Inorganic carbon acquisition and photorespiratory 2-phosphoglycolate metabolism in cyanobacteria

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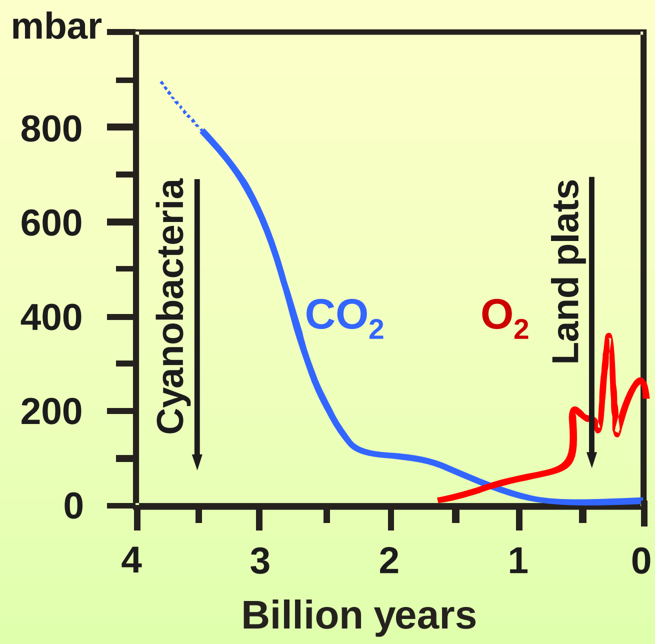
Inorganic carbon evolved as limiting factor
Rubisco: an enzyme with low affinity and specificity to CO_2

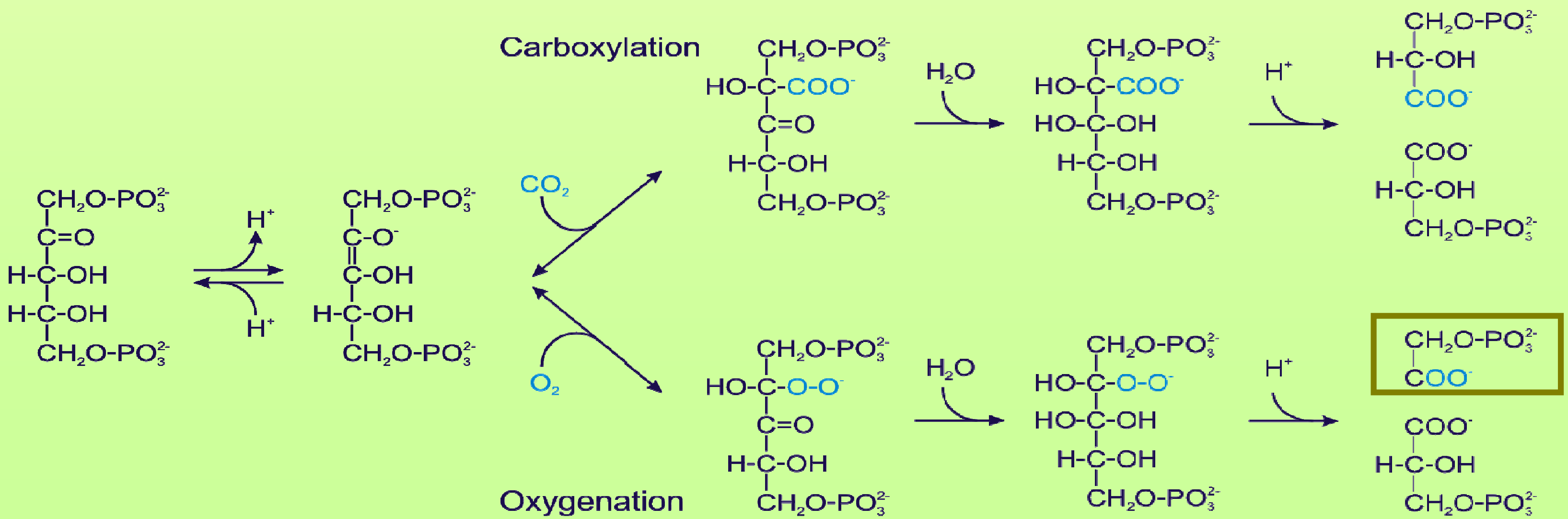
1. Problem: low affinity for CO_2

	Km (CO_2) [µM]	Reference
Spinacea oleracea	21 ± 1	Spreitzer et al. 2005
Anabaena variablis	293 ± 27	Badger 1980

→ high content of Rubisco compensates in higher plants (up to 30% leaf protein)



2. Problem: O_2 as competitive substrate



Phosphoglycolate (2-PG) or photorespiratory cycle in plants

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Metabolic pathway transforming 2-phosphoglycolate produced by oxygenase reaction of RubisCO to 3-phosphoglycerate

Functions:

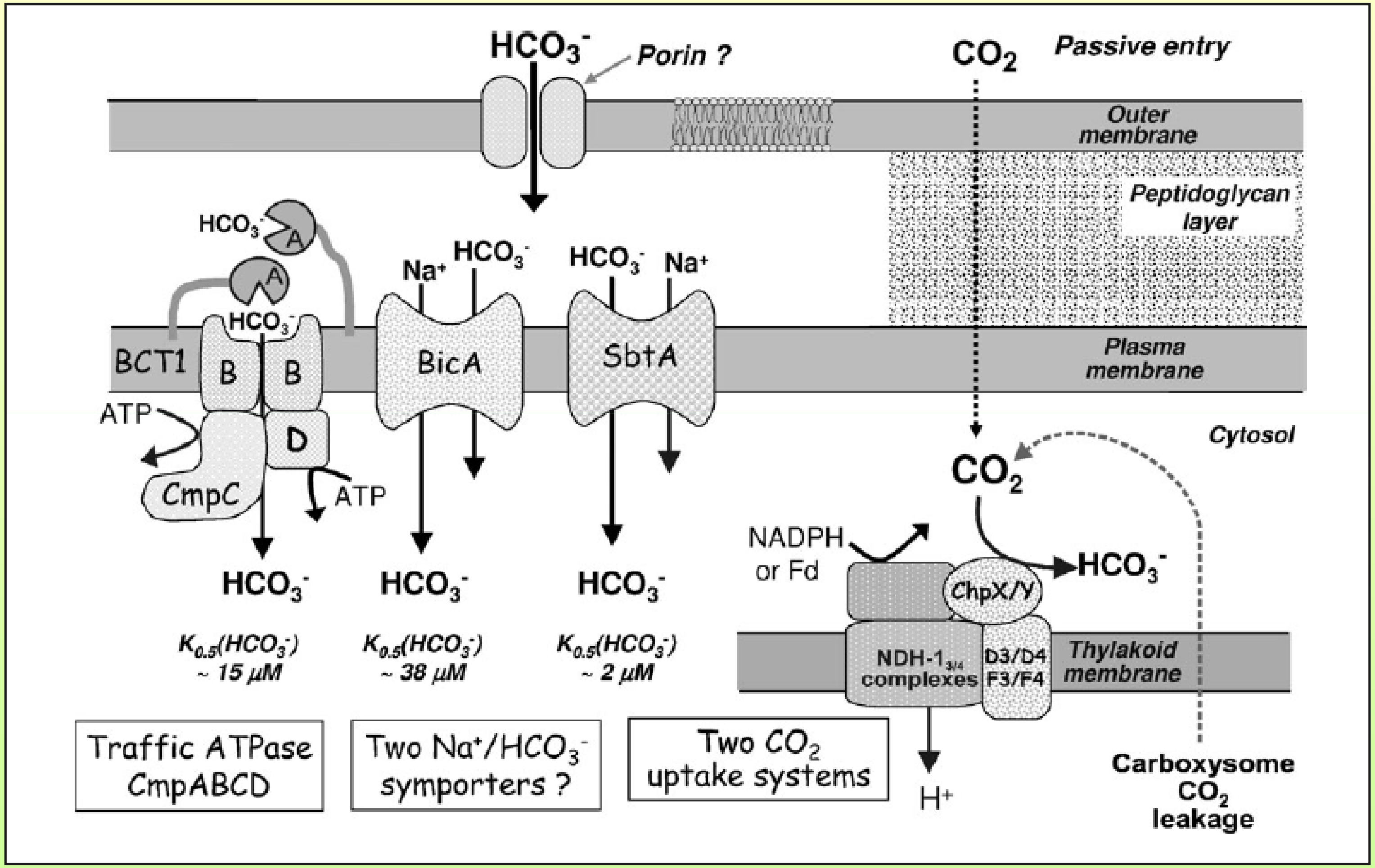
- Recycling of 75% of organic carbon from 2-PG
- Avoidance of accumulation of toxic intermediates
- Synthesis of intermediates
- Protection against high light

Problems:

- Loss of organic carbon
- Loss of energy

Photorespiratory plant mutants need high CO_2 – high CO_2-requiring phenotype

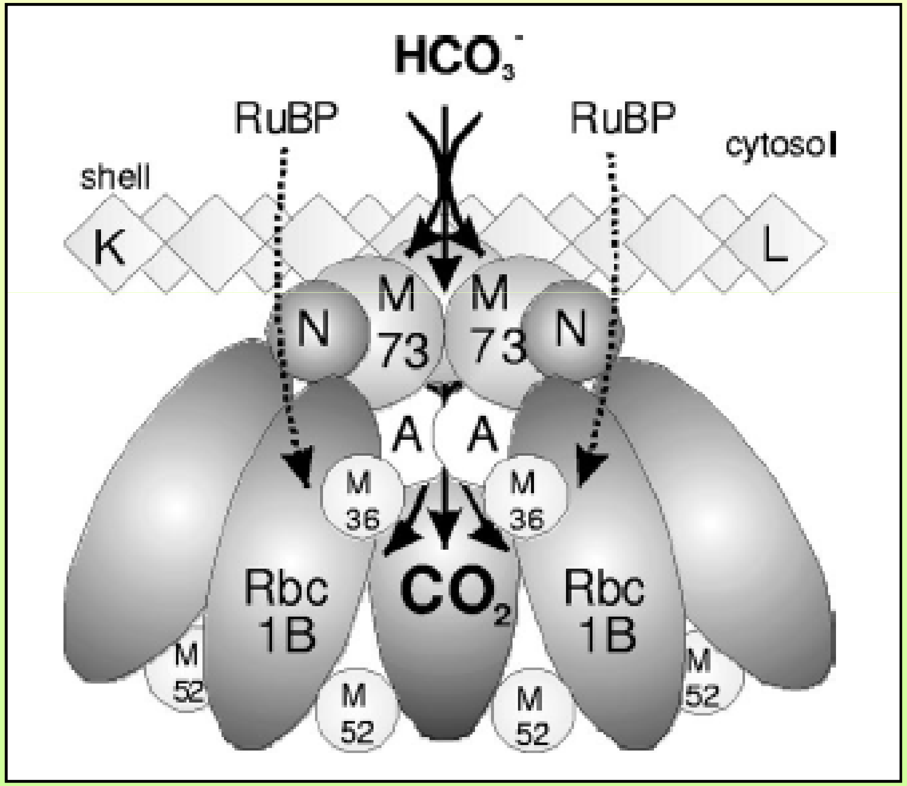
Cyanobacteria evolved a carbon concentrating mechanism (CCM) employing <u>C_i-uptake mechanisms</u> and carboxysomes

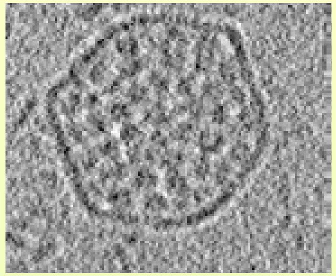


➡ Bicarbonate is concentrated inside the cell

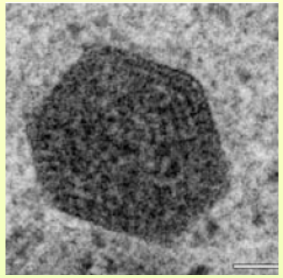
Cyanobacteria evolved a carbon concentrating mechanism (CCM) employing C_i-uptake mechanisms and <u>carboxysomes</u>

Model of the bicarbonate dehydrating complex inside the carboxysome (Cot, So & Espie, 2008)





Iancu et al. 2007



Kerfeld et al. 2005

Rubisco (Rbc) is concentrated in **carboxysomes**, where carbonic anhydrase (CcaA) releases high CO_2 amounts from HCO_3^-

➡ **CO_2 is released near RubisCO allowing efficient carboxylation, CCM mutants need high CO_2 – high CO_2-requiring phenotype**

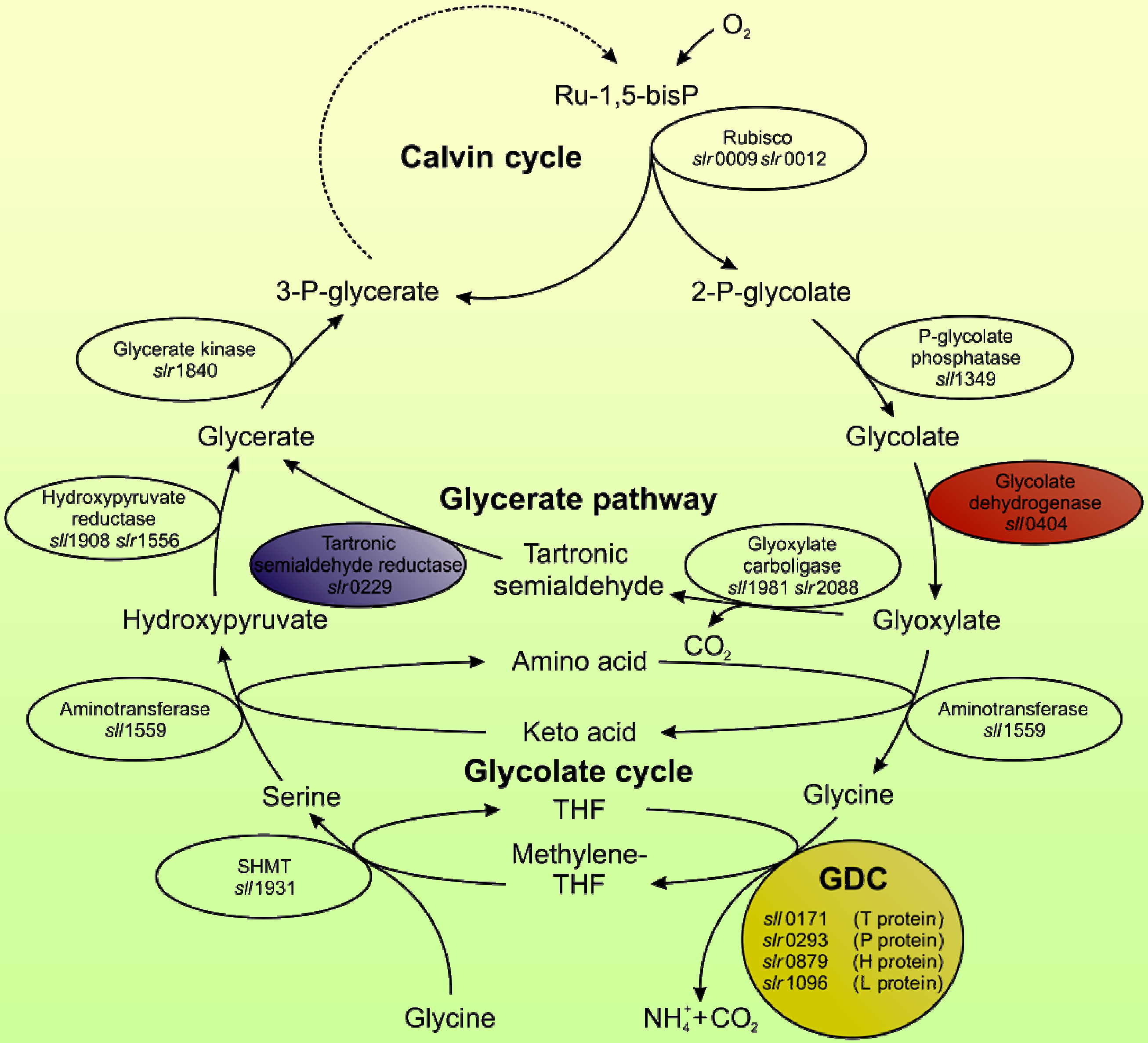
Search for 2-PG metabolizing proteins in *Synechocystis*: A mixture of plant and bacterial enzymes was found!

Sequence comparison of proteins participating in phosphoglycolate turnover with candidate proteins from *Synechocystis* sp. strain PCC 6803 using PSI- and PHI-BLAST (Altschul et al., 1997). The similar proteins from bacteria or *Arabidopsis thaliana* are in most cases biochemical characterized.

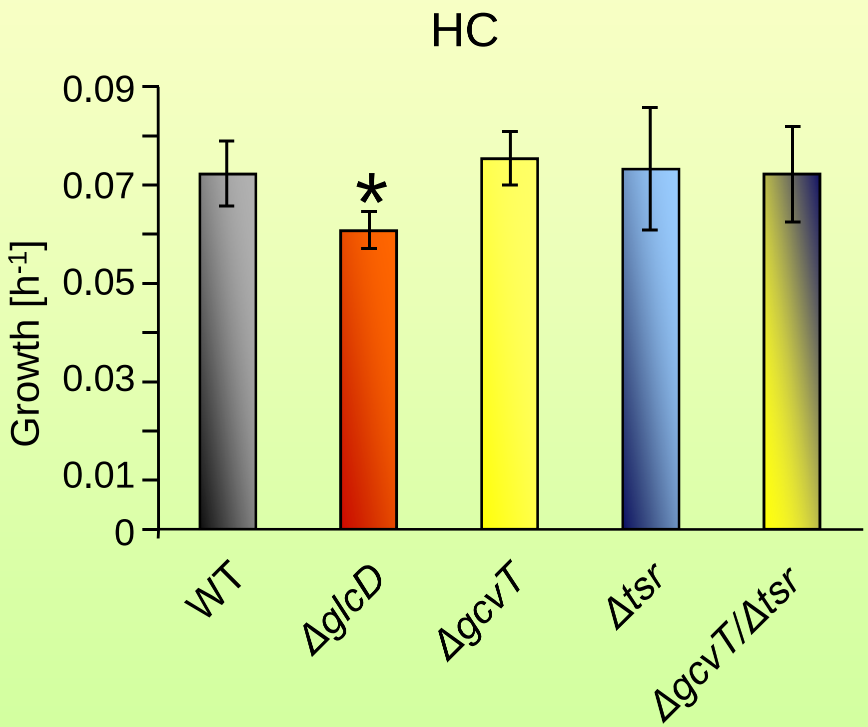
Protein (Abbreviation)	**Organism**	**Acc. No.**	**Literature**	**Similarity e-value**	**ORF in Synechocystis**
Phosphoglycolate phosphatase (PGP)	*A. eutrophus*	P40852	Schäferjohann et al., 1993	$3e^{-08}$	*slr*0458
				$9e^{-06}$	*sll*1349
Glycolate dehydrogenase subunit D (GlcD)	*E. coli*	AAC76015	Pellicer et al., 1996	$3e^{-126}$	*sll*0404
Serine:glyoxylate aminotransferase (AGT)	*A. thaliana*	At2g13360	Liepman and Olsen, 2001	$3e^{-59}$	*sll*1559
GDC, P protein (GcvP)	*A. thaliana*	At2g26080	Bauwe et al., unpubl.	0	*slr*0293
GDC, T protein (GcvT)	*A. thaliana*	At1g11860	Bauwe et al., unpubl.	$7e^{-52}$	*sll*0171
GDC, H protein (GcvH)	*A. thaliana*	At2g35120	Bauwe et al., unpubl.	$2e^{-28}$	*slr*0879
GDC, L protein (GcvL)	*A. thaliana*	At3g16950	Bauwe et al., unpubl.	$1e^{-164}$	*slr*1096
Serine hydroxymethyltransferase (SHMT)	*A. thaliana*	At4g37930	Voll et al., 2006	$6e^{-94}$	*sll*1931
Hydroxypyruvate reductase (HPR)	*A. thaliana*	At1g68010	Bauwe et al., unpubl.	$2e^{-28}$	*sll*1908
				$2e^{-26}$	*slr*1556
Glycerate kinase (GLYK)	*E. coli*	AAB93855	Cusa et al., 1999	$3e^{-64}$	*slr*1840
Glyoxylate carboligase (GCL)	*E. coli*	AAA23864	Chang et al., 1993	$1e-^{94}$	*slr*2088
				$3e-^{58}$	*sll*1981
Tartronic semialdehyde reductase (TSR)	*E. coli*	P77161	Cusa et al., 1999	$3e^{-35}$	*slr*0229

Hagemann et al. 2005

Cyanobacterial 2-PG metabolism: A combination of plant-like 2-PG cycle and bacterial-like glycerate pathway?

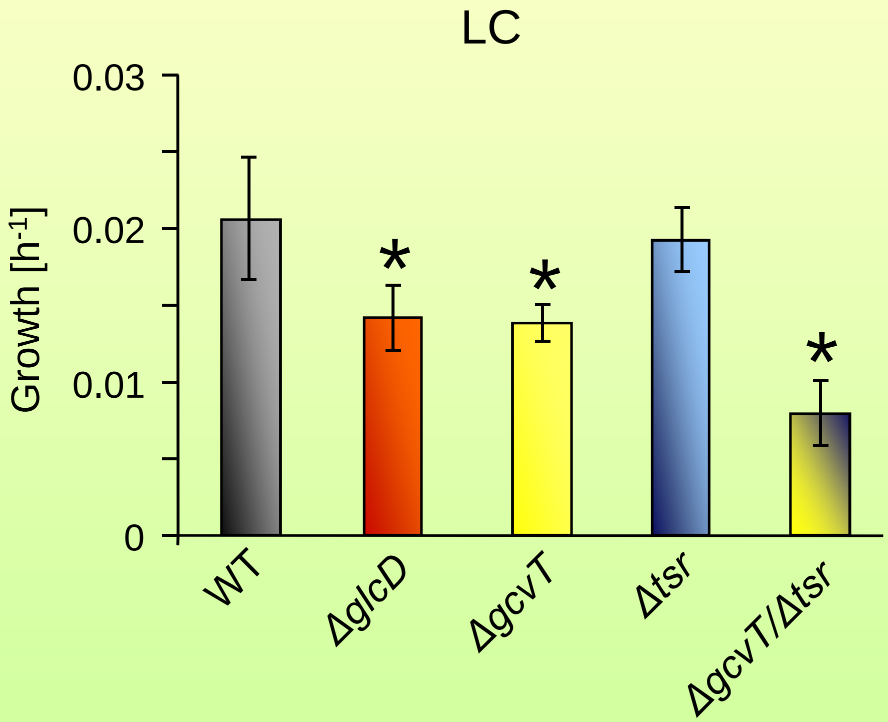


The glycolate dehydrogenase mutant showed diminished growth at HC, while other mutants grew like WT



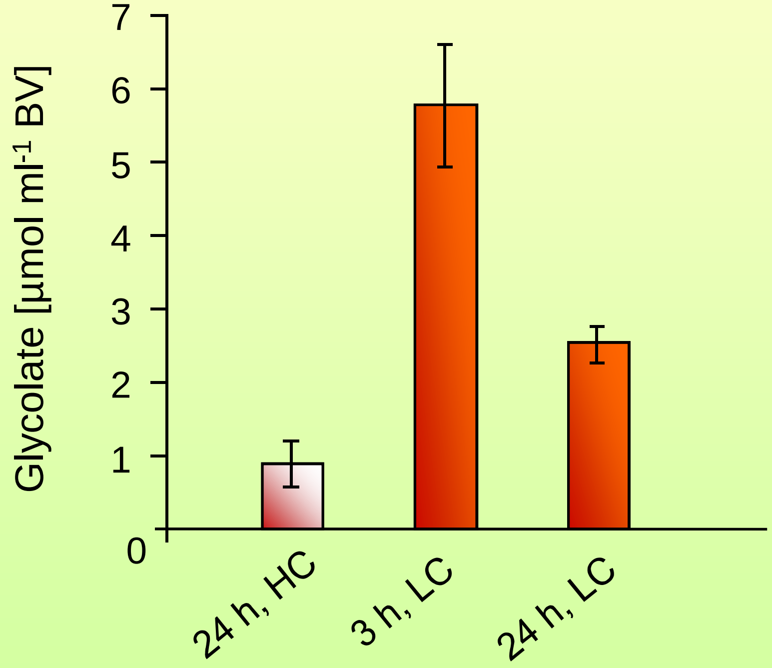
Eisenhut et al. 2006

Mutants defective in plant-like 2-PG cycle showed diminished growth at LC, while glycerate cycle mutant grew like WT



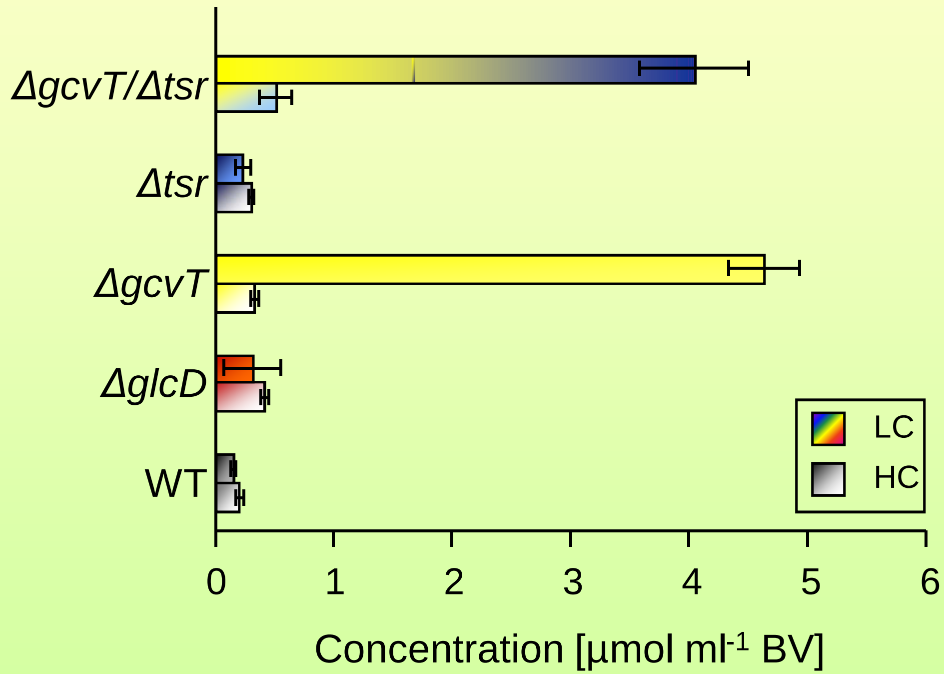
Eisenhut et al. 2006

Glycolate accumulation was only observed in cells of the glycolate dehydrogenase mutant, even at HC!



Eisenhut et al. 2006

Glycine accumulation in mutants of ORFs encoding plant-like 2-PG cycle or/and bacterial-like glycerate pathway proteins



➡ Glycolate metabolism exists but employs at least two pathways in cyanobacteria

Eisenhut et al. 2006