

Campylobacter concisus pathotypes isolated from diarrheal stool samples

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Objectives:

Campylobacter concisus is an emerging pathogen of genus *Campylobacter*. Its pathogenic potential in intestinal disease is still unclear, as this is a highly genetically diverse species. *C. concisus* strains can be divided into two intestinal pathotypes which are genetically different to commensal strains. Based on the presence of putative virulence determinants *C. concisus* can be divided into strains that are adherent and invasive (AICC) which poses gene for exotoxin 9 and strains that are adherent and toxigenic (AToCC) and have the ability to produce a zonula occludens toxin (Zot). In 2015 a more complex cultivation method of stool samples was introduced in Goriška region; consequently we have found significant increase in the number of *C. concisus* isolates (from 27 isolates in 2014 to 121 in 2015). The aim of our study was to determine which *C. concisus* isolates were a potential source of diarrhoea based on their virulence potential.

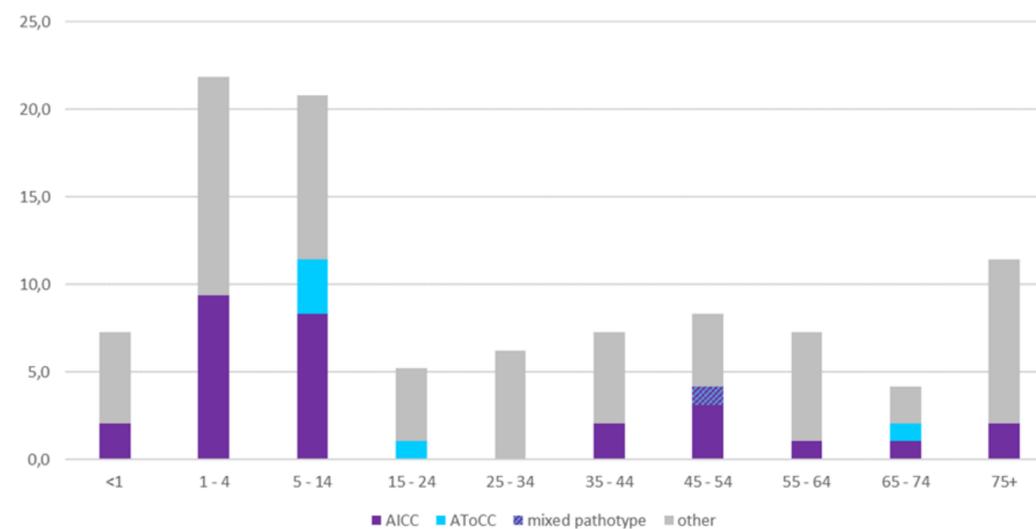
Methods:

In 2015 modified Cape Town membrane filtration technique with cultivation on a non-selective agar was included in diagnostic testing of stool samples. Colonies suspected to be non-thermophilic *Campylobacter* spp. were identified using MALDI-TOF (Bruker Daltonics, Bremen, Germany). DNA extraction was performed using LightCycler Advanced Lysis Kit (Roche Diagnostics, Mannheim, Germany). All *C. concisus* isolates were tested for the presence of putative virulence determinants exotoxin 9 and Zot using PCR as described previously (Kaakoush J Med Microbiol 2014, Mahendran PLoS One 2013). Isolates were divided into two intestinal pathotypes - AICC and AToCC.

Results:

C. concisus was isolated from 121 patients. Almost half of the isolates were obtained from children younger than 15 years (59/121; 48.8%). The median age of patients was 17 (average 28.4 years, age range 0 to 94 years); 63 out of 121 (52.1%) were female. Age distribution is presented in Figure 1). On average 10 cases per month were found with an increase in the last quarter. No seasonal variation have been observed. There was an almost constant three monthly prevalence of *C. concisus*, whereas *C. jejuni* has a typical seasonal variation with the peak in June-August. We have observed a peak in February-March and autumn months. In total 96 *C. concisus* isolates were available for molecular analysis; 29.2% (28/96) belonged to AICC, 5.2% (5/96) to AToCC, and 1% (1/96) to mixed pathotype, 64.6% (62/96) were negative for both virulence determinants. Distribution of pathotypes among age groups is presented in Figure 1, there were no statistically significant differences between age groups. On average one third (35%, range 25-60%) of *C. concisus* isolates per month belonged to intestinal pathotypes.

Figure 1: Distribution of *C. concisus* pathotypes among patients age groups



Conclusion:

Complex cultivation protocol yielded significantly more *C. concisus* isolates, however only one third of the isolates belonged to intestinal pathotypes, the remainder of isolates were probably commensal oral strains. In order to establish whether *C. concisus* is the probable cause of gastroenteritis the presence of putative virulence determinants (exotoxin 9 and ZOT toxin) must be determined.