

Table S6. Backbone and O-Island ORF Criteria. Details pertaining to inclusion of ORFs and to special adjustments in the Concatenated Backbone Reference Databases and the limited O-Island analysis.

	Backbone	O-Islands
<u>Inclusion Criteria</u>		
ORF:		Segments included if >10kb in length
	>45 nucleotides long	>45 nucleotides long
	>95% similar	>95% similar
	Equal length*	Equal length
	Non-duplicate (no other ORF with >95% similarity elsewhere in chromosome)	Non-duplicate: Lambdoid prophages that shared nearly identical sequences and duplicated EDL933 SpLE1 were excluded, but the late region of the Stx 2 phage was included because its nucleotide sequence is unique among lambdoid prophages
<u>ORF Adjustments</u>		
	Start and stop codons were not analyzed	Start and stop codons were not analyzed
	Overlapping nucleotides removed	Overlapping nucleotides removed
O157 Sakai and EDL933 genome comparison:**	We corrected upwards all corresponding ORFs in O157 Sakai and EDL933 that were of unequal lengths in the databases if different start codons had been assigned in the two annotations. We analyzed the rectified ORFs if they met all other criteria for inclusion. Using O157 Sakai as a reference, we included ORFs not entered as such in EDL933 in GenBank, if all criteria for analysis for these overlooked ORFs were met.	

* Identical length indels were not considered SNPs.

**All ORFs in the Cluster 3 strains (O157 Sakai and EDL933) that met these criteria were also compared visually to all ORFs listed on the websites of the projects that sequenced these two genomes (<http://genome.naist.jp/bacteria/o157/> and <http://www.genome.wisc.edu/sequencing/o157.htm>). Nucleotide positions in the O157 Sakai sequence were used as reference.