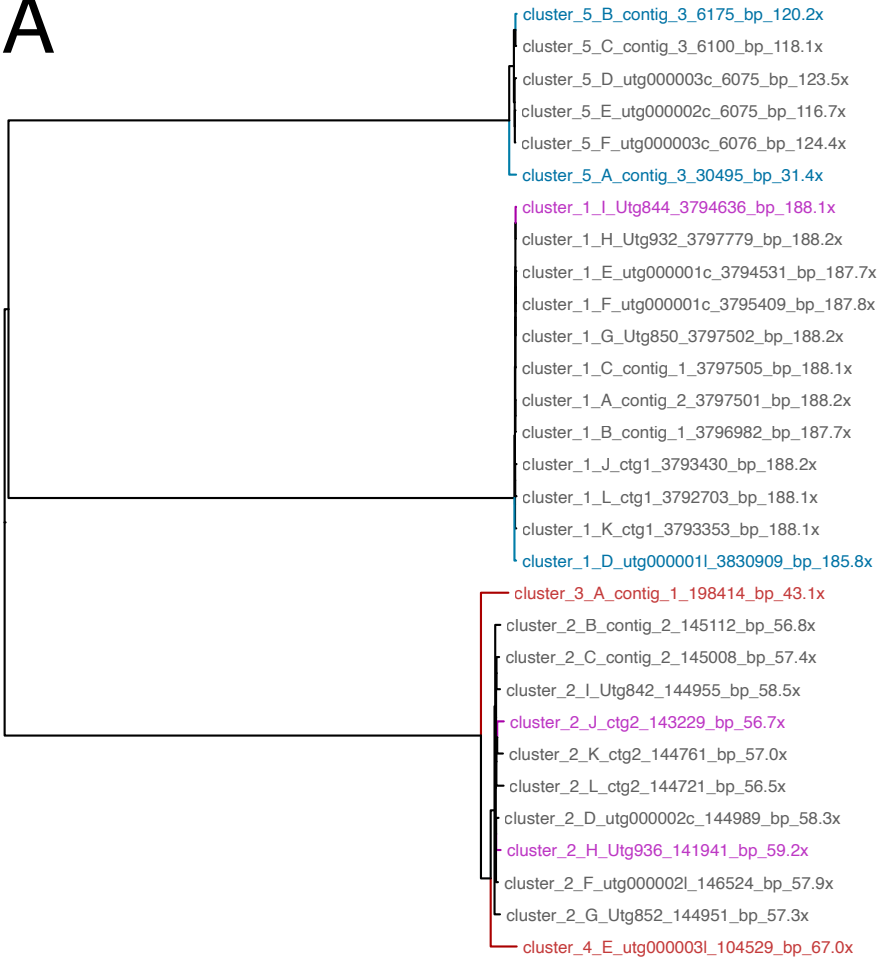
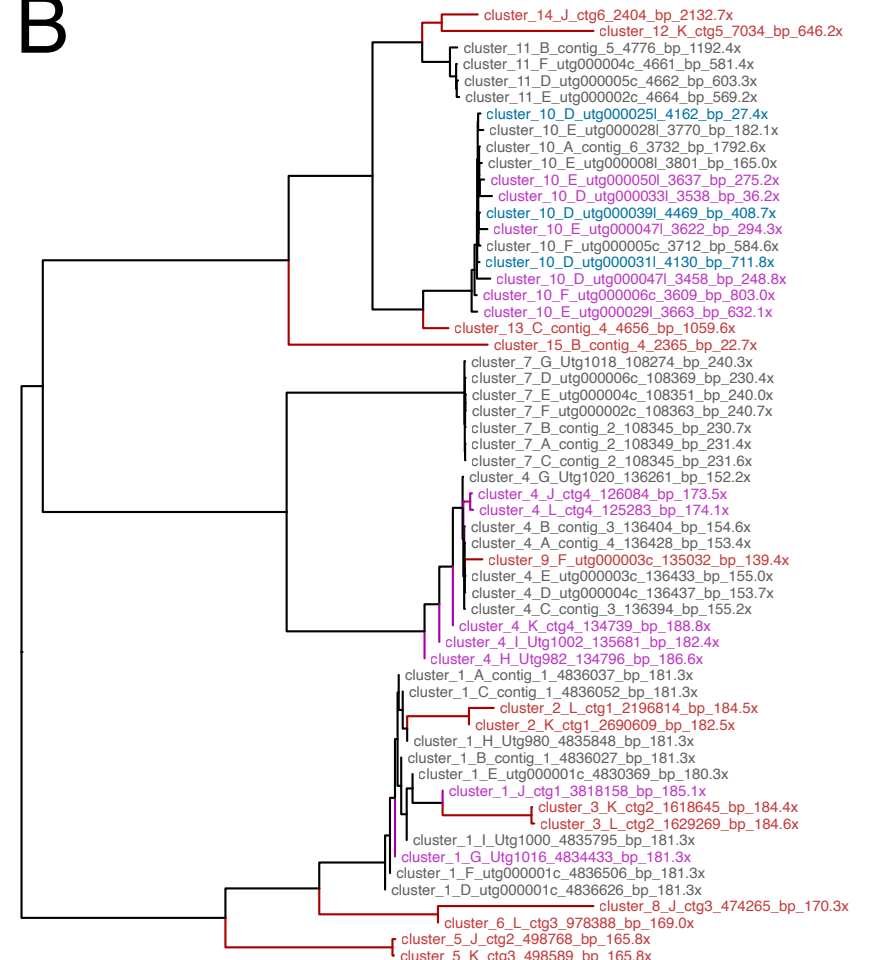


A



B



C

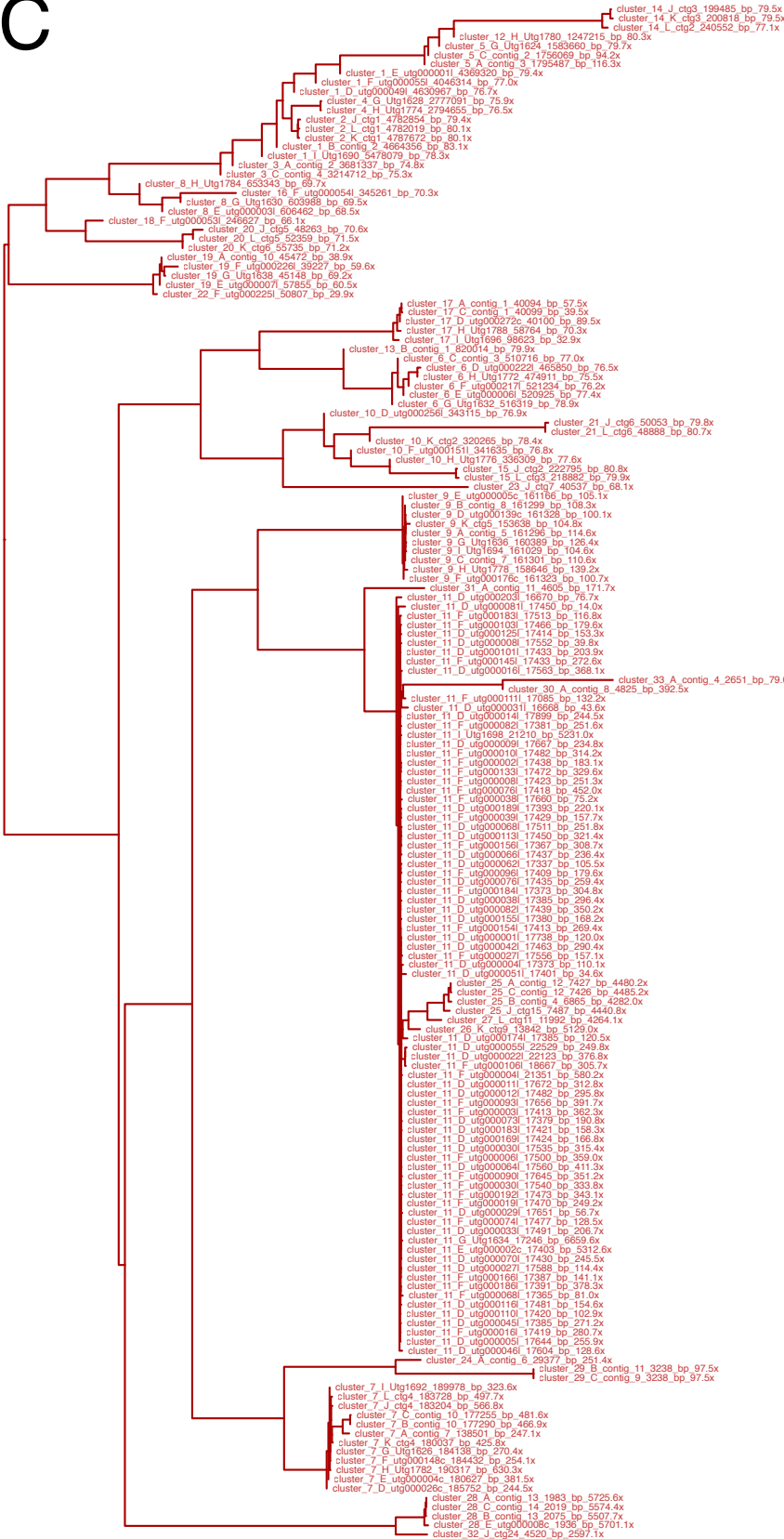


Figure S7: Examples of Trycycler clustering and manual intervention. This figure shows FastME trees generated by the Trycycler cluster command for three of the real read test genomes, illustrating good, mediocre and bad cases. For each genome, the author’s cluster choices are described, as are any manual interventions needed to reconcile the valid clusters. Clusters which were manually excluded are coloured **red**. Contigs in valid clusters that needed to be manually removed before reconciliation are coloured **violet**. Contigs in valid clusters which needed to be manually trimmed before reconciliation are coloured **blue**.

- A. *Acinetobacter baumannii* J9 ONT rapid:** a good case with clear clustering. Clusters 3 and 4 (misassembled contigs of the 145 kbp plasmid) were excluded, leaving valid clusters 1, 2 and 5. During cluster reconciliation, one cluster_1 contig had to be manually trimmed (due to low-quality start/end sequence), one cluster_1 contig had to be manually removed (due to poor pairwise alignment), two cluster_2 contigs had to be manually removed (due to being incomplete) and two cluster_5 contigs had to be manually trimmed (due to excessive length).
- B. *Enterobacter kobei* MSB1_1B ONT rapid:** a mediocre case with more complex clusters. Clusters 2, 3, 5, 6, 8 (misassembled contigs of the chromosome), 9 (misassembled contig of the 136 kbp plasmid), 12, 13, 14, and 15 (misassembled contigs of the small plasmids) were excluded, leaving valid clusters 1, 4, 7, 10 and 11. During cluster reconciliation, two cluster_1 contigs had to be manually removed (due to being incomplete or poor pairwise alignment), five cluster_4 contigs had to be manually removed (due to being incomplete), three cluster_10 contigs had to be manually trimmed (due to excessive length) and six cluster_10 contigs had to be manually removed (due to being incomplete or unable to circularise).
- C. *Serratia marcescens* 17-147-1671 ONT rapid:** a bad case where valid clusters were unclear. Insufficient read length and genome heterogeneity both contributed to the poor results. Without good clusters, it was not possible to proceed with Trycycler assembly.