Beata Fiecek, Tomasz Chmielewski, Grażyna Lewandowska,
Stanisława Tylewska –Wierzbanowska,

CHARACTERISTICS OF *BARTONELLA* SPP. INFECTIONS IN POLAND IN THE YEARS 2009-2012 IDENTIFIED IN THE LABORATORY OF NATIONAL INSTITUTE OF PUBLIC HEALTH - NATIONAL INSTITUTE OF HYGIENE

Unit of Rickettsiae, Chlamydiae and Spirochaetes
National Institute of Public Health – National Institute of Hygiene
Warsaw

ABSTRACT

INTRODUCTION. Various *Bartonella* species, (Gram-negative aerobic bacilli) are etiologic agents of zoonotic diseases called bartonelloses, which manifest with different symptoms depending on the bacterial species, reservoir and vector. In Poland and Europe, the most common bacterial species of the genus *Bartonella* is *Bartonella henselae*.
MATERIAL AND METHODS. Serum samples derived from patients with clinical symptoms suggesting *Bartonella* spp. infection, sent in 2009-2012 to the Laboratory of Rickettsiae, Chlamydiae and Spirochaetes of National Institute of Public Health - National Institute of Hygiene in Warsaw were tested. Levels of specific IgM and IgG antibodies to *B. henselae* and *B. quintana* antigens were detected with indirect immunofluorescence method (IFA).
RESULTS. Six hundred sixty three serum samples were examined from humans with clinical symptoms suggestive bartonellosis, in 2009-2012. Specific antibodies for *B. henselae* were detected in 435 patients (65,6%). IgM antibodies were found in 93 patients (21,4%) including 11 patients (2.5%) with IgM only. IgG antibodies were identified in 424 people (78,6%) of whom 342 had IgG antibodies only. The antibodies of both classes were detected in 82 people (18,9%). *B. quintana* infections were not found. The majority of samples for study of bartonellosis were submitted in the autumn. In patients with confirmed bartonellosis, the most common symptoms of disease were lymphadenopathy (86 people, 13%), fever (13 patients, 2%) and nodular changes in various organs (13 patients, 2%).
CONCLUSIONS. Infections caused by *Bartonella* spp. in Poland should be monitored to acquire the information on the frequency and distribution of disease in the country and their clinical course.

Key words: *Bartonella spp. infections, cat scratch disease (CSD), bartonellosis*

INTRODUCTION

Bartonella spp. are Gram-negative bacilli which are the etiological agent of bartonelloses, i.e. zoonotic diseases of diverse range of symptoms, dependent on bacteria genus, reservoir and vectors.

Bartonella henselae is the species of the genus *Bartonella* which is the most prevalent both in Poland and Europe. It is the causative agent of cat-scratch disease (CSD) which symptoms include: self-limited lymph nodes enlargement (of the head, neck, upper extremities) localized close to the bite or scratch site by cat, fever (ca 38ºC), headache, rash, fatigue and malaise.

Bartonella infection that involves liver, spleen, lungs, heart, pancreas, eyes, kidneys, joints and bones may occur in persons of decreased immunity system response, infected with HIV or HCV and organs transplant recipients. *B. henselae* can cause peliosis hepatis (PH), responsible for pathogenic liver lesions (hepatocytes necrosis) and haemorrhages (1,2,3).

B. quintana is another species of the genus *Bartonella* which can cause human diseases. This pathogen, which in the previous century was called *Rochalimaea quintana*, was the etiological agent of trench fever, occurring endemically during the World War I. *B. quintana* infections, transmitted by lice spread mainly

among soldiers in trenches. After the World War II, it was assumed that the bacteria was totally eliminated. However, at the turn of the years of 80. and 90. of the XX century, *B. quintana* infections were identified in the homeless and persons with alcohol dependence syndrome. It is considered that *B. quintana* is nowadays transmitted by cat fleas and the infection is manifested by the presence of fever, malaise, chills, sweating, anorexia, lymphadenopathy, conjunctivitis, endocarditis, persistent joint and muscle pain and numerous erythematic spots or nodules occurring in the area of thorax, abdomen and back (4,5).

Both *B. henselae* and *B. quintana* are responsible for bartonellosis manifestation called bacillary angiomatosis (BA). During the course of this disease the smooth or papillary nodules localized in brain, lymph nodes, respiratory tract, gastrointestinal tract and marrow may be present (6,7).

In 2002, the paper of Podsiadły et al. concerning the incidence of *B. henselae* and *B. quintana* in Poland in the years 1998-2001 was published. The tests were performed in the Unit of Rickettsiae of the Department of Bacteriology of NIPH-NIH. According to the obtained serological tests results for *Bartonella* spp., the highest percentage of infections was identified in mazowieckie voivodeship, i.e. 30.1% (265 serum samples were tested). The samples came also from dolnośląskie, podlaskie and łódzkie voivodeships and their respective share was 19.2%, 12% and 9.6. The lowest number of samples was sent from pomorskie and podkarpackie voivodeships. The highest incidence of bartonellosis was observed in autumn season (8).

Nowadays, bartonellosis is not subject to mandatory notification and registration in Poland. Thus, the only source of information on its occurrence is the data from diagnostics laboratories.

The paper was published many years ago; thus, it was agreed that the test results and clinical characteristics of persons diagnosed with bartonellosis would be analyzed again. The present paper aims at analyzing the current *Bartonella* spp. infections in Poland on a basis of serum samples test results which were sent to NIPH-NIH.

MATERIAL AND METHODS

The serum samples from sick persons presenting the symptoms suggestive of *Bartonella* spp. infection, which were sent in the years 2009-2012 to the Unit of Rickettsiae, Chlamydiae and Spirochaetes of National Institute of Public Health-National Institute of Hygiene, were enrolled in the study. The samples were delivered from out-patient clinics and hospitals localized in the territory of Poland.

B. henselae and *B. quintana-specific* IgM and IgG antibodies *were* evaluated by indirect immunofluorescence assay (IFA) using diagnostic kits *Bartonella* IFA IgM and *Bartonella* IFA IgG (FOCUS Diagnostic, USA) (9). Bartonella IFA IgM used suspension of *B. henselae* and *B. quintana* bacteria cultured in a yolk sac of chicken embryo, whereas Bartonella IFA IgG employed Vero cells infected with *B. henselae and B. quintana. Patients with* IgM antibodies titre of equal to or > 20 and IgG antibodies titre of equal to or > 64 were considered as indicative of infection.

RESULTS INTERPRETATION

As many as 663 serum samples collected in the years 2009-2012 from persons with symptoms typical of bartonellosis were tested. *B. henselae*-specific antibodies were detected in 435 persons (65.6%) (Tab. I). IgM antibodies were present in 93 persons (21.4%) of whom 11 persons (2.5%) had only antibodies of this class. IgG antibodies were detected in 424 persons (97.5%) and 342 of them (78.6%) had only antibodies of this class. Both IgM and IgG antibodies were detected in 82 persons (18.9%). The test results performed in the particular years were presented in Table I.

Table I. The presence of antibodies to *B. henselae* in serum samples of persons with the clinical symptoms of batonellosis, tested in NIPH-NIH

Year	Number of samples tested	Positive (%) samples by serology (total)	including the classes		
			IgM number (%)	IgG number (%)	IgM + IgG number (%)
2009	189	103 (54.5)	5 (1.2)	71 (16.3)	27 (6.2)
2010	182	121 (66.5)	2 (0.5)	100 (23.0)	19 (4.4)
2011	140	97 (69.3)	3 (0.7)	75 (17.2)	19 (4.4)
2012	152	114 (75.0)	1 (0.2)	96 (22.0)	17 (3.9)
Total	663	435 (65.6)	11 (2.5)	342 (78.6)	82 (18.9)

B. quintana infections were not identified which may be associated with limited prevalence of lice, being the main vector of these bacteria.

The serological diagnostic techniques used to diagnose bartonelloses are quick, simple and provide information on *Bartonella* spp. infection, preventing from surgical interventions when the lymph nodes are involved. Indirect fluorescent antibody test (IFA), which is of high sensitiveness (88%) and specificity (94%), is the reference method used in the diagnosis of bartonelloses (10).

It should be noted that only few patients (2.5%) had only IgM antibodies to *B. henselae*. The first symptoms of infection occur relatively late, often after more than 8 weeks from infection onset while IgM antibodies may

remain in the serum for approximately 3 months. Thus, IgM antibodies may be not detectable in many patients at the time of testing (11).

In patients referred to the Unit of Rickettsiae, Chlamydiae and Spirochaetes of NIPH-NIH, confirmed as bartonellosis cases by detecting the *Bartonella* spp.-specific antibodies, the most common symptom were lymphadenopathy (86 persons, 13%), fever (13 persons, 2%) and nodular lesions in various organs (13 persons, 2%) (Tab. II).

Table II. Clinical symptoms on the referrals for testing, occurring in patients positive to *Bartonella* spp.* infection in the years 2009-2012 in NIPH-NIH

Clinical symptoms	Number (%) of patients with positive serological test results
lymphadenopathy	86 (19.8)
fever	13 (3.0)
nodule/ abscess	13 (3.0)
CSD	9 (2.1)
hepatomegaly / splenomegaly	3 (0.7)
optic neuropathy	4 (0.9)
neurological disorders	2 (0.5)
changes in bones and joints	6 (1.4)
abdominal pain	6 (1.4)
lack of symptoms on the referral for testing	293 (67.4)

* all tested persons had only antibodies to *B. henselae*

The obtained data is convergent with the results presented by Podsiadły et al. in 2002. But it should be noted that the Unit of Rickettsiae of NIPH-NIH was the only centre performing these tests in the country in this time (8). Nowadays, the diagnosis of cat scratch disease is performed in many laboratories. Thus, the serological test results were compared only to the serum samples sent to NIPH-NIH. In relation to the years 1998-2001, much more samples are sent nowadays for diagnosis. It suggests that the physicians awareness regarding the possibility of bartonellosis occurrence and its course has raised.

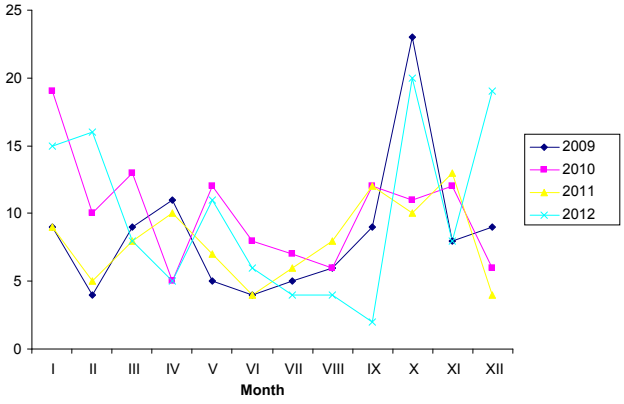

Fig. 1. Number of tested serum samples confirmed as positive to *B. henselae*, including the division on months in the years 2009-2012

The majority of samples to be tested for the presence of *Bartonella* spp. were sent in the autumn (Fig. 1.). The number of serum samples positive to *B. henselae* in September-February and March-August ranged from 60 to 35 and from 38 to 22, respectively. It is indicative of the seasonal distribution of bartonellosis, resulting from the highest activity of lice observed in autumn and the fact that the animals spend more time indoors in this time, constituting the source of infection to humans (12).

According to the obtained data, *B. henselae* infection affects mainly the patients at the age of 0-14 years as they eagerly spend time with pets and thus have close contact with them (Fig. 2).

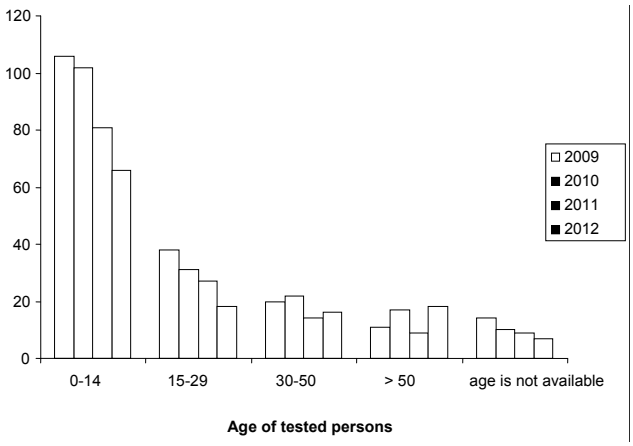

Fig. 2. Serum samples test results confirming the *B. henselae* infection in the years 2009-2012 with the division on age groups of persons tested

The distribution of detected bartonelloses in Poland is differentiated. Similarly to the previous years, the highest number of infections was notified in mazowieckie voivodeship - 173 (40%). The lowest number of infections (the lowest number of samples to be tested) was reported in the following voivodeships: śląskie – 3 (0.7%), lubuskie – 3 (0.7%) and opolskie – 4 (0.9%) (Tab. III).

In some years, no samples were sent for diagnosis from: śląskie (2009; 2012), lubuskie (2010), lubelskie (2011), łódzkie (2011) and opolskie (2012) voivodeships (Tab. III).

CONSLUSIONS

Apart from tularemia, toxoplasmosis, infectious mononucleosis, the bartonellosis should be also taken into account in the differential diagnosis of lymphadenopathy.

Table III. *B. henselae* infections identified in the particular voivodeships in the years 2009-2012

	2009	2010	2011	2012
Voivodeship	Number of positive persons by serology	Number of positive persons by serology	Number of positive persons by serology	Number of positive persons by serology
Dolnośląskie	4	10	16	15
Kujawsko-Pomorskie	4	6	4	2
Lubelskie	10	5	0	1
Lubuskie	0	0	2	1
Łódzkie	1	4	0	1
Małopolskie	4	4	1	1
Mazowieckie	33	59	34	47
Opolskie	1	2	1	0
Podkarpackie	8	3	3	6
Podlaskie	4	4	5	10
Pomorskie	4	8	6	11
Śląskie	0	2	1	0
Świętokrzyskie	9	4	6	3
Warmińsko-Mazurskie	11	7	4	2
Wielkopolskie	7	3	7	9
Zachodnio-Pomorskie	3	0	7	5
Total:	103	121	97	114

Bartonella spp. infections should be monitored in Poland to obtain the data on the frequency and distribution of disease in the country and their clinical course.

REFERENCES

1. Spinella A, Lumetti F, Sandri G, et al. Beyond cat scratch disease: a case report of bartonella infection mimicking vasculitic disorder. Case Rep Infect Dis 2012;2012:354625.
2. Boulouis HJ, Chang CC, Henn JB, et al. Factors associated with the rapid emergence of zoonotic Bartonella infections. Vet Res 2005;36:383-410.
3. Welc-Faleciak R. Current state of the knowledge of Bartonella infections. Przegl Epidemiol 2009;63:11-17.
4. Ehrenborg C, Byström R, Hjelm E, et al. High Bartonella spp. seroprevalence in a Swedish homeless population but no evidence of trench fever. Scand J Infect Dis 2008;40:208-215.
5. Vayssier-Taussat M, Le Rhun D, Bonnet S, et al. Insights in Bartonella host specificity. Ann N Y Acad Sci 2009;1166:127-132.
6. Koehler JE, Tappero JW. Bacillary angiomatosis and bacillary peliosis in patients infected with human immunodeficiency virus. Clin Infect Dis 1993;17:612-624.
7. Kaiser PO, Riess T, O'Rourke F, et al. Bartonella spp.: throwing light on uncommon human infections. Int J Med Microbiol 2011;301:7-15.
8. Podsiadły E, Sokołowska E, Tylewska-Wierzbanowska S. Występowanie zakażeń Bartonella henselae i Bartonella quintana w Polsce w latach 1998-2001. Przegl Epidem 2002; 56:399-407.
9. Pons I, Sanfeliu I, Nogueras MM, et al. Seroprevalence of Bartonella spp. infection in HIV patients in Catalonia, Spain. BMC Infect Dis 2008;8:58.
10. Karpathios T, Golphinos C, Psychou P, et al. Cat scratch disease in Greece. Arch Dis Child 1998;78:64–66.
11. Hermans M, Vermeulen MJ, Van de Kassteele J, et al. The use of *Bartonella henselae* – specific age dependent IgG and IgM in diagnostic models to discriminate diseased from non-diseased in Cat Scratch Disease serology. J Microbiol Methods 2007;71:107-113.
12. Sanguinetti-Morelli D, Angelakis E, Richet H, et al. Seasonality of Cat-Scratch Disease, France, 1999–2009. Emerg Infect Dis 2011;17:705–707.

Received: 20.03.2013
Accepted for publication: 23.09.2013

Address for correspondence:
Beata Fiecek
Unit of Rickettsiae, Chlamydiae & Spirochaetes
NIPH-NIH
Chocimska 24 Street, 00-791 Warsaw
tel. (22) 54 21 261
e-mail: bfiecek@pzh.gov.pl