

Genotyping of Human and Porcine *Yersinia enterocolitica*, *Yersinia intermedia*, and *Yersinia bercovieri* Strains from Switzerland by Amplified Fragment Length Polymorphism Analysis

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In this study, 231 strains of *Yersinia enterocolitica*, 25 strains of *Y. intermedia*, and 10 strains of *Y. bercovieri* from human and porcine sources (including reference strains) were analyzed using amplified fragment length polymorphism (AFLP), a whole-genome fingerprinting method for subtyping bacterial isolates. AFLP typing distinguished the different *Yersinia* species examined. Representatives of *Y. enterocolitica* biotypes 1A, 1B, 2, 3, and 4 belonged to biotype-related AFLP clusters and were clearly distinguished from each other. *Y. enterocolitica* biotypes 2, 3, and 4 appeared to be more closely related to each other (83% similarity) than to biotypes 1A (11%) and 1B (47%). Biotype 1A strains exhibited the greatest genetic heterogeneity of the biotypes studied. The biotype 1A genotypes were distributed among four major clusters, each containing strains from both human and porcine sources, confirming the zoonotic potential of this organism. The AFLP technique is a valuable genotypic method for identification and typing of *Y. enterocolitica* and other *Yersinia* spp.

Yersinia enterocolitica, a member of the family *Enterobacteriaceae*, is a well-known food-borne pathogen. *Y. enterocolitica* is isolated more often from children with diarrhea than from adults (11, 25). In children, infection with *Y. enterocolitica* can cause gastroenteritis with mild diarrhea, abdominal pain, low fever, and pseudoappendicitis. In adults, *Y. enterocolitica* can cause pharyngitis and various systemic infections and is frequently associated with autoimmune complications. In Germany, the incidence of *Y. enterocolitica* was reported to be 8.0 per 100,000 inhabitants in 2003 (26). In the same study, it was found that *Yersinia* spp. were the third most frequently isolated pathogens from patients with enteric diseases after *Salmonella* spp. and *Campylobacter* spp. An initial infection with *Yersinia* spp. is more often followed by postinfection reactive arthritis than an infection with *Salmonella* spp. or *Campylobacter* spp. is (16).

Y. enterocolitica can be differentiated into six biotypes (biotypes 1A, 1B, 2, 3, 4, and 5) and several serotypes. The most common bioserotypes associated with human disease are 1B/O:8, 2/O:5,27, 2/O:9, 3/O:3, and 4/O:3 (8). In Europe biotype 1B is only sporadically detected in France and Italy. The virulence spectra of the biotypes differ; bioserotype 1B/O:8 is considered highly pathogenic, while the pathogenicity of biotypes 2 to 4, including all commonly isolated serotypes, is lower, as shown in animal models (5). *Y. enterocolitica* biotype 1A lacks the *Yersinia* virulence plasmid pYV and therefore is considered avirulent (24). However, some evidence suggests

that biotype 1A strains have some pathogenicity, as strains belonging to this biotype are frequently isolated from humans suffering from diarrhea. Furthermore, *Y. enterocolitica* biotype 1A strains carry chromosomally encoded virulence factors and are able to invade cultured epithelial cells by a mechanism different from the mechanism used by pYV-bearing strains (31). Furthermore, a recent study showed that with regard to virulence genes, biotype 1A strains isolated from clinical samples did not differ significantly from strains isolated from other sources (32). Moreover, since *Y. enterocolitica* is difficult to differentiate from other *Yersinia* species by routine phenotypic tests (3, 4), strains identified as *Y. enterocolitica* biotype 1A and biotype 3 may belong to related species, such as *Y. intermedia* or *Y. bercovieri* (this study). As these *Yersinia* species lack classical virulence factors, they are considered nonpathogenic. These isolates originated from either clinical samples or environmental and food samples (2, 12, 29). In *Y. bercovieri* an enterotoxin different from that in *Y. enterocolitica* has subsequently been found (30).

Pork meat is one of the potential sources of infection of humans by *Y. enterocolitica*. In Switzerland the prevalence of *Y. enterocolitica* in pork meat at the retail level was reported to be 15.4% (20). However, only 0.7% of the strains isolated belonged to the potentially pathogenic biotypes 2, 3, and 4, while the majority of the strains belonged to apparently apathogenic biotype 1A (20). At the farm level, the prevalence of *Y. enterocolitica* was 63%, and 36.8% of the strains were biotype 1A strains in 2001 (20). Thus, the importance of pork meat as a source of *Y. enterocolitica* infection in Switzerland is unclear, given the assumptions concerning the virulence spectra of the different biotypes.

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