

# Supplementary Materials: Transcriptomic Analysis of Leaf in Tree Peony Reveals Differentially Expressed Pigments Genes

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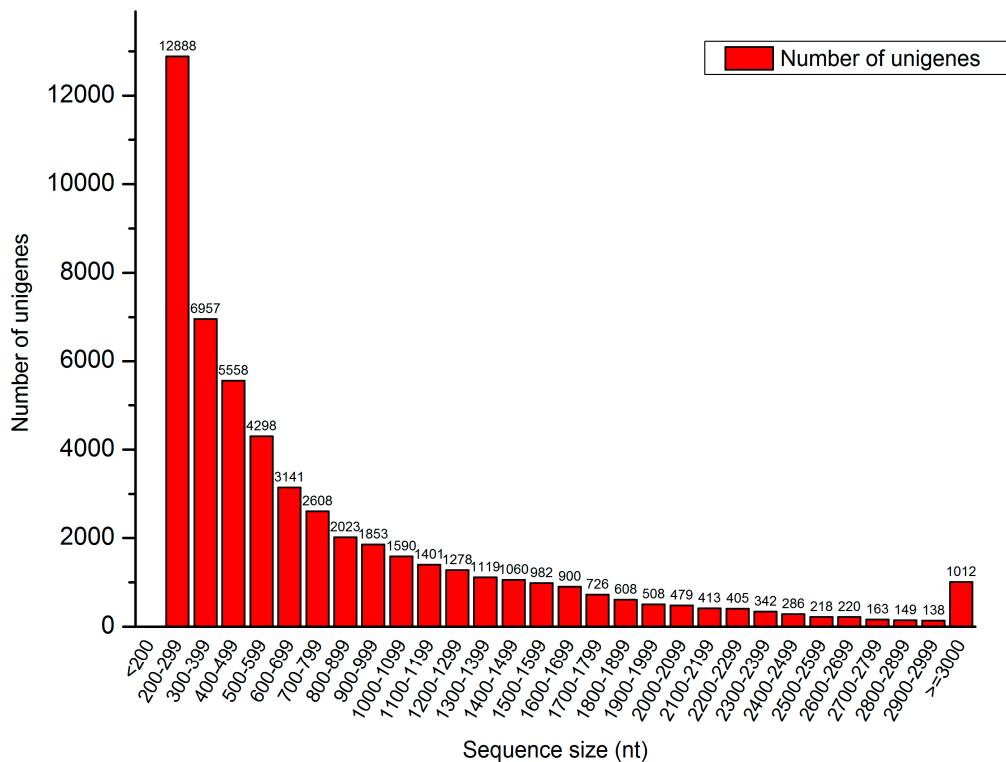


Figure S1. The size distribution of unigenes.

Table S1. KEGG pathway annotation of differentially expressed genes in S1 vs S3.

| No. | Pathway                                  | DEGs genes with pathway annotation (1436) | All genes with pathway annotation (5898) | Pvalue   | Qvalue   | Pathway ID |
|-----|--|---|--|----------|----------|------------|
| 1   | Ribosome                                 | 195 (13.58%)                              | 510 (8.65%)                              | 0        | 0        | ko03010    |
| 2   | DNA replication                          | 43 (2.99%)                                | 80 (1.36%)                               | 0        | 0.000001 | ko03030    |
| 3   | Photosynthesis                           | 33 (2.3%)                                 | 74 (1.25%)                               | 0.0001   | 0.004047 | ko00195    |
| 4   | Mismatch repair                          | 28 (1.95%)                                | 62 (1.05%)                               | 0.000256 | 0.007746 | ko03430    |
| 5   | Glycine, serine and threonine metabolism | 27 (1.88%)                                | 66 (1.12%)                               | 0.002079 | 0.046259 | ko00260    |
| 6   | Monoterpeneoid biosynthesis              | 8 (0.56%)                                 | 12 (0.2%)                                | 0.002294 | 0.046259 | ko00902    |
| 7   | Proteasome                               | 26 (1.81%)                                | 64 (1.09%)                               | 0.002803 | 0.048456 | ko03050    |
| 8   | Ribosome biogenesis in eukaryotes        | 53 (3.69%)                                | 157 (2.66%)                              | 0.004505 | 0.068133 | ko03008    |
| 9   | Base excision repair                     | 24 (1.67%)                                | 62 (1.05%)                               | 0.008179 | 0.105087 | ko03410    |
| 10  | Phenylpropanoid biosynthesis             | 44 (3.06%)                                | 130 (2.2%)                               | 0.008685 | 0.105087 | ko00940    |
| 11  | Spliceosome                              | 91 (6.34%)                                | 302 (5.12%)                              | 0.010883 | 0.119711 | ko03040    |

|    |   |            |             |          |          |         |
|----|---|------------|-------------|----------|----------|---------|
| 12 | Homologous recombination                              | 27 (1.88%) | 75 (1.27%)  | 0.015394 | 0.155219 | ko03440 |
| 13 | Limonene and pinene degradation                       | 6 (0.42%)  | 10 (0.17%)  | 0.017222 | 0.160293 | ko00903 |
| 14 | Alanine, aspartate and glutamate metabolism           | 18 (1.25%) | 47 (0.8%)   | 0.023012 | 0.182775 | ko00250 |
| 15 | Brassinosteroid biosynthesis                          | 8 (0.56%)  | 16 (0.27%)  | 0.023092 | 0.182775 | ko00905 |
| 16 | Photosynthesis - antenna proteins                     | 9 (0.63%)  | 19 (0.32%)  | 0.024169 | 0.182775 | ko00196 |
| 17 | Nucleotide excision repair                            | 31 (2.16%) | 92 (1.56%)  | 0.026538 | 0.188889 | ko03420 |
| 18 | Glyoxylate and dicarboxylate metabolism               | 29 (2.02%) | 86 (1.46%)  | 0.030894 | 0.207678 | ko00630 |
| 19 | Cutin, suberine and wax biosynthesis                  | 15 (1.04%) | 39 (0.66%)  | 0.034881 | 0.22214  | ko00073 |
| 20 | Nitrogen metabolism                                   | 10 (0.7%)  | 24 (0.41%)  | 0.046105 | 0.269627 | ko00910 |
| 21 | Carbon fixation in photosynthetic organisms           | 31 (2.16%) | 96 (1.63%)  | 0.046795 | 0.269627 | ko00710 |
| 22 | Purine metabolism                                     | 62 (4.32%) | 212 (3.59%) | 0.055633 | 0.305983 | ko00230 |
| 23 | Biotin metabolism                                     | 10 (0.7%)  | 25 (0.42%)  | 0.060611 | 0.306626 | ko00780 |
| 24 | Cysteine and methionine metabolism                    | 37 (2.58%) | 120 (2.03%) | 0.061388 | 0.306626 | ko00270 |
| 25 | Pyrimidine metabolism                                 | 48 (3.34%) | 161 (2.73%) | 0.063352 | 0.306626 | ko00240 |
| 26 | Pentose and glucuronate interconversions              | 21 (1.46%) | 63 (1.07%)  | 0.06723  | 0.31288  | ko00040 |
| 27 | Isoflavonoid biosynthesis                             | 3 (0.21%)  | 5 (0.08%)   | 0.096663 | 0.433192 | ko00943 |
| 28 | Cyanoamino acid metabolism                            | 21 (1.46%) | 66 (1.12%)  | 0.10282  | 0.441518 | ko00460 |
| 29 | Stilbenoid, diarylheptanoid and gingerol biosynthesis | 9 (0.63%)  | 24 (0.41%)  | 0.105818 | 0.441518 | ko00945 |
| 30 | Carotenoid biosynthesis                               | 13 (0.91%) | 38 (0.64%)  | 0.111346 | 0.449096 | ko00906 |
| 31 | Glycolysis / Gluconeogenesis                          | 42 (2.92%) | 147 (2.49%) | 0.133893 | 0.522614 | ko00010 |
| 32 | Starch and sucrose metabolism                         | 63 (4.39%) | 229 (3.88%) | 0.145001 | 0.548287 | ko00500 |
| 33 | Monobactam biosynthesis                               | 4 (0.28%)  | 9 (0.15%)   | 0.153581 | 0.5594   | ko00261 |
| 34 | Histidine metabolism                                  | 7 (0.49%)  | 19 (0.32%)  | 0.157187 | 0.5594   | ko00340 |
| 35 | Circadian rhythm - plant                              | 14 (0.97%) | 44 (0.75%)  | 0.162328 | 0.56119  | ko04712 |
| 36 | Vitamin B6 metabolism                                 | 6 (0.42%)  | 16 (0.27%)  | 0.172499 | 0.579788 | ko00750 |
| 37 | Fatty acid elongation                                 | 11 (0.77%) | 34 (0.58%)  | 0.184534 | 0.603477 | ko00062 |
| 38 | Degradation of aromatic compounds                     | 5 (0.35%)  | 13 (0.22%)  | 0.189681 | 0.603983 | ko01220 |
| 39 | One carbon pool by folate                             | 8 (0.56%)  | 24 (0.41%)  | 0.210419 | 0.652838 | ko00670 |
| 40 | Valine, leucine and isoleucine degradation            | 17 (1.18%) | 58 (0.98%)  | 0.228682 | 0.691763 | ko00280 |
| 41 | Ascorbate and aldarate metabolism                     | 13 (0.91%) | 44 (0.75%)  | 0.258604 | 0.74642  | ko00053 |
| 42 | Flavonoid biosynthesis                                | 9 (0.63%)  | 29 (0.49%)  | 0.259088 | 0.74642  | ko00941 |
| 43 | Plant hormone signal transduction                     | 59 (4.11%) | 226 (3.83%) | 0.288474 | 0.811752 | ko04075 |
| 44 | Fructose and mannose metabolism                       | 23 (1.6%)  | 85 (1.44%)  | 0.317098 | 0.872018 | ko00051 |
| 45 | Biosynthesis of amino acids                           | 73 (5.08%) | 286 (4.85%) | 0.339454 | 0.896371 | ko01230 |
| 46 | RNA transport   | 69 (4.81%) | 270 (4.58%) | 0.340769 | 0.896371 | ko03013 |
| 47 | Anthocyanin biosynthesis                              | 2 (0.14%)  | 5 (0.08%)   | 0.353416 | 0.909857 | ko00942 |
| 48 | Tryptophan metabolism                                 | 8 (0.56%)  | 28 (0.47%)  | 0.368838 | 0.929779 | ko00380 |
| 49 | Lysine biosynthesis                                   | 5 (0.35%)  | 17 (0.29%)  | 0.401105 | 0.952562 | ko00300 |
| 50 | Zeatin biosynthesis                                   | 6 (0.42%)  | 21 (0.36%)  | 0.405649 | 0.952562 | ko00908 |
| 51 | Arginine biosynthesis                                 | 10 (0.7%)  | 37 (0.63%)  | 0.412908 | 0.952562 | ko00220 |

|    |  |            |             |          |          |         |
|----|--|------------|-------------|----------|----------|---------|
| 52 | Phenylalanine, tyrosine and tryptophan biosynthesis    | 14 (0.97%) | 53 (0.9%)   | 0.413913 | 0.952562 | ko00400 |
| 53 | Flavone and flavonol biosynthesis                      | 1 (0.07%)  | 2 (0.03%)   | 0.427697 | 0.952562 | ko00944 |
| 54 | Terpenoid backbone biosynthesis                        | 16 (1.11%) | 62 (1.05%)  | 0.442377 | 0.952562 | ko00900 |
| 55 | Pentose phosphate pathway                              | 16 (1.11%) | 62 (1.05%)  | 0.442377 | 0.952562 | ko00030 |
| 56 | Synthesis and degradation of ketone bodies             | 2 (0.14%)  | 6 (0.1%)    | 0.450553 | 0.952562 | ko00072 |
| 57 | Sesquiterpenoid and triterpenoid biosynthesis          | 5 (0.35%)  | 18 (0.31%)  | 0.455329 | 0.952562 | ko00909 |
| 58 | Phagosome  | 24 (1.67%) | 95 (1.61%)  | 0.4566   | 0.952562 | ko04145 |
| 59 | Biosynthesis of unsaturated fatty acids                | 13 (0.91%) | 51 (0.86%)  | 0.47809  | 0.980491 | ko01040 |
| 60 | Protein processing in endoplasmic reticulum            | 79 (5.5%)  | 322 (5.46%) | 0.490509 | 0.989192 | ko04141 |
| 61 | Galactose metabolism                                   | 18 (1.25%) | 74 (1.25%)  | 0.547051 | 1        | ko00052 |
| 62 | Lysine degradation                                     | 10 (0.7%)  | 41 (0.7%)   | 0.557764 | 1        | ko00310 |
| 63 | N-Glycan biosynthesis                                  | 12 (0.84%) | 50 (0.85%)  | 0.577537 | 1        | ko00510 |
| 64 | Tyrosine metabolism                                    | 14 (0.97%) | 59 (1%)     | 0.594491 | 1        | ko00350 |
| 65 | Carbon metabolism                                      | 78 (5.43%) | 327 (5.54%) | 0.606832 | 1        | ko01200 |
| 66 | Arginine and proline metabolism                        | 12 (0.84%) | 51 (0.86%)  | 0.608136 | 1        | ko00330 |
| 67 | Linoleic acid metabolism                               | 4 (0.28%)  | 17 (0.29%)  | 0.623798 | 1        | ko00591 |
| 68 | Nicotinate and nicotinamide metabolism                 | 5 (0.35%)  | 22 (0.37%)  | 0.651118 | 1        | ko00760 |
| 69 | Caffeine metabolism                                    | 1 (0.07%)  | 4 (0.07%)   | 0.672541 | 1        | ko00232 |
| 70 | Fatty acid biosynthesis                                | 11 (0.77%) | 49 (0.83%)  | 0.675484 | 1        | ko00061 |
| 71 | Fatty acid metabolism                                  | 22 (1.53%) | 98 (1.66%)  | 0.707545 | 1        | ko01212 |
| 72 | Diterpenoid biosynthesis                               | 4 (0.28%)  | 20 (0.34%)  | 0.754873 | 1        | ko00904 |
| 73 | Glutathione metabolism                                 | 20 (1.39%) | 92 (1.56%)  | 0.757861 | 1        | ko00480 |
| 74 | Fatty acid degradation                                 | 12 (0.84%) | 57 (0.97%)  | 0.765751 | 1        | ko00071 |
| 75 | Phenylalanine metabolism                               | 9 (0.63%)  | 45 (0.76%)  | 0.80188  | 1        | ko00360 |
| 76 | Tropane, piperidine and pyridine alkaloid biosynthesis | 5 (0.35%)  | 28 (0.47%)  | 0.84765  | 1        | ko00960 |
| 77 | Isoquinoline alkaloid biosynthesis                     | 5 (0.35%)  | 28 (0.47%)  | 0.84765  | 1        | ko00950 |
| 78 | Other glycan degradation                               | 6 (0.42%)  | 33 (0.56%)  | 0.849645 | 1        | ko00511 |
| 79 | Non-homologous end-joining                             | 3 (0.21%)  | 18 (0.31%)  | 0.851219 | 1        | ko03450 |
| 80 | Steroid biosynthesis                                   | 7 (0.49%)  | 38 (0.64%)  | 0.852729 | 1        | ko00100 |
| 81 | RNA polymerase   | 12 (0.84%) | 63 (1.07%)  | 0.873266 | 1        | ko03020 |
| 82 | Pyruvate metabolism                                    | 19 (1.32%) | 96 (1.63%)  | 0.880455 | 1        | ko00620 |
| 83 | Arachidonic acid metabolism                            | 3 (0.21%)  | 20 (0.34%)  | 0.898131 | 1        | ko00590 |
| 84 | Protein export   | 12 (0.84%) | 65 (1.1%)   | 0.898781 | 1        | ko03060 |
| 85 | Taurine and hypotaurine metabolism                     | 2 (0.14%)  | 15 (0.25%)  | 0.911593 | 1        | ko00430 |
| 86 | Plant-pathogen interaction                             | 49 (3.41%) | 235 (3.98%) | 0.913604 | 1        | ko04626 |
| 87 | Butanoate metabolism                                   | 3 (0.21%)  | 21 (0.36%)  | 0.916187 | 1        | ko00650 |
| 88 | Inositol phosphate metabolism                          | 13 (0.91%) | 72 (1.22%)  | 0.921633 | 1        | ko00562 |
| 89 | Phosphatidylinositol signaling system                  | 13 (0.91%) | 72 (1.22%)  | 0.921633 | 1        | ko04070 |
| 90 | Glycerolipid metabolism                                | 10 (0.7%)  | 58 (0.98%)  | 0.92704  | 1        | ko00561 |
| 91 | Sulfur metabolism                                      | 5 (0.35%)  | 33 (0.56%)  | 0.931635 | 1        | ko00920 |

|     |   |            |             |          |   |         |
|-----|---|------------|-------------|----------|---|---------|
| 92  | Regulation of autophagy                               | 8 (0.56%)  | 49 (0.83%)  | 0.936286 | 1 | ko04140 |
| 93  | Glycosphingolipid biosynthesis - globo series         | 1 (0.07%)  | 10 (0.17%)  | 0.938739 | 1 | ko00603 |
| 94  | 2-Oxocarboxylic acid metabolism                       | 11 (0.77%) | 66 (1.12%)  | 0.950982 | 1 | ko01210 |
| 95  | alpha-Linolenic acid metabolism                       | 9 (0.63%)  | 56 (0.95%)  | 0.951535 | 1 | ko00592 |
| 96  | Thiamine metabolism                                   | 2 (0.14%)  | 18 (0.31%)  | 0.955467 | 1 | ko00730 |
| 97  | beta-Alanine metabolism                               | 7 (0.49%)  | 47 (0.8%)   | 0.960616 | 1 | ko00410 |
| 98  | Amino sugar and nucleotide sugar metabolism           | 28 (1.95%) | 150 (2.54%) | 0.962091 | 1 | ko00520 |
| 99  | RNA degradation                                       | 34 (2.37%) | 179 (3.03%) | 0.965533 | 1 | ko03018 |
| 100 | Citrate cycle (TCA cycle)                             | 12 (0.84%) | 75 (1.27%)  | 0.971161 | 1 | ko00020 |
| 101 | Peroxisome  | 21 (1.46%) | 121 (2.05%) | 0.975849 | 1 | ko04146 |
| 102 | Valine, leucine and isoleucine biosynthesis           | 2 (0.14%)  | 21 (0.36%)  | 0.978031 | 1 | ko00290 |
| 103 | mRNA surveillance pathway                             | 31 (2.16%) | 170 (2.88%) | 0.978552 | 1 | ko03015 |
| 104 | Sphingolipid metabolism                               | 4 (0.28%)  | 34 (0.58%)  | 0.979818 | 1 | ko00600 |
| 105 | Selenocompound metabolism                             | 2 (0.14%)  | 23 (0.39%)  | 0.98641  | 1 | ko00450 |
| 106 | Aminoacyl-tRNA biosynthesis                           | 11 (0.77%) | 76 (1.29%)  | 0.988119 | 1 | ko00970 |
| 107 | Glycosaminoglycan degradation                         | 1 (0.07%)  | 16 (0.27%)  | 0.988562 | 1 | ko00531 |
| 108 | Propanoate metabolism                                 | 5 (0.35%)  | 44 (0.75%)  | 0.990827 | 1 | ko00640 |
| 109 | Folate biosynthesis                                   | 2 (0.14%)  | 25 (0.42%)  | 0.991644 | 1 | ko00790 |
| 110 | Ubiquinone and other terpenoid-quinone biosynthesis   | 6 (0.42%)  | 58 (0.98%)  | 0.998054 | 1 | ko00130 |
| 111 | Ether lipid metabolism                                | 4 (0.28%)  | 46 (0.78%)  | 0.998374 | 1 | ko00565 |
| 112 | Glycosylphosphatidylinositol(GPI)-anchor biosynthesis | 2 (0.14%)  | 32 (0.54%)  | 0.998533 | 1 | ko00563 |
| 113 | Pantothenate and CoA biosynthesis                     | 3 (0.21%)  | 41 (0.7%)   | 0.998964 | 1 | ko00770 |
| 114 | SNARE interactions in vesicular transport             | 4 (0.28%)  | 52 (0.88%)  | 0.999573 | 1 | ko04130 |
| 115 | Endocytosis   | 34 (2.37%) | 229 (3.88%) | 0.99988  | 1 | ko04144 |
| 116 | ABC transporters                                      | 8 (0.56%)  | 86 (1.46%)  | 0.99991  | 1 | ko02010 |
| 117 | Oxidative phosphorylation                             | 23 (1.6%)  | 173 (2.93%) | 0.999911 | 1 | ko00190 |
| 118 | Porphyrin and chlorophyll metabolism                  | 4 (0.28%)  | 63 (1.07%)  | 0.999966 | 1 | ko00860 |
| 119 | Ubiquitin mediated proteolysis                        | 31 (2.16%) | 224 (3.8%)  | 0.999973 | 1 | ko04120 |
| 120 | Glycerophospholipid metabolism                        | 11 (0.77%) | 116 (1.97%) | 0.99999  | 1 | ko00564 |
| 121 | Basal transcription factors                           | 3 (0.21%)  | 77 (1.31%)  | 1        | 1 | ko03022 |

**Table S2.** Primers used for qRT-PCR analysis.

| Genes            | Primer                |
|------------------|-----------------------|
| Unigene0004270-F | CACTTAGGAGCGAGCGACTT  |
| Unigene0004270-R | CGCAGGAGTGAGAGGGATTGG |
| Unigene0018412-F | GGAGTCCCGAAATAGGTTGC  |
| Unigene0018412-R | AACGGCTTGACACAAGGTC   |
| Unigene0013558-F | TGCCCACTCGTTGAACTCAT  |
| Unigene0013558-R | TGCTGCAAATGTTGCCTGTC  |
| Unigene0016440-F | TGCCTTACTTGGCCTCACG   |
| Unigene0016440-R | TTCACCTCCTGCAGTAGCCC  |
| Unigene0022913-F | AGCCAAATCCTGCTCGCTAA  |

|                  |                      |
|------------------|----------------------|
| Unigene0022913-R | TAAGAGCTTGGGTGGGCAAG |
| Unigene0023761-F | AGTCCAAGCTGCCGTAGTTC |
| Unigene0023761-R | AAGCAACTTGGTAACCCGGA |
| Unigene0025170-F | ATCGATGGTGTACGCAACT  |
| Unigene0025170-R | CATGATGGCGCTGAAATG   |
| Unigene0027491-F | TTATCCACCAACCTCCGAA  |
| Unigene0027491-R | ACGCCTTCCACAATGAACCA |
| Unigene0028008-F | CAACCAGCGAATACGCCAAG |
| Unigene0028008-R | CTTCCATGCCACCGACTTCT |
| Unigene0029992-F | TGCACTGGGCTAACCTTCA  |
| Unigene0029992-R | TGGGGGAGGCTTTCAATGT  |
| Unigene0030040-F | GCGGATCAGAGCAAGGCTAA |
| Unigene0030040-R | AAGGGACGACATGGTCACA  |
| Unigene0031269-F | TTTGTTGAGCCAAATGCC   |
| Unigene0031269-R | TCCTGCAGGTTACGTGACAA |
| Unigene0032398-F | ACACTTACGCCCTCTCT    |
| Unigene0032398-R | ACTACCCGTCCAAGCGTATG |
| Unigene0032549-F | TCTGTTTGGCGCTGGTTG   |
| Unigene0032549-R | GGACATTAGCAACGGTCCT  |
| Unigene0034354-F | CCGAAATGTCCTCAACCGGA |
| Unigene0034354-R | GCCACCCACCTGATCTGAA  |
| Unigene0036679-F | TGGAAAGCCTGGATGTGTCC |
| Unigene0036679-R | ATGCGCCCTTCATTACAGA  |
| Unigene0039699-F | GATTICCAGTCGGAGCACCA |
| Unigene0039699-R | TGCAAGTGCCACAGCATTTC |
| Unigene0040930-F | CAAATGCACTGGCAAGGGTC |
| Unigene0040930-R | TGAATTGTCGGCTTGGGA   |
| Unigene0046696-F | CCATGTCACTCCAGCAGGTT |
| Unigene0046696-R | TGTGCGGAGAGTCGTTTCA  |
| Unigene0051809-F | GACGAGCTTCCACCAACAGA |
| Unigene0051809-R | ACTGTGGGTTAGGGCCATTG |
| Unigene0020903-F | GAAAAGGCCAGTTGCTTCGG |
| Unigene0020903-R | AGAAATGGAGTGGCACCTG  |

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