

# Clostridium difficile in intestinal contents and on carcasses of pigs and cattle at slaughterhouse in Belgium



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## INTRODUCTION

*Clostridium difficile* is a major cause of nosocomial acquired diarrhoea and colitis after use of antibiotics. With the recent isolation of this bacterium in healthy carrier food animals and retail meats, the possibility for foodborne transmission is a current concern.

## OBJECTIVE

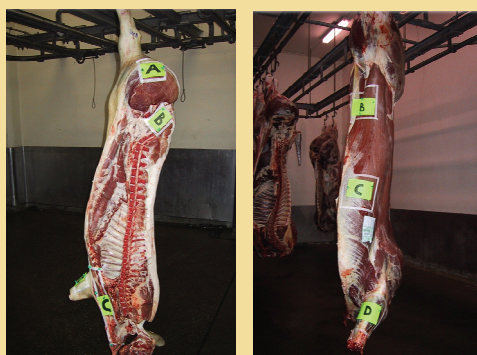
The objective of this study was to determine the presence of *Clostridium difficile* in intestinal samples and on carcasses of pigs and cattle at slaughterhouse in Belgium and identify the main PCR-ribotypes

## MATERIALS AND METHODS

### SAMPLES

Intestinal contents from pigs (100) and cattle (100) were collected from the slaughter line, directly from the last intestine in the viscera processing area.

Carcass swabs (100 from pigs and 100 from cattle) were collected post slaughter, after entry into the chilling room. Surface swabs of 1600cm<sup>2</sup> on cattle and 600cm<sup>2</sup> on pig carcasses were collected



### METHODS

Culture was carried out using an enrichment step. One gram of feces was inoculated into 9 ml of CCFBT and incubated anaerobically for 72h at 37 °C. As for the carcass samples, the four cottons used for swabbing each individual carcass were homogenized with 40 ml of CCFBT and incubated anaerobically for 72h at 37 °C. Subsequently, 10µl of the enrichment broth of each type of sample was spread onto CCFAT and incubated at 37 °C for two days.

An identification of the isolated colonies was done by PCR detection of *tpi*, *tcdA*, *tcdB* and *cdtA* genes. Toxic activity was also confirmed by a fecal cytotoxinimmunoassay. Further characterization was performed by PCR ribotyping



## RESULTS

Sample origin	PCR-ribotype	n° strains	Cytotoxicity assay	tcdA	tcdB	cdtA
Cattle fecal samples	BR078	3	POS	POS	POS	POS
	BR029	1	POS	POS	POS	NEG
	UCL118	1	POS	POS	POS	NEG
	UCL103	1	NEG	NEG	NEG	NEG
	UCL16r	1	POS	POS	POS	NEG
	UCL273	1	NEG	NEG	NEG	NEG
	UCL270	1	NEG	NEG	NEG	NEG
Cattle Carcasses	UCL254	1	POS	POS	POS	NEG
	BR015	1	POS	POS	POS	NEG
	BR023	1	POS	POS	POS	NEG
	UCL11	1	POS	POS	POS	NEG
	UCL16u	2	POS	POS	POS	NEG
Pig fecal samples	UCL5a	2	POS	POS	POS	NEG
	BR078	1	POS	POS	POS	POS
Pig carcasses	BR014	3	POS	POS	POS	NEG
	BR081	1	POS	POS	POS	NEG
	36UCL	2	POS	POS	POS	NEG

Table 1. PCR-ribotypes and toxin gene profiles of *Clostridium difficile* isolates

*Clostridium difficile* was found in 10% and 1% of the cattle and pigs intestinal samples respectively. Concerning carcass samples, *Clostridium difficile* was recovered from 7% of the swabs from cattle and 6% for pigs. A total of 15 different PCR ribotypes were identified including PCR ribotype 078, 029, 014, 015, 023 and 081. (Refer to table.1)

## CONCLUSIONS

The results of this study confirm that *Clostridium difficile* is carried in the intestinal tracts of food animals arriving at slaughterhouse and is present on carcasses at Belgium slaughterhouse. This fact shows that there is a clear potential for contamination of meat in the slaughterhouse.

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