tested in 33 isolates were: *hreP* (100%), *sat* (100%), *ymoA* (100%), *myfA* (94%), *inv* (54%), *ystA* (94%), *yadA* (94%), *virF* (85%), *rfbC* (85%), *ail* (85%), *tccC* (85%), *fepA* (33%), *fes* (15%), *fepD* (15%), *ystB* (6%), and *ystC* (0%). There were four virulence genes carriage patterns found. Isolates of bioserotype 1A/O:5 (n=2) were *hreF*, *fes*, *sat*, *fepD*, *ymoA*, *ystB* and *fepA*; bioserotype 1B/O:8 (n=3) were *hreF*, *myfA*, *fes*, *sat*, *fepD*, *inv*, *ymoA*, *ystA*, *yadA* and *fepA*; and bioserotype 3 variant/O:3 were either *hreP*, *virF*, *rfbC*, *myfA*, *sat*, *inv*, *ail*, *ymoA*, *ystA*, *tccC*, *yadA* and *fepA* (n=6) or without the *fepA* gene (n=22). Interestingly, the bioserotype 1A/O:5, that was grouped as non-pathogenic, carried some of the virulence genes which were not traditionally tested. We also found the loss of the *ail* gene (involved in adhesion, invasion, and protection from bactericidal effects) in the pathogenic bioserotype 1B/O:8 isolates.

**MB02: High Resolution Melt Curve Analysis for Mutation Detection in Quinolone-resistant Determining Region of DNA Topoisomerase IV Genes in Salmonella**

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The increasing prevalence of quinolone-resistant *Salmonella* serovars has raised public health concerns in Malaysia. The DNA topoisomerase IV enzyme plays an important role in bacterial DNA replication. Mutations in the quinolone-resistant determining region (QRDR) of the bacterial topoisomerase IV genes contribute to quinolone-resistance among the bacteria. However, mutations in the QRDR of these genes are not common among *Salmonella*. Hence, it is often too costly and ineffective to analyse large sample sizes via DNA sequencing. The objective of this study was to develop a high resolution melt curve (HRM) analysis for rapid mutation screening for topoisomerase IV genes. Two pairs of primers were designed to target the QRDRs of the *parC* and *parE* genes, which encode the subunits of the DNA topoisomerase IV enzyme. Twelve quinolone-resistant *Salmonella* Typhimurium strains were used to develop the HRM assay. The target regions were subsequently sequenced to confirm the presence of base substitutions. The HRM primers amplified a 219 bp and 193 bp region in *parC* and *parE* QRDR, respectively. Sequence analysis showed that only one strain consisted of multiple base substitutions in the *parC* QRDR, but others were silent mutations. The majority (n=1) of the strains consisted of the wild-type sequence in *parC* QRDR. In *parE* QRDR, base substitutions were detected in two strains, but only one resulted in an amino acid change (Met438-Ile). HRM analysis was able to detect mutations in the QRDR of the genes of interest. Different plots showed that the melt curves of the mutants were distinctly separated from the wild-type. In conclusion, HRM analysis was able to detect mutations in the QRDR of the topoisomerase IV genes. Hence, this method provides a rapid and sensitive screening tool for mutation detection, thus enabling high-throughput analysis of large sample sizes.