Supplementary material for

High quality reference genomes for toxigenic and non-toxigenic *Vibrio cholerae* serogroup O139.

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These supplementary materials include:

Table S1. List of strains and genome sequences used in this study (attached .xls

 spreadsheet).

Figures S1-S6.

Supplementary references. Additional references for cited data and supplementary figures.

Additional materials that support this study are available in Figshare:

https://dx.doi.org/10.6084/m9.figshare.6480266



Figure S1. Confirmation of the duplication of the CTXφ bacteriophage in 48853_H01. Short reads for 48853_H01 were mapped against the N16961 reference genome (see

Methods). Mapping data were visualised using Artemis⁸².



Figure S2. Confirmation of a duplication of VSP-1 in 48853_H01. (A): Short reads for 48853_H01 were mapped against the N16961 reference genome. Mapping data suggested that approximately double the number of reads mapping to VSP-1 were present relative to those mapping to the surrounding chromosome. (B): A region homologous to VSP-1 was detected on both chromosomes 1 and 2 in the 48853_H01 assembly. Data were visualised using Artemis and BamView ^{82,83} and ACT ⁶⁷.



Figure S3. BLAST atlas comparing the four closed assemblies from this study, and that of N16961, to MO10. The loci harbouring GI-11, GI-16, and SXT are also indicated. VPI-2 is not present in the MO10 genome. MO10 harbours an expanded SXT variant. The O139 LPS and capsule-production genes are absent from N16961 (Supplementary Fig. S4). The sequences of both *V. cholerae* chromosomes (*i.e.*, all MO10 contigs) were concatenated to generate this figure.



Figure S4. Comparison of the structure of capsule and lipopolysaccharide biosynthesis loci in five *V. cholerae*. The genes encoding the O1 antigen are absent in 48853_H01, replaced with those encoding the O139 antigen and polysaccharide capsule. These are common to MO10, and partially conserved in the non-toxigenic strain. Core LPS genes vary between MO10 and 48853_F01, and O-antigen genes are not shared between 48853_F01 and the three non-7PET *V. cholerae* to which it is most closely related (Fig. 3A), represented here by HE 09. Syntenic regions were visualised using ACT ⁶⁷.



Figure S5. Unedited V. cholerae species phylogeny. The non-7PET phylogeny reported in

Fig. 3A is presented without artificial branch shortening.



Figure S6. Phylogenetic trees computed under the GTR+ASC model. Phylogenies computed using an ascertainment bias correction model from the alignments used to generate Fig. 3A and Fig. 3B are presented (A and B, respectively).

Supplementary references. Additional references for cited data and supplementary figures.

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