

Campylobacter diversity on retail chicken – implications for source attribution and outbreak investigation

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EURL *Campylobacter* workshop

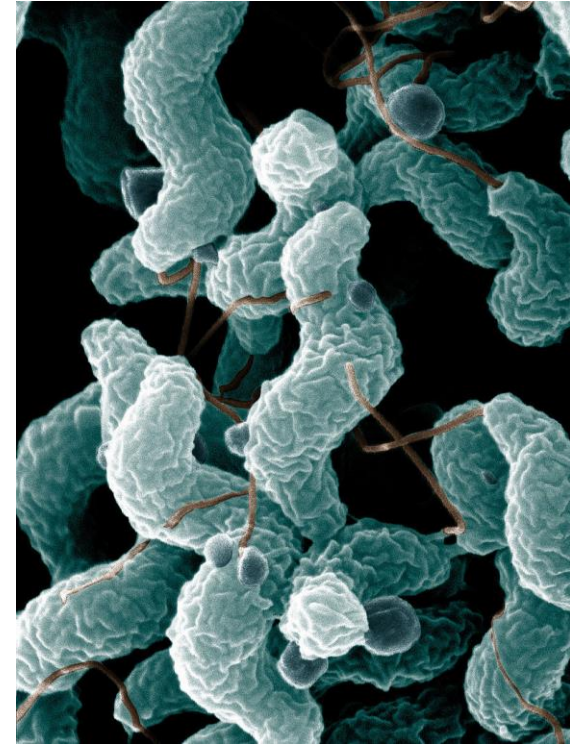
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Background

Campylobacter in poultry

- *Campylobacter*: Gram negative, spiral-shaped bacteria
 - Leading cause of gastroenteritis worldwide – over 500,000 estimated annual cases in the UK
 - Typically *C. jejuni*, but *C. coli* and other emerging species also implicated in disease
- Chicken meat: one of the main reservoirs of *Campylobacter* infection
 - Rise in diversity associated with chicken transport and meat processing
- Recovery of *Campylobacter* and identification of particular species and subtypes may be dependent on the culture method
 - Use different culture conditions
 - Investigate *Campylobacter* diversity within samples



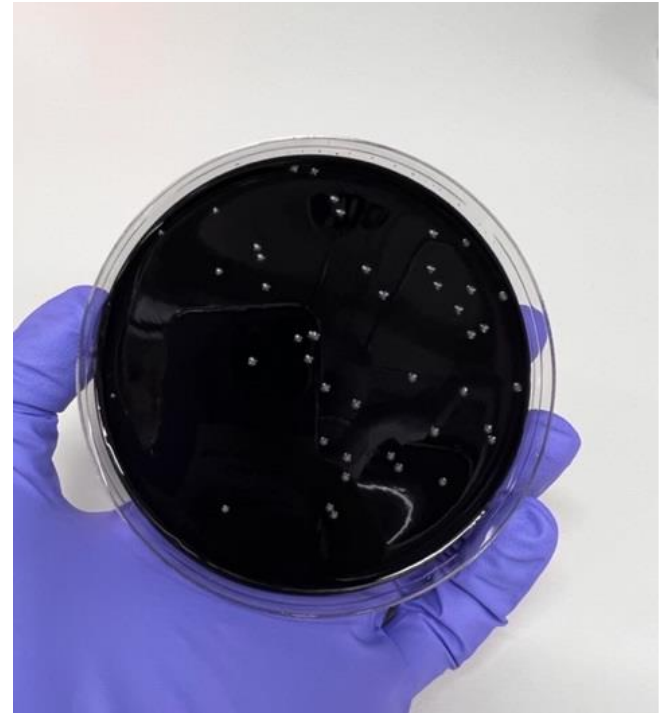
Scanning electron microscope image of
Campylobacter jejuni
De Wood, Pooley, USDA, ARS, EMU

Aims

Utilise a combination of culture method approaches to isolate *Campylobacter* from retail chicken

Determine the diversity of *Campylobacter* on culture-positive chicken samples

Illustrate the implications of the within-sample diversity on source attribution and outbreak investigation scenarios



Isolation workflow



45 chicken samples collected

100g homogenised in BPW and aliquots subjected to centrifugation

37°C

42°C

Direct plating

mCCDA

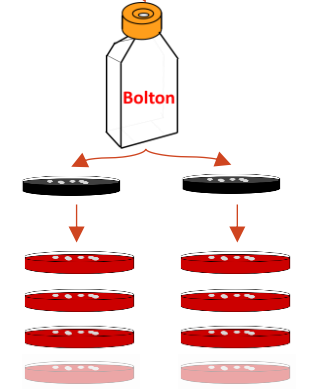
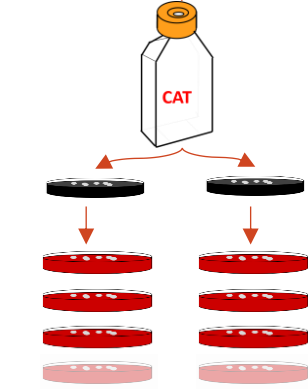
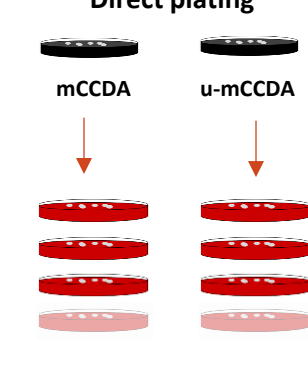
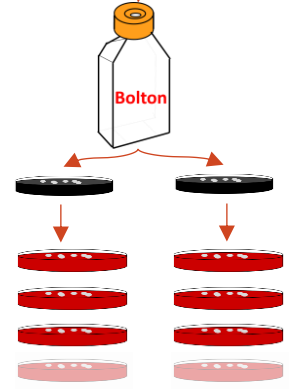
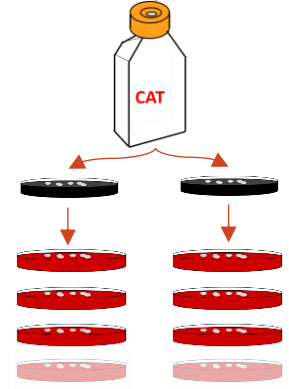
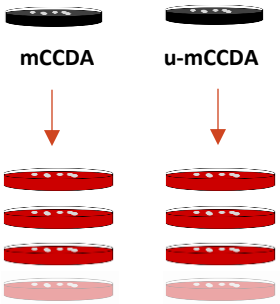
u-mCCDA



Direct plating

mCCDA

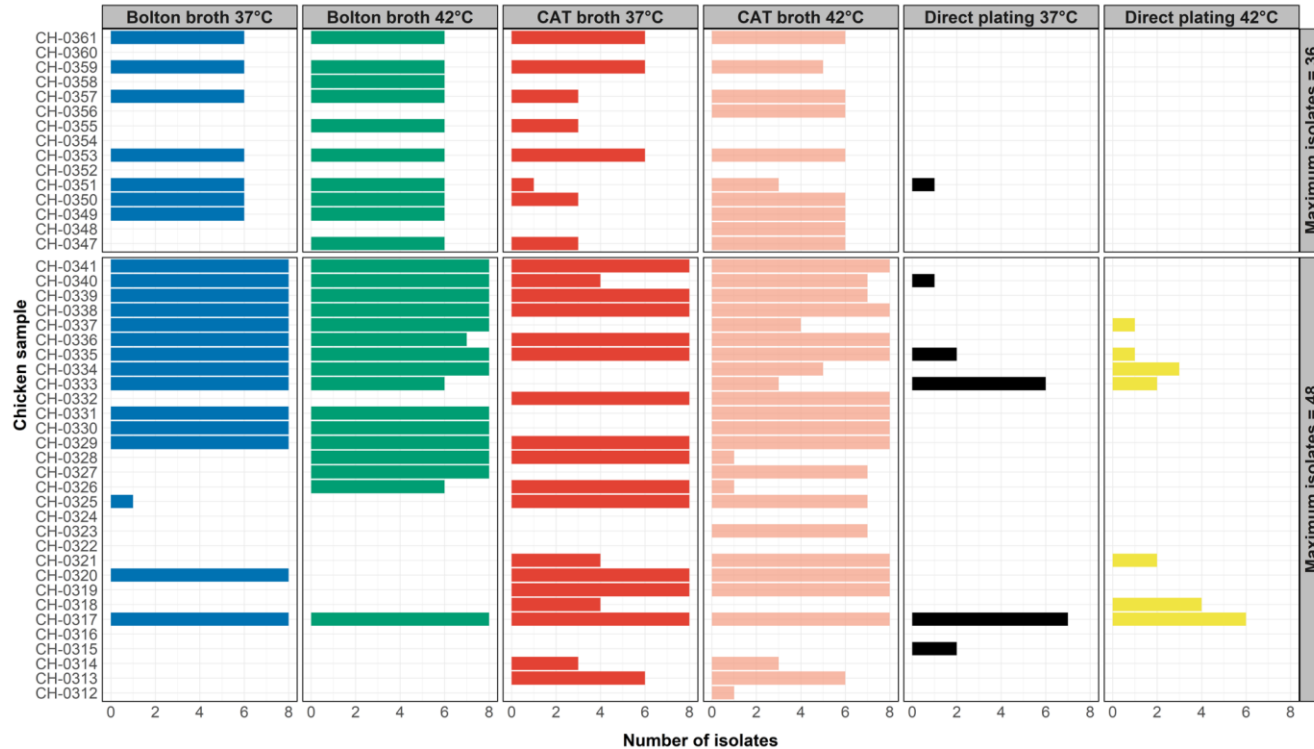
u-mCCDA



colonies from each positive plate isolated on CBA

Theoretical maximum of 48 (first 30 samples) and 36 (last 15 samples) isolates

Isolation of *Campylobacter* from chicken samples using methods tested



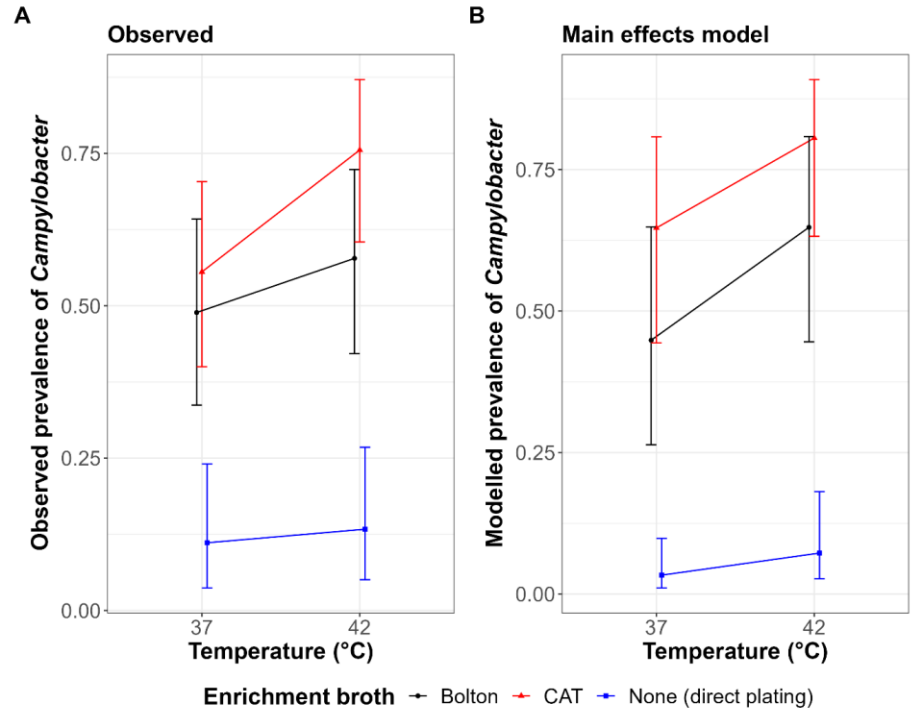
743 isolates obtained
0-45 isolates per sample

10 samples positive through CAT/Bolton enrichment or direct plating only

Campylobacter recovery modelling

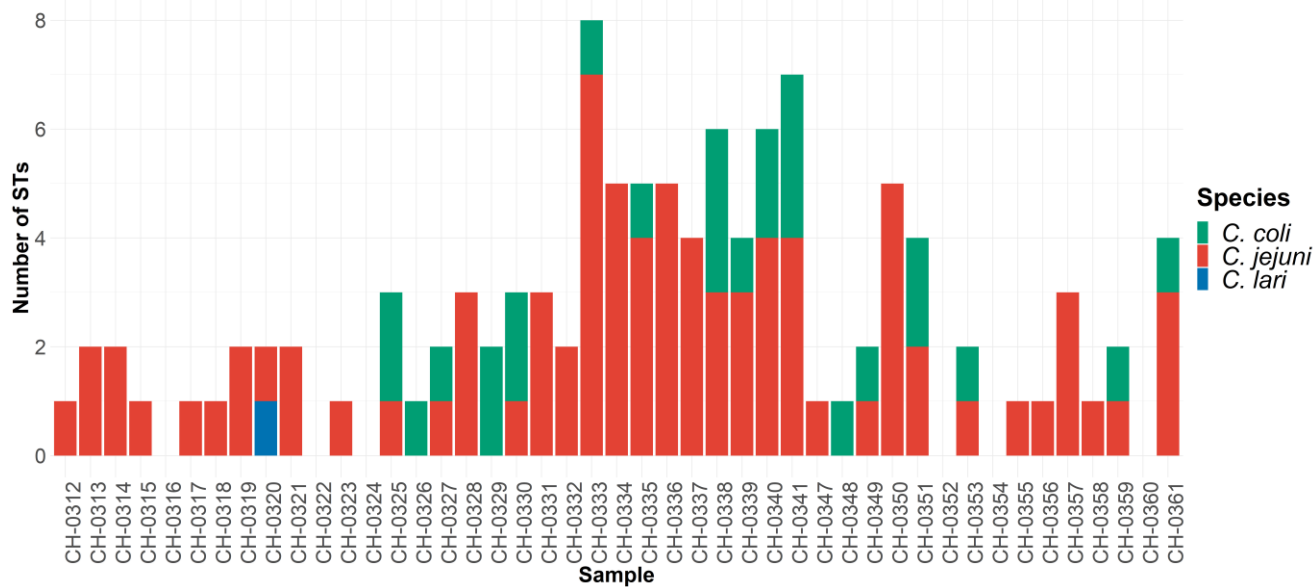
Effect of culture method on recovery

- Concordance between agar types was high
 - mCCDA more sensitive (McNemar $p=0.0003$)
- Multivariable mixed effects model testing effect of broth and temperature using mCCDA only
 - Higher detection at 42°C compared to 37°C ($p=0.02$)
 - Higher detection with Bolton broth vs direct plating ($p<0.001$) and with CAT vs Bolton broth ($p=0.039$)
- No evidence that the effect of temperature varied by Broth type
- Culture method can have implications on contamination rates and initial *Campylobacter* recovery



Campylobacter species and STs

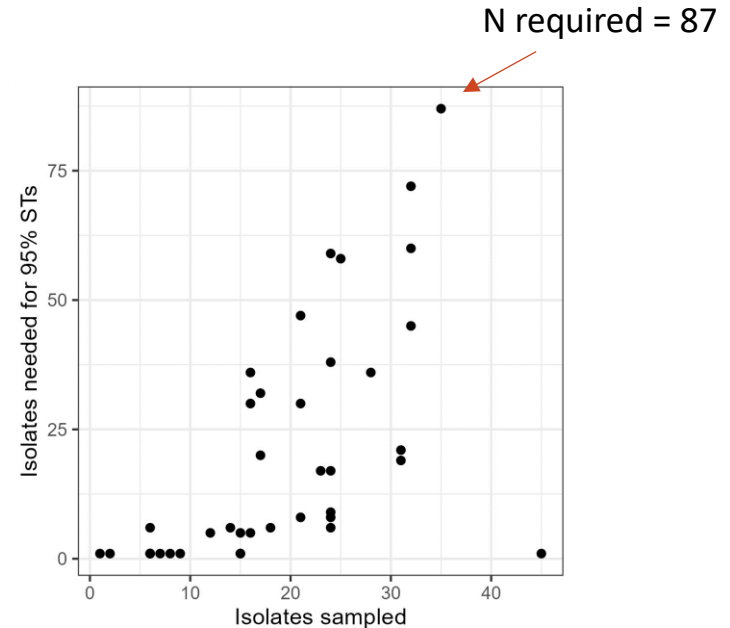
- Three species identified
 - *C. jejuni*, *C. coli* and *C. lari*
 - 33% of samples contained >1 species
- 62 STs identified (14 novel)
 - 1-8 STs per positive sample



Implications on source attribution and outbreak investigation

Probability of identifying theoretical outbreak STs and the number of isolates needed to sample ST diversity

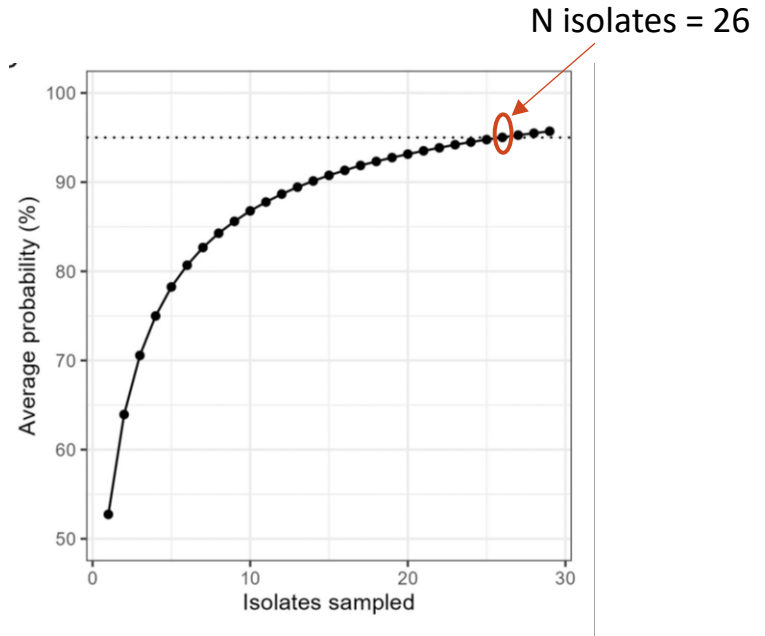
- Most studies take a limited number of isolates per sample
 - High number of isolates may be required for samples with many STs or rare STs
- Focusing on STs, a simulation (with replacement) was performed using resampling of the observed ST distribution
- Find the number of isolates required for the expected number of STs found to be at least 95% of the observed number



Implications on source attribution and outbreak investigation

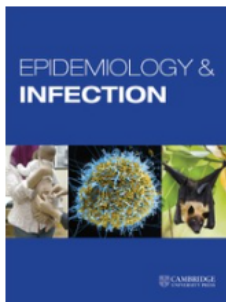
Probability of identifying theoretical outbreak STs and the number of isolates needed to sample ST diversity

- A theoretical outbreak scenario
 - Calculate number of isolates needed for the average probability of detecting a specific “outbreak” ST in the sample to reach 95%
- High diversity and limited sampling may affect outbreak investigation and source attribution
 - Underestimation of outbreak burden?
- Further diversity at the SNP/AMR genotype level – number of isolates required may further exceed estimated




Summary


- Isolation method may be important for the recovery of *Campylobacter* from chicken samples
- Chicken samples can contain diverse *Campylobacter* populations
- High diversity of *Campylobacter* within individual samples can have implications on source attribution and outbreak investigation



High *Campylobacter* diversity in retail chicken: epidemiologically important strains may be missed with current sampling methods

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