

**Title: Multiple isoforms of phosphoenolpyruvate carboxylase in the Orchidaceae (subtribe Oncidiinae): implications for the evolution of crassulacean acid metabolism**

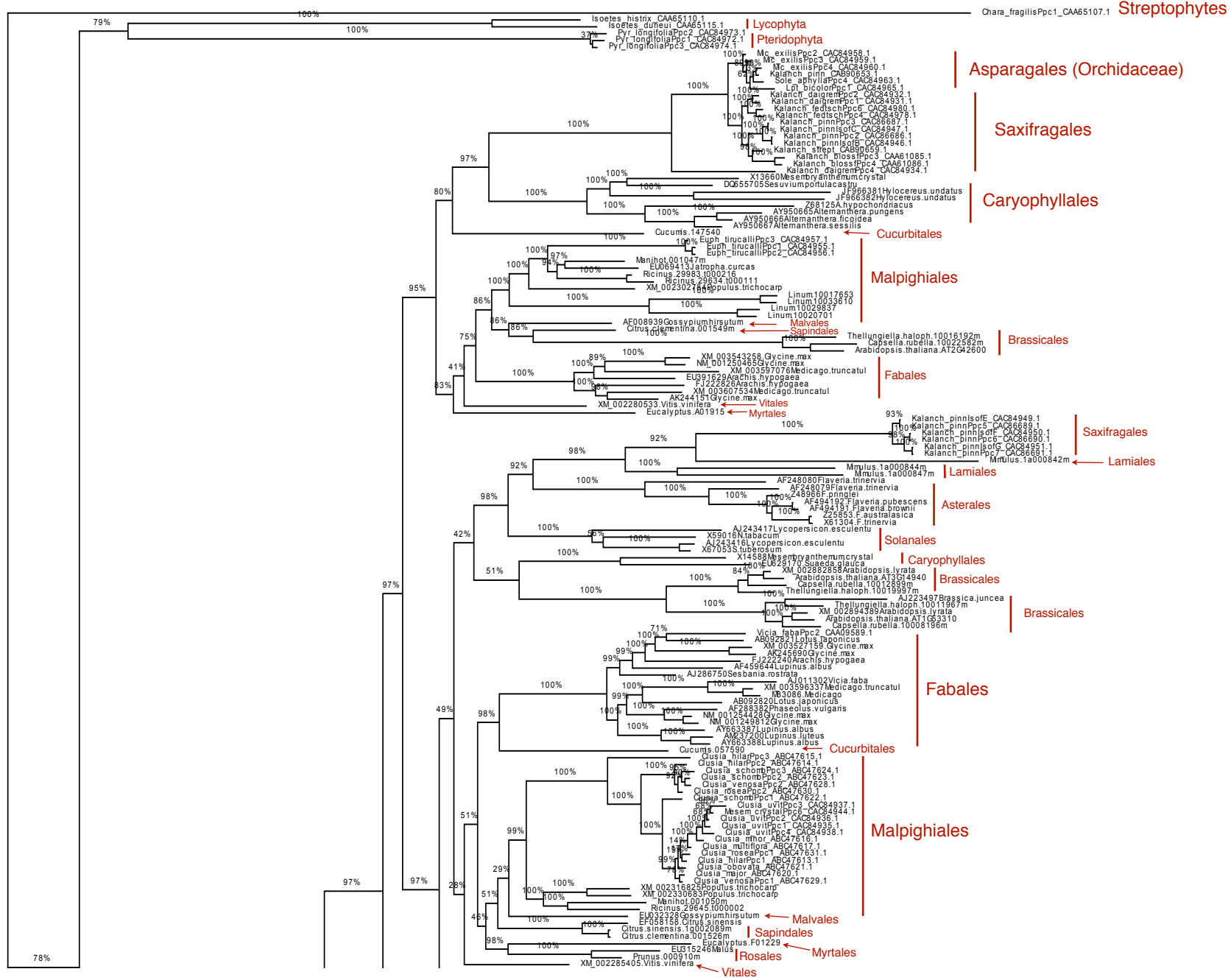
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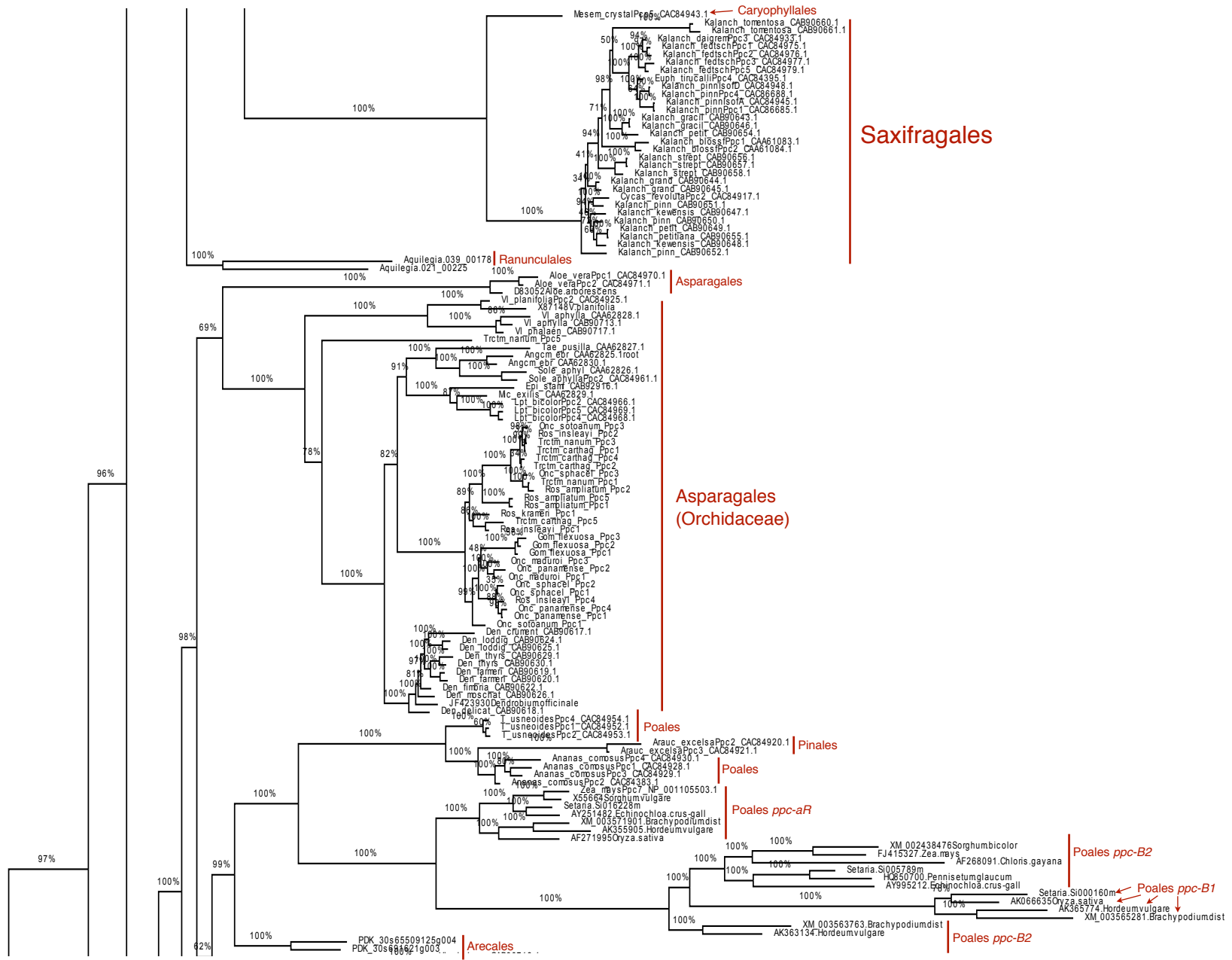
**Table S1.** Gene specific primer sets (GSP 5'– 3' position) used for 3'RACE amplification of the cDNA fragment coding for *Ppc* isoform for three species from the subtribe Oncidiinae. GSP1-1 to GSP1-23 were used for the first round of amplification, and GSP2-1 to GSP2-23 were used in the second round of amplification to reduce non-specific products

|         |                              |
|---------|------------------------------|
| GSP1-1  | AACTCTGAACGTCTGCCAAGCCTACGCG |
| GSP1-2  | AACTCTGAACGTTTGCCAAGCCTACACA |
| GSP1-3  | GACCCTGAATGTGTGCCAAGCCTTCACC |
| GSP1-4  | GACGCTGAATGTGTGCCAAGCCTTCACC |
| GSP1-5  | AACTCTGAACGTTTGCCAAGCCTACACG |
| GSP1-6  | TACTCTGAACGTTTGCCAAGCCTGCACA |
| GSP1-7  | GACCCTGAATGTGTGCCAAGCCTTCACT |
| GSP1-8  | GACCCTGAATGTGTGCCAAGCTTTCACC |
| GSP1-21 | CGACTCAGGCTCCGCTACCCG        |
| GSP1-22 | TTGGCAGTTGTACAAGGCTCAGGAG    |
| GSP1-23 | GGCAGCTATATAAGACTCAAGG       |
| GSP2-1  | GCGAATCAGAGATCCAAGCGATCATGG  |
| GSP2-2  | GCGGATCCGAGATCCAAGCAATGGTCA  |
| GSP2-3  | GCGAATCAGAGATCCAAGCGATCATCG  |
| GSP2-4  | GCGAATCAGAGATCCGAGCGATCATCA  |
| GSP2-5  | GCGAATCAGAGATCCGAGCGATCATCC  |
| GSP2-6  | GCGAATCAGAGATCCAAGCGATCATGG  |
| GSP2-7  | AAGGATCAGAGACCCAAATTTTCTTGT  |
| GSP2-8  | AAGGATCAGAGACCCAAAATTTTCTTGT |
| GSP2-9  | AAGGATCAGAGATCCAAATTTTCTTGT  |
| GSP2-10 | AAGAATCAGAGACCCAAATTTTCTTGTG |
| GSP2-12 | GCGGATCAGAGATCCAAGCGATCGTCA  |
| GSP2-13 | GCGGATCAGAGATCCAAGCGATCATCG  |
| GSP2-14 | GCGGATCAGAGATCCAAGCAATGGTCA  |
| GSP2-15 | GCGGATCCGAGATCCAAGCGACGGTCA  |
| GSP2-16 | AAGGATCAGAGACCCAAATTTTCATGT  |
| GSP2-21 | AACGTTTGCCAAGCCTACACGC       |
| GSP2-22 | GGCTCAGGAGGATCTCATAAAAAGTG   |
| GSP2-23 | GAAAGTAGCCAAGGAATTTGGA       |

**Table S2.** Gene specific primer sets (5'– 3' position) used for confirmation of the cDNA fragment recovered by RACE coding for *Ppc* isoform for three species from the subtribe Oncidiinae species. *Oncidium maduroi* (MAD), *Oncidium panamense* (PAN), and *Rossioglossum ampliatum* (AMP) forward (F) and reverse (R) primer sets were used for one round of amplification

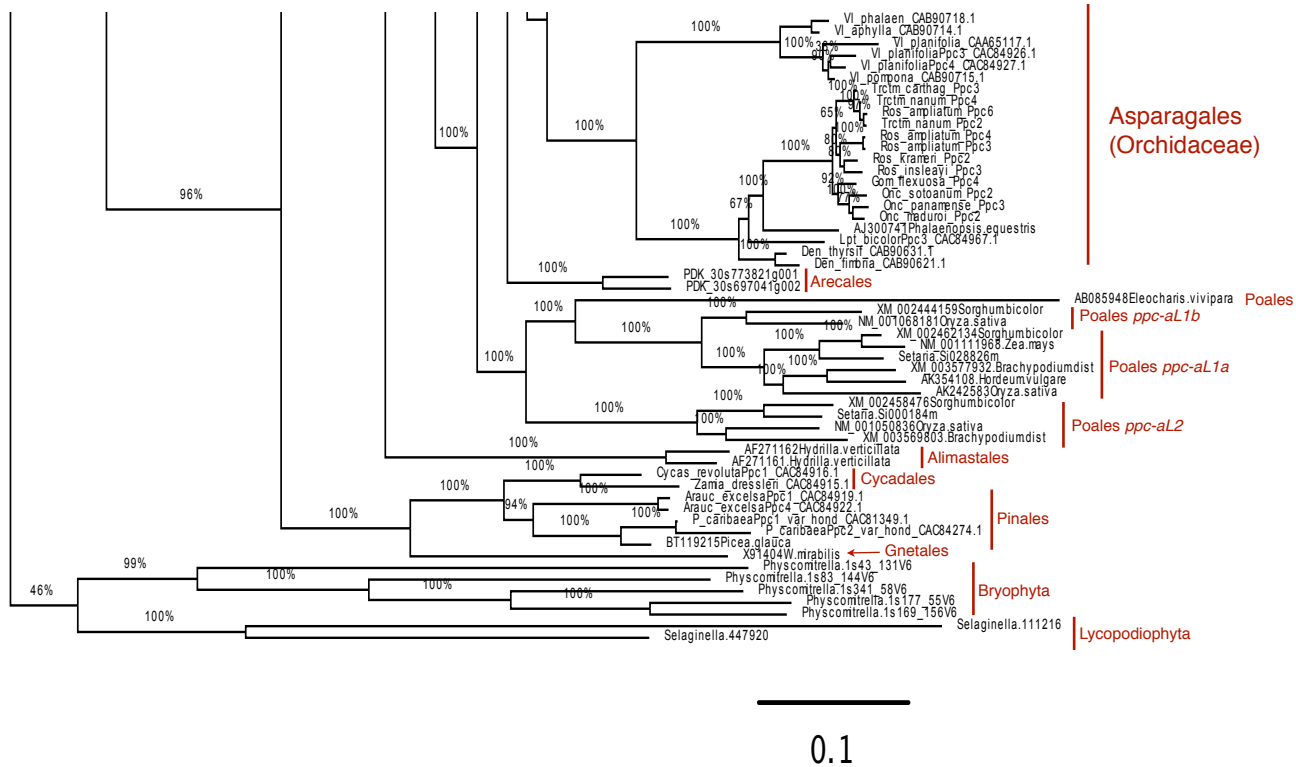
|          |                               |
|----------|-------------------------------|
| MAD1-F   | GGAGCTTGTGAAAGTAGCCAAGG       |
| MAD1-3R  | AATACAAAACACAGGAAGCGGGAG      |
| MAD2-F   | GAAGACCTAATAAAGGTTGCAAAAC     |
| MAD2-3R  | CAACTCCCTTCTCTCAATTATGTC      |
| MAD3F    | TTGGCAGTTGTACAAGGCTCAGGAGG    |
| PAN1-F   | GCTGGCCGTCTTTCTGCAGC          |
| PAN1-2R  | CAGTATCTCCCAATCTTTATTTAATATG  |
| PAN2F    | TTCGTGTTACCGTGCAGGGTGAAGTTATA |
| PAN3-F   | GAAGACCTAATAAAGGTTGCAAAAC     |
| PAN3-R   | CAGAAGTTTCAGCATCATTTTAACAC    |
| AMP1-F   | GAGCTTGTGGAAGTAGCCAAGC        |
| AMP2-F   | GAGCTTGTGAAAGTAGCCAAGG        |
| AMP1-2R  | TTGAACGTACATCACATTGAATGC      |
| AMP3-F   | CCATCTAAACGAAAACCTAGTGGT      |
| AMP4-F   | CCATCTAAACGAAAACCTAGCGGA      |
| AMP3-4-F | GAAGACCTAATAAAGGTTGCAAAAC     |
| AMP3-4R  | AGAAGCAAAGATAAAGCAAATC        |
| AMP3-5F  | GACGTCTTTCTGCAGTTGGCAGTTG     |
| AMP4-5F  | GACGTTTTTCTGCAGCTTGGCAGTTG    |
| AMP5R    | GATGCAAATTATAAGCACTTGGATGC    |





Saxifragales

Asparagales  
(Orchidaceae)



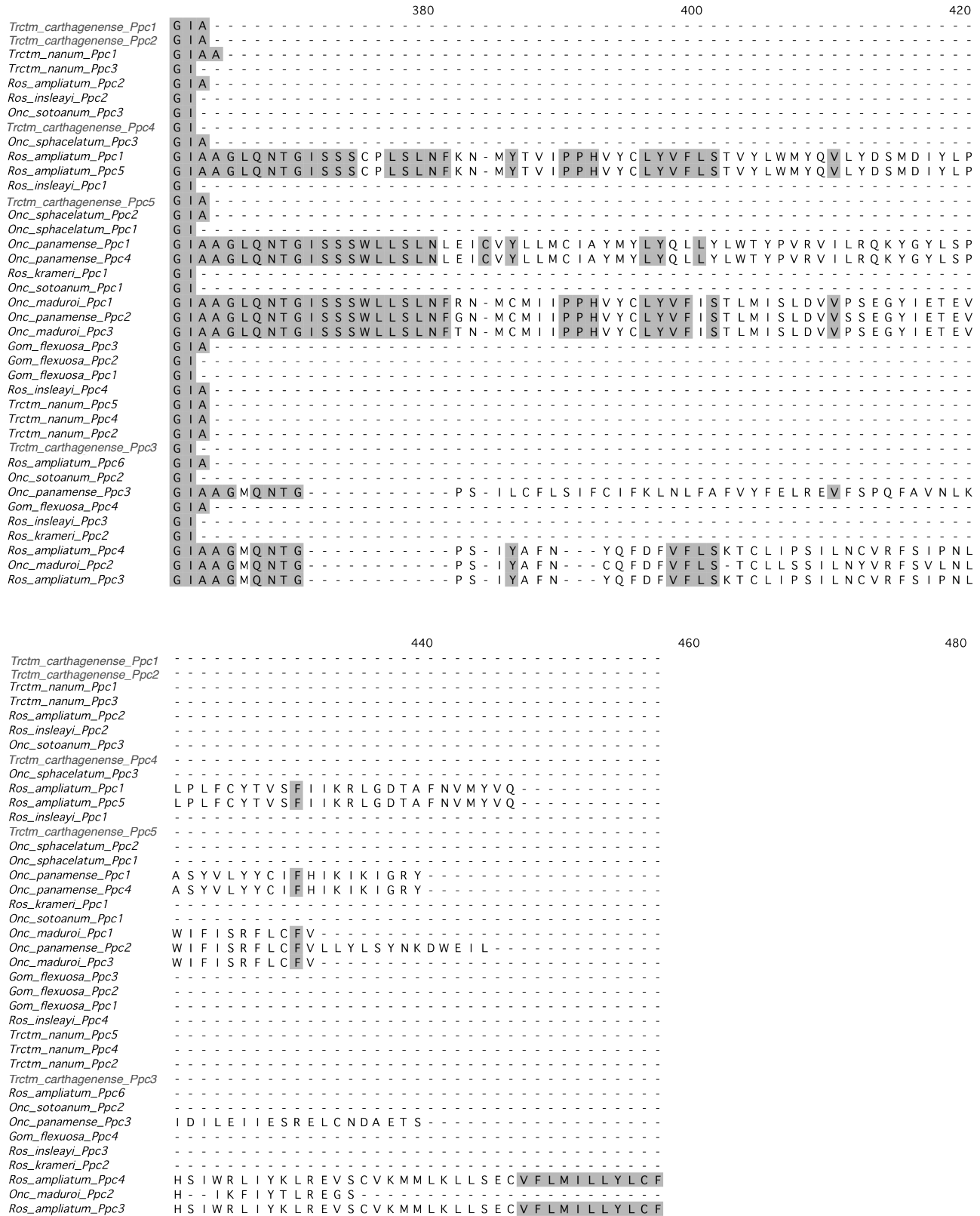
**Fig. S1** Phylogenetic tree of 312 *ppc-1* gene sequences. The tree was obtained through Bayesian analysis.











**Fig. S2** Protein sequence alignment for 28 PEPC isoforms, and 11 PEPC isoforms recovered by RACE for 10 species of the subtribe Oncidiinae