

Complete chloroplast genome sequence of *Picea engelmannii*, isolate Se404-851 from western Canada

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Abstract

Picea engelmannii, is an Engelmann spruce tree grown in western Canada. Here we present the complete chloroplast genome sequence of the *Picea engelmannii*, isolate Se404-851. This sequence contributes data to the study of the evolutionary phylogeny of the *Picea* organisms, and consequently, facilitating the improvement of Canada's forestry industry.

Genome Announcement

As part of the SpruceUp (1) and SMarTForests (2) projects, we sequenced, assembled and annotated the chloroplast genome of *Picea engelmannii* isolate Se404-851. This work contributes to improving the selection of spruce trees in breeding programs, a vital part of Canada’s forestry industry.

The needle tissue sample was collected from Kalamalka Forestry Centre in British Columbia (36°17’60”N, 105°24’0”W; elevation: 298 m). We then sequenced the sample at the British Columbia Genome Sciences Centre.

To sequence the sample, a modified version of TruSeq DNA PCR-Free kit (E6875-6877B-GSC, New England Biolabs) genome protocol was used to generate a 900-bp gap Illumina library on a Microlab NIMBUS liquid handling robot (Hamilton). Briefly, 5 μ g of genomic DNA was subjected to shearing by sonication (Covaris LE220) using a Duty Factor of 5 and Peak Incident Power of 450 for 70 seconds. The sonicated DNA products were concentrated with PCRClean DX magnetic beads (Aline Biosciences) and fractionated in 2 lanes of a 6% PAGE gel to recover fragments greater than 700-bp for library preparation. The isolated DNA fragments were end-repaired and bead-purified with a 1.8:1 ratio of beads, then A-tailed and ligated with full length indexed TruSeq adapters, and bead-purified. For quality check, an aliquot of the constructed library DNA was PCR amplified with Illumina universal primers to estimate the library gap size using the Agilent 2100 Bioanalyzer HSDNA assay, while the library concentration was determined using KAPA qPCR Library Quantification kit (KK4824). The PCR-Free library was sequenced with paired-end 150 base reads on the Illumina HiSeqX platform using V4 chemistry according to manufacturer recommendations.

For assembly, we subsampled the reads into a few million read pairs. ABySS v2.1.1 (3) assembled each subset. Then, BWA v0.7.17 (4) filtered for contigs greater than 500-bp that aligned with the reference chloroplast, *Picea glauca* isolate PG29. In the 3M assembly, there was one resulting contig ($n=1$; $N_{50}=1743$). QUAST v5.0.0 (5) revealed zero misassemblies and internal gaps.

To be consistent, we modified our assembly to match the reference using BLAST v2.7.1 (6). To ensure there were no missing sequences at the ends of our assembly we introduced a gap there, circularized the sequence and ran ABySS-sealer, closing the ‘end’ gap. Finally, Pilon v1.22 (7) polished the final assembly.

The Se404-851 chloroplast genome is 123,601-bp in length, with a GC content of 38.74%. GeSeq (8) annotated 114 genes: 74 protein-coding, 36 tRNA-coding, 4 rRNA-coding genes, using all available *Picea* chloroplast genomes as reference. We manually annotated four

genes: *rps12*, *petB*, *petD*, and *rpl16*. OGDRAW v1.2 (9) generated the genome map. By introducing this new chloroplast genome, further analysis of phylogeny and evolution of these spruce trees can be conducted to achieve the project goals.

Accession number(s). The complete chloroplast genome sequence of *Picea engelmannii*, isolate Se404-851 can be found in Genbank under MK241981. The tissue samples used can be found under BioSample: SAMN10388286; BioProject: PRJNA504036. The annotation NCBI references are as follows: *Picea abies* (NC_021456), *Picea asperata* (NC_032367), *Picea glauca* isolate PG29 (NC_028594), *Picea morrisonicola* (NC_016069), and *Picea sitchensis* (NC_011152).

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