

SEROEPIDEMIOLOGICAL STUDY OF LYME BORRELIOSIS AMONG FORESTRY WORKERS IN SOUTHERN POLAND

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Abstract: Forestry workers are a professional group particularly exposed to tick-borne infections; however, continuous monitoring of anti-*Borrelia burgdorferi* antibodies in this group enables faster diagnosis and more effective treatment for borreliosis. A group of 1,155 forestry workers from six forest inspectorates in southern Poland were examined with the immunoenzymatic method (ELISA test). The general level of anti-*Borrelia burgdorferi* antibodies was 12.8% in IgM class, and 25% in IgG class. Markedly more seropositive results were found in the group of fieldworkers (13.8% – IgM and 25.0% – IgG) than in office workers (10.0% – IgM and 13.7% – IgG). The highest proportions of infections both in IgM and IgG class (17.4% and 34.8%, respectively) were recorded in the group of persons over 50 years of age, the lowest – in the group of workers younger than 30 (IgM – 13.0%, IgG – 14.1%). Significant differences in the level of seropositive results in IgG class were related to the workers' gender – in women the percentage was 9.8%, in men – 28.1%. It was found that in the studied region of southern Poland, considered to be non-endemic, borreliosis occurs as a health risk to forestry workers.

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INTRODUCTION

Lyme borreliosis (LB) is one of the most common tick-borne disease in humans, both in North America [34] and in Europe [4, 18]. Borreliosis occurs also in Asia [16, 24, 26], Australia [7, 39], South America [40] and North Africa [5, 19]. Because of its high prevalence in various parts of the world, borreliosis is a serious public health problem, presenting a challenge to contemporary medicine. The etiological factor of LB is a spirochete *Borrelia burgdorferi* sensu lato (s.l.), which comprises 3 genospecies occurring in Europe, particularly pathogenic for humans: *Borrelia burgdorferi* sensu stricto, *Borrelia afzelii* and *Borrelia garinii* [36, 38]. In the United States, there is one predominant genospecies pathogenic for humans: *Borrelia*

burgdorferi sensu stricto [25]. These bacteria are transmitted to the host organism with the saliva of *Ixodes* ticks during feeding. *Ixodes ricinus* ticks (Linnaeus, 1758), as the main vector of the bacteria of *Borrelia* genus, usually live in mixed and deciduous forests [14, 15, 33]. A recent study, however, conducted in the vicinity of Bonn, Germany, suggests an expansion of *Ixodes ricinus* range towards cities and a year-round activity of ticks in some micro-habitats situated close to urban agglomerations [23]. Thus, the number of people directly exposed to tick attacks increases significantly, including city dwellers using recreation areas: parks, green squares, allotments. Nevertheless, the groups at the highest risk are forestry workers and farmers, as well as hunters, tourists, and forest undergrowth pickers staying in areas of the highest tick activity [9, 28]. A high



risk of borreliosis in forestry workers calls for continuous monitoring of professional groups particularly exposed to tick bites.

The aim of the study was to determine the level of anti-*Borrelia burgdorferi* antibodies in forestry workers occupationally exposed to contact with ticks in the south of Poland, considered as a non-endemic area for borreliosis. Another aim was to identify among the forest inspectorates those with the highest risk of borreliosis infection. The authors attempted to correlate the examined level of antibodies with the kind of work and job tenure, as well as with the gender of the examined forestry workers.

MATERIAL AND METHODS

Examined patients. A group of 1,155 foresters from 6 forestry inspectorates situated in southern Poland: Gidle, Herby, Kłobuck, Koniecpol, Przedbórz, Złoty Potok, were examined. Blood samples for coagulation were taken from the elbow vein of each worker in the autumn of 4 subsequent years, 2003–2006. The group of examined workers comprised 193 women and 962 men. According to the character of their occupation, 2 groups were distinguished: the first included fieldworkers (864 persons) and the second – office workers (291 persons). The frequency of anti-*Borrelia burgdorferi* sensu lato antibodies was assessed in patients in 4 age groups: below 30 years of age, from 30–40 and from 40–50, and over 50.

Serological tests. Altogether, 4,620 examinations were performed using ELISA test to detect the presence of IgM and IgG antibodies against *Borrelia burgdorferi* sensu lato, with commercial diagnostic kits (DRG Instruments, GMBH, Germany). The kits enable detection of infections caused by 3 pathogenic genospecies: *Borrelia garinii*, *Borrelia afzelii* and *Borrelia burgdorferi* sensu stricto. The level of serum antibodies lower than 9 DU (DRG Units = DU) was considered a negative result, results between 9–11 DU were qualified as the “grey zone”, and levels higher than 11 DU constituted a positive result.

Statistical analysis. To assess the infection risk for workers employed in individual forest inspectorates, the results were analyzed according to gender, age and occupation, with the use of χ^2 test (Pearson test).

RESULTS AND DISCUSSION

Borreliosis is a multisystemic disease, with skin, arthral and neurological symptoms whose specific character is due to particular genospecies constituting the complex *Borrelia burgdorferi* sensu lato [22, 29]. Immunoenzymatic methods are commonly used in the diagnosis of human borreliosis. ELISA tests with genetic recombinants, the use of antigens of proteins specific to individual genospecies inducing specific antibodies, increase sensitivity and specificity of the method [8].

Anti-*Borrelia burgdorferi* antibodies in IgM and IgG classes were examined in the sera of forestry workers from 6 forestry inspectorates in southern Poland in the years 2003–2006. The percentage of positive and uncertain results for IgM class ranged from 7.5%–25.0%, while for IgG class – from 16.3%–29.1%. IgM-class antibodies were found in 148 examined forestry workers (12.8%), while IgG-class antibodies (25.0%) were identified in twice as many workers (289). Most positive and uncertain results were detected in the workers of the Złoty Potok inspectorate; 65% of its area is covered in mixed and deciduous forests, believed to be the most favourable habitat for ticks [6, 17, 21, 30, 37]. Antibodies in IgM class were confirmed in 39 examined workers (17.4%), while in 17 workers (7.6%) the results were uncertain. In IgG-class, positive results reached the level of 14.3% (32 persons), and uncertain results – the level of 8.0% (18 persons). The least seropositive and uncertain results in IgM class were recorded in the Kłobuck inspectorate (7.5%), and for IgG antibodies – in the Przedbórz inspectorate (16.3%) (Tab. 1).

The study results point to a high risk of borreliosis in this professional group in the examined area considered as non-endemic for the disease. The detected percentage of positive results (12.8% for IgM and 25.0% for IgG) is

Table 1. Presence of antibodies against *Borrelia burgdorferi* in IgM and IgG classes in forestry workers in studied inspectorates.

Forest inspectorate	Anti- <i>Borrelia burgdorferi</i> IgM antibodies				Anti- <i>Borrelia burgdorferi</i> IgG antibodies				Total
	IgM+	IgM+/-	IgM ^a	IgM-	IgG+	IgG+/-	IgG ^b	IgG-	
Gidle	13 (6.0%)	4 (1.9%)	17 (7.9%)	199 (92.1%)	46 (21.3%)	12 (5.5%)	58 (26.8%)	158 (73.2%)	216 (100%)
Herby	10 (4.9%)	14 (6.9%)	24 (11.8%)	179 (88.2%)	45 (22.2%)	14 (6.9%)	59 (29.1%)	144 (70.9%)	203 (100%)
Kłobuck	2 (1.25%)	10 (6.25%)	12 (7.5%)	148 (92.5%)	33 (20.6%)	7 (4.4%)	40 (25.0%)	120 (75.0%)	160 (100%)
Koniecpol	13 (6.2%)	11 (5.2%)	24 (11.4%)	187 (88.6%)	50 (23.7%)	9 (4.3%)	59 (28.0%)	152 (72.0%)	211 (100%)
Przedbórz	9 (6.4%)	6 (4.2%)	15 (10.6%)	126 (89.4%)	20 (14.2%)	3 (2.1%)	23 (16.3%)	118 (83.7%)	141 (100%)
Złoty Potok	39 (17.4%)	17 (7.6%)	56 (25.0%)	168 (75.0%)	32 (14.3%)	18 (8.0%)	50 (22.3%)	174 (77.7%)	224 (100%)
Total	86 (7.4%)	62 (5.4%)	148 (12.8%)	1,007 (87.2%)	226 (19.6%)	63 (5.4%)	289 (25.0%)	866 (75.0%)	1,155 (100%)

IgM^a (sum of positive results IgM+ and grey zone results IgM+/-); IgG^b (sum of positive results IgG+ and grey zone results IgG+/-)



Table 2. Presence of antibodies against *Borrelia burgdorferi* in IgM and IgG classes in forestry workers according to character of work.

Year of examination	Office work					Field work					Total
	Anti- <i>Borrelia burgdorferi</i> IgM* antibodies		Anti- <i>Borrelia burgdorferi</i> IgG* antibodies		Total	Anti- <i>Borrelia burgdorferi</i> IgM* antibodies		Anti- <i>Borrelia burgdorferi</i> IgG* antibodies		Total	
	IgM ^a	IgM-	IgG ^b	IgG-		IgM ^a	IgM-	IgG ^b	IgG-		
2003	5 (11.4%)	39 (88.6%)	9 (20.5%)	35 (79.5%)	44 (100%)	25 (15.7%)	134 (84.3%)	48 (30.2%)	111 (69.8%)	159 (100%)	203 (100%)
2004	11 (13.6%)	70 (86.4%)	11 (13.6%)	70 (86.4%)	81 (100%)	23 (10.0%)	207 (90.0%)	43 (18.7%)	187 (81.3%)	230 (100%)	311 (100%)
2005	6 (7.2%)	77 (92.8%)	7 (8.4%)	76 (91.6%)	83 (100%)	29 (12.7%)	200 (87.3%)	61 (26.6%)	168 (73.4%)	229 (100%)	312 (100%)
2006	7 (8.4%)	76 (91.6%)	13 (15.7%)	70 (84.3%)	83 (100%)	42 (17.1%)	204 (82.9%)	64 (26.0%)	182 (74.0%)	246 (100%)	329 (100%)
Total	29 (10.0%)	262 (90.0%)	40 (13.7%)	251 (86.3%)	291 (100%)	119 (13.8%)	745 (86.2%)	216 (25.0%)	648 (75.0%)	864 (100%)	1,155 (100%)

^a total: IgM+ and IgM+/-; ^b total: IgG+ and IgG+/-; IgM* ($\chi^2=7.23$; $p=0.007$) in 2006; IgG* ($\chi^2=12.35$; $p=0.0004$) in 2005

comparable with results obtained in other regions of Poland. In south-eastern Poland, the determined percentage of specific anti-*Borrelia burgdorferi* antibodies in forestry workers' blood serum was ca. 40% [10, 41], in Lower Silesia – 35.0% [12], and in West Pomerania – from 35%–61.9% [27]. In farmers from the areas considered as endemic for borreliosis, the proportion of positive results was 33% [11]. In other European countries, the percentage of forestry workers and other employees particularly exposed to tick bites and showing antibodies against *B. burgdorferi* antigens varies: e.g., in Germany, it is 30% [31], in Slovakia – 12.8% [3], and in Italy – 7.5% [32]. In France, the antibodies were found in 14.1% of examined persons occupationally exposed to contact with ticks: the highest proportions of positive results were found in logging workers (17.5%) [35]. In Turkey, the presence of anti-*Borrelia burgdorferi* antibodies was confirmed in 10.0% of forestry workers and farmers [20].

Among the 1,155 persons examined in the present study, there were 291 office workers (25.2%) and 864 fieldworkers

(74.8%). Most anti-*Borrelia burgdorferi* antibodies were detected in the sera of the field workers. In IgM class, the percentage of positive results in both groups differed only slightly (10.0% of office workers and 13.8% of fieldworkers). However, the level of IgG-class antibodies found in fieldworkers (25.0%) was twice as high as that in office employees (13.7%). The study confirmed a statistically significant correlation between the seropositive results in 2005 in IgG class ($\chi^2 = 12.35$, $p = 0.0004$) and in 2006 – in IgM class ($\chi^2 = 7.23$, $p = 0.007$) according to the character of the examined persons' work (Tab. 2).

A greater number of seropositive results in fieldworkers confirms the particular exposure to tick attacks of those employees whose work requires their presence in forests – habitats of those arthropods. Despite this fact, as studies conducted in endemic areas of south-eastern Poland show, over 21% of persons exposed to contact with ticks use no prophylactics to reduce the risk of tick bites [2]. A greater risk of infection with *Borrelia burgdorferi* in persons more frequently exposed to ticks is also confirmed by other

Table 3. Presence of antibodies against *Borrelia burgdorferi* in IgM and IgG classes according to age of studied forestry workers.

Age interval	Anti- <i>Borrelia burgdorferi</i> IgM antibodies				Anti- <i>Borrelia burgdorferi</i> IgG* antibodies				Total
	IgM+	IgM+/-	IgM ^a	IgM-	IgG+	IgG+/-	IgG ^b	IgG-	
< 30 years	6 (7.1%)	5 (5.9%)	11 (13.0%)	74 (87.0%)	10 (11.8%)	2 (2.3%)	12 (14.1%)	73 (85.9%)	85 (100%)
30–40 years	32 (9.9%)	18 (5.5%)	50 (15.4%)	274 (84.6%)	45 (13.9%)	17 (5.2%)	62 (19.1%)	262 (80.9%)	324 (100%)
40–50 years	31 (7.6%)	19 (4.7%)	50 (12.3%)	357 (87.7%)	79 (19.4%)	18 (4.4%)	97 (23.8%)	310 (76.2%)	407 (100%)
> 50 years	37 (10.9%)	22 (6.5%)	59 (17.4%)	280 (82.6%)	92 (27.1%)	26 (7.7%)	118 (34.8%)	221 (65.2%)	339 (100%)
Total	106 (9.2%)	64 (5.5%)	170 (14.7%)	985 (85.3%)	226 (19.6%)	63 (5.4%)	289 (25.0%)	866 (75.0%)	1,155 (100%)

IgM^a (total: IgM+ and IgM+/-); IgG^b (total: IgG+ and IgG+/-); IgG* ($\chi^2=13.4935$; $p=0.0358$) in 2005



Tabela 4. Presence of antibodies against *Borrelia burgdorferi* in IgM and IgG classes according to gender of studied forestry workers.

Year of examination	Women					Men				Total	
	Anti- <i>Borrelia burgdorferi</i> IgM antibodies		Anti- <i>Borrelia burgdorferi</i> IgG* antibodies		Total	Anti- <i>Borrelia burgdorferi</i> IgM antibodies		Anti- <i>Borrelia burgdorferi</i> IgG* antibodies			Total
	IgM ^a	IgM-	IgG ^b	IgG-		IgM ^a	IgM-	IgG ^b	IgG-		
2003	4 (13.8%)	25 (86.2%)	2 (6.9%)	27 (93.1%)	29 (100%)	26 (14.9%)	148 (85.1%)	55 (31.6%)	119 (68.4%)	174 (100%)	203 (100%)
2004	7 (14.0%)	43 (86.0%)	7 (14.0%)	43 (86.0%)	50 (100%)	27 (10.3%)	234 (89.7%)	80 (30.7%)	181 (69.3%)	261 (100%)	311 (100%)
2005	7 (13.0%)	47 (87.0%)	4 (7.4%)	50 (92.6%)	54 (100%)	28 (10.9%)	230 (89.1%)	64 (24.8%)	194 (75.2%)	258 (100%)	312 (100%)
2006	8 (13.3%)	52 (86.7%)	6 (10.0%)	54 (90.0%)	60 (100%)	41 (15.2%)	228 (84.8%)	71 (26.4%)	198 (73.6%)	269 (100%)	329 (100%)
Total	26 (13.5%)	167 (86.5%)	19 (9.8%)	174 (90.2%)	193 (100%)	122 (12.7%)	840 (87.3%)	270 (28.1%)	692 (71.9%)	962 (100%)	1,155 (100%)

^a total: IgM+ and IgM+/-; ^b total: IgG+ and IgG+/-; IgG * ($\chi^2=7.7274$, $p=0.025$) in 2003–2006

studies in Poland [13]. In Slovenia, 23.8% of seropositive results in IgG class were determined in fieldworkers, while in the control group consisting of office workers the proportion of positive results was 9.7% [31].

The examined forestry workers were divided into 4 age groups: below 30 years of age (7.4% of examined population), between 30–40 (28.0%), between 40–50 (35.2%), and a group of workers older 50 (29.4%). The highest proportions of positive results were found in the forestry workers over 50 years of age, both in IgM (17.4%) and in IgG (34.8%) class. The lowest level of antibodies was detected in the group of persons less than 30 years old in IgG class (14.1%). Also in IgM class, their level of antibodies was one of the lowest and reached only 13.0%. A statistically significant correlation between the frequency of IgG antibodies and age of a worker was found for tests performed in 2005 ($\chi^2 = 13.4935$, $p = 0.0358$) (Tab. 3).

A distinct increase in the level of anti-*Borrelia burgdorferi* antibodies observed in forestry workers with an increasing time in the profession was also confirmed by the studies conducted in Lower Silesia, where during the period 2003–2005, the level of infections had grown by 20% [12].

A greater opportunity for contact with ticks due to job tenure determines also the seroprevalence of gender in examined forestry workers. A markedly greater percentage of positive results in IgG class was found in men (28.1%) than in women (9.8%). For IgM-class antibodies, similar proportions were recorded in both groups (13.5% in women and 12.7% in men). The study confirmed a statistically significant correlation between the frequency of antibodies in IgG class and the gender of workers over the whole observation time (2003–2006) ($\chi^2 = 7.7274$, $p = 0.025$) (Tab. 4).

The majority of positive results in men is related to the long period of employment in forestry, where 83.3% of 1,155 of the examined were men, mostly fieldworkers.

Women, 16.7% of the studied group, work mainly in the administration of the examined forestry inspectorates. It seems, therefore, that it is not the gender of a worker that significantly affects the percentage of seropositive results, but the character of his/her work (field or office). This conclusion is confirmed by research conducted, among others, in Spain [1].

The results obtained in the present study confirm the possibility of infection with spirochetes of *Borrelia burgdorferi* sensu lato in workers of forestry inspectorates in southern Poland. It is therefore necessary to undertake manifold activities to reduce the effects of tick parasitising; this may be achieved, among others, by propagating the principles of prophylactics against the risk of a tick bite, as well as taking appropriate measures concerning diagnosis and treatment of tick-borne diseases. The survey conducted among forestry workers shows that the relevant knowledge is still very limited [2]. A continuous monitoring of infections with *Borrelia burgdorferi* spirochetes, both in forestry workers and in other persons exposed to contact with ticks, as well as of ticks themselves, may improve the effectiveness of diagnosis and treatment of borreliosis cases.

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