Evaluation of a Phenotypic MicroArray (BIOLOG) for characterisation of bovine reproductive *Campylobacter fetus* subspecies and other *Campylobacter*-like isolates by comparison with standard phenotypic, PCR and 16S rDNA sequencing.

S.C. de Wet[A], *A. Koya*[B], *S. Moss*[A], *B. Venus*[C] *& A. Lew-Tabor*[C]

[A]*Department of Employment, Economic Development and Innovation, Biosecurity Queensland, Brisbane, QLD, 4001, Australia*
[B]*School of Veterinary Science, University of Queensland, Gatton Campus, QLD, 4343, Australia*
[C]*Queensland Alliance for Agriculture and Food Innovation, St Lucia, QLD, 4072, Australia*

Introduction: Distinguishing between *Campylobacter fetus* subsp *venerealis (Cfv)* and its close relative *Campylobacter fetus* subsp *fetus* (*Cff)* is essential due to differences in disease aetiology, epidemiology and clinical significance. It is advisable that all *Campylobacter*-like organisms isolated from the bovine prepuce be identified. Recent reports indicate that reliance on PCR alone can lead to false-negative and -positive results. Due to variance, glycine intolerance to screen for *Cfv* is unreliable. H_2S production for differentiation of *Cfv* biovars *intermedius* (*Cfvi)* and venerealis (*Cfvv)* is insensitive. The need to uncover reliable unique assays for differentiation is clear.

Methods and results: A number of *Campylobacter*-like laboratory and local abattoir isolates (n=88) was characterised and compared with a *Cfv* type strain (ATCC 19438). Eight independent tests (World Organisation for Animal Health (OIE) standards) were used. Eight species were identified: *Cfvv* (n=41)*, Cfvi* (n=11), *Cff* (n=4), *Arcobacter* spp (n=18), *C. sputorum* (n=5), *C. hyointestinalis* (n=4), *C. upsalensis* (n=3) and *C. mucosalis* (n=2). The isolates displayed varying results using two different PCR's targeting *parA*. 16S rDNA sequencing was used to verify the phenotypic results. Representative isolates from each group, including typical and a-typical

strains were compared with the type strain using Phenotypic MicroArrays (PM) (BIOLOG). A total number of 940 different assays including carbon, nitrogen, phosphorus and sulphur sources as well as chemical additives and antibiotics were tested. For the type strain, utilisation of 21 carbon sources of interest was identified. Utilisation of 54 compounds at high (9.5) and low pH (4.5) indicated a preference for tolerance of additives (including glycine) at pH 9.5. Resistance to at least 35 new and previously unpublished chemical additives were found.

Conclusion: The BIOLOG PM is a user-friendly and powerful tool providing vast quantities of phenotypic data for identifying additional biochemical assays, expanding phenotypic characterisation and improving growth, transport and enrichment procedures.