SHORT REPORT

Seasonality of *Yersinia enterocolitica* bioserotype 1B/O:8 infections in Poland

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SUMMARY

Both serological and bacteriological investigations revealed a cyclic, seasonal pattern of *Yersinia enterocolitica* 1B/O8 infections in Poland during the years 2008–2011. A large increase in incidence was observed in the second quarter and a decrease in the third quarter of each year. Such seasonal changes were not seen in the case of infections caused by the other enteropathogenic *Yersinia* bioserotypes.

Key words: Bacterial infections, infectious disease epidemiology, Yersinia enterocolitica.

Yersinia enterocolitica appears to be a common cause of enteritis in humans. The pathogen can be found in the natural environment, water, animals and food. Infection is most often acquired by eating raw or undercooked pork. Enterocolitis, abdominal pain and fever are the most common clinical manifestations, but symptoms such as polyarthritis or septicaemia may occur also [1].

Six biotypes (1A, 1B, 2, 3, 4, 5) and more than 70 serotypes have been described for *Y. enterocolitica*. This species encompasses two evolutionary lineages: American and European strains, considered separate subspecies: *Y. enterocolitica* ssp. *enterocolitica* and *Y. enterocolitica* ssp. *palearctica*, respectively. The highly pathogenic *Y. enterocolitica* bioserotype 1B/O8 is geographically limited to Northern America although it has also emerged in Japan and Europe [2–6]. According to recent epidemiological reports the most common pathogenic bioserotypes isolated from humans in Europe were 4/O:3 and 2/O:9 [7–10].

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In previously published studies on the prevalence of yersiniosis in Poland we reported a gradual increase of *Y. enterocolitica* bioserotype 1B/O:8 infections from 2004 to May 2008 [6]. The very high peak of incidences occurred during the first few months of 2008, when the number of positive *Y. enterocolitica* O:8 cases was much higher than for *Y. enterocolitica* serotypes O:3 and O:9. In this context we aim to show the prevalence of *Y. enterocolitica* bioserotype 1B/O:8 infections in Poland in the 4-year period from 2008 to 2011, with special emphasis on the seasonal distribution.

All data presented in this work were collected in the Bacteriology Department of the National Institute of Public Health – National Institute of Hygiene (NIPH-NIH) in Warsaw from 2008 to the end of 2011. *Y. enterocolitica* isolates were voluntarily sent from epidemiological and sanitary stations and hospital laboratories in Poland to NIPH-NIH for confirmation and biotyping and serotyping. Serum samples were received for routine serological diagnostics from healthcare practitioners throughout the country.

Bacteriological and serological investigations were conducted in different groups of patients. Only a few patients with bacteriologically confirmed yersiniosis were also tested by serological investigation. ELISAs were performed on polystyrene microtitre plates (Nunc, MaxiSorp, Denmark) coated with lipopolysaccharides (LPS) (25 µg/ml) and Yop proteins $(10 \,\mu\text{g/ml})$ in $0.05 \,\text{M}$ carbonate buffer according to a previous study [6]. LPS were extracted by trichloroacetic acid from Y. enterocolitica serogroups O:3, O:8, O:9 and Y. pseudotuberculosis I reference strains 105P, WA-314P, 96P and Y.psI, respectively (kindly provided by Dr Rakin, Max-von-Pettenkofer Institut, Munich, Germany) [11]. The bacteria strain Y. enterocolitica serogroup O:8 (WA-314) containing the 42 MDa plasmid pYV was used for production of Yop proteins according to Heesemann et al. [12]. The sensitivity and specificity of the ELISA with these antigens were as described in detail in a previous study [13]. Biotyping of Y. enterocolitica and Y. pseudotuberculosis was performed by classical biochemical tube test assay according to the EN ISO 10273 Standard and Tsubokura & Aleksic, respectively [14, 15]. All the strains were serotyped using a latex agglutination test [16] and examined for the CRMOX+ phenotype as described by Riley & Toma [17].

From the beginning of 2008 to the end of 2011, routine diagnostic serological tests for *Y. entero-colitica* and *Y. pseudotuberculosis* infection were performed on 9733 patients throughout the country who were suspected of yersiniosis during clinical investigation (2161 in 2008; 2508 in 2009; 2724 in 2010 and 2340 in 2011). There were no statistically significant differences in the number of tested sera per month. These patients were suffering mainly from abdominal pain with and without diarrhoea, reactive arthritis or erythema nodosum. The clinical manifestations of yersiniosis caused by *Y. enterocolitica* O:8 were restricted to the gastrointestinal tract in some of the cases with severe symptoms that can mimic appendicitis.

During the period under investigation, a significant diagnostic level of antibodies to Y. enterocolitical serotype O:8 was diagnosed in 194 (2.0%) patients, to Y. enterocolitical serotype O:3 in 329 (3.4%), to Y. enterocolitical serotype O:9 in 51 (0.5%) and to Y. pseudotuberculosis I in 72 (0.7%) patients. Quarterly analysis of the cases collected during these 4 years did not demonstrate a distinct seasonal differences in incidence of infections caused by Y. enterocolitical serogroups O:3, O:9 and Y. pseudotuberculosis I. By contrast, a large increase of serologically confirmed

Y. enterocolitica serogroup O:8 infections in Poland has been repeatedly observed during the second quarters of 2008, 2009, 2010 and 2011. This high prevalence of Y. enterocolitica O:8 infections ended quickly during the third quarter of each year.

The serological results presented in this study are in line with results of bacteriological investigations. A total number of 187 Y. enterocolitica bioserotype 1B/O:8, 306 Y. enterocolitica bioserotype 4/O:3, 12 Y. enterocolitica bioserotype 2/O:9 and four Y. pseudotuberculosis I strains were re-examined in our laboratory from 2008 to 2011. Twenty-seven (23.7%), 65 (49.6%), 40 (31.7%) and 55 (41.0%) Y. enterocolitica bioserotype 1B/O:8 strains were re-identified in 2008, 2009, 2010 and 2011, respectively. We did not observe any essential changes in the seasonal frequency of isolation of Y. enterocolitica bioserotypes 4/O:3, 2/O:9 and Y. pseudotuberculosis I strains from clinical samples in Poland. Similarly to the aforementioned results of the serological investigation, a large increase of Y. enterocolitica 1B/O:8 isolations was observed during the second quarter of each year. A large decrease in the number of isolations was noted during the third quarters of 2008, 2009, 2010 and 2011. The number of confirmed isolates and serum samples seropositive for Y. enterocolitica serotype O:8 and Y. enterocolitica serotype O:3 in Poland from 2008 to 2011 are shown in Figures 1 and 2, respectively.

According to the latest official epidemiological data, there were 253 and 326 yersiniosis cases reported in Poland in 2008 and 2009, respectively [18, 19]. The incidence rate was 0.66 and $0.85/100\,000$ inhabitants, respectively. It should be noted that the seasonal distribution of reported yersiniosis in Poland revealed the highest frequency during the second quarter of both years. Interestingly, this increase of reported yersiniosis cases in Poland corresponded to the reported increase of Y. enterocolitica serotype O:8 infections in the present study.

The seasonal prevalence of yersiniosis may depend on different factors, e.g. climate, extent of pork consumption or extent of pig slaughter [20]. In contrast to *Salmonella* or *Campylobacter* infections, which typically peak during the summer months, a higher frequency of yersiniosis was usually noted in some countries during the cooler periods of the year [21]. However, the latest epidemiological report from Germany showed, that during 2001–2008 the seasonal distribution of reported *Y. enterocolitica* infections (caused by serotype O:3 in 90 % of cases) was relatively uniform, with only a slight increase in June, July

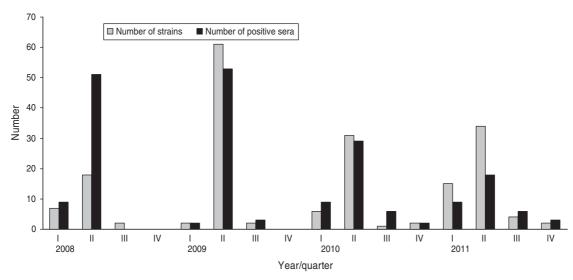


Fig. 1. Number of confirmed isolates and serum samples seropositive for *Yersinia enterocolitica* serotype O:8 in Poland, 2008–2011.

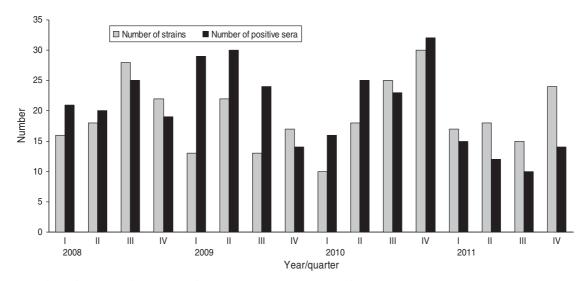


Fig. 2. Number of confirmed isolates and serum samples seropositive for *Yersinia enterocolitica* serotype O:3 in Poland, 2008–2011.

and September. Moreover, the lowest number of infections was reported in March and April [7].

So far, the reasons for the observed cyclic, seasonal pattern of *Y. enterocolitica* 1B/O8 infections in Poland are unknown. It is worth mentioning that we did not observe such changes during 2004–2007 when the incidence of these infections was much lower than in the period 2008–2011. Factors accounting for such variations may centre on the primary source of the infections. It is well known that swine are a recognized reservoir for human pathogenic *Y. enterocolitica* in Europe, where serotype O:3 has been isolated from the tongue, throat, tonsils, caecal contents, and faeces

of swine. Reports from Japan [2, 3] established rodents as an important source of *Y. enterocolitica* bioserotype 1B/O:8. However, data on the occurrence of this pathogen in both swine and rodents in Poland is lacking. Thus, the reservoir of *Y. enterocolitica* 1B/O8 in Poland has not yet been identified.

In a previous study, we showed that clinical isolates of *Y. enterocolitica* 1B/O8 collected in Poland were clonal and belonged to the same strain *sensu stricto* [22]. This strong clonality of Polish *Y. enterocolitica* 1B/O:8 isolates may suggest a common origin. However, the strains were isolated from patients in different regions of the country. Therefore, further

increased surveillance supported by local veterinary authorities is required to elucidate the reservoir of the pathogen in Poland and its route of transmission. This action appears to be important since the spread of highly pathogenic *Y. enterocolitica* 1B/O8 in Europe may be a serious public health threat.

DECLARATION OF INTEREST

None.

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