Whole Genome Analysis of *Streptococcus* pneumoniae Serotype 4 Causing Outbreak of Invasive Pneumococcal Disease, Alberta, Canada

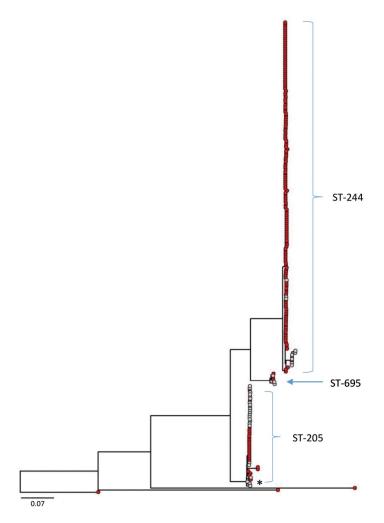
Appendix

Appendix Table. Number of single nucleotide variants in the core genome within and between the major phylogenomic clades of

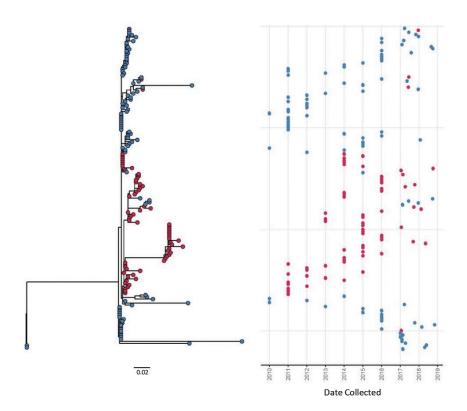
Streptococcus pneumoniae serotype 4 ST244 isolates collected in Alberta (Figure 3)

	No.	Avg. SNVs	Max. SNVs	Avg. SNVs	Min. SNVs	Max. SNVs
Clade	isolates	within clade	within clade	from next clade†	from next clade†	from next clade†
A1	21	4	12	12	6	23
A2	8	6	14	9	6	20
A3	10	2	10	15	6	29
A4	25	12	27	44	32	62
A5	19	4	19	38	30	52
A6	5	5	12	15‡	4‡	22‡

^{*}Avg, average; Max, maximum; Min, minimum; SNV, single nucleotide variants †Number of SNVs compared to the following closest ancestral clade in the phylogeny.



Appendix Figure 1. Maximum likelihood core SNV phylogenetic tree of *S. pneumoniae* serotype 4 isolates collected in Alberta (red nodes, n = 190) during 2010–2018, and other provinces (grey nodes, n = 37). All isolates from other provinces were collected in 2019 except 1 ST205 isolate from 2011 and 1 from 2012. We used a total of 3,075 sites in the phylogeny and included 77.2% of the core genome; we used isolate SC16-4549-P (the oldest outlier isolate) as the tree root and *S. pneumoniae* TIGR4 (NCBI accession number NC_003028.3) as a mapping reference.



Appendix Figure 2. Temporal distribution of isolates (n=159) collected in Alberta in the maximum likelihood core SNV phylogenetic tree of *S. pneumoniae* serotype 4 ST44 isolates during 2010–2018. We used a total of 615 sites in the phylogeny and 97.4% of the core genome was included. An internal isolate SC19-3744-P (oldest outlier) was used as a mapping reference and root. Isolates from the Calgary region are indicated with red circles, from Edmonton with blue circles.