Supplementary Material - Section 1: Lipidomics

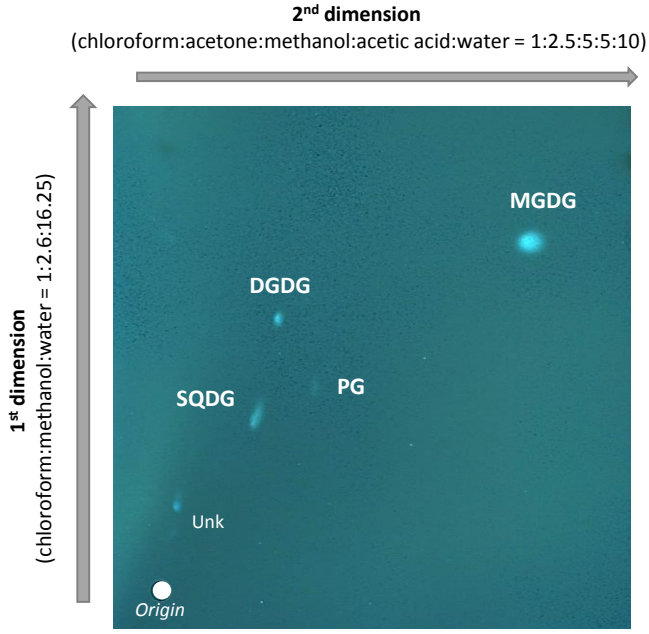


Figure S1: Separation of the four main classes of membrane lipids from *Synechococcus* sp. WH7803 by silica 2-dimensional thin layer chromatography, revealed by 8-anilino-1-naphthalene sulfonic acid under UV light. MGDG: monogalactosyldiacylglycerol; DGDG: digalactosyldiacylglycerol; SQDG: sulfoquinovosyldiacylglycerol; PG: phosphatidylglycerol; unk: unknown compounds.

Table S1: Global relative variations of fatty acids in long-term acclimated cells of the marine picocyanobacterium *Synechococcus* sp. WH7803 grown at different temperatures. The acyl chain data are expressed as percentages of total fatty acids. The length of the acyl chains is expressed in number of carbon atoms.

Growth temperature (°C)	14:0	16:0	18:0	14:1	16:1	16:2	18:1	30:2
16	35.2 ± 0.5	10.3 ± 0.6	0.0 ± 0.0	10.9 ± 0.5	31.2 ± 0.5	0.8 ± 0.1	1.1 ± 0.1	10.4 ± 0.6
18	41.0 ± 1.9	20.1 ± 1.1	0.0 ± 0.0	6.1 ± 2.6	30.1 ± 1.3	0.2 ± 0.2	0.0 ± 0.0	5.0 ± 2.8
22	39.9 ± 1.2	17.8 ± 1.2	0.0 ± 0.0	5.7 ± 0.4	30.9 ± 1.0	0.3 ± 0.0	0.0 ± 0.0	5.4 ± 0.3
25	40.3 ± 1.7	19.9 ± 0.9	0.0 ± 0.0	4.6 ± 0.3	29.8 ± 0.6	0.1 ± 0.0	1.8 ± 1.2	3.4 ± 0.3
28	41.1 ± 0.4	22.8 ± 0.7	0.0 ± 0.0	1.8 ± 0.1	32.3 ± 0.2	0.1 ± 0.0	0.0 ± 0.0	2.0 ± 0.1
30	43.1 ± 0.1	23.5 ± 1.0	0.0 ± 0.0	2.5 ± 0.1	29.3 ± 0.7	0.0 ± 0.0	0.2 ± 0.4	1.3 ± 0.1

Growth temperature (°C)	Average Carbon #	*sn*-1 Carbon #	*sn*-2 Carbon #	Saturated acyl chains	Unsaturated acyl chains
16	15.00 ± 0.02	15.60 ±0.02	14.39 ± 0.03	45.5 ± 0.6	54.5 ± 0.4
18	15.00 ± 0.05	15.67 ± 0.04	14.33 ± 0.06	61.9 ± 0.9	41.4 ± 0.9
22	15.04 ± 0.04	15.68 ± 0.02	14.39 ± 0.06	57.7 ± 0.5	42.3 ± 0.5
25	15.11 ± 0.07	15.79 ± 0.06	14.43 ± 0.08	60.2 ± 1.3	39.8 ± 1.3
28	15.12 ± 0.01	15.86 ± 0.01	14.39 ± 0.02	63.9 ± 0.4	36.1 ± 0.4
30	15.08 ± 0.01	15.79 ± 0.01	14.37 ± 0.01	66.6 ± 0.9	33.4 ± 0.9

A 30:2 diacyl molecular species (most likely C14:1-C16:1 but possibly C14:0-C16:2) was also detected during the MS analyses, but since it represented <5% in almost all samples, it was not taken into consideration in lipid composition calculations.

Table S2: Relative variations of the fatty acids esterified to the two stereospecific (*sn*) positions of the monogalactosyldiacylycerol (**MGDG**), digalactosyldiacylycerol (**DGDG**), sulfoquinovosolydiacylycerol (**SQDG**) and phosphatidylglycerol (**PG**), in long-term acclimated cells of the marine picocyanobacterium *Synechococcus* sp. WH7803 grown at different temperature. The data are expressed as percentages of total acyl chains per glycerolipid (note that in Figure 2, the data are expressed as percentages of total acyl chains for each *sn* position).

	Growth temperature (°C)	14:0 *sn*-1	14:0 *sn*-2	14:1 *sn*-1	14:1 *sn*-2	16:0 *sn*-1	16:0 *sn*-2	16:1 *sn*-1	16:1 *sn*-2	16:2 *sn*-1
MGDG	**16**	1.0 ± 0.0	44.8 ± 0.3	15.5 ± 0.1	5.2 ± 0.3	2.3 ± 0.0	0.0 ± 0.0	29.4 ± 0.2	0.0 ± 0.0	1.8 ± 0.1
	18	2.0 ± 0.6	48.5 ± 1.2	10.1 ± 2.3	1.5 ± 1.2	3.2 ± 0.4	0.0 ± 0.0	34.1 ± 1.7	0.0 ± 0.0	0.6 ± 0.4
	22	1.9 ± 0.1	48.4 ± 0.1	10.2 ± 0.2	1.6 ± 0.1	3.1 ± 0.0	0.0 ± 0.0	34.1 ± 0.3	0.0 ± 0.0	0.7 ± 0.0
	25	2.3 ± 0.0	49.2 ± 0.0	8.5 ± 0.2	0.8 ± 0.0	3.5 ± 0.1	0.0 ± 0.0	35.3 ± 0.2	0.0 ± 0.0	0.4 ± 0.0
	28	1.8 ± 0.1	49.9 ± 0.1	3.0 ± 0.1	0.1 ± 0.1	5.3 ± 0.1	0.0 ± 0.0	39.9 ± 0.1	0.0 ± 0.0	0.1 ± 0.0
	30	2.8 ± 0.2	49.9 ± 0.0	4.4 ± 0.3	0.1 ± 0.0	5.3 ± 0.2	0.0 ± 0.0	37.4 ± 0.4	0.0 ± 0.0	0.0 ± 0.0

	Growth temperature (°C)	14:0 *sn*-1	14:0 *sn*-2	14:1 *sn*-1	14:1 *sn*-2	16:0 *sn*-1	16:0 *sn*-2	16:1 *sn*-1	16:1 *sn*-2	16:2 *sn*-1
DGDG	**16**	5.2 ± 0.2	47.8 ± 0.2	9.5 ± 0.4	2.2 ± 0.2	9.0 ± 0.7	0.0 ± 0.0	25.6 ± 0.5	0.0 ± 0.0	0.6 ± 0.0
	18	5.7 ± 0.4	49.4 ± 0.3	4.6 ± 3.5	0.3 ± 0.5	31.2 ± 0.2	0.0 ± 0.0	9.6 ± 0.1	0.7 ± 0.5	0.0 ± 0.0
	22	5.9 ± 0.1	49.7 ± 0.3	3.7 ± 0.6	0.0 ± 0.0	26.8 ± 0.9	0.0 ± 0.0	13.3 ± 0.1	0.6 ± 0.5	0.0 ± 0.0
	25	5.9 ± 0.2	49.6 ± 0.3	3.1 ± 0.2	0.0 ± 0.0	30.2 ± 0.4	0.0 ± 0.0	10.3 ± 0.1	0.9 ± 0.7	0.0 ± 0.0
	28	3.4 ± 0.1	49.7 ± 0.2	2.0 ± 0.2	0.0 ± 0.0	31.7 ± 0.3	0.0 ± 0.0	12.5 ± 0.1	0.7 ± 0.7	0.0 ± 0.0
	30	5.5 ± 0.4	49.7 ± 0.2	1.4 ± 0.2	0.0 ± 0.0	37.3 ± 0.5	0.0 ± 0.0	5.5 ± 0.3	0.5 ± 0.5	0.0 ± 0.0

	Growth temperature (°C)	14:0 *sn*-1	14:0 *sn*-2	14:1 *sn*-1	14:1 *sn*-2	16:0 *sn*-1	16:0 *sn*-2	16:1 *sn*-1	16:1 *sn*-2	16:2 *sn*-1
SQDG	**16**	1.4 ± 0.1	30.0 ± 0.4	2.0 ± 0.1	0.1 ± 0.0	13.4 ± 0.2	17.2 ± 0.4	33.3 ± 0.2	2.8 ± 0.0	0.0 ± 0.0
	18	1.3 ± 0.3	26.9 ± 1.5	0.7 ± 0.2	0.0 ± 0.0	18.6 ± 2.9	20.7 ± 1.6	29.4 ± 3.0	2.4 ± 0.2	0.0 ± 0.0
	22	1.3 ± 0.2	24.6 ± 1.0	0.7 ± 0.1	0.0 ± 0.0	17.4 ± 0.1	21.8 ± 1.0	30.6 ± 0.2	3.6 ± 0.1	0.0 ± 0.0
	25	1.4 ± 0.1	26.6 ± 0.2	0.5 ± 0.0	0.0 ± 0.0	20.4 ± 0.3	21.4 ± 0.2	27.7 ± 0.2	2.0 ± 0.1	0.0 ± 0.0
	28	1.0 ± 0.1	28.0 ± 0.9	0.1 ± 0.0	0.0 ± 0.0	27.8 ± 0.2	21.2 ± 0.8	21.0 ± 0.1	0.8 ± 0.0	0.0 ± 0.0
	30	1.7 ± 0.2	26.0 ± 0.0	0.2 ± 0.3	0.0 ± 0.0	28.4 ± 0.2	23.0 ± 0.0	19.7 ± 0.4	1.0 ± 0.0	0.0 ± 0.0

	Growth temperature (°C)	16:0 *sn*-1	16:0 *sn*-2	16:1 *sn*-1	16:1 *sn*-2	18:0 *sn*-2	18:1 *sn*-1	18:1 *sn*-2
PG	**16**	0.6 ± 0.1	12.0 ± 0.5	36.9 ± 0.2	38.0 ± 0.5	0.0 ± 0.0	12.5 ± 0.2	0.0 ± 0.0
	18	0.0 ± 0.0	7.6 ± 1.0	50.0 ± 0.0	42.4 ± 1.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
	22	0.0 ± 0.0	6.9 ± 0.2	50.0 ± 0.0	43.1 ± 0.2	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
	25	0.8 ± 0.5	16.3 ± 5.1	34.3 ± 10.4	33.7 ± 5.1	0.0 ± 0.0	19.8 ± 9.9	0.0 ± 0.0
	28	0.5 ± 0.6	10.6 ± 0.5	49.5 ± 0.6	39.4 ± 0.5	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
	30	1.8 ± 0.7	16.7 ± 3.9	45.3 ± 6.0	33.2 ± 3.9	0.0 ± 0.0	2.9 ± 5.8	0.0 ± 0.0

Table S3: Relative variations of the acyl chain length and unsaturation of the monogalactosyldiacylycerol (**MGDG**), digalactosyldiacylycerol (**DGDG**), sulfoquinovosolydiacylycerol (**SQDG**) and phosphatidylglycerol (**PG**), in long-term acclimated cells of the marine picocyanobacterium *Synechococcus* sp. WH7803 grown at different temperatures. The acyl chain data are expressed as percentages of total fatty acids. The length of the acyl chains is expressed in number of carbon atoms.

	Growth temperature (°C)	Average Saturated	Average Unsaturated	Saturated at *sn*-1	Unsaturated at *sn*-1	Saturated at *sn*-2	Unsaturated at *sn*-2	Average Carbon #	*sn*-1 Carbon #	*sn*-2 Carbon #
MGDG	**16**	40.8 ± 0.7	59.2 ± 0.7	3.3 ± 0.1	46.7 ± 0.1	44.8 ± 0.3	5.2 ± 0.3	14.67 ± 0.00	15.34 ± 0.00	14.00 ± 0.00
	18	49.6 ± 3.3	50.4 ± 3.3	5.7 ± 0.1	44.3 ± 0.1	49.1 ± 0.0	0.9 ± 0.0	14.76 ± 0.03	15.52 ± 0.07	14.00 ± 0.00
	22	48.5 ± 0.1	51.5 ± 0.1	4.9 ± 0.1	45.1 ± 0.1	48.4 ± 0.1	1.6 ± 0.1	14.76 ± 0.01	15.52 ± 0.01	14.00 ± 0.00
	25	51.5 ± 0.1	48.5 ± 0.1	5.8 ± 0.0	44.2 ± 0.0	49.2 ± 0.0	0.8 ± 0.0	14.78 ± 0.01	15.57 ± 0.01	14.00 ± 0.00
	28	54.8 ± 0.2	45.2 ± 0.2	7.1 ± 0.1	42.9 ± 0.1	49.9 ± 0.1	0.1 ± 0.1	14.91 ± 0.00	15.81 ± 0.01	14.00 ± 0.00
	30	56.6 ± 0.3	43.4 ± 0.3	8.2 ± 0.2	41.8 ± 0.2	49.9 ± 0.0	0.1 ± 0.0	14.86 ± 0.01	15.71 ± 0.02	14.00 ± 0.00

	Growth temperature (°C)	Average Saturated	Average Unsaturated	Saturated at *sn*-1	Unsaturated at *sn*-1	Saturated at *sn*-2	Unsaturated at *sn*-2	Average Carbon #	*sn*-1 Carbon #	*sn*-2 Carbon #
DGDG	**16**	54.3 ± 1.2	45.7 ± 1.2	14.3 ± 0.9	35.7 ± 0.9	47.8 ± 0.2	2.2 ± 0.2	14.71 ± 0.00	15.41 ± 0.01	14.00 ± 0.00
	18	85.7 ± 0.3	14.3 ± 0.3	32.8 ± 0.8	17.2 ± 0.8	50.0 ± 0.0	0.0 ± 0.0	14.81 ± 0.02	15.61 ± 0.03	14.00 ± 0.00
	22	79.6 ± 0.8	20.6 ± 0.8	32.8 ± 0.8	17.2 ± 0.8	50.0 ± 0.0	0.0 ± 0.0	14.81 ± 0.02	15.61 ± 0.03	14.00 ± 0.00
	25	84.6 ± 0.0	15.4 ± 0.0	36.4 ± 0.0	13.6 ± 0.0	50.0 ± 0.0	0.0 ± 0.0	14.82 ± 0.00	15.64 ± 0.00	14.00 ± 0.00
	28	84.3 ± 0.2	15.7 ± 0.2	35.3 ± 0.3	14.7 ± 0.3	50.0 ± 0.0	0.0 ± 0.0	14.89 ± 0.01	15.78 ± 0.00	14.00 ± 0.00
	30	92.9 ±0.5	7.1 ± 0.5	42.9 ± 0.4	7.1 ± 0.4	50.0 ± 0.0	0.0 ± 0.0	14.86 ± 0.01	15.72 ± 0.01	14.00 ± 0.00

	Growth temperature (°C)	Average Saturated	Average Unsaturated	Saturated at *sn*-1	Unsaturated at *sn*-1	Saturated at *sn*-2	Unsaturated at *sn*-2	Average Carbon #	*sn*-1 Carbon #	*sn*-2 Carbon #
SQDG	**16**	60.3 ± 0.2	39.7 ± 0.2	14.7 ± 0.2	35.3 ± 0.2	47.1 ± 0.0	2.9 ± 0.0	15.33 ± 0.01	15.87 ± 0.01	14.80 ± 0.02
	18	66.6 ± 3.6	33.4 ± 3.6	19.9 ± 3.2	30.1 ± 3.2	47.6 ± 0.3	2.4 ± 0.3	15.42 ± 0.04	15.92 ± 0.01	14.92 ± 0.06
	22	63.9 ± 0.2	36.1 ± 0.2	18.7 ± 0.1	31.3 ± 0.1	46.4 ± 0.1	3.6 ± 0.1	15.47 ± 0.03	15.92 ± 0.01	15.02 ± 0.04
	25	69.2 ± 0.2	30.8 ± 0.2	21.8 ± 0.2	28.2 ± 0.2	48.0 ± 0.1	2.0 ± 0.1	15.43 ± 0.01	15.92 ± 0.00	14.94 ± 0.01
	28	77.9 ± 0.1	22.1 ± 0.1	28.8 ± 0.1	21.2 ± 0.1	49.2 ± 0.0	0.8 ± 0.0	15.42 ± 0.02	15.95 ± 0.00	14.88 ± 0.03
	30	78.8 ± 0.7	21.2 ±0.7	30.1 ± 0.4	20.0 ± 0.4	49.0 ± 0.2	1.0 ± 0.2	15.44 ± 0.02	15.92 ± 0.01	14.96 ± 0.03

	Growth temperature (°C)	Average Saturated	Average Unsaturated	Saturated at *sn*-1	Unsaturated at *sn*-1	Saturated at *sn*-2	Unsaturated at *sn*-2	Average Carbon #	*sn*-1 Carbon #	*sn*-2 Carbon #
PG	**16**	12.6 ± 0.5	87.4 ± 0.5	0.6 ± 0.1	49.4 ± 0.1	12.0 ± 0.5	38.0 ± 0.5	16.25 ± 0.00	16.50 ± 0.01	16.00 ± 0.00
	18	7.6 ± 1.0	92.4 ± 1.0	0.0 ± 0.0	50.0 ± 0.0	8.1 ± 0.2	41.9 ± 0.2	16.00 ± 0.00	16.00 ± 0.00	16.00 ± 0.00
	22	6.9 ± 0.2	93.1 ± 0.2	0.0 ± 0.0	50.0 ± 0.0	6.8 ± 0.2	43.2 ± 0.2	16.00 ± 0.00	16.00 ± 0.00	16.00 ± 0.00
	25	17.0 ± 5.6	83.0 ± 5.6	1.0 ± 0.2	49.0 ± 0.2	18.8 ± 0.5	31.2 ± 0.5	16.30 ± 0.20	16.60 ±0.40	16.00 ± 0.00
	28	11.1 ± 1.1	88.9 ± 1.1	0.3 ± 0.6	49.7 ± 0.6	10.3 ± 0.4	39.7 ± 0.4	16.00 ± 0.00	16.00 ± 0.00	16.00 ± 0.00
	30	18.6 ± 4.5	81.4 ± 4.5	2.0 ± 0.8	48.0 ± 0.8	17.9 ± 3.8	32.1 ± 3.8	16.08 ±0.14	16.16 ± 0.27	16.00 ± 0.00

Supplementary Material - Section 2:
Genes involved in membrane lipid synthesis

Fatty acid biosynthesis – The four enzymes (AccA-D) comprising the initiation module, *i.e.* the acetyl-coenzyme A (CoA) carboxylase (ACC) complex that catalyzes the conversion of acetyl-CoA to malonyl-CoA by addition of a carboxylic group, as well as the malonyl-CoA-ACP transacylase (MCAT) that transfers malonyl-CoA to the acyl-carrier protein (ACP) to produce malonyl-ACP, are present as single copy genes in all searched genomes (Table S4). The following FAS steps are catalyzed by four genes. The first one, β-ketoacyl-ACP synthase III (KAS III), initiates the condensation of the first acyl group and the malonyl-CoA-ACP, producing β-ketoacyl-ACP. The latter intermediate is reduced by the β-ketoacyl reductase (KR), giving rise to β-hydroxyacyl-ACP, which is in turn dehydrated by β-hydroxyacyl-ACP dehydratase (DH) to produce enoyl-ACP. This elongation cycle is completed by the enoyl-ACP reductase (ENR). Again, KAS III, KR, DH and ENR appear to be encoded by conserved single core genes (*fabH*, *fabG*, *fabZ* and *fabI*, respectively) in all marine *Synechococcus* and *Cyanobium*, while two *fabH* gene copies are present in *Synechocystis* sp. PCC 6803.

While in higher plants and *Escherichia coli*, subsequent rounds of elongation are initiated by two condensing enzymes KAS I (from C4 to C16) and KAS II (from C16 to C18), no KAS I enzyme has so far been found in cyanobacteria (including marine *Synechococcus*), suggesting that KAS II might be responsible for the entire elongation. KAS II condenses the growing acyl-ACP with malonyl-ACP to extend the fatty acid chain by adding two carbons at each cycle, using the so-called Claisen condensation reactions (White *et al.*, 2005). Once the elongated acyl-ACP is formed, two alternative reactions are possible: the acyl group can be cleaved by the acyl-ACP thioesterase to release a free fatty acid, or transferred to glycerol 3-phosphate (G3P) for incorporation into the membranes. Search for KAS II in *Synechococcus* genomes revealed that all of them possess at least one *fabF* gene copy, but 20 out of 53 strains (mostly belonging to clades II, III, IV and WPC1) actually possess a second copy, that we called *fabF2* (Table S4). Both copies are significantly more related to *E. coli fabF* (KAS II) than they are to *fabB* (KAS I).

Incorporation into the membranes and complex lipid synthesis - This pathway starts with the acylation of G3P, catalyzed by the PlsX-GPAT system, which is present in all the search genomes. Using the cytosolic acyl-ACP, the membrane-associated protein PlsX catalyzes the

formation of an acyl-phosphate (Acyl-P; Cross, 2016), which is used by the G3P acyltransferase (GPAT) to acylate the 1-position of G3P, forming lysophosphatidic acid (LPA). Finally, the LPA acyltransferase (LPAAT) acylates the *sn*-2 position of LPA to form a phosphatidic acid (PA), the central intermediate of membrane glycerolipids.

The biosynthetic pathways of membrane lipids in cyanobacteria then divide into two branches, leading to the synthesis of the glycolipids or to PG (Petroutsos *et al.*, 2014). In the galactolipid pathway, PA is dephosphorylated by the PA phosphatase to yield diacylglycerol (DG). An ortholog of the, biochemically characterized PA phosphatase of *Synechocystis* sp. PCC 6803 (Nakamura *et al.*, 2007) is present in four halotolerant strains (CB0101, CB0205, WH5701 and PCC 6307) but not in the 'truly' marine *Synechococcus* strains, suggesting that another enzyme is involved in this process in the latter strains. A possible candidate is a membrane protein possessing a PA phosphatase-like domain (Cyanorak cluster CK_0000099). In *Synechocystis* sp. PCC 6803, the DG produced is then used as a substrate for the synthesis of MGlcDG *via* the transfer of glucose from uridine 5'-diphosphate-1α-glucose (UDP-1α-glucose) to the *sn*-3 position of DG (Awai, 2016). In cyanobacteria, it is assumed that MGDG is produced by epimerization of the glucose moiety of the MGlcDG precursor into galactose. In many freshwater cyanobacterial strains, the MGlcDG epimerase is encoded by the *mgdE* gene (Awai *et al.*, 2014; Sato, 2015) that includes a C-terminus Rossmann fold domain and a fatty acid hydroxylase at the N-terminus, the function of which remains unclear (Awai, 2016). In marine *Synechococcus*, the best hit to *mgdE* is a gene that includes only the C-terminal Rossmann-fold domain of the *Synechocystis* gene (Table S5). DGDG is then synthesized from MGDG probably *via* UDP-1α-galactose, as in chloroplasts (Kelly and Dörmann, 2002). The candidate gene (*dgdA*) identified in *Synechocystis* sp. PCC 6803 (Sakurai *et al.* 2007) has an ortholog in all marine *Synechococcus*. Similarly, the UDP-sulfoquinovose synthase (SqdB) and SQDG synthase (SqdX) involved in the synthesis of SQDG (Sanda *et al.*, 2001) as well as the phosphatidyl-glycerophosphate synthase (PgsA) involved in PG biosynthesis are all core proteins in marine *Synechococcus* spp.

Table S4: Genome screening for genes involved in the biosynthetic pathways of the fatty acids in 53 marine *Synechococcus* and *Cyanobium* genomes. Cells filled with grey indicate the presence of one gene copy in the genome. Absence of color indicates that no orthologous gene was found in the genome. **ACP**: Acyl Protein Carrier; **ACC**: Acetyl-Coenzyme A Carboxylase; **MCAT**: Malonyl-Coenzyme A-Acyl carrier protein Transacylase; **KAS III**: β-Ketoacyl-Acyl carrier protein Synthase. **KR**: β-Ketoacyl Reductase; **DH**: β-Hydroxyacyl-acyl carrier protein Dehydratase; **ENR**: Enoyl- acyl carrier protein Reductase.

			Cyanobacterial Fatty Acid Synthase (FAS II)												
				Initiation					**ACP-acyl elongation and termination**						
Sub-Cluster[1]	**Clade**[2]	**Representative sequenced strains**	**ACP** ***acpP***	**ACCA** ***accA***	**ACCB** ***accB***	**ACCC** ***accC***	**ACCD** ***accD***	**MCAT** ***fabD***	**KAS III** ***fabH***	**KAS III** ***fabH2***	**KR** ***fabG***	**DH** ***fabZ***	**ENR** **fabI**	***KAS II*** ***fabF***	***KAS II*** ***fabF2***
5.1	**I**	CC9311, MVIR-18-1, PROS-9-1, ROS8604, SYN20, WH8016													
	II	A15-44, A15-62, CC9605, KORDI-52, PROS-U-1, RS9902, RS9907													
		M16.1, TAK9802, WH8109													
	III	A15-24, A15-28, A18-40, A18-46.1, BOUM118, RS9915, WH8102, WH8103													
	IV	BL107, CC9902													
	V	BMK-MC-1, WH7803													
	VI	MEDNS5, WH7805, PROS-7-1													
	VII	A15-60, A18-25c, NOUM97013													
	VIII	RS9909, RS9917, WH8101													
	IX	RS9916													
	CRD1	BIOS-E4-1, BIOS-U3-1, MIT9220													
	WPC1	A15-127, KORDI-49													
	XX	CC9616													
	UC-A	KORDI-100													
5.2		CB0101, CB205, PCC 6307, WH5701													
		NS01, PCC7001													
5.3		MINOS11													
		RCC307													
		Ortholog in *Synechocystis* sp. PCC 6803	*Ssl2084*	*sll0728*	*slr0435*	*sll0053*	*sll0336*	*slr2023*	*slr1511*	*slr1511*	*slr0886*	*sll1605*	*slr1051*	*sll1069*	*slr1332?*
		Cyanorak database gene cluster #	290	670	342	1134	978	222	221	5457	59	1081	261	66	9077

[1] *sensu* Herdman et al. (2001); [2] see Mazard et al. (2012) and Choi & Noh (2009).

Table S5: Genome screening for genes involved in the biosynthetic pathways of the four main membrane lipids in 53 marine *Synechococcus* and *Cyanobium* genomes. Cells filled with grey indicate the presence of one gene copy in the genome and dark grey cells indicate the presence of 2 gene copies in the genome. Absence of color indicates that no orthologous gene was found in the genome. **GPAT**: Glycerol-3-Phosphate AcylTransferase; **LPAAT**: LysoPhosphatidic acid AcylTransferase; **PAP**: Phosphatidic Acid Phosphatase; **MGlcDG**: MonoGlucoseDiacylGlycerol; **DGDG**: DiGalactylDiacylGlycerol; **UDP**: Uridine DiPhosphate; **SQDG**: SulfoquinovosylDiacylGlycerol; **DG**: DiacylGlycerol; **PGP**: PhosphatidylGlyceroPhosphate. Sub-clusters and clades are defined *sensu* Herdman et al. (2001); [2] see Mazard et al. (2012) and Choi & Noh (2009).

Sub-Cluster[1]	Clade[2]	Representative sequenced strains	*PlsX plsX*	*GPAT plsY*	*LPAAT1 plsC1*	*LPAAT2 plsC2*	*PAP-like -*	*PAP -*	*PAP-like -*	*MGlcDG Synthase mgdA*	*MGlcDG Epimerase mgdE*	DGDG Synthase dgdA	*UDP-SQDG Synthase sqdB*	*SQDG Synthase sqdX*	*CDP-DG Synthase cdsA*	*PGP-Synthase pgsA*	*PGP phosphatase pgpA / pgpB*
5.1	I	MVIR-18-1, ROS8604, SYN20, WH8016															
		CC9311, PROS-9-1															
	II	CC9605, KORDI-52, PROS-U-1, RS9902, RS9907M16.1, TAK9802, WH8109															
		A15-44															
		A15-62															
	III	A15-24, A15-28, A18-40, A18-46.1, BOUM118, RS9915, WH8102, WH8103															
	IV	BL107															
		CC9902															
	V	BMK-MC-1, WH7803															
	VI	MEDNS5, WH7805, PROS-7-1															
	VII	A15-60, A18-25c,															
		NOUM97013															
	VIII	RS9909, RS9917, WH8101															
	IX	RS9916															
	CRD1	BIOS-E4-1, BIOS-U3-1, MIT9220															
	WPC1	A15-127, KORDI-49															
	XX	CC9616															
	UC-A	KORDI-100															

Table S5 (continued)

5.2	CB0101, PCC 6307, WH5701															
	PCC7001, NS01															
	CB0205															
5.3	MINOS11															
	RCC307															
	Ortholog in *Synechocystis* sp. PCC 6803	*slr1510*	*sll1973*	*sll1848*	*sll1752*	-	*sll0545*	*slr1394*	*sll1377*	*sll1376 (C-term)*	*slr1508*	*slr1020*	*slr0384*	*slr1369*	*slr1522*	-
	Cyanorak database gene cluster #	220	1072	223	746	1999	7151	999	327	633	1040	123	333	760	991	-

[1] *sensu* Herdman et al. (2001); [2] see Mazard et al. (2012) and Choi & Noh (2009).

Supplementary Material - Section 3:
Histine-box motifs of the lipid desaturase enzymes

In freshwater cyanobacteria, DesC1 and C2 protein sequences exhibit histidine-rich motifs that correspond to the Δ9-1 and Δ9-2 motifs as defined by Chi *et al.* (2008). In marine *Synechococcus* and *Cyanobium* genomes, two DesC proteins display histidine-rich motifs typical of Δ9-3 and Δ9-4, with only a few mismatches for some strains (Fig. S2). We therefore named these proteins DesC3 and DesC4. The third *Synechococcus* DesC could not be assigned with confidence to one of the histidine-rich motifs and was therefore named DesC6.

For DesA, two proteins were clearly related to Δ12-b and Δ12-c histidine-rich motifs and were therefore named DesA2 and DesA3, respectively (Fig. S3). The third DesA protein cluster was not stable in the desaturase phylogenetic tree (Fig. 5). Its position varied between the DesA/B cluster or at the base of the DesC cluster. Furthermore, it displayed a somewhat intermediate signature between Δ9 and Δ12 histidine-rich motifs, although being closer to Δ12 (Fig. S3). Considering the role of these motifs in determining the function of the active site and the region specificity of the protein, we named this protein DesA4.

Figure S2: Comparison of the three conserved histidine-rich motifs of marine *Synechococcus* and *Cyanobium* DesC-like lipid desaturases with the closest motifs previously defined for cyanobacteria by Chi *et al.* (2008), *i.e.* Δ6, Δ9.1-5, Δ12a-c, Δ15. "X" correspond to non-conserved residues. "Consensus" indicates the motif found in most marine *Synechococcus* and *Cyanobium* sequences. Strain sequences exhibiting variations with regard to these motifs are also indicated and variable amino acids are surrounded by squares. A tentative assignment of the consensus and/or sequence specific motifs to a specific desaturase type or subtype, based on motif conservation, is shown on the right hand side.

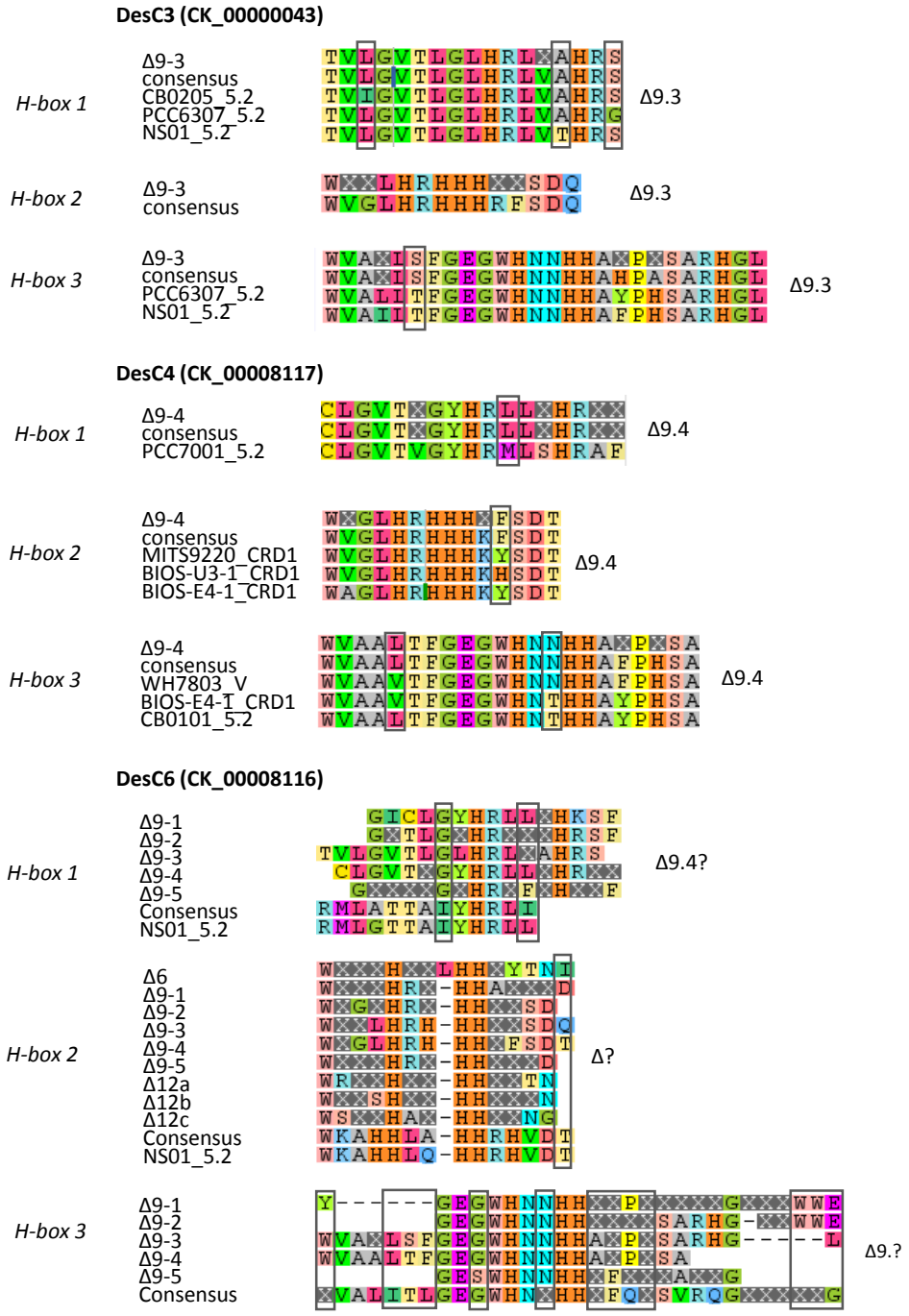


Figure S3: Same as Figure S2 but for marine *Synechococcus* and *Cyanobium* DesA-like lipid desaturases.

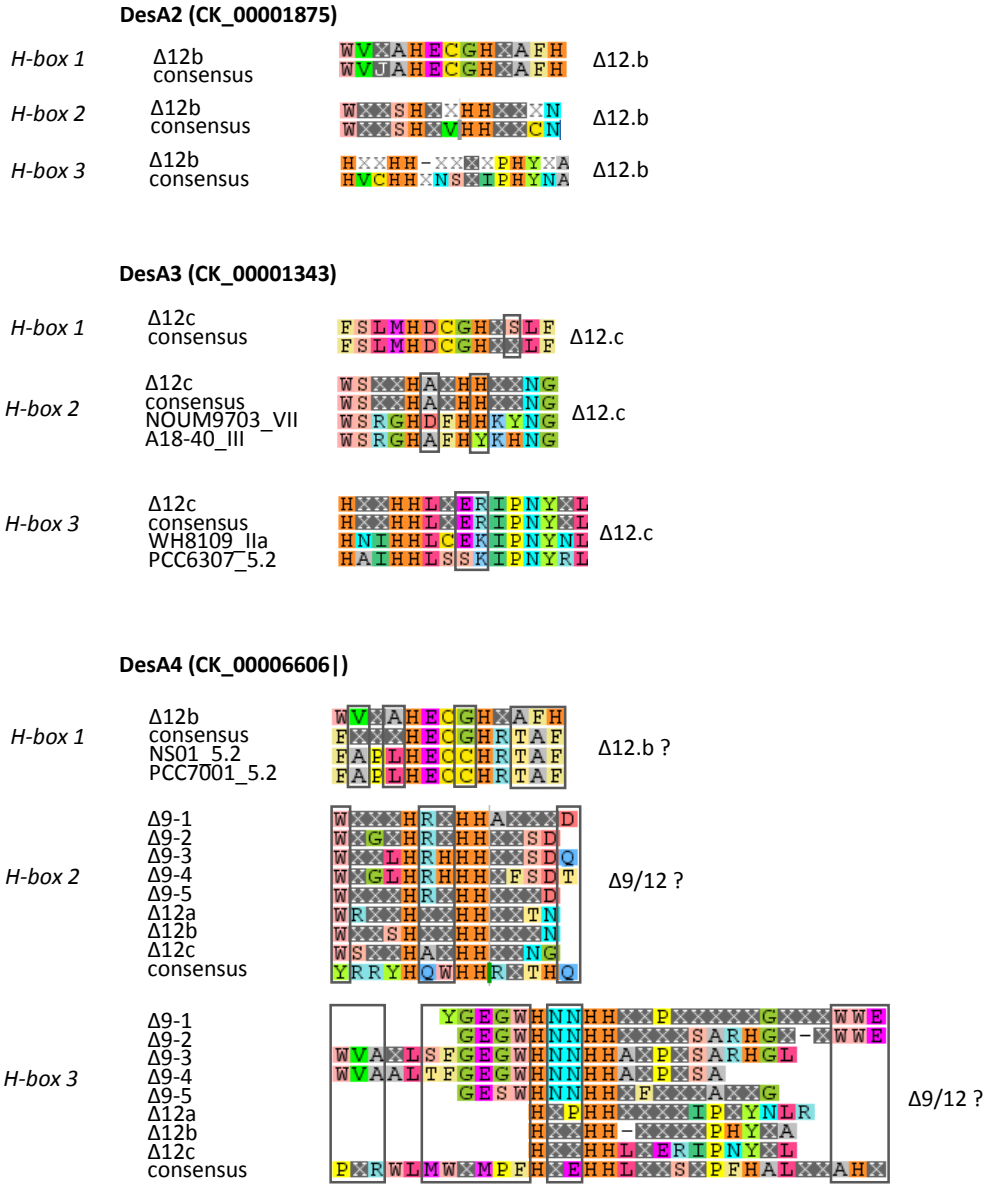
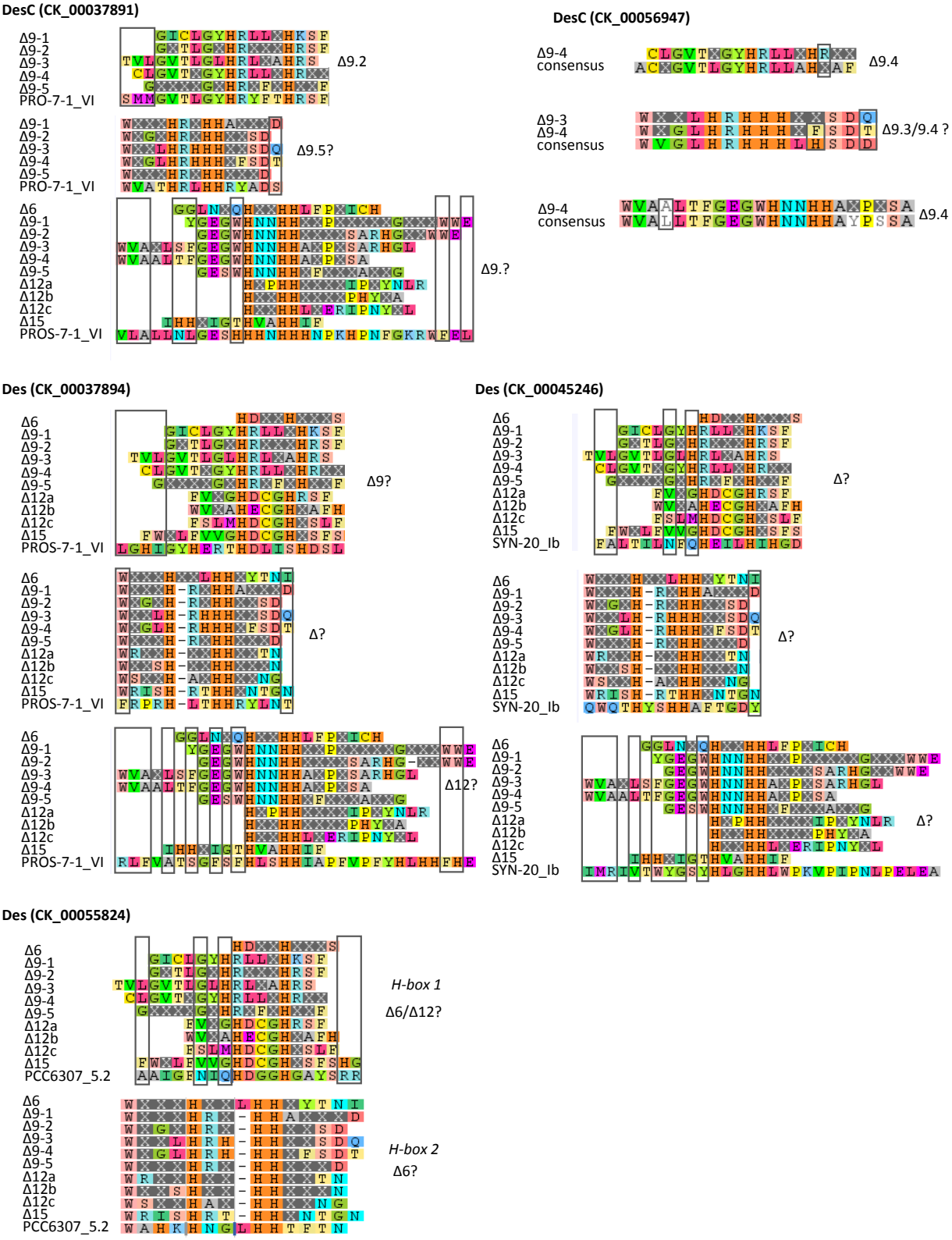


Figure S4: Same as Figure S2 but for other, hardly assignable lipid desaturase genes of marine *Synechococcus* and *Cyanobium*.



Supplementary Material - Section 3:
Analysis of horizontal transfers for lipid desaturase genes

Comparison of the phylogenetic trees for DesC3 and desC4 genes with that obtained with vertically inherited ribosomal proteins (core phylogeny) shows that they are globally congruent, except for a few branch switches (Fig. S5-S6). Besides the instability of the position of clade IX, represented by the sole RS9916 strain, the most notable shift concerns clade IV, associated with clade II based on ribosomal proteins, whereas it is found at the basis of clades II and III for DesC3 and to group with clade I for DesC4. These rearrangements could suggest that these genes have been laterally transferred at least once during the evolution of the marine *Synechococcus*/*Cyanobium* radiation. However, these genes were not detected as part of a genomic island by the Alien Hunter software (Fig. 6) nor did they show a G+C content significantly different from the surrounding genomic region consisting of core genes (data not shown).

The occurrence of multiple horizontal transfers for the DesA3 and desA4 genes is much clearer, given the analyses of the phylogeny, the genomic contexts and the presence of these genes in genomic islands.

Figure S5: Comparison of the phylogenetic maximum likelihood trees obtained for a concatenation of 52 vertically inherited ribosomal proteins (7,072 amino acid positions, left), and for the acyl-desaturases DesC3 (right). Strain description include strain names, sub-cluster, clades and sub-clades (e.g., WH8102_5.1_III_IIIa), as defined previously (Scanlan *et al.*, 2009; Mazard *et al.*, 2012; Farrant *et al.*, 2016). Each clade is shown with a different color.

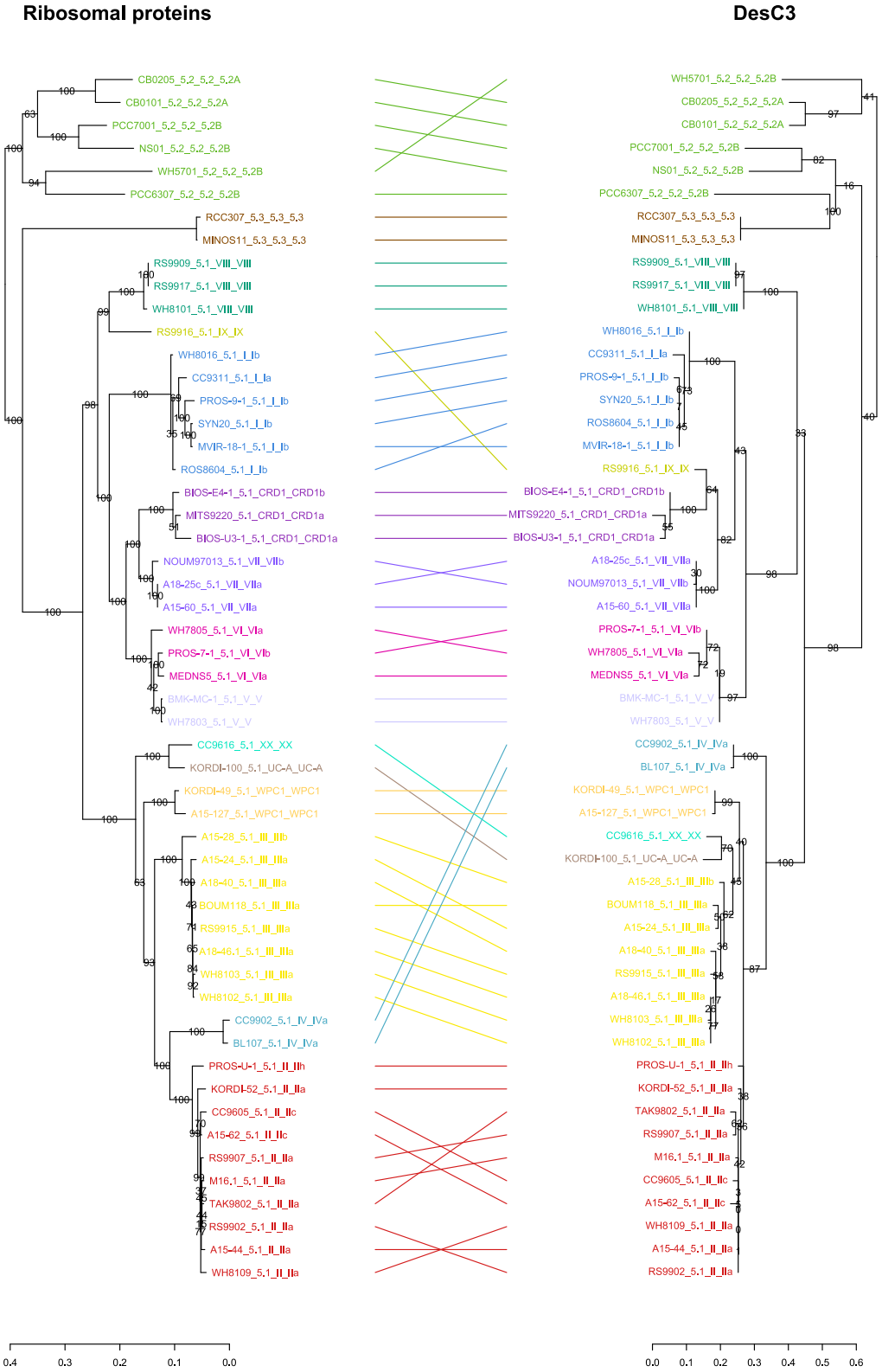


Figure S6: Same as Figure S5 but for DesC4 desaturases.

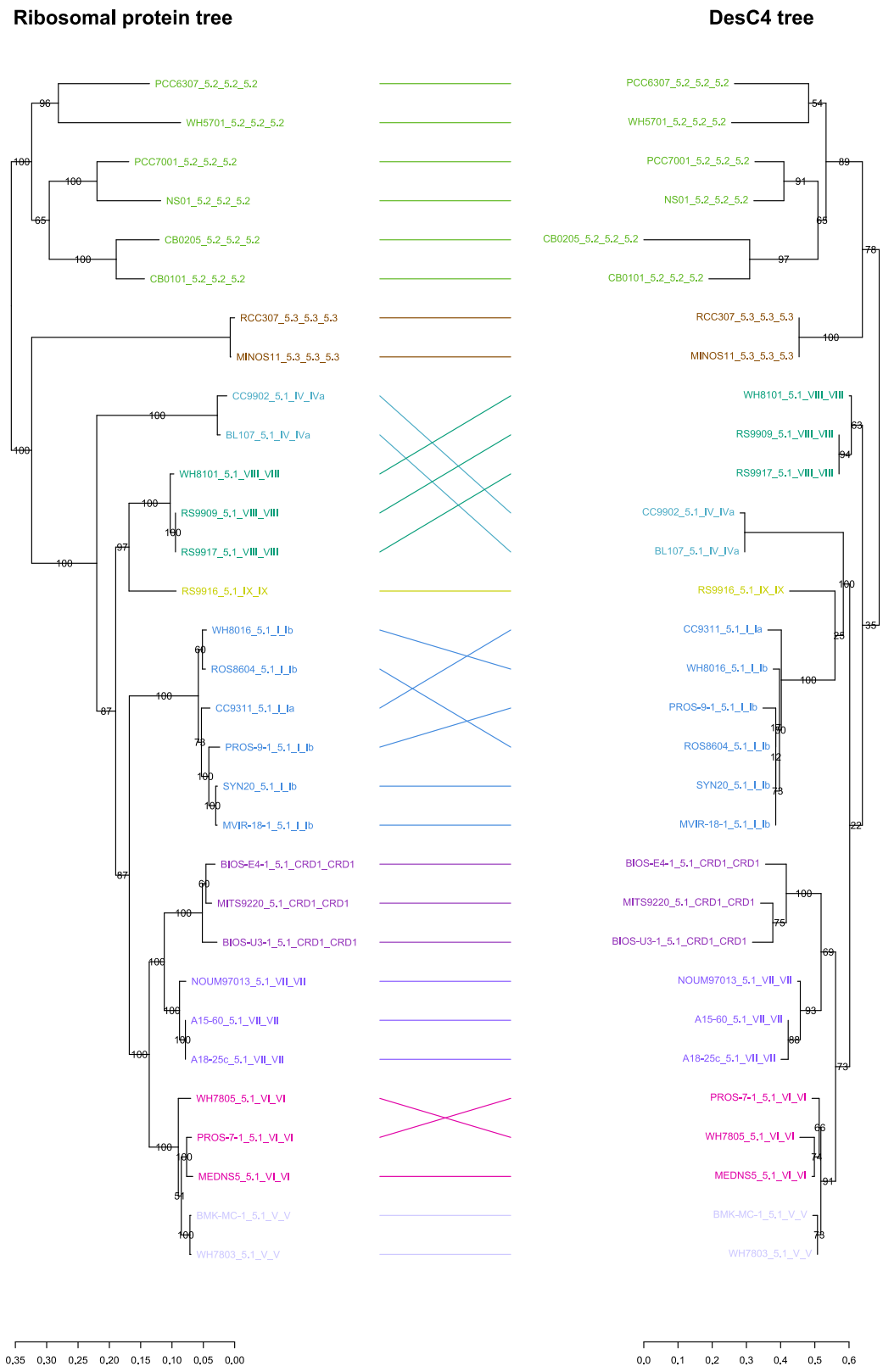
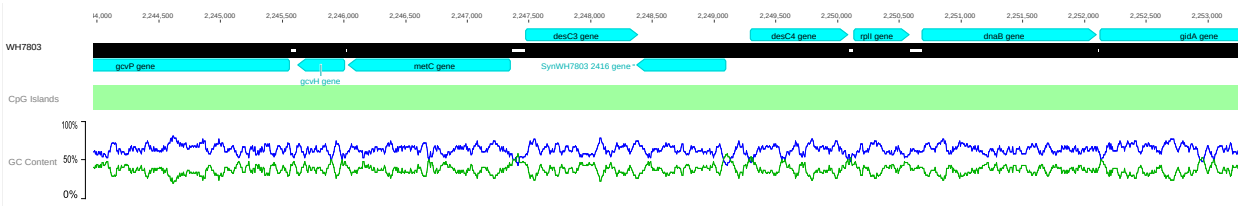
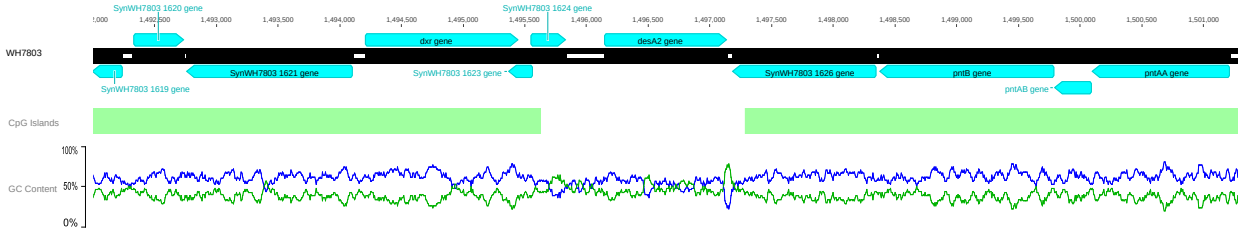


Figure S7: Variability of the local nucleotide composition around *desC3*, *desc4*, *desA2* and *desA3* genes for *Synechococcus* sp. WH7803 as assessed using Geneious® 8.1.5 (http://www.geneious.com, Kearse et al., 2012). Blue and green lines correspond to GC% and AT%, respectively (sliding window size: 50 bp).

desC3/desC4



desA2



desA3

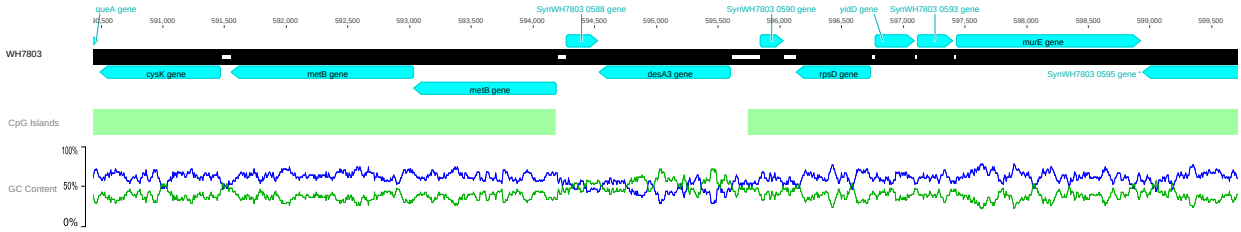


Figure S8: Same as Figure S5 but for DesA2 desaturases.

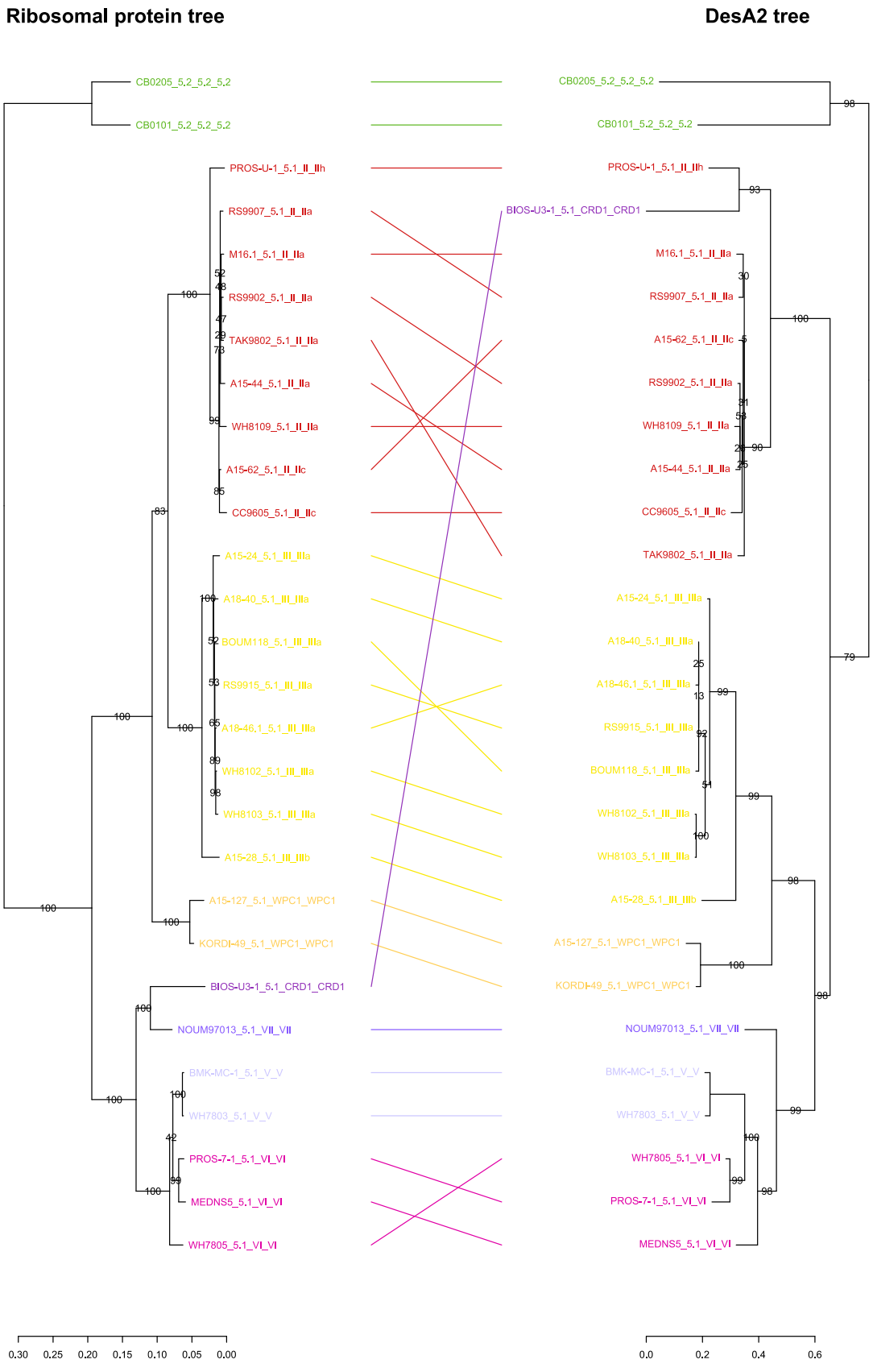


Figure S9: Same as Figure S5 but for DesA3 desaturases.