



FELINE BARTONELLOSIS – A MINI REVIEW

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Summary

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An up-to-date literature overview of bartonellosis in cats was made. Its seroprevalence, etiological characteristics, epidemiological features and clinical manifestations are outlined. The emphasis is placed on its laboratory diagnostics, therapy and prevention in cats. Human bartonellosis with data from Bulgaria is described.

Key words: *Bartonella henselae*, bartonellosis, cats, zoonosis

Feline bartonellosis, also known as cat-scratch disease (CSD) or felinosis, is a systemic vector-borne infectious diseases with a global distribution, encountered in humans and many animal species (Mogolon-Pasapera *et al.*, 2009). The disease has been clinically identified in people during the 1930s, but was first described in 1950 by Debré *et al.* It was initially believed that the causative agent was a virus and consequently, the Gram-negative bacterium *Afipia felis* was incriminated as the most probable infectious agent of CSD. Only in the early 1990s, the

leading role of *Bartonella henselae* (*B. henselae*) in CSD etiology was confirmed, and two years later (July 1992), the agent was proved in a cat with a healthy owner. The cats are acknowledged as the primary and most important reservoir of these bacteria, capable to transmit the pathogen to humans by scratching or biting (Skerget *et al.*, 2003). Although infected cats rarely present clinical signs, they remain sub-clinically infected over months and years. In humans, these bacteria cause a variety of disease manifestations ranging from

asymptomatic bacteraemia to chronic exhaustion and death (Pitassi *et al.*, 2015).

PREVALENCE

The seroprevalence rate among cats in Europe does not correspond to the small number of reported clinical cases mainly due to underestimated diagnostics of *Bartonella* spp. infections. In Greece, reported seropositivity rate among cats was 35.4% (Kokkinaki *et al.*, 2022). A comparable incidence – 54.8% was reported by an Italian team (Persichetti *et al.*, 2016) and by a survey in The Netherlands that detected a seropositivity rate of 51.8% (Bergmans *et al.*, 1997). Regarding the seroprevalence in Spain, reported data in tested cat populations ranged from 24.7% to 71.4% (Pons *et al.*, 2005; Ravicini *et al.*, 2016). A similar seroprevalence was reported from Denmark – 45.6% (Chomel *et al.*, 2002) and France – 41.1% (Gurfield *et al.*, 2001) among domestic and stray cats. A high seroprevalence among cats was evidenced in Germany – 68.7% (Mietze *et al.*, 2011) and in Portugal – 64.9% (Alves *et al.*, 2009). In Bulgaria, studies regarding the presence and seroprevalence of *B. henselae* among cats are not available.

ETIOLOGY

B. henselae is a small (0.3 to 0.6 by 1.0 to 1.7 µm), thin, short and slightly curved Gram-negative rod-shaped intracellular bacterium (Deng *et al.*, 2012). It is taxonomically related to the genus *Bartonella* from family *Bartonellaceae*. More than 30 *Bartonella* species have been isolated/detected in people, domestic and wild animals, with at least 13 species and subspecies known to be associated with human diseases (Álvarez-Fernández *et al.*,

2018). There is scientific evidence demonstrating that the major part of human clinical infections are associated with three *Bartonella* species – *B. bacilliformis*, *B. quintana* and *B. henselae*. These bacteria are extremely fastidious and grow slowly on nutrient media (Donovan *et al.*, 2018). *Bartonellae* can be cultured on enriched culture medium containing blood because they are highly dependent on hemin. The optimal growth temperature varies between species, being 25 to 30 °C for *B. bacilliformis* and 35 to 37 °C for *B. quintana* and *B. henselae*. The formation of visible colonies on enriched media may take 2 to 6 weeks. The isolation from clinical samples is more difficult due to previous antibiotic use and intracellular presence of the microorganism. Because of the slow growth of these bacteria, standard biochemical methods of identification may not be applicable. A new, chemically modified insect-based liquid culture medium (Bartonella alpha-Proteobacteria growth medium, BAPGM) was recently developed for the isolation of at least seven *Bartonella* species and other uncultured bacteria from blood and tissue samples (Diddi *et al.*, 2013). On the basis of 16S-rDNA sequences, *B. henselae* belongs to genotype I and genotype II groups (Duscher *et al.*, 2018). These strains have different point mutations, protein profiles, host specificity, prevalence and pathogenicity (Castel *et al.*, 2019). Genotype I (Houston) is encountered more often in humans and is spread in Asia, whereas the genotype II (Marseille) is prevalent mainly among cats and is encountered in Europe, Australia and the USA (Mazurek *et al.*, 2020). It is considered less pathogenic, as human CSD infections are more likely due to genotype I (Maggi *et al.*, 2013).

With regard to the susceptibility to disinfectants and high/low temperatures, information specific to *B. henselae* is not available, but most bacteria have been shown to be susceptible to low concentrations of chlorine, 70% ethanol, phenolics such as orthophenylphenol, ortho-benzylpara-chlorophenol, 2% aqueous glutaraldehyde, peracetic acid (0.001% to 0.2%) (Rutala *et al.*, 1998), and can be inactivated by moist heat (121 °C for 15–30 min) and dry heat (160–170°C for 1–2 hours) (Pflug *et al.*, 2001).

EPIDEMIOLOGY

Cats (*Felis catus*) are the primary but not the only reservoir of *B. henselae* (Jaffe *et al.*, 2018), the agent of cat-scratch disease. Besides, cats are the main hosts for *B. clarridgeiae* and *B. koehlerae* (Cheslock *et al.*, 2019; Logan *et al.*, 2019). They may become infected also with *B. bovis* (*ex weissii*) and *B. quintana*, yet the role of cats in the epidemiology of these two *Bartonella* spp. is not clearly acknowledged (Chomel *et al.*, 2004). The prevalence of *B. henselae* among male and female cats is not significantly different. Age-related predisposition does not exist, although studies on naturally and experimentally infected cats have established chronic bacteraemia mainly in cats younger than one year compared to adult cats (over 1 years of age). Also, stray cats are more frequently affected compared to domestic ones (Chomel *et al.*, 2006). Different *Bartonella* species have been isolated from many other animal and wild species, including bats, birds, dogs, cattle, deer, marine mammals, rodents, sheep and reptiles (Jones *et al.*, 2008; Alsarraf *et al.*, 2017).

The transmission of *B. henselae* occurs through blood-sucking vectors –

mainly fleas (*Ctenocephalides felis*), lice, sand flies, biting flies and ticks (*Ixodes ricinus* – transstadial transmission of *B. henselae*). Vector-borne transmission is realised through two main pathways: inoculation of faeces from blood-sucking *Bartonella*-infected arthropods (the organism survives 3 days in flea faeces) (Kosoy *et al.*, 2019) via cat scratches or bites (Mosbacher *et al.*, 2011) or direct bite of the host by a vector (Battisti *et al.*, 2015). Healthy cats reared together with infected ones in a specific ectoparasite-free environment are not seropositive, which emphasises the significance of vectors in disease transmission (Mosbacher *et al.*, 2011). There are no data for transmission of *B. henselae* between bacteraemic female cats and non-infected males during mating, as well as from infected pregnant females to kittens or during the neonatal period (no vertical transmission) in a flea-free environment (Guptill *et al.*, 1997). It should be noted that non-vector transmission e.g. to veterinarians through needle wounding (Oliveira *et al.*, 2010) or via blood transfusion as documented experimentally in cats, dogs and humans (Wardrop *et al.*, 2016; Lins *et al.*, 2019) is also possible.

CLINICAL SIGNS

Most cats, naturally infected with *B. henselae*, show no clinical signs and appear to tolerate chronic bacteraemia. There are however investigations demonstrating that persisting infections in cats are associated with a great variety of clinical signs with continuously increasing spectrum of manifestations – from subclinical bacteraemia (in immunocompetent animals and people) to endocarditis, left ventricular endomyocarditis, diaphragmatic myositis, mild non-specific fever, in-

creased risk of gingivostomatitis, lymphadenomegaly, transient mild behavioural or neurological signs; pyogranulomatous myocarditis and uveitis, conjunctivitis, keratitis and corneal ulcers (Breitschwerdt, 2017; Donovan *et al.*, 2018; Berkowitz *et al.*, 2020). The manifestation of these disease states is associated with the different virulence of *Bartonella* species and strains, the route of transmission, host immune status, concomitant infectious or non-infectious diseases, immunosuppression (*FeLV*) and malnutrition (Álvarez-Fernández *et al.*, 2018).

Despite the bacteraemia, the complete blood cell counts, blood serum biochemical profiles and urinalysis results of cats are often normal. In diseased cats, deviations in some laboratory indices are reported – anaemia, eosinophilia, hyperproteinaemia, hyperglobulinaemia, neutropaenia and thrombocytopaenia (Aylló *et al.*, 2012).

DIAGNOSTICS

So far, there is no diagnostic test whose negative result ensures the absence of *Bartonella* spp. infection. This is a real challenge to definitive diagnosis. In the beginning, the isolation of *Bartonella* spp. from blood samples was considered a gold standard (Breitschwerdt *et al.*, 2010). The growth of these bacteria takes too long (from 9 days to 6–8 weeks), and requires specific nutrient medium and cultivation conditions (Agan & Dolan, 2002). Also, it should be remembered that patients may be occasionally bacteraemic, as documented in experimental feline *B. henselae* infections. The culture method provides a definitive diagnosis (indicates active infection). Limitations of the study is the poor sensitivity from all specimen types (Rodino *et al.*, 2019).

One of the most employed methods of *Bartonella* spp. detection in peripheral blood and other clinical samples (cerebrospinal fluid, joint effusion, oral swabs, lymph nodes/other tissue samples or aspirates) is the PCR, targeting genes such as *gltA*, *groEL*, *pap31*, *ftsZ*, *16rRNA* and *16S-23S ITS* region (Urdapilleta *et al.*, 2020). During specimen collection, care should be taken to avoid the contamination of samples with flea faeces from the skin surface and thus, a false positive PCR result. *Bartonella*-specific RT-PCR is highly sensitive and specific in the examination of tissue samples. The limitations of the method are low sensitivity in the examination of blood samples. Sequencing and *16S rRNA* PCR reduces the need for targeted pathogen testing, but the sensitivity is lower compared with pathogen-specific PCR (Rodino *et al.*, 2019).

Serological tests used for detection of anti-*B. henselae* antibodies (IgG and IgM) comprise the immunofluorescence assay (IFA) and ELISA. IFA is preferred due to its better performance characteristics and ability to provide semiquantitative titers. The results from tests should be interpreted very carefully as the presence of antibodies indicates only a prior exposure to the pathogen, but not necessarily an active infection. IFA results of $\geq 1:64$ for IgG-class antibodies to *B. henselae* are considered positive, with sensitivity ranges of 88% to 98% reported across different assays (Vermeulen *et al.*, 2007). Seroprevalence studies performed across multiple countries have shown that 3% to 30% of otherwise healthy individuals may be seropositive for IgG-class antibodies to *B. henselae* using this threshold titre. However, the majority of these positive results are at titres of $\leq 1:128$, and for this reason a single-time-point, low-level positive IgG titre should be interpreted

with caution. Reasons for this reactivity are likely due to prior exposure to the bacterium or possibly cross-reactivity with other *Bartonella* spp. or organisms, including *C. burnetii*, *Chlamydophila pneumoniae*, or spotted fever group *Rickettsia*. Given these findings, IgG titres of $\geq 1:256$ and/or a 4-fold rise in IgG antibodies between acute and convalescent-phase samples are considered indicative of current or recent infection (Rodino *et al.*, 2019). Assays for detection of IgM-class antibodies against *B. henselae* are considerably less sensitive (range, 50% to 62%), with titres of $\geq 1:20$ considered positive. IgM-class antibodies to *Bartonella* species are typically present early in infection and wane 8 to 10 weeks thereafter, suggesting that IgM testing may not be helpful during later stages of disease (Vermeulen *et al.*, 2007). The reported specificity of IgM assays is high (95%), and notably higher than that reported for IgG assays (69% to 89%), although false-positive results with *C. burnetii* and *C. pneumoniae* infections may still occur. A positive anti-*Bartonella* IgM result alone should be considered cautiously and interpreted alongside the timing of the potential exposure and duration of patient symptoms. Convalescent testing of a new specimen collected 2 to 3 weeks later is recommended to show seroconversion of anti-*Bartonella* IgG and rule out a false-positive IgM result (Rodino *et al.*, 2019). Despite its good specificity, these tests are poorly sensitive regarding the prediction of bacteraemia in potentially ill cats. It is important to note that some infected cats and dogs have no antibodies against *B. henselae* regardless of the fact that the agent may be identified in blood/tissues through culturing or PCR (Golly *et al.*, 2017).

Histopathology/immunohistochemistry

method identifies directly the microorganism in tissue samples from cats, dogs and people and shows the relationship between antigen localisation and histopathological lesions, and may also be used for *Bartonella* spp. detection (Caponetti *et al.*, 2009; Buchmann *et al.*, 2010). Limitations on method are the risk of identifying a contaminant, lack of pathogen-specific stains, background/nonspecific staining, which make the interpretation difficult (Rodino *et al.*, 2019).

TREATMENT

While many antibiotics demonstrate good *in vitro* effects, their *in vivo* efficacy appears to be limited. The choice of antibiotic should be tailored to predominant clinical disease signs (ophthalmic, cardiac, neurological, bacteraemia). Therapeutic 4–6-week courses of several antibiotics (doxycycline, amoxicillin-clavulanic acid, marbofloxacin, azithromycin) have been used in cats with natural or experimental *Bartonella* spp. infection. In most instances, the treatment did not result in complete elimination of *Bartonella* spp. in all cats (Varanat *et al.*, 2009; Guptill, 2010). Due to the broad use of antibiotics and emerging antimicrobial resistance, antibiotic administration is not routinely recommended for infected adult cats (over 1 year of age) without clinical signs. In infected cats with clinical signs as well as asymptomatic young infected cats (up to 1-year-old) living in households with immunocompromised adults or children, antimicrobial therapy is necessary, and in most cases, a combination of antibiotics is needed to obtain a result. Despite insufficient clinical studies to determine treatment recommendations for feline bartonellosis, Nivy *et al.* (2022) described a clinical case in a 2.5-year-old

castrated male cat presented with fever and marked generalised lymphadenopathy, with a positive PCR for *B. henselae*. Dual antibiotic treatment comprising pradofloxacin (4.5 mg/kg, PO, q24h, for 62 days) and doxycycline (11 mg/kg, PO, q24h, for 62 days), resulted in resolution of clinical signs and agent elimination.

PREVENTION

At present, vaccines are not available. Prevention measures include control of fleas and ticks by regular application of acaricides, which interrupt efficiently the transmission of *B. henselae* among cats (Bradbury & Lappin, 2009). Humans and pets should avoid contacts with stray dogs and cats. If blood transfusion is indicated, recipient cats should not be treated with blood products from donors whose *Bartonella* spp. status is unknown or which are seropositive (Wardrop *et al.*, 2005). Nails of pets should be preferably cut occasionally to prevent scratches. In case of scratch or bite from a cat, the place should be washed profusely with water and soap and disinfected with a hand sanitiser (ethanol). Licking of open wounds by pets should not be allowed. Risk factors for bacteraemia in cats are young age, sites with high extensity of invasion with fleas and ticks, and outdoor access. If pet owners are acquainted with these recommendations and informed about the possible pathways of cat-to-cat or cat-to-human transmission of the bacterium, the zoonotic risk may be reduced to a minimum (Stützer & Hartmann, 2012).

HUMAN BARTONELLOSIS

The first *Bartonella* species pathogenic for people – *B. bacilliformis*, has been

identified in the beginning of 1900. This human-specific bacterium causes a two-phase disease (Carrión's disease) characterised with a sudden acute phase known as Oroya fever with high mortality (up to 90%), followed by a chronic phase with affection of the skin with prominent red-purple nodules known as Peruvian warts (*verruca peruana*). The pathogen is transmitted by the sand fly (*Lutzomyia verrucarum*). The second species pathogenic for humans is *B. quintana* transmitted by the body louse (*Pediculus humanus humanus*), which is associated with trench fever – with severe course with immunocompromised subjects with endocarditis, generalised lymphadenopathy and bacillary angiomatosis (Billeter *et al.*, 2008). *B. henselae* is the third most common zoonotic pathogen transmitted from cats to people, associated with the cat scratch disease (felinosis, feline bartonellosis). Different investigations in humans from the early and mid 1990s established a seroprevalence rate of 4–6% (Zangwill *et al.*, 1993). In a newer report, 23% of studied people had antibodies against *B. henselae*. No differences between humans owing or not a pet, as well as regarding age, sex, urban or rural residence, presence or not of concomitant illnesses were found out (Skerget *et al.*, 2003). The transmission of *B. henselae* from cats to people may occur either directly, usually through bites and scratches or indirectly, through vectors (fleas and ticks) (Stützer & Hartmann, 2012). In most immunocompetent people, *B. henselae* infection has no clinical manifestations. Nevertheless, in some subjects (most commonly in children) the disease course is outlined with a self-limiting regional lymphadenopathy and development of primary papular lesion at infection entrance door (Massei *et al.*, 2004). Sometimes, lymph

nodal abscesses or systemic signs (fever) are reported. In immunocompromised people, atypical and severe forms of the disease may develop, whose outcome may be fatal if left untreated. Symptoms include bacillary angiomatosis, parenchymal bacillary peliosis, recurrent fever with bacteraemia, endocarditis, neuroretinitis, aseptic meningitis and uveitis (Font *et al.*, 2011).

In Bulgaria, the first clinical case associated with *B. henselae* was described by Komitova *et al.* (2003) in a 14-year-old girl with Parinaud oculoglandular syndrome, after contact with kittens. A disease-specific clinical case was observed by Stanimirov (2013) in a 22-year-old woman with a typical skin papule in the entrance door area and protracted suppurative lymphadenitis in the parotid gland and neck region. In 2021, Todorova & Cherninkova described a unilateral neuroretinitis with laboratory confirmed *B. henselae* in a male patient, 30 years of age, living together with 10 cats, 6 dogs and a parrot.

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