SNP Types of *Campylobacter jejuni* Isolated from Different Hosts =

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Summary

In this study we applied Single Nucleotide Polymorphism (SNP) typing to 577 Campylobacter jejuni isolates from different host species, including chickens, feedlot cattle, dairy cattle, dogs, cats and humans. We found that SNP typing was an effective method for genotyping C. jejuni. The results show that some Campylobacter genotypes do show host specificity, which should be considered when investigating outbreaks of campylobacteriosis in humans. The SNP typing results clearly show that there are sources other than poultry meat associated with human campylobacteriosis.

Introduction

Campylobacter is the major cause of human gastrointestinal illness in Australia, with rates of infection approximately double those for Salmonella spp. each year. While poultry are a significant source of these infections, there is a considerable body of evidence that there are other sources, for example raw milk and pets (1, 2). Some types of *C. jejuni/coli* appear host specific (meaning that some types occur only in chickens) while other types can be found in multiple hosts (e.g. in both chickens and humans). There is a general agreement that the definitive method for typing C. jejuni/coli is Multilocus Sequence Typing (MLST) (3-5). The high cost of MLST, caused by the requirement to sequence around 500 base pairs in each of seven genes, has recently been overcome by the use of a combination of kinetic PCR and interrogative data analysis that provides the power of conventional MLST but at a much lower cost and with a more rapid response time. This new technology is called Single Nucleotide Polymorphism (SNP) analysis.

The objective of this study was to apply the technique of SNP analysis for typing *C. jejuni* to both poultry and non-poultry isolates. To achieve this we extended the current Agri-Science Queensland *Campylobacter* collection to include poultry isolates from other research groups and industry and we obtained non-poultry isolates from sources such as cattle, pets and humans. We were then able to compare and contrast the SNP types of the poultry *C. jejuni* with the non-poultry types.

Materials and Methods

In our study SNP typing as previously outlined (6) was applied to 577 *C. jejuni* isolates. The isolates consisted of:

- 32 Chicken faecal/caecal isolates from epidemiological studies (1999-2003)
- 36 Chicken Factory isolates (2008)
- 76 Chicken Factory isolates (2005-2006)
- 77 Chicken caecal isolates collected from a national survey (2003-2004)
- 93 Dairy Cattle isolates (2006-2008)
- 123 Feedlot Cattle isolates (2006-2008)
- 39 Dog and eight Cat isolates (2006-2008)
- 46 Human isolates (2000)
- 47 Human isolates (2008).

Results

In our study we applied SNP typing to 577 *C. jejuni* isolates from different host species. The 577 isolates were grouped into 39 different SNP types. Our results show that some genotypes are associated with multiple host species whereas other genotypes are predominantly associated with limited host

species. As an example, SNP type 44 was a genotype found only in humans, dogs and cats. SNP typing has also shown subtle differences in genotype distribution. SNP type 5 was associated with dairy cattle while SNP type 13 was associated with feedlot cattle.

Discussion

SNP typing is an effective method for genotyping C. jejuni, including the capacity to recognise associations. host Some Campylobacter genotypes do show host specificity, which should be considered when investigating outbreaks of campylobacteriosis in humans. The genotyping results clearly show that there are sources other than poultry meat associated with human campylobacteriosis. In particular, pets need to be considered as a source of C. jejuni for humans. Some preliminary data from our studies indicates that regional and company influences may play a role in the genotype distribution of Campylobacter isolates in poultry and we are currently investigating this further.

Overall, SNP typing has been shown to be a convenient first line tool for screening *C. jejuni* isolates. It is user friendly, easily transportable between research groups, is relatively cheap and has the advantage that it is directly linked to MLST. Unlike PFGE, SNP typing lends itself to robotics for sample preparation and assay set up. We would recommend SNP typing as a front line typing method when investigating outbreaks of campylobacteriosis or when looking for host associations with particular genotypes of *C. jejuni*.

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