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## Epidemiological study of *Yersinia pseudotuberculosis* in Belgium by multiple-locus variable-number tandem-repeat analysis

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**Background:** *Yersinia pseudotuberculosis* (YP) is an enteropathogenic bacterium which causes abdominal symptoms such as mesenteric adenitis and enteritis. Animals (wild mammals and birds, porks, bovines and pets) constitute the natural reservoir for this bacterial species which is transmitted to humans via consumption of contaminated food or water. YP presents 15 serotypes and several subserotypes, among which O:1 and O:2 are the most frequent serotypes isolated from humans. In Belgium, clinical laboratories send all YP strains found in clinical practice to the Belgian National Reference Center (NRC) for *Yersinia*. During the 2015 winter season, the NRC observed a 3,7-fold increase of YP cases (22 cases) as compared to the winters 2013 (6), 2014 (6) and 2016 (10).

**Material/methods:** In order to determine if that increase was due to a clonal spread, we performed Multiple-Locus Variable-Number Tandem-Repeat Analysis (MLVA) on the 44 YP isolates collected between 2012 and 2016 (39 serotype O:1, 3 serotype O:2, 1 serotype O:3 and 1 undetermined serotype). We used 7 sets of primers, as described before (Halkilahti et al., 2013). The different MLVA

profiles were named by the numbers of repetition of the 6 to 9 bp long tandem repeat of each locus. We considered as identical strains MLVA profiles presenting  $\leq 2$  loci variants. Grouping of strains was performed using BioNumerics (Applied Maths, Belgium).

**Results:** Among the 44 isolates, 37 different MLVA profiles were observed. A strictly identical profile was observed for two pairs of strains and a group of 6. The 5 strains which were not of serotype O:1 had unique profiles. During the 2015 winter peak, we found that the proportion of clustered cases exceeded the number of non-clustered cases, contrarily to other winter seasons. Overall 20/22 YP were from the O:1 serotype, of which 14/20 cases belonged to 3 different MLVA profiles (Table 1). The MLVA profile 4-7-3-5-6-5-2 which was reported 6 times during the 2014-2015 season is similar to a strain responsible of some food and human outbreaks reported in Finland since 2008 (Halkilahti et al., 2013).

**Conclusions:** In Belgium, during the 2015 winter season, we observed a 3,7-fold increase of YP cases. We found that 14/20 YP O:1 serotype (70%) were clustered using MLVA typing. Interestingly, a major cluster was similar to one previously reported in Finland. This situation illustrates the major need for integrated surveillance systems between veterinary medicine, food supply and human medicine at an international level. MLVA demonstrates here its usefulness for typing of YP isolates.

**Table 1:** Number of strains per winter season in Belgium and their MLVA profiles

	Profile	2013	2014	2015	2016	Total
Cluster 1	4-7-3-5-6-5-2	1		6	2	9
Cluster 2	5-8-3-2-3-5-10		2	6	1	9
Cluster 3	6-11-3-2-3-5-22			2		2
Cluster 4	7-9-3-2-4-7-12	1			1	2
Non-clustered		4	4	8	6	22