

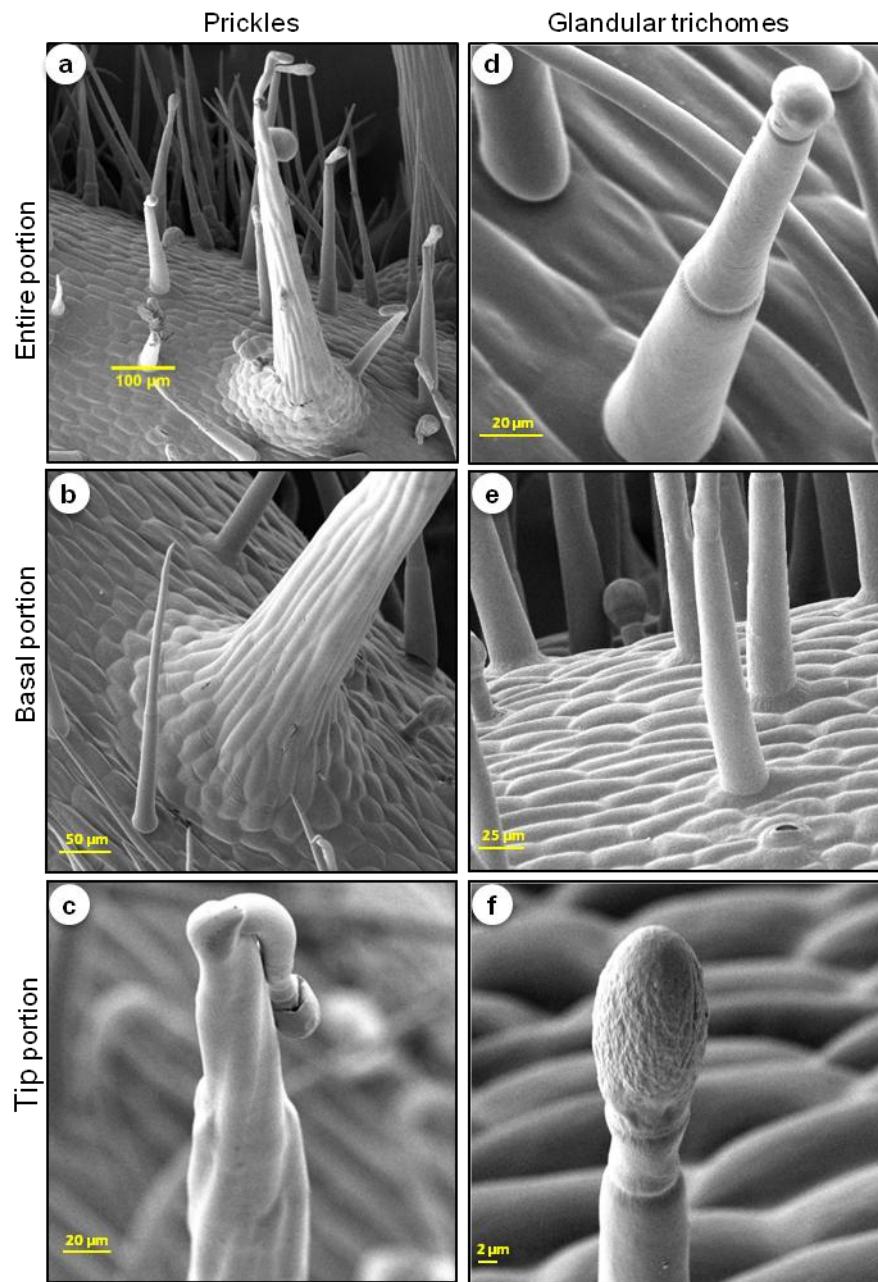
Transcriptome analysis provides insight into prickle development and its link to defense and secondary metabolism in *Solanum viarum* Dunal

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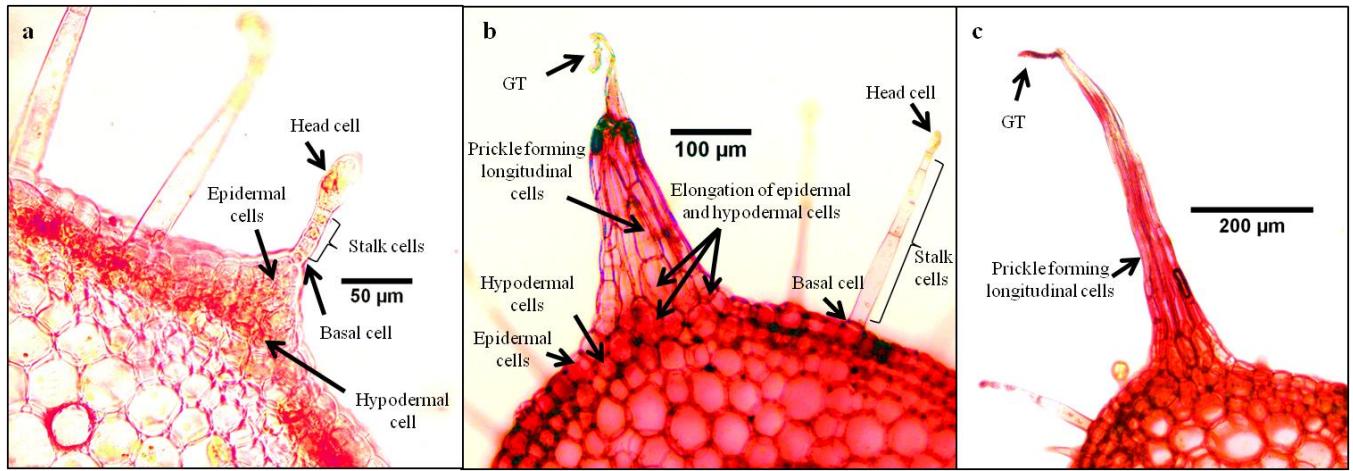
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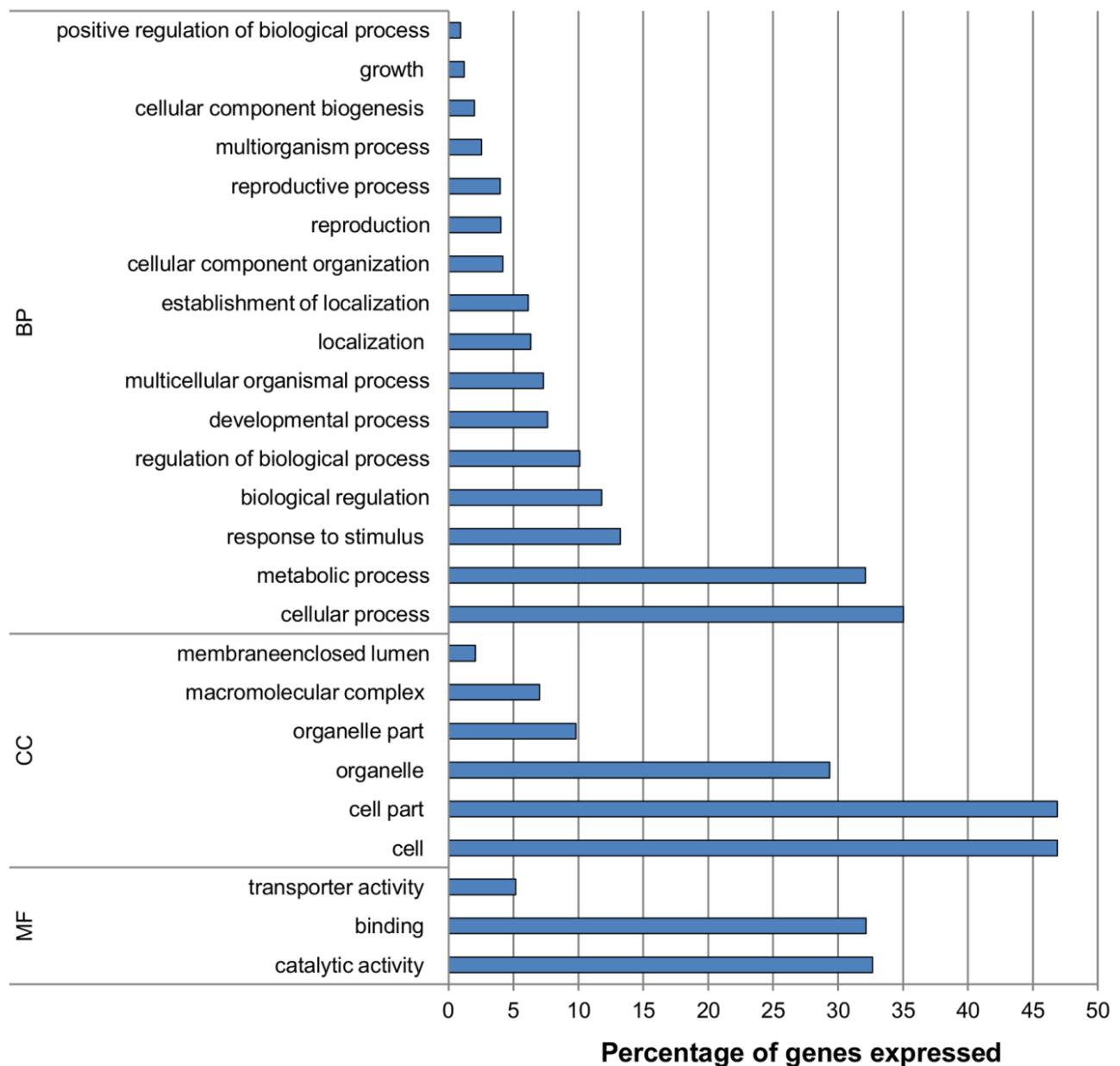
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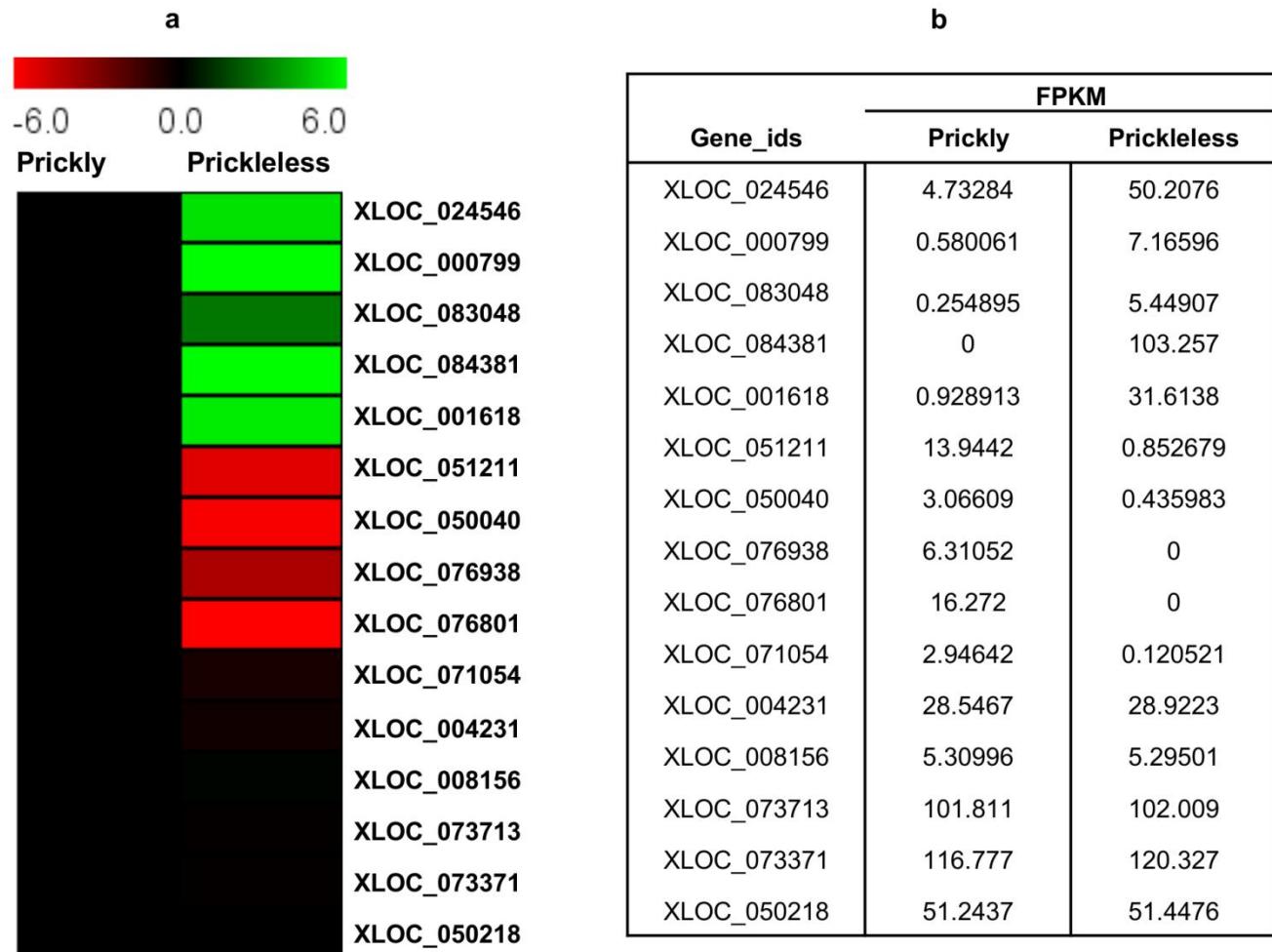
Supplementary Figure S1. Morphological similarity and differences between glandular prickle and glandular trichome of *S. viarum*. (a) Complete prickle. (b) Basal portion of prickle. (c) Tip portion of prickle showing GT attached to prickle. (d) Multicellular glandular trichome. (e) Base of trichomes. (f) Head of GTs.



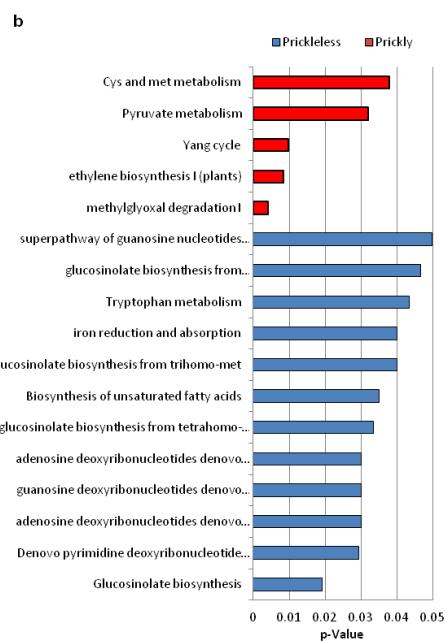
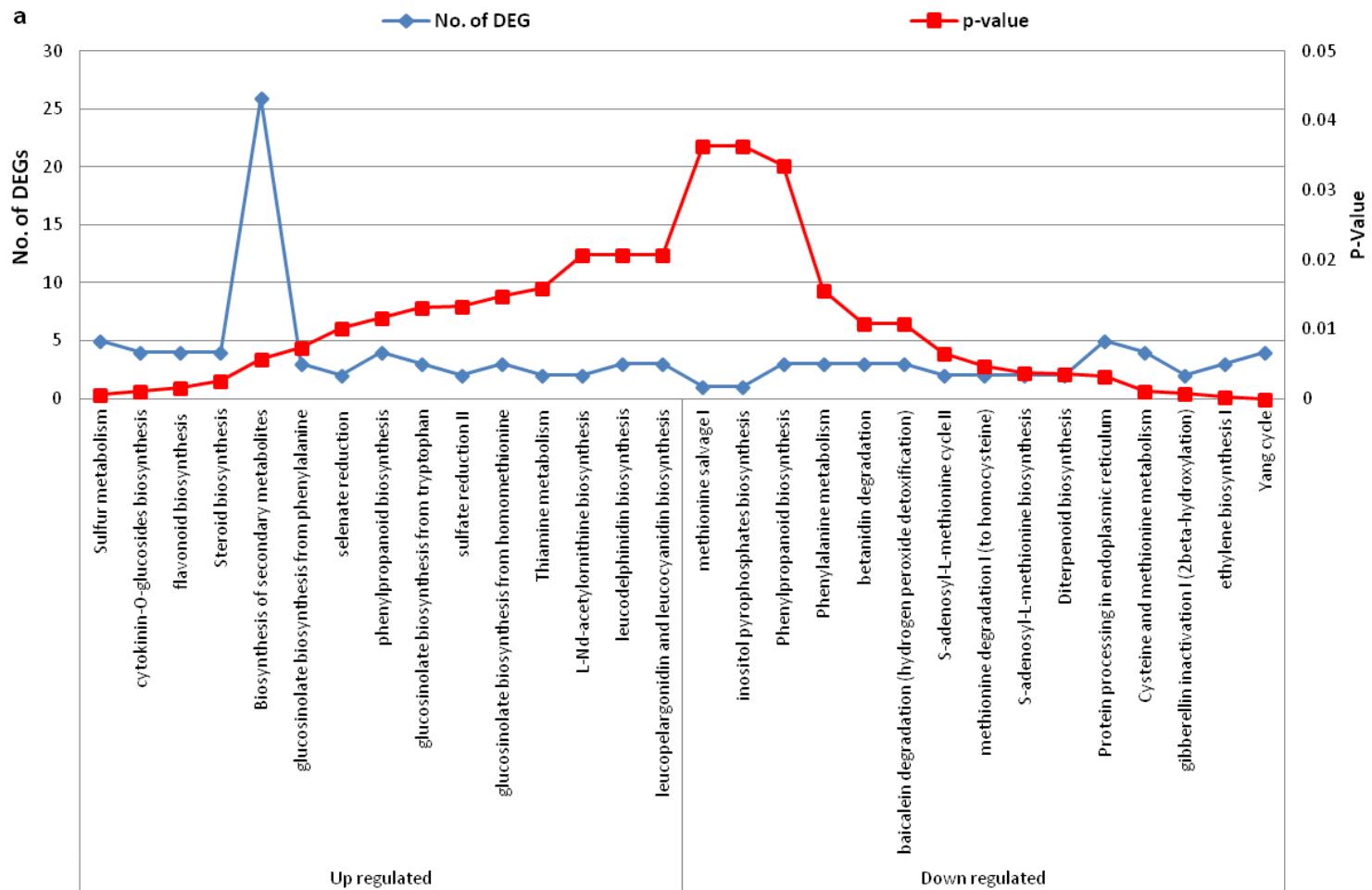
Supplementary Figure S2. T. S. of prickly stem showing different stages of prickle development in *S. viarum*. (a) Initiation of prickle formation beneath the basal cell of GT. (b) Developing prickle with GT attached to the tip. (c) Mature prickle with GT attached to the tip.



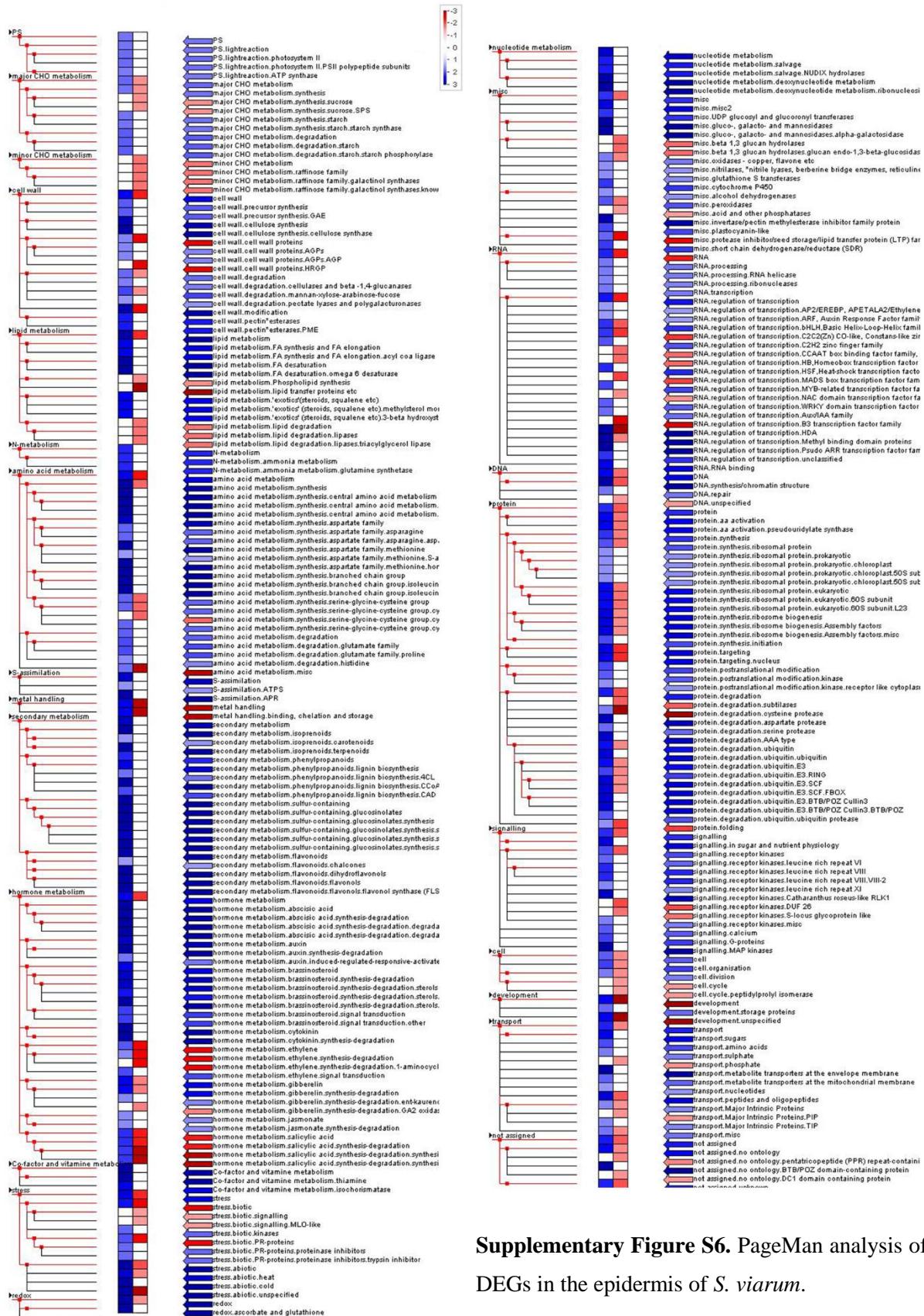
Supplementary Figure S3. Functional categorization of genes expressed in the epidermis of *S. viarum* through GO analysis.

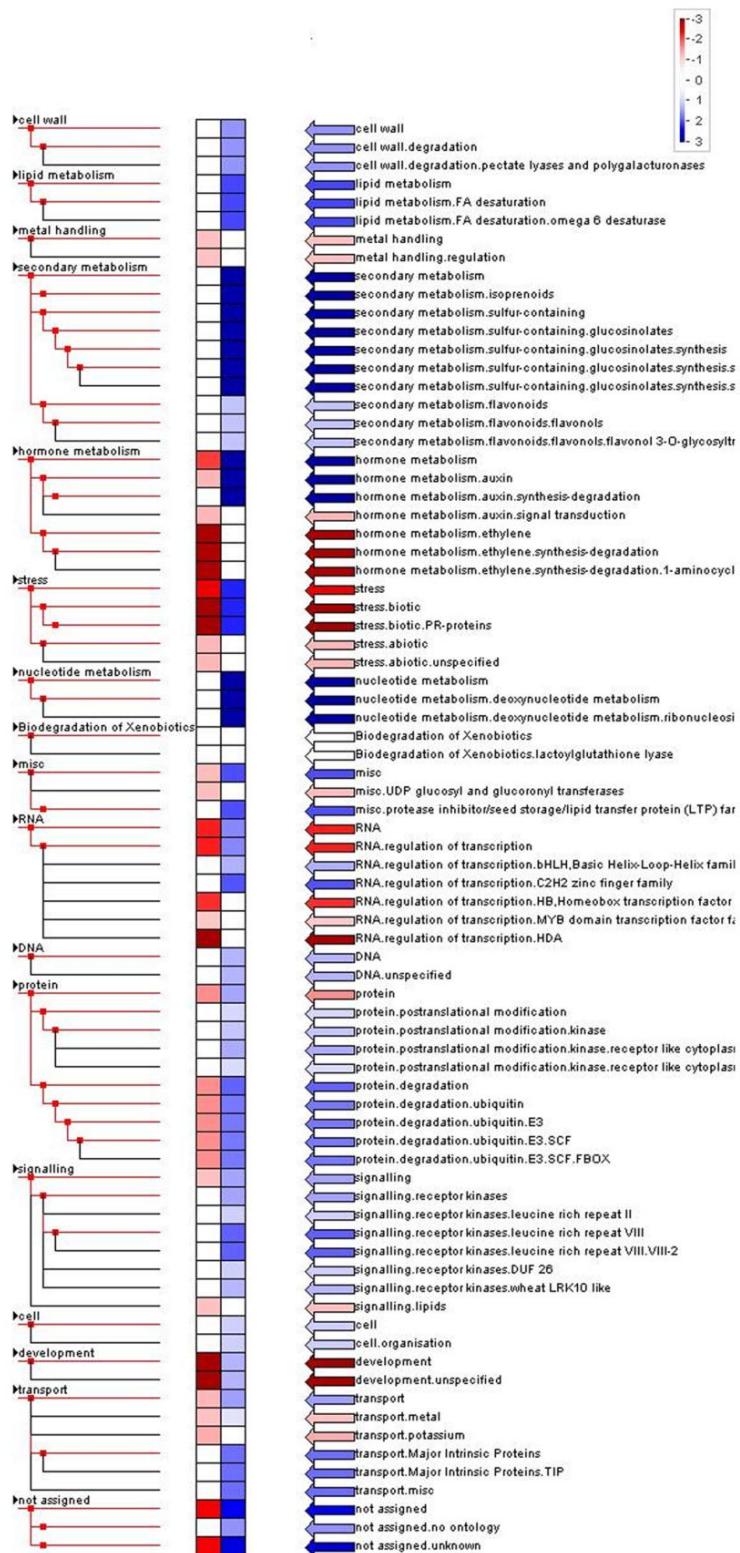


Supplementary Figure S4. qRT-PCR of some randomly selected highly up-regulated, down-regulated and similarly expressed genes for validation of transcriptome data of *S. viarum*. (a) Relative expression pattern analyzed through qRT-PCR. (b) FPKM based digital gene expression pattern analyzed through RNA-seq.

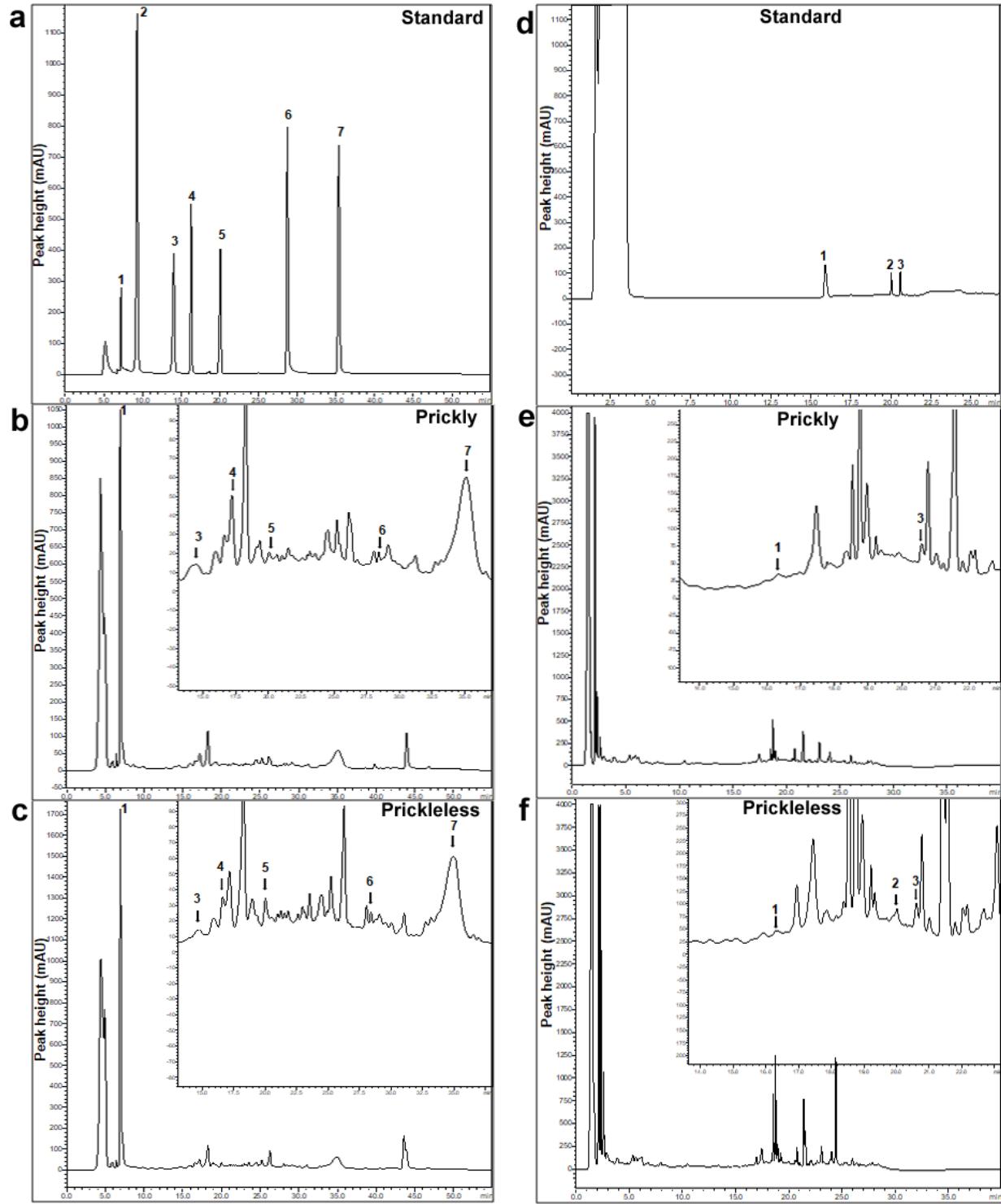


Supplementary Figure S5. Metabolic pathways altered in prickleless mutant of *S. viarum*. (a) Top 15 up and 15 down regulated pathways. (b) Uniquely expressed genes enriched to metabolic pathways significantly altered in prickly and prickleless mutant. Pathways with p-value ≤ 0.05 were considered statistically significant.





Supplementary Figure S7. PageMan analysis of uniquely expressed genes in epidermis of prickly or prickleless mutant of *S. viarum*.



Supplementary Figure S8. HPLC chromatogram of the standard and crude extract of prickly and prickleless *S. viarum*. a-c) phenolics and flavonoid [gallic acid -1, chlorogenic acid -2, caffic acid-3, rutin-4, ferulic acid-4, quercitin-6, kaempferol-7. (d-f) alkaloid [α -solanine-1, solanidine-2, solasodine-3].

Supplementary Table S1. Summary of the transcriptome sequencing, *de novo* assembly and annotation of *S. viarum*.

| | Prickly | | Prickleless | | | |
|---|-------------------|-------------------|-------------------|-------------------|--|--|
| | R1 | R2 | R1 | R2 | | |
| No. Of Reads | 18175222 | 15851932 | 17096127 | 13586099 | | |
| No. of HQ reads | 17444985 (95.98%) | 15153863 (95.60%) | 16429445 (96.10%) | 12992487 (95.63%) | | |
| No. of Contigs | 150880 | | 142861 | | | |
| Largest Contig (bp) | 59569 | | 46421 | | | |
| Smallest contig(bp) | 201 | | 201 | | | |
| Average Length (bp) | 1126.75 | | 1117.51 | | | |
| GC% | 46.11% | | 46.70% | | | |
| NR Annotated transcript | 73189 | | 70970 | | | |
| No. of trancripts after clustering | 156926 | | | | | |
| Largest Contig | 59569 | | | | | |
| Smallest contig | 201 | | | | | |
| Average Length | 1110.9 | | | | | |
| GC% | 39.30% | | | | | |
| TAIR Annotated transcripts | 62975 | | | | | |
| TAIR Unannotated transcripts | 93951 | | | | | |
| Tomato Annotated transcripts | 67752 | | | | | |
| Tomato Unannotated transcripts | 89174 | | | | | |

Supplementary Table S4. List of the top 15 up and 15 down-regulated genes in epidermis of prickly vs. prickleless mutant of *S. viarum*.

| Gene_Ids | Prickly vs. Prickleless (fold change) | p-value | Plant GD ID | Description | TAIR10 Ids | Tomato Ids |
|-------------|---|---------|--------------|--|-------------|--------------------|
| XLOC_064742 | -5.118538307 | 0.00005 | YCX91_PHAAO | Uncharacterized protein ORF91 | NA | NA |
| XLOC_079911 | -4.697050957 | 0.00005 | NA | NA | NA | NA |
| XLOC_066158 | -4.656741736 | 0.00005 | YCF1A_MARPO | Putative membrane protein ycf1 N-terminal part; AltName: ORF 1068; | NA | NA |
| XLOC_071054 | -4.611606642 | 0.00005 | C5XHE6_SORBI | Putative uncharacterized protein Sb03g011550 | NA | NA |
| XLOC_065300 | -4.596655786 | 0.00005 | Q40502_TOBAC | Extensin | NA | NA |
| XLOC_051211 | -4.031518631 | 0.00005 | B9HID6_POPTR | DEFORMED ROOTS AND LEAVES 1 family protein | AT1G13870.1 | Solyc08g008250.1.1 |
| XLOC_076801 | -4.024319679 | 0.00005 | F6HF70_VITVI | Putative uncharacterized protein | AT2G03440.1 | Solyc04g005480.1.1 |
| XLOC_067196 | -3.927747592 | 0.00015 | F6HH27_VITVI | Putative uncharacterized protein | AT3G22440.1 | Solyc11g071340.1.1 |
| XLOC_062280 | -3.897727126 | 0.00035 | Q0MX19_ARAH | | NA | NA |
| XLOC_085802 | -3.82662512 | 0.00005 | E3W9P7_TOBA | Putative PR-10 type pathogenesis-related protein | AT1G70890.1 | Solyc04g007010.2.1 |
| XLOC_065725 | -3.796462781 | 0.00005 | B3VI97_SOLDE | Putative disease resistance protein CC-NBS-LRR (Fragment) | AT3G14460.1 | Solyc08g007630.1.1 |
| XLOC_051029 | -3.684004197 | 0.0001 | Q93YD2_TOBA | Putative translocation transactivator | NA | Solyc08g044450.1.1 |
| XLOC_042610 | -3.682356673 | 0.00005 | Q39467_CAPAN | Sn-2 protein (MLP-like protein 34) | AT5G28010.1 | Solyc04g007770.2.1 |
| XLOC_024732 | -3.652715931 | 0.00005 | D7LTE8_ARAL | | NA | NA |
| XLOC_038231 | -3.596861021 | 0.00005 | E7D4U5_TOBA | Amino acid transporter, putative | AT5G45890.1 | Solyc02g076910.2.1 |
| XLOC_084381 | 6.690095778 | 0.00005 | F2CZR9_HORV | Predicted protein (Uncharacterized protein) | NA | NA |
| XLOC_007885 | 6.004369091 | 0.00005 | B9GNI9_POPTR | Uncharacterized protein | AT5G48850.1 | Solyc06g007970.2.1 |
| XLOC_031857 | 5.987186837 | 0.00005 | B9S545_RICCO | Putative uncharacterized protein | AT5G48850.1 | Solyc06g007970.2.1 |
| XLOC_002160 | 5.647052243 | 0.00095 | B3SP99_CAPAN | Fatty acid desaturase | AT3G12120.2 | Solyc01g006430.2.1 |
| XLOC_085535 | 5.481334466 | 0.00005 | PRS2_SOLTU | Pathogenesis-related protein STH-2 | NA | Solyc09g091000.2.1 |

| | | | | | | |
|-------------|-------------|---------|------------------|--|-------------|--------------------|
| XLOC_023238 | 5.201494901 | 0.00005 | A5BSN3_VITVI | Putative uncharacterized protein | AT1G22400.1 | Solyc04g074330.2.1 |
| XLOC_005604 | 5.10395771 | 0.00005 | Q9LM08_CAPA N | Pepper esterase | AT5G62180.1 | Solyc06g035520.2.1 |
| XLOC_001618 | 5.088867165 | 0.00005 | B9HUS6_POPTR | Homocysteine S-methyltransferase 3 family protein | AT3G63250.1 | Solyc09g082460.2.1 |
| XLOC_034413 | 5.07700713 | 0.0002 | B9RP37_RICCO | Putative uncharacterized protein | NA | NA |
| XLOC_018784 | 4.961102747 | 0.0003 | A2Q1J3_MEDT R | DUF506 family protein | AT1G12030.1 | Solyc11g012120.1.1 |
| XLOC_015461 | 4.911130691 | 0.00005 | D7MJF9_ARAL L | Putative uncharacterized protein | NA | NA |
| XLOC_030037 | 4.709704193 | 0.00005 | D5KXD2_SOLL C | Beta caryophyllene/humulene synthase (Caryophyllene/alpha-humulene synthase) | AT3G14540.1 | Solyc06g059930.2.1 |
| XLOC_019256 | 4.564999303 | 0.00005 | C6TGT6_SOYB N | Uncharacterized protein | AT3G63030.1 | Solyc06g084400.2.1 |
| XLOC_022696 | 4.457186326 | 0.00005 | B9TIC7_RICCO | Cytochrome P450, putative (EC 1.14.13.76) (Fragment) | NA | NA |
| XLOC_083048 | 4.418035045 | 0.0002 | P93789_SOLTU | UDP-galactose:solanidine galactosyltransferase | AT2G36790.1 | Solyc07g043490.1.1 |

Supplementary Table S6. List of the primers used for qRT-PCR analysis.

| Selected primers used for transcriptome data validation | | | | |
|--|-------------|----------------------------|--------------------------|------------------------|
| Sl. No. | Gene_Ids | Primer Sequence 5' to 3' | | |
| | | Forward | Reverse | |
| 1 | XLOC_024546 | CCATTCCACCAGCCCATCT | CGCCCACATCGTTGTTG | |
| 2 | XLOC_000799 | CAAAAAATGAGCACGTGAGAAAA | CGCCCCAAGCTAACTTCTT | |
| 3 | XLOC_083048 | CCAGCCTACCATTGATGACGA | TGGCAGTGTGAAGCTCTCA | |
| 4 | XLOC_084381 | GGAAAGCCGGTGACTCTCAA | TCGTCTGCTCTGCTTCGTT | |
| 5 | XLOC_001618 | AGGCAACTCTTCAGGGATT | TCTCTACGCTCTTTGAGCA | |
| 6 | XLOC_051211 | TGTTTGGCTAGACGACGAGG | ACGGTCTCCTCTCTCACGA | |
| 7 | XLOC_050040 | GCTTCATTGCGTGCTTGTG | TATGCCTTATCTCACCATCC | |
| 8 | XLOC_076938 | CGAAAGGAGATAGAGAACATGGAAGA | CCTTTCTAACTCCACCGTGTG | |
| 9 | XLOC_076801 | CGGTTGTTGTGGTAGA | ATTCGTCGGAAAAGCAGAG | |
| 10 | XLOC_071054 | TCAGGCATTGAGTTGTTGA | CTCGGGCATTGTTGTTTAC | |
| 11 | XLOC_004231 | TGCGAACGAGAGACCATAAGT | GGAGGACCCAGAACGGAATT | |
| 12 | XLOC_008156 | GGTGATTCTTGGTCTCCTTCC | CACGCTAAGGAGGCTGCAA | |
| 13 | XLOC_073713 | GCATTCAACCCATCAGAGCAA | TTGTCTCCCACCACTCTGTGA | |
| 14 | XLOC_073371 | CGGCTGTGGTGGTGAAGAG | TCTGGCAGGGCGTGTAC | |
| 15 | XLOC_050218 | CCACAGCGGGCAACAGTAA | TCTCCGTGGTGGTTTTAGGTT | |
| Selected primers used for the expression analysis of transcription factors | | | | |
| Sl. No. | Gene_Ids | Gene names | Forward | Reverse |
| 1 | XLOC_067399 | REM | TTGAAGCTGAATGGACATCG | CCCAATTGCACATTATGATCC |
| 2 | XLOC_040694 | MSM1 | TGGACTCATTGTTTTCTGCTACTG | CCCTTTCTGACTGCGTCTTGT |
| 3 | XLOC_050040 | C2C2-CO-like | GCTTCATTGCGTGCTTGTG | TATGCCTTATCTCACCATCC |
| 4 | XLOC_051211 | DRL1 | TGTTTGGCTAGAGCAGCAGG | ACGGTCTCCTCTCTCACGA |
| 5 | XLOC_027183 | Homeobox | TCTTCCATCTAGCTCGCTG | TGCTAAAGAGTTGGGTGTCC |
| 6 | XLOC_075558 | R2R3-MYB | ATGTGGTCATATTCCCTGGTTG | GGACCTCTCTCAATCCAACCT |
| 7 | XLOC_035637 | HAP2 | GACGAATGGAAGCATCACGA | TGCATACATCAGACTGCACAAC |
| Selected primers used for expression analysis of secondary metabolite pathways | | | | |
| 1 | XLOC_009213 | FLS1 | ATTTGTGAGCCCCCTTCCAC | TTGCACCCATTCTCCA |
| 2 | XLOC_034589 | CHS | GAAGGTGCAAGCCCAAGAA | GCCAAGCCAAGTCCAAAAT |
| 3 | XLOC_084597 | CFI | TTCCATGTCCCCTCAAAGGT | TGAAGTGGTGGTGGATGAAA |
| 4 | XLOC_025921 | COMT | CAAAAAGCGTGGTGGAGTT | TTTGGTTCTCCACGCAAT |
| 5 | XLOC_052049 | 4CL | GGCAAGGTCTCTCCACTTCA | TGAGGAAACAGACCGGTATGG |