PLASTOME ANNOUNCEMENT

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Complete chloroplast genome sequence of *Solanum hjertingii*, one of the wild potato relatives

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ABSTRACT

Solanum hjertingii is a wild tuber-bearing species classified in the Solanaceae family. The chloroplast genome of *S. hjertingii* was completed via *de novo* assembly using Illumina paired-end sequencing data. Total length of the chloroplast genome of *S. hjertingii* is 155,545 bp consisting of 85,976 bp in a large single copy, 18,383 bp in a small single copy, and 25,593 bp in a pair of inverted repeat regions. Its structure is circular and typically quadripartite. It contains 158 predicted genes in total, including 105 protein-coding, 45 tRNA, and eight rRNA genes. Maximum likelihood phylogenetic analysis of the sequence along with 33 species in the *Solanaceae* family revealed that *S. hjertingii* belongs to a large clade with other *Solanum* species including *S. tuberosum* and is most closely grouped in the clade with *S. hougasii* and *S. stoloniferum* in the clade.

The wild tuber-bearing Solanum hjertingii Hawkes 1963 is a relative of the potato (Solanum tuberosum) originating from Mexico. It has been identified as a potential source of resistance to blackspot bruising because it exhibits neither enzymatic browning nor blackspot caused by impact or compression damage during harvest or storage (Hawkes 1990; Sim et al. 1997; Culley et al. 2002; Hara-Skrzypiec and Jakuczun 2013). It was also determined in contemporary researches to be resistant to both biotic and abiotic stresses such as Phytophthora infestans, drought and salt (data not shown). For these reasons, the wild species can be used for introgression of certain traits into the cultivated potatoes. However, S. hjertingii and S. tuberosum are not conventionally crossable, although both are tetraploids. The endosperm balance numbers (EBNs) for these species are 2 and 4, respectively (Hawkes 1990; Ortiz and Ehlenfeldt 1992; Cho et al. 1997). As a result, more advanced methods such as bridge crossing and somatic hybridization can be used for potato breeding as applied with other wild Solanum species (Hermsen 1966; Hermsen and Ramanna 1973; Binding et al. 1982; Iwanaga et al. 1991; Park et al. 2005; Luthra et al. 2019). The bridge crossing method was applied once (Culley et al. 2002), but somatic hybridization has not yet been tried with S. hjertingii. Therefore, we have been trying it and developing molecular markers to identify cytoplasm genome composition after obtaining hybrids via somatic hybridization.

The wild *S. hjertingii* species (PI186559) was obtained from the Highland Agriculture Research Institute, South Korea (37°68′05.4″N 128°73′09.1″E) and the specimen deposited at the National Agrobiodiversity Center, South Korea (http://genebank.rda.go.kr/, Hyun-Jin Park, rosa2125@korea.kr) as voucher number IT301488. Chloroplast genome sequencing was performed via the Phyzen bioinformatics pipeline (Kim et al. 2015). Total genomic DNA was isolated from one of the S. hjertingii lines by using a Genomic DNA Extraction kit for plants (RBC, New Taipei City, Taiwan). An Illumina paired-end (PE) genomic library was constructed with the genomic DNA by following the PE standard protocol (Illumina, San Diego, USA) and was sequenced at Macrogen (http://www.macrogen.com/kor/) using an Illumina HiSeq2000 platform. Approximately 1.48 Gbp of the sequence raw data obtained in total was trimmed and low-guality bases with a raw Phred score of 20 or less were removed using the CLC quality trim program in the CLC assembly cell package version 4.2.1 (CLC Inc, Rarhus, Denmark). Finally, approximately 1.27 Gbp of high-quality PE reads were applied for de novo assembly using the CLC de novo assembly program in the same package followed by retrieving the principal contigs representing the chloroplast genome and arranging the representative chloroplast contigs using Nucmer (Kurtz et al. 2004) and BLASTZ analysis (Schwartz et al. 2003) with the chloroplast genome sequence of S. hougasii (MF471372, Cho et al. 2018; Kim and Park 2020a). Gene annotation and manual curation were performed with the GeSeg program (Tillich et al. 2017) and BLAST searches. Phylogenetic analysis was performed using the chloroplast coding sequences of S. hjertingii and 33 published species belonging to the Solanaceae family by using a maximum likelihood method with the Kimura 2-parameter model and 1,000 bootstrap options in MEGA 6.0 (Tamura et al. 2013).

Total length of the complete *S. hjertingii* chloroplast genome (MK690623) is 155,545 bp consisting of a large single copy (LSC) region of 85,976 bp, a small single copy (SSC) region of 18,383 bp, and a pair of inverted repeat (IRa and IRb) regions of 25,593 bp with the typical circular and quadripartite structure like most plastids. Overall GC content was

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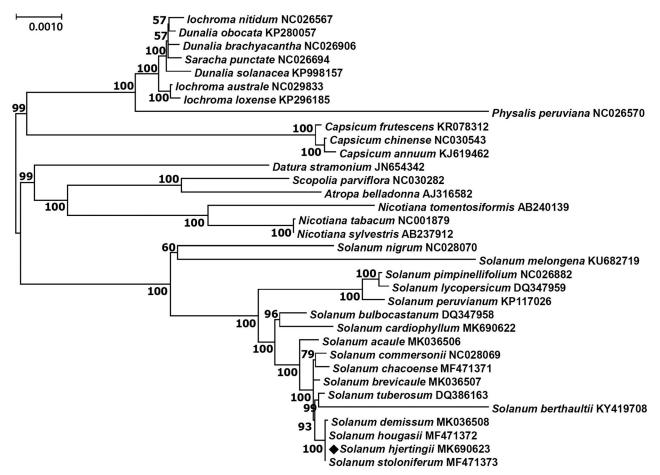


Figure 1. Maximum likelihood phylogenetic tree of *S. hjertingii* with 33 species belonging to the Solanaceae family based on chloroplast protein coding sequences. Numbers in the nodes are bootstrap values from 1000 replicates. The data were partially adopted from Park (2021).

37.88%. The closest *Solanum* species were *S. hougasii* (MF471372) and *S. stoloniferum* (MF471373, Park 2018; Kim and Park 2020b) each with a very high sequence identity of 99.97% and 99.96%, respectively. A total of 158 genes were annotated with an average length of 583.3 bp and gene features were typically identical to those of higher plants. The chloroplast genome consists of 105 protein-coding genes, 45 tRNA genes and eight rRNA genes with average sizes of 765.0 bp, 62.0 bp and 1131.0 bp, respectively.

Results from phylogenetic analysis revealed that *S. hjertingii* belongs to the same clade with other *Solanum* species as expected, and is most closely grouped in the clade with *S. hougasii* and *S. stoloniferum* (Figure 1). These results can be explained by the fact that the plastid DNA data generated a four-clade phylogeny and the three species originating from Mexico belong to the same clade (Spooner et al. 2008). Their genome compositions evolutionarily identified by gene and genomic in situ hybridization (GISH) analyses also support an AABB genome constitution (allotetraploid) for *S. hjertingii* and *S. stoloniferum*, and AABBPP (allohexaploid) for *S. hougasii* (Spooner et al. 2008; Pendinen et al. 2012; Ono et al. 2016).

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Ethical approval: This study did not involve endangered or protected species, and the plant was collected under the permission from Highland Agriculture Research Institute (37°68′05.4″N 128°73′09.1″E), Pyeongchang, South Korea.

Disclosure statement

No potential conflict of interest was reported by the author(s).

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Data availability statement

The data that support the findings of this study are openly available in the NCBI under accession number MK690623 (https://www.ncbi.nlm.nih. gov/nuccore/MK690623). The associated BioProject, SRA, and BioSample numbers are PRJNA704091 (https://www.ncbi.nlm.nih.gov/bioproject/PR JNA704091), SRR13766187 (https://www.ncbi.nlm.nih.gov/sra/SRR1376 6187), and SAMN18029792 (https://www.ncbi.nlm.nih.gov/biosample/SA MN18029792), respectively.

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