**Table 4. Gene expression in day vs. night by functional categories in Sparkling Lake.**  We aggregated timepoints by day (9AM, 1PM, 5PM) and night (9PM, 1AM, 5AM) to compare differential gene expression. This analysis includes the top 20,000 most expressed genes. Functional categories were determined based on gene annotations – only categories with at least 10 expressed genes were tested. Significance (\*) was considered to be p < 0.05. Genes with cyclic trends were detected using RAIN (Thaben and Westermark, 2014), while p-values of day vs. night read totals per sample were calculated using a two-tailed t-test. The day/night ratio is the sum of reads assigned to that category in day divided by the sum at night. A ratio greater than one indicates higher expression in day, while a ratio less than one indicates higher expression at night.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
|  | Number of genes | % Genes more expressed in day | % Genes more expressed at night | % Cyclic genes (12 hr phase) | p-value from t-test of day vs. night read totals | Day/night ratio |
| Photosynthesis | 573 | 30.89 | 10.47 | 16.23 | 0.00\* | 2.76 |
| Opsins | 95 | 6.32 | 1.05 | 2.11 | 0.77 | 0.95 |
| RuBisCO | 97 | 0.00 | 1.03 | 0.00 | 0.08 | 0.66 |
| Polyamines | 23 | 4.35 | 13.04 | 0.00 | 0.18 | 0.80 |
| Alkaline phosphatase | 12 | 0.00 | 0.00 | 0.00 | 0.13 | 0.62 |
| Reactive oxygen species | 68 | 29.41 | 1.47 | 14.71 | 0.00\* | 1.87 |
| Protease | 278 | 10.79 | 0.00 | 2.16 | 0.14 | 1.46 |
| Carboxylate transport | 27 | 7.41 | 0.00 | 3.70 | 0.15 | 1.26 |
| Ribose transport | 13 | 0.00 | 7.69 | 0.00 | 0.07 | 0.69 |
| General sugar transport | 102 | 6.86 | 3.92 | 1.96 | 0.48 | 0.88 |
| Raffinose/stachyose/melibiose transport | 11 | 0.00 | 0.00 | 0.00 | 0.02\* | 0.53 |
| Glucose/mannose transport | 15 | 0.00 | 0.00 | 0.00 | 0.14 | 0.77 |
| Xylose transport | 17 | 0.00 | 0.00 | 0.00 | 0.28 | 0.76 |
| Fructose transport | 13 | 0.00 | 0.00 | 0.00 | 0.09 | 0.69 |
| Amino acid transport | 158 | 3.16 | 0 | 0.63 | 0.31 | 1.18 |