Prevalence and genetic characterization of *Campylobacter* spp. isolates from swine in Quebec, Canada.

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**Summary**: In order to assess the importance of swine in the transmission of *Campylobacter*, it is relevant to evaluate its prevalence and distribution in the animal population and to further characterize this microorganism. A total of 850 animals were screened at slaughterhouse for the presence of *Campylobacter*. Pulse-field gel electrophoresis (PFGE) was performed by digestion of DNA with *KpnI* and *SmaI*. A total of 660 (78% of samples) *Campylobacter* isolates were recovered from swine feces. *C. coli* was the most frequently observed species (91%) while no *C. jejuni* was recovered. PFGE gave up to 72 genetic patterns with digestion by *KpnI* for 94 strains tested. Up to five distinct isolates were characterized from the same animal fecal sample. These isolates were digested with both *KpnI* and *SmaI*. Four animals over 10 (40%) had two or more genetically distinct macrorestriction profiles. A large genetic diversity was thus observed among *Campylobacter* of swine origin.

**Keywords**: Food-borne pathogen, swine, slaughterhouse, genetic variations, PFGE

**Introduction**: Pigs are frequently colonized by *Campylobacter* spp. and their contribution to the human infection is currently not fully understood (Wassenaar, 1997). In order to assess the importance of swine in the transmission of bacteria such as *Campylobacter* spp. it is relevant to evaluate the prevalence and distribution of various genotypes within the animal population. To acquire a better knowledge on the population genetic diversity, it is also necessary to further characterize the bacteria. Among the available techniques, pulse-field gel electrophoresis (PFGE) is one of the most discriminatory technique for *Campylobacter*. The aims of this study was to evaluate the prevalence and distribution of *Campylobacter* spp. in swine and swine herds in the province of Quebec, Canada, and to genetically and phenotypically characterize the strains recovered from healthy swine within a limited geographical area.

**Material and Methods**: A total of 850 pigs were randomly selected over a six months period in a slaughterhouse in Quebec, Canada. After the evisceration
process and inspection, 1 g of caecal content was aseptically collected. Samples were inoculated directly onto charcoal-based selective medium with supplements and incubated at 37°C under microaerophilic atmosphere for 48 h. Typical colonies were tested based on the classical biochemical identification for *Campylobacter* spp. PFGE was performed as described previously with some modifications (Gibson et al., 1994) for 101 strains isolated. For lysis, plugs were placed in 2 ml of a solution of lysis buffer with 0.5 mg/ml of lysozyme overnight at 37°C before digestion with proteinase K (Harrington et al., 1999). A pre-migration was performed by submitting plugs to 60 V for 45 min (Whatling & Thomas, 1993). Plugs were digested with 20 U of *KpnI* or *SmaI* restriction endonuclease as recommended by the manufacturer (On et al., 1998) for 18 hours at 37°C. PFGE was performed at 200 V and 14°C with a pulse time of 5 s for 9 h, 20 s for 9 h and 5 s for 2 h for a total time of 20 h in a Gene Navigator® apparatus. DNA bands were visualized and then analyzed using the Image Master® software version 3.01.

**Results:** A total of 660 (78% ± 2.81%) specimens of swine caecal content out of 850 yielded *Campylobacter* spp. Of those, 600 (91%) were identified as *C. coli* while no *C. jejuni* or *C. lari* were detected. The remaining isolates were variable in their biochemical profiles or lost after freezing process. Following guidelines of Tenover et al. (1995) for small sets of isolates, a criteria of homology of 85% was selected to establish relatedness between strains. A total of 72 different profiles were observed. A large genetic diversity and distribution according to the strains profiles by farms over the 51 farms sampled was observed. The same farm can exhibit as many different profiles as the number of strains tested. PFGE was used to analyze the genotype of five different colonies by animal. Among the 10 animals sampled, a unique pattern of digestion of the various strains was observed in 4 with *KpnI*. Two animals harboured 2 closely related strains using *KpnI* while they were found indistinguishable when digested with *SmaI*. Two animals beard strains with 2 distinct patterns with the two enzymes and finally, two animals had more than 2 genetically distinct strains.

**Discussion:** The prevalence of 78% for *Campylobacter* spp. observed in swine fecal material in this study is in accordance with previous studies (Jacobs-Reitsma, 2000). The analysis of swine DNA profiles obtained with *KpnI* restriction endonuclease digestion revealed a large variety of profiles. It has been suggested that such variation may results from large-scale genomic recombination in genetically related *C. jejuni* or *C. coli* strains (Duim et al., 1999). A similar study conducted in Ontario in different abattoirs showed up to 86 subgroups when PFGE was performed with *SmaI* and 78 subgroups when genotyped with *SalI* for 141 *Campylobacter* isolates (Steele et al., 1998). Within the same farm, the animals sampled showed different genetic profiles. Most of the time, each animal from a given farm possessed its own profile. It can thus be suggested that there is a
potential for contamination by a large variety of strains at the abattoir (Steele et al., 1998). The genetic variability in *Campylobacter* can explain why PFGE patterns were so variable in the present study. On the other hand, this wide genetic diversity would likely make it more difficult to establish genetic link between strains from different species.

References: