

**Fig. S1** Scan of Coomassie-stained 1-DE SDS gels of protein extracts from cultivation on 12C-galactose (A) and three-fold replicated cultivation on 13C-galactose, replicates I (B), II (C) and III (D), after different incubation times. Bands that were cut from the gel for further analysis are indicated.

**Table S1**: List of all proteins identified in the 12C samples at all time points.

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **Accession** | **Coverage** | **# Peptides** | **# AAs** | **MW [kDa]** | **calc. pI** | **Score** | **Description** |
|  |  |  |  |  |  |  |  |
| gi198282835 | 91.3 | 66 | 551 | 59 | 5.3 | 9472 | chaperonin GroEL [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198282766 | 63.5 | 24 | 471 | 52 | 5.3 | 1170 | glutamine synthetase, type I [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi121382 | 54.3 | 18 | 468 | 52 | 5.8 | 1022 | RecName: Full=Glutamine synthetase; AltName: Full=Glutamate--ammonia ligase |
| gi198284881 | 55.6 | 24 | 514 | 56 | 5.4 | 932 | ATP synthase F1, alpha subunit [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198284378 | 41.5 | 19 | 634 | 68 | 5.2 | 751 | chaperone protein DnaK [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198282178 | 23.1 | 10 | 499 | 53 | 9.1 | 350 | Pyrrolo-quinoline quinone [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198283506 | 14.0 | 9 | 766 | 79 | 9.2 | 287 | carboxysome structural protein CsoS2 [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198282694 | 18.5 | 9 | 493 | 54 | 4.9 | 277 | NusA antitermination factor [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198284705 | 17.4 | 8 | 570 | 63 | 5.3 | 256 | phosphoenolpyruvate-protein phosphotransferase [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198283027 | 20.0 | 9 | 581 | 64 | 6.4 | 253 | acetolactate synthase, large subunit, biosynthetic type [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198283505 | 22.1 | 9 | 533 | 59 | 7.0 | 231 | carboxysome shell carbonic anhydrase [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198284201 | 15.2 | 8 | 547 | 57 | 6.2 | 215 | putative mercuric reductase [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198282630 | 6.2 | 9 | 1362 | 152 | 5.6 | 178 | DNA-directed RNA polymerase, beta subunit [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198282634 | 8.0 | 4 | 699 | 77 | 5.2 | 175 | translation elongation factor G [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198283153 | 14.4 | 8 | 568 | 63 | 5.0 | 148 | ribosomal protein S1 [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198282605 | 13.0 | 8 | 784 | 88 | 6.3 | 146 | 5-methyltetrahydropteroyltriglutamate--homocysteine S-methyltransferase [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi218666536 | 5.3 | 3 | 722 | 78 | 6.2 | 146 | transketolase pyridine binding domain protein [Acidithiobacillus ferrooxidans ATCC 23270] |
| gi194239376 | 13.6 | 5 | 557 | 60 | 7.4 | 138 | sulfate adenylyltransferase [Acidithiobacillus ferrooxidans] |
| gi218666292 | 9.1 | 4 | 663 | 72 | 6.5 | 127 | transketolase [Acidithiobacillus ferrooxidans ATCC 23270] |
| gi198284388 | 6.0 | 3 | 567 | 61 | 5.1 | 115 | hypothetical protein Lferr\_2301 [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198283897 | 8.3 | 3 | 506 | 55 | 5.2 | 113 | Aldehyde dehydrogenase (NAD(+)) [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198283746 | 3.4 | 3 | 866 | 97 | 5.7 | 112 | ATP-dependent chaperone ClpB [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198283489 | 5.9 | 4 | 557 | 58 | 6.6 | 106 | dihydroxy-acid dehydratase [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198283477 | 4.6 | 3 | 825 | 92 | 7.8 | 102 | hypothetical protein Lferr\_1358 [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198283105 | 7.2 | 4 | 545 | 60 | 5.9 | 100 | CTP synthetase [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198284798 | 6.0 | 4 | 588 | 65 | 5.9 | 98 | flavodoxin/nitric oxide synthase [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198282947 | 5.7 | 3 | 558 | 59 | 6.4 | 95 | dihydroxy-acid dehydratase [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198283191 | 3.8 | 2 | 530 | 59 | 5.5 | 86 | ABC transporter related [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198282623 | 5.6 | 2 | 396 | 43 | 5.5 | 84 | elongation factor Tu [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198284876 | 3.6 | 2 | 611 | 66 | 6.5 | 78 | glucosamine/fructose-6-phosphate aminotransferase, isomerizing [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198283921 | 9.4 | 4 | 459 | 50 | 6.1 | 78 | ribulose bisphosphate carboxylase [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198282489 | 4.0 | 2 | 579 | 64 | 6.3 | 68 | arginyl-tRNA synthetase [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198283146 | 5.7 | 2 | 527 | 56 | 6.0 | 64 | D-3-phosphoglycerate dehydrogenase [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi198283699 | 4.2 | 2 | 572 | 62 | 5.7 | 58 | DNA polymerase III, subunits gamma and tau [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi218668112 | 2.6 | 3 | 1384 | 153 | 7.4 | 52 | DNA-directed RNA polymerase, beta' subunit [Acidithiobacillus ferrooxidans ATCC 23270] |
| gi198282456 | 3.4 | 2 | 612 | 65 | 8.3 | 51 | Putative lipoprotein-like protein [Acidithiobacillus ferrooxidans ATCC 53993] |
| gi218665890 | 4.2 | 2 | 762 | 78 | 9.2 | 46 | Tat (twin-arginine translocation) pathway signal sequence domain protein [Acidithiobacillus ferrooxidans ATCC 23270] |
| gi198282192 | 9.2 | 2 | 336 | 37 | 9.1 | 42 | putative sulfate-binding protein [Acidithiobacillus ferrooxidans ATCC 53993] |
|  |  |  |  |  |  |  |  |
| gi148261486 | 74.9 | 49 | 549 | 58 | 5.5 | 4172 | chaperonin GroEL [Acidiphilium cryptum JF-5] |
| gi148259108 | 57.5 | 29 | 593 | 64 | 5.8 | 1479 | dihydroxy-acid dehydratase [Acidiphilium cryptum JF-5] |
| gi148259766 | 40.9 | 25 | 575 | 63 | 5.3 | 810 | 30S ribosomal protein S1 [Acidiphilium cryptum JF-5] |
| gi148259090 | 25.5 | 9 | 522 | 56 | 6.2 | 477 | cell division protein FtsZ [Acidiphilium cryptum JF-5] |
| gi148259927 | 32.7 | 13 | 440 | 49 | 4.8 | 473 | trigger factor [Acidiphilium cryptum JF-5] |
| gi148261444 | 26.8 | 12 | 559 | 62 | 5.5 | 429 | putative ABC transporter ATP-binding protein [Acidiphilium cryptum JF-5] |
| gi148260643 | 18.4 | 9 | 635 | 68 | 5.1 | 400 | chaperone protein DnaK [Acidiphilium cryptum JF-5] |
| gi148260097 | 28.6 | 14 | 531 | 59 | 6.1 | 399 | isocitrate lyase [Acidiphilium cryptum JF-5] |
| gi148261802 | 35.9 | 10 | 449 | 48 | 5.0 | 361 | pyruvate dehydrogenase subunit beta [Acidiphilium cryptum JF-5] |
| gi148260236 | 23.0 | 8 | 544 | 57 | 6.4 | 331 | anthranilate phosphoribosyltransferase [Acidiphilium cryptum JF-5] |
| gi148259232 | 14.9 | 10 | 895 | 97 | 5.9 | 321 | aconitate hydratase [Acidiphilium cryptum JF-5] |
| gi148261454 | 16.6 | 6 | 567 | 60 | 6.8 | 317 | dihydroxy-acid dehydratase [Acidiphilium cryptum JF-5] |
| gi148259028 | 17.8 | 10 | 596 | 64 | 5.1 | 285 | acyl-CoA dehydrogenase domain-containing protein [Acidiphilium cryptum JF-5] |
| gi148262038 | 13.2 | 5 | 477 | 53 | 6.0 | 271 | glutamine synthetase, type I [Acidiphilium cryptum JF-5] |
| gi148259243 | 13.6 | 7 | 506 | 56 | 4.8 | 267 | transcription elongation factor NusA [Acidiphilium cryptum JF-5] |
| gi148260879 | 20.0 | 7 | 551 | 59 | 6.8 | 265 | phosphoenolpyruvate carboxykinase [Acidiphilium cryptum JF-5] |
| gi148260676 | 20.2 | 9 | 510 | 56 | 5.6 | 244 | F0F1 ATP synthase subunit alpha [Acidiphilium cryptum JF-5] |
| gi148260975 | 12.4 | 5 | 468 | 51 | 6.3 | 243 | phospho-2-dehydro-3-deoxyheptonate aldolase [Acidiphilium cryptum JF-5] |
| gi148260875 | 14.4 | 8 | 603 | 65 | 6.6 | 241 | succinate dehydrogenase flavoprotein subunit [Acidiphilium cryptum JF-5] |
| gi148259959 | 13.8 | 7 | 596 | 64 | 5.5 | 228 | arginyl-tRNA synthetase [Acidiphilium cryptum JF-5] |
| gi148259482 | 15.4 | 8 | 604 | 64 | 6.8 | 214 | phosphogluconate dehydratase [Acidiphilium cryptum JF-5] |
| gi148261029 | 8.5 | 6 | 821 | 87 | 7.4 | 209 | TonB-dependent receptor [Acidiphilium cryptum JF-5] |
| gi148260320 | 15.4 | 6 | 508 | 53 | 9.2 | 196 | protease Do [Acidiphilium cryptum JF-5] |
| gi148260379 | 2.6 | 2 | 544 | 57 | 5.4 | 195 | chaperonin GroEL [Acidiphilium cryptum JF-5] |
| gi148262049 | 6.0 | 4 | 882 | 96 | 5.8 | 186 | ATPase [Acidiphilium cryptum JF-5] |
| gi148262052 | 14.7 | 7 | 497 | 55 | 6.3 | 170 | hypothetical protein Acry\_3072 [Acidiphilium cryptum JF-5] |
| gi148259198 | 14.6 | 4 | 474 | 51 | 6.8 | 167 | pyruvate kinase [Acidiphilium cryptum JF-5] |
| gi148260340 | 11.1 | 5 | 548 | 60 | 6.6 | 166 | tartrate/fumarate subfamily Fe-S type hydro-lyase beta subunit [Acidiphilium cryptum JF-5] |
| gi148261780 | 7.4 | 2 | 557 | 59 | 8.7 | 162 | Pyrrolo-quinoline quinone [Acidiphilium cryptum JF-5] |
| gi148259579 | 16.2 | 4 | 519 | 55 | 6.5 | 157 | RND efflux system outer membrane lipoprotein [Acidiphilium cryptum JF-5] |
| gi148260620 | 16.3 | 4 | 410 | 44 | 6.1 | 139 | 2-oxoglutarate dehydrogenase, E2 subunit, dihydrolipoamide succinyltransferase [Acidiphilium cryptum JF-5] |
| gi148259640 | 8.6 | 5 | 573 | 62 | 6.4 | 134 | AMP-dependent synthetase and ligase [Acidiphilium cryptum JF-5] |
| gi148260961 | 7.5 | 4 | 770 | 82 | 5.7 | 133 | carbon-monoxide dehydrogenase (acceptor) [Acidiphilium cryptum JF-5] |
| gi148260979 | 8.2 | 4 | 583 | 62 | 6.0 | 132 | NAD synthetase [Acidiphilium cryptum JF-5] |
| gi148259097 | 8.1 | 4 | 801 | 85 | 6.2 | 129 | aldehyde oxidase and xanthine dehydrogenase, molybdopterin binding [Acidiphilium cryptum JF-5] |
| gi148260941 | 8.9 | 3 | 395 | 43 | 5.5 | 127 | elongation factor Tu [Acidiphilium cryptum JF-5] |
| gi148261174 | 6.3 | 6 | 888 | 96 | 5.5 | 124 | pyruvate phosphate dikinase [Acidiphilium cryptum JF-5] |
| gi148261633 | 10.8 | 5 | 528 | 56 | 6.1 | 123 | D-3-phosphoglycerate dehydrogenase [Acidiphilium cryptum JF-5] |
| gi148260621 | 3.9 | 4 | 949 | 104 | 6.2 | 120 | 2-oxoglutarate dehydrogenase, E1 subunit [Acidiphilium cryptum JF-5] |
| gi148243830 | 7.6 | 3 | 529 | 58 | 8.4 | 117 | CRISPR-associated Cse1 family protein [Acidiphilium cryptum JF-5] |
| gi148261447 | 11.3 | 4 | 506 | 53 | 5.7 | 107 | AMP-dependent synthetase and ligase [Acidiphilium cryptum JF-5] |
| gi148260223 | 7.5 | 3 | 677 | 72 | 7.2 | 105 | transketolase [Acidiphilium cryptum JF-5] |
| gi148260238 | 10.2 | 6 | 628 | 66 | 9.6 | 103 | hypothetical protein Acry\_1235 [Acidiphilium cryptum JF-5] |
| gi148261803 | 10.4 | 4 | 425 | 43 | 6.8 | 101 | pyruvate dehydrogenase complex dihydrolipoamide acetyltransferase [Acidiphilium cryptum JF-5] |
| gi148259867 | 6.8 | 3 | 503 | 55 | 7.3 | 100 | DEAD/DEAH box helicase domain-containing protein [Acidiphilium cryptum JF-5] |
| gi148261200 | 4.9 | 3 | 576 | 62 | 6.3 | 98 | dihydroxy-acid dehydratase [Acidiphilium cryptum JF-5] |
| gi148259386 | 4.5 | 3 | 717 | 77 | 5.5 | 94 | polynucleotide phosphorylase/polyadenylase [Acidiphilium cryptum JF-5] |
| gi148261275 | 3.9 | 2 | 644 | 71 | 6.0 | 92 | acetate--CoA ligase [Acidiphilium cryptum JF-5] |
| gi148260054 | 5.8 | 4 | 617 | 65 | 6.2 | 91 | dihydroxy-acid dehydratase [Acidiphilium cryptum JF-5] |
| gi148259064 | 4.2 | 2 | 575 | 61 | 8.9 | 89 | gluconate 2-dehydrogenase (acceptor) [Acidiphilium cryptum JF-5] |
| gi148260241 | 5.9 | 3 | 543 | 60 | 6.2 | 86 | CTP synthetase [Acidiphilium cryptum JF-5] |
| gi148261773 | 4.9 | 3 | 615 | 67 | 8.8 | 82 | Pyrrolo-quinoline quinone [Acidiphilium cryptum JF-5] |
| gi148259245 | 2.8 | 2 | 887 | 95 | 6.9 | 82 | translation initiation factor IF-2 [Acidiphilium cryptum JF-5] |
| gi148260944 | 2.2 | 3 | 1388 | 154 | 7.8 | 79 | DNA-directed RNA polymerase subunit beta' [Acidiphilium cryptum JF-5] |
| gi148261021 | 10.9 | 4 | 487 | 52 | 5.7 | 76 | isocitrate dehydrogenase [Acidiphilium cryptum JF-5] |
| gi148260274 | 3.2 | 2 | 594 | 67 | 5.6 | 71 | glycoside hydrolase 15-related protein [Acidiphilium cryptum JF-5] |
| gi148259675 | 7.3 | 3 | 548 | 59 | 6.1 | 59 | AMP-dependent synthetase and ligase [Acidiphilium cryptum JF-5] |
| gi148259274 | 6.0 | 3 | 567 | 61 | 5.9 | 58 | AMP-dependent synthetase and ligase [Acidiphilium cryptum JF-5] |
| gi148259266 | 6.2 | 4 | 695 | 77 | 5.4 | 53 | translation elongation factor G [Acidiphilium cryptum JF-5] |
| gi148261666 | 3.6 | 2 | 633 | 68 | 6.7 | 46 | hypothetical protein Acry\_2683 [Acidiphilium cryptum JF-5] |
| gi148260280 | 2.7 | 2 | 943 | 101 | 6.2 | 36 | bifunctional transaldolase/phosoglucose isomerase [Acidiphilium cryptum JF-5] |
| gi148260246 | 3.7 | 2 | 722 | 78 | 6.5 | 32 | malate synthase G [Acidiphilium cryptum JF-5] |
| gi148260060 | 4.3 | 2 | 417 | 45 | 8.1 | 22 | extracellular ligand-binding receptor [Acidiphilium cryptum JF-5] |
|  |  |  |  |  |  |  |  |