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Fachmann, Mette Sofie Rousing; Löfström, Charlotta; Josefsen, Mathilde Hasseldam; Hoorfar, Jeffrey

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Low cost semi-continuous quantification of *Campylobacter* by air sampling in broiler houses

Mette S.R. Søndergaard, Charlotta Löfström, Mathilde H. Josefsen, and Jeffrey Hoorfar

National Food Institute, Technical University of Denmark, Søborg
E-mail: msrso@food.dtu.dk

Aim

To evaluate a semi-continuous air sampling method coupled with quantitative real-time PCR (qPCR) for detection of *Campylobacter* contamination in European broiler houses.

Introduction

Current boot swab sampling is not accurate enough. More sensitive methods like analysis of cecal droppings or cecal sacks requires time-consuming collection of specific droppings or sacrifice of animals [1]. To identify control measures that would be universally applicable sampling was carried out in conventional broiler houses in the mid-eastern part of Poland from July to October 2012 in addition to preliminary samplings performed in Denmark.

Experiment

- Each measurement consisted of air samples on gelatine filters, conventional boot swab faecal samples and particle counts.
- Three Polish broiler flocks were sampled over a period of eight weeks and the samples subsequently sent to Denmark for analysis.
- The presence and levels of *Campylobacter* in the boot swabs and air samples were assessed using culture and qPCR.
- The particle counts were used to analyse size distribution in airborne particles (0.3-10 µm) in the broiler houses in relation to bacterial distribution.



Table 1. Test results for the three Polish broiler flocks for week zero (W0, before introduction of chickens to the house), week 1 (W1, first week of the rearing period) and for the remaining weeks (W2-W8).^a

		Air sampling (filters + qPCR)					
		W0		W1		W2-W8	
		Positive	Negative	Positive	Negative	Positive	Negative
Fecal sampling (boot swabs + qPCR)	Positive	0	0	0	0	17	0
	Negative	3	0	3	0	0	0

^a A flock was regarded positive for *Campylobacter* contamination if one of either sample types were found positive.

Materials & Methods

- Culturing: Culturing was performed in complete Bolton Broth and on mCCDA at $41.5 \pm 1^\circ\text{C}$ under microaerophilic conditions (6% O₂, 7% CO₂, 7% H₂ and 80% N₂).
- DNA extraction: DNA purified on an automated KingFisher (Thermo Labsystems).
- Standards for filters: These were prepared by spiking *Campylobacter* free filters with enumerated *Campylobacter* culture and extracting DNA.
- Standards for socks: These were prepared by spiking a solution obtained by massaging socks with *Campylobacter* free faeces and buffered peptone water, with enumerated *Campylobacter* culture and extracting DNA.
- qPCR: performed as previously described [2].

Conclusions

- *Campylobacter* detection with air sampling coupled with qPCR is a good alternative to boot swabs

Results

- Airborne *Campylobacter* was not found to correlate with a specific particle size
- *Campylobacter* contamination could be detected in air samples up to two weeks before boot swabs and in 1-2 logs higher levels

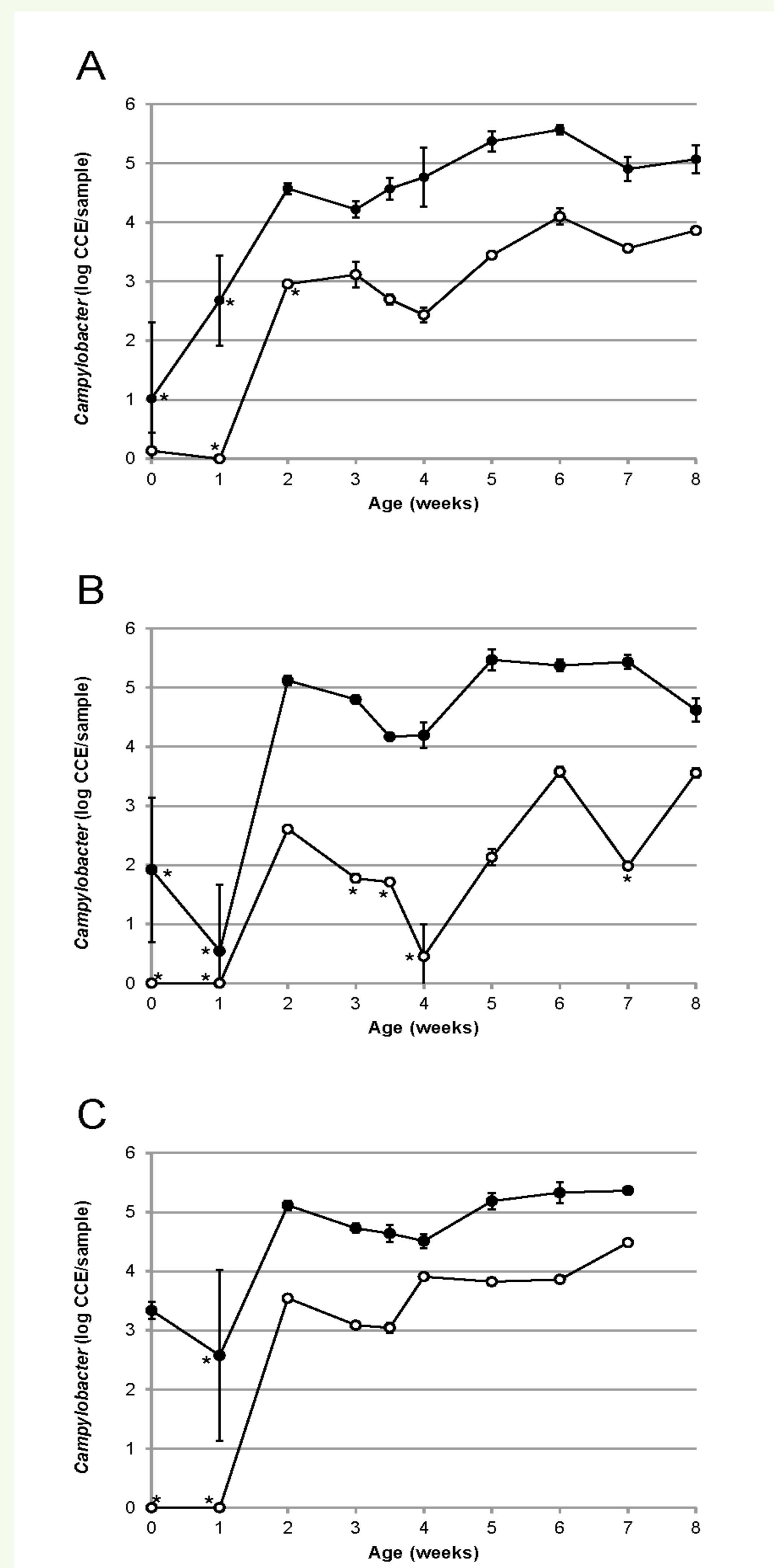


Fig 1. The log-transformed *Campylobacter* cell equivalents (log CCE) per sample (e.g. per filter (●) or boot swab (○)). The represented data is the CCE per sample for each of the tree flocks (panel A-C) for each week (0: week zero, before introduction of chickens to the house; 1-8: week one through eight, the remaining sampling period). The error bars show the standard deviation related to the qPCR analysis. The data points marked with * are samples that gave Ct values outside the quantifiable range.

References

1. EFSA Journal 2012;10(6):2764
2. Josefsen et al., 2010. Appl Environ Microbiol. 76:5097-5104

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