**Supplementary Material**

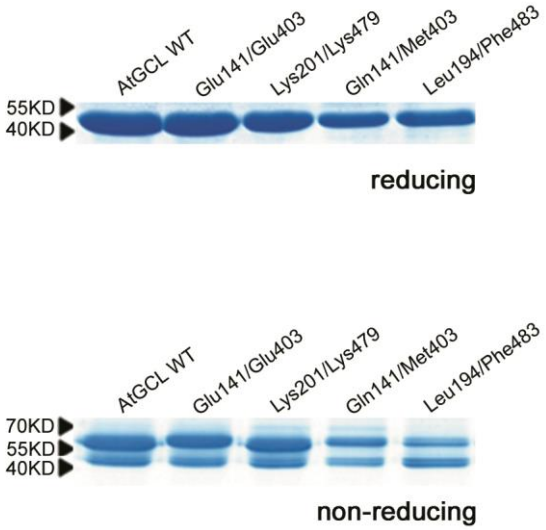


**Figure S1**

**Analysis of WT and mutant AtGCL proteins via reducing (top panel) and non-reducing (bottom panel) SDS-PAGE**

WT and mutated AtGCL proteins were recombinantly expressed in *E. coli* and after purification separated by SDS-PAGE. Under reducing conditions (+ßME) (top panel) each GCL protein is detected as a single band at ~ 50 kDa, whereas under non-reducing conditions (-ßME) (bottom panel) GCL proteins run as double bands, reflecting the oxidized (upper band) and reduced (lower band) status of the regulatory disulfide bridge. Proteins were stained with Coomassie blue. Arrowheads correspond to Mr markers.

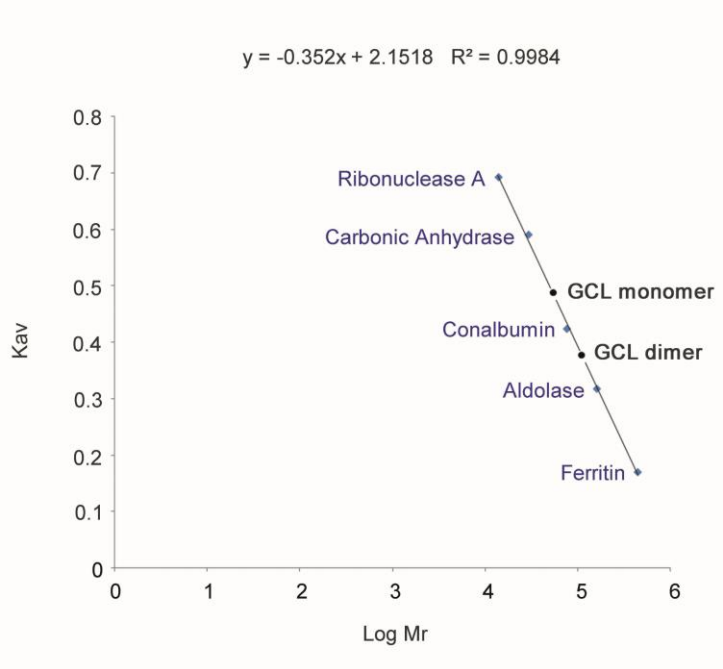


**Figure S2**

**Calibration curve used to estimate molecular weights for GCL monomer versus dimer.**

Size exclusion chromatography column HiLoad 16/600 Superdex 200 pg was calibrated using Ribonuclease A (13700 Da), Carbonic Anhydrase (29000 Da), Conalbumin (75000 Da), Aldolase (158000 Da), and Ferritin (440000 Da). Blue diamonds indicate the known calibration standards, black circles correspond to the positions of *K*av values for GCL dimer and monomer, the molecular weights of which were estimated to be around 106.000 Da and 51.000 Da, respectively.

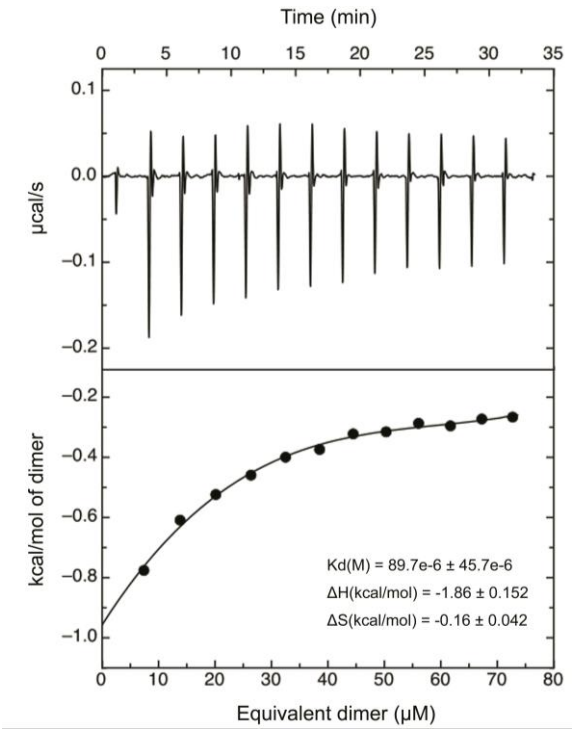


**Figure S3**

**Isothermal titration calorimetry analysis of recombinant AtGCL dissociation.**

Protein samples were dialyzed overnight in ITC buffer (see Materials and Methods). ITC experiments were performed in triplicate on a MicroCal PEAQ-ITC machine (Malvern Instruments). For determination of the dissociation constant ($K_D$) of *A. thaliana* GCL the syringe was loaded with GCL (450 µM) and titrated into the sample cell filled with ITC buffer. ITC data were processed using the MicroCal PEAQ-ITC Analysis Software (Malvern Instruments) and thermodynamic parameters were obtained by fitting the data to a dissociation model (for further details see Materials and Methods).

**Table S1**

Primer sequences

| | |
|---|---|
| P1 | 5'-CCGCTCGAGTTAGTACAGCAGCTCTTCGAACACGG-3' |
| P2 | 5'-CATGCCATGGCGGCAAGTCCTCCAACG-3' |
| P3 | 5'-CACACAGCCTTTCCCAGGGACCTC-3' |
| P4 | 5'-GAGGTCCCTGGGAAAGGCTGTGTG-3' |
| P5 | 5'-TCCCATTCAAATCTCTGAGCGATACC-3' |
| P6 | 5'-TCTTAATGGTATCGCTCAGAGATTTGAATG-3' |
| P7 | 5'-ACACAGCCTCATCCAGGGACCTC-3' |
| P8 | 5'-AGGTCCCTGGATGAGGCTGTGTGC-3' |
| P9 | 5'-CCAATTCCCATTTCCTTAGCAACTGCTTTTAC-3' |
| P10 | 5'-TCAGGTAAAAGCAGTTGCTAAGGAAATGG-3' |
| P11 | 5'-GCTTTTACCTGCAAAAGATGTGAATTGACT-3' |
| P12 | 5'-GAAGTCAATTCACATCTTTTGCAGGT-3' |
| P13 | 5'-CTTTGCATCAAACTTCTGCTGAAGTCAATTC-3' |
| P14 | 5'-GAATTGACTTCAGCAGAAGTTTGATGCAAAG-3' |

* Note that the primers used for cloning reporter gene constructs contained restriction sites and protective bases.

**Table S2**

**Data collection and refinement statistics**

| Data collection | GSM-GCL complex |
|---|---|
| PDB accession code | PDB ID 6GMO |
| Space group | P21 |
| Unit cell parameters (Å) | a = 58.77, b = 109.86, c = 84.75, β = 98.72 |
| X-ray source (Beamline) | ESRF (BM16) |
| Wavelength (Å) | 0.975 |
| Resolution range a (Å) | 19.89 - 1.75 (1.80-1.75) |
| No. of unique reflections | 105,763 (7,902) |
| Multiplicity | 3.72 (3.09) |
| I/σ(I) | 14.09 (2.57) |
| Rsym (%)a | 8.2 (53.8) |
| Completeness (%) | 98.9 (91.4) |
| Refinement | |
| Resolution range (Å) | 19.72 - 1.75 (1.77-1.75) |
| Rwork (%)b/ Rfree (%)c | 16.1 / 19.3 |
| r.m.s.d. bonds (Å) | 0.006 |
| r.m.s.d. angles (°) | 1.072 |
| Average B factor (Å2)/ No. of atoms | 19.65 / 8476 |
| Protein | 17.7 / 7179 |
| GSM and Mg2+ | 34.0 / 44 |
| water | 31.0 / 1233 |

a Values in parentheses are for the highest resolution shell

b as defined in XDS [17]

c as defined in Refmac5 [20]