Unravelling the genotypic and phenotypic diversity of the psychrophilic *Clostridium estertheticum* complex, a meat spoilage agent

Background:

Clostridium estertheticum and *C. estertheticum*-like spp. are obligate anaerobic psychrophiles causing "blown pack" spoilage of chilled vacuum-packed meat.

WP1: Relevant aspects of *Clostridium estertheticum* as a specific spoilage organism of vacuumpacked meat – a review

https://www.mdpi.com/2076-2607/7/5/142

WP2: Occurrence of Clostridium estertheticum and C. estertheticum-like spp. in the food chain

Detection of psychrophilic *Clostridium* spp. in fecal samples from cattle of different ages sampled at the slaughterhouse level

https://pubmed.ncbi.nlm.nih.gov/32818242/

Insight: Forty (39%) of 102 samples were RT-PCR positive. The frequency of positive samples was the following: 9 (45%) of 20 in calves (aged ≤ 160 days), 23 (43%) of 54 in young cattle (aged 161 to 1,000 days), and 8 (29%) of 28 in cows or bulls (aged >1,000 days). Six strains were isolated from 6 of 40 RT-PCR-positive samples. The six isolates were identified as *C. estertheticum* (n = 1), *Clostridium frigoriphilum* (n = 1), and *C. estertheticum*-like spp. (n = 4).

Detection of psychrophilic *Clostridium* spp. causing "blown pack" spoilage in meat juice samples from chilled vacuum-packed beef and lamb meat imported from different countries to Switzerland https://pubmed.ncbi.nlm.nih.gov/31825674/

Insight: One hundred fifty-four MJS (*n* = 78 from beef; *n* = 76 from lamb meat) were screened for psychrophilic *Clostridium* spp. by quantitative PCR. Psychrophilic *Clostridium* spp. were detected in 10 MJS, of which 2 were from beef and 8 were from lamb meat. The two beef MJS originated from Spain and Lithuania, whereas the lamb MJS originated from New Zealand and Australia.

WP3: Sequences and Genomics of *Clostridium estertheticum* and *C. estertheticum*-like spp.

3.1 Sequences:

https://pubmed.ncbi.nlm.nih.gov/32817160/ https://pubmed.ncbi.nlm.nih.gov/33060280/ https://pubmed.ncbi.nlm.nih.gov/33154005/

3.2 Genomics:

Comparative genome analysis and phenotypic characterization of *Clostridium gasigenes* CGAS001 isolated from chilled vacuum-packed lamb meat https://www.frontiersin.org/articles/10.3389/fmicb.2020.02048/full

Insight: The genetic and phenotypic analyses suggest that CGAS001 constitutes a novel subspecies of *C. gasigenes* adapted to a saprophytic lifestyle and can synthesize narrow spectrum antimicrobial compounds.

Whole genome sequence-based identification of *Clostridium estertheticum* complex strains supports the need for taxonomic reclassification within the species *Clostridium estertheticum* https://www.frontiersin.org/articles/10.3389/fmicb.2021.727022/full

Insight: We have used WGS-based methods, including pan- and core-genome phylogenomics and pairwise nucleotide-level comparisons (dDDH and ANI), to identify 13 MJS and BFS CEC isolates to the species level and determine the phylogeny of CEC species. The WGS-based analyses provided a basis to propose the abolishment of the current subspecies classification within species *C. estertheticum*.

Genotypic and phenotypic diversity of the psychrophilic Clostridium estertheticum complex

https://www.frontiersin.org/articles/10.3389/fmicb.2022.856810/abstract

Insight: The spoilage of vacuum-packed meat by *Clostridium estertheticum* complex (CEC), which is accompanied with or without production of copious amounts of gas, has been linked to the acetone-butyrate-ethanol (ABE) fermentation, but the mechanism behind the variable gas production has not been fully elucidated. Here we provide data, that variable inter- and intra-species gas production in meat by *C. estertheticum* and *C. tagluense* is associated with the distribution of the [NiFe]-hydrogenase *hyp* gene cluster whose absence or presence is associated with occurrence or lack of pack distention, respectively.