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Identification of Cold and Disease Resistance Genes *via* Chromosome-Level Genome Reference

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DESCRIPTION

Poncirus Raf., also known as Citrus, is a citrus species that belongs to the Rutaceae subfamily and includes two species: Poncirus polyandra and Poncirus trifoliata. Poncirus is a Chinese plant that differs from citrus, kumquats, and other genera. Poncirus plants have been widely used as germplasm resources for rootstocks in citrus production due to a number of advantageous characteristics, including efficient nutrient absorption and utilisation, cold hardiness, and resistance to a number of major diseases and pests [1-2].

Poncirus polyandra, a new species, is a member of the Rutaceae family's poncirus genus. It is sexually compatible with Citrus species but differs from citrus and poncirus polyandra in many ways, including flower size, petals, and stamen number. It is evergreen, has three leaflets on each leaf, large thorns on the shoots, and produces bitter, inedible fruits. The Poncirus polyandra has potential as a rootstock material for Citrus species and may provide useful information on the nature of the ancient Citrus-Poncirus divergence. It is a valuable medicinal resource that inhibits angiogenesis and locally reduces tumour cell expression. Poncirus trifoliata, the only other species of poncirus, has been widely used as a rootstock for citrus production due to resistance genes associated with biotic and abiotic stresses. In the poncirus trifoliata genome, for example, many candidate genes involved in disease tolerance and cold stress have been well characterised, including *Cystathionine Beta Synthase* (CBF) genes, *Nucleotide Binding Site* (NBS) genes, and other disease resistance genes [3-4].

A high-quality reference genome is required for the crop genome mining to be effective and efficient. Initially, already existing labels were used to gain an overall understanding of how the genes are classified into families. This allowed us to identify a preliminary set of genes or gene families of interest, including the candidate genes associated with the cold signalling, resistance to Huanglongbing (HLB), Citrus Tristeza Virus (CTV), and citrus nematodes, rapidly evolving gene families, poncirus polyandra-specific genes, and single-copy genes, by

matching with the poncirus trifoliata or arabidopsis thaliana [5-6].

The Nucleotide Binding Site (NBS) domain is a common domain found in the protein products of resistance genes that encode signal transduction pathways. The most common type of resistance gene in many plant species, *Nucleotide Binding Site-Leucine Rich Repeat* (NBS-LRR) genes, encode proteins with variable N-terminal domains connected by a predicted NBS domain and a short LRR motif. The CBF1-CBF3 protein family, also known as Dehydration Responsive Element Binding factor 1 (DREB1) proteins, consists of three members [7-8].

Using sequence alignment, three candidate genes associated with the cold signalling pathway in poncirus polyandra were identified based on the hypothesis that poncirus polyandra has cold tolerance-related genes that may be homologous to arabidopsis thaliana cold signalling pathway genes. Other candidate genes were discovered as a result of the availability of the poncirus polyandra genome. In addition to the NBS and CBF genes mentioned above, the poncirus polyandra genome contains 41 gene families containing 83 genes related to the arabidopsis thaliana cold signalling pathway, 19 *HLB* tolerance genes, 11 *CTV* genes, and one *citrus nematode* resistance gene. Overall, the enriched functions of lineage-specific and rapidly evolving genes in poncirus polyandra may be important for adaptive evolution and may contribute to the species' unique characteristics [9-10].

The goal of this study was to find genes with functional properties that improve resistance to abiotic and biotic stresses. This study provides not only a valuable genetic resource for poncirus polyandra with disease resistance and cold tolerance, but also a better understanding for future developments on gene function mining in poncirus polyandra, a plant species.

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