CamCon

Campylobacter control - novel approaches in primary poultry production

Deliverable 3.1.2. Definition of level of sensitivity
Deliverable was due and delivered Month 24 – a revised version was delivered Month 30.

General comments
The inventor of the integrated lab-on-chip (ILOC) technology platform, Ilochip A/S, went bankrupt during the preparation for the CamCon project. We managed to transfer the intellectual property rights to another Danish company, Delta A/S. However, the priorities of Delta A/S shifted towards clinical point-of-care diagnostics and further development of the ILOC technology and air sampling was therefore not made. Technical problems with the equipment, available, experienced during the project resulted in the exclusion of this technology platform from the project. Altogether, this resulted in a re-planning of the work in WP3. Hence, the feasibility of air sampling was validated and the deliverables met as reported below using the Sartorius AirPort MD8 (Sartorius Stedim Biotech GmbH) technology along with conventional boot sock sampling.

A total of four flocks of broilers were followed as planned. Yet, one of the flocks was slaughtered ahead of time due to *Salmonella* infections and another two became *Campylobacter*-positive too late during their life course to allow comprehensive analyses. It means that the data on size distribution in relation to production systems and quantities of airborne *Campylobacter* in relation to size distribution of airborne particle is insufficient for publication. The data from two flocks are reported below and are to be compiled with data achieved from ongoing studies in Poland for publication.

Definition of level of sensitivity
No standard for the quantification of *Campylobacter* (or for any other bacterium, as we are informed) in air exists, and as we have seen previously (Olsen et al., 2009) there is a large discrepancy between cell count and viable count of *Campylobacter* in air, probably due to the micro-aerophilic nature of *Campylobacter* and limited tolerance to oxygen. As the issue here is the evaluation of a method to detect the presence of *Campylobacter* in a flock or a building, level of detection should be targeting cell count or genome equivalent rather than viable count.

It was the intention to use the ILOC technology platform to define a detection limit and use this as a unit of quantification. A 10-channel ILOC sampler had previously been developed and by sampling for a series of defined time periods an ILOC unit or an $ILOC_{50}$ unit could have been defined as the limit of detection. Given a detection limit of signal generation on the microchip chamber of 1 genome equivalent this approach could have given a very precise measure of genome equivalents in air and a golden standard for the evaluation of more user-friendly air detection technologies to be developed in the future. Due to the bankruptcy of the company inventing the ILOC technology and lack of perspective in pursuing ILOC technology platform this intention had to be aborted.

Instead we have made a tentative estimate of the level of sensitivity using the Sartorius Airport MD8 air sampler.

Making the assumption that viable count is equivalent to cell count in a fresh culture it is possible to use the standard curve to make an approximation to the limit of detection using the method adopted in Task 3.1.1. From Fig. 1 an approximation of the limit of detection can be defined as 1 $\log_{10}$ cells per 1/8 section of the filter. As one filter contains the cells of 750 L of air this is equivalent to a detection limit of approximately 2 $\log_{10}$ cells per m^3 of air.

As volume of air sampled through the gelatin filter can be scaled up and down and rough assumptions were made during calculations, this level of sensitivity also is scalable – proportionate to volume of air filtered, among other parameters - and of limited value except in the comparative evaluation of the Sartorius Airport MD8 air sampler to alternative air sampling devices in the future. An estimate of the level of sensitivity of detection of *Campylobacter* in air and discussion of

relevance in relation to monitoring in air is to be published together with the method of quantification of *Campylobacter* in air (D 3.1.1).

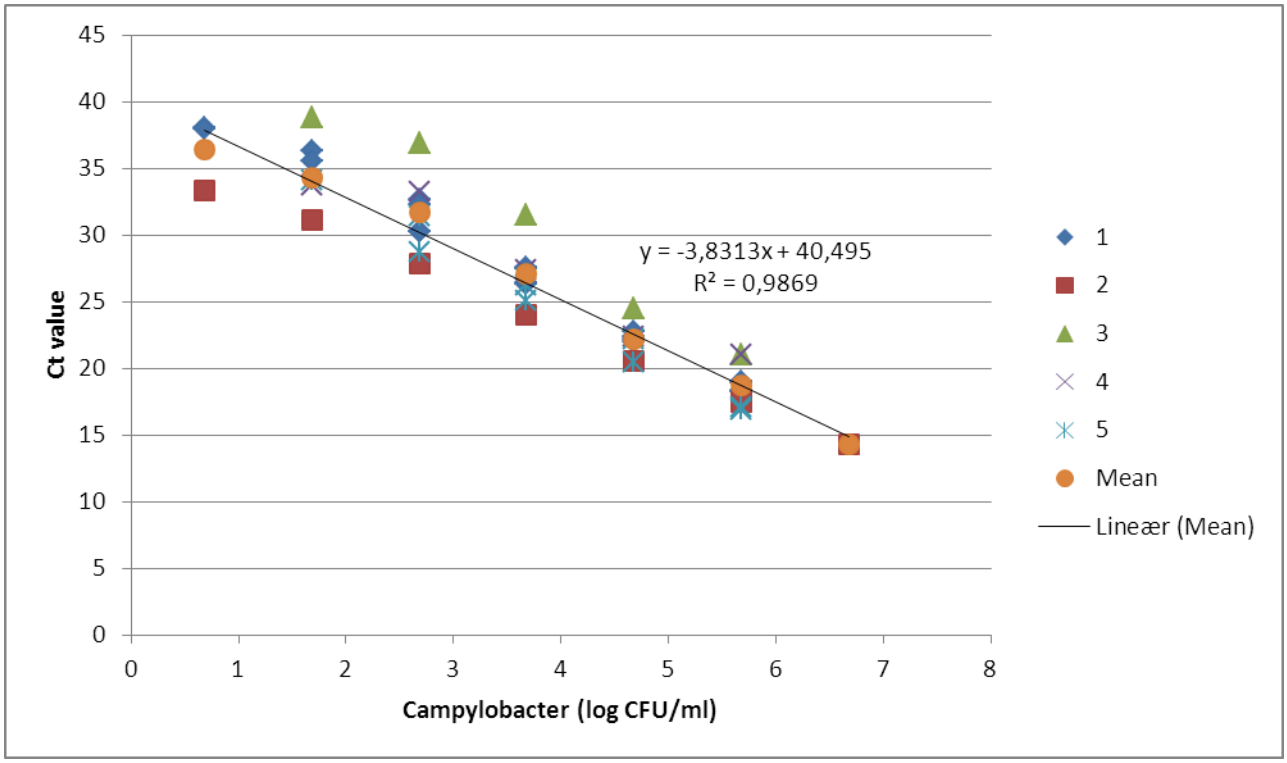

Figure 1. Standard curves produced from five series of DNA extraction from gelatin filters spiked with 100 µL *Campylobacter jejuni* CCUG 11284 (ATCC) from a 10-fold serial dilution of a standard stock suspension. A linear standard curve was drawn based on the mean values of Ct-values at each dilution.