Comparison of four different selective media for the quantification of Campylobacter in poultry meat and rapid confirmation of suspect colonies

De Zutter Lieven¹, Van Damme Inge¹, Mugg Philip² and Karen Ukaegbu Karen²

¹ Department of Veterinary Public Health and Food Safety, Faculty of Veterinary Medicine, Ghent University, Salisburylaan 133, 9820 Merelbeke, Belgium;

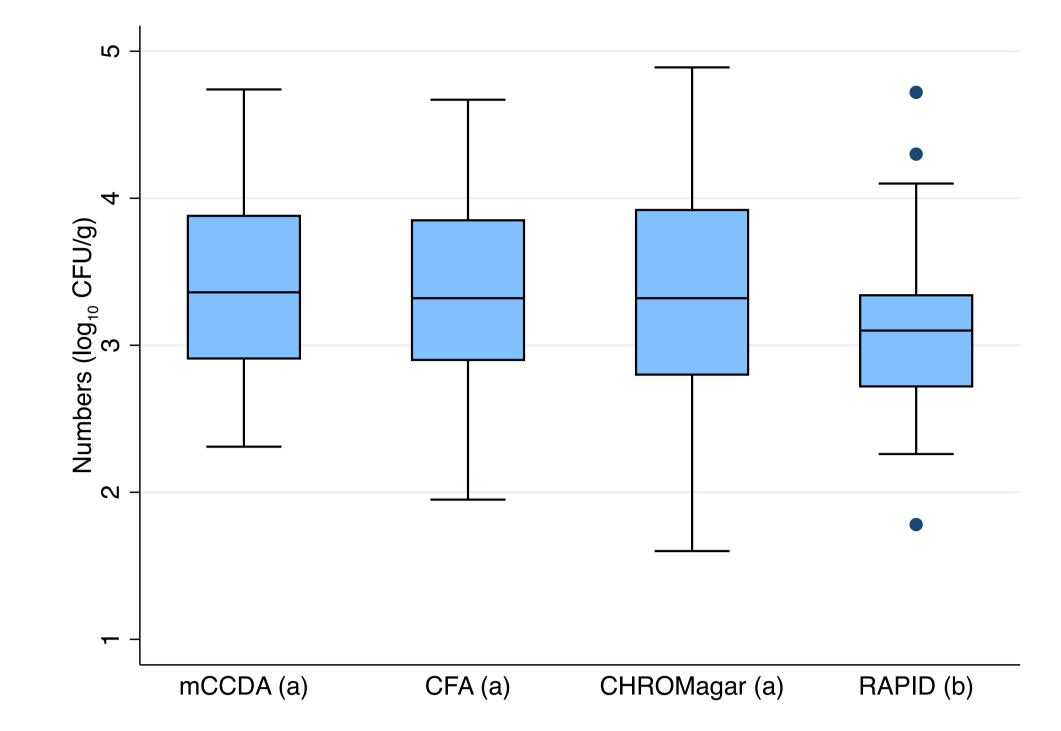
² Microgen Bioproducts Ltd, 1 Admiralty Way, Camberley, Surrey, UK

INTRODUCTION

Quantification of *Campylobacter* in food products, especially in those products known to be highly contaminated (such as fresh poultry meat), is becoming common. Over the last years, different new selective media for the enumeration of *Campylobacter* have been developed.

The present study aimed to evaluate the performance of four commercially available selective media for the quantification of *Campylobacter* in broiler meat

Figure 1: Box plots of Campylobacter counts in naturally contaminated samples obtained by the 4 different selective media. Average counts on media with a different letter are significantly different (p < 0.05).



and a latex test kit for the identification of thermophilic Campylobacter.

MATERIAL & METHODS

Naturally contaminated samples

- 47 samples with skin were purchased in a broiler slaughterhouse and connected cutting plant.
- At the laboratory, from each sample 10g was collected for analysis.

Artificial inoculation of broiler skin samples

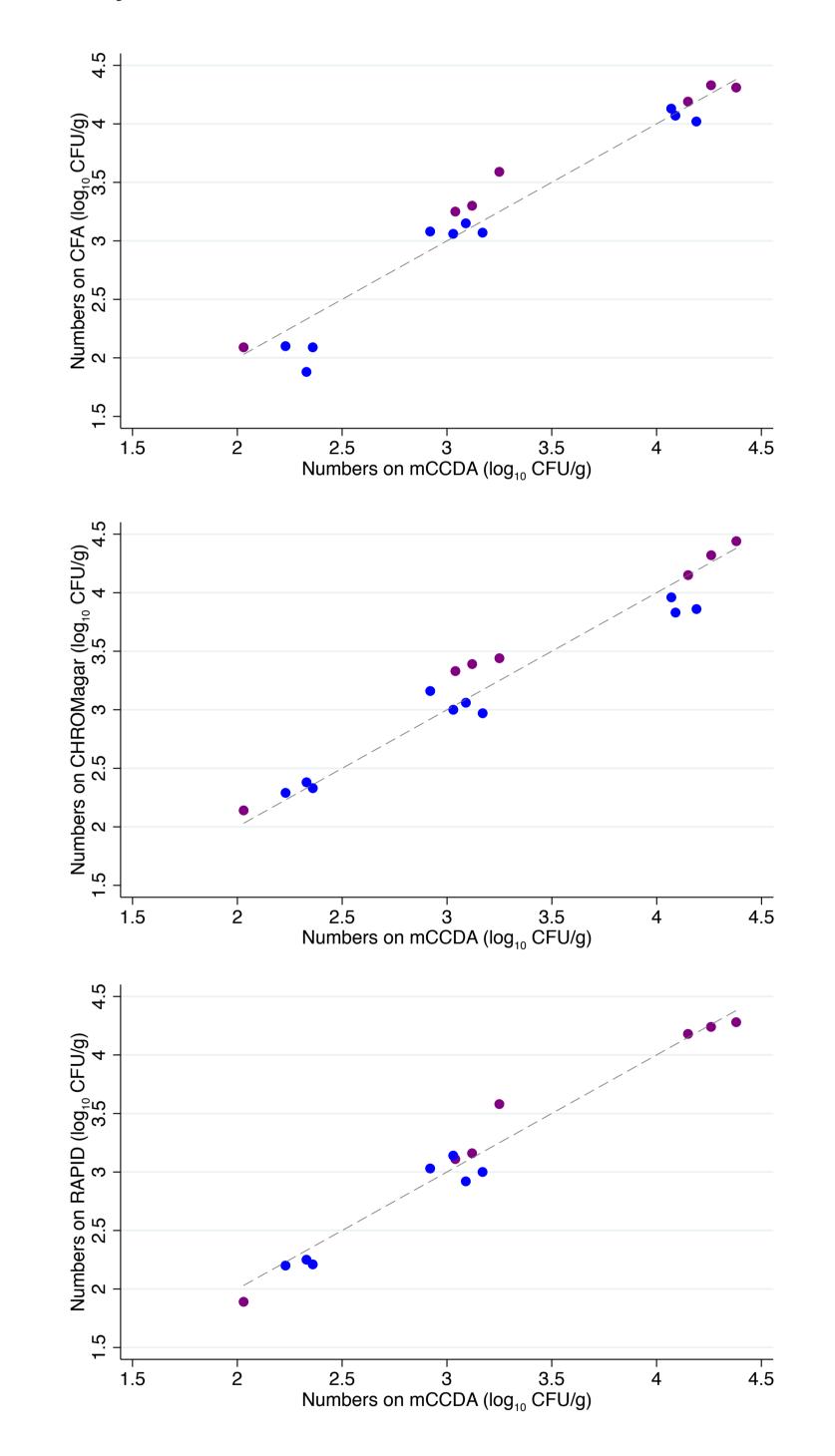
- 10 samples were artificially inoculated with a
- *C. coli* isolate originating from a broiler skin sample and
- C. lari isolate LMG 8846.
- Broiler carcass parts containing skin (stored at -20°C) were thawed overnight.
- 20 samples of 5g skin were collected and inoculated at three levels $(10^2, 10^3)$ and 10⁴/g).

Campylobacter were quantified in all samples using the following selective media:

- 1/ Modified Charcoal Cefoperazone Deoxycholate Agar (mCCDA) (Oxoid, UK)
- 2/ CHROMagar[™] Campylobacter (CHROM) (CHROMagar, France)
- 3/ RAPID'Campylobacter Agar (RAPID) (Bio-Rad, France)
- 4/ Campy Food Agar (CFA) (bioMérieux, France).

b/ Artificially contaminated broiler skin samples

Figure 2: Correlation between Campylobacter counts on mCCDA and CHROMagar, CFA and RAPID. Samples that are artificially contaminated with C. coli and C. lari are indicated in purple and blue, respectively.



After incubation under microaerobic conditions at 41.5°C during 48h, colonies with typical and non-typical *Campylobacter* morphology on each agar medium were counted. From each countable plate, 3 colonies (if present) were picked for species identification of *C jejuni* and *C. coli* using PCR.

The 2 isolates used for inoculation of skin samples, 10 C. jejuni and 29 C. coli isolates present in the laboratory collection were agglutinated by Microgen Campylobacter Latex after growth on CHROM.

Counts (log₁₀-transformed) obtained with the different media from the naturally contaminated skin samples were compared using a random-effects generalized least squares regression analysis with the samples as group variable. Bonferroni corrections were applied from multiple testing. A significance level of 5% was used.

RESULTS

a/ Naturally contaminated broiler skin samples

From 12 of the 47 tested samples, the results could not be used to evaluate the performance of the different Campylobacter media, due to the absence of Campylobacter colonies on one or more selective plates or the spreading of colonies over the agar so that accurate counting was not possible. Moreover, for 20 samples, mCCDA plates showed growth of colonies with a non-typical Campylobacter morphology, while this was only the case for 1, 1 and 4 samples using CHROM, RAPID and CFA medium respectively. Speciation of colonies picked from positive samples were confirmed by PCR as C. jejuni.

c/ Identification of isolates





All isolates tested by Microgen Campylobacter Latex resulted in a clear positive agglutination reaction.

CONCLUSIONS

- The use of mCCDA, CHROM and CFA medium resulted n similar counts when testing naturally contaminated skin samples, whereas the average counts are significantly lower for RAPID medium.
- Colonies with non-typical *Campylobacter* morphology were only frequently present on mCCDA
- Similar counts were obtained with the four selective agar media when testing samples artificially contaminated with a *C. coli* or *C. lari* isolate.
- Microgen Campylobacter Latex seems a reliable test to confirm colonies with a typical *Campylobacter* morphology on CHROM agar.