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SHORT COMMUNICATION

High Prevalence of *Bartonella henselae* and *Bartonella quintana* Antibodies in Croatian Patients Presenting with Lymphadenopathy

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Abstract

Between 2007 and 2010, a total of 268 Croatian patients with lymphadenopathy were tested for IgM/IgG antibodies to *Bartonella* (*B.*) *henselae* and *B. quintana*. Samples from 44.4% patients showed positive IgG antibodies: 35.8% to *B. henselae*, 6.7% to *B. quintana* and 1.9% to both *Bartonella* species. There was no difference in seropositivity between males and females (47.4% *vs.* 41.5%). Seroprevalence was high in all age groups (40.4–60.9%). Patients from urban and rural areas showed a similar seroprevalence rate (44.1% *vs.* 44.8%). Positive IgM antibodies were found in 28.3% patients varying from 17.5% and 37.5% among age groups. Most cases were reported from August to March.

Key words: *Bartonella*, croatia, lymphadenopathy, prevalence

Bartonella (*B.*) *henselae* and *B. quintana* are the causative agents of cat-scratch disease, the most common zoonosis caused by *Bartonella* spp. (Boulouis *et al.*, 2005). Domestic cats are the main reservoir of *B. hensellae*, which is transmitted among cats by the cat flea (Chomel *et al.*, 2004). Seroepidemiological studies have demonstrated the worldwide distribution of *B. henselae* infection in domestic cats with antibody prevalence from 5–86% (Podsiadly *et al.*, 2002; Fabbi *et al.*, 2004; Boulouis *et al.*, 2005; Celebi *et al.*, 2009). Transmission to humans mainly occurs directly by a cat scratch and possibly *via* a cat bite (Boulouis *et al.*, 2005; Breitschwerdt *et al.*; 2007; Westling *et al.*, 2008). In immunocompetent individuals, cat-scratch disease is characterized by a benign regional lymphadenopathy. Low-grade fever, malaise and aching are often reported (Ridder *et al.*, 2002; Chomel *et al.*, 2004). Since the isolation of *Bartonella* spp. is difficult, serologic tests are commonly used for etiologic diagnosis of cat-scratch disease (Vermeulen *et al.*; 2007, Hoey *et al.*, 2009).

Human *Bartonella* infections have been reported from several continents, including Europe (Chomel *et al.*, 2004; Lamas *et al.*, 2008). The prevalence of antibodies varies in different geographic regions and among the individuals living in the same geographic area (Maruyama *et al.*, 2000; Massei *et al.*, 2003; da Costa *et al.*, 2005). Seropositivity is lower among blood donors (McGill *et al.*, 2005) and up to 53.3% in certain risk groups such as veterinarians and cats owners (Chmielewski *et al.*, 2007).

In Croatia, data on the prevalence of bartonellosis are very limited (Pandak *et al.*, 2009). The aim of this study was to determine the prevalence of *B. henselae* and *B. quintana* in patients presented with lymphadenopathy.

During a four year period (2007–2010), a total of 268 serum samples from children and adults aged 1–73 years presented with lymphadenopathy were tested for the presence of specific IgM and IgG antibodies to *B. henselae* and *B. quintana*. Serologic tests were performed using commercial indirect immunofluorescence assay (Euroimmun, Lubeck, Germany). According to manufacturer recommendation, titer ≥100 for IgM and ≥320 for IgG were considered positive. Chi-square test was used to compare differences between groups. $P < 0.05$ was considered to be statistically significant.

Serum samples from 119/44.4% (95% CI = 38.6–50.4) patients showed IgG antibodies to *Bartonella* spp. Ninety-one patients (35.8%) were seropositive to *B. henselae*, 18 (6.7%) to *B. quintana* and 5 (1.9%) to both *Bartonella* species.

Prevalence of *B. henselae*/*B. quintana* antibodies according to characteristic of participants is shown in Table I. The difference in IgG seropositivity between males and females was not significant (47.4% vs. 41.5%, $p = 0.397$). According to age, IgG seropositivity rates varied from 40.4% to 60.9% with no statistically significant differences between age groups ($p = 0.669$). A similar IgG seroprevalence was found in patients residing in urban areas (44.1%) and patients residing in rural areas (44.8%, $p = 0.999$).

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Table I
Prevalence of *B. henselae*/*B. quintana* antibodies according to characteristics of participants

Characteristic	Tested N/%	IgM positive N/%	95% CI	p value	IgG positive N/%	95% CI	p value
Gender				0.066			0.397
Male	133/49.6	45/33.8	26.3–42.2		63/47.4	39.1–55.8	
Female	135/50.4	31/22.9	16.6–30.8		56/41.5	33.5–49.9	
Age group (years)				0.386			0.669
<10	82/30.6	27/32.9	23.7–43.7		36/43.9	33.7–54.7	
10–19	57/21.3	17/29.8	15.5–42.7		23/40.4	28.6–53.3	
20–29	24/8.9	9/37.5	21.1–57.4		11/45.8	27.9–64.9	
30–39	42/15.7	9/21.4	11.5–36.1		17/40.5	27.0–55.5	
40–49	23/8.6	7/30.4	15.4–51.1		14/60.9	40.7–77.9	
50+	40/14.9	7 /17.5	8.3–32.3		18/45.0	30.7–60.2	
Area of residence				0.661			0.999
Urban	152/56.7	41/26.9	20.5–34.5		67/44.1	36.4–52.0	
Rural	116/43.3	35/30.2	22.5–39.1		52/44.8	36.1–53.9	

Acute *B. henselae*/*B. quintana* infection was found in 76/28.3% (95% CI = 23.3–34.0) patients. The prevalence of IgM positive patients was high in all age groups (17.5–37.5%, p = 0.386) (Table I). Most cases of bartonellosis were reported from August to March with a peak incidence (%) in November (Fig. 1).

Cat-scratch disease is frequently reported in children and young adults (Podsiadly *et al.*, 2002; Massei *et al.*, 2002), but many cases may go undiagnosed in older adults (Lamas *et al.*, 2008). In this study, acute *B. henselae*/*B. quintana* infection (positive IgM antibodies) was demonstrated in 28.3% patients presented with lymphadenopathy. No significant difference in IgM seropositivity was found between age groups (17.5–37.5%, p = 0.386). A Thai study showed similar results suggesting that *Bartonella* infection may occur in various age groups (Maruyama *et al.*, 2000).

Seasonality is different in the Southern and Northern hemispheres. In Peru, most cases of cat-scratch disease occur in December and January (summer school vacation and exposure to pets) (Huarcaya *et al.*, 2002). In contrast, a study conducted in France (1999–2009) showed that the majority of cases (87.5%) were reported during September-April and peaked in December (Sanguinetti-Morelli *et al.*, 2011). This study demonstrated a seasonality of *B. henselae* and *B. quintana* infection similar to that reported for France, but shifted one month earlier (from August to March and peaked in November). This pattern may be explained by seasonality in cat reproductive behavior. In the Northern hemisphere cat reproduction increases in spring and summer, and kittens stay with their mother until 12–16 weeks of age (Chomel *et al.*, 1995). In addition, during summer cats spend most time outside the house, whereas during autumn they stay indoors (Sanguinetti-Morelli *et al.*, 2011).

In this study, the overall IgG seropositivity to *B. henselae* and/or *B. quintana* was 44.4%. A total of 35.8% patients were seropositive only to *B. henselae*, 6.7% patients only to *B. quintana* and 1.9% patients to

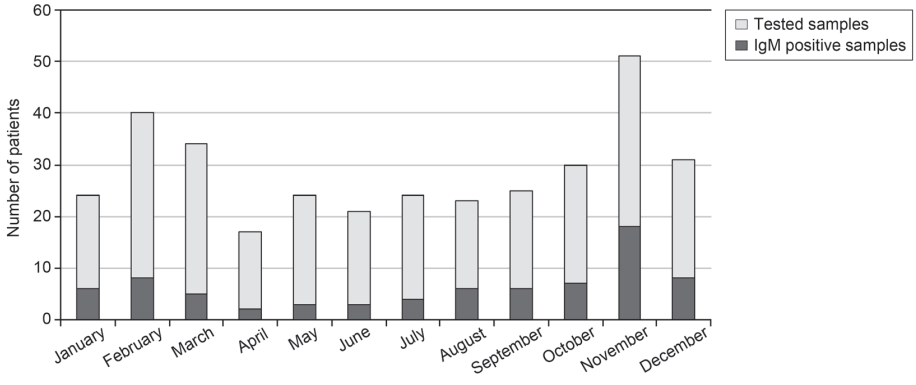

Fig. 1. Seasonal distribution of *Bartonella henselae/quintana* cases

both *Bartonella* species. Serologic studies of bartonellosis in healthy population across the Europe showed the IgG seropositivity of 8.7% in Spain (Pons *et al*., 2008), 16.3% in the United Kingdom (Harrison and Doshi, 1999), 22.4% in Greece (Tea *et al*., 2003) and 30% in Germany (Sander *et al*., 1998). However, a Polish study conducted in a group of patients with lymphadenopathy showed a higher IgG seropositivity rate (57%) (Podsiadly *et al*., 2002). In addition, a study conducted in Italy showed very high IgG prevalence (61.6%) to *B. henselae* among Italian children without evidence of cat-scratch disease (Massei *et al*., 2003).

Similar to other published studies (Harrison *et al*., 1999; Maruyama *et al*., 2000; Tea *et al*., 2003; Pons *et al*., 2008; Pandak *et al*., 2009), we observed no significant difference in the IgG seropositivity between males (47.4%) and females (41.5%). In addition, no difference in IgG seropositivity was found between children and adults which is consistent with a previous Croatian study (Pandak *et al*., 2009). The IgG seroprevalence rate was high in all age groups varying from 40.4% to 60.9%.

According to place of residence, there was no difference in the IgG positivity among patients who live in urban areas (44.1%) and patients who live in rural areas (44.8%). The other authors reported similar results (Pons *et al*., 2008; Pandak *et al*., 2009).

In conclusion, the results of this study indicate a high prevalence of cat-scratch disease (28.3%) both in children and adults presented with lymphadenopathy. Therefore, testing to *Bartonella* antibodies should be included in differential diagnosis of lymphadenitis in children as well as in adults.

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