The main drawback of this method is that it depends entirely on the biological samples being transported and stored properly to maintain the organism viability. There may also be contamination problems caused by other Gram-negative bacterial species, which can lead to false-positive reactions and erroneous diagnosis, depending on the kind of sample submitted for analysis and the detection technique used. At least two of the veterinary pathogens, *C. psittaci* and *C. abortus*, are zoonotic and can seriously infect humans, therefore handling these pathogens raises concerns about safety. This method was not used in the articles under review (Sachse et al., 2009). Since most commercially available antigen detection tests developed over the past 25 years are based on the family-specific LPS antigen, they should theoretically also be suitable for detecting chlamydial infections in animals. However, they are primarily and extensively used for the detection of *Chlamydia trachomatis* infections in human clinical specimens. These immunoassays consist of solid-phase ELISAs, plate-based ELISAs, and direct fluorescent antibody (DFA) tests. Apart from the fact that immunoassays take less time to perform, one of the primary benefits of using them instead of cell culture for infection diagnosis is that they do not rely on viability to detect soluble LPS antigen in secretions and both viable and non-viable EBs (Sachse et al., 2009). Antibody detection methods are commonly used in studies of both humans and animals. However, in recent years, PCR methods have been employed for diagnosing this disease in European countries. In China, manufactured tests like indirect hemagglutination test (IHA) were used, but there is no information or description of studies conducted with these tests in Europe. In a study using IHA conducted in China, the prevalence of *Chlamydia* was identified in dogs with antibodies against *Chlamydia*. In another study, specific *C. felis* antibodies were found in both dogs and cats. However, the methodology of these studies did not include information on the specificity or sensitivity of the method, nor did it address potential errors in the research (Sachse et al., 2009; Wu et al., 2013; Tian et al., 2014). In Europe, a study into antibody detection was performed using the direct immunofluorescence (DIF) method. Conducted in Lithuania, this study assessed the prevalence of *Chlamydia* among dogs with clinical symptoms and among those without. The study did not differentiate between *Chlamydia* species. It found that the prevalence of *Chlamydia* in the examined population was as high as 61.9% (Liutkeviciene et al., 2009). Many studies using the PCR method have been conducted to identify *Chlamydia* species and to apply this method for diagnosing the disease. In the literature, numerous PCR protocols have been proposed. Targets in the ompA gene or the ribosomal RNA operon are the basis for most of the published conventional PCR techniques (Sachse et al., 2009). In a study conducted in Germany using the PCR method, a dog was identified with *C. psittaci*, confirming the disease and indicating that this pathogen could potentially be transmitted to humans (Sprague et al., 2009). While conventional PCR can only confirm the presence or absence of a given pathogen, real-time PCR additionally enables the diagnostician to quantitate the amount of this agent present in the sample. One major benefit of real-time PCR is that it eliminates the need for post-PCR sample handling, which leads to more rapid and high-throughput assays and eliminates the possibility of PCR product carry-over contamination (Sachse et al., 2009). In a study using real-time PCR to assess the prevalence of

chlamydiosis in dogs conducted in Sweden, no positive cases were found. However, in Poland, only one dog was found to have this pathogen out of 130 dogs tested, although its exact species was not determined (Domrazek & Jurka, 2024; Holst et al., 2010). There are numerous methods and assays available for diagnosing chlamydial infection, including those that either identify antigen in tissue and swab samples directly or identify anti-chlamydial antibodies in blood samples. The introduction of an alternative gold standard consisting of a combination of independent DNA tests seems like a viable and realistic course of action to follow, given the well-established benefits of PCR and other DNA amplification tests over chlamydial cell culture in terms of sensitivity, throughput, and time consumption. A significant number of studies using PCR are conducted among humans and animals, with ongoing improvements and standardisation of the methodology for diagnostic purposes (Sachse et al., 2009). In conclusion, diagnosing chlamydial infections in both domestic and farm animals involves a range of methods, from traditional pathogen isolation and serological testing to more recent PCR techniques. While traditional methods rely on the viability of samples and can be hindered by contamination, PCR and immunoassays offer faster, more reliable results without needing viable organisms. The adoption of real-time PCR has further enhanced diagnostic accuracy and efficiency, yet variability in methods and reporting, particularly across different regions, underscores the need for standardised protocols. The combination of various diagnostic approaches, including the potential for new DNA-based tests, promises to improve the detection and management of chlamydial infections in both animals and humans.

TICK TRANSMISSION OF BRUCELLOSIS AND CHLAMYDIOSIS

The possibility of ticks carrying a variety of diseases that humans, livestock, wildlife, and even domestic animals can contract makes tick-borne diseases (TBDs) extremely concerning. Over the past few years, both microclimate and macroclimate have changed, as has human behaviour, which can increase the risk of TBDs (Ma et al., 2024).

Chlamydia was first detected in ticks in 2014, in China, from several different climatic regions within the country (Tian et al., 2014). In this study Croxatto et al. used the real time PCR method and found that 28.1% of ticks were infected with chlamydia. The positive samples were categorised according to the tick developmental stages: 31.1% of nymphs and 28.1% of adult ticks and 38.9% in mixed adult/nymph pool were found to be infected. However, the *Chlamydia* species were not identified (Croxatto et al., 2014). In 2015 and 2020, similar studies were conducted in European countries, specifically in Switzerland and Italy (Pilloux et al., 2015; Chisu et al., 2020). In Switzerland, a study using the PCR method was conducted to assess the overall prevalence of *Chlamydia* in ticks. It was found that 6.4% of ticks were infected. Specifically, the infection rates were 6.9% in nymphs and 5.8% in adults. This study was carried out in various locations across the country with different climatic conditions. However, the *Chlamydia* species were not identified during it (Pilloux et al., 2015). In Italy, a study investigated ticks collected from animals such as dogs and cats to determine the potential pathogens they carried. Despite the small sample size, which warrants cautious interpretation, *Chlamydia* was detected. Specifically, 46% of the ticks (17 ticks) were found to carry *Chlamydia*. Among these, *C. abortus* was identified in most ticks, while one tick was found to carry *C. psittaci*. However, it was not confirmed whether this pathogen could have been transmitted from animals or if the tick was a vector for the pathogen. Additionally, this study was conducted in a single location within the country, so the prevalence across the entire country cannot be assessed (Chisu et al., 2020).

Several studies have shown that ticks can carry *Brucella* bacteria. In 2024, a review article was conducted analysing the role of ticks in the transmission of brucellosis. One of the earliest studies conducted in 1937 demonstrated

that ticks could act as carriers of brucellosis. The study found that the pathogen could survive in ticks for a certain period while the research was ongoing. Additionally, live pathogens were detected in the faeces of the ticks. In the analysed studies, brucellosis was detected in 16 different tick species, with a prevalence reaching as high as 33.87% (2524 ticks). In different developmental stages of ticks, brucellosis was found in 40.9% of larvae and 4.6% of female ticks. The above studies found the pathogen in various tick tissues, such as the salivary glands; also, the pathogen adapted to primary intracellular environment within the tick cells. It has been established that ticks are one of the hosts for this pathogen and can transmit it to animals. However, it has not yet been proven that ticks can transmit this pathogen between themselves. From 1963 to 2019, several sources suggested that ticks might transmit the *Brucella* pathogen to humans. However, due to insufficient information, it is difficult to confirm this. The disease can be transmitted to humans through various routes, which complicates the validation of tick-borne transmission (Ma et al., 2024).

Tick-borne diseases are still not fully understood, making it challenging to determine whether ticks can act as carriers for these diseases in some cases. As tick prevalence changes with shifting climates, it poses a risk to farm animals, domestic animals, and wildlife, which may potentially transmit diseases to humans. Ticks also represent a significant risk factor for disease transmission to humans, not only for known diseases but also for those that have not yet been identified, which can lead to serious health issues. For instance, these two zoonoses cause various symptoms, and their diagnosis remains complex for both humans and animals (Ma et al., 2024; Chisu et al., 2020; Croxatto et al., 2014).

CONCLUSIONS

Brucella canis has become an important cause of canine brucellosis, especially in environments with a high density of dogs, such as shelters and breeding facilities. This pathogen exhibits distinct characteristics, including a rough phenotypic appearance and specific infection mechanisms, which contribute to its unique clinical and diagnostic challenges. *B. canis* is primarily recognised for its impact on canine health, there is also a growing concern about its potential zoonotic risk.

Chlamydia bacteria, significant pathogens for both humans and animals, present a diverse array of acute, chronic, and asymptomatic infections across various hosts. In domestic animals, especially dogs, *Chlamydia* infections can lead to a range of clinical symptoms, from ocular and respiratory diseases to reproductive issues.

Both *B. canis* and *Chlamydia* spp. are Gram-negative intracellular and zoonotic pathogens that can be found in certain organ systems. Several of this overlap and could facilitate the detection of these pathogens: the reproductive system, urinary system, spleen, liver, and respiratory tract. These two diseases either do not cause inflammatory reactions or cause them periodically. In both diseases, transmission occurs through secretions from the nose, eyes, urine, vagina, or semen. Their detection can be challenging due to their ability to localise in multiple organs and bodily fluids.

The complexities of diagnosing canine brucellosis, exacerbated by the ability of the pathogento persist in various tissues and evade immune responses, highlight the need for advanced and accurate diagnostic techniques. Current methods, ranging from serological tests like RSAT and AGID to molecular techniques such as PCR and LAMP, each have their own strengths and limitations, therefore more sensitive and specific tests are needed. The detection of *Chlamydia* spp. in dogs, including instances of *C. psittaci*, suggests diagnostic techniques, including PCR and serological assays, have improved detection capabilities. While traditional culture methods remain sensitive, they are labour intensive and prone to contamination. Modern molecular techniques like real-time PCR offer enhanced sensitivity and specificity, though their application can be variable based on regional practices and available resources.

Tick-borne diseases (TBDs), including those caused by *Brucella* and *Chlamydia*, have been on the rise, and studies have identified ticks as potential carriers for both pathogens. In recent years, *Chlamydia* was detected in a substantial percentage of ticks in various regions, though the exact species and vector potential remain unclear. Similarly, *Brucella* has been found in ticks, with historical and recent studies indicating their role in pathogen transmission.

Both *Brucella* and *Chlamydia* pose significant health risks to animals and humans, with their complex biology and broad host ranges presenting ongoing challenges for diagnosis and management.

Received 4 August 2024
Accepted 21 August 2024

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Viktorija Petrauskaitė, Gintaras Zamokas, Inga Stadalienė, Birutė Karvelienė

BRUCELLA CANIS IR *CHLAMYDIA* SPP.: ĮŽVALGOS APIE ŠUNŲ UŽSIKRĖTIMĄ, DIAGNOSTIKĄ IR GALIMĄ INFEKCIJOS PLATINIMĄ PER ERKES

Santrauka

Brucella ir *Chlamydia* spp. yra zoonotiniai sukėlėjai, turintys įtakos tiek žmonių, tiek gyvūnų sveikatai. *Brucella* rūšys sukelia bruceliozę, pasireiškiančią įvairiais klinikiniais simptomais, ir kitus reikšmingus zoonotinius pavojus. *Brucella* infekcijos, nors ir rečiau nustatomos šunims, kelia susirūpinimą dėl galimo perdavimo žmonėms, tad būtina nuolat stebėti šias tendencijas veterinarijos praktikoje. *Chlamydia* spp. būdingas didelis patogeniškumas pasireiškia tiek lengvomis infekcijomis, tiek sunkiomis sisteminėmis ligomis. *Chlamydia* infekcijos šunims gali sukelti konjunktyvitą, kvėpavimo sistemos ir reprodukcinius sutrikimus, įskaitant nevaisingumą ir priešlaikinį palikuonių atvedimą. Labai svarbu sistemingai stebėti ir kontroliuoti zoonotinę riziką turinčius *Chlamydia* sukėlėjus, ypač *Chlamydia felis*, kadangi jie veikia ir naminius gyvūnus, ir žmones. Manoma, kad perduodant šiuos patogenus erkės vaidina svarbų vaidmenį. Mokslinių tyrimų metu erkėse buvo aptikta *Chlamydia* spp., bet epidemiologinė reikšmė dar nėra aiški. Nors *Brucella* taip pat buvo rasta erkėse, tačiau trūksta tikslių įrodymų apie erkių perduodamą infekciją žmonėms ar gyvūnams. Siekiant valdyti *Brucella* ir *Chlamydia* infekcijų riziką tiek gyvūnams, tiek žmonėms tobulinami diagnostikos metodai ir ruošiami kontrolės planai daugiausia dėmesio skiriant erkėms kaip galimoms platintojoms.

Raktažodžiai: *Brucella canis*, šunų bruceliozė, *Chlamydia*, zoonozė, erkės, perdavimas